



Special Program and Abstract Issue

2014 Intersociety Meeting: Comparative Approaches to Grand Challenges in Physiology

Guyton Educator of the Year Award

Herbert F. Janssen



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The humble gratitude I feel as recipient of the Guyton Educator of the Year Award necessitates that I acknowledge my friend and colleague David Osborne for the nomination, the colleagues and students who supported the nomination, and the numerous students who, over the years, helped develop my teaching style and philosophy of education. I must also thank the American Physiological Society, the APS Selection Committee, and Elsevier for recognizing the importance of educators in APS.

Over a century ago, the Flexner Report concluded that didactic lectures were hopelessly antiquated and belonged to an age of accepted dogma, “when the professor ‘knew’ and the students ‘learned’” (1). Abraham Flexner encouraged medical educators to provide students the opportunity to learn rather than be taught. Replacing the “Sage on the Stage” with a learner-focused approach is a worthy goal. I, along with teachers at all levels of the educational process, echo this philosophy. While doing this, it is also important for educators to provide students with the opportunity to master critical thinking skills and to learn to apply content and skills appropriately. For the student who wishes to become a researcher, this means learning to design, conduct, and interpret results. For the medical student, it means learning to engage patients while providing a nonjudgmental and humanitarian application of physiological principles that encourages mental and physical healing. For the student who wishes to become an educator, it means learning the art and science required to advance our understanding of how knowledge is conveyed, recalled, and applied.

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A Matter of Opinion
from the President

What Does Peer Review Need?

Research funding decisions require a strong peer review process. After all, who but the experts can identify the most promising science? At the same time, complaining about peer review has always been one of scientists' favorite pastimes. Although these complaints are not new, I believe our current peer review system for grants is facing serious challenges.

Full Participation

A major problem is that too few senior, experienced reviewers choose to serve on peer review panels. It is easy to blame the difficult funding

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Mentoring Forum

Success in Research at an Undergraduate Institution

Trevor A. Day

Mount Royal University, Calgary, Alberta, Canada



Trevor A. Day

I am honored to have been asked to contribute to the APS Mentoring Forum, especially at such an early point of my own career. In terms of my own training journey, I trained at the University of Calgary, Faculty of Medicine, which is a research-focused institution. Here, faculty face high exceptions to land external grants to fund their research and focus on research with advanced trainees (masters,

doctoral, and postdoctoral), and are evaluated almost exclusively on research productivity. The teaching they do amounts to a few lectures or course coordination in graduate level classes in their area of expertise. It was an incredible place to train in neurobiology. However, most trainees, when they get out, do not end up with jobs at institutions like this. Following completion of my PhD, I landed a tenure-track instructor position at an undergraduate teaching college, where students completed the first 2 years of their programs and transferred to bigger institutions. At the time, I had heard there were efforts to transform the institution into a bachelor degree granting institution, and I wanted to be a part of the process. We completed this transition in 2010, in both name and membership in the Association of Universities and Colleges of Canada. We rolled out many new degree programs, and I was and continue to be involved in both new curriculum and course development. The transition of Mount Royal College to Mount Royal University in 2010 has come with a lot of changes and growing pains, not the least of which were around academic rank and work pattern transitions to include scholarship. My focus in this article will be around workload and scholarship support in a new university setting that focuses its efforts on teaching and training undergraduates. My comments are aimed at relatively new faculty who are trying to start a research program with the inclusion of undergraduate students at a teaching-focused undergraduate institution. Additionally, instructors at small liberal arts colleges

may find my experience helpful if they want to get started, either at their own institutions or in collaboration with other universities. If you are a Chair or a Dean interested in supporting the development of research programs, my experiences outlined below may help in framing the justification for and in securing resources to support these activities.

Post-PhD, not everyone is going to get jobs in research-intensive institutions. If you are one of those new to mid-career faculty members at a small teaching-focused undergraduate institution trying to get started with a research program, this column is aimed at you.

Why Bother?

This article is about working with undergraduate students in the laboratory. Labs are an important part of biology courses, but students in biology degrees often ask what the point of labs are. Their complaint is often seated in the extra cost and time associated with labs in science courses. The fact that most students are not going to go into laboratory-based work post-graduation is often cited as a reason why this extra cost and effort is not worth it. I think this concern misses the point of undergraduate training – it's not just about what we know, it's also important to understand how we know it. Exposing students to experiential learning opportunities is important not only in demonstrating the scientific process but also in student engagement. Canned or cook-book-type lab demonstrations may not be as strong in these outcomes, but taking a step toward projects that allow students to ask a question, troubleshoot equipment, and solve problems takes it to the next level. In an era when college and university teachers are looking for ways to increase student engagement and hand responsibility for learning back to students (e.g., flipped classrooms), one need look no further than the good old-fashioned laboratory research project as a means to foster student engagement.

There is something special about student lab projects. Students who are not academically strong in the traditional classroom setting (e.g., exams) can shine in

laboratory research projects, and I have witnessed this first-hand time and again. When students have some choice over the topics they are investigating, they develop a sense of ownership over the project and often put a tremendous amount of effort into it and do their best work. Our role is then to facilitate these opportunities through securing space and equipment, integrating these experiences into the curriculum, having a list of ideas to give to them as they try to come up with their own questions, guiding the protocol development, data collection, and analysis, and helping with dissemination opportunities. These are not trivial endeavors, but as I will outline below, they are well worth the efforts.

With the utility of the undergraduate research project established, we need to move toward the challenges and strategies in implementing these activities into the undergraduate student experience.

A Framework for Research Support in Physiology

In the context of a small undergraduate institution in transition, with research support being in its infancy, I continue to look for ways to build and maintain a program that is sustainable. Part of that includes aiming to align my relatively high teaching load with my research interests. Also, over the past 4-5 years, I have been very active in collaborating with investigators and advanced trainees at other institutions, as well as building a research program at Mount Royal University. University faculty members in all disciplines need support for their scholarly endeavors, primarily in terms of protected time. However, laboratory scientists have unique and specific needs to be able to carry out their research activities, which can cumulatively conspire against you if they are not in place. If you are at a small institution trying to develop your own program of research, organizing these arguments in this way may help with lobbying efforts to find support. I'll outline a framework for research support, with examples of my own challenges into the following six categories.

1. Equipment. By far, the single most important element that has contributed to my ability to do research at my home institution has been the purchase of equipment. The new degree implementation and the building of a new science wing came with an influx of capital money to purchase equipment for labs. I got organized with respect to what equipment I wanted (even some blue sky

stuff). It paid off, and, in hindsight, I am surprised with what we were able to obtain for the lab. The Integrative Physiology Lab now has a pretty comprehensive list of equipment that we can use to ask a variety of straightforward cardiorespiratory and cerebrovascular questions; most of this has been purchased since 2009 when I set out to start building a physiology program at MRU. Most of this equipment has been used by undergraduate students for lab demonstrations or projects, and much of it has been used in collaboration at other universities or in field work (e.g., high altitude), so I can now make contributions to collaborations as well. I think it is important to frame the purchase of equipment for teaching (and expand how you define that), but then make sure you get items that are research quality.

2. Time. There are two work patterns at MRU. In the teaching/service (TS) work pattern, faculty teach four courses each in the fall and winter semesters (eight total per year) and serve on committees at the departmental, faculty, and/or university level. There is no requirement to be involved in scholarly activities, although many faculty members do get involved. In the teaching/service/scholarship work pattern, faculty teaching loads are reduced to three and three, and they are expected to engage in scholarly activities, which they are also evaluated on for rank and promotion. The choice to keep the teaching load of the TSS work pattern relatively high reflects the teaching-focused emphasis of our institution but amounts to little protected time for research. Many TSS faculty spend their summers doing research, forgoing holidays, leading to inequity between the work patterns. However, it is really the only way to make progress, particularly if it is with students, as the academic year fills up with teaching, marking, course development, and service obligations.

3. Space. If you are at a small institution, you are unlikely to have a lab with your name on the door. You'll likely be using space that is dedicated to teaching. To carry out projects, we need to work around the scheduled courses booked in the room, which take precedent. The result is that we are often collecting data during evening and weekends, and we can't always leave equipment set up after collecting data because there is often a scheduled lab the next day. I have been able to lobby for a space that is dedicated to human physiology courses and then have renovations done to make it more conducive to these activities (e.g., removal of lab benches, installation

of moveable tables/chairs and power cables on pulleys from the ceiling, equipment on moveable carts).

4. Trainees. Science is a team sport, and unlike many academic disciplines, we rely on advanced trainees to do a lot of the data collection and analysis. I have been fortunate enough to have been successful in a number of grants to cover for student salaries for two full-time research assistants for the last four consecutive summers. You can do a lot more with two than with one, and our summers have been very productive, sometimes also in thanks to student volunteers. The trick is to prepare well in advance for grants, ethics applications, and project ideas so these are in place to hire students when they are looking.

It's hard when you only have them for a summer or a semester, but you can break projects into small components and build a data set over a period of time with a group of students. However, one shouldn't underestimate the power of a few motivated students. After training, you can easily get a few data sets collected over a few months. For one study this past summer, a small group of us collected data on 21 participants for a 3-hour protocol in 1 week! It took a lot of organization ahead of time, but it can be done.

It's also difficult to complete manuscripts when students have moved on to other programs or jobs. It is likely that you will be doing the bulk of the writing, but I have been pleasantly surprised at the level of commitment and motivation students show in helping see a project through to completion, even after they have moved on to another phase of their lives. Given the developmental level of an undergraduate student, you are going to have to mentor things like reading and critiquing manuscripts, writing and editing, analyzing data, making figures and statistical analysis. It's helpful if the curriculum can ladder these skills so that students arrive in senior years more prepared.

5. Ethics. Most biological disciplines require some sort of ethical approval before they begin their research, but having animal and biomedical ethics in place can be a more complex endeavor. Animal care and an animal ethics process are separate entities but are interconnected, given that appropriate housing is a part of humane treatment. Smaller institutions may not have the budget or expertise to develop or maintain

an animal care facility. Also, small institutions may not have the internal expertise to handle the safety concerns associated with human biomedical research. At my institution, the Office of Research Services arranged for the human biomedical ethics board at the nearby research-intensive University in Calgary, such that human biomedical applications from MRU could be cleared there. In the mean time, I have worked with the MRU human research ethics board, which has traditionally focused on behavioral studies, to develop a biomedical process that can at least allow student projects to go through. After a few years of successful applications, they have since allowed me to submit low-risk applications from my own research program as well.

6. Funding. Typically, when scientists negotiate a new position at a university, a part of that negotiation is start-up funds for equipment and lab space renovations. Often, larger pots of external money can be applied for, which the university would match. This kind of arrangement isn't usually in place at smaller institutions, so we are almost entirely dependent on an internal capital expense process, which will understandably be geared toward teaching in the laboratory. We have a number of small internal grant processes (\$4,000-10,000), which are very competitive and can help faculty get on their feet. I am often looking for external sources for student salaries, and my success in finding them has improved over the years. Most of us at small institutions are likely not going to be competitive for the larger governmental agency grants, so it's best to look for other avenues, at least while you build your track record. We only just became eligible for our large national governmental awards. In addition, we just don't have the grant-writing support (internal peer review) that larger places have. Sometimes there are private donations for equipment or private funding opportunities. It's worth a conversation with the fundraising office of your institution to see whether there are ways to align yourself with strategic initiatives. It might mean having a donor's name on a door, which can give a much-needed injection of cash to get things rolling.

Aside from student salaries, operational funds, and equipment, we need money for conference and collaboration travel. In addition, some journals charge page charges, adding another roadblock for

dissemination if you don't have a large grant. It's likely that some of these costs, at least early on, will come out-of-pocket. That is a deterrent to some, but I enjoy travel enough and the payoff is high enough that I see it as an investment in my career.

What Kind of Research Should I Do?

I am acutely aware of the limitations that a small institution imposes on my own research program ambitions. This, to some degree, was the choice I made when I took this position. However, I continue to strive toward finding ways to make research with undergraduates in my institution feasible and sustainable. One of those strategies was to change to the kinds of research I was doing and to aim to integrate my teaching and research activities together. I trained in respiratory neurobiology using amphibian and rodent models. When I started my faculty position, I was teaching various courses in "human" anatomy and physiology, and there were neither animal physiology courses nor animal care facilities. It was clear that doing animal work was not in the cards anytime in the near future. So, I switched my focus to working with human participants, and aimed to merge my teaching and research together. My strategy was to leverage the capital purchase of equipment for teaching that would allow me to get started with some straightforward questions in integrative human physiology. I retrained by starting collaborations with other human physiologists at nearby universities and took courses on new techniques using new equipment.

In smaller institutions with limited research infrastructure and support, it is helpful to expand our view of scholarship. I think a broader perspective on what scholarship could be includes three elements: 1) creating new knowledge, new understanding, or new perspectives, that is 2) peer-reviewed by some expert and is 3) disseminated to some stakeholder audience. Seeing scholarship in this way, beyond just traditional experimental research, opens up your view of what kinds of professional activities could be included as a part of your workload. I try to view scholarship in three possible categories: 1) traditional experimental research that I can present as posters at conferences and publish in peer-reviewed journals, 2) educational scholarship, and 3) science communication.

Experimental Research

It goes without saying that primary experimental research published in peer-reviewed academic journals is the cornerstone of scholarly output in our field. Aside from doing studies that are aimed at publishing in peer-reviewed journals, there are other things you can focus on, if only to get you started to gain some momentum. Abstract and poster presentations are obvious ways, and these can be both peer-reviewed and non-peer-reviewed. You can build up a research question with a series of small preliminary data sets, getting feedback and training students along the way. Similarly, student literature reviews that result from directed reading courses could be expanded, formatted, edited, and submitted to undergraduate journals.

Educational Scholarship in Science

There are many avenues to pursue educational scholarship. Faculty can get involved in developing teaching resources and textbook publishing. One of my colleagues is a co-author on a widely adopted introductory anatomy and physiology textbook. Our institution has identified the Scholarship of Teaching and Learning (SoTL) as a major initiative across disciplines, and we now have an SoTL institute with a scholars program that comes with money for successful applicants. Moving toward educational research is not for everybody, but it is a possible solution for teaching intensive physiologists who wish to focus on pedagogy and are willing to spend some time with the educational literature and learn to use qualitative methodologies. An interesting possibility is to publish standard laboratory research techniques as laboratory demonstrations for adoption in laboratory courses. The APS journal *Advances in Physiology Education* has a new "Source Book" series, which is a great place to publish laboratory methods and demonstrations if you have a unique idea and the equipment to collect data and show representative traces.

Science Communication

In the summer of 2008, I participated in a 2-week science communication residency program at the Banff Centre. Involvement in this program and staying engaged with an ever-growing community introduced me to a whole other group of scientists who were interested in what the public thinks about science. We learned about events and performances, and writing for the

popular press (newspapers, magazines, and books), podcasting, television, web content, and social media. Although this kind of work is not usually on the radar at most institutions, small institutions are in a position to expand their view of scholarly work and can play leadership roles in science outreach to the media and the general public. I have a colleague in my department who is aiming to publish popular articles about science and how science is done by embedding herself in field studies, participating in the data collection, and writing these stories in the popular press. There is a large and active community of science communicators on social media. You can start following and reading work by these people to see whether this kind of work interests you. Personally, I am actively involved in science communication performances and have a lot of ideas about articles for the popular press. I'll get to them in my spare time!

How Do I Get It Going?

Starting a research program is a massive challenge at any institution, especially if you are also building a lab from scratch. These challenges are compounded with the usual constraints of small institutions where the teaching and service loads are high, there is minimal space, and equipment is limited. I have tremendous sympathy for my colleagues who do not teach in bachelor degrees, where they are teaching service courses in the first and second years only. Similarly, some faculty are doing work that an undergraduate can simply not participate in (e.g., mathematics, philosophy). However, I think physiology is a discipline that students at any level can be involved in, even if it's just helping with instrumentation, holding a mouthpiece, or entering comments and tags in a software program. I have been able to build a program in this context with some hard work, lobbying, and taking a long view. Below are a few strategies that might help in getting your own program off the ground.

Get Research into the Curriculum

One of the most important things I was able to do early on was develop ways to include research-like activities into the curriculum. I coordinate a third-year, full-year course in human physiology that has a lab. We ladder skill development across the year in the lab to start with some simple data collection and analysis, move to a full lab manuscript on a "canned" demonstration, and finally,

in the second semester, after some training, students can design their own experiments ($n = 1$) to demonstrate a known phenomenon. This experience ladders them into a more advanced fourth-year option in applied physiology I created, where the students can do real research by investigating a novel question with a larger data set ($n = 6$ or higher). This course has allowed me to feed the students questions I have in mind for proof-of-principle. Last year, three group projects from this course submitted their abstracts to a local symposium to present talks. This year's course (ongoing as I write this) will likely do the same to a different local symposium I found. If successful, these projects can be expanded on in the summer months, and these more advanced students are great candidates for full-time paid or volunteer work with larger projects in the summer. We also implemented "directed reading" and "independent project" courses into the curriculum as senior options. Here, we can work one-on-one with students on a research project. I haven't had any difficulty finding students who are interested in these courses, especially as I meet the keen ones in the third-year course, and these independent project courses have represented invaluable opportunities to try out an experimental question or expand a data set. It's important that other courses at the lower levels begin skill development early. Make them read and make them write. The most important developmental step is for them to stop accepting textbook information as fact and to start seeing the caveats in experimental models and designs. So many of them have come to me to say they don't know what to believe anymore, which I think is great!

Integrate Your Teaching and Research

It was clear early on when I started at MRU that I was not going to be able to do any animal work in the near future. Given I was teaching and developing courses across the curriculum in human physiology to various audiences, I decided to switch my focus toward research with human participants so my teaching and research could feedback on each other. Here is where collaborations were critical to help me retrain in new methods with new equipment. Ethics and safety are also different considerations when working with humans. The fact that I can teach and coordinate courses that I now have some expertise in allows me to bring this expertise in the classroom and enrich the student experience in the lab. Similarly, small student projects in these courses can often lead to larger research projects. Laboratory support and lab instructor

expertise are issues I have had to contend with, and I have had to do many of the things myself that faculty at large universities can take for granted in terms of laboratory support. Through some effort, lobbying, and support of the department and the chair, we have a dedicated full-time lab instructor who helps with the third-year physiology course, and a new lab resource person has been hired to support the lab's activities. The lab is now renovated and dedicated to courses and research in human physiology year round.

Frame Your Research Activities as Teaching and Mentorship

"Teaching release" can be a dirty word at smaller teaching-intensive universities. In addition, teaching hours are easy to count, giving the administration the illusion of equity in terms of workload. Research activities are very hard to quantify, and there are colleagues at my institution that see research as a distraction from the core business of the institution, despite the fact that MRU is expecting it from those of us on the TSS work pattern. I try, wherever possible, to frame my research activities as teaching and mentorship, as that is what it is, even if it doesn't look like 3 contact hours of lectures per week and exams. We keep lobbying to have these activities throughout the year and in the summer count as a part of our quantifiable teaching workload.

Get Students Involved

No matter the student population you work with, I am convinced there are ways to get students involved in laboratory projects in physiology. My recent experience tells me that they are hungry for it and enjoy spending time in the lab. The lab is a lot more relaxed (until it's business time) and engaging than the typical classroom lecture. Of course there is a place for everything, but the lab is a great addition to the training experience of students, especially if they are pushed to build things, troubleshoot, problem solve, and collect data. As I write this line, a group of fourth-year undergraduate students are finding middle and posterior cerebral blood velocity measurements using transcranial Doppler ultrasound on a participant and are about to start a lower-body negative pressure protocol. With some effort, this kind of thing need not be a unique experience for undergraduate students. This group has told me that this project is the most fun they have had in their degree, and their commitment to the project has led to an incredible amount of development in just a semester.

I think it's also useful for students to see a project to completion beyond just handing in an assignment for a grade. Perhaps there are ways to get them to make posters and present to the college or university community. If you can expose students to manuscript writing, editing, and the peer-review process, they can gain insight into how the publication and knowledge creation process is carried out. Perhaps you can submit abstracts to local, national, or international symposia or conferences from their work. Our university has a number of student research days, and the buzz around the posters as people ask questions is exciting for everyone. Departments and student associations often have pots of money to fund students for conference travel. In the summer of 2013, two of my fourth-year students self-funded a trip to Birmingham, UK, to present posters at the International Union of Physiological Sciences. They have both since gone off to graduate school, and these experiences were invaluable for their training and their resumes. As much as I hate that students might go out-of-pocket for these kinds of opportunities, it is a tremendous investment in their education, and many students seem to understand the impact.

Lastly, I always look for ways for students to mentor each other. I love watching more senior students teach and mentor more junior students. The usual ladder of trainees in science is not there at a small institution (masters, PhD, post-doc). Finding ways to ladder mentorship within an undergraduate degree is important for both the new and soon-to-be graduating students. Recently, I have found opportunities to get fourth-year students to present to third-year students about their projects. The fourth-year students get experience presenting, and the third-year students can see where their efforts can take them. Start early in their training through the curriculum or identify interested and motivated students and get them involved in the lab early. If you can find students early in their training, you can develop a relationship and their skills over a longer period of time and have their help for a longer period of time.

Research opportunities with students can include 1) volunteer experiences, 2) laboratory projects in courses, 3) independent project courses, and 4) paid research assistant work in the summer or throughout the year. By pushing these initiatives, you can be a feeder program for graduate training with colloquies who run labs at other universities. This strengthens your relationships

with other labs, and you can continue to collaborate with these students. Many of my previous research trainees have gone on to graduate programs in the labs of friends and collaborators at other universities, and it has been incredible to watch their development and keep working with them on new projects.

Collaborate

Science is performed in teams, and the single most important thing I have done to get my own program off the ground is to collaborate with other investigators and their teams at other institutions. Small places often have “one of everything,” so there often aren’t groups of people working in a specific area to develop a collaborative team, leaving you alone to do your work. I have only just begun to find potential collaborations at my own institution through a multidisciplinary project. More importantly, build an international community, both for development of expertise to work on bigger projects and to place graduates from your program into graduate programs in other labs. Perhaps getting adjunct status at other institutions can allow you to co-supervise more advanced trainees. You are an asset to a larger lab because you come with new ideas, you are trained, and you already have a salary. Take a sabbatical if you can. You can get a tremendous amount of work done with time to focus, especially if you incorporate visits to other labs into the time. I was a part of a large international expedition to high altitude in Nepal in 2012, and this experience did so much for my own development track record by maintaining and making new international relationships and new ideas for future projects. Much of this has stemmed from friends and collaborators resulting from conference travel. It’s critical to travel to meetings and to collaborate to maintain relationships, generate new ideas, and stay up-to-date.

Build a Program

Many colleagues at my own institution frame scholarship in terms of “projects.” It is more helpful to think in terms of a “program,” however modest and diverse your program shapes up to be. This perspective has improved my own development of proposals and grant applications, and will help immensely if you ever want to be successful in larger external funding opportunities. External granting agencies want to see you developing an independent program, not just be involved in a series of projects. I also think it’s a useful exercise to articulate what your program is about and

where it is going. However, there is nothing wrong with working with other labs on projects where you have the interest and the time. What’s important is deciding what your goals are.

Write Grants

Without a large grant and advanced trainees, it seems unlikely that I could get any work done at all. In fact, a reviewer for a large federal grant application I submitted recently stated as much – that it was unlikely that I could be productive working only with undergraduates in the summer. I aim to demonstrate that it is possible, and I have been relatively productive through the model I have outlined here. Part of the reason is that I have consistently landed small grants that have allowed me to travel for training, conferences, and collaboration, as well as pay student salaries in the summer. These grants have ranged between \$4,000 and \$15,000. All researchers know how much work grant writing is, but this is not a reason not to try. If you identify small pots of money and chase them regularly, you’ll get better at the process, develop templates for grant structure, and improve your writing. I am also amazed at how much clearer my research ideas are forced to get just by the process of writing a proposal. Don’t let early “failures” in landing money discourage you. Your proposal writing will improve, your ideas will get better, and eventually you’ll start bringing money in.

Reinvent Yourself

Don’t be afraid to change directions if there seems to be a more fruitful path. I trained in animal physiology and made the switch to working with human participants because I saw this as more feasible in the long run, despite the efforts in retraining. I trained in laboratory work and am developing an interest in fieldwork. My training in respiratory neurobiology is leading to an interest in the kidney. Perhaps you will find a grant that is for something that is only tangentially related to your expertise and interests. It might be worth the effort. Similarly, collaborations can take your work in a whole new direction. Being flexible and open to moving in new directions can help move your productivity.

Get on Committees

Many researchers are tempted to limit their service work so they can focus on developing their programs. I understand the impulse. However, strategic committee

membership can help bring to light the various needs of laboratory research and the roadblocks that exist in your institutions. If the committees don't exist, spearhead their development. We have a department, faculty, and institutional level research and scholarly activities committee, a scholarship review committee, and a committee aimed at supporting promotion and celebrating undergraduate research. I have served on most of these at some point, and having a voice at the table from someone who is actively involved in research is important to raise awareness and lobby for support. The added benefit is that people will see that you are still serious about making service contributions to your institution and are not just focusing on your own research.

Promote Yourself

I was initially hesitant to promote the work I was doing with students, but it is the only way for people inside and outside your institution to be aware of your activities. Develop a relationship with people in the communications office and feed them stories. You never know where a little promotion can lead in term of new opportunities for support.

Challenges and Opportunities

Having outlined a few strategies to get your research going above, it's worth outlining some challenges that you might face in the process and ways to navigate them.

Finding Your Place

Let's be honest, you are not going to become a "big player" if your platform is a small undergraduate institution without advanced trainees and you have a high teaching and service load. That's okay, but calibrating your expectations of yourself is important. I don't have any research mentors at my own institution, nor have I had collaborators until very recently where an interdisciplinary project is coming together. I am in a constant state of not knowing where I stand. When I collaborate at other institutions, I feel like I have only just begun, that my potential contributions are small, and that I can't keep up with their pace. When I am at my home institution, I feel ahead of the curve and find it difficult to be patient with the slow pace of change and the level of support. The truth of my own potential is likely somewhere in between, and much of my own ability to build a program and make contributions is

limited first by my own efforts and imagination, which is what I try to focus on. Much can be done to find your place, but it's worth being conscious of the challenges we face so we can calibrate our own expectations of the institution and ourselves, and the challenges we face can be articulated and systematically addressed when the opportunities to lobby arise.

Dabbling in Science

Once you go down the rabbit hole, it's hard to hold back. However, the workload demands can make it such that you might only have time to dabble, at least during the academic year. It's hard to dabble in science. Dabbling will not get you external grant funding, which is likely necessary in terms sustainability if the institution is not putting a large amount of resources into the initiative. The difficulty is that we can't change how science is done, and we are a part of a larger community, regardless of the internal rhetoric of the institution being unique or student-focused. My own view is that involving students in research is not contrary to a student-focused mandate but rather augments it. Ultimately, release from formal classroom teaching is what faculty need to get a program off the ground, which is frowned upon by many (e.g., "It seems that research is more important to you than teaching," I often hear). It's a challenge that administrators need to grapple with. What kind of institution are we going to be? What role will research play and will we support it? Either the university is going to fund it (e.g., equipment, student salaries, travel money) so that you can focus on your high teaching load while doing some research with students with little pressure to publish, or they need to give you start-up funds and teaching release so you can be competitive with other investigators at larger institutions while you apply for grants, write ethics applications and proposals, and build a record of productivity. The latter is likely financially impossible and unpalatable to the sensibilities of a small institution, and the former is likely financially impossible too. This leaves you to navigate how ambitious you want to be in the context you are in.

Expectations, Support, and the Pace of Change

You may be frustrated by the slow pace of change, especially if the institution is asking you to do scholarship but has not yet worked out the kinks in terms of support. At my own institution, we are expected to be scholars and include students where possible, and we are evaluated

on these activities for rank and promotion, but the university has not yet addressed all of the roadblocks I addressed above. In a changing academic culture, you may experience naysayers at your home institution by people who feel threatened by change and who see the research endeavor as a distraction to teaching. It's probably not worth too much effort to worry about these opinions; rather, find ways to show that what you are doing is indeed teaching of a kind – providing training opportunities and mentorship to students seeking experiential training in physiology. Research training is both teaching and mentorship, and needs to be recognized as such in terms of value, support, and workload credit. In the short term, you will likely not get any workload credit for these efforts, and you will also likely be out-of-pocket for expenses. These things are worth bringing to the attention of administrators, but it's likely best to just get over it. Being a faculty member comes with job stability and flexibility, so these personal costs are a likely consequence of starting a research program, especially early on.

Some faculty at my own institution, sometimes after trying for a while, have concluded that it's too hard and move to a work pattern that does not require scholarship (i.e., more teaching). This is a justifiable response to the difficulties of building a program, depending on your own motivation and life circumstances. However, if you have the interest and the energy, it represents an important leadership opportunity that can pay huge dividends in terms of student training opportunities and career satisfaction.

Measuring Success

Given your other commitments, it's important not to measure your success, as many research-intensive institutions do, on the amount of external funding you bring in. Measure it instead on the number and impact of student experiences you facilitate. These might include non-peer-reviewed abstracts and student presentations, student publications in undergraduate journals, or more traditional outcomes like peer-reviewed abstracts and manuscripts.

Advantages of a Small Place

One advantage I have stems from a significant disadvantage. We do not have advanced trainees, but that comes with the added benefit of not having to fund graduate student and postdoctoral salaries. Now that

we have some equipment in place, the most significant expense I encounter is funding undergraduate salaries in the summer, which are significantly less than those of pre-doc or post-doc trainees. I have managed to find grants to pay two full-time salaries for the past four summers. It's not without significant effort, some luck, and timing, and I have had to be organized well in advance with project ideas for grants, and identify students early.

Another advantage is the diversity of the institution I work in. I am not surrounded by neurobiologist like the institution I trained in. However, I have constant interactions with faculty of different disciplines, which expands my perspective. I also teach more broadly, far outside of my own areas of expertise. This diversity has led to research ideas that I may not have envisioned in a more focused research context. I am grateful for these perspectives, despite the fact that I feel like I am always learning new areas, and I will likely not come fully to grips with the literature in every area I am working in. This does not stop me from coming up with new research ideas in new areas all the time and enjoying the new ideas students bring to the table.

My main limitations are my time, motivation, and imagination. The latter is continually fostered by working with students in the laboratory. I find working with students in a laboratory setting a continual source of inspiration, and it is now one of the most rewarding aspects of my career.

Conclusions

When embarking on the development or continued maintenance of a research program at an undergraduate institution, it's worth considering your motivations and to be clear about why you are doing it. Is it to build your CV to make a move to another institution? Is it to facilitate student experiences? Is it to fulfill a workload obligation? Is it for your own intellectual curiosity and interest? Is it to make a contribution to your field? These are all valid reasons to justify the endeavor, but a frank assessment of motivations and limitations, as well as strengths and creative ways to overcome the limitations, is a worthwhile exercise.

Working with and mentoring senior undergraduate students has been one of the most rewarding experiences in my early career, despite some of the difficulties

associated with getting it off the ground and making it a sustainable part of my workload. I have learned, if only from making the effort to getting it rolling, that it can be done and that it's worth the effort.

I hope this will be helpful to some readers in supporting their own activities. I welcome your thoughts, feedback and questions. Feel free to contact me at tday@mtroyal.ca.

Acknowledgments

I want to thank the many students who have worked with me in recent years, all of whom have made contributions to

my research program, and many have done work that has or will lead to peer-reviewed abstracts and manuscripts. Their comments to me over the past 5 years have helped shaped this column. I also want to thank my research mentors and collaborators (both PIs and graduate students at other universities) in helping me in the continued development of my skills, helping me with my record of productivity, and in getting my own research program off the ground. You all know who you are! ●



MentorNet

E-Mentoring for Diversity in Engineering and Science

The American Physiological Society (APS) has partnered with MentorNet, the award-winning non-profit online mentoring network for women and those underrepresented in science, technology, engineering, and mathematics (STEM).

MentorNet's One-on-One Mentoring Program pairs APS mentors with students from over 100 campuses. 95% of MentorNet students persist to graduation and 91% remain in the STEM fields three years after they complete the program.

Communicate via email in **less than 15 minutes** per week
Network with other professionals
Change a student's life

Become a Mentor Today:

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Continued from page 1: A Matter of Opinion from the President

environment and the need to focus on grant writing. Furthermore, once tenured, what is the incentive for thankless tasks such as reviewing grants? It requires a lot of time and hard work, and offers few rewards. It can also be frustrating – and even demoralizing – to review grants when so few of them will get funded. In this climate, altruism can only go so far. But for peer review to work as it should, a balance of investigators at every career stage – early, mid-career, and senior individuals – must be willing to participate.

Strategic Decision-Making

Peer review is not a perfect process. Bias and inconsistency are inherent problems, and there is also a potential for abuse. The current system seems to favor a negative tone, but since all experiments have limitations, I believe that the interests of science would be better served if reviewers spent more effort providing positive statements about how best to improve the work rather than simply pointing out the flaws. To address some of these problems, funding bodies such as NIH should take a more active role in evaluating the quality of the peer review and as strategic decision makers.

When I served on study section, I observed problems with peer review grow to the point where many scientists came to believe that getting a fundable score on a grant was driven more by chance than by the quality and significance of their work. One obvious challenge is that the current system does not readily embrace broad strategic decision-making. If we want to “fix” peer review, the solution will require that scientific review and program officers as well as study section chairs take a more active role in assessing the quality of the reviews themselves and how the work fits with the funding agency’s overall goals.

Effect of a Hypercompetitive Environment

Adding to the issues described above is the imbalance between research funding and demand from the

scientific community. The doubling of the NIH budget during the 1990s led to a rapid expansion at medical schools. New buildings went up, and institutions hired new faculty to fill them. Now that the NIH budget has flattened and in fact has decreased in real terms, the workforce is too large to sustain with the research dollars available.

A recent blog post by NIH Deputy Director of Extramural Research Sally Rockey included data showing that nearly 50% of all first-time NIH grant recipients never get another grant (<http://nexus.od.nih.gov/all/2014/10/28/retention-of-first-time-r01-awardees/>). This attrition rate should be a wakeup call. The challenge before us is how to achieve the kind of diverse, talented, and sustainable scientific work force that can lead us in the 21st century. Achieving this will require asking hard questions and keeping the greater good foremost when we answer them. Are we bringing too many people into the system? Are we bringing in people who lack the breadth and depth of talent and/or training to succeed after their first grant? Is it a combination of these and perhaps other factors? And what are the implications of continuing a Darwinist approach after the first grant is awarded? Step one is to diagnose the problem accurately.

Conclusion

The big question is how to productively address these problems. Part of the solution can be improving peer review, and one way to do that is to ensure the full participation of the scientific community. I want to challenge the APS membership – especially those who have experience as reviewers – to step up and do your part: We have met the enemy, and s/he is us. Unless we do something to improve the system, we will share the responsibility and consequences if it fails. ●

David M. Pollock

Here are some resources to help APS members who want to take part in peer review

- Peer review advice for trainees, junior faculty, and mentors: <http://the-aps.org/peerreview101>
- Editorial: “Civil, sensible, and constructive peer review in APS Journals”: <http://ajpregu.physiology.org/content/305/3/R171>
- How to join a peer review board: <http://the-aps.org/PeerReviewBoard>
- More peer review resources: <http://the-aps.org/peerreview>

Continued from page 1: Guyton Educator of the Year Award

Preparing students for different careers is a challenge for any educator. Dispensing knowledge is important but falls short of what the educator hopes to accomplish. Likewise, teaching critical thinking skills is important, but without knowledge and practice, the practical aspects of our discipline will be lost. The grandmaster of chess may possess outstanding critical thinking skills but would be lost attempting to explain even the rudimentary concepts of cardiovascular or renal physiology. Paraphrasing Kahlil Gibran's comments "On Teaching" from *The Prophet* (2), the teacher gives not of his or her wisdom but rather faith and lovingness. For if the teacher is indeed wise, s/he does not bid you enter the house of wisdom but rather leads you to the threshold of your own mind. The astronomer may speak to you of his or her understanding of space, but s/he cannot give you understanding. For the wisdom of one person lends not its wings to another (2).

How is this best accomplished in our modern-day educational setting? Students communicate using short text messages and 140-character tweets. They attempt to multitask while studying, even though research clearly demonstrates this learning technique is flawed. Modern-day technology has replaced many aspects of teaching that were considered essential in Flexner's time. The aspects of lecturing that were considered appropriate by Flexner, i.e., introducing new material, etc., is being replaced with online computer-based systems. If we as educators feel we can resist this cultural shift, we are sadly mistaken. Quite possibly the most appropriate approach would be to embrace both the culture and the technology and use them to our advantage whenever possible.

What resources and approaches are available to help students become critical thinkers, and are we using these as effectively as possible? Many who teach the basic preparatory science courses feel underutilized when their teaching consists almost entirely of reading PowerPoint slides in class that contain the same information a conscientious student could acquire by reading a textbook chapter. The modern educator should promote a paradigm shift that assigns responsibility for obtaining foundational information to the student before class. As a result, the value of face time is elevated, and this in turn allows for activities

that encourage development of critical-thinking skills. This seems to be a much more reasonable pedagogical approach that capitalizes on available technology and the culture of the current learner.

Physiology is a practical discipline and is seldom learned for its own sake. This being the case, the student needs to acquire the advanced critical thinking skills required to apply physiology in the research laboratory or healthcare setting. Adapting the basics principles of critical thinking set forth by Paul and Elder (5) is helpful in designing our pedagogical approach. Steps in the critical thinking process follow a basic format in most, if not all, situations (3). These steps include the following:

1) Define the purpose for undertaking the current endeavor. Obviously this is an important step that dictates the course of the action that is to be followed. This is true whether the goal is completing a research project or caring for a sick or injured patient.

2) Formulate questions that address the identified purpose. This important step can easily be overlooked if one rushes to a conclusion, diagnosis, or treatment plan. Failure to ask the appropriate question(s) can easily lead to faulty assumptions, inadequate information, misdiagnosis, and flawed treatment. In addition to the problem-specific question(s), basic questions also need to be asked. What information is needed to resolve the research question or make a correct diagnosis? What alternative explanations might also account for the research or clinical findings? What tests would be needed to differentiate between the various explanations? What assumptions am I making? Are they valid? Other questions will vary based on the goal or purpose determined previously.

3) Obtain the information required to answer the previously formulated questions. This is an important part of the critical-thinking process. What do I know? What can I discover in the available literature? If conflicting information is present in the literature, which conclusion is supported by the preponderance of data? What information is unavailable to me? What conclusions does the research or clinical data support? Each of these questions is important and must be answered before a valid answer can be obtained.

4) Derive a solution to the formulated questions based on your knowledge and the acquired information.

This is one of the most challenging steps for students to learn. Some students are able to rely on innate abilities or prior experience, whereas others struggle. As noted in *The Prophet* (2), educators cannot give students their understanding. Each student must master his or her own understanding using inductive and deductive logic to solve the research and medical problems encountered. Simply put, one cannot endow students with an algorithm for drawing logical conclusions. Instead, each student must practice this skill repeatedly and search for his or her own algorithm. The educator can, and should, participate in this process by scaffolding the critical thinking process for the student. They can also encourage the student to engage the mental process that can lead each student to the development of expert-type critical-thinking skills.

5) Apply the solution to the given situation. In the classroom, the instructor can propose scenarios based on the available literature. In this setting, the student should evaluate the possible outcomes purposed by the educator and then compare each to the solution that is anticipated. By pondering each situation and then choosing from the most likely solutions, the student can learn to build his or her own problem-solving algorithm. S/he must continue this learning process in the laboratory or clinical setting where the data are not always clear and the symptoms seldom follow the textbook example.

6) Determine whether the application of the proposed solution produced the desired results. Here again, the educator can help the student predict cause-and-effect relationships. Which physiological system is activated, what is the most logical outcome of this response, and which treatment will most likely produce the desired result? Again, the classroom example is usually well defined, whereas the real-world situations are more variable, and the outcome does not always follow a set pattern described in the textbook.

7) Reflect on the process to determine whether it produces the most desirable outcome in the most effective and efficient manner. What did we do right and what could we do better? This step will help

each person evaluate his or her own problem-solving approach and hone it to a finer edge. We learn from what we do, which in turn will help each person advance his or her own set of critical-thinking skills.

Approaches such as the flipped classroom (7), problem-based learning (PBL) (6), and team-based learning (TBL) (4) have been shown to enhance the value of face-time while allowing students the opportunity to master content outside the classroom. These pedagogical approaches require students to come to class having prepared adequately. Textbook material, reliable online sources, and published scientific reports provide this information. These sources can be enhanced using voice-over PowerPoint slides or short video clips prepared by the educator that contain basic content and problem sets. This leaves class time for discussions emphasizing the application of that content. This approach allows the student to engage in critical-thinking activities through interaction with other students and the professor. The professor is no longer a presenter of facts and becomes a participant in the learning process and is able to analyze the student's thought process.

Although the formal elements of thought and the process of scientific inquiry have been defined, a didactic presentation of those elements will never improve a student's critical-thinking ability. Developing logical reasoning skills, in research and clinical practice, happens best under the tutelage of a well trained educator who encourages frequent, deliberate practice. There is no shortcut or magic solution that will allow this to happen without repeated practice where the student takes responsibility for his or her own learning. Using this model, the educator becomes a coach who uses in-class problem-solving exercises that challenge students to expand their knowledge and problem-solving skills. The educator monitors performance and provides feedback to improve the learning behavior of students. Students cease to function as passive note takers and become active learners. This role shift places the student in a position to increase his or her knowledge of the content, develop critical thinking skills, and more importantly learn to combine skills and knowledge to solve problems in the research laboratory and healthcare setting. ●

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Meetings and Conferences

Experimental Biology 2015

March 28-April 1, 2015 • Boston, Massachusetts

14th International Conference on Endothelin: Physiology, Pathophysiology and Therapeutics

September 2-5, 2015 • Savannah, Georgia

Physiological Bioenergetics: From Bench to Bedside

September 9-12, 2015 • Tampa, Florida

Cardiovascular, Renal and Metabolic Diseases: Physiology and Gender

November 17-20, 2015 • Annapolis, Maryland

the-aps.org/mm/Conferences

Chapter News

Nebraska Physiological Society Meeting

On behalf of the Nebraska Physiological Society (NPS) and the NPS council, I am pleased to report on the activities of our society for the year. Many thanks go to the members of the NPS Council, NPS staff, and staff of UNMC's Department of Cellular and Integrative Physiology for their work with NPS throughout the year and especially in helping to organize this year's annual meeting. We also thank our generous sponsors that helped make this meeting possible.

One of the best parts of being a physiologist is knowing that my research and teaching helps us understand more about life. Physiology is the study of the integrative nature of life and is a translational science that incorporates information from an array of fields, from biochemistry to environmental science. As physiologists, it is easy for us to see the relevance of our work, but we also appreciate that it is not only our responsibility to advance our knowledge in the field of physiology but also to communicate the relevance of this work to the public. One of the goals of the American Physiological Society and this Nebraska chapter is to foster the communication of physiology concepts and research to the public, either to teachers and students in area schools through our PhUn week activities or through other public outreach campaigns. This year, we decided to focus the majority of our efforts on public outreach events, including making public outreach a focus of this year's annual meeting.



David Epstein at book signing with NPS member Tamra Llewellyn

Some of NPS's outreach activities this year included an outreach booth at the 2014 NE SciFest event, an outreach presentation and booth at the Nebraska Association of Teachers of Science (NATS) annual meeting, a successful APS Chapter Activity Grant award (written by Alicia Diener) to help fund future outreach activities involving the Vernier LabQuest system, numerous outreach events at Omaha-area schools, and, lastly, NPS council members gave APS awards to high school students for excellence in physiology research. At the GNSEF, Jameson Collier from Wayne Junior Senior High School was given an APS award for his project "Gender as a Variable of Hypoxic Sensitivity" and at the NJAS, Brittany Boyd from Zoo-Academy-Papillion La Vista G12 was given an APS award for her project "The Effect of Temperature on the *Cyanea Lamardckii* Polyp Strobilation."

Our annual meeting this past year was held on Saturday, October 11th at the Durham Research Center on the University of Nebraska Medical Center's campus from 9 AM to 12:30 PM. The theme of the event was exercise physiology and was officially titled "Athletic Performance, the Human Factor." The keynote address "The Sports Gene: Inside the Science of Extraordinary Athletic Performance" was given by David Epstein, an acclaimed science writer and New York Times bestselling author. Following the keynote address, we had a physiology, health and wellness expo and break-out sessions. The speakers featured during the



David Epstein, author of *Sports Gene: Inside the Science of Extraordinary Athletic Performance*

breakout session included Patrick Lambert (Creighton University), Anne Stanco (Innovative Wellness), Kris Berg (UNO), Christie Toland (Natural Grocers), Jenna Yentes (UNO), Sandy Bikus (Mind Body Spirit), Dusty Slivka (UNO), and Sarah Paasch (Karma Yoga). There were 21 sponsors/organizations that had a booth at the

expo or contributed items for door prizes for the event. With the help of Vicky Cerino from UNMC's Public Relations Department, this event received significant media attention, with interviews and event information presented on local radio (KIOS FM), newspaper (Omaha World Herald), TV (KETV channel 7), and numerous other media outlets.



Vernier LabQuest system demonstration. Left to right: Peter Pellegrino, Bryan Becker, Irving Zucker, Keshore Bidasee, and Sean Bidasee

We had a record attendance for an annual NPS meeting, with 187 attendees. Of those 187, over 100 were non-NPS members, a good indicator that our goal of increasing public awareness about NPS, APS, and communicating the importance of physiology-related research to nonscientists at this event was achieved. Although we will likely not sponsor an outreach event like this every year, the success of this year's meeting sets the stage for future NPS councils to try alternative annual meeting formats that can continue to make this chapter of the American Physiological Society one of the most active and innovative in the country. ●

Carol Fassbinder-Orth
Nebraska Physiological Society President



Join the American Physiological Society at EB 2015 in Boston!

March 28 - April 1, 2015 • Boston, Massachusetts

Experimental Biology is an annual meeting comprised of more than 14,000 scientists representing six participating Societies and 30 guest Societies. Primary focus areas include anatomy, physiology, biochemistry and molecular biology, investigative pathology, nutrition, and pharmacology.



Deadlines

Abstract: January 21, 2015
Registration: February 2, 2015
Housing: February 23, 2015

apsebmeeting.org

Experimental Biology

Experimental Biology 2015 March 28-April 1, 2015, Boston PHYSIOLOGY PLATFORM SESSIONS

Saturday, March 28, 2015

Room			
205B	9:30 AM-11:30 AM <i>MCS Symp</i> Microcirculation: Oxygen/Blood Flow Frame	1:30 PM-3:00 PM <i>MCS Symp</i> Microcirculation: Inflammation	3:30 PM-5:00 PM <i>MCS Symp</i> Microcirculation: Signaling/Channels Butcher
205C	2:15 PM-5:15 PM <i>WEH Section Award</i> WEH Trainee Award Finalists Session and Data Diuresis		
206A	1:00 PM-3:00 PM <i>APS Workshop</i> Big Data Workshop Larkin/Lindsey	3:15 PM-5:15 PM <i>APS Workshop</i> Proteomics for the Physiologist Adhikari/Rinehart	
207	3:15 PM-5:15 PM <i>Communications Comm Symp</i> Connecting with the Media Goodman		
209	1:00 PM-3:00 PM <i>Science Policy Comm Symp</i> Reproducibility in Research: What Are the Problems? How Can We Fix Them? What Happens if We Don't? Northcott		
210A	8:00 AM-12:00 PM <i>Education Comm Symp</i> It's All in Your Head – A Refresher Course on the Brain and Systems Control Young/Rodenbaugh		
210B	5:30 PM-6:30 PM <i>Cannon Award Lecture</i> Yanagisawa		
211	1:00 PM-5:00 PM Second Annual APS PG Group Conference		
212	3:00 PM-5:00 PM <i>NCAR Section Award</i> Data NCARnation Haack/Dick		

Sunday, March 29, 2015

Room	8:00-10:00 AM	10:30 AM-12:30 PM	3:15-5:15 PM
205B	CV Section FT Cardiovascular Responses to Trauma Hester	Physiologists in Industry Comm Symp Targeting Gut Microbiome in Human Diseases and as Novel Therapeutics Moreno Quinn/Intapad	MCS Landis Award Lecture Fukumura
205C	CV Section FT Cerebrovascular Abnormalities in Hypertensive Disease Dorrance/Warrington	10:30 AM-12:30 PM NCAR Section Symp Angiotensin Type 2 Receptors in the Brain: A Functional Coming-of-Age in Cardiovascular Control Sumners/Bruce	2:00 PM-3:00 PM CAMP Section Davson Lecture Aperia 3:15 PM-5:15 PM CV Section FT Vascular Endothelial Cell Insulin Resistance: A New Target for Reducing Vascular Risk in Diabetes? Pierce
206A	NCAR Section FT NCAR Trainee Featured Topic Xia/Moraes	CV Section Symp New Insights into Vascular Function from In Vivo Vascular Imaging Wier/Zhang	E&M Section Symp The Yin/Yang of Estrogen Signaling in the Control of Energy Homeostasis Kelly
206B	EEP Section FT New Insights into the Physiology and Pathophysiology of Diving and Hyperbaric Environments Florian	CAMP Section and AJP:Cell Symp Morphogen Signaling Pathways in Tissue Patterning and Disease Processes Adams/Yuan	CNS Section Symp Gliotransmission and Behavior Parpura
207	PG Group Symp Revolutionary Systems – Medicine Approaches to Understand Disease and Drug Response Physiology Blackman	AFMR Symp Omics of Brain Injury Lo	PG Group FT Physiologic Effects of Sex Chromosome Complementation and Chromosome Y Genetic Variants Deschepper
208	CV Section Symp Cardiopulmonary Consequences of Perinatal Exposures Rogers/Velten	CNS Section FT Spinal Plasticity Dale	CV Section Symp The Role of Store-Operated Calcium Entry in Cardiovascular Physiology and Disease Lompré/Collins
209	Hypoxia Group Symp Molecular Oxygen: At the Crossroads of Inflammation and Metabolism Haase	Biomedical Engineering Society Symp Stem Cells for Tissue Regeneration TBD	NCAR Section Featured Topic Baroreflex and Chemoreflex Controls of the Human Cerebral Circulation Rickards
210A	CV Section Symp Ion Channels in Health and Disease Jaggar/Navedo	10:30 AM-11:30 AM Teaching Section Bernard Lecture Macknight	3:15-4:15 PM WEH Section Starling Lecture Reckelhoff 4:15 PM-5:15 PM WEH Section New Investigator Award Lecture

Sunday, March 29, 2015, *cont.*

210B	<i>TPIG Oral</i> Highlights in Translational Physiology	<i>Cross Sectional Symp</i> The Host-Microbe Interface and Control of Barrier Function: The Path from Pathology to Therapy McCormick	<i>President's Symp Series</i> <i>Physiology: Answers to Big Questions Symp</i> The Future of Diabetes Research Hall 5:45 PM-6:45 PM Bowditch Award Lecture LaMarca
211	<i>Teach Section FT</i> Innovations in Classroom Teaching Miller/Golden	<i>Renal Section FT</i> Immune Cells, The Kidney and Hypertension Sullivan/Bell	<i>Renal Section Symp</i> Control of Electrolyte Balance by Novel Pathways in Intercalated Cells Sansom/Grimm
212	<i>WEH Section FT</i> The Heart of the Matter: Menopause, Sex Steroids, and Cardiovascular Disease Reckelhoff/Wenner	<i>Resp Section Symp</i> Mechanochemical Background in the Intact Lung and the Role of Contextual Cell Biology for the Study of Lung Injury and Repair Kuebler/Waters	<i>Resp Section FT</i> Respiratory Related Disorders in Aging and Neurodegeneration Greising/Sieck

Monday, March 30, 2015

Room	8:00-10:00 AM	10:30 AM-12:30 PM	3:15-5:15 PM
205B	<i>CV Section Symp</i> The Physical Biology of Thrombus Formation McCarty/Neeves	<i>CV Section and the MCS FT</i> Kaley Lecture and Complementary Presentations Nelson	3:15 PM-4:15 PM Renal Section Gottschalk Lecture Ellison 5:30 PM-6:30 PM <i>APS and The Physiological Society (UK) Conversation</i> A Conversation with Denis Noble and Michael Joyner on the Integration of Evolutionary Biology with Physiological Science Paterson
205C	<i>Publications Comm Symp</i> Publishing 101: How to Get Your Work Published and Avoid Ethical Minefields Sigmund/Scheman	<i>CNS Section Symp</i> Brainstem Mechanisms Underlying Cardiorespiratory Signaling: From Synapses to Circuits Derbenev/Andresen	3:15-4:15 PM CNS Section Erlanger Lecture Deisseroth 4:15 PM-5:15 PM <i>CNS Section Minisymposium</i>
206A	<i>EEP Section Symp</i> Limitations to and Potential of Exercise for the Spinal Cord Injured Taylor	<i>Resp Section FT</i> Lung Epithelium and Endothelium: Injury, Repair, and Remodeling Zhao/Petrache	<i>AFMR Symp</i> Protecting and Restoring Functional Beta Cell Mass in Type 1 Diabetes: Research from Bench to Bedside Evans-Molina/Fueger
206B	<i>Renal Section FT</i> Recent Advances in Renal Physiology and Kidney Disease I Welling	<i>WEH Section FT</i> Hypertension: Developing Concepts Sullivan/Ryan	<i>CAMP Section Symp</i> Organoids and Physiology – The Way of the Future and Beyond? Hamilton

Monday, March 30, 2015, *cont.*

207	<i>WEH Section Symp</i> Gastro-Renal Communication Jose	<i>Hypoxia Group FT</i> Cellular, Molecular and Systems Integration Underlying Adaptation and Maladaptation to Hypoxia Ramirez	<i>WEH Section FT</i> Water, Electrolyte, and Blood Pressure Homeostasis: Neural and Humoral Regulators and Stressors Gao/Veelken
208	<i>CV Section FT</i> Cellular Membrane Repair in Cardiovascular Physiology and Pathophysiology Weisleder	<i>GI&L Section FT</i> Targeting Colonic Contents for Treatment of Disease Keely	<i>CV Section Symp</i> Treating Cardiovascular Disease with Exercise: Mechanistic Insight Translated from Animal Models Emter/Libonati
209	<i>CAMP Section FT</i> Ion Channels and Transporters in Health and Disease O'Grady/Sarathy	<i>NCAR Section FT</i> Hypothalamic Autonomic Control of Metabolism Jiang/Zsombok	
210A	8:00 AM-9:00 AM NCAR Section Ludwig Lecture Mitchell 9:00 AM-10:00 AM NCAR Section Minisym Neural Control of the Circulation during Exercise in Normal and Disease States Mitchell	10:30 AM-11:30 AM E&M Section Berson Lecture Hardie	2:00 PM-3:00 PM EEP Section Adolph Lecture Laughlin 3:15 PM-5:15 PM Resp Section Symp Pathogenesis of Sudden Infant Death Syndrome: Is it Just a Breathing Disorder? Nattie/Paterson
210B	<i>TAC FT</i> Recent Advances in Obesity Research Haack	<i>Cross Sectional Symp</i> Neurohormonal Mechanisms in Blood Pressure Control Haywood/Cowley	<i>President's Symp Series</i> <i>Physiology: Answers to Big</i> <i>Questions Symp</i> The Future of Obesity Research Hasty
211	<i>Teach Section Symp</i> Resources and Experiences in Developing Flipped Classrooms for Graduate and Medical Physiology Wilson	<i>Renal Section Symp</i> Novel Mechanisms in Renal Function and Blood Pressure Regulation Weisz/Caceres	<i>Teach Section Symp</i> What's Your Major? The Rise of the Undergraduate Physiology Degree Halliwill/Wehrwein
212	<i>Resp Section FT</i> Cellular Quality Control in the Lung: Role of ERAD, Autophagy, and Mitophagy in Health and Disease Beers/Priolo	<i>CEPS Section FT</i> CEPS Abstract-driven Trainee Featured Topic Pamenter/Kirkton	<i>CEPS Section Symp</i> Comparative Biology of Mitochondria: From Physiology to Molecules and Back Jastroch/Perocchi

Tuesday, March 31, 2015

Room	8:00 AM-10:00 AM	10:30 AM-12:30 PM	3:15 PM-5:15 PM
205B	<i>GI&L Section FT</i>	<i>CV Section FT</i> Wiggers Award Featured Topic Spaan/Chilian	<i>Renal Section Symp</i> Emerging Role of AMPK in Kidney Epithelial Transport, Metabolism and Disease Singh/McDonough
205C	<i>Physoc, JPhys, and APS Symp</i> Voltage-gated Calcium Channels and the Function of Excitable Cells: From Basic Mechanisms to Disease Dolphin	<i>E&M Section Symp</i> Rejuvenating the Beta Cell Lynch/Samson	<i>GI&L Section Davenport Lecture</i> Lund
206A	<i>E&M Section FT</i> Diet, Nutrition, and Adipose Tissue: You are What You Eat Yosten	<i>NCAR Section FT</i> Interactions Between Cardiovascular and Ingestive Behavioral Signals at the Circumventricular Organs Collister/Ferguson	<i>MBG Symp</i> Membrane Repair in Muscle Cells: Molecular Mechanisms and Therapeutic Approaches Michele/Metzger
206B	<i>CNS Section FT</i> CNS Mechanisms of Blood Pressure Regulation Wainford	<i>WEH Section FT</i> The Biology of Oxygen Homeostasis and Hypoxia Polichnowski/Evans	<i>CAMP Section Symp</i> Cation Channels Controlling Intracellular Functions Kozak/Brueggemann
207	<i>Careers in Physiology Committee Symp</i> Resilience is Power: Dealing with the Ups and Downs of Your Scientific Career Wehrwein/Schnackenberg	<i>EEP Section FT</i> Sex Hormone Effects on Autonomic and Endothelial Function Stachenfeld	<i>EEP Section Symp</i> Muscle Atrophy, Hypertrophy and MicroRNA Carson/Wang
208	<i>ETG FT</i> Epithelial Transport: Cell Biology Møller/Blount	<i>ETG FT</i> Epithelial Transport: Pathophysiology Bomberger/Rieg	<i>ETG Symp</i> Autophagy: Importance in Health, Epithelial Transport and Aging Levi/Klein
209	<i>CAMP Section FT</i> Cell Signaling: Proteins, Pathways and Mechanisms Jones/Bradbury	<i>CAMP Section FT</i> Oxidative Stress: Mechanisms and Responses White	<i>E&M Section Symp</i> Inflammation in Obesity is not All Bad Ye/McGuinness
210A	<i>NCAR Section Symp</i> Autonomic Denervation in Cardiovascular Disease: Mechanisms and Therapeutic Potential Zucker/Paton	10:30 AM-11:30 AM <i>Resp Section Comroe Lecture</i> Raj	2:00 PM-3:00 PM <i>CV Section Berne Lecture</i> Ping 3:15 PM-4:15 PM <i>CEPS Section</i> Krogh Lecture Sponsored by Novo Nordisk Fndn. DeVries

Tuesday, March 31, 2015, *cont.*

210B		Cross Sectional Symp "Omics" and Epithelial Systems Physiology Delpire/McCormick	3:15 PM-5:15 PM <i>President's Symp Series</i> <i>Physiology: Answers to Big Questions Symp</i> The Future of Hypertension Research Jacob 5:45 PM-7:00 PM APS Business Meeting
211	<i>Renal Section FT</i> Recent Advances in Renal Physiology and Kidney Disease II Ortiz	<i>GI&L Section FT</i> Inflammatory Responses in Gastrointestinal and Liver Cancer: Current Insights into Mechanism and Treatment Rao	<i>PG Group</i> Trainee Highlights in Physiological Genomics
212	<i>EEP Section Symp</i> Exercise at the Molecular Level: Myokines and Other Novel Therapeutic Opportunities White	<i>CEPS Section FT</i> The Cost of Physiological Plasticity Hindle/van Breukelen	<i>Resp Section Symp</i> The Brain on Intermittent Hypoxia Watters/Dougherty
Westin Hotel, Room TBD			1:00 PM-2:00 PM <i>History Group Lecture</i> Ryan

Wednesday, April 1, 2015

Room	8:00 AM-10:00 AM	10:30 AM-12:30 PM	2:30 PM-4:30 PM
205B	<i>Women in Physiology Committee Symp</i> Mentoring for Diverse Careers: Mentor and Protégé Perspectives Dwinell/Mathis	<i>NCAR Section FT</i> Refreshing Perspectives on the Role of the Chemoreflexes in the Control of Cardiorespiratory Functions: New Pathways and Players Marcus	
205C	<i>Pan-American Physiological Societies Symp</i> Salt-Sensitive Hypertension: The Brain or the Kidney to Blame Antunes/Reyes		
206A	<i>TPIG Symp</i> Translational Strategies for Musculoskeletal Regenerative Medicine Mendias/Funai	<i>TAC Symp</i> Scientists as Supervisors: Hiring, Firing, and Beyond Banek	
206B	<i>MBG FT</i> Neuromuscular Diseases: Novel Therapeutic Strategies Ljubicic	<i>CV Section FT</i> Molecular Regulators of Skeletal Muscle Angiogenesis in Health and Disease Haas/Gustafsson	<i>CV Section FT</i> Systemic and Pulmonary Vascular Function at High Altitude/Hypoxia: Learning from Maladaptations Scherrer

Wednesday, April 1, 2015, cont.

207	<i>EEP Section Symp</i> Who can Tolerate Blood Loss? New Insights into Mechanisms of Compensation to Hemorrhage Convertino/Hinojosa-Laborde	<i>EEP Section FT</i> Sympathetic Control of the Vasculature in Clinical Populations Alexander/Greaney	<i>EEP Section FT</i> Autophagy in Muscle Hood
208	<i>GI&L Section Symp</i> Wanted and Unwanted Paracellular Passage in Intestinal Epithelia Fromm/Schulzke	<i>Resp Section Symp</i> Neonatal Lung Development and Adult Lung Homeostasis: Common Molecular Mechanisms in Lung Disease Nelin/Abman	<i>CV Section FT</i> Autophagy and miRNA in Diabetic Heart Failure Mishra
209	<i>E&M Section FT</i> Circadian Clock in Metabolic Regulation Wang	<i>MBG FT</i> Muscle Diseases: Recent Advances in Disease Mechanisms Beedle/Selsby	
210A	<i>Resp Section and CEPS Section FT</i> Evolution of Air Breathing Harris/Wilson	<i>History Group Symp</i> Neuroplasticity in Space: Reflections from the STS-90 Neurolab Space Shuttle Crew Dean	
210B		<i>Cross Sectional Symp</i> Contributors to the Slowed Aging Phenotype: Exercise and other Common Mediators Hamilton	4:45 PM-5:45 PM <i>President's Symp Series</i> <i>Physiology: Answers to Big</i> <i>Questions Lecture</i> APS Nobel Prize in Physiology or Medicine Lecture Lefkowitz
211	<i>CV Section Symp</i> The Pathophysiology of Drug- induced Vascular Injury (DIVI) Wamhoff/Turk	<i>GI&L Section Symp</i> Enteric Nervous System Regulation of GI Function Mittal	<i>AFMR Symp</i> Emerging Concepts in Hypoxia- related Disease Signaling Rasouli/Colgan
212	<i>AFMR Symp</i> From Basic Science to Precision Medicine: The Use of Genomic, Epigenomic and Translational Research to Develop Personalized Treatments Payne/Colburn	<i>CEPS Section Symp</i> Effects of a Changing Climate on Insect Physiology Greenlee/Harrison	<i>Biomedical Engineering Society</i> <i>Symp</i> Vascular Bioengineering Dai/Tien

Comparative Meeting

The 2014 APS Intersociety Meeting: Comparative Approaches to Grand Challenges in Physiology

San Diego, California, October 5-8, 2014

The 2014 APS Intersociety Meeting: Comparative Approaches to Grand Challenges in Physiology was held at the iconic Town & Country Resort and Convention Center located in sunny San Diego, California. The well attended meeting was sponsored in part by five guest societies including The Society for Integrative and Comparative Biology (SICB), Society for Experimental Biology (SEB), the Australian & New Zealand Society for Comparative Physiology and Biochemistry (ANZCPB), Canadian Society of Zoologists (CSZ), and the Crustacean Society.

The meeting program had a packed schedule of dynamic plenary lectures, concurrent symposia, workshops, interactive poster sessions, and networking opportunities, which made the meeting a valuable experience for all attendees, was organized by Chair Bernard Rees, University of New Orleans, along with an organizing committee that was equally dedicated to putting on a great scientific program and providing opportunities for young investigators. The committee members included Siribhinya Benyajati, University of Oklahoma Health Science Center; Heidy Contreras, University of La Verne; Peter Frappell, University of Tasmania, Australia; Greg Goss, University of Alberta, Canada; Jon Harrison, Arizona State University; Scott Kirkton, Union College; Mathew McHenry, University of California, Irvine; Holly Shiels, University of Manchester, United Kingdom; Jonathon Stillman, University of California, Berkeley; Tobias Wang, Aarhus University, Denmark; and Cassandra Williams, University of California, Irvine.

The meeting was attended by 411 total registrants, of whom 67 (17%) identified themselves as APS members, 22 (5%) registered as members of one of the participating sponsoring societies, 28 (7%) registered as nonmembers, 44 (11%) registered as postdoctoral fellows, followed by a large contingent of young investigators (145 attendees or 35%). Rounding out the attendance at 98 (24%) were invited chairs and speakers, along with exhibitors at 1%. Table 1 shows the breakdown of the different registration types. This meeting mainly

attracted comparative scientists and students from the United States; however, of the 411 registrants, 134 (33%) represented many countries from around the world. Table 2 depicts the breakdown of the different regions that attendees came from.

Table 1. Registration Statistics

Registration Type	Number of Attendees (%)
APS Member	71 (17%)
Guest Society Member	22 (5%)
Nonmember	28 (7%)
Postdoctoral Fellows	44 (11%)
Students	145 (35%)
Invited Chairs/Speakers	98 (24%)
Exhibitors	3 (1%)
Total	411 (100%)

Table 2. Registration Geographic Region Statistics

Region	Number of Attendees (%)
USA	277 (67%)
Canada	58 (14%)
Europe	40 (10%)
Australia/New Zealand	12 (3%)
Central and South America	9 (2%)
Asia	6 (2%)
Africa	5 (1%)
Middle East	4 (1%)
Total	411 (100%)

The meeting program consisted of 2 plenary lectures, 15 concurrent symposia sessions, 2 workshops, numerous oral presentations sessions based on the abstracts that had been submitted for the meeting, and the exciting Scholander Competition Finals, where 9 attendees participated. The audience was encouraged to share their ideas and thoughts with the speakers at the end of

their talks. The meeting also had several social activities including a Welcome and Opening Reception, which was designed to give attendees a chance to meet with long-time colleagues, create new friendships, and enjoy some desserts and beverages after the opening plenary lecture. There were three afternoon poster sessions, which included the Best Poster Competition, where scientists presented their work and discussed their comparative work with other attendees, as well as a closing banquet and award ceremony at the conclusion of the meeting.

A total of 340 abstracts were submitted for the meeting, with 283 of these abstracts programmed as either a poster or oral presentation. The remaining 57 abstracts were submitted by invited speakers. Of the abstracts submitted for the meeting, 146 (43%) were submitted by a female first author compared with 194 (57%) submitted by male first authors.

At the meeting, Janet Gonzalez-Rosario, University of New Orleans; Felix Jimenez, Brigham Young University; Alexis Jones, Oklahoma State University Center for Health Sciences; Bridget Martinez, University of California, Merced; and Karl Rodriguez, University of Texas Health Science Center at San Antonio, were the recipients of the American Physiological Society Minority Travel Fellowship Award, which is provided to encourage participation of underrepresented minority individuals in the physiological sciences. The fellowship provides reimbursement of all expenses associated with travel and participation in the workshop. Moreover, each recipient is paired with an APS member who serves as his/her mentor during the meeting. Thank you to Alice Villalobos, Texas A&M University; Colleen Farmer, University of Utah; Theodore Garland, Jr., University of California, Riverside; Shane Kanatous, Colorado State University; and Hannah Carey, University of Wisconsin, Madison.

The grand finale of the successful meeting was during the Closing Banquet and Awards Ceremony. During the banquet, Rees thanked the attendees and all of the individuals involved in the various parts of planning the meeting. Excitement mounted among the attendees as the 26 APS Abstract-based Travel Awardee's were congratulated by Rees (*Figure 1*). Next, Scott Kirkton, Organizer for the Best Poster Competition, announced the winners of both the undergraduate and the graduate categories. Sherry Du, McMaster University, Canada, won first place for her poster presentation, followed by second place Jennifer Jung, University of North Texas (*Figure 2*). In the graduate category of the competition, Bryan Helm, North Dakota State University, claimed first place, followed by Christopher Hardy, University of Nevada, Las Vegas, and Anders Findsen, Aarhus University, Denmark, for second and third place, respectfully. Finally, the time had come to announce the winner of the 2014 Comparative and Evolutionary Physiology Section (CEPS) prestigious Scholander Award. Award Organizer Sinya Benyajati praised the excellent work presented by the nine competitors during the Scholander Award Finals (*Figure 3*). However, after deliberation with the Scholander judges, Katie Barott, Scripps Institute on Oceanography, La Jolla, was awarded first place, followed by Heath MacMillan, Aarhus University, Denmark, with second place and Joshua Pemberton, University of Alberta, Canada, with third place (*Figure 4*).



Figure 1. Meeting Organizer Bernard Rees (*left*) congratulates the APS Abstract-based Travel Award winners. Each winner receives a certificate and travel reimbursement stipend.

The American Physiological Society and the Organizing Committee gratefully acknowledge the financial support provided through generous educational grants from the National Science Foundation, The Society for Integrative and Comparative Biology, Society for Experimental Biology, Comparative Biochemistry and Physiology Journal, the Australian & New Zealand Society for Comparative Physiology and Biochemistry, International Society for Neuroethology, Loligo Systems, Canadian Society of Zoologists, The Crustacean Society, and the Exercise Medicine and Sports Sciences Initiative at University of California, Irvine. ●



Figure 3. Scholander Award Organizer Sinya Benyajati (*third from left*) with the nine Scholander Award Finalists. The finalists are (*in alphabetical order*): Katie Barott, Anne Dalziel, Yusuke Kumai, Heath MacMillan, Milica Mandic, Katie Marshall, Joshua Pemberton, Cosima Porteus, and Matthew Regan.

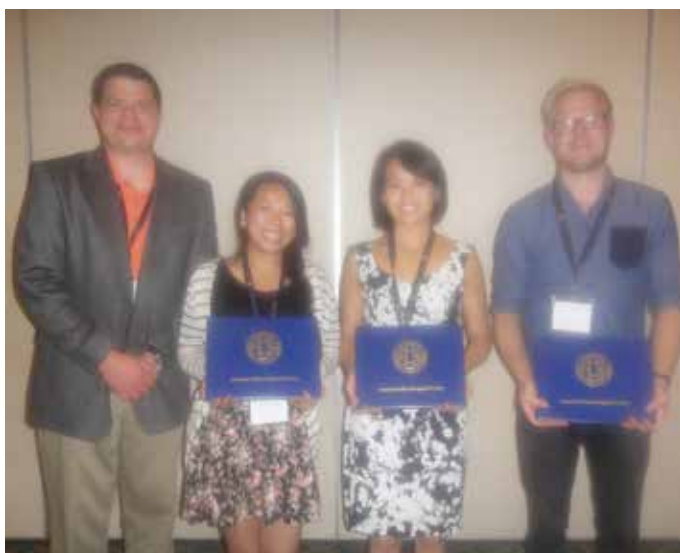


Figure 2. Best Poster Competition Organizer, Scott Kirkton (*left*) congratulates Sherry Du (*middle*) for first place and Jennifer Jung (*second from left*) for second place in the undergraduate category, and Anders Findsen (*right*) for third place in the graduate category. (First and second place winners Bryan Helm and Christopher Hardy are not pictured.)



Figure 4. Meeting Organizer Barney Rees (*right*) and Sinya Benyajati (*left*) congratulate Katie Barott (*middle*) for winning the prestigious 2014 CEPS Scholander Award.

Science Policy

APS Updates Guiding Principles

On October 15, 2014, the Council approved an update to the APS Guiding Principles for the Care and Use of Vertebrate Animals in Research and Training. The updated version is available on the APS website at <http://www.the-aps.org/Guiding-Principles>.

The Guiding Principles serve as an ethical code for APS members and authors. The Animal Care and Experimentation Committee recommended changes to ensure that the Guiding Principles are equally applicable to researchers living in countries other than the U.S. and to expand on or clarify certain passages.

Previously, the Guiding Principles referred to the U.S. Government Principles for the Utilization and Care of Vertebrate Animals Used in Testing, Research, and Training to underscore the importance of both animal welfare and the knowledge gained from animal studies. However, given the increasing number of APS members and authors who work outside the U.S., the ACE Committee recommended replacing this reference with a comparable statement from an international body. The revised Guiding Principles now includes the following excerpt from the International Guiding Principles for Biomedical Research Involving Animals of the Council for International Organizations of Medical Sciences (CIOMS):

The advancement of scientific knowledge is important for improvement of human and animal health and welfare, conservation of the environment, and the good of society. Animals play a vital role in these scientific activities and good animal welfare is integral to achieving scientific and educational goals.

In addition, the statement that research must comply with “federal, state, and local laws and regulations” was broadened to require compliance with “national and local laws and regulations.”

Optimizing study design to promote both scientific rigor and animal welfare was another topic of concern. The revised Guiding Principles addresses this by urging

investigators to “carefully consider the three principles of reduction, refinement, and replacement (3Rs) in the study design, focusing on how the scientific or educational goals can be accomplished with the least animal morbidity and mortality.”

The final change was to provide context about what is considered appropriate husbandry. This was accomplished by adding a reference to the research setting to the following statement: “Animals used in research and education must be housed, fed, and maintained in an appropriate setting for their species, condition, and the research to be conducted.” ●



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The APS Office of Science Policy is on Twitter with the handle **@SciPolAPS**. Follow us for news about research funding, animal research, and science advocacy. Also, check out the hashtag **#HillDayAPS** to learn about APS members who visit Capitol Hill.



APS Advises NIH on Sex as a Biological Variable

On September 11, 2014, the National Institutes of Health (NIH) published a request for information (RFI) seeking input on consideration of sex as a biological variable in preclinical research. The RFI posed six questions. These questions and edited excerpts of the APS responses are below. To read the full APS response, see <http://www.the-aps.org/mm/SciencePolicy/About/Comments-Letters/Sex-as-a-Biological-Variable.html>.

Is consideration of sex as a biological variable an issue that affects the reproducibility, rigor, and/or generalizability of research findings?

Sex as a biological variable is an issue of particular concern with respect to generalizability of research findings, and it should be considered wherever appropriate and feasible. An important first step would be to require researchers to report the sex of all animal subjects and of the organisms from which all biological materials used in experiments were derived.

Reproducibility of findings in animal and cell studies is a related issue that depends in part on genetic and epigenetic differences between animals and cell lines. However, sex is just one of several relevant factors that affect reproducibility.

What are the areas of science or phases of research conducted with animals that have the greatest opportunity or need for considering sex as a biological variable?

Translational and clinical research with immediate implications for human medicine should include consideration of sex differences unless there is a sound reason not to do so. Analysis of sex differences is particularly important in proof-of-concept studies such as translational studies in animal models.

Research into conditions and diseases that affect only one sex should be exempted from requirements to consider sex differences. Examples include pregnancy, menopause, cervical cancer, and prostate cancer.

Basic research exploring fundamental biological principles without a direct translational or clinical relevance may not always need to consider sex as a

biological variable, and policies should be put in place to allow exemptions when determined to be appropriate.

What are the areas of science or phases of research conducted with cells and/or tissues that have the greatest opportunity or need for considering sex as a biological variable?

Some research performed at the molecular and cellular level may be less likely to be affected by sex differences unless there is a chromosomal effect. However, when research on cellular and molecular mechanisms has a strong and direct relevance to more complex systems at the organ and whole organism level, the inclusion of material derived from both sexes may facilitate consideration of sex as a variable in later studies.

What are the main impediments (e.g., scientific, technical, and other) to considering sex as a biological variable in research?

Since many sex-based differences are due to differences in sex steroid levels, for some studies it may be necessary to study female animals at either one specific phase of the estrous cycle or at various phases of the cycle. When females in various stages of the estrous cycle must be studied, the number of subjects will have to be increased substantially. In some cases, this could be addressed by spaying the animals to eliminate hormonal fluctuations.

Additional resources will be needed to increase the number of animals or cell lines in an existing study because of the need to cover the costs of acquiring and maintaining the additional animals and materials, along with the cost of conducting additional procedures, assays, etc.

Researchers will need time to develop new experimental designs that accommodate analysis of sex differences, and they will also need additional time to complete and analyze the additional studies.

How can NIH facilitate the consideration of sex as a biological variable in NIH-supported research?

NIH and members of the scientific community, including professional societies, should undertake a broad campaign of education outlining why consideration

of sex as a biological variable is important and how to begin to incorporate it. This campaign should be targeted to a wide audience including students, faculty, and reviewers.

As a first step, the NIH should begin asking grant applicants to provide information about the sex of all animal subjects and of the organisms from which all biological materials used in experiments were derived. Determination of whether additional experiments will be required to address sex differences should be considered subsequently.

NIH should provide guidelines for when using both sexes is required and when it is not.

Some investigators may need training in how to implement experimental protocols using both sexes. Related education should include how to do the best power analysis to make sure the investigator has the correct study design and how to craft a justification statement when exemptions are sought. These areas of education would best be targeted to faculty writing the grants.

In addition, NIH reviewers and grant-management staff will need training in experimental design that includes both sexes. They will also need clear guidelines for evaluating justification statements when exemptions are sought and for determining when exemptions are appropriate. Policies should also be developed for reviewing progress reports of funded grants to determine whether both sexes are being used as proposed in the original application.

Modifying experimental design to include consideration of sex as a biological variable will add cost to research

budgets. Providing additional resources to investigators who are increasing the scope of their proposed research is essential. NIH should also consider additional incentive such as RFAs or supplements to understand the implications of previously published findings in animals or specimens of the other sex.

Given the practical and financial implications of incorporating sex as a biological variable, this policy should be phased in, and priority should be given to areas such as translational research. NIH should allow investigators adequate time to modify their experimental designs to optimize the value of previous work.

Are there any additional comments you would like to offer to NIH about the development of policies for considering sex as a biological variable in research involving animals, tissues, or cells?

In addition to sex, other biological variables may be important to consider, including:

- age of the animals being studied
- genomic variation
- microbiome variation between colonies of animals
- differences related to the time of day that animals are handled and disruption of circadian rhythms
- differences based on the type and brand of feed animals receive.

Because resources are not available to explore every possible variable, it will be necessary to consider where the best value lies for each particular research study. For each of the variables listed above, and for sex, detailed factual reporting is a critical first step toward understanding the role that each variable plays in determining experimental outcomes. ●

Education

APS Promotes Physiology to Biology Educators at National Convention

The APS once again highlighted physiology for K-12 biology teachers at the National Association of Biology Teachers (NABT) 2014 Conference in Cleveland, OH. The annual national conference, held the first week of November, attracts middle and high school teachers as well as 2- and 4-year college faculty from across the nation. APS sponsored an exhibit booth and featured speaker, and presented a hands-on workshop that highlighted resources from the *LifeSciTRC*.

This year's sponsored speaker was APS member Merry Lindsey, Director, Mississippi Center for Heart Research, Director, San Antonio Cardiovascular Proteomics Center, and Professor Department of Physiology and Biophysics University of Mississippi Medical Center. Lindsey delivered her talk titled, "Cardiac Wound Healing After a Heart Attack" to an audience of 60+ educators interested in learning more about the latest research. In her 75-minute presentation, Lindsey explained that, following myocardial infarction (MI), the left ventricle (LV) responds by undergoing a series of changes that involve wall thinning, dilation, and infarct expansion; inflammation and necrotic myocyte resorption; and fibroblast accumulation and scar formation. Collectively, these events are referred to as LV remodeling. Lindsey went on to explain that LV remodeling is initially a compensatory response, the transition to adverse remodeling frequently culminates in the development of congestive heart failure (CHF), and CHF is a significant contributor to high cardiovascular morbidity and mortality rates for the MI patient. Her talk further defined LV remodeling, with particular emphasis on the inflammatory cell (macrophage)- and enzyme (matrix metalloproteinase)-dependent mechanisms that stimulate the extracellular matrix wound-healing process. Throughout her talk, Lindsey inserted videos of members of her lab reminiscing about how they first became interested in science. Many of them remembered teachers who impacted their career decisions. This was of great interest to the teachers in attendance, who mentioned it numerous times after the completion of the session.



Merry Lindsey was the sponsored speaker at this year's NABT meeting in Cleveland, OH

In a hands-on workshop led by Miranda Byse, Program Manager, Professional Skills Training & Life Science Teaching Resource Community, and Margaret Stieben, Program Manager, K-12 Education Programs, teachers were given a brief introduction to the *Next Generation Science Standards*. The newly released standards create a challenge for K-12 teachers and the way science will be taught in their classrooms. During this well attended workshop, sample resources and ideas for using the *Next Generation Science Standards* were presented, giving teachers the opportunity to actually try some of the hands-on engineering and modeling lab lessons that are currently available in the *LifeSciTRC*.

Interest in undergraduate programs was a highlight at the exhibit booth throughout the 3-day conference as resources and program materials were provided and discussed with community college attendees. Next year's conference will be held in Providence, RI. For further information, please contact Margaret Stieben, Program Manager, K-12 Education Programs (mshain@the-aps.org). ●

APS Promotes Physiology to Mid-Level Educators at National Convention

The APS highlighted physiology for middle school science teachers and administrators at the annual Association for Middle Level Education (AMLE) Conference held in Nashville, TN, November 5-8. This was the fourth year for an APS presence at the AMLE Conference, which is attended by over 4,000 teachers, administrators, and counselors from across the country. Teachers were as excited as ever for a science society's presence since so few opportunities are available for science-related materials at this meeting. The APS booth was extremely busy and well received, with many questions about the Life Science Teaching Resource Community website, careers materials including high interest in the newly released APS Career Trading Cards, and the Frontiers in Physiology Program. Promotion of the new Six Star Science Online Professional Development Fellowship was the main focus in this year's booth, with the application window opening on November 1.

Next year's conference will be held in Columbus, OH, October 15-18. For more information, please contact Margaret Stieben, Program Manager, K-12 Education Programs (mshain@the-aps.org). ●



Teachers took part in a hands-on workshop on the sense of touch starting with a two-point discrimination test



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LifeSciTRC.org

APS Hosts the Second Annual NSF IOS BP PI Meeting

As part of the three Integrative Organismal Systems (IOS) Broadening Participation (BP) awards to the APS, the Council on Undergraduate Research (CUR), and the Society for Developmental Biology (SDB), this year was our second PI meeting and was hosted by APS and SDB at the FASEB Headquarters.

The IOS BP PI meeting kicked off with an informal dinner on Sunday, September 14. On Monday, September 15, 2014, the group convened on the FASEB campus. In attendance were NSF staff; representatives from APS, CUR, and SDB; as well as staff from American Society for Nutrition, American Society of Toxicology, American Society of Microbiology, American Society for Pharmacology and Experimental Therapeutics, Genetics Society of America, The Leadership Alliance, and the Society for Conservation Biology.

To gain insight into the target groups for these programs, two speakers were invited to discuss both specific needs for each group and best practices for programs to increase participation in STEM. Clifton Poodry, Science Education Fellow, HHMI, and former Director of NIGMS Diversity Programs, presented "Who Cares, So What and What Were We Thinking?" discussing best practices in promoting STEM participation among underrepresented

minority groups. Mark Leddy, Program Director, Division of Human Resource Development, Directorate for Education and Human Resources, NSF, then discussed "Broadening the Participation of People with Disabilities in STEM." Both talks were recorded and will be available in the Life Sciences Teaching Resource Community (<http://www.lifescitrc.org/collection.cfm?collectionID=3261>).

Time was made available for networking among the societies, speakers, and NSF staff. In addition, an update on potential funding opportunities was presented by NSF.

Graciela Unguez, Professor, Department of Biology, New Mexico State University, provided an update on the SDB "Choose Development!" program activities; Mary Crowe, Associate Provost of Experiential Education, Florida Southern University, provided an update on the CUR BP Institute and other activities; and Brooke Bruthers, APS Senior Program Manager, Diversity Programs, provided an update on APS IOSP program.

The group will meet again in fall of 2015. SDB will host this meeting. For more information, please view the IOSP website (www.the-aps.org/iosp) and/or contact Brooke Bruthers (education@the-aps.org). ●

Awards, Grants, and Fellowships of the APS

- ✓ Student/Trainee Awards
- ✓ Section Awards
- ✓ Society Awards
- ✓ Teacher Awards

For more information, please visit
the-aps.org/awards



APS at SACNAS 2014

The APS was an exhibitor at the 2014 Society for the Advancement of Chicanos and Native Americans in Science (SACNAS) annual meeting at the LA Convention Center in Los Angeles, CA from October 16 to 18. The theme this year was “Creativity, Vision & Drive: Toward Full Representation in STEM.”

The APS, represented by William Johnson, University of South Florida and Porter Physiology Development and Minority Affairs Committee Member, and Brooke Bruthers, Senior Program Manager, Diversity Programs, staffed the exhibit booth, located in the FASEB Row. Rudy Ortiz, University of California-Merced and APS Councilor, also attended the conference and spoke at the APS booth with students interested in physiology.

The SACNAS National Conference is designed to motivate, inspire, and engage participants to achieve their highest goals in pursuing education and careers in all disciplines of science, technology, engineering, and mathematics from across the country. Conference programming is specifically tailored to support undergraduate and graduate students, postdoctoral researchers, and career professionals at each transition stage of their career as they move toward positions of science leadership. The conference showcases cutting-edge science and features mentoring and training sessions for students and scientists at all levels. Over 3,800 attended the conference, and 300 exhibits shared training, research, grad school,

and job opportunities. For more information about the SACNAS National Conference, visit www.sacnas.org. For more information on APS diversity programs, visit www.the-aps.org/diversity. ●



SACNAS exhibit booth

Mark Your Calendars for Professional Development Symposia at Experimental Biology 2015!

It's All in Your Head – A Refresher Course on the Brain and Systems Control (Medical Education Refresher Course)

Get an update on content from leading experts in the field: “The Brain and the Cardiovascular System” (Roger Dampney, Univ. of Sidney), “The Brain and the Immune System” (Francois Abboud, Univ. of Iowa), “The Brain and the Respiratory System” (Gordon Mitchell, Univ. of Wisconsin-Madison), and “The Brain and the Gut” (Michael Gershon, Columbia Univ.).

Saturday, March 28, 8:00 AM to 12:00 PM
Boston Convention Center, Rm. 210A

Resilience is Power: Dealing with the Ups and Downs of Your Scientific Career (Career Symposium)

A career in science is filled with challenges that can often have nothing or little to do with the research itself. Hear from experts in the psychology of resilience as well as scientists who can offer advice on adaptability and resilience specific to the discipline.

the-aps.org/resilience

Tuesday, March 31, 8:00 AM to 10:00 AM
Boston Convention Center, Rm. 207

Mentoring for Diverse Careers: Mentor and Protégé Perspectives (Mentoring Symposium)

Get information on how to approach your mentor or advisor about taking “alternative” career paths and how to handle these situations if a mentee approaches you for advice on a non-traditional career path.

the-aps.org/mentoringdiversecareers

Wednesday, April 1, 8:00 AM to 10:00 AM
Boston Convention Center, Rm. 205B

Scientists as Supervisors: Hiring, Firing, and Beyond (Trainee Symposium)

Supervisory and management skills are essential for scientists’ success both inside and outside of research. Learn about 1) hiring and firing (finding the right employees), 2) resource and personnel management, and 3) conflict resolution.

the-aps.org/supervisor

Wednesday, April 1, 10:30 AM to 12:30 PM
Boston Convention Center, Rm. 206A ●



APS Professional Skills Training 2015 Course Offerings www.the-aps.org/PST

Manuscript Writing Skills

Writing and Reviewing for Scientific Journals
January 15-18 (Orlando, FL)

Work with leading experts in-person to improve your first-author draft manuscript while learning the essentials of scientific writing and reviewing.

Writing and Reviewing for Scientific Journals
July 6–August 21 (Online)

Work with leading experts online to improve your first-author draft manuscript while learning the essentials of scientific writing and reviewing.

Interviewing Skills

Interviewing for an Academic Position
May 7-17 (Online)

Work with experienced faculty on how to start a job search, prepare a curriculum vitae and research statement, have a successful interview, and present an engaging job talk.

Interviewing for an Industry Position
September 10-20 (Online)

Work with industry professionals on how to start a job search, prepare a cover letter & resume, have a successful interview, and present an engaging job talk.

Meeting and Presentation Skills

Creating a Poster for a Scientific Meeting
February 12–18 (Online)

Learn how to organize and create an effective and engaging scientific meeting poster.

Presenting a Scientific Poster
February 26–March 4 (Online)

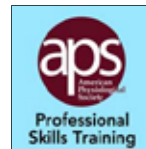
Learn the essentials of presenting a poster to multiple audiences at a scientific meeting.

Networking at a Scientific Meeting
March 12-18 (Online)

Learn how to successfully network at a scientific meeting.

Abstract Writing for Scientific Meetings
October 15-21 (Online)

Receive feedback on your first-author abstract while improving your abstract writing skills.



For More Information Visit or Follow Us
Website: www.the-aps.org/PST
Facebook: APS.PST
Twitter: @APS_PST

Missed **Experimental Biology** 2014?



OR

Attended EB 2014 But Missed
APS Career/Trainee/Mentoring/Education Sessions?

You can still attend them!

Listen to the talks and view the PowerPoint presentations for:

Refresher Course

Exercise Physiology: The Role of Exercise in Disease Prevention,
Treatment, and Optimal Aging

the-aps.org/refresher-exercise

Career Symposium

Conscious Choice and Serendipity in Your Career Trajectory

the-aps.org/career-trajectory

Mentoring Symposium

Ahead of the Curve: Taking the Lead

the-aps.org/taking-lead

Trainee Symposium

The Other Side of the Submit Button: The Ins and Outs of the
Manuscript Review Process

the-aps.org/peer-review-process

People and Places

Six APS Members Elected to the Institute of Medicine

The Institute of Medicine (IOM) has announced its new members. We're excited that six APS members are among the ranks. Congratulations Walter F. Boron, Case Western Reserve University; Nancy J. Brown, Vanderbilt University; Pamela B. Davis, Case Western Reserve University; Gerard Karsenty, Columbia University; Kelle Harbart Moley, Washington University; and Michael N. Shadlen, Columbia University! The six APS members were elected as part of a class of 70 new members and 10 foreign associates during the IOM's 44th annual meeting.

The IOM is both an honorific membership organization and an advisory organization. Established in 1970 by the National Academy of Sciences, IOM has become recognized as a national resource for independent, scientifically informed analysis and recommendations on health issues. With their election, members make a commitment to volunteer their service on IOM committees, boards, and other activities. With the election of the new class, IOM's total active membership is 1,798 and 128 foreign associates. ●

David S. Bruce Awards for Undergraduates in Research

Application deadline: January 12, 2015

the-aps.org/bruce

The David S. Bruce Undergraduate Awards are presented annually to undergraduate students who are first authors on an Experimental Biology (EB) abstract and presenting their research at the EB meeting. There are two types of Bruce Awards that students can apply for through a single application. See the website for more details and apply online at the-aps.org/awardapps.

David S. Bruce Outstanding Undergraduate Abstract Awards

This award provides \$100 and a 2-year complimentary membership with APS. The student must be enrolled as an undergraduate at the time of the abstract submission, be the first author on a submitted abstract for the EB meeting, and be working with an APS member. Selection of awardees is based on the abstract, letter of application, and letter of support from the research host. Receipt of the award is contingent on presenting the research at EB.

David S. Bruce Excellence in Undergraduate Research Awards

To be considered for this award, students must be a David S. Bruce Outstanding Undergraduate Abstract Awardee and must attend and present a poster at the EB meeting. The recipients receive \$400 and a certificate. The highest ranked awardee receives another \$250, thanks to the generous contribution of an APS member. Selection of awardees is based on the quality of the poster and oral presentation of the poster to the David Bruce Award Selection Committee.

Membership

Obituary: Stanley G. Schultz (1931-2014)

Ray Frizzell, University of Pittsburgh
Jack Byrne, University of Texas, Houston



Stan Schultz

Stanley “Stan” Schultz, a world-renowned scientist, educator, administrative leader, and past-president of the APS died of cancer on October 24, 2014 at his home in Mountain View, CA. He was 82.

Stan was the Dean of the University of Texas Medical School at Houston (2002-2006), Associate Dean for Institutional Advancement (2006-2010), and

professor in the Department of Integrative Biology and Pharmacology and the Department of Internal Medicine. He was chair of the Department of Cell Biology and Physiology (1979-1995). He retired from UT with the title of emeritus professor on August 31, 2010.

Stan is widely recognized as an outstanding scientist and educator who made fundamental contributions to the understanding of epithelial ion, sugar, and amino acid transport. His early work was the first to demonstrate sodium-coupled sugar and amino acid absorption by the small intestine, establishing the “sodium-gradient” hypothesis of solute transport and providing the rationale for the development of oral rehydration therapy. He was one of the first to recognize the significance of the paracellular pathways that traverse the epithelial barrier, and with colleagues he proposed a model for chloride secretion by epithelial cells that is widely accepted. In short, his work provided a functional identification of many of the channels and transporters that mediate transepithelial transport, which are now defined at the molecular level.

He was a native of New York City, graduating with a B.A. from Columbia University summa cum laude in 1952 and from New York University with his medical degree in 1956, where he was a member of AOA. Following an internship and residency in internal medicine at Bellevue Hospital, he became a fellow in cardiology and developed his interest

in electrophysiology. His affinity for an exigent description of mechanism led Stan to the Biophysical Laboratories at Harvard Medical School in 1959 as a National Research Council postdoctoral fellow, where he met and worked with Peter F. Curran, a gifted theoretician. Together, they rigorously evaluated and modeled the “Coupled transport of sodium and organic solutes,” the title of their seminal *Physiological Reviews* article published in 1970.

In 1967, Stan joined the Department of Physiology at the University of Pittsburgh School of Medicine as associate professor and was soon promoted to professor. It was during this period that we came to know him also as an outstanding teacher, as evidenced by his award of several “Golden Apples” by the medical students. During our time together as junior faculty at Pittsburgh, there was always talk among the faculty about pedagogy, which was a hot topic because we all had to lead small group conferences (now called team-based learning) on various topics in the course ranging from electrical signaling in nerve cells to the counter-current multiplier system in the kidney. We were all trying to outdo each other to find ways to explain difficult topics, not just to the students but also to each other. Of all the great teachers in the department, Stan was clearly the master of pedagogy and a mentor to us all. Stan had the unique ability to explain difficult concepts to the students by using examples and humor. We fondly remember his vivid explanations about how a pair of charged molecules move sequentially through a membrane channel to establish a diffusion potential by saying it was just like a poor swimmer like him being tethered with a large rubber band to an Olympic swimmer like Mark Spitz. To emphasize that he was a poor swimmer, Stan would say “To me, swimming is an attempt to stay alive in water.” So you could envision the two “swimmers,” the graceful Spitz followed by the floundering Schultz moving through that channel together at the same speed, but with Spitz always in front! If Stan could make molecules moving through a channel interesting, we felt that we had a chance at making our lectures interesting and understood. His storytelling abilities enmeshed basic science with history (e.g., the Broad Street Pump as the source of the cholera outbreak

in Soho, London), providing a bit of epidemiology with secretory pathology. Another example of his colorful style was his conjecture of how the “man on the street” would describe the function of the kidneys: they make urine. But, Stan would assert, this is like saying that the goal of Michelangelo in carving the magnificent statue of David, which stands in the Accademia di Belle Arti di Firenze, was to make marble chips. Rather, the role of the kidneys is to “make the man.” At both Pittsburgh and Houston, Stan had a profound impact in mentoring students and in developing the faculty, and we were fortunate to be recipients of his gifts.

At the University of Texas Medical School at Houston, Stan received recognition for his research, administrative leadership, and popularity among the students and faculty for his teaching abilities. One of his first successes upon becoming Chair of the Department of Physiology and Cell Biology was to argue that the floundering systems-based curriculum be replaced with a discipline-based approach. The newly constituted curriculum was a great success, and the physiology course that he ran consistently was voted by the students to be the best basic science course. In recognition of his enormous contributions to education, Stan received the 1999 President’s Scholar Award from the Health Science Center, the highest honor bestowed on a faculty member by the UT Health Science Center. Although his tenure as dean of the Medical School was cut short because of health reasons, Stan was both an effective and beloved dean. He was particularly proud of his creation of the school’s Global Health Initiative, his creation of a unique Surgical and Clinical Skills Center, featuring the latest in robotic education, and his oversight of the design and construction of the school’s new research building.

Stan is the author of nearly 200 peer-reviewed research papers and the author, coauthor, or editor of 14 books, including one with one of us (J. Byrne; *Introduction to Membrane Transport and Bioelectricity*) that grew from lecture notes in the Pittsburgh physiology course. His many editorial roles for professional journals in his field include editor-in-chief of the *American Journal of Physiology* and the *Journal of Applied Physiology* (sections on gastrointestinal physiology), *Physiological Reviews*, the five-volume *Handbook of Gastrointestinal Physiology*, and *News in Physiological Sciences*.

A member of the American Physiological Society since 1966, Stan served as its president from 1992 to 1993. He has also served as president of the Association of Chairmen of Departments of Physiology, where he received the society’s award for distinguished scientific accomplishments. He served also as chairman of the U.S. National Committee of the International Union of Physiological Sciences, chairman of their Scientific Program Committee for the meeting held in St. Petersburg, Russia in 1997, and he served on the Executive Committee of the Federation of American Societies for Experimental Biology.

Stan was the recipient of numerous awards, including the 1978 Hoffman-LaRoche Prize for Outstanding Contributions to Gastrointestinal Physiology as well as the 1999 Arthur C. Guyton Best Teacher of the Year Award, and the 2003 Daggs Award from the APS, given by the Society for lifetime contributions to physiology. In 2003, he was honored with the Solomon A. Berson Alumni Achievement Award for contributions to clinical science from his alma mater, the NYU College of Medicine. He was elected to membership in the Association of American Physicians in 1981 and to membership in the European Academy of Sciences in 2004, and in 2006 he received the Prince Mahidol Award in Medicine from the King of Thailand in recognition of his work that established the scientific foundation of oral rehydration therapy (ORT). ORT has been credited with saving the lives of millions of patients suffering from dehydration due to diarrheal diseases. In 2012, the World Health Organization cited ORT as second only to vaccinations as a life-saving intervention. In September 2007, Stan received the “Seeds of Hope” award from RESULTS for his contributions benefiting the health of impoverished children.

Stan is survived by Harriet, his wife of 53 years, sons Jeffrey and Kenneth, and five grandchildren.

Gifts in the memory of Stan may be made to the Stanley G. Schultz, M.D. Student Travel Award in Global Health or The Doris Simon Student Fund. Gifts should be mailed to: UT Health, Office of Development, P.O. Box 1321, Houston, TX 77251-1321. ●

New Regular Members

***transferred from student membership**

Alessandra Adami

Los Angeles BioMedical Res. Inst. at
Harbor, Torrance, CA

Michael Edward Adams

Univ. of California, Riverside,
Riverside, CA

Olushola Emmanuel Adeleye*

Federal Univ. of Agriculture,
Abeokuta, Nigeria

David William Adelson

David Geffen UCLA Sch. of Med., Los
Angeles, CA

Tomader Ali

Univ. of Alabama at Birmingham,
Birmingham, AL

Matthew Peter Anderson

Harvard Med. Sch./Beth Israel
Deaconess Med. Coll., Boston, MA

Luca Angius

Sch. of Sport and Exercise Sciences,
Chatham, Kent, United Kingdom

Hnin Hnin Aung

Univ. of California at Davis, Davis, CA

Christopher T. Banek

Univ. of Minnesota, Saint Paul, MN

Andrew Edward Beaudin*

Univ of Calgary Mackimmie Lib.,
Calgary, Canada

Gwenaëlle Begue

Ball State Univ., Muncie, IN

Frank Bremmer

Univ. of Marburg, Marburg, Germany

Rodney Britt

Mayo Clinic, Rochester, MN

Rebecca Sue Bruning*

Indiana Univ. Sch. of Med.,
Indianapolis, IN

Viviane Callier

Duke Univ., Rockville, MD

Guan Cao

Univ. of Texas at Austin, Austin, TX

Stephen Jordan Carter*

Univ. of Alabama at Birmingham,
Birmingham, AL

Jie Chao

Southeast Univ., Nanjing, China

Bernadette Chen

The Res. Inst. at Nationwide Children's
Hosp., Columbus, OH

Gerard Clarke

Univ. College Cork, Cork, Ireland

Heidy Lorena Contreras

Univ. of La Verne, La Verne, CA

Frederic Crevecoeur

Univ. Catholique De Louvain,
Louvain-la-Neuve, Belgium

J. Thomas Curtis

Oklahoma State Univ. Ctr. for Health
Scienc, Tulsa, OK

Numa Dancause

Univ. De Montreal, Montreal, Canada

Jason M. De Freitas

Oklahoma State Univ., Stillwater, OK

Douglas Dayton Deming

Loma Linda Univ., Loma Linda, CA

Olga Dergacheva

The George Washington Univ.,
Washington, DC

Jennifer T. Durham

Tufts Univ., Boston, MA

Jhansi Rani Dyavanapalli

The George Washington Univ.,
Washington, DC

Lashauna Evans

Univ. of Texas Health Sci. Ctr. at San
Antonio, San Antonio, TX

Carmella Evans-Molina

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Qilong Feng

West Virginia Univ., Morgantown, WV

Jerome Fleuriot

Univ. of Washington/Primate Ctr.,
Seattle, WA

Heather Francis

Baylor Scott & White Healthcare,
Temple, TX

Eric Christopher Freese*

Pepsico, Inc., New York, NY

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Indianapolis, IN

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Australia

R. C. Fernanda Giachini

Univ. Federal do Mato Grosso, Barra
Do Garcas, Brazil

Diane Godin-Ribuot

Univ. Joseph Fourier, Grenoble, France

Edward John Golob

Tulane Univ., New Orleans, LA

Elisa J Gonzalez-Rothi
Univ. of Florida, Alachua, FL

Christopher G. Guglielmo
Univ. of Western Ontario,
London, Canada

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Univ. of Cincinnati Blue Ash College,
Blue Ash, OH

Michelle Harrison*
Univ. of Texas at Austin, Austin, TX

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Loughborough Univ., Loughborough,
United Kingdom

Daniel M. Hirai
Queen's Univ., Kingston, Canada

Else Kay Hoffmann
Univ. of Copenhagen, Copenhagen,
Denmark

Adam Gordon Hotchkiss
Dalhousie Univ., Halifax, Canada

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Dallas, TX

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Sri Kasturirangan
Univ. of Wisconsin, Middleton, WI

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Bronx, NY

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Univ. of Florida, Gainesville, FL

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Oakland Univ. William Beaumont Sch.
of Med., Rochester, MI

Ivana Y. Kuo*
Yale Univ. Sch. Med., New Haven, CT

Tim Lahm
Indiana Univ., Indianapolis, IN

Benjamin Lauzier
L'Institut Du Thorax, Nantes, France

Gina Marie Leininger
Michigan State Univ., East Lansing, MI

Erica S. Levitt
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Heng Lin
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Taipei, Taiwan

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Laredo, TX

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Rocky Vista Univ. COM, Parker, CO

Chetan N. Patil
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Jackson, MS

Lisa Pierce
Tripler Army Med. Ctr.,
Tripler AMC, HI

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Univ. of Sydney, Lidcombe, Australia

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The Univ. of Alabama at Birmingham,
Birmingham, AL

Christopher Ronald Rathbone
United States Army Inst. of Surgical
Res., San Antonio, TX

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Al-Qura U Makkah, Saudi Arabia

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and Res., Bangalore, Whitefield,
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Hilary SeifertLouisiana State Univ. Health Sci. Ctr.,
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Port Aransas, TX**Bun Tsuji**

JSPS, Tsukuba City, Japan

Sara Freiberg Turner

Univ. of Florida, Gainesville, FL

Marta Noa Valcarcel Ares

Univ. of Oklahoma, Oklahoma City, OK

Aleksandr Vasilyev

NYIT COM, Old Westbury, NY

Erin V. VasudevanSUNY Stony Brook Univ.,
Stony Brook, NY**Gerry Wagenaar**Leiden Univ. Med. Ctr., Leiden, The
Netherlands**Xin Wang**The George Washington Univ.,
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Montreal, Canada**Lufang Zhou**Univ. of Alabama, Birmingham,
Birmingham, AL**Graeme Zosky**

Univ. of Tasmania, Hobart, Australia

**New Graduate
Student Members****Elizabeth Lee Adams**

Univ. of Connecticut, Manchester, CT

Farjana Akther

Univ. of the Pacific, Stockton, CA

Asghar Ali

Colorado State Univ., Fort Collins, CO

Jacob Matthew Allen

Univ. of Illinois, Champaign, IL

Amy Arnold

Robert Gordon Univ., Aberdeen,
United Kingdom

Gunisha Arora

Northern Illinois Univ., Dekalb, IL

Amirhossein Arzani

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Berkeley, CA

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Heights, OH

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David Stewart Bayless

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Emma Leigh Beamish

Lancaster Univ., Lancaster,
United Kingdom

Anson Blanks

Virginia Commonwealth Univ.,
Richmond, VA

Philip Apraku Bondzie

Boston Univ. Sch. of Med., Boston, MA

Igor Dutra Braz

Univ. of Birmingham, Birmingham,
United Kingdom

Joseph John Candela

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Adedayo Catlett

Northeastern Univ., Salisbury, MD

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Fort Worth, TX

David Douglas Church

Univ. of Central Florida, Orlando, FL

Kristin Claflin

Univ. of Iowa, Iowa City, IA

Matthew Steven Clark

Univ. of Oregon, Eugene, OR

Daniel Craighead

The Pennsylvania State Univ.,
Univ. Park, PA

Kaylin Dix Didier

Univ. of Oklahoma, Norman, OK

Chidinma Ijeoma Eziuzo

Univ. of Port Harcourt, Nnewi, Nigeria

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Charmain Angela Fernando

Univ. of Missouri, Columbia,
Columbia, MO

Liam Fitzgerald

Univ. of Massachusetts,
Southampton, MA

Jean Fredrick

St. John's Med. Coll., Bangalore,
Karnataka, India

Joanne Garbincius

Univ. of Michigan, Ann Arbor, MI

Christine P. Gibson

Midwestern Univ., Glendale, AZ

Taneisha Renee Gillyard

Meharry Med. Coll., Nashville, TN

Maria Alejandra Gonzalez

Mayo Clinic, Rochester, MN

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KU Leuven, Leuven, Belgium

Kofi-Kermit Horton

Univ. of South Florida, Tampa, FL

Catherine Margaret Ivy

McMaster Univ., Hamilton, Canada

Heather Jameson

George Washington Univ.,
Washington, DC

Melanie Jannaway

Univ. of Southampton, Southampton,
United Kingdom

Sachini Jayaratne

Univ. of Sydney, Camperdown,
Australia

Kevin L. Kelly

Michigan State Univ., Okemos, MI

Obaid Khurram

Mayo Clinic, Rochester, MN

Noélie Egídia Watanabe Kiill

Univ. Estadual Paulista,
Araçatuba, Brazil

Ayeeshik Kole

Indiana Univ. Sch. of Med.,
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Stephanie Paige Kurti

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Porter Physiology Development Fellowships

Application deadline: January 15, 2015

<http://www.the-aps.org/porter>

The goal of the Porter Physiology Development Program is to encourage diversity among students pursuing full-time studies toward the Ph.D. in the physiological sciences and to encourage their participation in the Society.

The Porter program provides a full-time graduate fellowship (\$28,300 during the academic year) to students in programs leading to the Ph.D. in the physiological sciences at U.S. institutions. The program is open to underrepresented racial and ethnic minority applicants who are citizens or permanent residents of the U.S. or its territories and student members of the Society. Apply online at <https://www.the-aps.org/awardapps>.

Publications

Interview with David Pollock



David Pollock



David Pollock is the Editor-in-Chief of *Comprehensive Physiology* and President of the American Physiological Society (APS), and is the NRTC Endowed Professor in

the Division of Nephrology Department of Medicine at the University of Alabama at Birmingham (UAB). He serves as Director of UAB's Cardio-Renal Physiology and Medicine section, a translational research program supported jointly by the Division of Cardiovascular Disease and Division of Nephrology.

Comprehensive Physiology is published on behalf of the APS by Wiley. Launched in 2011, it is a relatively new publication, as the APS published its first journal in 1898. In this interview, Pollock discusses some new initiatives being developed by APS and Wiley to introduce *Comprehensive Physiology* to a wider audience.

All APS members are being given access to *Comprehensive Physiology* through the end of 2015. Why is this important?

I thought it was important that our 11,000 APS members are able to access all the content in *Comprehensive Physiology* so that they can read it, use it for teaching, and of course cite it. We are extending this benefit through the end of 2015 so that our members can experience and appreciate what a great resource *Comprehensive Physiology* can be for them.

It certainly is a valuable resource. Can you tell us how an APS member gets access to the publication?

APS members only need to click on the link to *Comprehensive Physiology* from that journal's page

on the APS website [<http://www.the-aps.org/mm/Publications/Journals/CompPhys>] and enter his or her APS membership credentials. They are then brought to Wiley's *Comprehensive Physiology* site, where they can search or browse for content within the publication and instantly access it without logging in again.

Your institution did not have a subscription to *Comprehensive Physiology* until you requested it. Can you tell us more about how that happened?

When I arrived at UAB in early 2014, I quickly learned that we did not have a subscription to *Comprehensive Physiology*. I let our librarian know that I was the editor and told her about the history and usefulness of this publication, and within a few weeks they had arranged a licensing agreement with Wiley. Now all researchers, students, and others at UAB have access to all the content. Librarians make purchasing decisions based on what their faculty use and want to use; they find it helpful when the faculty let them know. If you want your library to subscribe to *Comprehensive Physiology*, please ask your librarian! Also, you will find a Librarian Recommendation form on the *Comprehensive Physiology* website [<http://www.comprehensivephysiology.com/WileyCDA>].

You mention the history of this publication. Can you tell us more about that?

Comprehensive Physiology is published as an online-only quarterly invited review journal, but it was actually created to update the APS's venerable *Handbook of Physiology* series, which has long been essential to all physiologists, medical students, and those non-physiologists who need to know something about how the body functions. The volumes of the *Handbooks* were published based on the systems of the body, for instance,

the cardiovascular or renal system, and *Comprehensive Physiology* is also divided into sections and topics based on those systems.

So how does *Comprehensive Physiology* differ from the Handbook series?

Unlike the book series, the most up-to-date articles are published online in *Comprehensive Physiology* as they are written and accepted, regardless of which section they are in. This means that researchers and faculty can get access to the most up-to-date articles right away, without having to wait years or sometimes decades for new editions of the books to be published.

Can you tell us more about the ongoing updates and development of *Comprehensive Physiology*?

The first section of articles invited for and published in *Comprehensive Physiology* were about the respiratory system. New articles are added every quarter and now include exercise, renal, gastrointestinal, and others. I am very excited that we have now started publishing cardiovascular and neurophysiology articles – these were some of the most popular *Handbook* topics, and the quality of the new articles has been impressive. The original *Handbook* chapters have also been made available through the *Comprehensive Physiology* website, and I have been impressed by how much of the original content still gets used.

What message would you like to give authors who have been invited to write for *Comprehensive Physiology*? How would you distinguish it from APS's other review journals?

First of all, I want to thank the authors who have written articles for *Comprehensive Physiology*. There are some real gems in there that will be read and cited for years to come. For those who are invited, I would say, *Comprehensive Physiology* is really like a state-of-the-art textbook rather than a review journal. The title “comprehensive” confuses a lot of people. I tell people that it really refers to the breadth of the journal, not the breadth of individual articles. Don Kohan and I wrote a huge review for *Physiological Reviews* (PRV) a couple years ago and then turned around and wrote an article for *Comprehensive Physiology* on the exact same topic. The PRV article covered every little detail and was very long with 800-some references. The *Comprehensive Physiology* article focused on what we see as the key findings – as if I were teaching my post-doc. Another way to look at it is that *Comprehensive Physiology* is meant to teach people who might be new to the field, and the other journals are speaking to people who are mostly experts.

Thank you for sharing this information with us. *Comprehensive Physiology* is destined to become an essential resource, and APS members are sure to benefit from the access that has been provided to them. ●

Dale J. Benos Early Career Professional Service Award

Application deadline: January 23, 2015

the-aps.org/benos

The Dale J. Benos Early Career Professional Service Award honors an early career stage (graduate student, postdoctoral fellow, assistant professor, or equivalent position) member of APS. The award will honor someone who is judged to have made outstanding contributions to the physiology community and demonstrated dedication and commitment to furthering the broader goals of the physiology community. This can be by serving on professional committees, by participating in K-12 education outreach, participating in scientific advocacy and outreach programs, or by otherwise strengthening and promoting the physiology community. See the website for more details and apply online at the-aps.org/awardapps.

Publications

Calls for Papers

Physiological Genomics

- Gut Microbiota in Health and Disease
- Systems Biology and Polygenic Traits

Journal of Neurophysiology

- Neurobiology of Deep Brain Stimulation
(Submission deadline: March 1, 2015)
- Decision Making: Neural Mechanisms
(Submission deadline: March 1, 2015)
- Correlating Neuronal Activity and Neural Imaging
(Submission deadline: March 1, 2015)
- Neurophysiology of Tactile Perception: A Tribute to Steven Hsiao
(Submission deadline: June 1, 2015)
- Neuronal Diversity: Categorizing Types of Neurons
(Submission deadline: July 1, 2015)
- Control of Autonomic Function: Insights From Neurophysiological Studies in Conscious Animals (Including Humans)
(Submission deadline: July 1, 2015)

Advances in Physiology Education

- Pre-Professional Education in Transition

American Journal of Physiology – Gastrointestinal and Liver Physiology

- Physiology and GI Cancer
- Intestinal Stem Cells in GI Physiology and Disease
- Innovative and Emerging Technologies in GI Physiology and Disease

American Journal of Physiology – Heart and Circulatory Physiology

- Cardiac Regeneration and Repair: Mechanisms and Therapy
(Submission deadline: January 15, 2015)
- Impact of Sympathoexcitation on Cardiovascular Function in Humans
(Submission deadline: January 15, 2015)
- Cardiovascular Responses to Environmental Stress
(Submission deadline: May 15, 2015)
- Exercise Training in Cardiovascular Disease: Mechanisms and Outcomes
(Submission deadline: May 15, 2015)

American Journal of Physiology – Lung Cellular and Molecular Physiology

- Sex Differences in the Respiratory System
- Translational Research in Acute Lung Injury and Pulmonary Fibrosis
- Biomarkers in Lung Diseases: From Pathogenesis to Prediction to New Therapies
- Bioengineering the Lung: Molecules, Materials, Matrix, Morphology, and Mechanics
- Nanoparticles and the Lung: Friend or Foe?
(Submission deadline: March 1, 2016)

American Journal of Physiology – Regulatory, Integrative and Comparative Physiology

- Central Control of Cardiovascular Function
(Submission deadline: January 31, 2015)
- Oxygen as a Regulator of Biological Systems
(Submission deadline: April 30, 2015)

For a complete list of current Calls for Papers, visit the APS homepage and click on the tab for Calls for Papers.

News from Distinguished Physiologists

Letter to Lois Jane Heller

Paul Hill writes: "Thank you for the invitation to write to you.

"It is now 20 years since I took early retirement from the University of Auckland, and I am no longer active in physiology. But children and grandchildren, writing, music, film and theatre, woodwork, tramping, and travel make for a full and fulfilling life. Now that I'm 80, I can look back on a life that has had its share of ups and downs but which has been satisfying and enjoyable.

"My academic career began in 1962 when, an educational novice, I was appointed to the Fiji School of Medicine (FSM) to teach physiology, biochemistry, and introductory medicine. The FSM began in 1875 as a school to train native vaccinators, with smallpox having been brought to Fiji with indentured Indian labor.

"When I joined it, FSM offered a 5-year course leading to a qualification as an Assistant Medical Officer (AMO). It also trained a variety of paramedicals – laboratory technicians, radiologists, dieticians, and health inspectors. Its students came from all over the Pacific, with entry qualifications ranging from having completed primary school to having passed the New Zealand University Entrance examinations.

"My students in Fiji taught me as much if not more than I taught them. They taught me to respect them and their dedication; they taught me the difference between intelligence and education; they taught me that, in every discipline, the first requirement is a sound knowledge of the vocabulary of that discipline. I learned that some of them had no words for abstract concepts in their native languages and that, for some, English was not merely a second language but a third, fourth, fifth, or even sixth language. That in turn taught me that simple grammatical construction could illuminate the understanding of complex concepts. I learned to avoid subordinate clauses like the plague they are and use the simple sentence structure of the Authorized Version of the Bible as my model.

"In many ways, that was one of the most rewarding experiences of my life. It was immeasurably satisfying, and it was fun. They were lovely young people to teach.

"Looking back on my career, there are other lessons I have learned. I learned that we often spend too much time thinking about teaching and not enough about learning. I learned that, in addressing any topic, the first and most difficult task is deciding what to leave out. I learned that the regurgitation of lecture content is a poor educational objective. I learned that we all learn by making mistakes, and students should never be mocked for having the courage to make mistakes. I learned to be sceptical of fashionable educational dogmas such as problem-based learning.

"In my experience, problem-based learning is not without problems. First, it is expensive of both staff and student time and other resources. Second, problems are most easily introduced when the student has acquired the necessary vocabulary for defining the problem. That vocabulary is most efficiently provided through didactic teaching. Third, the problem is usually defined within known parameters and assumes the successful solution of the problem by logical analysis and the application of current knowledge. It too often directs the student to search for the single "right" answer rather than recognizing that some problems may have several valid solutions.

"In New Zealand, the majority of students enter medicine directly from school where they have been brainwashed into the belief that there is always a single right answer to any question, the one that rewards them with high marks in the examination. They are uncomfortable with uncertainty. Bertrand Russell wrote in the introduction to his *History of Western Philosophy* that one of the functions of philosophy was 'To teach how to live without certainty, and yet without being paralyzed by hesitation . . .' (p. 11). In my view, that is also the function of medical education. Medical practice is full of uncertainties but often cannot afford hesitation. We cannot always solve a problem but we must always make a decision. To that end, I think our focus would be better fixed on robust decision making than on problem-solving techniques.

"It is also important that we encourage our students to think imaginatively. Einstein repeatedly stated his belief

that 'Imagination is more important than knowledge. For knowledge is limited, whereas imagination embraces the entire world' (Albert Einstein, *Cosmic Religion: With Other Opinions and Aphorisms*. 1931, p. 97). Our function is not merely to train students for the practice of medicine according to the best standards of current knowledge but to train our successors to advance the standards of practice and the quality and breadth of our knowledge. We will only achieve that function if we encourage our students to use their imaginations.

"It can be salutary to ask ourselves why we are teaching what we are teaching and whether what we teach serves the purpose we intend. Asking myself these questions led me to rethink laboratory teaching for third-year bachelor of science students taking physiology. It seemed to me that the purpose of this course was to inform students about the nature of physiological investigation and how the evidence acquired is evaluated. Accordingly, I reorganized the course as follows.

"Students worked together in small groups. They were first exposed to a simple experimental technique and allowed to familiarize themselves with this over two or three sessions. During this time, they were told to think up a question to be answered using the technique and to develop an experimental plan to be

carried out over the following weeks of the course. The results of their experiments and the conclusions drawn from these results were to be presented as a poster at the end of the course. The posters were displayed in a final session attended by all departmental staff who circulated around all the posters, questioning, discussing, and assessing the students' findings. Staff assessments were averaged, and a mark was awarded to each group. To introduce students to the idea of peer review, each group decided how the group mark should be distributed among the members of the group. Their judgements were generally fair but uncompromising. Non-contributors got short shrift.

"In general, good students enjoyed the challenges of this approach, whereas others hankered after a more conventional, directed approach. There were problems, but I thought the experiment was well worthwhile.

"Finally, I was once told that, as an academic, I had a responsibility first to myself, second to my discipline, and third to my students. There is some truth in the dictum, but I have come to believe that ranking these responsibilities is less important than striking a balance between them. My students were always far too rewarding ever to rank them as last in importance." ●

Undergraduate Summer Research Fellowships

Application deadline: February 1, 2015

<http://www.the-aps.org/summerresearch>

APS is proud to offer four programs that allow undergraduate students to participate in research during the summer (IOSP, STRIDE, UGSRF, UGREF). Recipients spend an average of 10 weeks in the laboratory of an established scientist and APS member. Each program recruits undergraduate students nationwide, two internationally. Some programs are open to students from disadvantaged backgrounds, students from underrepresented racial and ethnic groups, and students with disabilities. Each Fellow receives a stipend plus additional funds for travel to present his or her research at a scientific meeting. Research hosts receive funds for student lab supplies. See the website for more details and apply online at <https://www.the-aps.org/awardapps>.

Positions Available

Postdoctoral Research Associate: A NIH-funded postdoctoral research associate position is available immediately at the Cardiovascular Research Center of Rhode Island Hospital and the Alpert Medical School of Brown University to study the role of cardiac fibroblasts and cross talk between myocytes and fibroblasts in the cardiac remodeling response to hemodynamic stress and injury. The laboratory of Dr. Ulrike Mende takes integrative and multi-scale approaches at the single cell, bio-engineered microtissue, organ and whole animal level to address clinically relevant questions in cardiac remodeling, using molecular, biochemical, cell biological, and physiological approaches and genetically modified mouse models. The interdisciplinary Cardiovascular Research Center (<http://cvrc.brownmedicine.org>) is home to scientists, physician-scientists, and students investigating molecular mechanisms of cardiac disease, with focal areas in cardiac hypertrophy and failure, ischemia/reperfusion, arrhythmias and sudden cardiac death. Investigators are supported by Hospital- and Brown-based core facilities. Candidates should have the following qualifications: PhD, MD, or equivalent degree; relevant research experience; and ability to work with small rodents. Prior experience in animal research and rodent microsurgery are preferred. Must be highly motivated, have effective verbal and written communication skills, and work well independently as well as in a team. Interested candidates should send a letter of application (summarizing research experience and career goals), curriculum vitae, and the names and contact information for three references to Dr. Ulrike Mende, Associate Professor of Medicine; e-mail: ulrike_mende@brown.edu. The salary will be based on years of experience, commensurate with NIH guidelines. Rhode Island Hospital is part of Lifespan, an equal opportunity employer; women and minority applicants are encouraged to apply.

Postdoctoral Position: The Chapkin laboratory at Texas A&M University has an opening for a recent PhD graduate with an interest in one or more of the following areas: obesity, intestinal stem cells, epigenetics, metabolic profiling in preclinical models, gut-microbe cross talk, membrane biology, immunology, and/or computational biology/bioinformatics. Our lab studies the effects of natural bioactive molecules on chronic diseases, with particular interest in molecular mechanisms of action. Methods include flow cytometry, cell culture, confocal (FLIM, FRET, TIRF) super-resolution microscopy,

metabolite analysis, RNA Seq, ChIP Seq, microRNA analysis, among others. Pathways of interest include Wnt signaling, lipid rafts/cytoskeletal interaction, AhR, an Myc and EGFR/Ras-dependent signaling. The successful candidate will be highly motivated, comfortable with technical challenges and problem solving, and able to work collaboratively. Experience with microscopy is an asset. Competitive salary and benefits are available commensurate with experience. Fluent English, a track record of strong publications, and a cooperative attitude are a must for this position. Please submit your CV and statement of current interests to Dr. Laurie Davidson, L-davidson@tamu.edu.

Tenure/Tenure-Track Full-Time Chair: California State University, Sacramento, seeks to hire a qualified person to serve as full-time Chair of the Department of Kinesiology and Health Science. The new Chair will have an excellent opportunity to shape the future direction of a dynamic multidisciplinary department. The Department of Kinesiology and Health Science is looking for an individual with a documented record of academic innovation and progressive leadership. The Department of Kinesiology and Health Science is part of the College of Health and Human Services. The department offers bachelor of science degrees in Kinesiology, Health Science, and Athletic Training. A master of science is also available and offers two concentrations of study (Exercise Science and Movement Studies). The successful candidate must qualify for retreat rights as a faculty member in Kinesiology (Athletic Training, Exercise Science, or Physical Education) or Health Science (Community Health Education, Health Care Administration, or Occupational Health & Safety). Applications are only accepted through the Sacramento State jobs website. For full vacancy announcement, including application procedure, please see <http://www.csus.edu/about/employment/>. The review of applications begins December 1, 2014 and will continue until the position is filled. Address questions only to: Dr. Patty Woodward, Assistant Professor; phone 916 278-7478. AA/EEO. Clery Act Statistics available. Mandated reporter requirements. Criminal background check may be required.

Faculty Positions (Tenure-Track Assistant, Tenured Associate, and Tenured Full Professor): The Department of Physiology and Biophysics invites applications for two tenure-track faculty positions for candidates of

exceptional promise and achievement. All ranks will be considered. We seek extraordinary applicants working at the frontiers of molecular and cellular physiology, broadly defined. The research program of the successful candidate will add to or complement the department's existing strengths in structural biology, proteomics, cell signaling, immunology, ion channels, and metabolism (<http://www.physiology.uci.edu/>). Candidates must hold a PhD or MD degree and will be expected to teach medical and graduate students. Inquiries about the position should be directed to Professor Albert Zlotnik, Search Committee Chair, Department of Physiology and Biophysics, School of Medicine, D340 Medical Science I, University of California, Irvine, CA 92697-4560. URL for online applicants: <https://recruit.ap.uci.edu/apply/JPF02199> (full professor, tenured); <https://recruit.ap.uci.edu/apply/JPF02198> (associate professor, tenured); <https://recruit.ap.uci.edu/apply/JPF02245> (assistant professor, tenure-track). Applicants should complete the application profile and upload the following application materials electronically to be considered for the position: 1) cover letter, 2) curriculum vitae, 3) summary of research interests (up to 3 pages), 4) summary of teaching experience, 5) names and contact information for 3-5 referees, 6)

PDFs of 3-5 recent publications. Applications will be considered until the position is filled. The University of California, Irvine is an Equal Opportunity / Affirmative Action Employer advancing inclusive excellence. All qualified applicants will receive consideration for employment without regard to race, color, religion, sex, national origin, disability, age, protected veteran status, or other protected categories covered by the UC nondiscrimination policy.

Assistant Professor: The Department of Biological Sciences at California State University San Marcos (CSUSM) invites applications for a tenure-track position at the assistant professor level in the field of Animal Physiology to begin Fall 2015. Applicants must have a PhD in the biological sciences with training and research in animal physiology and some postdoctoral experience. All subdivisions of physiology will be considered, including but not limited to comparative physiology, evolutionary physiology, and developmental physiology. Researchers who focus on invertebrate systems are especially encouraged to apply. Previous teaching experience is preferred. The successful applicant will have a strong commitment to undergraduate

**FACULTY AND VICE-CHAIR OF RESEARCH POSITION
DEPARTMENT OF SURGERY
CARDIOVASCULAR-RENAL RESEARCH CENTER
University of Mississippi Medical Center**

The Department of Surgery and the Cardiovascular-Renal Research Center (CRRC) at University of Mississippi Medical Center invites applicants for a state supported, tenure track faculty position at the rank of Assistant, Associate, or full Professor. The applicant must have a Ph.D. and/or M.D. degree with appropriate research experience and extramural research funding. Special consideration will be given to candidates with strong backgrounds in one or more of the following areas: 1) vascular biology, 2) ischemia-reperfusion injury 3) organ transplantation biology. The successful candidate is expected to develop a nationally recognized research program, manage departmental research activities and mentor post-doctoral M.D. and Ph.D. fellows within the Department of Surgery. The large group of CRRC scientists offers excellent opportunities for collaboration at molecular, cellular or systems levels of integration. The department offers generous laboratory space in a new state-of-the-art building and excellent core facilities within the CRRC. New faculty members receive highly competitive salaries and start-up packages. Applicants should send a curriculum vitae, a statement of research plans, previous and current extramural research funding, and the names of at least three references to: Dr. Joey Granger, Director, Cardiovascular-Renal Research Center, University of Mississippi Medical Center, 2500 North State Street, Jackson, MS 39216-4505. E-mail: jgranger@umc.edu. Equal opportunity employer, M/F/D/V

education and should have a demonstrated ability or potential to effectively instruct undergraduate and graduate students, establish community partnerships, and develop an independent research program that includes undergraduate and graduate students. Teaching responsibilities will include some combination of a biology major core animal physiology course, upper division physiology electives (preferably with a laboratory section), upper division general education courses, and graduate/advanced undergraduate courses in the candidate's area of expertise. Preference will also be given to candidates with demonstrated intercultural competence with diverse groups in teaching, research, and/or service. For position details and application instructions, please see our website (www.csusm.edu/biology). Questions can be directed to ANIMPHYS1415@csusm.edu. Screening of applications will begin November 15, 2014 and continue until position is filled. California State University San Marcos is an Affirmative Action/Equal Opportunity/Title IX Employer. The University has a strong commitment to the principles of diversity and, in that spirit, seeks a broad spectrum of candidates including women, members of minority groups, and people with disabilities.

Assistant/Associate Professor: The Department of Physiology and Biophysics at Boston University School of Medicine invites candidates to apply for the position of assistant professor; exceptional candidates for a more senior position will also be considered. The Department is composed of 24 faculty with interests in structural and membrane biology, visual, neural, and muscle physiology, and lipoprotein structure and metabolism. The collegiate environment in Boston allows excellent opportunities to collaborate within as well as outside the School of Medicine. Website: <http://www.bumc.bu.edu/phys-biophys/>. The successful candidate will be expected to develop or have a strong, independently funded research program, become involved in the training and education of graduate students, and participate in teaching in the School of Medicine preclinical Physiology and/or Neuroscience courses. Candidates from any field will be considered but priority will be given to those individuals whose research interests complement those of the Department's faculty. The Department can offer a competitive startup package including generous laboratory space. For full consideration, please send a single PDF file containing a cover letter, a curriculum vitae, a statement of research and teaching interests,

and the names and contact information of at least three references to Raphael A. Zoeller, PhD, Faculty Search Chair, at rzoeller@bu.edu. Boston University School of Medicine is an equal-opportunity/affirmative-action employer. Contact information: Raphael A. Zoeller, PhD, Department of Physiology & Biophysics, Boston University School of Medicine, 700 Albany St., W302, Boston, MA 02118.

Assistant Professor of Biology: The Department of Biology at Wilmington College of Ohio is seeking a broadly trained biologist with particular expertise in anatomy and physiology for a tenure-track position starting Fall Semester 2015. The successful candidate will teach lecture and laboratory sections of vertebrate anatomy and physiology, human anatomy, and physiology, contribute to the department's general biology and Research and Seminar sequences, and mentor undergraduate research projects and theses. PhD required; teaching experience and familiarity with small college environment preferred. Wilmington College is a career-oriented liberal arts institution affiliated with the Society of Friends (Quakers). The main campus of Wilmington College is located in southwestern Ohio. Additional information about Wilmington College can be found at <http://www.wilmington.edu>. Send resume, letter of application, and names and contact information for three references to: Wilmington College, Director of Human Resources, Pyle Center 1187, 1870 Quaker Way, Wilmington, Ohio 45177-2499. E-mail submissions to humanresources@wilmington.edu are preferred. Review of applications will begin immediately and continue until position is filled. Wilmington College is an equal opportunity employer and will not discriminate unlawfully in employment matters on the basis of race, religion, gender, color, ancestry, national origin, age, disability, or any other category protected by law. EOE.

Pharmacology Faculty Position: The Department of Veterinary Physiology and Pharmacology, College of Veterinary Medicine and Biomedical Sciences at Texas A&M University (vetmed.tamu.edu) invites applications for a clinical-track or tenure-track faculty position focused on teaching pharmacology. The position is open for all academic ranks, depending on experience and qualifications. The candidate must have a demonstrated interest in teaching pharmacology; preference will be given to those with recognized expertise in teaching. Required training has either 1) a PhD degree in an area of biomedical sciences

related to pharmacology or therapeutics or 2) a DVM (or equivalent) and board eligibility in an AVMA-recognized veterinary specialty (www.avma.org/ProfessionalDevelopment/Education/Specialties). The successful applicant will be expected to devote a significant portion of his or her effort to teaching pharmacology in the veterinary professional program. The successful applicant will participate in interdisciplinary research and, if in the tenure track, will be expected to establish a sustainable, extramurally funded research program. Intra- and interdepartmental collaboration is expected and encouraged. The college serves ~500 students in the DVM professional degree program as well as 2,000 undergraduate students and 200 graduate students in a dynamic Biomedical Sciences program. Undergraduate, graduate, and professional students participate in traditional pharmacology courses. Development of additional elective courses in various topics in pharmacology at the undergraduate and graduate levels is encouraged. Review of applications will begin December 1, 2014 and continue until the position is filled. Applicants should send a letter of interest outlining their qualifications, current curriculum vitae, statement of specific research and teaching interests and philosophies,

and the names of three potential references, along with complete contact information, to: Dr. Randolph Stewart, Department of Veterinary Physiology and Pharmacology, Texas A&M University, 4466 TAMU, College Station, TX 77843-4466; phone: 979-862-7764; fax: 979-845-6544; searchpharmacology@cvm.tamu.edu. Applications may be submitted either electronically by e-mail with attachments (preferred) or by mail to Dr. Stewart. Texas A&M University is the oldest public institution of higher education in Texas and one of the nation's largest and most dynamic universities. Approximately 58,000 students are enrolled in the University's 10 academic colleges, with approximately 9,000 pursuing graduate or professional degrees. Texas A&M is a major research university with a growing international focus and outstanding public and private support. For more information about Texas A&M University, please visit <http://www.tamu.edu>. For more information about the Department of Veterinary Physiology and Pharmacology at the College of Veterinary Medicine and Biomedical Sciences, please visit <http://vetmed.tamu.edu/vtpp>. Texas A&M University is an Equal Opportunity Employer and encourages applications from all underrepresented groups. ●

**RESEARCH FACULTY POSITION
DEPARTMENT OF EMERGENCY MEDICINE
CARDIOVASCULAR-RENAL RESEARCH CENTER
University of Mississippi Medical Center**

The Department of Emergency Medicine and the Cardiovascular-Renal Research Center (CRRC) at University of Mississippi Medical Center invite applicants for a state supported, tenure track faculty position at the rank of Assistant, Associate, or full Professor. The applicant must have a Ph.D. and/or M.D. degree with appropriate research experience and extramural research funding. Special consideration will be given to candidates with strong backgrounds in one or more of the following areas: 1) vascular/endothelial biology, 2) microcirculation (in vitro flow based and in vivo techniques to monitor and assess), 3) mitochondrial function (tissue prep, oximetry, individual complex assays), 4) platelet function/coagulation (TEG, platelet mapping). The successful candidate is expected to develop a nationally recognized research program, manage departmental research activities and mentor post-doctoral M.D. and Ph.D. fellows within the Department of Emergency Medicine. The large group of CRRC scientists offers excellent opportunities for collaboration at molecular, cellular, or systems levels of integration. The department offers generous laboratory space in a new state-of-the-art building and excellent core facilities within the CRRC. New faculty members receive highly competitive salaries and start-up packages. Applicants should send a curriculum vitae, a statement of research plans, previous and current extramural research funding, and the names of at least three references to: Dr. Joey Granger, Director, Cardiovascular-Renal Research Center, University of Mississippi Medical Center, 2500 North State Street, Jackson, MS 39216-4505. E-mail: jgranger@umc.edu. Equal opportunity employer, M/F/D/V

Book Reviews

EMG: A Guide

Alex Lehn, Caroline Airey, Sasha Dionisio,
and John Cameron
ANZAN, February, 2014, 175 p., \$7.99

This e-book is an electronic bedside reference for clinical EMG and nerve conduction studies (NCS). It is designed to provide a quick review of basic neuromuscular anatomy and disease, as well as the basics of electrophysiological testing, in addition to acting as a useful reference when performing clinical studies.

The first chapter, "Introduction," is divided into five sections. The first section, "Basics," begins with a brief refresher of peripheral nerve anatomy and the principles of electrophysiological testing, including movies of typical normal and abnormal EMG findings. These are a particular strength of the text, as the ability to jump straight from a written description of a waveform to an audio and video recording makes the activity much easier to recognize. This section concludes with a brief summary of the timeline of EMG and NCS findings following peripheral nerve injury.

The second section, "Innervation," consists of a series of tables organized by anatomy, showing the nerve innervating each limb muscle and the degree to which individual roots contribute to that muscle's nerve. There is no accompanying text. The third section, "Normal Values," consists of a series of tables with typical normal values for NCS studies, some of which are segregated by age. The fourth section consists of a single page describing the calculation of the *F* ratio and a link to a self-contained app that performs this calculation.

The remaining chapters of the book are organized by anatomic regions: "Face and Neck," "Upper Limbs," "Shoulder and Torso," and "Lower Limbs." With the exception of the "Upper Limbs" chapter, each chapter is divided into three sections: 1) an introduction, with animated anatomical diagrams of each nerve and its branches followed by discussion of common sites of nerve entrapment and normal variants; 2) nerve conduction studies, with full-page descriptions of each NCS performed, including electrode location, distances, and normal values, and accompanied by clear and relevant photographs; and 3) EMG, with full-page descriptions

of each muscle typically used in EMG testing, including photographs of surface anatomy with needle insertion sites and anatomic drawings, as well as a text summary of muscle origins, insertions, innervation, needle insertion points, and activating maneuvers.

The "Upper Limbs" chapter contains the same information but subdivides it by nerve rather than topic, so that the introduction and NCS of the median nerve are covered before introducing the ulnar and radial nerves, and then the EMG of the entire limb closes out the chapter. Throughout the book, muscle and nerve names are hyperlinked, and selecting them takes the reader directly to the summary EMG or NCS page, respectively.

Overall, the text succeeds in its goal of being a media-rich, fast and easy bedside reference for clinical EMG/NCS studies, but it is limited by issues in organization and by omissions in its coverage. Although cases can be made for breaking down each anatomic region's chapter either by nerve or by study type, using one structure for the upper limbs and another for the other chapters is confusing. That being said, when the book is being used as a reference work (and an electronically searchable and hyperlinked one, at that), this is less of an issue.

More concerning is the uneven coverage of different topics. Although the lumbosacral plexus is covered in significant detail, with multiple figures and relevant clinical correlates, the brachial plexus appears in only a single figure at the beginning of the median nerve section and is not discussed in the text. On the other hand, the "Shoulder and Torso" chapter covers the innervation, musculature, and testing (both bedside and electrophysiological) of the shoulder region with remarkable completeness and clarity, and is highly recommended.

One final observation regarding this book is that, as an e-book, it may be easily revised and updated in the future. It is my hope that the authors will continue to develop this text into the easily accessible and clinically relevant reference and review book that it has every potential to become. ●

Julian Bragg
Midtown Neurology
Atlanta, GA

History of Exercise Physiology

Edited by Charles M. Tipton

Human Kinetics, 2014, 608 p., illus., \$119 (hardback)

ISBN: 9780736083690

This history book of exercise physiology is a more extensive sequel to the APS publication “People and Ideas” published in 2003 and also edited by C. M. Tipton. It is appropriate that Tipton serves as editor of these volumes given his rich history in the discipline – notably his pioneering development of a biology-driven graduate program in exercise physiology in the 1960s at the University of Iowa, his own research in the morphologic and physiological effects of exercise training, which spanned four decades, and his more recent extensive publication record in the history of the exercise sciences.

The book contains 22 chapters in two broad sections, with the first seven chapters exploring antiquity and the influence of selected laboratories in the emergence of exercise physiology as a scientific discipline. The remaining chapters are devoted primarily to each of the major organ systems and are written by leading investigators in these fields.

In general, I found this text to be an easy and highly informative read that goes well beyond the simple brokerage of information. Rather, most of the authors have managed to address the essential history of important findings in their specialty as well as to critically evaluate these findings retrospectively, so that one can trace the scientific evolution of a hypothesis. We are also provided some personal information about several of the key scientists and a few details on what drove their passion for their research. However, this personal aspect is one part of the book that I felt could have used more attention. Two especially appealing characteristics are the use of a table of “milestones of discovery” for each topic as well as the inclusion of numerous recordings of original findings from landmark experiments. Most chapters also do an excellent job of weaving together the history with present-day approaches to similar research problems.

From the antiquity and early history section we learn that several present-day major controversies have

extensive early histories. For example, debates over the preferred intensity of exercise training for maximal physiological benefit emerged in the form of the “Ying-Yang” theory in China during the Shang dynasty (1800–800 BCE) and also received extensive treatment in a *Physiological Reviews* publication on exercise training in 1933 written by A. H. Steinhaus. Furthermore, major influences on the cardioventilatory responses to exercise via central command vs. peripheral neural feedback from working muscle were outlined by German and Danish physiologists between 1880 and 1910, although circulating CO_2/H^+ was a stimulus thought to be common to both feedforward and feedback mechanisms in these early hypotheses. The rich early contributions of the Germans and Scandinavians to exercise physiology is detailed from the late 19th to the early 21st centuries. The timeline from Zuntz and Krogh through the three Danish musketeers (Christiansen, Asmussen, and Nielsen) is presented, culminating in a detailed account of the present-day, unparalleled accomplishments of the Copenhagen Muscle Research Laboratory spearheaded by the prolific contributions of the late Bengt Saltin (1935–2014), Bente Pedersen, Erik Richter, and their colleagues to exercise biochemistry, physiology, and endocrinology and especially to the training of scientists in exercise biology throughout the world. Finally, authors Neuffer and Tipton and then Bouchard and Malina provide a detailed accounting of the contributions to the field from the muscle biochemistry eras of the 1970s and 1980s, leading to applications of molecular biology to exercise science beginning in the late 1980s. They also outline how genomics was incorporated into exercise biology – beginning with twin studies of the early 1970s through animal selective breeding regimens and the Heritage Family studies of the 1990s and 2000s – aimed in large part at determining the heritable basis of individual differences in exercise performance and in the trainability of physiological traits.

The remaining 15 chapters in the second section of the text include 3 each devoted to the cardiovascular system and to muscle, with single chapters devoted to other organ systems and integrative chapters on oxygen transport, immunity, and temperature regulation.

I especially enjoyed the Sensorimotor chapter authored by Gardiner and Edgerton. They provide detailed accounts of research findings in healthy

and neurologically compromised humans together with evidence from chronically instrumented animal models to the development of such landmark concepts as exercise-induced patterns of muscle recruitment, feedback inhibition of central locomotor output, and exercise training “plasticity” effects – including training-induced modulation of reflex effects and the concept of “spinal learning.”

Among the three cardiovascular chapters, I found there to be significant overlap of content, especially that concerned with training effects. However, the writing style and emphasis of each of the sets of authors was sufficiently diverse that I carried away new appreciation for the rapidity with which newly developed techniques for studying the neural and hemodynamic aspects of the cardiovascular system were applied to exercising humans and chronically instrumented animals. These advances provided substantial progress over the past century in understanding the role of neural control and local vascular modulators in the regulation of cardiac output and its distribution during exercise and its modification with exercise training. The three chapters devoted to skeletal muscle were also sufficiently diverse so that the evolution of important concepts such as muscle atrophy and hypertrophy, lactate formation and utilization, and mitochondrial biogenesis were clearly outlined. The late Brian Whipp (1937-2011) played a large role as an investigator in the history of exercise/respiratory physiology. His treatment of this topic – together with wife Susan Ward – was

beautifully written (no doubt reflecting the authors’ UK education) and provided insights into the long-standing, controversial dilemma of exercise hyperpnea. Olympic champion/scientist Peter Snell together with cardiologist Ben Levine and the legendary Jere Mitchell provided thorough, objective, and insightful history from A. V. Hill to Saltin of the classic, ongoing debate concerning determinants of «overdot» $\text{VO}_{2\text{max}}$ and exercise performance. Finally, single chapters concerned with less studied topics in exercise physiology including renal, endocrine, and GI, as well as the integrative topic of immunology are necessarily limited in volume – but each of these authors provide thorough, excellent historical reviews. Temperature regulation is an integral part of exercise physiology. Authors Schneider and Moseley provide a detailed, century-long timeline of discovery from training-induced heat acclimation and set-point temperature regulation theories to heat shock proteins and from the eras of Sid Robinson and David Dill through to Carl Gisolfi.

Hearty congratulations to editor/historian Sir Charles “Tip” Tipton for envisioning and compiling this unique and important history. In turn, sincere thanks to the 36 eminent investigators who took time from their research programs to painstakingly research the history of their specialty so that we could all benefit from their insightful views of our profession’s heritage. ●

Jerome Dempsey
University of Wisconsin, Madison

Meetings & Congresses

2015

March 18-22

AD/PD 2015, Nice, France. *Information:* internet: <http://www2.kenes.com/adpd/Pages/Home.aspx>

March 19-21

The International Society for Evolution, Medicine, & Public Health Inaugural Meeting, Tempe, AZ. *Information:* internet: <https://sites.google.com/a/asu.edu/cemph/cemph-events/emph-society-meeting>

March 28 to April 1

2015 Experimental Biology, Boston, MA.

June 6-11

International Neuromodulation Society 12th World Congress, Montreal, Canada. *Information:* internet: <http://www.neuromodulation.com/in-congress>

August 3-7

14th International Congress on Amino Acids, Peptides and Proteins, Sao Paulo, Brazil. *Information:* Professor Gert Lubec, c/o Medical University of Vienna, Wahringer Gurtel 18, A-1090 Vienna, Austria; e-mail: icapp@meduniwien.ac.at; internet: <http://www.meduniwien.ac.at/icaap>

September 2-5

APS Conference: 14th International Conference on Endothelin: Physiology, Pathophysiology and Therapeutics, Savannah, GA. *Information:* internet: <http://www.endothelins.com/Conferences/ET-14/>.

September 9-12

APS Conference: Physiological Bioenergetics: From Bench to Bedside, Tampa, Florida

November 17-20

APS Conference: Cardiovascular, Renal and Metabolic Diseases: Physiology and Gender, Annapolis, Maryland

2016

April 2-6

2016 Experimental Biology, San Diego, CA.

July 21-25

12th International Congress of Cell Biology, Prague, Czech Republic. *Information:* internet: <http://www.cscb.cz/>

APS/NIDDK STEP-UP Undergraduate Summer Research Fellowships

Application deadline: February 15, 2015

<http://www.the-aps.org/stepup>

STEP-UP Fellows spend an average of 8-12 weeks in the laboratory of an established scientist conducting research in the NIDDK mission areas. This program is open to students from disadvantaged backgrounds, students from underrepresented racial and ethnic groups, and students with disabilities. Each Fellow receives a stipend plus additional funds for travel to present his or her research at the STEP-UP summer research symposium. Research hosts receive funds for student lab supplies. See the website for more details and apply online at <https://stepup.niddk.nih.gov/ApplyOnline.aspx>.

2014 Contributions

APS is extremely grateful for contributions received throughout the past year from individuals, corporations, foundations, and other organizations. Since membership dues only support a very small portion of our program initiatives, APS relies heavily on donations and grants to support efforts in education, diversity, and science policy and to fund numerous recognition awards for our members each and every year.

APS recognizes all donors contributing \$100 or more from January 1st thru December 5th of 2014.

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aps Intersociety Meeting:
Comparative Approaches to
Grand Challenges in Physiology



Meeting Program & Abstracts



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Canadian Society of Zoologists

Town and Country Resort and Convention Center
San Diego, California • October 5-8, 2014

View a meeting preview video at <http://bit.ly/CEPPreview>

the-aps.org/comparative



2014 APS Intersociety Meeting

Comparative Approaches to Grand Challenges in Physiology

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National Science Foundation

The Society for Integrative and Comparative Biology

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Comparative Biochemistry and Physiology Journal

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Exercise Medicine and Sport Sciences Initiative, University of California, Irvine

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2014 APS Intersociety Meeting
Comparative Approaches to Grand Challenges in Physiology

Sunday, October 5	Monday, October 6	Tuesday, October 7	Wednesday, October 8
2:00 PM Registration	7:00 AM Registration	7:30 AM Registration	7:30 AM Registration
	8:00—10:00 AM Concurrent Symposia Physiological Adaptations to Extremes: Providing Novel Animal Models for Investigating Health and Disease J.W. Hicks and T. Wang Genomics in Integrative and Comparative Physiology D. Crawford Frontiers in Insect Homeostasis-Advantages and Exploitation J.A.T. Dow and M. F. Romero	8:00—10:00 AM Concurrent Symposia Overcoming a Major Physiological Barrier: Adaptation from Saline to Freshwater Habitats G. Charmantier and C. Lee Challenges from the Very Beginning: Developmental Physiology, Epigenetics, and Critical Windows J. Eme and C. A. Mueller Comparative Gastrointestinal Physiology: From Genes to Animal Performance D. P. German	8:00—10:00 AM Concurrent Symposia Molecular and Physiological Features of Animal Diapause S. C. Hand New Perspectives on the Ecology and Evolution of Homeostasis L. B. Martin and H. A. Woods Linking Behavior and Physiology in Animal Navigation and Orientation J. Smolka and B. el Jundi
	10:30 AM—12:30 PM Concurrent Symposia Cardiorespiratory Physiology of Vertebrate Extremophiles G. R. Scott Diverse Approaches in Evolutionary Physiology T. Garland, Jr. Recent Ideas and Technological Advances in Comparative Epithelial Physiology M. Tresguerres and G. G. Goss	10:30 AM—12:30 PM Concurrent Symposia Responses to Global Change: Acclimatize, Adapt or Die G. Hofmann, M. Kelly, and T. G. Evans Evolutionary and Developmental Origins of Endothermy T. Owerkowicz and E. Dzialowski Determinants of Skeletal Muscle Diversity M. Azizi and L. P. Hernandez	10:30 AM—12:30 PM Concurrent Abstract Oral Presentations (<i>see daily schedule for more details</i>)
	12:30—3:30 PM Poster Presentations 3:30—5:30 PM Concurrent Abstract Oral Presentations (<i>see daily schedule for more details</i>)	12:30—3:30 PM Poster Presentations 3:30—5:30 PM Concurrent Abstract Oral Presentations (<i>see daily schedule for more details</i>)	12:30—3:30 PM Poster Presentations 3:30—6:00 PM Scholander Award Oral Presentations
4:50—5:00 PM Welcome B. Rees 5:00—6:00 PM Plenary Lecture: Participant: P. Schulte 6:00—8:00 PM Opening Reception	5:45—7:30 PM Workshop 1 Trainee Workshop: Non-Traditional Career Paths for Comparative Physiologists C. Williams and B. Rees	5:45—7:30 PM Workshop 2 The Challenge of Teaching Physiology in a Changing Environment: Innovation and Resources T.W. Ecay and K. Sweazea	7:00—10:00 PM Awards Banquet 8:30—9:30 PM Plenary Lecture: Participant: S. L. Chown

GENERAL INFORMATION

Location:

The 2014 APS Intersociety Meeting: Comparative Approaches to Grand Challenges in Physiology will be held October 5—8, 2014 at the Town & Country Resort and Conference Center, 500 Hotel Circle North, San Diego, CA 92108, telephone (619) 291-7131, FAX: (619) 294-4681.

Onsite Registration Hours:

Sunday, October 5.....2:00—8:30 PM
Monday, October 6.....7:00 AM—6:00 PM
Tuesday, October 7.....7:30 AM—6:00 PM
Wednesday, October 8.....7:30 AM—5:00 PM

On-Site Registration Fees:

APS Member.....\$450
Guest Society Member.....\$450
APS Retired Member.....\$300
Guest Society Member.....\$300
Nonmember.....\$550
Postdoctoral.....\$350
Student.....\$250

The registration fee includes entry into all scientific sessions, opening reception, and award banquet dinner.*

*Must get ticket for entry.

Payment Information:

Registrants may pay by institutional or personal check, traveler's check, MasterCard, VISA or American Express or in United States Dollars. Checks must be payable to "*The American Physiological Society*" and drawn on a United States bank payable in US dollars.

Student Registration:

Any student member or regularly matriculated student working toward a degree in one of the biomedical sciences is eligible to register at the student fee. Nonmember postdoctoral fellows, hospital residents and interns, and laboratory technicians do not qualify as students. Nonmember students who register onsite must provide a valid university student ID card. APS student members should present their current APS membership card indicating their student category status.

Postdoctoral Registration:

Any person who has received a Ph.D. degree in physiology or related field, **within four years** of this meeting, as attested to by the department head is eligible to register at the postdoctoral fee. **A statement signed by the department head must accompany the registration form and remittance when registering.**

Press:

Press badges will be issued at the APS registration desk, only to members of the working press and freelance writers bearing a letter of assignment from an

editor. Representatives of allied fields (public relations, public affairs, etc.) must register as nonmembers.

Program Objective:

Comparative physiology takes advantage of the diverse evolutionary histories and ecological settings of animals. By definition, comparative physiology is broad, spanning a variety of animal taxa occurring in diverse environmental settings, and studied at many levels of biological organization (from molecular physiology to physiological ecology). This breadth allows comparative physiology to (a) understand basic physiological processes and (b) identify novel mechanisms used by animals to solve specific physiological challenges. This meeting will draw comparative and evolutionary physiologists from around the world to present and discuss recent advances in animal physiology. The three and a half day meeting will feature fifteen symposia, two plenary lectures, two workshops, and multiple sessions for contributed abstracts as oral or poster presentations. This meeting will include the 2014 Scholander Award competition for young comparative physiologists, plus other trainee awards and activities.

Target Audience:

The intended audience for this meeting includes all professionals involved in teaching, research, and clinical fields related to comparative and evolutionary biology.

Photography is *not* permitted during the meeting sessions or in the poster room

Don't forget to join us at the Welcome Reception directly following the Opening Plenary Lecture

**Golden Foyer
6:00—8:00 PM**

SUNDAY, OCTOBER 5, 2014

Plenary Lecture I

1.0**PLENARY LECTURE**

Sun., 5:00–6:00 PM, Pacific Ballroom.

Sponsored by the Society for Integrative Comparative Biology.

5:00 PM

1.1 Opening the Black Box: How Physiology Links Genomes to Animal Function. **Patricia Schulte**. *Univ. of British Columbia, Vancouver, Canada.***MONDAY, OCTOBER 6, 2014**

Concurrent Symposia I

2.0**PHYSIOLOGICAL ADAPTATIONS TO EXTREMES: PROVIDING NOVEL ANIMAL MODELS FOR INVESTIGATING HEALTH AND DISEASE**

Mon., 8:00–10:00 AM, Pacific Ballroom.

Chairs:

James W. Hicks, *Univ. of California, Irvine.*
Tobias Wang, *Aarhus Univ., Denmark.*

8:00 AM

2.1 Is Physiology Redundant? Why Rodent Models in Biomedical Research are Failing. **Michael J. Joyner**. *Mayo Clinic.*

8:30 AM

2.2 Mammalian Hibernation as a Model of Disuse Osteoporosis: The Effects of Physical Inactivity on Bone Metabolism, Structure, and Strength. **Seth W. Donahue**. *Colorado State Univ.*

9:00 AM

2.3 Extreme Phenotypic Plasticity: How the Burmese Python Provides Clues to a Healthy Heart. **Leslie A. Leinwand**. *Univ. of Colorado, Boulder.*

9:30 AM

2.4 Hypoxia Tolerance in the Vertebrate Brain: Insights from Comparative Physiology. **Philip Bickler**, *Univ. of California, San Francisco.*

Concurrent Symposia II

3.0**GENOMICS IN INTEGRATIVE AND COMPARATIVE PHYSIOLOGY**

Mon., 8:00–10:00 AM, Pacific Salon 4/5.

Chair:

Douglas L. Crawford, *Univ. of Miami.*

8:00 AM

3.1 Evolutionary Genetics of Energetics: Effects of Mitochondrial-nuclear Interactions on Metabolism and Genome Evolution. **Kristi Montooth**, *Univ. of Nebraska, Lincoln.*

8:30 AM

3.2 Polymorphism in Oxygen Exchange Capacity and the Physiology of a Model Organism for Ecology. **James Marden**, *Penn. State Univ.*

9:00 AM

3.3 Comparative Physiological Genomics of Salinity Tolerance. **Andrew Whitehead**, *Univ. of California, Davis.*

9:30 AM

3.4 Transcriptomics as a Tool of Functional Genomics: Possibilities and Limitations with Hypoxia Response of Fish as a Case Study. **Mikko Nikinmaa and Jenni Prokkola**, *Univ. of Turku, Finland.*Join us at the Welcome
Reception following the
Opening Plenary Lecture

Concurrent Symposia III

4.0**FRONTIERS IN INSECT HOMEOSTASIS-ADVANTAGES AND EXPLOITATION**

Mon., 8:00–10:00 AM, Pacific Salon 6/7.

Chairs:

Julian A. T. Dow, *Univ. of Glasgow, UK.*
Michael F. Romero, *Mayo Clinic.*

8:00 AM

4.1 GPCR's Role in Regulation of Diuresis or Water Movement in Insects. **Patricia V. Pietrantoni**. *Texas A&M Univ.*

8:30 AM

4.2 PA1b, A Natural Peptidic Insecticidal Agent Against Vacuolar H⁺-ATPases. **Markus Huss**. *Univ. of Osnabrück, Germany.*

9:00 AM

4.3 Regulation of Ion Transport by Tyramine in the *Drosophila* Malpighian Tubule: Identification of Multiple Tyramine Receptors. **Edward M. Blumenthal**. *Marquette Univ.*

9:30 AM

4.4 Exploiting the Renal Homeostatic Mechanisms of Mosquitoes for Novel Vector Control. **Peter Piermarini**. *Ohio State Univ.*

Concurrent Symposia IV

5.0**CARDIORESPIRATORY PHYSIOLOGY OF VERTEBRATE EXTREMOPHILES**

Mon., 10:30 AM–12:30 PM, Pacific Ballroom.

Chair:

Graham R. Scott, *McMaster Univ., Hamilton, Canada.*

10:30 AM

5.1 Breathe Deep to Dive Deep: Respiratory Volumes in Penguins. **Paul Ponganis**. *Scripps Inst. of Oceanography, Univ. of California, San Diego.*

11:00 AM

5.2 Metabolic Strategies for Surviving the Winter in Hibernating Mammals. **Brian Barnes**. *Univ. of Alaska, Fairbanks.*

11:30 AM

5.3 Cardiorespiratory Performance in Anoxia Tolerant Fish and Reptiles. **Goran E. Nilsson**. *Univ. of Oslo, Norway.*

12:00 Noon

5.4 Evolution of Cardiorespiratory Physiology in High-altitude Mammals and Birds. **Graham R. Scott**. *McMaster Univ., Hamilton, Canada.*

Concurrent Symposia V

6.0**DIVERSE APPROACHES IN EVOLUTIONARY PHYSIOLOGY**

Mon., 10:30 AM–12:30 PM, Pacific Salon 4/5.

Chair:

Theodore Garland, Jr., *Univ. of California, Riverside.*

10:30 AM

6.1 Comparative Genetics and Genomics of Behavioral Phenotypes in Mice: Lessons for Evolutionary Physiology. **Scott A. Kelly**. *Ohio Wesleyan Univ.*

11:00 AM

6.2 Physiology and Genomics of the Evolution of Life Histories and Senescence in Garter Snakes. **Anne Bronikowski**. *Iowa State Univ.*

11:30 AM

6.3 Causes of Parallel Biochemical Adaptation: Insights from Hemoglobins of High-Altitude Vertebrates. **Jay F. Storz**. *Univ. of Nebraska, Lincoln.*

12:00 Noon

6.4 Molecular Evolution of Hormones and their Receptors. **Joe Thornton**. *Univ. of Chicago.*

DAILY SCHEDULE

Concurrent Symposia VI

7.0

RECENT IDEAS AND TECHNOLOGICAL ADVANCES IN COMPARATIVE EPITHELIAL PHYSIOLOGY

Mon., 10:30 AM—12:30 PM, Pacific Salon 6/7.

Chairs:

Martin Tresguerres, *Scripps Inst. of Oceanography, Univ. of California, San Diego.*
Greg G. Goss, *Univ. of Alberta, Canada.*

10:30 AM

7.1 Rainbow Trout Use Acid Sensing Ion Channels (ASICs) for Na⁺ Uptake in Dilute Freshwater. **Greg G. Goss**, *Univ. of Alberta, Canada.*

11:00 AM

7.2 Fish Intestinals HCO₃⁻ Secretion: From Molecules to the Oceanic Inorganic Carbon Cycle. **Martin Grosell**, *RSMAS, Univ. of Miami.*

11:30 AM

7.3 The Na-K-Cl Cotransporter: Recent Advances in Structure, Function, and Regulation. **Michelle Monette**, *Western Connecticut State Univ.*

12:00 Noon

7.4 Evolutionary Conserved Mechanisms for Acid/base Sensing. **Martin Tresguerres**, *Scripps Inst. of Oceanography, Univ. of California, San Diego.*

Poster Presentation

8.0

METABOLISM, ENERGETICS, AND NUTRITION

Mon., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

1

8.1 Investigation of Algal Feeding Preferences of the Solar-powered Sea Slug, *Elysia clarki*. **M. Arnette***, **A. Shelton**, and **J. Genz**, *Univ. of West Georgia.*

2

8.2 Light-induced Oxidative Stress and Defense in the Mantle of a Giant Clam. **B. Ching**, **K. C. Hiong**, **M. V. Boo**, **Y. L. Choo**, **J. L. Y. Ong**, **X. L. Chen**, **Y. R. Chng**, **W. P. Wong**, **S. F. Chew**, and **Y. K. Ip**, *Natl. Univ. of Singapore, and Natl. Inst. of Education, Singapore.*

3

8.3 Physiological Responses to Environmental Stress in a Mollusc: Why is Being a Hybrid an Advantage? **K. Alter***, **A. Morash**, **S. Andrewartha**, **N. Elliott**, and **P. Frappell**, *CSIRO, Univ. of Tasmania, Australia.*

4

8.4 Mitochondrial Mechanisms of Hypoxia Tolerance in Marine Bivalves. **A. Ivanina**, and **I. Sokolova**, *Univ. of North Carolina at Charlotte.*

5

8.5 Withdrawn.

6

8.6 Scorpion Burrow Structure and Ventilation. **A. Adams***, **S. Turner**, **P. Berliner**, and **B. Pinshow**, *Ben-Gurion Univ., Israel, and SUNY-ESF.*

7

8.7 The Effect of Humidity on the Metabolic Rate of *Gromphadorhina portentosa*. **C. Toogood**, and **H. L. Contreras**, *Univ. of La Verne.*

8

8.8 Activation of cGMP-dependent Protein Kinase Reduces *Drosophila* S2 Cell Injury Caused by Anoxia and Oxidative Stress. **O. Makhnyeva***, **K. Dawson-Scully**, and **S. Milton**, *Florida Atlantic Univ.*

9

8.9 Metabolism and Locomotion of Anoxic *Drosophila*. **J. Campbell***, **V. Callier**, **S. Hand**, and **J. F. Harrison**, *Arizona State Univ., and Louisiana State Univ.*

Poster Board

10

8.10 Small RNA Regulation of Diapause in the Flesh Fly, *Sarcophaga bullata*. **J. Reynolds**, and **D. Denlinger**, *Ohio State Univ.*

11

8.11 How do You Like Your eggs? Egg Cannibalism and Digestibility in the California Grunion, *Leuresthes tenuis* (Teleostei: Atherinopsidae). **A. R. Braciszewski**, **A. Carrillo**, **M. H. Horn**, **A. Carter**, and **D. P. German**, *Univ. of California, Irvine, and California State Univ., Fullerton.*

12

8.12 The Effect of Diet and Intestinal Microbiome Manipulation on Intestinal Cellular Pathways. **C. Bucking**, **C. LeMoine**, **P. Craig**, **T. Moon**, and **A. Poulain**, *York Univ., Toronto, and Univ. of Ottawa, Canada.*

13

8.13 Fasting-induced Morphological Reorganization of the Colon May Not Drive Concomitant Changes in the Microbiome. **C. Passemant**, **K. Kohl**, **D. Meyerholz**, and **M. McCue**, *St. Mary's Univ., Univ. of Utah, and Univ. of Iowa.*

14

8.14 Glucose Can Fuel Metabolism in RBCs from Normoglycemic But Not Hypoglycemic Fish. **W. Driedzic**, **K. Clow**, and **C. Short**, *Memorial Univ. of Newfoundland, Canada.*

15

8.15 Activity of the 20S Proteasome is Not Correlated with the Expression of Oxygen-binding Proteins in Antarctic Notothenioid Fishes. **C. Oldham**, and **K. O'Brien**, *Univ. of Alaska, Fairbanks.*

16

8.16 Mechanisms and Costs of Mitochondrial Thermal Acclimation in the Common Killifish *Fundulus heteroclitus*. **D. Chung***, and **P. Schulte**, *Univ. of British Columbia, Vancouver, Canada.*

17

8.17 Mitochondrial Responses to Sustained and Intermittent Hypoxia in Killifish *Fundulus heteroclitus*. **S. N. N. Du***, and **G. R. Scott**, *McMaster Univ., Hamilton, Canada.*

18

8.18 How Tolerant is an Anoxia-tolerant Vertebrate? **S. Lefevre***, **M-K. Torp**, **J. A. W. Stecyk**, and **G. E. Nilsson**, *Univ. of Oslo, Norway, and Univ. of Alaska, Anchorage.*

19

8.19 Paradoxical Anaerobism in Desert Pupfish. **F. van Breukelen**, and **S. Hillyard**, *Univ. of Nevada, Las Vegas.*

20

8.20 Highly Efficient Mitochondria Fuel Bluefin Tuna Red Muscle Within Distinct Temperature Ranges. **M. Jastroch**, **J. Treberg**, **M. Brand**, and **B. Block**, *Helmoltz Ctr., Munich, German Res. Ctr. for Environmental Hlth., Germany, Univ. of Manitoba, Winnipeg, Canada, The Buck Inst. for Res. on Aging, Novato, CA, and Stanford Univ.*

21

8.21 Changes in Expression of Two Genes Involved in Arginine Synthesis, and Concentrations of Arginine and Nitric Oxide, in an Aestivating African Lungfish. **Y. R. Chng**, **B. Ching**, **J. L. Y. Ong**, **X. L. Chen**, **W. P. Wong**, **S. F. Chew**, and **Y. K. Ip**, *Natl. Univ. of Singapore, and Natl. Inst. of Education, Singapore.*

22

8.22 The Effects of Temperature on the Metabolic Fate of Lactate During Recovery from Anoxia in the Painted Turtle. **C. Hill**, and **D. Warren**, *St. Louis Univ.*

23

8.23 MsrA as an Important Neuroprotective Mechanism in the Anoxia Tolerant Model: *Trachechys scripta elegans*. **M. Reiterer**, and **S. Milton**, *Florida Atlantic Univ.*

Poster Board

- 24 **8.24** Cervical Osteoderms Reveal Pattern of Whole Body Growth in Juveniles of the American Alligator. **D. Vasconcellos, T. Owerkowitz, J. Eme, J. Blank, R. Elsey, and J. Hicks.** *California State Univ., San Bernardino, McMaster Univ., Hamilton, Canada, California Poly, San Luis Obispo, Rockefeller Wildlife Refuge, Grand Chenier, LA, Univ. of California, Irvine.*
- 25 **8.25** Phenotypic Plasticity Across the Annual Cycle in a Migratory Bird. **P. Schaeffer, K. Corde, A. Hamilton, K. Demoranville, and J. Huss.** *Miami Univ., and Beckman Res. Inst., Duarte, CA.*
- 26 **8.26** Bats and Birds Share Digestive Adaptations to an Aerial Lifestyle. **E. Price, A. Brun, E. Caviedes-Vidal, and W. Karasov.** *Univ. of Wisconsin, Madison, and Univ. Natl. de San Luis, Argentina.*
- 27 **8.27** Withdrawn.
- 28 **8.28** Heart Rate Dynamics in a Marsupial Hibernator. **S. Swoap, G. Kortner, and F. Geiser.** *Williams Coll., Williamstown, MA, and Univ. of New England.*
- 29 **8.29** Apoptotic Regulation During Mammalian Hibernation. **M. Treat*, and F. van Breukelen.** *Univ. of Nevada, Las Vegas.*
- 30 **8.30** A Cytosolic Protein Factor Located in Multiple Tissues in the Naked Mole-rat Protects the Proteasome from Inhibition and Activates the Proteasome in Other Species. **K. Rodriguez*, P. Osmulski, A. Pierce, S. Weintraub, M. Gaczynska, and R. Buffenstein.** *Univ. Texas Hlth. Sci. Ctr., San Antonio, and Univ. of Texas Med. Branch Galveston.*
- 31 **8.31** Lipids and Myoglobin: New Insights from Non-model Species. **S. Kanatous, and A. Schlater.** *Colorado State Univ., and McMaster Univ., Hamilton, Canada.*
- 32 **8.32** Thyroid Gland Remains Responsive to Thyroid Stimulating Hormone with Sensitivity Increasing with Fasting Duration in a Prolonged Fasted Mammal. **B. Martinez*, D. Somo, D. Ensminger, H. Peck, D. Lee, D. Crocker, and R. Ortiz.** *Univ. of California, Merced, Sonoma State Univ., Rohnert Park, CA.*
- 33 **8.33** Purine Nucleoside Phosphorylase Activity in Erythrocytes from Bottlenose Dolphins (*Tursiops truncatus*) in Response to Breath-hold Diving and Exercise. **I. Del Castillo Velasco Martinez, R. I. Lopez-Cruz, C. J. Hernandez-Camacho, L. C. Mendez-Rodriguez, and T. Zenteno-Savin.** *CIBNOR, Baja California Sur, Mexico, and IPN-CICIMAR, Baja California Sur, Mexico.*
- 34 **8.34** Opposite Trends in Over-summer Mass Change: Post-parturient Weddell Seals Gain Weight While Non-reproductive Females Lose Mass and Condition. **A. Kirkham*, R. Beltran, M. Shero, and J. Burns.** *Univ. of Alaska, Anchorage, and Univ. of Alaska, Fairbanks.*
- 35 **8.35** Linking Caloric Intake to Growth and Blubber Deposition in Walrus Calves. **S. Noren, M. Udevitz, and C. Jay.** *Univ. of California, Santa Cruz, and U. S. Geological Survey, Anchorage.*
- 36 **8.36** Lactating Mice Increase Villus Surface Area Thru Increased Enterocyte Width in Response to Low Protein Diets. **K. Short, M. Cook, and E. Derrickson.** *Loyola Univ., Baltimore.*
- 37 **8.37** Macrophage Infiltration in the Adipose Tissue of Dairy Cows During Negative Energy Balance. **G. A.**

Poster Board

- Contreras, E. Kabara, J. Brester, L. Neuder, and M. Kiupel.** *Michigan State Univ.*
- 38 **8.38** The Effect of Circadian Organization on Energy Use and Immune Function in C57B Mice. **C. Richardson, X. Jean, K. Xiao, B. Dumont, and F. Davis.** *Northeastern Univ.*

Poster Presentation

9.0

FIELD PHYSIOLOGY

Mon., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 39 **9.1** Entrainment of Circatidal Gene Expression Rhythms in California Mussel *Mytilus californianus*. **J. Lin, and A.Y. Gracey.** *Univ. of Southern California.*
- 40 **9.2** The Stress Response is Associated with Both Cortisol and Aldosterone Release in Marine Mammals. **C. Champagne, M. Tift, D. Houser, and D. Crocker.** *Natl. Marine Mammal Fdn., San Diego, and Sonoma State Univ., Rohnert Park, CA.*
- 41 **9.3** Persistent Tissue Differences in Fatty Acid Profiles of Weddell Seals (*Leptonychotes weddellii*) Reflect Tissue Roles. **L. Pearson*, D. Costa, and J. Burns.** *Univ. of Alaska, Fairbanks, Univ. of California, Santa Cruz, and Univ. of Alaska, Anchorage.*

Poster Presentation

10.0

EVOLUTIONARY PHYSIOLOGY

Mon., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 42 **10.1** Tales from the Precambrian: TNF-induced Apoptosis Remains Unchanged 550 million Years Later. **S. Quistad, A. Stotland, K. Barott, C. Smurthwaites, B. Hilton, J. Grasis, R. Wolkowicz, and F. Rohwer.** *San Diego State Univ. and Univ. of California, San Diego.*
- 43 **10.2** Muscular Dystrophy Genes and the Early Metazoan Transition from Dynein- to Myosin-Powered Locomotion. **A. Mead, A. Malik, R. Krishnakutty, and H. Stedman.** *Univ. of Vermont, and Univ. of Pennsylvania.*
- 44 **10.3** Evolution of the Urea Transporter Family in Vertebrates. **C. LeMoine, and P. Walsh.** *Univ. of Ottawa, Canada.*
- 45 **10.4** Differential Gene Regulation Underlies Responses to Acute Heat Stress and Local Adaptation in *Tigriopus californicus*. **S. Tangwancharoen, G. Moy, and R. Burton.** *Univ. of California, San Diego.*
- 46 **10.5** Relationships Between Mitochondrial Thermal Performance and Heat Tolerance Among Populations of the Intertidal Copepod *Tigriopus californicus*. **A. Harada, and R. Burton.** *Scripps Inst. of Oceanography, La Jolla.*
- 47 **10.6** Obesity-induced Cardiac Dysfunction in Starvation-Selected *Drosophila*. **C. Hardy*, R. Birse, M. Wolf, and A. Gibbs.** *Univ. of Nevada, Las Vegas, Sanford Burnham Med. Res. Inst., La Jolla, and Duke Univ.*
- 48 **10.7** Sex and Nutrient Effects on Energy Allocation Among Body Parts Within an Individual. **G. Davidowitz.** *Univ. of Arizona.*
- 49 **10.8** Mitochondrial Genomes and Oxidative Phosphorylation from Populations of *Fundulus heteroclitus* Distributed Along a Thermal Cline. **J. Nunez*, T. Baris, D. Crawford, and M. Oleksiak.** *Univ. of Miami.*

DAILY SCHEDULE

Poster Board

- 50 **10.9** Reciprocal Osmotic Challenges Reveal Mechanisms of Divergence in Phenotypic Plasticity in the Killifish *Fundulus heteroclitus*. **R. Brennan, F. Galvez, and A. Whitehead.** *Univ. of California, Davis, and Louisiana State Univ.*
- 51 **10.10** The Role of Lipids and Proteins in the Diet of Common Hamsters (*Cricetus cricetus*) on Hibernation and Reproductive Success. **M. Weitten, J.-P. Robin, and C. Habold.** *Univ. de Strasbourg, France.*
- 52 **10.11** Phylogenetic Correlation Between Maximum Oxygen Consumption and Home Range Area Among Species of Mammals. **R. Albuquerque, and T. Garland.** *Univ. of California, Riverside.*
- 53 **10.12** Effects of Voluntary Wheel Running on Body Mass and Composition in Selectively-bred High-Runner Mice. **L. Hiramatsu*, G. Claghorn, and T. Garland.** *Univ. of California, Riverside.*

**Photography is *not* permitted
during the meeting sessions or
in the poster room**

Poster Presentation

11.0 HUMAN NUTRITION AND PHYSIOLOGY EDUCATION

Mon., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 54 **11.1** Gender Difference in Nutritional Status of a Primary School Children of Bangladesh. **M. A. Salam, and M. U. A. Khan.** *THC Araihaajar, Narayanganj, Bangladesh, and Noakhali Med. Coll., Bangladesh.*
- 55 **11.2** Nutritional Status Of MBBS Students of a Selected Government Medical College of Bangladesh. **M. U. A. Khan, and M. Nasiruzzaman.** *Noakhali Med. Coll., Bangladesh.*
- 56 **11.3** Is the Growth of Bangladeshi School Children Proportionate to Their Age? **R. U. Ahmed, M. U. A. Khan, and M. Abedin.** *Dhaka Med. Coll. Hosp., Bangladesh, and Noakhali Med. Coll., Bangladesh.*
- 57 **11.4** Gender Difference in the Result of 1st Term Physiology Examination of a Government Medical College of Bangladesh. **M. U. A. Khan, and A. Salam.** *Noakhali Med. Coll., Bangladesh.*
- 58 **11.5** Use of Fellow Examinee as Subject in Observed Structured Practical Examination (OSPE) in 1st Term Physiology Examination. **M. U. A. Khan, and D. Hossain.** *Noakhali Med. Coll., Bangladesh.*
- 59 **11.6** Effectiveness of Frequent Discussion Embedded Interactive Lecture (FDEIL) in Learning Medical Science. **M. U. A. Khan, A. Abbasi, and M. A. Salam.** *Noakhali Med. Coll., Bangladesh, and THC Araihaajar, Narayanganj, Bangladesh.*
- 60 **11.7** Reading the Primary Literature: Skills of First-year and Senior Undergraduate Biology Majors. **J. Blank, K. McGaughey, E. Keeling, and J. Scaramozzino.** *California Poly. State Univ., San Luis Obispo.*

Oral Abstract Presentations

12.0 CARDIOVASCULAR AND RESPIRATORY PHYSIOLOGY

Mon., 3:30—5:30 PM, Pacific Ballroom.

Chairs: **Michael Hedrick,** *California State Univ., East Bay.*
Holly Shields, *Univ. of Manchester, UK.*

- 3:30 PM **12.1** Getting to the Heart of Plasma-accessible Carbonic Anhydrase in Fish. **Sarah Alderman.** *Univ. of British Columbia, Vancouver, Canada.*
- 3:45 PM **12.2** Electrical Activation and Repolarization Sequence in the Rainbow Trout Heart. **Holly Shields.** *Univ. of Manchester, UK.*
- 4:00 PM **12.3** Antioxidant Defense Mechanisms in the Red Blood Cells of Longhorn Sculpin (*Myoxocephalus octodecemspinosus*) in Response to Hypoxia and Rapid Re-oxygenation. **Johanne M. Lewis.** *Georgia Southern Univ.*
- 4:15 PM **12.4** Heart Rate and Blood Oxygen Depletion in Loggerhead Turtles, *Caretta caretta*. **Cassandra Williams.** *Univ. of California, Irvine.*
- 4:30 PM **12.5** Mechanisms for Suppression of Oxygen Delivery and Consumption in Hibernating Brown Bears. **Angela Fago.** *Aarhus Univ., Denmark.*
- 4:45 PM **12.6** Respiratory Physiology in Cetaceans. **Andreas Fahlman.** *Texas A&M Univ.*
- 5:00 PM **12.7** The Evolution of Unidirectional Pulmonary Airflow. **C. G. Farmer.** *Univ. of Utah.*
- 5:15 PM **12.8** Nature Versus Nurture: Hypoxic Cardiovascular and Respiratory Responses in Bar-headed Geese (*Anser indicus*) and Related Waterfowl. **Sabine Lague.** *Univ. of British Columbia, Vancouver, Canada.*

Oral Abstract Presentations

13.0 EVOLUTIONARY PHYSIOLOGY

Mon., 3:30—5:30 PM, Pacific Salon 4/5.

Chair: **Frank van Breukelen,** *Univ. of Nevada, Las Vegas.*

- 3:30 PM **13.1** Evidence of Countergradient Variation and Adaptive Slow Growth Rate in a Marine Isopod (*Idotea baltica*) Locally Adapted to Low Salinity. **Hannah Wood.** *Univ. of Gothenburg, Sweden.*
- 3:45 PM **13.2** Hybrid Breakdown and Physiological Compensation in Gene Expression in the Copepod *Tigriopus californicus*. **Ronald Burton.** *Univ. of California, San Diego.*
- 4:00 PM **13.3** Variation Between Stickleback Populations in Cold Tolerance and Mechanisms of Freshwater Ionoregulation. **Taylor Gibbons.** *Univ. of British Columbia, Vancouver, Canada.*
- 4:15 PM **13.4** Transgenerational Inheritance of a Stress Proteome in the Least Killifish, *Heterandria formosa*, Exposed to Copper During Early Life Stage. **Frédéric Silvestre.** *Univ. of Namur, Belgium.*
- 4:30 PM **13.5** Rapid Evolution of Physiology in Laboratory Populations of *Drosophila*. **James N. Kezos.** *Univ. of California, Irvine.*
- 4:45 PM **13.6** Physiological Synergism and Antagonism in the Evolution of Life Histories. **Goggy Davidowitz.** *Univ. of Arizona, Tucson.*

5:00 PM **13.7** Does MMR Regulate of Life-history Traits? Correlated Changes in Mice Selected for Mass-independent MMR. **Cynthia Downs**. *Univ. of Nevada, Reno.*

5:15 PM **13.8** Molecular Evolution and Adaptation of Insulin-like/TOR Signaling Across Amniotes. **Suzanne McGaugh**. *Univ. of Minnesota, St. Paul.*

Oral Abstract Presentations

14.0 METABOLISM, ENERGETICS, AND PERFORMANCE

Mon., 3:30—5:30 PM, Pacific Salon 6/7.

Chairs: **Sarah Andrewartha**, *Commonwealth Sci. & Ind. Res. Org., Hobart, Australia.*
Jonathon Stillman, *Univ. of California, Berkeley.*

3:30 PM **14.1** Ontogenetic and Interspecific Metabolic Scaling in Insects. **James Maino**. *Univ. of Melbourne, Australia.*

3:45 PM **14.2** Combining X-ray Synchrotron Imaging of Amber Fossils with Body Size Changes in the Insect Fossil Record to Elucidate the Effect of Atmospheric Oxygen on Paleophysiology. **John VandenBrooks**. *Midwestern Univ.*

4:00 PM **14.3** Diel Changes in Coral Metabolism: Potential Regulation by Phosphorylation. **Lauren Linsmayer**. *Univ. of California, San Diego.*

4:15 PM **14.4** Specific Dynamic Action in Decapod Crustaceans. **Iain McGaw**. *Memorial Univ. of Newfoundland, Canada.*

4:30 PM **14.5** Real-time Physiology: Can it Assist Aquaculture Productivity? **Sarah Andrewartha**. *Commonwealth Sci. & Ind. Res. Org., Hobart, Australia.*

4:45 PM **14.6** Temperature and Acidification Variability Reduce Physiological Performance in the Intertidal Zone Porcelain Crab *Petrolisthes cinctipes*. **Adam Paganini**. *San Francisco State Univ.*

5:00 PM **14.7** What Limits Performance in Wild Pacific Bluefin Tuna? **Barbara Block**. *Stanford Univ.*

5:15 PM **14.8** Validation of the Relationship Between 3-dimensional Body Acceleration and Oxygen Consumption in Trained Steller Sea Lions (*Eumetopias jubatus*) Diving with Increased Oxygen Depletion. **Beth L. Volpov**. *Deakin Univ., Burwood, Australia.*

Oral Abstract Presentations

15.0 OSMOTIC AND ION REGULATION: JUNCTIONS AND TRANSPORTERS

Mon., 3:30—5:30 PM, Royal Palms Salon 1/2.

Chair: **David Goldstein**, *Wright State Univ.*

3:30 PM **15.1** A Role for Septate Junction Proteins in the Regulation of Salt and Water Balance in Larval Mosquito (*Aedes aegypti*). **Sima Jonusaitė**. *York Univ., Toronto, Canada.*

3:45 PM **15.2** Tricellular Tight Junction Proteins and Their Contribution to Paracellular Occlusion in the Fish Gill Epithelium. **Dennis Kolosov**. *York Univ., Toronto, Canada.*

4:00 PM **15.3** Abundance and Localization of Branchial Claudins in Rainbow Trout (*Oncorhynchus mykiss*) and Implications in Hypoosmoregulation. **Joanna Bujak**. *Univ. of Arkansas.*

4:15 PM **15.4** Hypotonicity Stimulates K⁺ Flux Through the WNK-SPAK/OSR1 Kinase Cascade and the Ncc69 Sodium-Potassium-2-Chloride Cotransporter in the *Drosophila* Renal Tubule. **Aylin Rodan**. *Univ. of Texas, Southwestern.*

4:30 PM **15.5** Anuran-specific Aquaporin 2 Homolog in a Urodele, Evidence for Early Gene Duplication in Amphibian Evolution. **Stanley Hillyard**. *Univ. of Nevada, Las Vegas.*

4:45 PM **15.6** Sequencing and Gene Expression of Aquaporin-9 in Freeze Tolerant Cope's Gray Treefrogs. **Brian Stogsdill**. *Wright State Univ.*

5:00 PM **15.7** Gene Expression of Three Isoforms of Urea Transporter in an Aestivating African Lungfish. **Biyun Ching**. *Natl. Univ. of Singapore.*

5:15 PM **15.8** Dehydration and Thirst of Hydrophiline Sea Snakes. **Harvey Lillywhite**. *Univ. of Florida, Gainesville.*

Workshop I

16.0

TRAINEE WORKSHOP: NON-TRADITIONAL CAREER PATHS FOR COMPARATIVE PHYSIOLOGISTS

Mon., 5:45—7:30 PM, Pacific Ballroom.

Chairs: **Cassandra Williams**, *Univ. of California, Irvine.*
Bernard Rees, *Univ. of New Orleans.*

5:45 PM **16.1** The Soft Money Research Position: How Does it Work? **Shawn Noren-Kramer**. *Univ. of California, Santa Cruz.*

6:05 PM **16.2** Science for the Public! Careers in Science Centers and Museums. **Karen Kalumuck**. *Emeritus, San Francisco Exploratorium.*

6:25 PM **16.3** Taking the Road Less Traveled: Non-Governmental Organizations. **Dorian Houser**. *Natl. Marine Mammal Fdn., San Diego.*

6:45 PM **16.4** The Undergraduate University Path. **Scott Kirkton**. *Union Coll.*

7:05 PM **16.5** The Transition to Industry. **Robert Swezey**. *SRI Intl., Menlo Park, CA.*

TUESDAY, OCTOBER 7, 2014

Concurrent Symposia VII

17.0

OVERCOMING A MAJOR PHYSIOLOGICAL BARRIER: ADAPTATION FROM SALINE TO FRESHWATER HABITATS

Tues., 8:00—10:00 AM, Pacific Ballroom.

Sponsored by The Crustacean Society.

Chairs: **Guy Charmantier**, *Univ. of Montpellier 2, France.*
Carol Lee, *Univ. of Wisconsin, Madison.*

8:00 AM **17.1** Rapid Evolution of Ionic Regulation During Saline to Freshwater Habitat Invasions. **Carol Lee**. *Univ. of Wisconsin, Madison.*

8:30 AM **17.2** Colonization of Freshwater Habitats from the Marine Environment: Lessons from Stickleback and Killifish. **Patricia Schulte**. *Univ. of British Columbia, Canada.*

DAILY SCHEDULE

9:00 AM **17.3** Osmoregulation in the Anadromous Lamprey *Petromyzon marinus*. **Jonathan M. Wilson**. *Wilfrid Laurier Univ., Waterloo, Canada*.

9:30 AM **17.4** Evolutionary Transition to Freshwater in the Shrimp *Macrobrachium amazonicum*: Ecophysiological Adaptations. **Guy Charmantier**. *Univ. of Montpellier 2, France*.

Concurrent Symposia VIII

18.0 CHALLENGES FROM THE VERY BEGINNING: DEVELOPMENTAL PHYSIOLOGY, EPIGENETICS, AND CRITICAL WINDOWS

Tues., 8:00—10:00 AM, Pacific Salon 4/5.

Sponsored by the Comparative Biochemistry and Physiology Journal.

Chairs: **John Eme**, *McMaster Univ., Hamilton, Canada*.
Casey A. Mueller, *McMaster Univ., Hamilton, Canada*.

8:00 AM **18.1** Critical Windows in Animal Development: Stressor Dose, Effect Size and Experimental Design. **Casey A. Mueller**. *McMaster Univ., Hamilton, Canada*.

8:30 AM **18.2** Noisy Embryos? The Potential Evolutionary Importance of Variation in the Timing of Developmental Events. **Simon Rundle**. *Univ. of Plymouth, UK*.

9:00 AM **18.3** Mitigating the Risks Associated with Accelerated or Deficient Perinatal Growth. **Robert D. Roghair**. *Univ. of Iowa*.

9:30 AM **18.4** Epigenetic Influences in Developmental Comparative Anatomy and Physiology. **Warren Burggren**. *Univ. of North Texas*.

Concurrent Symposia IX

19.0 COMPARATIVE GASTROINTESTINAL PHYSIOLOGY: FROM GENES TO ANIMAL PERFORMANCE

Tues., 8:00—10:00 AM, Pacific Salon 6/7.

Sponsored by the Society for Integrative Comparative Biology and Comparative Biochemistry and Physiology Journal.

Chair: **Donovan P. German**, *Univ. of California, Irvine*.

8:00 AM **19.1** How the Gut Limits Nutrition, and the Influence of this on the Ecology and Evolution of an Insect Herbivore, the Grasshopper. **Fiona Clissold**. *Univ. of Sydney, Australia*.

8:30 AM **19.2** Evolutionary and Molecular Mechanisms Underlying Intestinal Flexibility for Snakes. **Stephen M. Secor**. *Univ. of Alabama, Tuscaloosa*.

9:00 AM **19.3** The Role of Gut Microflora in the Nutrition of Marine Herbivorous Fishes. **Kendall D. Clements**. *Univ. of Auckland, New Zealand*.

9:30 AM **19.4** Amylase Genetics and Biochemistry Underlie a Digestive Specialization in Prickleback Fishes. **Donovan P. German**. *Univ. of California, Irvine*.

Photography is *not* permitted during the meeting sessions or in the poster room

Concurrent Symposia X

20.0 RESPONSES TO GLOBAL CHANGE: ACCLIMATIZE, ADAPT OR DIE

Tues., 10:30 AM—12:30 PM, Pacific Ballroom.

Sponsored by the Society for Experimental Biology, Division of Comparative Physiology & Biochemistry.

Chairs: **Gretchen Hofmann**, *Univ. of California, Santa Barbara*.
Morgan Kelly, *Louisiana State Univ.*
Tyler G. Evans, *California State Univ., East Bay*.

10:30 AM **20.1** Trait-based Approaches to Predicting the Responses of Species to Global Change. **Sarah Diamond**. *Case Western Reserve Univ.*

11:00 AM **20.2** Ocean Acidification Effects on Temperate Rockfishes. **Cheryl Logan**. *California State Univ., Monterey Bay*.

11:30 AM **20.3** Mechanistic Overlap Between Plastic and Evolved Responses to Heat Stress. **Morgan Kelly**. *Louisiana State Univ.*

12:00 Noon **20.4** Comparing Physiological Plasticity Vs. Evolutionary Adaptation Vs. Phylogenetic Constraint on Species Distributions; *Drosophila* and Beyond. **Ary Hoffmann**. *Univ. of Melbourne, Australia*.

Concurrent Symposia XI

21.0 EVOLUTIONARY AND DEVELOPMENTAL ORIGINS OF ENDOTHERMY

Tues., 10:30 AM—12:30 PM, Pacific Salon 4/5.

Sponsored by Australian & New Zealand Society for Comparative Physiology and Biochemistry

Chairs: **Tomasz Owerkowicz**, *California State Univ.*
Edward Dzialowski, *Univ. of North Texas*.

10:30 AM **21.1** Thermoregulation and Thermogenesis in Reptiles. **Glenn Tattersall**. *Brock Univ., St. Catherine's, Canada*.

11:00 AM **21.2** Development of Endothermy in Marsupial and Placental Mammals. **Sarah Andrewartha**. *Univ. of Tasmania, Australia*.

11:30 AM **21.3** Testing Competing Hypotheses of the Evolution of Endothermy. **Marek Konarzewski**. *Univ. of Bialystok, Poland*.

12:00 Noon **21.4** Development of Endothermy in Altricial and Precocial Birds. **Edward Dzialowski**. *Univ. of North Texas*.

Concurrent Symposia XII

22.0 DETERMINANTS OF SKELETAL MUSCLE DIVERSITY

Tues., 10:30 AM—12:30 PM, Pacific Salon 6/7.

Sponsored by the Comparative Biochemistry and Physiology Journal.

Chairs: **Manny Azizi**, *Univ. of California, Irvine*.
L. Patricia Hernandez, *George Washington Univ.*

10:30 AM **22.1** Evolutionary Selection of Myofibrillar Protein Isoforms for Specific Muscle Function. **Peter Reiser**. *Ohio State Univ.*

11:00 AM **22.2** Comparative Physiology of Body Weight-sensitive Skeletal Muscle Plasticity. **Ruud Schilder**. *Penn State Univ.*

11:30 AM **22.3** Cardiac Myosin Alpha and Ventricular Hypertrophy Protect Ground Squirrels in Hibernation. **Bryan Rourke**. *California State Univ., Long Beach.*

12:00 Noon **22.4** The Diversity and Evolution of Locomotor Muscle Properties in Anurans. **Henry Astley**. *Georgia Inst. of Tech.*

Poster Presentations

23.0 CARDIOVASCULAR AND RESPIRATORY PHYSIOLOGY

Tues., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

61 **23.1** Do *Drosophila* Larvae Experience Functional Oxygen Limitation Late in the Instar? **J. Harrison, and V. Callier**. *Arizona State Univ.*

62 **23.2** A Study on Embryonic Development Rate and Cardiorespiratory Performance of the Norway Lobster, During Acute Exposure to Elevated PCO₂, Manganese and Hypoxia. **H. Styf, H. N. Sköld, H. L. Wood, A-S. Krång, and S. Eriksson**. *Univ. of Gothenburg, Sweden.*

63 **23.3** Repeatability and Morphological Correlates of Fish Behavior in Hypoxia. **L. Matute*, K. Bricker, and B. Rees**. *Univ. of New Orleans.*

64 **23.4** Effects of Hypercapnia on Gill Ventilation, Standard Metabolic Rate, Oxygen Supply Capacity and Swimming Performance in Red Drum (*Sciaenops ocellatus*). **R. Ern, and A. Esbaugh**. *Univ. of Texas, Marine Sci. Inst., Port Aransas.*

65 **23.5** Optimization of *in vitro* Incubation of Gills from *Fundulus grandis*. **K. Farragut*, J. Diaz, and B. Rees**. *Univ. of New Orleans.*

66 **23.6** Purification and Characterization of Antibodies Against Killifish HIF-1 α . **J. Gonzalez-Rosario*, and B. Rees**. *Univ. of New Orleans.*

67 **23.7** Acclimation to Overnight Hypoxia and Increased Temperature Improve Aerobic Performance in Salmon (*Salmo salar*) and Charr (*Salvelinus alpinus*). **K. Anttila, J. Prokkola, and M. Nikinmaa**. *Univ. of Turku, Finland.*

68 **23.8** Long-term Acclimation to Hypoxia Does Not Confer Cross-tolerance to High Temperature in Steelhead Trout (*Oncorhynchus mykiss*). **T. Norin*, R. Motyka, and A. K. Gamperl**. *Memorial Univ. of Newfoundland, Canada.*

69 **23.9** Hypoxia-induced Apoptosis in the Hearts of Hypoxia-tolerant Tilapia (*Oreochromis Hybrid sp.*) and Hypoxia-sensitive Striped Bass (*Morone saxatilis*). **A. Reynolds*, and J. M. Lewis**. *Georgia Southern Univ.*

70 **23.10** Acute Thermal Challenges of Cardiac Function in Pacific Bluefin Tuna. **H. Shiels, G. Galli, and B. Block**. *Univ. of Manchester, UK., and Stanford Univ.*

71 **23.11** Temperature Dependence of Blood-Oxygenation in Juvenile Sandbar Sharks (*Carcharhinus plumbeus*). **P. Morrison*, T. Harter, R. Brill, P. Bushnell, and C. Brauner**. *Univ. of British Columbia, Vancouver, Canada, Natl. Marine Fisheries Service and the Virginia Inst. of Marine Sci., Gloucester Point, and Indiana Univ., South Bend.*

Poster Board

72 **23.12** Effect of Temperature Acclimation on Hemoglobin-oxygen Binding Properties in Pacific Bluefin Tuna (*Thunnus orientalis*) and Yellowfin Tuna (*Thunnus albacare*). **L. Lilly, J. Bonaventura, M. Lipnick, and B. Block**. *Stanford Univ., Duke Univ., and Univ. of California, San Francisco.*

73 **23.13** Aerial Respiration in Polypterids. **C. Jew, and J. Hicks**. *Univ. of California, Irvine.*

74 **23.14** The Effects of *Umbellularia californica* Essential Oil on the Cutaneous Vasculature of Frogs. **H. Wagstaff, S. Maman, M. J. Tufte, and M. Weeg**. *Southern Utah Univ.*

75 **23.15** Baroreflex Characteristics of Anuran Amphibians from Different Environments. **M. Hedrick, and D. Crossley**. *California State Univ, East Bay, and Univ. of North Texas.*

76 **23.16** Does the Right-to-Left Shunt Affect Assimilation Efficiency, Digesta Transit, and Postprandial Metabolism in Alligators? **C. Slay*, J. Eme, and J. Hicks**. *Univ. of California, Irvine, and McMaster Univ., Hamilton, Canada.*

77 **23.17** Hypercarbic Ventilatory Response in Lizards (*Tropidurus torquatus*) Acclimated to Different Temperatures. **L. de Souza Porto, K. C. Bicego, W. Klein, and L. H. Gargaglione**. *São Paulo State Univ., Jaboticabal, Brazil, and Univ. of São Paulo, Ribeirão Preto, Brazil.*

78 **23.18** The Avian Paradox: Avian Resistance to Protein Glycation. **K. Sweazea, C. Borges, and S. Rayle**. *Arizona State Univ.*

79 **23.19** Nature Hibernation Protects Myocardial Ischemia. **D. Vatner, L. Yan, and S. Vatner**. *Rutgers Univ.-New Jersey Med. Sch.*

80 **23.20** Characterizing Respiratory Parameters in Captive Belugas and from Excised Lungs. **M. Piscitelli, A. Fahlman, M. Brodsky, D. Garcia, M. Haulena, L. Loseto, S. Raverty, and R. Shadwick**. *Univ. of British Columbia, Vancouver, Canada, Texas A&M Univ., V.M.D. Consulting, Miami Shores, FL, L'Océanografic, Valencia, Spain, Vancouver Aquarium, Canada, Vancouver Oceans, Winnipeg, Canada, Dept. of Fisheries and Oceans, British Columbia Ministry of Agriculture, Abbotsford, BC, Canada.*

81 **23.21** Grand Paradigm Shift in Diving Physiology: the Likelihood of the Bends in Marine Vertebrates. **Y. B. de Quiros, M. Moore, D. García-Párraga, P. Jepson, J. L. Crespo-Picazo, A. Fahlman, and A. Fernández**. *Texas A&M Univ., Woods Hole Oceanographic Inst., L'Océanografic, Valencia, Spain, Zoological Soc. of London, UK, and Univ. of Las Palmas de Gran Canaria, Spain.*

82 **23.22** Adaptation to High Altitude is Linked to Growth of the Lungs and Higher Alveolar Surface in Mice but Not in Rats. **A. Jochmans-Lemoine*, G. Villalpando, M. Conzaes, I. Valverde, R. Soria, and V. Joseph**. *Laval Univ., Quebec, Canada, and IBBA, La Paz, Bolivia.*

83 **23.23** Aerobic Physical Training Increases Contractile Response and Reduces the Cardiac Fibrosis in Rats Submitted to Early Estrogen Deprivation. **H. C. D. De Souza, A. C. S. Felix, and S. G. V. Dutra**. *Univ. of São Paulo, Ribeirão Preto, Brazil.*

DAILY SCHEDULE

Poster Board

- 84 **23.24** Dynamics of Oxygen Utilization During Passive and Active Cycling Exercise. **T. Saitoh, and K. Niizeki.** *Yamagata Univ., Japan.*
- 85 **23.25** The Effects of Modulating Endothelial Nitric Oxide Synthase (eNOS) Activity and Coupling in Coronary, Hindlimb, Renal, and Mesenteric Vascular Inflammation Models. **A. Lopez, K-A. Perkins, A. Koon, A. Bottex, Q. Chen, R. Barsotti, and L. Young.** *Philadelphia Coll. of Osteopathic Med.*
- 86 **23.26** The Effect of Streptokinase Infusion on Cardiac Biomarkers & ST Segment of Electrocardiogram Post Myocardial Infarction in Humans. **N. Habibolahy*.**
- 87 **23.27** Protein Kinase C (PKC) Delta (δ) Activator Attenuates N^G-nitro-L-Arginine-Methyl-Ester (L-NAME) Induced Leukocyte-Endothelial Interactions in Rat Mesenteric Postcapillary Venules. **H. Pham, A. Bottex, A. Koon, J. Adams, Q. Chen, R. Barsotti, and L. Young.** *Philadelphia Coll. of Osteopathic Med.*

Poster Presentations

24.0 DEVELOPMENTAL PHYSIOLOGY

Tues., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 88 **24.1** Genetically Determined Variation in Metabolic Allocation-Potential for Adaptation to Environmental Change. **S. L. Applebaum, T-C. F. Pan, B. A. Lentz, D. Hedgecock, and D. T. Manahan.** *Univ. of Southern California.*
- 89 **24.2** Exposure to Lowered pH and Acute Thermal Stress Increases Mortality in Embryonic Porcelain Crabs. **E. Armstrong, T. Page, N. Miller, E. Papineau, P. Calosi, and J. Stillman.** *Univ. of California, Berkeley, San Francisco State Univ., and Plymouth Univ., UK.*
- 90 **24.3** Reallocation of ATP in Response to the Stress of Ocean Acidification. **T-C. F. Pan, S. L. Applebaum, C. A. Frieder, and D. T. Manahan.** *Univ. of Southern California.*
- 91 **24.4** Characterizing Metamorphosis of the Alfalfa Leafcutting Bee Using Micro-computed Tomography. **B. Helm*, S. Payne, J. Rinehart, G. Yocum, J. Bowsher, and K. Greenlee.** *North Dakota State Univ., and USDA-ARS-NPA, Fargo, ND.*
- 92 **24.5** Shifting Incubation Temperatures Alter Heart Rate and Oxygen Consumption of Lake Whitefish Embryos and Hatchlings. **J. Eme, C. Mueller, R. Manzoni, C. Somers, D. Boreham, and J. Wilson.** *McMaster Univ., Hamilton, Canada, Univ. of Regina, Canada, Northern Ontario Sch. of Med., Canada.*
- 93 **24.6** Physiological Consequences of Compensatory Growth in the Checkered Garter Snake, *Thamnophis marciatus*. **K. Holden, A. Bronikowski, and N. Ford.** *Iowa State Univ., and Univ. of Texas at Tyler.*
- 94 **24.7** Hypoxia During Critical Windows of Ontogeny Alters Organ Mass and Cardiovascular Function in the American Alligator (*Alligator mississippiensis*). **K. Tate, J. Eme, J. Crossley, T. Rhen, R. Elsey, Z. Kohl, and D. Crossley.** *Univ. of North Texas, McMaster Univ., Hamilton, Canada, Univ. of North Dakota, Grand Forks, Louisiana Dept. of Wildlife and Fisheries, Grand Chenier.*
- 95 **24.8** Temperature Effects on Heart Rate and Baroreflex Function of Embryonic American Alligator (*Alligator mississippiensis*). **D. Nelson, K. Tate, R. Elsey, and D. Crossley.** *Univ. of North Texas, Louisiana Dept. of Wildlife and Fisheries, Grand Chenier.*

Poster Board

- 96 **24.9** The Effects of Chronic and Acute Hypoxia on Cardiac Function in Embryonic Chickens. **D. Crossley, M. Espinoza, E. Davis, G. Giraud, and S. Jonker.** *Univ. of North Texas, Oregon Hlth. & Sci. Univ., and Portland VA Med. Ctr.*
- 97 **24.10** Developmental Physiology of the Pekin Duck (*Anas pekin*) Ductus Arteriosus. **F. Mascarenhas*, and E. Dzialowski.** *Univ. of North Texas.*
- 98 **24.11** Developmental Changes in mRNA Levels of avANT and PGC-1 α in Duck Liver and Heart. **J. Jung*, T. Sirsat, and E. Dzialowski.** *Univ. of North Texas.*
- 99 **24.12** Thyroid Hormone and Development of Endothermy in King Quail. **M. Pineda*, S. Sirsat, T. Sirsat, and E. Dzialowski.** *Univ. of North Texas.*
- 100 **24.13** How to Build a Furnace: The Role of T3 in Development of Endothermy in the Altricial Red-winged Blackbird (*Agelaius phoeniceus*). **S. G. Sirsat*, T. Sirsat, M. Pineda, and E. Dzialowski.** *Univ. of North Texas.*
- 101 **24.14** Conditional Pulmonary Overexpression of Claudin 6 (Cldn6) During Embryogenesis Delays Lung Morphogenesis. **F. R. Jimenez*, S. T. Belgique, S. A. Albright, C. M. Jones, and P. R. Reynolds.** *Brigham Young Univ.*

Visit the exhibit booths daily during the poster sessions

Poster Presentations

25.0 ENDOCRINOLOGY AND REPRODUCTION

Tues., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 102 **25.1** Hyperglycemic and Putative Hyperlipidemic Activities of the Recombinant Crustacean Hyperglycemic Hormone CHH-B1 Isoform in the Pacific White Shrimp *Litopenaeus vannamei*. **L. C. Jiménez, E. P. Rivas, M. E. M. Márquez, and F. Díaz.** *CICESE, Ensenada, Mexico, and Univ. Autónoma de Baja California, Tijuana, Mexico.*
- 103 **25.2** Biased Signaling by Two Endogenous GnRH Isoforms Differentially Regulates Total LH and GH Availability in Goldfish Pituitary Cells. **J. Pemberton*, M. Orr, J. Stafford, and J. Chang.** *Univ. of Alberta, Canada.*
- 104 **25.3** Perchlorate Exposure Does not Affect Hormone Cycling Over Diel and Reproductive Season Schedules in Threespine Stickleback. **A. Gardell*, F. von Hippel, W. Cresko, J. Postlethwait, and C. L. Buck.** *Univ. of Alaska, Anchorage, and Univ. of Oregon.*
- 105 **25.4** Exogenous AVT Suppresses Courtship Behavior in *Xenopus laevis*. **H. Rhodes, C. Ego, and K. Es-selburn.** *Denison Univ., Granville, OH.*
- 106 **25.5** TRH Increases GH, but no Thyroid Hormones Release During Cold Exposure in Green Iguana. **J. Avila-Mendoza*, M. Luna, M. Carranza, D. Avila-Mendoza, and C. Arámburo.** *Neurobiology Inst., Juriquilla, Mexico.*

Poster Board
107

25.6 Overwinter Changes in Weddell Seal Body Condition and Hormone Profiles: Implications for Pregnancy? **M. Shero, R. Krotz, D. Costa, J. Richmond, and J. Burns.** *Univ. of Alaska, Anchorage, Univ. of North Florida, and Univ. of California, Santa Cruz.*

108

25.7 Ameliorative Capacity of Quercetin on Alcohol and Nicotine Induced Infertility in Experimental Rats. **C. Akintayo*, and Y. Raji.** *Afe Babalola Univ., Ado Ekiti, Nigeria, and Univ. of Ibadan, Nigeria.*

109

25.8 Withdrawn.

Poster Presentations

26.0

THERMAL PHYSIOLOGY

Tues., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

110

26.1 Water Before Ions? Early Chill Coma Ion Balance Challenges the Current Mechanistic Model of Chill Coma. **L. Des Marteaux*, and B. Sinclair.** *Univ. of Western Ontario, Canada.*

111

26.2 Thermal Sensitivity of Muscle Performance in the Chill Susceptible Locust, *Locusta migratoria*. **A. Findsen*, T. H. Pedersen, and J. Overgaard.** *Aarhus Univ., Denmark.*

112

26.3 The Potential to Resist Chill Coma: How Temperature Affects Flight Muscle Resting Membrane Potential and Heart Rate in Drosophilids. **J. Andersen*, and J. Overgaard.** *Aarhus Univ., Denmark.*

113

26.4 Variation in Thermal Tolerance, Hypoxia Tolerance and Metabolic Rate in the Atlantic Killifish, *Fundulus heteroclitus*. **T. Healy, and P. Schulte.** *Univ. of British Columbia, Vancouver, Canada.*

114

26.5 Sub-lethal Heat Stress Causes Apoptosis in Cold-adapted Antarctic Fishes. **B. Buckley, and I. Sleadd.** *Portland State Univ.*

115

26.6 Behavioural Regulation of Water Loss in Four Australian Skinks. **E. Pirtle*, C. Tracy, and M. Kearney.** *Univ. of Melbourne, Australia, and California State Univ., Fullerton.*

116

26.7 Thermal Preference During Metabolic Recovery from Anoxic Hibernation in Painted Turtles. **E. Cantrell*, C. Hill, M. Oxendine, D. Odegard, and D. Warren.** *St. Louis Univ.*

117

26.8 The Role of Ambient Temperature on Toxin Ingestion by a Mammalian Herbivore. **P. Kurnath*, and M. D. Dearing.** *Univ. of Utah.*

118

26.9 From Fur to Blubber: Evolutionary and Ontogenetic Transitions in Mammalian Insulation. **H. Liwanag, L. Pearson, N. Gmuca, A. Berta, D. Costa, J. Burns, S. Budge, and T. Williams.** *Adelphi Univ., Garden City, NY, Univ. of Alaska, Fairbanks, San Diego State Univ., Univ. of California, Santa Cruz, Univ. of Alaska, Anchorage, and Dalhousie Univ., Halifax, Canada.*

Oral Abstract Presentations

27.0

CONSERVATION PHYSIOLOGY

Tues., 3:30—5:30 PM, Pacific Ballroom.

Chair:

Johannes Overgaard, *Aarhus Univ., Denmark.*

3:30 PM

27.1 Exploring Local Adaptation to Ocean Acidification in *Mytilus californianus* During Simulated Up-

welling Events. **Geoffrey Dilly.** *California State Univ., Channel Islands, Camarillo, CA.*

3:45 PM

27.2 High Pressure Neurological Syndrome in Shallow-water Marine Invertebrates: Implications on Climate Driven Bathymetric Range Shifts and Acclimatization to Depth. **James P. Morris.** *Univ. of Southampton, UK.*

4:00 PM

27.3 Adaptive Variability in Salinity Tolerance Explains Habitat Variability Between Genetically Distinct Populations of Sacramento Splittail. **Christine Verhille.** *Univ. of California, Davis.*

4:15 PM

27.4 Temperature and Hypoxia Affect Swimming Energetics and Kinematics of Brown Trout (*Salmo trutta*). **Karlina Ozolina.** *Univ. of Manchester, UK.*

4:30 PM

27.5 Behavioural Responses of Black Perch to Marine *Synechococcus cyanobacteria*. **Trevor Hamilton.** *MacEwan Univ., Edmonton, Canada.*

4:45 PM

27.6 Brevetoxin Metabolism and Physiology Using Freshwater Turtles as a Model to Measure Morbidity in Endangered Sea Turtles. **Sarah Milton.** *Florida Atlantic Univ.*

5:00 PM

27.7 Avian Thermoregulation in the Heat: Tolerance to Heat Stress Varies Greatly Among Species. **B. Wolf.** *Univ. of New Mexico.*

5:15 PM

27.8 Physiological and Behavioral Responses to Environmental Challenges in the Western Terrestrial Garter Snake, *Thamnophis elegans*. **E. Gangloff, A. Sparkman, and A. Bronikowski.** *Iowa State Univ., and Westmont Coll., Santa Barbara.*

Oral Abstract Presentations

28.0

DEVELOPMENTAL PHYSIOLOGY

Tues., 3:30—5:30 PM, Pacific Salon 4/5.

Chair:

Dane Crossley, *Univ. of North Texas.*

3:30 PM

28.1 Molecular Biological and Genomic Analysis of the Developmental Physiology of Integumental Transport. **Donal T. Manahan.** *Univ. of Southern California.*

3:45 PM

28.2 Ontogenetic Changes in the Osmotic Stress Response of Blue Mussels. **Melissa May.** *Univ. of Maine, Orono.*

4:00 PM

28.3 Tracheal System Structure and Function Changes Within an Instar in the Caterpillar, *Manduca sexta*. **Kendra Greenlee.** *North Dakota State Univ.*

4:15 PM

28.4 Effects of Calcium Availability on Growth and Survival of *Acipenser fulvescens* in Early Life Stages. **Janet Genz.** *Univ. of West Georgia, Carrollton.*

4:30 PM

28.5 Gene Expression Patterns of Alternative Developmental Trajectories in Embryos of an Annual Killifish. **Amie Romney.** *Portland State Univ.*

4:45 PM

28.6 Evidence of Hypoxic Metabolic Programming in Developing Alligator Hearts. **Gina Galli.** *Univ. of Manchester, UK.*

5:00 PM

28.7 Effect of Thyroid Hormone Manipulation on Endothermic Development in Double-Crested Cormorants (*Phalacrocorax auritus*). **Tushar Sirsat.** *Univ. of North Texas.*

5:15 PM

28.8 Quantification of Left Ventricular Function in Embryonic Chickens (*Gallus gallus domesticus*) at 70% of

DAILY SCHEDULE

Incubation Using Ultrasonic Imaging. **Zachary Kohl**. *Univ. of North Texas*.

Oral Abstract Presentations

29.0 METABOLISM: HYPOXIA AND ANOXIA

Tues., 3:30—5:30 PM, Pacific Salon 6/7.

- Chairs: **Leslie Buck**, *Univ. of Toronto, Canada*.
Andrea Morash, *Univ. of Tasmania, Hobart, Australia*.
- 3:30 PM **29.1** Erythropoietin Through Evolution: A Speculative View. **Max Gassman**. *Univ. of Zurich, Switzerland*.
- 3:45 PM **29.2** Evolution of Cytochrome Oxidase Subunit 4-2 as a Hypoxia Responsive Gene. **Christopher Moyes**. *Queen's Univ., Kingston, Canada*.
- 4:00 PM **29.3** Cytochrome C Oxidase Oxygen Binding Affinity Varies with Hypoxia Tolerance in Intertidal Fishes. **Gigi Lau**. *Univ. of British Columbia, Vancouver, Canada*.
- 4:15 PM **29.4** Characterizing the Influence of Anoxia Exposure on the Isolated Hagfish Heart. **Todd Gillis**. *Univ. of Guelph, Canada*.
- 4:30 PM **29.5** Deep Sequencing of the Hepatopancreas Transcriptome Reveals New Isoforms of Hemocyanin and their Regulation in Response to Low O₂ High CO₂ in the Pacific Whiteleg Shrimp, *Litopenaeus vannamei*. **Jillian Johnson**. *Coll. of Charleston*.
- 4:45 PM **29.6** Anoxia-responsive Small RNA Gene Expression in Annual Killifish Embryos. **Claire L. Riggs**. *Portland State Univ.*
- 5:00 PM **29.7** Is Anoxia and ROS-mediated GABA Receptor Inhibitory Shunting in Turtle Cortical Neurons Mediated by Tonic, Fast or Slow Phasic Currents? **Leslie Buck**. *Univ. of Toronto, Canada*.
- 5:15 PM **29.8** Can Ketone Bodies Protect the Heart Against the Effects of Chronic Hypoxia? **Andrea Morash**. *Univ. of Tasmania, Hobart, Australia*.

Oral Abstract Presentations

30.0 OSMOTIC AND ION REGULATION: SALINITY, OSMOLYTES, AND pH

Tues., 3:30—5:30 PM, Royal Palms Salon 1/2.

- Chairs: **Dietmar Kultz**, *Univ. of California, Davis*.
Lars Tomanek, *California Poly State Univ., San Luis Obispo*.
- 3:30 PM **30.1** Physiological and Functional Genomic Mechanisms of Seawater to Freshwater Transitions in the Alewife. **Jonathan Velotta**. *Univ. of Connecticut*.
- 3:45 PM **30.2** From Comparative Intestinal Transcriptome Analysis to Characterization of Transporters Linking Nutrient Absorption with Ion and Acid-base Regulation. **Avner Cnaani**. *Agricultural Res. Org., Bet Dagan, Israel*.
- 4:00 PM **30.3** The Regulation and Function of Polyamines in *Fundulus* Species During Salinity Stress. **Fernando Galvez**. *Louisiana State Univ.*
- 4:15 PM **30.4** Regulation of Organic Osmolyte Concentration in Tissues of Euryhaline Teleosts. **Dietmar Kultz**. *Univ. of California, Davis*.
- 4:30 PM **30.5** Cellular Mechanisms for Acid/Base Sensing and Regulation in Elasmobranch Gills. **Jinae N. Roa**. *Scripps Inst. of Oceanography, La Jolla*.

- 4:45 PM **30.6** Ocean Acidification Stimulates Respiratory Plasticity in the Estuarine Red Drum (*Sciaenops ocellatus*). **Andrew Esbaugh**. *Univ. of Texas Marine Sci. Inst., Port Aransas*.
- 5:00 PM **30.7** Changes to Intestinal Transport Physiology at Varying Levels of Hypercapnia in the Gulf Toadfish (*Opsanus beta*). **Rachael Heuer**. *RSMAS, Univ. of Miami*.
- 5:15 PM **30.8** Crab Proteomics: Responses to Simultaneous Low pH and Temperature Stress. **Lars Tomanek**. *California Poly State Univ., San Luis Obispo*.

Workshop II

31.0 THE CHALLENGE OF TEACHING PHYSIOLOGY IN A CHANGING ENVIRONMENT: INNOVATION AND RESOURCES

Tues., 5:45—7:30 PM, Pacific Ballroom.

- Chairs: **Tom W. Ecy**, *East Tennessee State Univ.*
Karen Sweazea, *Arizona State Univ.*
- 5:45 PM **31.1** Vision and Change Update: Progress in Implementing Report Goals in Undergraduate Biology Education. **Cynthia Bauerle**. *HHMI*.
- 6:05 PM **31.2** Teaching and Learning by Inquiry. **Barbara Goodman**. *Univ. of South Dakota*.
- 6:25 PM **31.3** Assessing Student Learning After Converting to Inquiry. **Douglas Luckie**. *Michigan State Univ.*
- 6:45 PM **31.4** Constructing Concept Inventories and Assessing their Value. **Jenny McFarland**. *Edmonds Comm. Coll.*
- 7:05 PM **31.5** APS Resources for Teachers and Students. **Miranda Byse**. *American Physiological Soc.*

WEDNESDAY, OCTOBER 8, 2014

Concurrent Symposia XIII

32.0 MOLECULAR AND PHYSIOLOGICAL FEATURES OF ANIMAL DIAPAUSE

Wednes., 8:00—10:00 AM, Pacific Ballroom.

- Chair: **Steven C. Hand**, *Louisiana State Univ.*
- 8:00 AM **32.1** AMPK Buffers Adverse Epigenetic Change and Consequent Transgenerational Reproductive Defects Following Acute Energy/Nutrient Stress. **Richard Roy**. *McGill Univ., Montreal, Canada*.
- 8:30 AM **32.2** Insulin Signaling as a Key Regulator of Insect Diapause. **David Denlinger**. *Ohio State Univ.*
- 9:00 AM **32.3** The Role of Maternal Provisioning and Micro-RNA Regulation of Diapause in the Annual Killifish *Austrofundulus limnaeus*. **Jason Podrabsky**. *Portland State Univ.*
- 9:30 AM **32.4** Bioenergetics of Diapause in a Crustacean Extremophile. **Steven Hand**. *Louisiana State Univ.*

Photography is not permitted during the meeting sessions or in the poster room

Concurrent Symposia XIV

33.0 NEW PERSPECTIVES ON THE ECOLOGY AND EVOLUTION OF HOMEOSTASIS

Wednes., 8:00—10:00 AM, Pacific Salon 4/5.

*Sponsored by the Society for Integrative Comparative Biology.*Chairs: **Lynn B. Martin**, *Univ. of South Florida*.
H. Art Woods, *Univ. of Montana*.8:00 AM **33.1** Integrating Physiological Assessments of Animal Health to Population Models. **Erica Crespi**, *Washington State Univ.*8:30 AM **33.2** The Reactive Scope Model: Predicting the Effects of Challenges to Homeostasis. **Molly Dickens**, *Univ. of California, Berkeley*.9:00 AM **33.3** Physiological Regulatory Networks: The Orchestra of Life? **Lynn B. Martin**, *Univ. of South Florida*.9:30 AM **33.4** Information Theory, Homeostasis, and Evolution. **H. Art Woods**, *Univ. of Montana*.

Concurrent Symposia XV

34.0 LINKING BEHAVIOR AND PHYSIOLOGY IN ANIMAL NAVIGATION AND ORIENTATION

Wednes., 8:00—10:00 AM, Pacific Salon 6/7.

*Sponsored by the International Society for Neuroethology*Chairs: **Jochen Smolka**, *Lund Univ., Sweden*.
Basil el Jundi, *Lund Univ., Sweden*.8:00 AM **34.1** Using Genetics to Reveal Migratory Flight Orientation Mechanisms in the Monarch Butterflies. **Christine Merlin**, *Texas A&M Univ.*8:30 AM **34.2** 3D Neural Compass in the Bat Brain. **Arseny Finkelstein**, *Weizmann Inst. of Sci., Rehovot, Israel*.9:00 AM **34.3** Neural Representation of the Hierarchy of Celestial Cues in the Dung Beetle Brain. **Basil el Jundi**, *Lund Univ., Sweden*.9:30 AM **34.4** Polarized Light Navigation in *Drosophila*. **Michael Dickinson**, *Univ. of Washington*.

Oral Abstract Presentations

35.0 THERMAL PHYSIOLOGY

Wednes., 10:30—12:30 PM, Pacific Ballroom.

Chair: **Edward Dzialowski**, *Univ. of North Texas*.10:30 AM **35.1** An in vivo Investigation of Low Temperature Energetics in *Drosophila melanogaster* Using ³¹P NMR. **Caroline Williams**, *Univ. of California, Berkeley*.10:45 AM **35.2** New Approaches to Understanding Insect Freeze Tolerance. **Brent J. Sinclair**, *Univ. of Western Ontario, London, Canada*.11:00 AM **35.3** Mitochondrial and Nuclear Genetic Variation Relate to Heat and Cold Tolerance in a Montane Leaf Beetle. **Nathan Rank**, *Sonoma State Univ., Rohnert Park, CA*.11:15 AM **35.4** Never Mind Thermal Performance Curves-It's All About Tolerating the Extremes! Sensitivity to Thermal Extremes Predict Current (and Future?) Distribution of*Drosophila* Species. **Johannes Overgaard**, *Aarhus Univ., Denmark*.11:30 AM **35.5** Thermal Performance, Aerobic Scope, and Relevance of the OCLTT Hypothesis. **Fredrik Jutfelt**, *Univ. of Gothenburg, Sweden*.11:45 AM **35.6** Thermal Tolerance and Molt Cycle-dependent Gene Expression in Juvenile Dungeness Crabs. **Astrid Wittmann**, *Alfred Wegener Inst., Helmholtz Ctr. for Polar & Marine Res., Bremerhaven, Germany*.12:00 Noon **35.7** Variation in Transcriptomic Signatures of Thermal Acclimation in Four Key Aquatic Insects in California Riverine Food Webs. **Jonathon Stillman**, *Univ. of California, Berkeley*.12:15 PM **35.8** Seasonally Induced Hepatotranscriptomic Changes in the Freeze Tolerant North American Wood Frog *Rana sylvatica*. **Andor Kiss**, *Miami Univ.*

Oral Abstract Presentations

36.0 ENDOCRINOLOGY AND REPRODUCTIVE PHYSIOLOGY

Wednes., 10:30—12:30 PM, Pacific Salon 4/5.

Chair: **Stephen Trumble**, *Baylor Univ.*10:30 AM **36.1** Elucidating the Regulatory Mechanisms of Circadian Control of Ovarian Ecdysteroid Production and Release During Egg Development in Adult Female *Rhodnius prolixus*. **Andrea Durant**, *York Univ., Toronto, Canada*.10:45 AM **36.2** One Cell or Two? Direct Visualization of Ligand-Receptor Interaction Provides Novel Insights into the Evolution of Insect Renal Function. **Kenneth Halberg**, *Univ. of Glasgow, UK*.11:00 AM **36.3** Brain Monoamines and Behavior- Relationship to Personality Traits and the Effects of Social Interaction. **Svante Winberg**, *Uppsala Univ., Sweden*.11:15 AM **36.4** Withdrawn.11:30 AM **36.5** Corticosterone Responses and the Ability of Birds to Cope with Environmental Change. **John F. Cockrem**, *Massey Univ., Palmerston North, New Zealand*.11:45 AM **36.6** Influence of Corticosterone on Growth, Home-cage Activity, Wheel Running, and Maximal Oxygen Consumption in Replicate Lines of House Mice Selectively Bred for High Voluntary Wheel-running Behavior. **Jennifer Singleton**, *Univ. of California, Riverside*.12:00 Noon **36.7** Molecular Resolution of an Acute Stress Response in a Free-ranging Marine Mammal. **Jane Khudiyakov**, *Sonoma State Univ., Rohnert Park, CA*.12:15 PM **36.8** Breaking Diapause: Successful Use of Ultrasonography Shows Intra-specific Variation in the Probability and Timing of Embryo Implantation in Weddell Seals. **Michelle Shero**, *Univ. of Alaska, Anchorage*.

Oral Abstract Presentations

37.0 HIBERNATION, FLIGHT, AND SUBSTRATE METABOLISM

Wednes., 10:30—12:30 PM, Pacific Salon 6/7.

Chairs: **Marshall McCue**, *St. Mary's Univ., San Antonio*.
Allyson Hindle, *Massachusetts Gen. Hosp.*10:30 AM **37.1** Seasonal Metabolism of Brown Adipose Tissue in Hibernating Thirteen-Lined Ground Squirrels. **Mallory Ballinger**, *Univ. of Minnesota, Duluth*.

DAILY SCHEDULE

- 10:45 AM **37.2** Is Saponin-permeabilization Appropriate for Characterizing Mitochondrial Metabolism? Assessing Mitochondrial Respiration in Hibernating and Euthermic Ground Squirrels. **James Staples**. *Univ. of Western Ontario, London, Canada*.
- 11:00 AM **37.3** Brown Fat Transcriptome Dynamics: Preservation of Selected mRNAs Across a Torpor Bout Supports Rapid Thermogenesis During Arousal. **Sandy Martin**. *Univ. of Colorado, Denver*.
- 11:15 AM **37.4** Characterizing Cardiac Molecular Mechanisms of Mammalian Hibernation Via Quantitative Proteogenomics. **Katie Vermillion**. *Univ. of Minnesota, Duluth*.
- 11:30 AM **37.5** Do Polyunsaturated Fatty Acids Improve Migratory Flight Performance? **Morag Dick**. *Univ. of Western Ontario, London, Canada*.
- 11:45 AM **37.6** Cold and Exercise Training Produce Similar Increases in Maximal Metabolic Output in House Sparrows. **Yufeng Zhang**. *Univ. of South Dakota*.
- 12:00 Noon **37.7** Hydrogen Isotope ($\delta^2\text{H}$) Discrimination in Tilapia. **Seth Newsome**. *Univ. of New Mexico*.
- 12:15 PM **37.8** The Breath Becomes Isotopically Heavier as the Body Burns More Carbohydrates During Intense Exercise: Exploiting the Natural Differences in ^{13}C Between Lean and Lipid Tissues. **Marshall McCue**. *St. Mary's Univ., San Antonio*.

Oral Abstract Presentations

38.0 BIOMECHANICS, LOCOMOTION, AND FUNCTIONAL MORPHOLOGY

Wednes., 10:30—12:30 PM, Royal Palms Salon 1/2.

Chair: **Scott Kirkton**, *Union Coll.*

- 10:30 AM **38.1** Musculoskeletal Plasticity of the Hindlimb/tail Locomotor Module in the American Alligator (*Alligator mississippiensis*). **Jessica Joneson**. *California State Univ., San Bernardino*.
- 10:45 AM **38.2** Skeletal Muscle performance Under Submaximally Activated Conditions. **Natalie Holt**. *Univ. of California, Irvine*.
- 11:00 AM **38.3** Size Matters: The Impact of Body Mass on biochemical and Structural Properties in Harbor Seal Muscles. **Jennifer Burns**. *Univ. of Alaska, Anchorage*.
- 11:15 AM **38.4** Structure and Mechanics of the Cetacean Diaphragm and its Contribution to Thorax Pressurization During a Dive. **Margo Lillie**. *Univ. of British Columbia, Vancouver, Canada*.
- 11:30 AM **38.5** Shrimp Exoskeleton Morphology, Mineralization, and Biophotonics Under Ocean Acidification Conditions. **Jennifer Taylor**. *Univ. of California, San Diego*.
- 11:45 AM **38.6** ExerFlyzer: A High-Throughput System for Inducing and Quantifying Flight Behavior in *Drosophila* Over Extended Time Periods. **Andrew Mead**. *Univ. of Vermont*.
- 12:00 Noon **38.7** Age Related Changes in Flight Muscle Ultrastructures of the Hawk moth, *Manduca sexta*: A novel Non-vertebrate Animal Model for Investigating Vertebrate Skeletal Muscle Function, Disease, Degeneration, and Aging. **Bernard Wone**. *Univ. of Nevada, Reno*.

- 12:15 PM **38.8** Effect of Gravity on Jump Performance and Muscle Physiology in the American Locust. **Scott Kirkton**. *Union Coll.*

Poster Presentations

39.0

OSMOTIC AND IONIC REGULATION

Wednes., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 119 **39.1** Potential Soluble Adenylyl Cyclase Isoforms in Coral. **M. Barron, K. Barott, and M. Tresguerres**. *Scripps Inst. of Oceanography, La Jolla*.
- 120 **39.2** Differential Localization of Ion-transporting Proteins Suggest Species-specific Physiological Mechanisms in Corals. **S. Perez, K. Barott, and M. Tresguerres**. *Scripps Inst. of Oceanography, La Jolla*.
- 121 **39.3** Extreme Stress Tolerance of the Intertidal Tardigrade *Echiniscoides sigismundi*. **N. Møbjerg, K. Andersen, T. Hygum, S. Mortensen, K. Halberg, L. Clausen, and A. Jørgensen**. *Univ. of Copenhagen, Denmark*.
- 122 **39.4** Branchial Chamber of *Litopenaeus vannamei* During Post-embryonic Development. **J. Chong-Robles, I. Giffard-Mena, D. Rodarte-Venegas, and K. K. Munguía-Ortega**. *Univ. Autónoma de Baja California, Mexico*.
- 123 **39.5** Allatostatin A-like Factors in the Aquatic Larvae of *Chironomus riparius*: Regulation of Hindgut Motility, Ion Reabsorption and Implications for Salinity Exposure. **L. Robertson, H. Chasiotis, V. Galperin, and A. Donini**. *York Univ., Toronto, Canada*.
- 124 **39.6** Sodium and Ammonia Transport in *Aedes aegypti* Larvae Reared in Varying Water Na Levels. **M. Patrick, K. Fazio, J. Wang, N. Day, N. Lane, J. Capaldi, V. Simon, E. Williams, A. Dollesin, A. Sulaeman, J. Castoreale, N. Graige, L. Harder, T. McReynolds, K. Monahan, A. Oh, A. Rwamashongye, D. Serna, H. Shaprio, D. Stuart, S. Syed, and L. York**. *Univ. of San Diego*.
- 125 **39.7** RNAi Knockdown of Ammonia Transporter and Osmoregulatory Transcripts in Larval Mosquitoes, *Aedes aegypti*. **H. Chasiotis, and A. Donini**. *York Univ., Toronto, Canada*.
- 126 **39.8** Functional Characterization of Heterologously Expressed *Drosophila melanogaster* Organic Cation Transporter ORCT in *Xenopus laevis* Oocytes. **M. Cruz*, B. Matier, and M. Rheault**. *Univ. of British Columbia, Kelowna, Canada*.
- 127 **39.9** Effects of Diuretics on Renal Calcium Oxalate Crystallization in a *Drosophila* (fly) Model of Oxalate Nephrolithiasis. **G. Landry, C. Gallo, K. Strohmaier, P. Williams, T. Hirata, J. Lieske, J. Dow, E. Furrow, and M. Romero**. *Mayo Clinic, Univ. of Glasgow, UK, and Univ. of Minnesota, St. Paul*.
- 128 **39.10** Conservation of the Osmosensitive and Thermosensitive ΔN -TRPV1 Ion Channel in Osmoregulating Animals. **C. Zaelzer, and C. W. Bourque**. *McGill Univ. Hlth. Ctr., Montreal, Canada*.
- 129 **39.11** The Role of Pulsatile Urea Excretion in Chemical Communication and Predator Avoidance in Gulf toadfish (*Opsanus beta*). **M. Cartolano, M. D. McDonald, H. Eskelinen, and J. Borger-Turner**. *Univ. of Miami, and Dolphins Plus, Key Largo, FL*.

Poster Board

- 130 **39.12** Characterization of the Serotonin Transporter (SERT) and Effects of Fluoxetine Treatment on SERT in the Gulf Toadfish, *Opsanus beta*. **M. H. Broome, C. M. Alexander, and M. D. McDonald.** *Univ. of Miami.*
- 131 **39.13** The Roles of Glutamate and Putrescine in γ -Aminobutyric Acid (GABA) Synthesis in *Fundulus heteroclitus* During Osmotic Stress. **K. Munley, D. Liu, and F. Galvez.** *Louisiana State Univ.*
- 132 **39.14** Tight Junction Proteins and the Regulation of Salt and Water Balance in an Extant Agnathan, *Petromyzon marinus*. **D. Kolosov*, P. Bui, A. Donini, M. Wilkie, and S. Kelly.** *York Univ., Toronto, Canada, and Wilfred Laurier Univ., Waterloo, Canada.*
- 133 **39.15** Homologous Serum and Heparin Alter Tight Junction Protein Abundance and the Permeability of a Primary Cultured Salmonid Gill Model. **C. C. Chen, and S. P. Kelly.** *York Univ., Toronto, Canada.*
- 134 **39.16** Regulation of Gill Claudin Isoforms in Mozambique Tilapia (*Oreochromis mossambicus*) by Salinity and Cortisol. **C. Tipsmark, J. P. Breves, D. B. Rabeneck, R. T. Trubitt, D. T. Lerner, and E. G. Grau.** *Univ. of Arkansas, Skidmore Coll., and Univ. of Hawaii.*
- 135 **39.17** Paracellular Pathway Regulation in Response to Salinity Changes in the Japanese Medaka (*Oryzias latipes*). **M. Bomane-Bossus*, S. Madsen, and C. Tipsmark.** *Univ. of Arkansas, and Univ. of Southern Denmark, Odense, Denmark.*
- 136 **39.18** Expression of Gill Na^+K^+ -ATPase α -Subunit Isoforms in Euryhaline Japanese Medaka (*Oryzias latipes*) During Salinity Challenges. **R. Bollinger*, S. Madsen, M. Bomane-Bossus, and C. Tipsmark.** *Univ. of Arkansas, and Univ. of Southern Denmark, Odense, Denmark.*
- 137 **39.19** Four Na^+K^+ -ATPase α -Subunit Isoforms in the Three Electric Organs and the Skeletal Muscle of the Electric Eel. **B. Ching, J. M. Woo, K. C. Hiong, W. P. Wong, S. F. Chew, P. K. L. Ng, and Y. K. Ip.** *Natl. Univ. of Singapore, and Natl. Inst. of Edu., Singapore.*
- 138 **39.20** Characterizing Sodium Uptake in the Blackskirt Tetra (*Gymnocorymbus ternetzi*). **N. Day*, A. Casciato, K. Galacgac, R. Gonzalez, and M. Patrick.** *Univ. of San Diego.*
- 139 **39.21** The Effects of Water Ionic Composition on the Rate and Degree of Acid-base Regulation in Rainbow Trout, *O. mykiss*, During Hypercarbia at Rest and Sustained Exercise. **K. Tovey, and C. Brauner.** *Univ. of British Columbia, Vancouver, Canada.*
- 140 **39.22** Gut Carbonate Excretion by Fish Increases Exponentially Within Natural Ocean Salinity Range (25-40 psu). **C. E. Stephens*, C. T. Perry, and R. Wilson.** *Univ. of Exeter., UK.*
- 141 **39.23** Phenotypic Plasticity in Response to Hypercapnia Induced Acid-base Disturbances in Red Drum (*Sciaenops ocellatus*). **E. Brown*, and A. Esbaugh.** *Univ. of Texas Marine Sci. Inst., Port Aransas.*
- 142 **39.24** Changes in Expression of *Aquaporin* Isoforms in an Aestivating African Lungfish. **Y. R. Chng, B. Ching, J. L. Y. Ong, X. L. Chen, W. P. Wong, S. H. Lam, S. F. Chew, and Y. K. Ip.** *Natl. Univ. of Singapore, and Natl. Inst. of Edu., Singapore.*
- 143 **39.25** Effects of Temperature on Hypotonic Swelling Induced by Water and Glycerol in Hepatocytes from

Poster Board

- Copes' Gray Treefrog.* **D. Goldstein, J. Frisbie, A. Backs, K. Bobka, and L. Finatti.** *Wright State Univ., and MacKenzie Univ., São Paulo, Brazil.*
- 144 **39.26** Cardiac pH Regulation and Buffering of the Western Painted Turtle (*Chrysemys picta bellii*). **M. J. Oxendine*, C. Grombka-Murphy, and D. Warren.** *St. Louis Univ.*
- 145 **39.27** Control of the Osmotic Function of the Avian Lower Gastrointestinal Tract. **E. Braun, J. Vranish, and P. Warner.** *Univ. of Arizona, and Univ. of Nebraska, Omaha.*
- 145A **39.28** Evidence of Rapid Physiological Evolution in an Introduced Lake Population of Threespine Stickleback. **J. Divino, M. Monette, S. McCormick, and E. Schultz.** *Univ. of Connecticut, Storrs, Western Connecticut State Univ., Danbury, and U. S. Geological Survey, Turner Falls, CT.*

Poster Presentations

40.0

"OMICS" IN COMPARATIVE PHYSIOLOGY

Wednes., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 146 **40.1** Enhancing the Restoration of California's Estuaries by Exploring the Genetic Basis of Environmental Tolerance in Olympia Oysters (*Ostrea lurida*). **A. Maynard, J. Bible, E. Sanford, and T. Evans.** *California State Univ., East Bay, and Univ. of California, Davis.*
- 147 **40.2** Proteomic Responses of the Mussel *Mytilus galloprovincialis* to Aerial Exposure Induced Hypoxia and Sirtuin Inhibition. **M. C. Vasquez, M. Rosner, J. Campbell, N. Petersen, M. Zuzow, and L. Tomanek.** *California Poly. State Univ., San Luis Obispo.*
- 148 **40.3** The Proteomic Response of Subtidally and Tidally-entrained California Ribbed Mussel *Mytilus californianus* to Anoxia Stress. **A. Fowler, M. Zuzow, and L. Tomanek.** *California Poly. State Univ., San Luis Obispo.*
- 149 **40.4** Transcriptomic Analysis of *Daphnia pulex* Response to Interactive Effects of Temperature and Salinity Variability. **E. Papineau*, S. Fay, and J. Stillman.** *San Francisco State Univ., Romberg Tiburon Ctr. for Environmental Studies, Tiburon, CA, and Univ. of California, Berkeley.*
- 150 **40.5** Taking a Comparative Approach to Understanding MicroRNA Expression and its Functional Consequences After Fluoxetine Exposure in Two Related Species, *Carassius auratus* and *Danio rerio*. **P. Craig, B. Cameron, T. Moon, and V. Trudeau.** *Univ. of Waterloo, Canada, and Univ. of Ottawa, Canada.*
- 151 **40.6** Whole-Genome Methylation Profiling of Threespine Stickleback Reared in High and Low Salinities. **D. C. H. Metzger, and P. M. Schulte.** *Univ. of British Columbia, Vancouver, Canada.*
- 152 **40.7** Determination of the Gill Transcriptome of the Fathead Minnow (*Pimephales promelas*). **S. Wentworth, K. Thede, V. Aravindabose, I. Monroe, A. Thompson, J. Garvin, and R. Packer.** *George Washington Univ., and Case Western Res. Univ.*
- 153 **40.8** Hypoxia Induces Global Reprogramming of the Acetylome in the Zebrafish Brain. **R. Dhillon, and J. Richards.** *Univ. of Wisconsin, Madison, and Univ. of British Columbia, Vancouver, Canada.*

DAILY SCHEDULE

Poster Board

- 154 **40.9** Evolution of Embryonic Diapause in the Annual Killifish *Austrofundulus limnaeus*: Searching for Clues in the Genome and Epigenome. **J. Wagner***, **F. Chavez**, and **J. Podrabsky**. *Portland State Univ.*
- 155 **40.10** Withdrawn.
- 156 **40.11** Effects of Ocean Acidification on Juvenile Rockfish (*Sebastes spp.*) Gene Expression. **A. Makukhov**, **L. Tobosa**, **G. Bernardi**, **H. Fennie**, **S. Hamilton**, and **C. Logan**. *California State Univ., Monterey Bay, Univ. of California, Santa Cruz, and San Jose State Univ.*
- 157 **40.12** Proteomic Profile and Proteogenomic Analysis of Skeletal Muscle in a Mammalian Hibernator. **K. Anderson***, **P. Jagtap**, **T. Markowski**, **L. Higgins**, **J. Johnson**, **T. Griffin**, and **M. Andrews**. *Univ. of Minnesota, Duluth, Univ. of Minnesota, St. Paul, and Univ. of Minnesota, Minneapolis.*
- 158 **40.13** A Functional Genomic Analysis of Weddell Seal Diving Adaptations: Vascular Biology. **A. G. Hindle**, **J. Turner-Maier**, **A. M. Berlin**, **J. U. Meir**, **J. Johnson**, **J. Alfoldi**, **P. Y. Sips**, **K. Lindblad-Toh**, **W. M. Zapol**, and **E. S. Buys**. *Massachusetts Gen. Hosp., Broad Inst., Cambridge, MA, Brigham and Women's Hosp., Boston.*

Poster Presentations

41.0 CONSERVATION PHYSIOLOGY

Wednes., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 159 **41.1** Ocean Acidification: Effects of CO₂ on Behavior and GABA Functions in Teleost Fish. **L. Vossen**, **A. Cocco**, **J. Sundin**, **B. Birnir**, **F. Jutfelt**, and **S. Winberg**. *Uppsala Univ., and Univ. of Gothenburg, Sweden.*
- 160 **41.2** Contrasting Sensitivity to Ocean Acidification-induced Behavioural Change in Atlantic Cod (*Gadus morhua*) and Three-spined Stickleback (*Gasterosteus aculeatus*). **F. Jutfelt**, **M. Hedgärde**, **K. B. DeSouza**, **J. Sturve**, and **J. Sundin**. *Univ. of Gothenburg, and Uppsala Univ., Sweden.*
- 161 **41.3** Pace of Life Effects Critical Windows for Disease Emergence and Transmission in Amphibians. **R. Warne**. *Southern Illinois Univ.*
- 162 **41.4** Ecosystem Health and Environmental Influences on Innate Immune Function in the Loggerhead (*Caretta caretta*) and Green (*Chelonia mydas*) Sea Turtle. **S. Milton**, **P. Keating**, and **P. Sposato**. *Florida Atlantic Univ.*
- 163 **41.5** Decoupling the Relationship Between Immune Response and Stress Hormones: An Immunologic Profile of the Northern Elephant Seal. **H. Peck**, and **D. Crocker**. *Sonoma State Univ., Rohnert Park, CA.*
- 164 **41.6** Withdrawn.
- 165 **41.7** Heat Tolerance of Australian Birds: The Importance of Body Size, Phylogeny and Evaporative Pathway. **A. Gerson**, **W. Talbot**, **E. Smith**, **T. McWhorter**, and **B. Wolf**. *Univ. New Mexico, and Univ. Adelaide, Australia.*

Poster Presentations

42.0 BIOMECHANICS, LOCOMOTION, AND FUNCTIONAL MORPHOLOGY

Wednes., 12:30—3:30 PM, Golden Ballroom.

*Denotes presenter is competing in the Best Poster Competition.

Poster Board

- 166 **42.1** The Mechanistic Basis of Unreliable Signals of Strength in Males of the Two-toned Fiddler Crab, *Uca vomeris*. **C. Bywater**, **C. White**, **F. Seebacher**, and **R. Wilson**. *Univ. of Melbourne, Univ. of Queensland, and Univ. of Sydney, Australia.*
- 167 **42.2** The Effects of Selection For Desiccation or Starvation Resistance on Takeoff Flight Performance in *Drosophila melanogaster*. **M. L. Brewer**, **L. M. Peterson**, and **A. Gibbs**. *Univ. of Nevada, Las Vegas.*
- 168 **42.3** Material and Structural Characterization of Mineralized Elasmobranch Cartilage: Lessons in Repeated Tiling Patterns in Mechanically Loaded 3D Objects. **R. Seidel**, **D. Knoetel**, **P. Zaslansky**, **D. Baum**, **J. Weaver**, and **M. Dean**. *Max Planck Inst., Potsdam, Germany, Zuse Inst., Berlin, Germany, Charité Hosp., Berlin, Germany, and Harvard Univ.*
- 169 **42.4** The Elasmobranch Heart Doesn't Twist: A Speckle-tracking Echocardiography Study. **Y. Hirasaki**, **S. Minamisawa**, and **M. Okabe**. *The Jikei Univ. Sch. of Med., Tokyo, Japan.*
- 170 **42.5** Trends in Morphology and Biomechanics of the Aquatic Gill Ventilatory System of Ray-finned Fishes (*Actinopterygii*). **S. Farina***, **L. Ferry**, **T. Near**, **A. Summers**, and **W. Bemis**. *Cornell Univ., Arizona State Univ., Yale Univ., and Univ. of Washington.*
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- 174 **42.9** Improving Exercise Adherence and Physical Measures in Hispanic Women. **L. Martin**, **A. C. Perry**, **B. E. Kahn**, **J. F. Signorile**, **S. Ahn**, and **A. W. Perkins**. *Univ. of California San Diego, Univ. of Miami, Univ. of Pennsylvania, and Washington State Univ.*

Scholander Award Competition

43.0 SCHOLANDER AWARD

FINALISTS

Wednes., 3:30—5:30 PM, Pacific Ballroom.

Chairs:

Scott Kirkton, *Union Coll.*

Siribhinya Benyajati, *Univ. of Oklahoma Hlth. Sci. Ctr.*

3:30 PM

43.1 Does Local Adaptation of Exercise Physiology Limit Acclimation Capacity Among Lake Whitefish (*Coregonus clupeaformis*) Ecotypes? **Anne Dalziel**. *Univ. of Laval, Quebec, Canada.*

3:45 PM

43.2 Coral Host Cells Acidify Symbiotic Algal Microenvironment to Promote Photosynthesis. **Katie Barott**. *Scripps Inst. of Oceanography, La Jolla.*

4:00 PM

43.3 GnRH-Selective Signal Transduction Networks and the Control of Pituitary Cell Function. **Joshua Pemberton**. *Univ. of Alberta, Canada.*

4:15 PM

43.4 Convergent and Divergent Patterns of Gene Expression in Sculpins that Vary in Hypoxia Tolerance.

Milica Mandic. *Univ. of British Columbia, Vancouver, Canada.*

- 4:30 PM **43.5** Ocean Acidification Directly Impairs Olfactory Sensitivity in a Marine Teleost. **Cosima Porteus.** *Univ. of Exeter, UK.*
- 4:45 PM **43.6** What has K^+ Got to do with it? The Differing Roles of Extracellular K^+ in Onset and Recovery of Insect Chill Coma. **Heath MacMillan.** *Aarhus Univ., Denmark.*
- 5:00 PM **43.7** Changes in MO_2 , Anaerobic Glycolysis and Metabolic Heat with Decreasing Water PO_2 in Goldfish. **Matthew Regan.** *Univ. of British Columbia, Vancouver, Canada.*
- 5:15 PM **43.8** Integrating the Effects of Repeated Cold Exposure from Transcriptome to Whole-organism in the Eastern Spruce Budworm. **Katie Marshall.** *Univ. of British Columbia, Vancouver, Canada.*
- 5:30 PM **43.9** The Role of Transcription Factor Glial Cell Missing 2 (*gcm2*) in Ca^{2+} Balance in Zebrafish Larvae. **Yusuke Kumai.** *Case Western Res. Univ.*

Plenary Lecture II

44.0

PLENARY LECTURE

Wednes., 8:30—9:30 PM, Pacific Ballroom.

*Sponsored by the Society for Experimental Biology,
Division of Comparative Physiology & Biochemistry.*

- 8:30 PM **44.1** Macrophysiological Forecasting for a Policy in a Changing World. **Steven L. Chown.** *Monash Univ., Australia.*

**Don't forget to attend the
Closing Banquet and Awards Ceremony
Wednesday, October 8 at 7:30 PM**

**Enjoy a meal with your colleagues,
congratulate the Scholander Awardee,
and listen to the Plenary Lecture
by Steven Chown**

**Get your *free ticket* at the
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The Crustacean Society

Thank you!

**2014 APS Intersociety Meeting
Comparative Approaches to Grand Challenges in Physiology**

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1.0: PLENARY LECTURE

1.1 OPENING THE BLACK BOX: HOW PHYSIOLOGY LINKS GENOMES TO ANIMAL FUNCTION

Patricia Schulte¹

¹Zoology, Univ. of British Columbia, 6270 University Blvd., Vancouver, B.C., V6T 1Z4, Canada.

Comparative physiologists are ideally placed to elucidate the links across levels of biological organization from genomes to the functions of animals in their environments—questions that are particularly relevant as we face the urgent task of predicting how animals may respond to anthropogenic environmental change. With the advent of new methods in molecular biology and genomics that allow the collection of unprecedented amounts of data at the molecular level, even in “non-model” species, the role of the physiologist is to open the black box that connects processes at the molecular level to functional variation at the level of organisms and populations in the context of a changing environment. Here, I provide an example of such an integrative approach examining how killifish (*Fundulus heteroclitus*) respond to stressors such as changes in temperature and oxygenation at levels from the gene to the organism. This work demonstrates how information at each level of organization enriches our understanding of processes at adjacent levels, allowing more nuanced predictions of how organisms respond to complex combinations of environmental parameters.

2.0: PHYSIOLOGICAL ADAPTATIONS TO EXTREMES: PROVIDING NOVEL ANIMAL MODELS FOR INVESTIGATING HEALTH AND DISEASE

2.1 IS PHYSIOLOGY REDUNDANT? WHY RODENT MODELS IN BIOMEDICAL RESEARCH ARE FAILING

Michael J. Joyner¹

¹Anesthesiology, Mayo Clinic, 200 First St., SW, Rochester, MN, 55905.

The purpose of this presentation is to discuss the limitations of rodent models in biomedical research. Many such models have been bred or engineered to generate genotype-equals-phenotype animals with specific susceptibilities for human clinical conditions like obesity, aging, Alzheimer's disease and others. Such results are then interpreted as part of a self-fulfilling prophecy indicating that genotype is deterministic for disease phenotype. Interventions that cure such animals are then developed only to fail when translation to humans is attempted. Why? Several possible explanations will be explored: 1) Genetically constrained animal models are also typically exposed to limited environmental and behavioral variability. This is not the case for humans and might explain some of the failure to translate. 2) Contrary to expectation, the Human Genome Project has shown no clear cut pattern of “risky gene variants” for essentially any complex non-communicable diseases in humans. Thus the bred or engineered models are false analogues for human disease. I close by calling for a “back to the future” approach that includes a wider appreciation of physiological redundancy and also a wider variety of animal models to understand human disease and facilitate translational research.

2.2 MAMMALIAN HIBERNATION AS A MODEL OF DISUSE OSTEOPOROSIS: THE EFFECTS OF PHYSICAL INACTIVITY ON BONE METABOLISM, STRUCTURE, AND STRENGTH

Seth W. Donahue¹

¹Mechanical Eng., Colorado State Univ., Flint Animal Cancer Ctr. (1620), 300 West Drake Rd., Fort Collins, CO, 80523-1620.

Physical inactivity leads to increased bone resorption, elevated serum and urinary calcium concentrations, bone loss, bone mechanical property loss, and increased fracture risk in humans and other animals. Grizzly and black bears, yellow-bellied marmots, and 13-lined ground squirrels do not lose bone mass or mechanical properties during prolonged (4–6 months) hibernation. Bone remodeling (i.e., bone resorption and formation) continues during hibernation, although at significantly reduced levels compared to summer levels. Hibernating bears are anuric, yet serum calcium concentration remains at homeostatic levels throughout the entire year. These findings suggest that hibernating bears and rodents have biological mechanisms to preserve bone tissue integrity when challenged with prolonged physical inactivity. Reduced bone remodeling in hibernators likely contributes to the conservation of metabolic energy. Neural signals and circulating factors (e.g., calcium regulatory hormones) likely contribute to the changes at the bone cell level that are involved in bone tissue preservation. Normal balance between bone resorbing osteoclasts and bone forming osteoblasts is likely maintained to preserve normal serum calcium concentrations during anuria. Identification of the molecular mechanisms that regulate bone cell function during hibernation may contribute to the development of new therapies for osteoporosis and inform our understanding of how hibernators have adapted to survive extreme environmental conditions. Funding from NIH (NIAMS AR050420).

2.3

EXTREME PHENOTYPIC PLASTICITY: HOW THE BURMESE PYTHON PROVIDES CLUES TO A HEALTHY HEART

Leslie A. Leinwand¹

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We employ the postprandial Burmese python as an animal model to discover novel pathways of beneficial cardiac adaptation. An infrequent feeder, the Burmese python exhibits a ~40 fold increase in overall metabolic rate and a 160 fold increase in plasma triglycerides within a day of eating a meal that can equal its body mass. This extreme metabolic demand activates rapid and reversible organ growth, including beneficial cardiac enlargement. Postprandial cardiac hypertrophy is accompanied by mechanisms that activate beneficial changes in gene expression and prevent cardiac lipid accumulation. Instead, the heart increases activity of genes involved in lipid handling, mitochondrial oxidative capacity, and free radical scavenging. Importantly, we identified a novel combination of 3 fatty acids (FAs) (myristic, palmitic, and palmitoleic) in postprandial python plasma that when administered to either pythons or mice promotes cardiac adaptation with no signs of pathologic lipid signaling. Moreover, we found that an aquaglyceroporin gene, aquaporin 7 (AQP7), is the most potently activated gene in rat cardiac myocytes treated with these FAs. We have made cardiac myocyte-specific AQP7 null mice and are examining its phenotypes. Since many aspects of the python postprandial cardiac responses resemble the beneficial mammalian cardiac response to exercise, we plan to explore the preventive and therapeutic potential of the FAs before or after a pathologic cardiac stimulus. We believe that the biology of the Burmese python will reveal novel signaling pathways that may lead to therapeutics for cardiovascular disease. (NIH 1R01HL119937-01).

2.4

HYPOXIA TOLERANCE IN THE VERTEBRATE BRAIN: INSIGHTS FROM COMPARATIVE PHYSIOLOGY

Philip Bickler¹

¹Anesthesia, Univ. of California, San Francisco, 513 Parnassus Ave., San Francisco, CA, 94143-0542.

Hypoxic and ischemic brain injury remains a major cause of human death and disability. With the exceptions of induced mild hypothermia (32°C) for neonatal asphyxia and adult survivors of cardiac arrest and thrombolysis for stroke, no neuroprotective therapy has proved effective. Numerous pharmacologic treatments have been studied clinically but have failed due to side effects or lack of efficacy. Dozens of therapeutic targets have been exploited in developing these treatments, based on molecular mechanisms of acute and delayed cell injury and death. The study of anoxia-tolerant vertebrates has provided important insights into the direction of some of this pharmacologic research, most notably in understanding the neuroprotective roles of adenosine and endogenous opioids. However, overall, it has proved exceedingly difficult to translate comparative models of anoxia-tolerance into clinical therapy. A major problem has been that rodent models have not predicted neuroprotection in humans. A major new area of interest is in enhancing recovery and repair programs, including new drugs and cell-based therapies (i.e. stem cells). Comparative physiology models may contribute to this understanding by exploiting the regeneration processes and synaptic remodeling that occurs after dormancy. Another major area of future contribution is understanding hypothermia tolerance since it is now recognized that mammalian neurons have limited tolerance of therapeutic hypothermia.

3.0: GENOMICS IN INTEGRATIVE AND COMPARATIVE PHYSIOLOGY

3.1

EVOLUTIONARY GENETICS OF ENERGETICS: EFFECTS OF MITOCHONDRIAL-NUCLEAR INTERACTIONS ON METABOLISM AND GENOME EVOLUTION

Kristi Montooth¹

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Effects of mtDNA mutations are predicted to be conditional on variation in nuclear genomes, because mitochondrial and nuclear gene products interact to fuel eukaryotic performance. We have found that interactions between mitochondrial tRNAs and their nuclear-encoded tRNA synthetases impact organismal traits via their effects on metabolism. I will show that tRNA synthetases that interact with mitochondrial tRNAs accumulate more molecular divergence across diverse taxa than do those that interact with cytoplasmic tRNAs, consistent with both the differential expression of mitochondrial and nuclear synthetases and the different rates of evolution in the two classes of tRNAs, and supporting models of molecular coevolution. Yet, the fitness effects of intergenomic interactions are complex. Effects of these interactions on metabolic rate, development time, pupation height and reproduction in *Drosophila* are highly conditional on the thermal environment. We find that deleterious effects of these interactions are ameliorated when larvae develop at 16°C and exacerbated at higher temperatures, consistent with temperature accelerating biological processes and increasing energy demand. Yet, even at 16°C, these genetic interactions impact larval metabolic thermal plasticity (i.e., the Q_{10} for metabolic rate). The expression of

intergenomic interactions in only some environments weakens the efficacy of selection on this type of genetic variation and may promote the accumulation of mitochondrial-nuclear incompatibilities whose fitness effects will depend upon the environment in which hybrids between closely related species occur. References: Meiklejohn et al. 2013 PLoS Genetics 9:e1003238. Hoekstra et al. 2013 Genetics 195:1129.

3.2 POLYMORPHISM IN OXYGEN EXCHANGE CAPACITY AND THE PHYSIOLOGY OF A MODEL ORGANISM FOR ECOLOGY

James Marden¹

¹Biology, Penn State Univ., 208 Mueller Lab, University Park, PA, 16802.

Glanville fritillary (*Melitaea cinxia*) butterflies, a model organism in ecology, have allelic variation in a metabolic enzyme (succinate dehydrogenase) that regulates the hypoxia inducible factor (HIF) pathway. One *Sdh* allele is associated with reduced SDH activity, elaboration of tracheoles in flight muscle, and better flight performance. Butterflies with less tracheal development have greater post-flight hypoxia signaling, mitochondrial damage, oxidation of membrane lipids, and aging of metabolic performance. Experimentally elevated succinate in pupae increased HIF-1 α and expression of genes responsive to HIF activation, including tracheal morphogenesis genes. In a separate study in Finland, allelic variation in *Sdh* and another metabolic enzyme locus (*Pgi*) interacted with patch size to explain a large portion of the variation in year-to-year size of local populations. These results indicate that the hypoxia inducible pathway, even in lowland populations, can be an important axis for genetic variation underlying intraspecific differences in oxygen delivery, physiological performance, life history and ecology. (NSF EF-0412651, IOS-0950416). References: Marden, J.H., Fescemyer, H.W., Schilder, R.J., Doerfler, W.R., Wheat, C.W. (2013) Genetic variation in HIF signaling regulates quantitative variation in physiological and life history traits within lowland butterfly populations. *Evolution* 67, 1105-1115. Wheat, C.W., Fescemyer, H.W., Kvist, J., Tas, E., Vera, J.C., Frilander, M.J., Hanski, I., Marden J.H. (2011) Functional genomics of life history variation in a butterfly metapopulation. *Molecular Ecology* 20, 1813-1828. Marden, J.H. (2013) Nature's inordinate fondness for metabolic enzymes: why alleles at metabolic enzyme loci are so frequently targets of selection. *Mol Ecol*, 22, 5743-5764.

3.3 COMPARATIVE PHYSIOLOGICAL GENOMICS OF SALINITY TOLERANCE

Andrew Whitehead¹

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Salinity limits the distributions of aquatic species, and most large clades of fish are either exclusively marine or freshwater. Yet, euryhaline species harbor the physiological plasticity necessary to invade new osmotic niches, and may thereby seed speciation across osmotic boundaries. Killifish (*Fundulus* sp.) is a comparative model system for studying the genomic basis of physiological plasticity in different osmotic environments. Euryhalinity is the ancestral state within the genus. Several estuarine species have derived extreme euryhalinity that has enabled exploitation of the entire continuum of osmotic niches. In contrast, repeated diversification across osmotic boundaries is particularly labile within the genus, coinciding with repeated assimilation of physiological plasticity in freshwater species. By comparing gill transcriptomic responses to osmotic challenges between populations and species, we are uncovering some of the molecular mechanisms, and the genomic architecture, that underpins osmoregulation, and that may represent molecular features that enable physiological plasticity in general. Regulation of paracellular permeability is a consistent strategy for compensating for osmotic challenge across taxa. Mechanisms associated with regulation of cell volume, polyamine synthesis, and ion transport, appear to consistently evolve between populations and species with different osmotic compensatory abilities. The functional genomic response to osmotic challenge is canalized in populations that have derived exceptional osmoregulatory abilities, and these genes appear to have simplified trans-regulatory complexity indicating modularity that may enable evolutionary fine-tuning.

3.4 TRANSCRIPTOMICS AS A TOOL OF FUNCTIONAL GENOMICS: POSSIBILITIES AND LIMITATIONS WITH HYPOXIA RESPONSE OF FISH AS A CASE STUDY

Mikko Nikinmaa¹, and Jenni Prokkola¹

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Oxygen availability has been an important factor driving the evolution of fish. Recently, hypoxia has become a major problem in aquatic ecosystems. Hypoxia responses in animals are often studied with transcriptomic techniques, such as quantitative PCR and microarrays. Their use calls for understanding what can be said about the observed changes. Naturally, only transcribed genes can be expressed, but the mRNA levels do not always correlate with changes in protein activity. The correspondence between mRNA and protein levels is protein-specific; e.g., the level of HIF (hypoxia inducible factor) is mainly post-transcriptionally regulated, and its in-

crease in response to hypoxia can occur without mRNA level changes. Gene expression in response to hypoxia can also be regulated at its energetically most costly level, translation, and by rapid mechanisms not involving *de novo* protein synthesis. Thus, transcriptional changes indicate the genes that should be studied further to reveal the affected functional pathways and the activities of gene products (proteins). Notably, hypoxia responses may depend on circadian rhythms of animals and can be affected by environmental pollution via mechanisms not currently well understood. Uncovering these mechanisms requires work both at transcriptional and protein activity levels, both of which are needed for full characterization of hypoxia responses keeping in mind that the transcriptional changes must be related to protein activity changes to evaluate the functional significance of the response.

4.0: FRONTIERS IN INSECT HOMEOSTASIS-ADVANTAGES AND EXPLOITATION

4.1

GPCR'S ROLE IN REGULATION OF DIURESIS OR WATER MOVEMENT IN INSECTS

Patricia V. Pietrantoni¹

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Insect renal organs, the Malpighian tubules, are simple epithelia useful to study hormonally controlled fluid transport. Renal fluid secretion is achieved in insects by the establishment of ion gradients that drive fluid towards the M. tubule lumen. This fluid is emptied into the hindgut for excretion of metabolic wastes, where water may be reabsorbed or excreted when in excess. Females of the mosquito *Aedes aegypti* (L.) feed mainly on human blood, which is hypoosmotic with respect to the mosquito hemolymph. Upon blood feeding, diuretic factors are released from the nervous system into the hemolymph, increasing the rate of secretion in the M. tubules, and allowing females to quickly excrete excess sodium and chloride ions, and water. We have investigated the G protein-coupled receptors for these diuretic factors in adult mosquitoes. These GPCRs are, in family A, 1) The insect kinin (leucokinin) receptor, and in family B, 1) The corticotropin-releasing factor-like (diuretic hormone 44) receptor and, 2) The calcitonin-like (diuretic hormone 31) receptor. A variety of approaches, including receptor functional analyses, peptidomimetics testing (SAR), IHC, RNAi and fluid secretion/excretion measurements in vitro and in vivo, were utilized to assess their function. The receptors are expressed in multiple tissues, have unusual, cell-specific spatial distribution patterns and, through their orchestrated diuretic and myotropic actions contribute to the successful arthropod adaptation to blood-feeding. Kwon, H.S. et al. PLoS One 2012; 7(11):e50374. (Texas AgriLife Research; USDA 2008-35302-18820).

4.2

PA1B, A NATURAL PEPTIDIC INSECTICIDAL AGENT AGAINST VACUOLAR H⁺-ATPASES

Markus Huss

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Vacuolar H⁺-ATPases (V-ATPases) are constituent enzymes in every eukaryotic cell. They are heteromultimeric proton pumps, which energize many essential transport processes across plasma- and endomembranes. In insects they play a key role for the physiology of the gut and the Malpighian tubules. Interference with the correct function of the V-ATPase either by mutations or by inhibitors is usually lethal. Therefore the V-ATPase is an attractive target for the development of insecticides. Recently, PA1b, a peptide derived from peas (*Pisum sativum*) was described to be the first peptidic and insect specific V-ATPase inhibitor. The structure of PA1b exhibits a unique cysteine knot fold which is also common to toxins from e.g. cone snail, spider and scorpion. However, its natural origin from a plant being a component of the regular human diet makes PA1b highly attractive for the exploration of its binding site and inhibition mechanism for the further development as a potential bio-insecticide for pest control. Using a radioactive labeled PA1b in UV-induced cross linking experiments and a second derivative modified with biotin-streptavidin-HRP in single particle electron microscopy, it was possible to localize the binding site of PA1b at the interface of subunits c and e. This led to the development of the first idea how PA1b silences the H⁺-transport across the membrane. Funding: Deutsche Forschungsgemeinschaft (SFB 431, SFB 944).

4.3

REGULATION OF ION TRANSPORT BY TYRAMINE IN THE DROSOPHILA MALPIGHIAN TUBULE: IDENTIFICATION OF MULTIPLE TYRAMINE RECEPTORS

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The biogenic amine tyramine (TA) is an important signaling molecule in insects, with roles in reproduction and locomotion. We have previously identified TA as a potent diuretic agent in the *Drosophila* Malpighian tubule (MT). Application of nanomolar TA to isolated MTs causes a rapid depolarization of the transepithelial potential associated with an increased transepithelial chloride conductance, resulting in diuresis.

We and others have found that the action of TA is associated with calcium signaling in the stellate cells of the MT (Blumenthal 2003, Cabrero et al, 2013). To identify the TA receptor, we studied tubules mutant for the previously-reported TA receptor *TyrR*. The allele *TyrR⁶⁵⁰²*, which lowers expression by >100-fold, or knockdown of *TyrR* expression in the stellate cells, reduces but does not eliminate TA sensitivity. Deletion of the adjacent homologous gene *TyrRII* has no effect. In contrast, deletion of both *TyrR* and *TyrRII* eliminates TA sensitivity, as does knockdown of *TyrRII* expression in *TyrR⁶⁵⁰²* mutants. We conclude that while *TyrR* encodes the primary TA receptor in the MT, *TyrRII* also contributes to the TA response. Surprisingly, mutation or knockdown of *TyrR* also eliminates the response of MTs to the related amine octopamine (OA), suggesting that OA can also activate TyrR. This finding contrasts with the function of *TyrR* in heterologous cells, where it displays high selectivity for TA over OA. NSF IOS-0744619. Reference: G Cazzamali, DA Klaerke, CJP Grimmelikhuijzen. A new family of insect tyramine receptors. BBRC 338, 1189-96 (2005). Cloning and functional characterization of *Drosophila TyrR*.

4.4 EXPLOITING THE RENAL HOMEOSTATIC MECHANISMS OF MOSQUITOES FOR NOVEL VECTOR CONTROL

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¹Entomology, The Ohio State Univ./Ohio Agricultural Res. and Dev. Ctr., 1680 Madison Ave., Wooster, OH, 44691, ²Biomedical Sci., Cornell Univ., Tower Rd., Ithaca, NY, 14853, ³Anesthesiology and Pharmacology, Vanderbilt Univ., 1161 21st Ave., S., Nashville, TN, 37232.

The efficacy of insecticides used to control mosquito vectors of disease is eroding due to the emergence of resistance. Thus, new chemicals are needed to improve our capabilities for mosquito control. Here we review efforts by our group that aim to disrupt inward-rectifying K⁺ (Kir) channels expressed in the renal (Malpighian) tubules of mosquitoes; we hypothesized that inhibiting renal Kir channels would disrupt hemolymph K⁺ homeostasis in mosquitoes with lethal consequences. We describe: 1) the expression, localization, and functional characterization of Kir channels in the Malpighian tubules of adult female mosquitoes (*Aedes aegypti*); 2) the discovery of small molecule inhibitors of mosquito Kir channels; and 3) the disruption of mosquito renal functions by these inhibitors. Our data indicate that the mosquito Malpighian tubules and Kir channels are valuable physiological and molecular targets, respectively, for the development of novel insecticides. Funded by a grant from the Foundation for the NIH, VCTR program. Raphemot, R., Rouhier, M.F., Hopkins, C.R., Gogliotti, R.D., Lovell, K.M., Hine, R.M., Ghosalkar, D., Longo, A., Beyenbach, K.W., Denton, J.S., and Piermarini, P.M. Eliciting renal failure in mosquitoes with a small-molecule inhibitor of inward-rectifying potassium channels. *PLoS One* 8: e64905, 2013.

5.0: CARDIORESPIRATORY PHYSIOLOGY OF VERTEBRATE EXTREMOPHILES

5.1 BREATHE DEEP TO DIVE DEEP: RESPIRATORY VOLUMES IN PENGUINS

Paul Ponganis¹

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The penguin respiratory system constitutes 30-45% of the total body oxygen store. The ratio of air sac volume to lung air capillary volume may also prevent barotrauma to the rigid avian lung, and limit maximum dive depth. In order to assess lung and air sac volumes, computerized tomographic scans and 3D anatomical reconstructions were conducted on anesthetized animals. In Adelie, king, and emperor penguins, respectively, mass specific lung volumes were 25, 19, and 18 ml kg⁻¹, consistent with allometrically predicted values, while maximum air sac volumes (at 30 cm H₂O inflation pressure) were 296, 357, and 363 ml kg⁻¹ (2.2, 2.9, and 3.0 x allometric predictions, and also greater than end-of-dive total diving air volumes estimated in free-diving penguins). If emperor penguins inhale to such a large air volume prior to deep dives, the total body oxygen store would be increased from prior estimates by 71% to 116 ml O₂ kg⁻¹ with 61% of all O₂ in the respiratory system. Based upon previous lung morphometry, and the measured lung volumes and maximum air sac volumes, the air sac to lung air capillary volume ratios were 66, 105, and 108, respectively, in the three species. These ratios are equivalent to safe compression to depths of 650, 1040 and 1170 m, all greater than their maximum recorded depths. Whether penguins can maximally inspire to such a large air sac volume is unknown, but larger than predicted air volumes have been previously documented in other diving birds. In addition, exhalation of air prior to ascent may decrease measured end-of-dive air volumes in free-diving penguins. More physiological and morphometric studies are required.

5.2 METABOLIC STRATEGIES FOR SURVIVING THE WINTER IN HIBERNATING MAMMALS

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Mammals that hibernate are most known for their ability to lower body temperature, but body temperature changes are a consequence of genomic and physiological repro-

gramming that alter energy demand and metabolic fuel use. It is these adaptive changes that allow a variety of species to withdraw from the environment and subsist solely on endogenous energy and/or stored food for up to 9 months. This talk will review our research on hibernators that represent physiological extremes for their ability to survive metabolism reduced to 2% basal rates and body temperatures of -3 °C - arctic ground squirrels - and metabolism reduced to 25% basal with only a 3-4 °C reduction in body temperature-black bears. Changes in the cardiorespiratory systems of hibernating black bears include the development of profound sinus arrhythmias between heart beat rate and breathing and a fatal susceptibility to cardiac arrhythmias and fibrillation if body temperature declines below 30 °C. In both species metabolic fuel use is restricted to fats and ketones, which, along with reduction in metabolic demand and a desensitization of immune response, is hypothesized to protect the heart and brain from damage due to low blood flow rate and, in extreme experiments in ground squirrels at body temperatures of 30 °C, heart stoppage for 45 minutes followed by reperfusion.

5.3 CARDIORESPIRATORY PERFORMANCE IN ANOXIA TOLERANT FISH AND REPTILES

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Shallow lakes and ponds in Northern Europe often become anoxic for several months every winter due to ice coverage. The only fish that survives in these waters is the crucian carp (*Carassius carassius*), the wild cousin of the goldfish (*C. auratus*). The crucian carp is arguably the most anoxia-tolerant fish species. Among vertebrates, its anoxia tolerance is only matched by that of some North American freshwater turtles (genera *Trachemys* and *Chrysemys*). However, unlike turtles, the crucian carp remains active during anoxia. The key adaptation allowing a high level of glycolytic ATP production in anoxia is its ability to convert lactate to ethanol. Turtles lack this ability and are forced to enter into deep metabolic depression to limit the lactate load. In addition, when faced with hypoxia, the crucian carp has a remarkable ability to remodel its gills, resulting in a 7 fold increase in the respiratory surface area, which of course boosts its capacity for oxygen uptake. In contrast to turtles, which strongly suppresses cardiac work in anoxia, crucian carp maintain cardiac out-put, and also ventilation, at normal levels even after several days in anoxia. This suggests that being active in anoxia demands an active circulatory system for shuttling glycolytic substrates and removing ethanol. At the same time, it appears that it is the ability of the crucian carp to avoid lactic acidosis allows its heart to function in anoxia. Supported by the Research Council of Norway.

5.4 EVOLUTION OF CARDIORESPIRATORY PHYSIOLOGY IN HIGH-ALTITUDE MAMMALS AND BIRDS

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The hypoxic and cold environment at high altitudes requires that endothermic animals sustain high rates of O₂ consumption for both locomotion and thermogenesis while facing diminished O₂ availability. We are examining the integrative respiratory mechanisms that are used by highland mammals and birds to overcome this challenge and help maintain performance during hypoxia. High-altitude adaptation appears to have altered several steps in the oxygen transport pathway of many highland taxa to improve O₂ supply in hypoxia. These adaptations include a more effective hypoxic ventilatory response, large lungs, haemoglobin with a high O₂ affinity, changes in the control of blood flow, and a dense capillary network in the skeletal muscle and heart. The increases in O₂ supply that are afforded by these unique traits support mitochondrial respiratory capacities that are maintained or even elevated in highlanders compared to lowlanders. The transcriptomic basis for increases in capillarity and metabolic capacity in the skeletal muscle includes changes in the expression of metabolic genes as well as regulators of metabolism and angiogenesis. As a consequence of their unique physiology, highland animals can sustain impressive feats of aerobic performance in the extremely harsh environment at high elevation. (Supported by NSERC of Canada).

6.0: DIVERSE APPROACHES IN EVOLUTIONARY PHYSIOLOGY

6.1 COMPARATIVE GENETICS AND GENOMICS OF BEHAVIORAL PHENOTYPES IN MICE: LESSONS FOR EVOLUTIONARY PHYSIOLOGY

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Genes, the environment, and gene-by-environment interactions simultaneously influence complex traits, such as behavior. Moreover, underlying the genetic architecture and the environment are individual-specific elements (e.g., pleiotropy, diet) that each contributes to the total phenotypic variation. Utilizing emerging and established mouse resources (e.g. Advanced Intercrosses, Collaborative Cross, Diversity Out-

bred) studies have begun to reveal the genetic determinants of behavioral phenotypes with increasingly high precision. Investigations have utilized linkage, genome-wide association, and expression approaches in an attempt to not only identify genetic variants underlying behavior, but also to better understand how these variants may be functioning in physiologically complex systems that are phenotypically plastic. Voluntary physical activity is one example of a behavior where systems approaches have been utilized to simultaneously understand the underlying genetic architecture, physiological systems, and their interactions. Although investigations into the biological basis of exercise have distinct human health implications, locomotion is also clearly important from an ecological and evolutionary perspective. In general, utilizing laboratory mouse resources to uncover the genomic architecture may lead to a more comprehensive understanding of adaptive behavioral phenotypes, the physiological systems that underlie them, and the potential for both to respond proximately and evolutionarily. References: Kelly S. A., et al. Genetic determinants of voluntary exercise. *Trends Genet.* 29:348-357, 2013. Logan RW, et al. High precision genetic mapping of behavioral traits in the diversity outbred mouse population. *Genes Brain Behav.* 12:424-437, 2013. Threadgill DW, et al. Ten years of the collaborative cross. *Genetics* 190:291-294, 2012.

6.2

PHYSIOLOGY AND GENOMICS OF THE EVOLUTION OF LIFE HISTORIES AND SENESENCE IN GARTER SNAKES

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For a species to persist in a changing environment, individuals need to respond appropriately to physiological/environmental stress, and the molecular networks underlying these responses need to acclimate or evolve. Evidence from laboratory model systems indicate that molecular networks regulating stress response may often be the networks underlying life-history trade-offs between longevity and growth reproduction. To understand how these molecular networks are functioning in natural populations, we are comparing stress responses between naturally evolved, closely related populations of garter snake ecotypes. These ecotypes include a fast-living ecotype with faster growth, higher reproductive output, and shorter lifespan relative to a slow-living ecotype. In laboratory experiments, we have used heat stress as an activator of metabolic and oxidative stress responses to investigate if these molecular networks have diverged in between the ecotypes. Our results indicate that the ecotypes respond differently to heat stress in their 1) levels of circulating free radicals (superoxide and hydrogen peroxide), 2) production of hydrogen peroxide in the liver mitochondria, 3) amount of DNA damage in blood cells, 4) innate immune function, and 5) liver gene expression. Furthermore, we have identified genetic variants in genes related to oxidative stress that differentially associate with these ecotypes. These results support the hypothesis that closely situated populations with divergent life histories can evolve different responses to physiological stress. (NSF IOS0922528, DEB1011350, IOS1253896, T. Schwartz & AM Bronikowski, 2013. Dissecting molecular stress networks: identifying nodes of divergence between life-history phenotypes *Molec Ecol* DOI:10.1111/j.1365-294X.2012.05750.x.)

6.3

CAUSES OF PARALLEL BIOCHEMICAL ADAPTATION: INSIGHTS FROM HEMOGLOBINS OF HIGH-ALTITUDE VERTEBRATES

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Is it possible to predict which molecular mechanisms are most likely to contribute to biochemical adaptation? Can we predict which mutations – or which types of mutation – are most likely to contribute to adaptive changes in protein function? To address these questions about the inherent predictability of adaptive evolution at the molecular level, we are conducting systematic comparative studies of hemoglobin (Hb) function in high-altitude vertebrates. Specifically, we are conducting phylogenetically replicated comparisons of Hb function between high- and low-altitude species of birds and small mammals. This work combines evolutionary analyses of sequence variation with protein engineering experiments based on site-directed mutagenesis. Results to date suggest several mechanistic explanations for why parallel changes in protein function may or may not involve parallel substitutions at the amino acid level. Grant funding: NIH-HL087216. References: Natarajan, C., N. et al. (2013). Epistasis among adaptive mutations in deer mouse hemoglobin. *Science* 340: 1324-1327. Projecto-Garcia, J., et al. (2013). Repeated elevational transitions in hemoglobin function during the evolution of Andean hummingbirds. *Proceedings of the National Academy of Sciences USA* 110: 20669-20674. Revsbech, I., et al. (2013). Hemoglobin function and allosteric regulation in semi-fossorial rodents (family Sciuridae) with different altitudinal ranges. *Journal of Experimental Biology* 216: 4264-4271.

7.0: RECENT IDEAS AND TECHNOLOGICAL ADVANCES IN COMPARATIVE EPITHELIAL PHYSIOLOGY

7.1

RAINBOW TROUT USE ACID SENSING ION CHANNELS (ASICS) FOR NA⁺ UPTAKE IN DILUTE FRESHWATER

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The current model for Na⁺ uptake in rainbow trout gill proposes two mechanisms: NHE-Rh protein metabolon and/or epithelial Na⁺ channel (ENaC) coupled to H⁺-ATPase. However, while extensive evidence for NHE mediated Na⁺ uptake has been gathered, there is no molecular evidence for existence of ENaC. We propose that in rainbow trout the role of epithelial sodium channels is served by acid-sensing ion channels, members of the ENaC/DEG family. We cloned ASIC gene homologues ASIC1 and ASIC4 from the gill and demonstrated their expression in mitochondrion-rich cells. Moreover, using immunohistochemistry we co-localized ASIC protein to the MRCs rich in NKA. We also report that in adult rainbow trout, ASIC specific inhibitors decreased Na⁺ uptake in a dose-dependent manner. Our findings suggest that ASIC channels play a role in Na⁺ uptake in freshwater fish, and therefore we propose that they provide an alternative mechanism for Na⁺ uptake to NHEs for rainbow trout in very low ionic strength waters. Supported by an NSERC Discovery grant to GGG. References: Dymowska A. K., Schultz A. G., Blair S. D., Chamot D., Goss G. G. (2014) Acid-sensing ion channels (ASICs) are involved in epithelial Na⁺ uptake in rainbow trout (*Oncorhynchus mykiss*). *American Journal of Physiology-Cell Physiology* Published 4 June 2014 Vol. no. DOI: 10.1152/ajpcell.00398.2013.

7.2

FISH INTESTINAL HCO₃⁻ SECRETION: FROM MOLECULES TO THE OCEANIC INORGANIC CARBON CYCLE

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Hypertonic fluid absorption by the marine teleost intestinal epithelium is driven by Na⁺ and Cl⁻ absorption, with ~50% of Cl⁻ absorption being attributable to anion exchange resulting in high luminal concentrations of HCO₃⁻. This luminal HCO₃⁻ reacts with Ca²⁺ to form CaCO₃ precipitates, a reaction that reduces luminal osmotic pressure by as much as 100 mOsm. Further reductions in osmotic pressure occur as H⁺ secreted by distal intestinal segments is titrated with luminal HCO₃⁻, facilitating continued water absorption. Intestinal anion exchange is thus important for solute coupled as well as osmotic fluid absorption and is mediated by the apical a6 member of the SLC26 transporter family, an electrogenic nHCO₃⁻/Cl⁻ exchanger. HCO₃⁻ for intestinal secretion comes from hydration of endogenous CO₂, catalyzed by cytosolic carbonic anhydrase (CAc), and HCO₃⁻ imported across the basolateral membrane via the NBC1 member of the SLC4 transporter family, the latter forming the rate limiting step for intestinal HCO₃⁻ secretion rates. The formation of CaCO₃ precipitates appears to be facilitated by matrix proteins and the release of these CaCO₃ precipitates to the environment may be mediated by a diuretic response. The distal intestine responds to peptides of the guanylin family with a transient reversal to a secretory Cl⁻ current mediated by NKCC1 and CFTR, a response which is associated with a secretory water flux. The excretion of CaCO₃ precipitates contributes to the inorganic oceanic carbon cycle and likely explains observations of increased titratable alkalinity with depth above the aragonite lysocline (Ω=1).

7.3

THE NA-K-CL COTRANSPORTER: RECENT ADVANCES IN STRUCTURE, FUNCTION, AND REGULATION

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Na-K-Cl cotransporters (NKCCs) mediate the movement of Na⁺, K⁺, and Cl⁻ across the cell membrane. NKCC1 is expressed in most vertebrate cells and is part of the Cl⁻ secretory pathway in salt-secreting epithelia. NKCC2 is responsible for Na⁺ and Cl⁻ reabsorption in the kidney. NKCCs have cytoplasmic N- and C-termini and a central 12-transmembrane (TM) domain. Activation of NKCCs occurs through phosphorylation of threonine residues in the N-terminus, whereas ion translocation occurs in the TM domain. How the event of phosphorylation in the N-terminus is translated to the TM domain to activate ion transport is completely unknown. To define structure-function relationships of NKCCs, we examined both inter- and intramolecular motions of NKCC1. Using fluorescence resonance energy transfer, we demonstrate that activation of NKCC1 is accompanied by a large movement between two positions in the C-termini of a dimeric cotransporter. Using a cysteine cross-linking approach, we demonstrate that NKCC1 activation involves conformational change at the TM10 and TM11/12 interface. Our results suggest that NKCC activation involves movement of TM12 relative to TM10, which may be tied to movement of the cytoplasmic C-terminus. We provide a novel model for understanding the molecular

motions that bring about NKCC activation. Support: NIH GM083340 and NIH P01-DK17433 to B. Forbush.

7.4 EVOLUTIONARY CONSERVED MECHANISMS FOR ACID/BASE SENSING

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Epithelial cells can achieve diverse physiological functions that rely on the transport of ions, including acid/base (A/B) and osmotic regulation, nutrient absorption, and calcification. Despite the functional diversity of epithelia, many of these functions are achieved by the combined function of the enzymes V-type ATPase (VHA), carbonic anhydrase (CA) and soluble adenylyl cyclase (sAC). The general mechanism involves hydration of metabolic or externally derived CO₂ by CA into HCO₃⁻ and H⁺. HCO₃⁻ stimulates the A/B sensor sAC, which can modulate the activity of multiple downstream targets via the cAMP pathway, while H⁺ is promptly transported out of the cell by VHA. The ultimate physiological function of the CA/sAC/VHA complex is determined by the polarization of proteins to basolateral or apical membranes that result in differential permeability to CO₂/HCO₃⁻, secretion or absorption of ions, and presence of other cell-specific accessory proteins. The CA/sAC/VHA complex has already been established to be essential for A/B regulation in shark gills and mammalian nephron, and for NaCl and water absorption in the marine teleost intestine. Other epithelia in which the CA/sAC/VHA promise to play essential roles include mantle and hemocytes from mollusks, the "root" epithelium from the bone-eating worm *Osedax*, and corals, as well as multiple epithelia from vertebrates. Funded by the National Science Foundation (#EF-1220641) and by an Alfred P. Sloan Research Fellowship (#BR2013-103).

8.0: METABOLISM, ENERGETICS, AND NUTRITION

8.1 INVESTIGATION OF ALGAL FEEDING PREFERENCES OF THE SOLAR-POWERED SEASLUG, *ELYSIA CLARKI*

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The sacoglossan sea slug, *Elysia clarki*, is one of only four species of sea slugs that is capable of incorporating the chloroplasts from different algal species into their own bodies and utilizing them long term for up to 3 to 4 months. The chloroplasts remain functional, allowing *E. clarki* to derive nutrition from ongoing photosynthesis, which makes this animal part of a quite unique symbiosis. Little information is available on how the chloroplasts of algal species maintain their structural and functional integrity following phagocytosis into the digestive tract where they are stored. Furthermore, associations with several algal species have been reported for *E. clarki*, but the nature of these associations is currently unclear. Feeding experiments were administered to determine which macroalgal species *Elysia clarki* will eat, and to investigate if there are any differences in feeding characteristics between adults and juveniles. The macroalgal species tested include: *Bryopsis hypnoides*, *Caulerpa brachypus*, *C. prolifera*, *C. racemosa*, *C. sertularioides*, *Halimeda incrassata*, and *Penicillus capitatus*. Sea slugs were presented with each of these species, and the amount of algae consumed and growth rate of the *E. clarki* were measured. Preliminary results from this experiment indicate that *Elysia clarki* do not consume *C. brachypus*, but will eat *B. hypnoides*. To our knowledge, these feeding associations represent previously unreported symbioses of *Elysia clarki* with macroalgae.

8.2 LIGHT-INDUCED OXIDATIVE STRESS AND DEFENSE IN THE MANTLE OF A GIANT CLAM

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The giant clam, *Tridacna squamosa*, has symbiotic microalgal zooxanthellae that can transfer photosynthetic products to the clam as food. However, algal photosynthesis in the presence of light increases the formation of oxygen and reactive oxygen species (ROS), increasing susceptibility of the clam to oxidative stress. The exodus of photosynthetic products may also induce downstream physiological responses in the clam. We examined the antioxidant response in the inner and outer mantle of clams in 12 h of light or 12 h of darkness, and hypothesized that the mantle would increase metabolic activity in the presence of light and display an enhanced antioxidant response. Results showed that 12 h of light exposure significantly increased activities of several antioxidant enzymes in the inner mantle, indicating light-induced oxidative stress, although oxidative damage products remained unchanged in both the inner and outer mantle. More importantly, they revealed that the inner mantle was metabolically more active, thereby producing more ROS and increasing oxidative defense, under light exposure, which warrants further investigation. This study was funded by SMF through TMSI.

8.3

PHYSIOLOGICAL RESPONSES TO ENVIRONMENTAL STRESS IN A MOLLUSC: WHY IS BEING A HYBRID AN ADVANTAGE?

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In Australian mollusc aquaculture, two abalone species, *Haliotis rubra* and *H. laevis* are cross-bred to create a hybrid that shows 25% faster growth. On land-based farms animals are held in water sourced from the ocean, which varies in temperature and oxygen (O₂) level. Yet, tanks are supplied with O₂ until abalones are 5 months old. Thus, temperature stress is experienced by individuals throughout their life on the farm, while O₂ stress is a factor experienced by older individuals. Suboptimal environmental conditions, management practices and parental origin may change the behaviour of an individual. Furthermore, under suboptimal conditions, energy is used to mitigate stress rather than channelling it into growth. This study examined how *H. laevis*, *H. rubra* and their hybrid cope with temperature and O₂ stress to reveal the physiological and behavioural strategies which result in the improved fitness of the hybrid. Respiration rate (MO₂) and activity level of abalones were measured during short-term exposure to 10, 15, 19 and 23°C and decreasing oxygen levels (21–0 kPa). Experiments were conducted with 4 and 8 month old individuals, to reveal different strategies not only between types of abalones but also across development. Four month old hybrids had a similar tissue mass but had grown larger shells in comparison to the two pure species. Yet, all types of abalone had similar MO₂, which correlated positively with increasing temperatures and negatively with O₂ levels.

8.4

MITOCHONDRIAL MECHANISMS OF HYPOXIA TOLERANCE IN MARINE BIVALVES

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Marine organisms are exposed to oxygen deficiency in estuaries due to the tidal cycles and/or the benthic "dead zones". Energy limitation and oxidative damage are major stressors during hypoxia and post-hypoxic recovery, and it is not well known how mitochondria of hypoxia-tolerant organisms cope with these challenges. We studied mitochondrial responses to hypoxia in the hard clam *Mercenaria mercenaria* and the bay scallop *Argopecten irradians*. Membrane potential (Δψ) and kinetics of substrate oxidation, proton leak and phosphorylation subsystems were measured in clams and scallops exposed to hypoxia (17 h at <1% O₂) followed by a 1 h recovery. In scallops, hypoxia suppressed the capacity of all three mitochondrial subsystems, and mitochondrial condition further deteriorated during reoxygenation, with strong depolarization of mitochondria and decreased capacity for the substrate oxidation and phosphorylation. In contrast, in clams hypoxia increased the Δψ-dependent capacity of the substrate oxidation subsystem and had weak inhibitory effects on the phosphorylation and proton leak subsystems. During reoxygenation, the substrate oxidation capacity of clam mitochondria further increased and the capacity of the phosphorylation subsystem returned to normal. Upregulation of the substrate oxidation in hypoxia poises clams for a quick recovery upon reoxygenation, while scallops suffer from mitochondrial deterioration limiting their ability to survive hypoxia. Supported by UNC Charlotte

8.5 WITHDRAWN

8.6

SCORPION BURROW STRUCTURE AND VENTILATION

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Many animals spend much of their time underground in burrows, which serve as refuges from predators and adverse environmental conditions. The intimate association between animal and burrow leads to the question, how has burrow architecture been shaped by natural selection? Burrowing scorpions are found on all continents except Antarctica. We are using scorpions and their burrows to test the hypothesis that burrow structure acts as a physiological control system to regulate temperature and moisture levels. We predict that burrows are built to minimize convective ventilation in order to maintain high relative humidity thereby reducing the occupant's evaporative water loss, with burrow structure being modified depending on soil moisture and temperature. We cast 20 natural burrows of *Scorpio maurus palmatus*, and four of *Opisthophthalmus setifrons* with molten aluminum and used a 3D scanner to capture burrow shape and dimensions. We related how structure differs with soil moisture, temperature, and body size, while testing which features are common among individuals and species. We found that burrow volume decreases as soil

moisture increases and all burrows have in common a horizontal platform just below the surface. Little is known about scorpion burrows, especially how this environment functions to meet their physiological needs, our findings demonstrate how an ectotherm modifies their surroundings to act as external organs of physiology. Funded by the Israel Science Foundation.

8.7 THE EFFECT OF HUMIDITY ON THE METABOLIC RATE OF GROMPHADORHINA PORTENTOSA

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Environmental humidity is a significant part of the ecosystem for all living organisms. Animals cope with a particular range of humidity levels depending on the habitat that they are found in by employing behavioral and physiological adaptations. However, how organisms will deal with the rapid changes in ambient humidity predicted with global climate change is still unclear. Previous studies have suggested that insects may decrease their metabolic rates, and alter their respiratory patterns, when ambient humidity is decreased. In this study we used *Gromphadorhina portentosa*, an insect normally found in humid environments, to investigate how these insects respond to a rapid change in ambient humidity. Using flow-through respirometry we measured the resting metabolic rates of adult, male roaches when placed in decreasing humidity levels (80, 70, 60, 50, 40, 30 and 0% R.H.). We found that as relative humidity decreased, the resting metabolic rates of these roaches significantly decreased. Furthermore, roaches showed a continuous respiratory pattern at high humidity levels but switched to a discontinuous pattern at low humidity levels. We therefore conclude that this organism has the ability to conserve water through a physiological and behavioral adaptation.

8.8 ACTIVATION OF cGMP-DEPENDENT PROTEIN KINASE REDUCES DROSOPHILA S2 CELL INJURY CAUSED BY ANOXIA AND OXIDATIVE STRESS

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Ischemic stroke is one of the leading causes of human death worldwide. It occurs due to the high susceptibility of neurons to anoxia and reoxygenation. Unlike mammals, the fruit fly *Drosophila melanogaster* withstands low oxygen levels without showing pathology. In the present study, *Drosophila* Schneider (S2) cells were employed to investigate the role of the cGMP-dependent protein kinase (PKG) signaling pathway in *Drosophila* anoxia tolerance. Cells were subjected to chemical anoxia and oxidative stress concurrently with treatments by pharmacological agents affecting specific targets of the PKG pathway and cell injury was assessed. Treatment of S2 cells with the PKG pathway activators 8-Br-cGMP or sildenafil citrate reduced cellular damage compared to cells exposed to CoCl₂ or H₂O₂ alone. Results thus indicate that activation of the PKG pathway preserves S2 cell plasma membrane integrity from chemical anoxia and oxidative stress. The results of this study may lead to a better understanding of the fruit fly's innate strategies of anoxia tolerance. Subsequently, this knowledge may be used to identify potential therapeutic targets to prevent detrimental neurological effects of an ischemic stroke in humans. The study was funded by FAU Division of Research SEED Grant, McGinty Foundation Grant, and Research Priority Grant: Brain Function, Damage and Repair.

8.9 METABOLISM AND LOCOMOTION OF ANOXIC DROSOPHILA

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Although it is well known that insects have the highest aerobic metabolic rates in the animal kingdom during flight, a much less appreciated trait is their ability to survive and perform under anoxic conditions. Despite being a premier model for organism for the study of anoxia tolerance, we still lack a basic understanding of the physiological responses of *Drosophila* to anoxia. Here we present data on survival, locomotion, heat production (calorimetry), gas exchange and multiple measures of metabolites for larvae and adult *Drosophila melanogaster* in anoxia. Additionally, we conduct a Genome-Wide Association Study using the *Drosophila* Genetics Reference Panel (DGRP) to identify genes correlated with variation in anoxia survival. Adults become quickly paralyzed by anoxia, larvae continue to exhibit escape locomotion for over 30 minutes powered by high rates of lactate production. While both stages produce lactate during anoxia, larvae produce lactate at a 3x higher rate. Adults and larvae had similar capacities to suppress metabolism with longer-term anoxia; at 1-3 hours of anoxia, both larvae and adults suppress metabolic heat production to 3% of normoxic rates. In larvae, heat production can be quantitatively explained by lactate production, but other end-products must be important in adults. Interestingly, nearly 100% of adults survive anoxia up to 4 hours while less than 50% of third instar larvae survive anoxic bouts greater than 90 minutes. Our data suggest that historical ex-

posure to anoxic micro-sites may have selected for metabolic and behavioral traits that allow escape in larvae, at a cost of reduced survival. *Drosophila* adults retain the more common pattern among insects, a strategy that may be particularly useful for survival of rain or flooding. Supported partially by NSF IOS 1256745.

8.10 SMALL RNA REGULATION OF DIAPAUSE IN THE FLESH FLY, SARCOPHAGA BULLATA

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Understanding the molecular basis of diapause, a phenotypically plastic, alternative developmental pathway that includes a period of dormancy, is one key to predicting the impact of global climate change on seasonal distributions of insects. Diapause, which is characterized by developmental arrest and metabolic restructuring, is associated with wide-spread changes in gene expression. Though diapause-related changes in gene expression have been characterized for species ranging from crickets to mosquitoes, the mechanisms that mediate the observed changes remain largely unknown. We predict that small regulatory RNAs (sRNAs), one type of epigenetic mechanism, mediate pupal diapause in the flesh fly, *Sarcophaga bullata*. Specifically, elevated expression of genes encoding core components of the piwi-RNA and small-interfering RNA pathways in photosensitive first instar larvae reared in diapause-inducing conditions compared to those reared in diapause-averting conditions, suggests a role for these sRNAs in diapause initiation. In addition, a 2-fold increase in *Argonaute1*, a core component of the microRNA pathway, in diapausing pupae suggests a role for this class of sRNAs in regulating diapause maintenance. Next-generation sequencing of small-RNAs isolated from diapause and non-diapause pupae will provide significant new information about the role of these regulatory molecules in initiating and maintaining diapause [National Science Foundation Grant IOS-1354377].

8.11 HOW DO YOU LIKE YOUR EGGS? EGG CANNIBALISM AND DIGESTIBILITY IN THE CALIFORNIA GRUNION, LEURESTHES TENUIS (TELEOSTEI: Atherinopsidae)

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California grunion (*Leuresthes tenuis*) spawn on the beach during spring tide events. In preparation for spawning, adult grunion fast and have empty guts. Recently, grunion have been found with conspecific eggs in their intestines after spawning, leading to a hypothesis that their eggs provide a potential food resource. However, grunion eggs are structurally resilient, withstanding up to six developmental weeks buried in beach sand, and in vitro tests have failed to destroy eggs with formalin or commercial digestive enzyme preparations. We examined egg digestibility in grunion to determine if the fish can digest their eggs. Grunion were separately fed fertilized and unfertilized eggs, and serially dissected over 10 hours. Comparisons of egg numbers, and egg visual quality, in the proximal, mid, and distal intestine showed unfertilized and fertilized eggs being broken down and disappearing (i.e., digested) during the experiment. The amount of force (N) needed to crush eggs taken from grunion digestive tracts was significantly lower than that needed to crush uneaten fertilized and unfertilized eggs. Analyses of nutrient concentrations (protein, lipid, and carbohydrate) of eggs recovered from different intestinal regions, and digestive enzyme activities in those intestinal sections, are underway and should affirm egg digestibility. Overall, our study confirms that grunion are capable of digesting their eggs, and thus, this food resource may be important after spawning.

8.12 THE EFFECT OF DIET AND INTESTINAL MICROBIOME MANIPULATION ON INTESTINAL CELLULAR PATHWAYS

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The diet affects cellular pathways in fish enterocytes. For example, carnivorous diets enhance protein catabolic pathways, while herbivorous diets enhance carbohydrate catabolic pathways. We proposed that manipulation of the microbiome inhabiting the gastrointestinal tract (GIT) will likewise affect cellular pathways. Furthermore, manipulation of both the diet and microbiome would produce additive effects. We used antibiotics to eliminate the GIT microbiome in zebrafish. We then re-introduced a native GIT microbiome from zebrafish, or a non-native GIT microbiome from goldfish. Additionally, we manipulated the diet, providing either standard zebrafish food or goldfish food. Subsequently, we analyzed several intestinal enzymatic pathways at a tissue level, as well as intestinal transcriptional regulation using a specific zebrafish microarray. Results revealed surprising effects on intestinal cellular pathways and physiology. Our prediction that manipulating both the diet and microbiome would be

additive was upheld for certain pathways, while others were only affected by diet or GIT microbiome but not both. Overall, cellular pathways in the GIT of fish are dynamic processes and their interactions with the GIT microbiome are not clearly understood. All experiments involving animals were conducted in conformance with guidelines for experimental animals and with an approved institutional animal use protocol. Funded by NSERC.

8.13

FASTING-INDUCED MORPHOLOGICAL REORGANIZATION OF THE COLON MAY NOT DRIVE CONCOMITANT CHANGES IN THE MICROBIOME

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We previously documented varied changes in the colonic microbiomes of animals representing five classes of vertebrates (i.e., tilapia, toads, leopard geckos, quail, and mice) over the course of fasting. In the current study we tested the hypotheses that the extent of tissue reorganization in the fasted colon was correlated with the observed changes in the microbiome. Colon segments adjacent to those used for the genomic study were fixed in Camoy's solution, mounted on slides, and stained with hematoxylin and eosin. We used ImageJ software to quantify cross-sectional and mucosal surface areas as well as thicknesses of mucosa, submucosa, and tunica muscularis. We found no fasting-induced differences in the morphology of colons of the mice (3days), quail (7days), or geckos (28 days). The toads that exhibited a general increase in phylogenetic diversity of their microbiome also exhibited reduced mucosal circumference at 14 and 21 days. The tilapia that increased their phylogenetic diversity also exhibited a thickened tunica muscularis in 21 days, but this change is unlikely to explain the dramatic changes in their microbiome that we documented. Given that the mice and quail exhibited fasting-induced increases and reductions, respectively, in their microbial diversity but did not exhibit detectable changes in colon morphology, we conclude that structural reorganization is not the primary factor shaping changes in microbial diversity within the fasted colon.

8.14

GLUCOSE CAN FUEL METABOLISM IN RBCS FROM NORMO-GLYCEMIC BUT NOT HYPOGLYCEMIC FISH

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Energy metabolism was assessed in red blood cells (RBCs) from Atlantic cod (*Gadus morhua*) and short-horned sculpin (*Myoxocephalus scorpius*). Blood glucose level was 2.5 mM in Atlantic cod and 0.2 mM in short-horned sculpin, respectively. Atlantic cod are normoglycemic and short-horned sculpin hypoglycemic by fish standards. Oxygen consumption, lactate production, and glucose utilization were determined in whole blood and related to g RBC. Glucose utilization was assessed by measuring both glucose disappearance and by the production of tritiated H₂O from glucose tritiated in the number 2 position. RBCs from both species have an aerobic based metabolism. In Atlantic cod, extracellular glucose is sufficient to provide the sum of glucosyl equivalents to support both oxidative metabolism and lactate production. In contrast, extracellular glucose can account for only 10% of the metabolic rate in short-horned sculpin RBCs. In this species energy metabolism by RBCs must be supported by alternative fuels. The difference in rates of extracellular glucose utilization is related to the extremely low levels of blood glucose in short-horned sculpin.

8.15

ACTIVITY OF THE 20S PROTEASOME IS NOT CORRELATED WITH THE EXPRESSION OF OXYGEN-BINDING PROTEINS IN ANTARCTIC NOTOTHEOID FISHES

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Antarctic icefishes (suborder Notothenioidei, family Channichthyidae) lack hemoglobin (Hb) as adults, and 6 of the 16 species of icefishes lack myoglobin (Mb) in their heart ventricle. All notothenioids lack Mb in oxidative skeletal muscle. As iron centered proteins, both Hb and Mb can promote the production of reactive oxygen species, which damage biological macromolecules. Consistent with this, we find higher levels of oxidized proteins in tissues of red-blooded notothenioids compared to icefishes. We hypothesized that the activity of the 20S proteasome, which degrades oxidized proteins, would be higher in +Hb and +Mb notothenioids compared to icefishes. Activity of the proteasome was measured in heart ventricle, pectoral adductor, and liver of six species of notothenioids varying in the expression of Hb and Mb. Proteasome activity was lowest in heart ventricle of *Chaenocephalus aceratus* (-Hb/-Mb), but highest in hearts of *Champscephalus gunnari* (-Hb/-Mb). Similarly, in liver, proteasome activity was highest in the icefish *C. gunnari*. Proteasome activity in pectoral adductor was equivalent among all species. These results suggest that the activity of the 20S proteasome is not correlated with the expression of Hb and Mb in notothenioids. Differences in proteasome activity may be related to phylogeny or differ-

ences in lifestyle and aerobic metabolic capacities among notothenioids. Funded by a grant from the *National Science Foundation* to K.OB (ANT-1043781).

8.16

MECHANISMS AND COSTS OF MITOCHONDRIAL THERMAL ACCLIMATION IN THE COMMON KILLIFISH (*FUNDULUS HETEROCLITUS*)

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Processes acting at the level of the mitochondria have been suggested to determine the thermal limits of organisms. To explore these questions, we have examined how mitochondrial properties are affected by thermal acclimation in the eurythermal killifish, *Fundulus heteroclitus*. We hypothesized that acclimation to lower temperatures could incur a functional cost with acute exposure to high temperatures (e.g., loss of proton motive force; PMF), perhaps accounting for shifts in thermal limits at the whole-organism level. We measured mitochondrial respiration rates through multiple complexes following thermal acclimation (5, 15, 33°C), and assessed mitochondrial membrane potential (i.e., PMF), and rates of reactive oxygen species (ROS) production. Acclimation to 5°C resulted in compensation of mitochondrial respiration, but these mitochondria were able to maintain PMF with acute exposure to high temperatures, and ROS production did not differ between acclimations, suggesting that increases in mitochondrial capacity do not reduce mitochondrial thermal sensitivity. Acclimation to 33°C caused suppression of mitochondrial respiration that was caused by effects on NADH-dehydrogenase (complex I). This work demonstrates that killifish mitochondria can successfully acclimate to low temperatures without incurring a cost to function, and that high temperature acclimation results in a suppression of metabolism, consistent with patterns observed at the organismal level. Funding: NSERC.

8.17

MITOCHONDRIAL RESPONSES TO SUSTAINED AND INTERMITTENT HYPOXIA IN KILLIFISH (*FUNDULUS HETEROCLITUS*)

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Fish experience diurnal cycles of hypoxia in the wild, but most previous research has focused on sustained hypoxia. We therefore compared the responses of liver mitochondria of killifish acclimated to normoxia, constant hypoxia, or nocturnal intermittent hypoxia using high-resolution respirometry and fluorimetry, coupled with measurements of antioxidant enzyme activities and oxidative stress. Acclimation to both hypoxia patterns increased the mitochondrial capacity for pyruvate oxidation, without affecting the capacities of electron transport complexes, oxygen kinetics, or phosphorylation efficiency. Both patterns also reduced the emission of reactive oxygen species (ROS) from mitochondria, and elevated cytosolic (but not mitochondrial) superoxide dismutase activity. Only intermittent hypoxia increased the activity of catalase. Our results suggest that there are multi-faceted cellular and mitochondrial responses to both intermittent and sustained hypoxia to alter ROS-mediated signalling and/or minimize oxidative stress. (Supported by NSERC of Canada).

8.18

HOW TOLERANT IS AN ANOXIA-TOLERANT VERTEBRATE?

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The crucian carp *Carassius carassius* is capable of surviving without oxygen for several months, by carefully matching ATP supply and demand. It does so by exploiting its unique ability to produce ethanol as an anaerobic end-product and undergo partial metabolic rate suppression, shutting down non-vital functions such as vision. Even so, it is unknown if crucian carp are fully able to protect themselves from the damage (i.e. cell death), normally caused by absence and particularly re-entrance of oxygen. Here, we investigated the occurrence of apoptosis in brain of crucian carp exposed to normoxia, anoxia and re-oxygenation in the laboratory, using Terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL). We also measured mRNA and protein levels of the apoptosis executor protein Caspase 3 using quantitative real-time PCR and Western blotting. Additionally, Caspase 3 expression levels were measured in brains from crucian carp captured in their natural habitat at different months of the year. Overall, crucian carp appear to maintain apoptotic activity in anoxia, but there is a transient increase in the early re-oxygenation phase. In its natural habitat transcription of Caspase 3 is generally much down-regulated during the winter, but protein levels are maintained, with only a slight increase in the spring. The study was financially supported by the Research Council of Norway and the Carlsberg Foundation (DK).

8.19

PARADOXICAL ANAEROBISM IN DESERT PUFFFISH

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Aerobic metabolism results in production of ~15-fold more ATP from glucose than anaerobic metabolism. We report that desert pupfish (*Cyprinodon spp.*) experience periods of up to nearly 5h of virtually no oxygen consumption despite being maintained in oxygen-saturated water; a process we call paradoxical anaerobism. Although most bouts are only a few minutes in duration, the longest continuous bout that we observed was 149 min. Fish randomly cycle between periods of paradoxical anaerobism and stable oxygen consumption patterns. Yet, there is no evidence for compensatory oxygen use. Fish produce ethanol as an alternate end product of anaerobic metabolism. Addition of exogenous ethanol induces rapid onset of paradoxical anaerobism.

8.20

HIGHLY EFFICIENT MITOCHONDRIA FUEL BLUEFIN TUNA RED MUSCLE WITHIN DISTINCT TEMPERATURE RANGES

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Pacific bluefin tuna (*Thunnus orientalis*) maintain elevated body temperatures that are 25-30°C in the slow oxidative "red" muscle that powers endurance swimming. Here we show red muscle mitochondria contribute to thermogenesis by efficient ATP production but not proton leak as found for mammals. Changing assay temperatures (20 - 35°C), we found that maximal mitochondrial substrate oxidation rate is temperature-sensitive but not rate-limiting for temperature-insensitive ADP-induced respiration. Interestingly, the low proton conductance is stable within the physiological temperature range of this species but increases rapidly above 30°C, possibly counteracting increasing mitochondrial reactive oxygen species production-but decreasing mitochondrial ATP production. Membrane-potential adjusted respiration measurements of ATP synthesis and proton leak revealed that red muscle mitochondria, compared to liver and white muscle, maintain high efficiency of ATP production, losing as little as 4 - 7 % energy to proton leak at physiologically relevant temperatures up to 30°C. In contrast, liver mitochondrial efficiency is less adapted to low temperatures, caused by decreased substrate oxidation rates. Endothermy in Pacific bluefin tuna is fueled by highly efficient mitochondria that are tightly coupled within the physiological range enabling high ATP output to fuel locomotion and heat production but less efficient at high temperatures.

8.21

CHANGES IN EXPRESSION OF TWO GENES INVOLVED IN ARGinine SYNTHESIS, AND CONCENTRATIONS OF ARGinine AND NITRIC OXIDE, IN AN AESTIVATING AFRICAN LUNGFISH

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Argininosuccinate synthase (Ass) and argininosuccinate lyase (Asl) are involved in arginine synthesis for multiple purposes. This study aimed to clone and sequence *ass* and *asl* from the liver of the African lungfish, *Protopterus annectens*, and to examine their mRNA expression in various tissues/organs during the induction, maintenance and arousal phases of aestivation. In the liver, significant increases in the mRNA expression of *ass* and *asl* could increase arginine production to support increased urea synthesis during the induction phase or increased nitric oxide (NO) production during the maintenance and arousal phases of aestivation. In the kidney, the *ass* mRNA expression level decreased significantly after 6 mon of aestivation, and there could be decreases in the synthesis of arginine and its supply to other tissues/organs. In the brain, changes in *ass* and *asl* mRNA expression levels could be related to regulating the supply of arginine for NO synthesis in response to ischemia and ischemia-reperfusion during the maintenance and arousal phases of aestivation, respectively. The decrease in *ass* mRNA expression, accompanied with decreases in the concentrations of arginine and NO, in the muscle of aestivating *P. annectens* might ameliorate the potential of disuse muscle atrophy. This study was approved by the NUS IACUC.

8.22

THE EFFECTS OF TEMPERATURE ON THE METABOLIC FATE OF LACTATE DURING RECOVERY FROM ANOXIA IN THE PAINTED TURTLE.

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Western painted turtles (*Chrysemys picta bellii*) survive anoxia for 160 days at 3°C and tolerate up to 200 mM plasma lactate. Stable isotope tracers were used to study the effect of temperature on the metabolic fate of lactate during and following anoxic submergence. A bolus of [U-¹³C]-lactate tracer was infused via an intra-arterial cath-

eter during anoxia and allowed to equilibrate before the start of recovery. ¹³C enrichment of plasma and tissue metabolites was determined using gas chromatography and mass spectrometry, and ¹³C enrichment of CO₂ from expired air, blood, and bone was determined using isotope ratio mass spectrometry. At 20°C, M3-glucose enrichment slowly increased throughout the recovery, and liver glycogen accounted for ~30% of the label at the end of recovery. Maximum urinary excretion rates were ~9 mmol glucose • kg⁻¹h⁻¹ and ~7 mmol lactate • kg⁻¹h⁻¹. At 10°C and 20°C, bone CO₂ represented ~16% and ~20% of the label, respectively, indicating its importance as a sink for metabolically produced CO₂ at all temperatures. Expired CO₂ accounted for ~1% of the injected ¹³C at 10°C and ~6% at 20°C. We conclude that at both 10°C and 20°C, painted turtles oxidize part of the lactate load initially during recovery, but the primary fate is gluconeogenesis. At 10°C, significantly less lactate is oxidized. Urinary excretion of lactate and glucose should be included when calculating the energetic cost of recovery. *This work was funded by the National Institutes of Health.*

8.23

MSRA AS AN IMPORTANT NEUROPROTECTIVE MECHANISM IN THE ANOXIA TOLERANT MODEL: *TRACHEMYS SCRIPTA ELEGANS*

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The effects of Reactive Oxygen Species (ROS) and oxidative stress have been recognized as key factors in aging, senescence and several neurodegenerative diseases such as Parkinson's disease and stroke (ischemia/reperfusion). The freshwater turtle, *Trachemys scripta elegans*, provides an alternative model to examine "physiology without pathology" due to its ability to tolerate long periods of anoxia and subsequent reoxygenation without any apparent functional damage. Oxidative stress resistance occurs through high antioxidant levels and an inhibition of ROS production. One potential antioxidant mechanism is the Methionine Sulfoxide Reductase System (Msr) that catalyzes the reduction of oxidized Methionine (Met) residues. Met is one of the most readily oxidized amino acids and Msr may restore activity to damaged proteins; it has also been shown that Msr can catalytically scavenge ROS before cellular damage occurs. The present study examined the role and regulation of MsrA during anoxia/reoxygenation in the turtle *in vitro* by knocking down MsrA expression as well as inducing FOXO3a which has been shown to regulate MsrA in other animal models. We found that knocking down MsrA expression with target specific siRNA resulted in decreased cell viability during anoxia/reoxygenation and that induction of FOXO3a protects against cell death. These results suggest that MsrA may be an important survival mechanism in turtles for anoxia and oxidative stress. This research was funded by NIH Project No. 1R15AG033374-01 Molecular mechanisms of oxidative stress resistance in an animal model of aging without senescence.

8.24

CERVICAL OSTEODERMS REVEAL PATTERN OF WHOLE BODY GROWTH IN JUVENILES OF THE AMERICAN ALLIGATOR

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Osteoderms (ODs), small bone nodules within the sauropsid integument, are increasingly used to infer the physiology and growth patterns of extinct species, such as non-avian dinosaurs. How ODs grow in extant species, and whether their mineral apposition rate (MAR) correlates with the animal's whole body growth, has not been determined. Effect of activity or cardiovascular shunts on OD growth is also unknown. We examined the relationship between MAR in cervical ODs and growth in two-year old American alligators, reared at 30°C. A surgical procedure was performed to convert the circulation pattern to in-series (n=24), or conserve the in-parallel pattern (sham, n=36). Animals were assigned to one of 3 groups: sedentary, running or swimming, exercise routine 18 months. Measurements of body mass and length were taken every other week, and animals received injections of fluorescent dyes during the growth period. We found regional growth heterogeneity between the apical, basal and lateral OD facets, which may reflect multiple functions of ODs. There is significant positive correlation between MAR and whole body growth, not affected by either activity level or circulatory pattern. However, no correlation exists between MAR in early and late ontogeny. We conclude ODs may be useful in inferring growth physiology of extinct archosaurs, regardless of circulatory design or activity pattern.

8.25

PHENOTYPIC PLASTICITY ACROSS THE ANNUAL CYCLE IN A MIGRATORY BIRD

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Prior to their migration, birds make physiological and behavioral adjustments to quickly accumulate fat stores and increase muscle performance to undertake their journey. The majority of the fat stores are utilized during their long-distance flights. However, residual fat may be detrimental due to impaired flight performance and increased predation risk, thus return to lean body composition is also rapid. This need for swift and substantial changes in a bird's physiology marks migration as a unique life history stage requiring phenotypic flexibility. Specific mechanisms directing stage transitions in migrating birds are largely unknown. Our efforts seek to characterize the role of peroxisome proliferator-activated receptors (PPARs) in the regulation of the migratory phenotype of the gray catbird (*Dumetella carolinensis*). PPARs are a family of nuclear receptors involved in mammalian energy metabolism, are highly conserved in our study species and ligand responses are similar to mammalian receptors. PPAR involvement in migratory adiposity is supported by higher rates of glycerol release from adipose tissue during migratory compared to non-migratory stages. Investigation of the involvement of PPARs in flight muscle hypertrophy during migration are ongoing. This work was funded by the NSF, grant no. IOS-1257455 to PJS and JH.

8.26

BATS AND BIRDS SHARE DIGESTIVE ADAPTATIONS TO AN AERIAL LIFESTYLE

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Powered flight evolved at least twice in vertebrates. We tested for shared digestive adaptations in two extant volant lineages. Bats and birds, compared with nonflying mammals, share shorter intestines and smaller nominal intestinal surface areas (NSA), which lowers digestive mass carried and thus improves flight maneuverability and economy. Intestinal hydrolytic enzyme and nutrient transport activities appear similar among these groups per unit intestine, but lower over the entire intestine in the fliers. Nutrients can also be absorbed paracellularly by passing through the tight junctions that link adjacent enterocytes. Seven bat species and 14 bird species, with a variety of natural diets, absorbed significantly more of ingested L-arabinose and other similarly sized, metabolically inert, nonactively transported monosaccharides than 18 species of nonflying mammals. These differences in nutrient-sized probe absorption were demonstrated at the tissue level comparing results from perfusion experiments (7 bat species, 1 bird, 5 nonflying mammals) that control for several potential confounding factors. Greater amplification of digestive surface area by villi and differences in expression patterns of junctional proteins (i.e., claudins and occludin) may provide mechanistic explanations for the observation of higher paracellular absorption in bats and birds relative to nonflying mammals. Supported by USA NSF and Argentina CONICET.

8.27 WITHDRAWN

8.28

HEART RATE DYNAMICS IN A MARSUPIAL HIBERNATOR

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The pygmy-possum (*C. nanus*) is a marsupial that undergoes spontaneous short bouts of torpor, as well as multi-day bouts of deep hibernation. To examine heart rate (HR) control at varying ambient temperatures (Ta) and varying body temperatures (Tb) during torpor, we used radiotelemetry for ECG and Tb. The HR during euthermia was at its min (321±34 bpm) at a Ta of 31°C and increased linearly with Ta to a max of 630±19 bpm at a Ta of 20°C. During entry into torpor at a Ta of 20°C, HR slowed primarily as a result of episodic periods of cardiac activity. Electrical activity of the heart occurred in groups of 3 or 4 heart beats that coincided with ventilation, followed by pauses of multiple seconds with no electrical activity. At a Tb of 23°C in these torpor bouts, the HR was regular (i.e. no asystoles) with a rate of 40±3 bpm. In multi-day bouts of torpor in the absence of food and water, Ta was lowered to 4°C. Tb of during these deep bouts of torpor reached a minimum of 5°C, with a minimum HR of 6 bpm. Bouts of shivering that lasted 20 sec were detected in the ECG tracings. Shivering occurred every 3-4 minutes, during which ventilation occurred, and HR was briefly elevated to 20 bpm. The duration of the QRS complex rose from 10ms

during euthermia to 140ms at a Tb of 5°C. Similarly, the amplitude of the complex fell from 1.891 mV at euthermia to 0.736 mV at a Tb of 5°C. These findings demonstrate the dynamic functioning range of the heart of this marsupial to be >100 fold.

8.29

APOPTOTIC REGULATION DURING MAMMALIAN HIBERNATION

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During hibernation, ground squirrels experience cycles of profound metabolic depression. Body temperature may approach ambient (to below 0°C) and oxygen consumption may be as low as 1/100th of active rates. Mammalian hibernators naturally experience conditions known to cause widespread apoptosis in other systems e.g. hypothermia, bradycardia, and ischemia/reperfusion injury. We found that hibernating golden-mantled ground squirrels (*Spermophilus lateralis*) experience partial activation of the caspase cascade. Importantly, winter squirrels experience a 2-fold increased cleavage of the p32 procaspase 3 to liberate the active p17 caspase 3. Such a liberation might be expected to result in 20,000 times more proteolytic activity. However, activity assays for caspase 3 suggest limited activity under the conditions of torpor. Similar regulation of other caspase activities suggest a global event. We found no evidence for ICAD activation, PARP cleavage, or DNA nicking consistent with downstream evidence of caspase activation. In other words despite seeming activation of caspases, there was no evidence of apoptosis. These data highlight the need for systems level approaches in understanding complex physiological states like hibernation.

8.30

A CYTOSOLIC PROTEIN FACTOR LOCATED IN MULTIPLE TISSUES IN THE NAKED MOLE-RAT PROTECTS THE PROTEASOME FROM INHIBITION AND ACTIVATES THE PROTEASOME IN OTHER SPECIES

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The naked mole-rat maintains robust proteostasis and high levels of proteasome-mediated proteolysis for most of its exceptional (~31y) life span. Here, we report that the highly active proteasome from the naked mole-rat liver, muscle, and brain resists attenuation by a diverse suite of proteasome-specific small molecule inhibitors. Moreover, mouse, human, and yeast proteasomes exposed to the proteasome-depleted naked mole-rat cytosolic fractions, recapitulate the observed inhibition resistance, and the mammalian proteasomes also show increased activity. Gel filtration coupled with mass spectrometry and atomic force microscopy indicates that these traits are supported by a protein factor that resides in the cytosol. This factor interacts with the proteasome and modulates its activity. Although HSP72 and HSP40 (Hdj1) are among the constituents of this factor, the observed phenomenon, such as increasing peptidase activity and protecting against inhibition cannot be reconciled with any known chaperone functions. Furthermore, this phenomenon seems to correlate with longevity in seven different rodent species. This novel function may contribute to the exceptional protein homeostasis in the naked mole-rat, and other long-lived rodents, to allow them to successfully defy aging.

8.31

LIPIDS AND MYOGLOBIN: NEW INSIGHTS FROM NON-MODEL SPECIES

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During endurance exercise, terrestrial mammals rely mainly on oxygen delivered by the blood to support lipid-fueled aerobic metabolism. Physiological changes associated with endurance training increase muscular blood flow and subsequent oxygen delivery; yet, terrestrial endurance athletes have more myoglobin (Mb) than their sedentary counterparts. Interestingly, Mb has a high affinity for oxygen, requiring extremely low partial pressures of oxygen to release oxygen stores. Accordingly, the traditional functional paradigm pertaining to Mb as an important oxygen store and transporter does not appear to be fully applicable to healthy, terrestrial mammals in vivo. Recent studies in our lab have provided new insight on Mb function in relation to lipids that challenge this traditional paradox. These studies first identified a link between lipid supplementation and Mb expression in diving mammals, and then extended those findings to terrestrial mammals while elucidating an apparent tie to ROS signaling. These combined studies provide data offering an alternative paradigm, whereby in the context of lipid metabolism, Mb as a ROS scavenger or lipid

transporter appears more relevant than Mb as an oxygen store. Thus, as mammals increase their aerobic capacity, and lipid utilization, there is an increase in Mb expression to facilitate lipid metabolism by offsetting negative byproducts associated with a high fat diet.

8.32

THYROID GLAND REMAINS RESPONSIVE TO THYROID STIMULATING HORMONE WITH SENSITIVITY INCREASING WITH FASTING DURATION IN A PROLONGED FASTED MAMMAL

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Food deprivation in mammals is associated with reduced thyroid hormones (TH), especially tri-iodothyronine (T3), to suppress metabolism. However, in prolonged-fasted, metabolically active elephant seal pups, cellular thyroid hormone-mediated components are up-regulated with fasting duration. The functional relevance of this apparent paradox is unknown and demonstrates variability in the regulation of TH levels, metabolism and function among food-deprived mammals. To address our hypothesis that the thyroid gland remains responsive with fasting duration, we infused early and late fasted pups with thyroid stimulating hormone (TSH) and measured the concentration of total and free thyroxine, (fT4 and fT4) as well as total tri-iodothyronine, (fT3). During the early fast, concentrations of fT4, fT4 and fT3 increased throughout the infusion period in response to TSH, with levels peaking at 120 minutes, $58 \pm 9\%$, $76 \pm 12\%$, and $54 \pm 6\%$, respectively. During the late fast, concentrations of fT4, fT4 and fT3 also increased throughout the infusion period in response to TSH, with levels peaking at 120 minutes, $56 \pm 10\%$, $59 \pm 12\%$, and $60 \pm 9\%$, respectively. Additionally, although levels returned to normal at 24 hours during the early fast, levels remained 115% and 142% elevated in fT4 and fT4, respectively after 24 hours in the late fast. Combined with our previous data demonstrating that cellular TH pathways are active and functional during prolonged food-deprivation in metabolically and physically active seals, the data suggest that the regulation and function of the thyroid gland and of thyroid hormones of the northern elephant seal is atypical.

8.33

PURINE NUCLEOSIDE PHOSPHORYLASE ACTIVITY IN ERYTHROCYTES FROM BOTTLENOSE DOLPHINS (*TURSIOPS TRUNCATUS*) IN RESPONSE TO BREATH-HOLD DIVING AND EXERCISE

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Under hypoxia, adenosine triphosphate (ATP) is degraded and hypoxanthine accumulates. Exercise and breath-hold diving can induce tissue hypoxia. During exercise, skeletal muscle energy demand and oxygen consumption increase. If oxygen consumption exceeds its supply, hypoxic conditions ensue. During breath-hold diving, oxygen supply decreases and, although oxygen utilization is regulated by bradycardia (low heart rate), peripheral vasoconstriction and ischemia, oxygen reserves decrease. The goal of this study is to evaluate changes in the purine metabolism in response to diving and exercise in bottlenose dolphin (*Tursiops truncatus*). Blood samples were taken from captive *T. truncatus* (n=8) following an exercise routine and after a dive to 1.5 m deep for 90 s. Hematocrit and plasma and intraerythrocyte activity of purine nucleoside phosphorylase (PNP, enzyme related to increased ATP degradation under hypoxia) was measured. PNP activity in erythrocytes from bottlenose dolphins after diving was not significantly different from that after exercise, but was higher than that under basal conditions. PNP activity increases in terrestrial mammals during high energy demanding activities. Results suggest that PNP activity increases under hypoxic conditions associated to exercise and breath-hold diving in *T. truncatus*.

8.34

OPPOSITE TRENDS IN OVER-SUMMER MASS CHANGE: POST-PARTURIENT WEDDELL SEALS GAIN WEIGHT WHILE NON-REPRODUCTIVE FEMALES LOSE MASS AND CONDITION

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To determine how summertime changes in mass and body condition in female Weddell seals correspond with pup-production and the annual molt, we handled prime-age multiparous females that successfully pupped (moms) or that did not pup (skips) during both the breeding (Nov/Dec) and molt (Jan/Feb) periods. At first handling

(~35dpp; end of lactation), moms were significantly lighter and leaner than skips (283.8 ± 9.5 kg, $29.8 \pm 1.1\%$ lipid vs. 449.4 ± 13.6 kg, $38.2 \pm 1.7\%$ lipid). Two months later, the body mass and condition (% lipid) of moms and skips were converging, as moms had regained mass at 0.9 ± 0.2 kg/day, nearly all as lean tissue, while skips had lost mass at 1.1 ± 0.2 kg/day (0.4 ± 0.2 kg/day lean). As a result, skips lost condition while moms maintained but did not increase lipid reserves. Moms were just starting to molt at the second handling, and they may prioritize lean tissue deposition during summer in anticipation of the protein costs associated with generation of new hair. In contrast, skips were already fully molted by Jan/Feb, and their loss of both fat and protein likely reflects the metabolic costs of hair growth as well as the increased haul-out times and reduced foraging effort evident during the molt. These findings suggest that percent lipid is not always an accurate measure of an animals' nutritional status and that protein stores may influence the timing of molt in a high latitude marine predator. Funding for this research was provided by NSF award ANT-124643.

8.35

LINKING CALORIC INTAKE TO GROWTH AND BLUBBER DEPOSITION IN WALRUS CALVES

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Walrus may be unable to meet daily energetic demands in their changing Arctic ecosystem. Immature walrus, with limited ability to capture prey, may be vulnerable as they transition from a complete reliance on their mother's milk for nutrition. Mature walrus have the ability to buffer short term energetic deficits by using energy stored as blubber, but little is known about caloric intake or the sequestration and mobilization of blubber in immature walrus. Two orphaned wild walrus calves raised in human care provided the opportunity for controlled study of daily caloric intake and body mass, as well as quarterly measurements of morphology and blubber thickness (21 sites). During the two year study, the animals had a 2.9 to 5.1-fold increase in body mass and a 1.3 to 1.4-fold increase in body length. Caloric intake ranged from 0 to 27,777 kcal day⁻¹, with mass-specific intake decreasing with age. Blubber thickness varied topographically (range: 1.45 - 6.6 cm), with average blubber depth generally increasing with age. Estimates of the proportion of blubber indicated that 0-2 year-old walrus are comprised of 24 - 30% fat. Ultimately, these food consumption and body morphology data will provide realistic inputs to construct meaningful bioenergetics models for free-ranging walrus. References: Noren, S.R., Udevitz, M.S., Jay C.V. 2012. Bioenergetics model for estimating food requirements of female Pacific walrus (*Odobenus rosmarus divergens*). *Mar Ecol Progr Series* 460:261-275.

8.36

LACTATING MICE INCREASE VILLUS SURFACE AREA THRU INCREASED ENTEROCYTE WIDTH IN RESPONSE TO LOW PROTEIN DIETS

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Animals have evolved the ability to flexibly change the size of their gut to match intake. However, under some conditions, animals can respond to dietary challenges in ways that counterbalance nutrient availability. Previous work in our lab has demonstrated that lactating mice on a low protein isocaloric diet increase small intestine mass and diameter, which may improve amino acid absorption. In this study, we compared aminopeptidase activity and villus and enterocyte morphology to determine how lactating females modify their gut at the tissue level when challenged with insufficient dietary protein. Mice were fed isocaloric diets containing either 11.5% or 23 % (control) protein from one week prior to pairing until peak lactation, or for a comparable time for nonreproductive females, at which time females were sacrificed and tissues were harvested. Diet did not affect aminopeptidase activity, whereas villus morphology was modified with diet. Villus height and intestinal surface area in the proximal region increased by more than 25% in lactating mice on the low protein diet. The increase in surface area was matched by an increase in enterocyte width, indicating that larger cells are produced in lactating mice experiencing protein insufficiency. These changes were seen only in reproducing mice, supporting the interpretation that modification of the gut serves to compensate for low dietary protein under conditions of high protein needs.

8.37

MACROPHAGE INFILTRATION IN THE ADIPOSE TISSUE OF DAIRY COWS DURING NEGATIVE ENERGY BALANCE

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Excessive lipolysis during periparturient and early lactation periods elevates plasma nonesterified fatty acids (NEFA), predisposing cows to clinical diseases including displaced abomasum (DA), ketosis and fatty liver. While in humans, uncontrolled lipolysis is commonly associated with adipose tissue macrophage (ATM) infiltration in obesity and metabolic syndrome, it is unknown if it leads to ATM infiltration in dairy cows. The goal of this study was to characterize ATM infiltration into different adipose tissue depots in early lactation cows with DA (DIM<50, n=7) and non-lactating non-gestating animals (n=7). Serum samples and biopsies from the omental (OM) and subcutaneous (SC) fat depots were obtained. Adipose macrophage infiltration was assessed using flow cytometry and immunohistochemistry. Cows with DA were ketotic and had a plasma NEFA>1.0 mEq/L. These animals also had significant infiltration of ATMs in OM and SC as characterized by increased numbers of CD117a⁺ and CD14⁺ cells compared to dry cows. Both macrophage markers were expressed by a significantly higher number of SVC from OM compared to SC in cows with DA. These results indicate that lipolysis induces ATM infiltration during negative energy balance stages in dairy cows. Future studies will evaluate the role of this inflammatory response in the adipose tissue remodeling process associated with demand lipolysis. Funded by USDA-NIFA grant 2012-67012-19832 and the CVM Venture and Last Lecture Fund/Weiser.

8.38

THE EFFECT OF CIRCADIAN ORGANIZATION ON ENERGY USE AND IMMUNE FUNCTION IN C57B MICE

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Circadian organization is thought to be important for efficiency and energy balance, and may therefore affect metabolic rate. Using two strains of C57B mice (*Mus musculus*): wildtype (WT) and transgenics lacking vasoactive intestinal peptide (VIP), we examined the effects of disrupted circadian rhythms in VIP-/- mice on energy expenditure and immune function under two different circadian conditions (entrained and free-running). We examined two measures of energy expenditure: basal metabolic rate (BMR) and daily food intake (FI), and two measures of immune function: total white blood cell (WBC) count and neutrophil/lymphocyte (N/L) counts. The BMR of entrained VIP-/- mice was higher than that of WT mice and decreased under free-running conditions; but still remained higher than that of WT mice. FI of entrained VIP-/- mice was lower than that of WT mice and increased under free-running conditions. VIP-/- mice had lower total WBC counts as compared to the WT mice, regardless of circadian condition. Circadian condition had no effect on N and L counts in WT mice, but L counts decreased and N counts increased in VIP-/- mice between entrained and free-running conditions. Although VIP-/- mice are known to have disrupted circadian organization, the loss of VIP might also affect other systems. Our observations that energy expenditure and immune function differ between circadian conditions in VIP-/- mice suggest that circadian organization contributes to the regulation of energy use.

9.0: FIELD PHYSIOLOGY

9.1

ENTRAINMENT OF CIRCADIAN GENE EXPRESSION RHYTHMS IN CALIFORNIA MUSSEL *MYTILUS CALIFORNIANUS*

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Since the ebb and flow of the tide is a regular temporal occurrence associated with highly contrasting environmental conditions, it follows that intertidal organisms should have a dedicated, endogenous time-keeping mechanism that aids in anticipating these environmental fluctuations and making the appropriate physiological changes. As a first step in elucidating the molecular mechanism of the circatidal clock, we sought to identify a set of candidate genes likely to be involved in the central oscillator in *M. californianus*. Our previous work has shown that *M. californianus* exhibit circatidal gene expression rhythms while exposed to simulated intertidal conditions, as well as intertidal and subtidal conditions in the field. If any of those rhythmic genes are involved in the circatidal clock, then they should continue to show circatidal rhythmicity even in the absence of environmental cues. In this study, *M. californianus* were acclimated to field subtidal conditions, then quickly moved to constant conditions. Gill samples were taken every 2 hours for 48 hours. RNA was isolated and microarray and RNAseq analysis were performed. Results show that *M. californianus* continue to show circatidal gene expression, even in constant conditions. Moreover, all of the circatidal genes identified in this study were found to be circatidal in our previous studies as well. This set of candidate genes will allow us to further investigate the molecular mechanism of the circatidal clock.

9.2

THE STRESS RESPONSE IS ASSOCIATED WITH BOTH CORTISOL AND ALDOSTERONE RELEASE IN MARINE MAMMALS

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Numerous environmental conditions potentially disrupt homeostasis in animals. Sufficient disturbance results in a stress response and activation of the hypothalamus-pituitary-adrenal (HPA) axis. Stress hormones may therefore be useful indicators of disturbance in animals of conservation concern but we must understand the variability in HPA axis response among species to interpret stress levels in wildlife. We therefore investigated the stress response in two groups of marine mammals: free-ranging elephant seals (n=16) and captive bottlenose dolphins (n=7). Stress was induced by administering adrenocorticotrophic hormone (ACTH) and blood samples were collected over ~2 hrs. Seals received 0.25 U/kg ACTH and showed increases in both cortisol and aldosterone. Several downstream effects were detected including increases in reverse T3 and energy substrates (glucose and free fatty-acids). Captive dolphins received varied doses of ACTH (0.02 to 0.25 U/kg) combined with out-of-water sampling that contributed to greater variation in the magnitude of hormone release but dolphins also displayed increases in cortisol and aldosterone. Corticosteroid concentrations were far lower in dolphins than in seals. ACTH is not a strong aldosterone secretagogue in terrestrial mammals but several studies have measured increased aldosterone in response to stress in marine mammals, indicating its potential importance in osmoregulation or vasoconstriction in this group. This work was funded by the Office of Naval Research.

9.3

PERSISTENT TISSUE DIFFERENCES IN FATTY ACID PROFILES OF WEDDELL SEALS (*LEPTONYCHOTES WEDDELLII*) REFLECT TISSUE ROLES

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Phocid seals store the majority of ingested lipid as a subcutaneous blubber layer, which acts as the main energy store and thermal barrier. However, lipids are also stored as intramuscular lipid droplets and are an immediate source of energy. To determine if the energetic roles of each tissue (storage vs. immediate energy) result in differential storage of specific lipids, we compared blubber and muscle fatty acid (FA) profiles of adult female Weddell seals in January and October. There were seasonally persistent significant differences (mean difference \pm SEM) among lipid classes, and individual FAs between tissues. Muscle had significantly greater proportion of saturated FAs (SFAs; 14:1 \pm 3.4%), driven by significantly higher proportions of C16:0 (4.0 \pm 0.6%) and C18:0 (5.5 \pm 0.6%), while blubber contained a significantly greater proportion of monounsaturated FAs (MUFA) (13.9 \pm 1.7%) due to much higher levels of 16:1n7 (5.7 \pm 0.7%) and 18:1n9 (9.0 \pm 1.4%). Overall polyunsaturated FA (PUFA) accounted for 19.0 \pm 0.3% in both muscle and blubber, though more individual PUFAs were found in significantly greater proportion in the muscle than blubber. The exception was 22:6n3, which was significantly higher in the blubber (2.9 \pm 0.4%). Overall, tissue specific storage of FA reflects the metabolic roles of each tissue; with blubber containing more MUFA, which retain fluidity at cold temperatures, and muscle containing more energetically dense SFAs in support of phocids lipid based metabolism.

10.0: EVOLUTIONARY PHYSIOLOGY

10.1

TALES FROM THE PRECAMBRIAN: TNF-INDUCED APOPTOSIS REMAINS UNCHANGED 550 MILLION YEARS LATER

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The Precambrian explosion led to the rapid appearance of most major animal phyla alive today. It has been argued that the complexity of life has steadily increased since that event. Here we challenge this hypothesis through the characterization of apoptosis in reef-building corals, representatives of some of the earliest animals. The Tumor Necrosis Factor (TNF) Receptor-Ligand Superfamilies (TNFRSF/TNFSF) are central mediators of apoptosis and the predicted proteome of *Acropora digitifera* contains more putative coral TNFRSF members than any organism described thus far, including humans. This high abundance of TNFRSF members led us to wonder what would happen if corals were exposed to a member of the Human TNFSF (HuTNF α). HuTNF α was found to bind directly to coral cells, increase Caspase activity, cause apoptotic blebbing and cell death and finally induce coral bleaching. Next immortalized human T-cells (Jurkats) expressing a functional Death Receptor Pathway (Wild-Type) and a corresponding FADD-knockout cell line were exposed to a coral TNFSF member (AdTNF1) purified here. AdTNF1 treatment resulted in significantly higher cell death (p<0.0001) in Wild-Type Jurkats compared to the cor-

responding FADD-knockout demonstrating that coral AdTNF1 activates the *H. sapien* Death Receptor Pathway. Taken together these data show remarkable conservation of the TNF-induced apoptotic response representing 550 million years of functional conservation.

10.2 MUSCULAR DYSTROPHY GENES AND THE EARLY METAZOAN TRANSITION FROM DYNEIN TO MYOSIN-POWERED LOCOMOTION

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During early animal evolution, the primary source of mechanical power shifted from flagellar dynein to densely arrayed class II myosin. Although muscle conferred the advantage of three-dimensional scalability, the transition brought with it the problem of safely transmitting huge forces across the cell membrane - a task that in vertebrates is accomplished in part through the dystrophin-dystroglycan-sarcoglycan complex (DGC), which is implicated in most forms of muscular dystrophy. We present the inferred earliest steps in the molecular evolution of these cell surface proteins, using genome sequence data from all early branching metazoan phyla and a broad sampling of unicellular taxa. Surprisingly, the phylogeny suggests that a DGC emerged before the 'sarcomeric' clade of myosins, implying conserved function(s) among unicellular lineages closely related to the Metazoa. Furthermore, linkage of the DGC to the cytoskeleton occurred before the tandem-repeat expansions seen in scaffolding proteins of the titin superfamily. Finally, intron positions in the inferred dystrophin gene in a common ancestor to all Metazoa provide an important clue to the molecular basis of Duchenne muscular dystrophy and emerging therapies. Our reconstruction suggests that evolution of membrane-spanning muscular dystrophy protein complexes was an essential process as one geometric constraint on power transduction was traded for another.

10.3 EVOLUTION OF THE UREA TRANSPORTER FAMILY IN VERTEBRATES

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Urea transporters (UT) are facilitative transporters that allow urea to move down its concentration gradient across cellular membranes. While it would be expected that urea excreting (ureotelic) animals express these proteins, surprisingly several non-ureotelic species also express these transporters in multiple tissues. In the current study, we explored UT evolution and tissue distribution in vertebrate lineages to further our understanding of their role and function throughout vertebrate history. We reconstructed a phylogeny of UT proteins in vertebrates, and using RT-PCR generated a tissue distribution of these transporters in multiple vertebrate species. Our results suggest that a single copy of an ancestral UT present in bacteria was initially duplicated several times early in vertebrates history. However, these duplicates were secondarily lost in lobe finned fishes, and a subsequent series of tandem duplications in amniotes and mammals generated the UT-A and UT-B mammalian genes. Interestingly, the tissue distribution of these transporters in ammonotelic, ureotelic and uricotelic species suggests that regardless of organismal ureogenic capacity, a majority of tissues express different UT isoforms. Thus, we propose that these transporters may have primarily evolved and been conserved throughout evolution for the cellular control of urea transport rather than for an organismal nitrogenous waste excreting strategy. Funded by NSERC (Canada).

10.4 DIFFERENTIAL GENE REGULATION UNDERLIES RESPONSES TO ACUTE HEAT STRESS AND LOCAL ADAPTATION IN *TIGRIOPUS CALIFORNICUS*

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Temperature is one of the main environmental factors that influences local adaptation of conspecific populations along the latitudinal gradients. However, the molecular mechanisms underlying local adaptation to temperature gradients are not well understood. The intertidal copepod *Tigriopus californicus* is a good model for studying acute heat stress response and thermal adaptation. Populations of *T. californicus* inhabit high intertidal rock pools along the west coast of North America from Baja California to Alaska. These pools present extreme thermal environment and previous studies have shown evidences of thermal adaption of *T. californicus* populations along the latitudinal gradient. Southern populations survive acute heat stress at higher temperatures than northern populations. Transcriptome studies have shown that thermal tolerance is associated with levels of expression of numerous heat shock protein (Hsp) genes. We hypothesize that genetic variation in the heat shock transcription factor (HSF) underlies differential responses to acute heat stress among different populations of *T. californicus*. There is one copy of the HSF gene with 529 amino acids in the *T.*

californicus genome. For example, we found 10 amino acid substitutions in the HSF gene between Santa Cruz and San Diego populations. This suggests functional differences of HSF from different populations along the latitudinal gradient. HSF is activated by heat stress and it subsequently induces transcription of Hsp genes. However the heat shock regulatory network has not been well studied in the ecological and evolutionary context. Here we investigate differential regulations of heat stress responses by comparing the activation temperatures of HSF from different populations of *Tigriopus californicus*.

10.5 RELATIONSHIPS BETWEEN MITOCHONDRIAL THERMAL PERFORMANCE AND HEAT TOLERANCE AMONG POPULATIONS OF THE INTERTIDAL COPEPOD *TIGRIOPUS CALIFORNICUS*

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Populations of conspecifics that are distributed across a wide latitudinal range allow for the study of adaptation. *Tigriopus californicus* is a copepod found in high rocky tide pools along the west coast of North America, making it an ideal study system for examining the evolution of heat tolerance. Previous studies have shown that the southernmost populations of *T. californicus* have the highest survivorship following acute heat stress. In this study, we examine the physiological basis of heat tolerance. We hypothesize that adaptation among populations leads to differences in mitochondrial thermal performance and confers enhanced tolerance to southern populations, allowing them to survive higher thermal stress. After common garden acclimation, we isolated mitochondria from six populations across the geographic range and tested ATP production at temperatures between 10-45 degrees C. While mitochondria of northern and southern populations performed similarly at lower temperatures, the ATP production of northern populations dropped as the temperature was raised until it reached a lethal temperature (no ATP production). Southern populations produced more ATP at high temperatures than northern populations, and their lethal temperature was higher. These results suggest that increased mitochondrial performance contributes to the ability of southern populations of copepods to survive thermal stress more effectively than their northern conspecifics. Supported by NSF DEB1051057.

10.6 OBESITY-INDUCED CARDIAC DYSFUNCTION IN STARVATION-SELECTED *DROSOPHILA*

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Drosophila has recently emerged as a model to study the pathophysiological effects of obesity on the heart. When fed high fat or sugar diets, *Drosophila* become obese, leading to heart defects including dilation, steatosis and myofibrillar disorganization. We have selected populations of *Drosophila* for starvation resistance for over 80 generations. In response, the starvation-selected (SS) lines have evolved physiologies that mimic obesity in mammals, including much higher lipids and lower metabolic rates than their unselected controls. Using video microscopy and OCT, we found SS hearts to be dilated and less contractile than their controls. We demonstrated a direct relationship between fat storage and heart dysfunction, as we rescued the SS hearts by making them lean through a period of starvation. These findings suggest that the genetic basis of heart disease in the SS lines is dependent on lipid homeostasis. We hypothesize the major source of dysfunction to be the physical interference of excess adipose tissue between the dorsal cuticle and the heart, which we observed upon dissection. We found no evidence of cardiac steatosis in the SS lines, nor any indication of myofibrillar disorganization. These data suggest that the mechanism of heart dysfunction in the SS lines is fundamentally different from previously reported models of diet-induced heart disease in *Drosophila*. Overall our findings provide a new model to study the physiology and genetics of obesity-induced heart disease.

10.7 SEX AND NUTRIENT EFFECTS ON ENERGY ALLOCATION AMONG BODY PARTS WITHIN AN INDIVIDUAL

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We examined relative energy allocation among body parts (head, thorax, legs, wings, abdomen) within individuals as a function of body size, diet quality, and sex, in the hawkmoth *Manduca sexta*. 50-90 moths per sex, per diet (478 total), were dissected and each body part dry weighed. Calibration curves for the caloric content of each body part, diet, and sex were generated using a bomb calorimeter. These calibration curves were used to calculate the predicted caloric content of each of the 5 body parts in the 478 moths. Males and females allocated resources very differently within and across diet qualities. For example, mass-specific (cal/g) allocation to the

thorax in females increased with thorax size on high quality diet but decreased on low quality diets. This pattern was reversed in males. Similar inverse relationships for males and females were found in other body parts. Tradeoffs in caloric investment were seen only between the abdomen and other body parts. These patterns of trade-offs held for both sexes and all diets. This pattern of tradeoffs disappeared, however, when examined on a mass specific level. Our results show that a life history strategy that favors dispersal is more expensive (per gram) than a life history strategy that favors investment in reproduction. Together these results show that males and females have different rules of mass-specific caloric investment into individual body parts when diet quality varies. Funding was provided by NSF-IOS.

10.8

MITOCHONDRIAL GENOMES AND OXIDATIVE PHOSPHORYLATION FROM POPULATIONS OF *FUNDULUS HETEROCLITUS* DISTRIBUTED ALONG A THERMAL CLINE

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The whole mitochondrial genome was examined in the teleost fish *Fundulus heteroclitus* to gain insight into the physiological effect of mitochondrial DNA sequence variation. *Fundulus* is distributed along a steep thermal gradient (1°C/degree latitude) on the eastern coast of the United States and display a northern and a southern mitochondrial haplotype with an admixture of both haplotypes found in northern New Jersey. The whole mitochondrial genome was sequenced from five populations along the thermal cline to define all functional nucleotide polymorphisms. To better understand the effects of nucleotide mitochondrial variation on physiological function, we measured mitochondrial respiration on 180 individuals in an admixture population containing both the northern and southern haplotype. Mitochondrial activity included state 3, E state, complex I, complex II, and LEAK at three assay temperatures (12°C, 20°C, and 28°C). Mitochondrial polymorphisms will be correlated with mitochondrial function and analyzed to gain information on divergence due to the thermal gradient and possible signatures of natural selection.

10.9

RECIPROCAL OSMOTIC CHALLENGES REVEAL MECHANISMS OF DIVERGENCE IN PHENOTYPIC PLASTICITY IN THE KILLFISH *FUNDULUS HETEROCLITUS*

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The killifish *Fundulus heteroclitus* is an estuarine species with broad physiological plasticity enabling acclimation to diverse stressors. Previous work suggests freshwater-adapted populations have expanded their physiology to accommodate low salinity environments, however, their hyper-osmotic tolerance is unknown. We employed a comparative approach to investigate the mechanistic basis of a derived freshwater phenotype and the fate of an ancestral euryhaline phenotype after invasion of a static freshwater environment. We compared physiological and transcriptomic responses to high and low salinity stress in fresh and brackish water populations and found an expansion of plasticity to hypo-osmotic conditions in the freshwater-native population. Improved hypo-osmotic tolerance was coupled with a contraction of resilience to high salinity. Transcriptomic data identified genes coupled with a conserved common response, a conserved salinity dependent response, and responses associated with micro-evolutionary divergence. Genes that diverged in their expression between populations were primarily those that showed salinity-specific expression. When populations were matched with their native salinity, expression patterns were consistent with the concept of transcriptomic resilience, suggesting local adaptation. These findings provide insight into the fate of a plastic phenotype after a shift in environmental salinity and help to reveal mechanisms allowing for euryhalinity.

10.10

THE ROLE OF LIPIDS AND PROTEINS IN THE DIET OF COMMON HAMSTERS (*CRICETUS CRICETUS*) ON HIBERNATION AND REPRODUCTIVE SUCCESS

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Hibernation is an effective strategy used by endotherms to cope with seasonal decline in food resources. The energy savings achieved by hibernation depends on the duration and the depth of torpor bouts. Long and deep torpor bouts allow animals to have a better body condition at emergence, on which greatly depend reproductive performances. Although this is a crucial point in an evolutionary perspective, the effect of food quality (lipid and protein content) on hibernation and reproductive success has not yet been fully investigated. In this study we evaluated such an effect in female common hamsters. Hibernation pattern, body condition and reproductive success were compared in two groups (R1: n=17, R2: n=15) fed with diets similar energy and carbohydrate contents, but with different lipid (5 vs 10%) and protein (19 vs 13%)

contents. The protocol agreed by the relevant Ethical Committee, followed the directives on animal experiments. Surprisingly, females fed the high-fat low-protein diet (R2) had the worst hibernation quality by spending less time in hypothermia. Thus, R2 females lost more fat and lean mass during hibernation. They also had less and smaller pups and the worst reproductive success. We show here that the gross-food quality influence both hibernation and next reproduction. In a next step we will determine the exact contribution of lipids and proteins according to their composition on both hibernation and/or reproductive success, and ultimately on fitness.

10.11

PHYLOGENETIC CORRELATION BETWEEN MAXIMUM OXYGEN CONSUMPTION AND HOME RANGE AREA AMONG SPECIES OF MAMMALS

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Home range (HR) can be defined as the area an animal uses during its daily activities. All else being equal, larger HRs should entail greater daily movement distances (DMD), accomplished by a greater amount of time moving and/or higher average speed. The latter could require a higher maximal aerobic speed (MAS), which is primarily determined by maximal oxygen consumption (VO₂max), so correlated evolution of VO₂max and HR is expected to occur. We tested for this correlation using literature data for 60 species of mammals. We used three models to compute regressions on body mass, using log-transformed data, and then correlate HR and VO₂max residuals: Ordinary Least Squares, Phylogenetic Generalized Least Squares, and Phylogenetic Regression with Ornstein-Uhlenbeck branch length transformation (RegOU) that mimics stabilizing selection. Preliminary results indicate that RegOU was the best-fitting model for both the HR and VO₂max regressions (allometric scaling exponents = 0.99 and 0.84, respectively). Residuals from the RegOU regressions did not show a statistically significant correlation, contrary to our prediction. HR is highly variable, and a substantial portion of this variation may represent measurement in the broad sense (e.g. variation in method of calculation, habitat quality, population density), which would obscure correlations with VO₂max. Alternatively, simple measures of HR may not accurately reflect variation in DMD or aerobic exercise requirements.

10.12

EFFECTS OF VOLUNTARY WHEEL RUNNING ON BODY MASS AND COMPOSITION IN SELECTIVELY-BRED HIGH-RUNNER MICE

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Selective breeding for high voluntary wheel running in four replicate High Runner (HR) lines produced an approximately 3-fold increase in number of wheel revolutions per day as compared with four unselected control (C) lines. Mice from generation 69 were measured for fat mass, lean mass, and water mass using an EchoMRI-100H scanner before and after the routine six-day period of wheel access as used to select breeders each generation. ANOVA indicated that males had greater body mass than females before wheel testing, and an interaction between sex and linetype such that C males were heavier than HR males, but female C and HR did not differ statistically. Analysis of covariance with body mass as a covariate indicated females had greater fat mass than males before wheel testing. Females lost more mass than males over the six days of wheel access, and C males gained mass while HR males lost mass. All groups lost fat mass, though there was a significant interaction between sex and linetype in females, HR mice lost less fat than C, while in males, HR mice lost more fat than C. Future analyses will consider the quantitative effects of amount of wheel running and food consumption on body mass and composition. This long-term selection experiment offers a unique model for studying sex-dependent body composition patterns and the effects of predisposition for voluntary exercise. Supported by NSF grant IOS-1121273 to T.G.

11.0: HUMAN NUTRITION AND PHYSIOLOGY EDUCATION

11.1

GENDER DIFFERENCE IN NUTRITIONAL STATUS OF A PRIMARY SCHOOL CHILDREN OF BANGLADESH

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Background: About 62% children of Bangladesh are malnourished. In this country, although, boys are assumed to be provided with more food than girls, this discrepancy has not yet been examined. Comparison of the nutritional status between boys and girls may be a good indicator to examine this difference. **Objectives:** To compare the nutritional status between school-boys and girls of Bangladesh. **Method:** This cross-sectional comparative study was conducted in the department of Physiology, Noakhali Medical College during April–June'12. 219 students of a selected public primary school of Bangladesh were enrolled for the study by convenient sampling. Permission

of the authority and the students were taken. Height and weight were measured and BMI was calculated. Statistical analysis was done by SPSS (version 16). Comparison between two groups was done by Student 't' test. **Result:** There were 91(41.6%) boys and 128(58.4%) girls. The mean age of all participants was 9.39 ± 1.72 years. There was no significant difference in age between boys and girls (9.59 vs 9.26 years, $p=1.64$). The mean \pm SD height, weight and BMI of all participants were 1.27 ± 0.10 meter, 21.94 ± 5.07 kg and 13.54 ± 1.63 respectively. No significant difference was observed between boys and girls in mean \pm SD height (1.27 ± 0.09 vs 1.26 ± 0.11 meter, $p=0.426$), weight (22.38 ± 4.70 vs 21.63 ± 5.31 kg, $p=0.284$) and BMI (13.71 ± 1.69 vs 13.43 ± 1.59 , $p=0.205$). **Conclusion:** There was no significant difference in nutritional status between boys and girls of a public primary school of Bangladesh. **Limitations:** As the study was done in a single public primary school of Bangladesh, it does not represent the whole country's children. **Funding source:** Navana, Beximco, SK&F & ACI pharma Bangladesh Ltd.

11.2

NUTRITIONAL STATUS OF MBBS STUDENTS OF A SELECTED GOVERNMENT MEDICAL COLLEGE OF BANGLADESH

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Background: Early adults are considered to be a nutritionally vulnerable segment of the population. Poor nutritional status during early adulthood is an important determinant of health outcomes at a later stage of life. In Bangladesh, various survey reports showed that approximately 85% of the people intake insufficient food, 76% of rural households is deficient in protein intake and 93%, 88% and 87% of households had deficient intake of calcium, Vitamin A and Vitamin C respectively. Medical students pass their early adulthood periods during their stay at medical college. They may have nutritional abnormalities as they are not able to take proper nutritious food in their dormitories. **Objectives:** To examine the nutritional status of medical students of Bangladesh. **Methods:** This cross-sectional descriptive study was conducted in the department of Physiology, Noakhali Medical College, Bangladesh. Permission of the authority and consent of the students were taken. Height and weight were measured and BMI was calculated and recorded on a data sheet. Statistical analysis was done by SPSS (version 16). **Results:** A total of 201 medical students were enrolled in the study. The mean \pm SD age of the students was 20.93 ± 1.57 years. Among the participants 84 (40.4%) were male students. The mean \pm SD BMI of the male, female and total students were 22.80 ± 3.69 , 20.71 ± 3.19 and 21.56 ± 3.55 respectively. Among the total students 138 (69%) had normal weight, 37(18.5%) were under weight, 21(10.5%) pre-obese and 4(2.0%) obese. **Conclusion:** Almost one-third of medical students suffer from nutritional abnormality. 18.5% suffer from underweight and 12.5% from overweight. **Funding sources:** Beximco, ACI, Navana & Square Pharma, Bangladesh Ltd.

11.3

IS THE GROWTH OF BANGLADESHI SCHOOL CHILDREN PROPORTIONATE TO THEIR AGE?

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Background: 62% Bangladeshi children are malnourished. **Objectives:** To test if the growth of school children is proportionate to their age. **Method:** This study was conducted in the department of Physiology, Noakhali Medical College Bangladesh during April-June'12. 174 students of a public primary school of Bangladesh were enrolled for the study by convenient sampling. Permission of the authority and the students was taken. Height, weight and BMI were measured. Statistical analysis was done by SPSS (version 16). Comparison between groups was done by Student 't' test. **Result:** Among 174 children 6,7,8,9,10,11,12,13, and 14 years old were 3(1.4%), 22(10%), 32(14.6%), 44(20.1%), 35(16.0%), 20(9.1%), 12(5.5%), 4(1.8%) and 2(0.9%) respectively. The mean \pm SD height (m), weight (kg) and BMI of 6, 7, 8, 9, 10, 11, 12, 13 and 14 years old children were 1.07 ± 0.08 , 18.67 ± 7.22 , 15.86 ± 4.05 ; 1.15 ± 0.06 , 17.26 ± 2.28 , 12.94 ± 1.39 ; 1.19 ± 0.07 , 18.56 ± 2.57 , 12.1 ± 1.2 ; 1.26 ± 0.07 , 21.23 ± 3.35 , 13.31 ± 1.32 ; 1.3 ± 0.08 , 23.14 ± 4.47 , 13.54 ± 1.40 ; 1.36 ± 0.07 , 26.75 ± 3.71 , 14.49 ± 1.69 ; 1.4 ± 0.04 , 27.5 ± 5.73 , 14.0 ± 2.5 ; and 1.37 ± 0.02 , 25.5 ± 2.48 , 13.61 ± 0.92 and 1.45 ± 0.01 , 36.0 ± 8.49 , 17.22 ± 3.9 respectively. The height, weight and BMI were rising as the child grew older. However the growth showed the following exception. The height of the 13 years group was less than 12 years group ($p=1.88$). The weight of the 7 years and 13 years group was less than 6 years ($p=461$) and 12 years group ($p=517$) respectively. The BMI of the 7 years, 12 years and 13 years group was less than the 6 years ($p=0.14$), 11 years ($p=519$) and 12 years ($p=650$) group respectively. **Conclusion:** The growth of primary school children of Bangladesh was not proportionate to their age. **Funding source:** Navana, Beximco, ACI pharma.

11.4

GENDER DIFFERENCE IN THE RESULT OF 1ST TERM PHYSIOLOGY EXAMINATION OF A GOVERNMENT MEDICAL COLLEGE OF BANGLADESH

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Background: In recent years more female students than males qualify to study in medical colleges of Bangladesh. Whether female students do better than males in their course exam is not known. **Objective:** To compare the 1st term result between male and female undergraduate medical students. **Method:** This cross-sectional comparative study was conducted in the department of Physiology, Noakhali Medical College during the period of June-August'13. 115 students of a selected public medical college of Bangladesh were enrolled for the study purposively. Consent of students and authority was taken. Data on admission merit-score and 1st term marks were collected. The written marks were scored out of 70 and oral out of 100. The result was classified as 'passed', 'failed' and 'absent'. Data was analyzed by SPSS (version 16). The continuous data was compared by Student's 't' test and categorical data by Chi-square test. P value <0.05 was significant. **Result:** There were 48 (41.74%) males. The mean admission merit-score of male and female students was similar (156.27 ± 5.78 vs 158.39 ± 6.40 vs, $p=0.70$). In 1st term exam the male and female students obtained similar marks in written (47.25 ± 7.15 vs 47.89 ± 8.15 , $p=0.679$, 95% CI, -3.733 to 2.442) and in oral (63.19 ± 8.049 vs 66.36 ± 9.13 , $p=0.72$, 95% CI, -6.632 to 2.92). Among the male students 20 (41.7%) passed, 22 (45.8%) failed and 6(12.5%) were absent and among the female students 26(38.8%) passed, 34(50.7%) failed and 7(10.7%) were absent. There was no significant difference between male and female students' result ($p=0.861$). **Conclusion:** The result of male and female undergraduate medical students in the 1st term physiology exam was similar. **Funding sources:** Unihealth, Globe, Ibne Sina & Acme Pharma Ltd.

11.5

USE OF FELLOW EXAMINEE AS SUBJECT IN OBSERVED STRUCTURED PRACTICAL EXAMINATION (OSPE) IN 1ST TERM PHYSIOLOGY EXAMINATION

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Background: Department of Physiology finds difficulty in managing 'subjects' in practical procedure. To avoid this difficulty fellow examinees of other group may be used as subjects. **Objective:** To find out the merits and demerits of using fellow examinees as subjects in the practical procedure. **Method:** This cross-sectional descriptive study was conducted in the department of Physiology, Noakhali Medical College, Bangladesh during May-June'14. Forty-two 1st year undergraduate medical students of a selected public medical college of Bangladesh were enrolled for the study purposively. Consent of students and authority was taken. Eighteen of them were selected as subjects and designated as subject-examinees. Other fellow examinees (non-subject) examined their blood pressure and pulse as part of 'observed structured practical examination' (OSPE). The opinion of all examinees regarding the merits and demerits of using fellow examinee as subjects in the practical procedure was recorded. **Result:** Examinees stated that they could perform their practical procedure without nervousness (24/42, 57.14%), accurately and comfortably (14/42, 33.33%) and subjects were made available without wasting time (2/42, 4.76%). Nineteen students (45.24%) found no disadvantage and 2(4.76%) felt embracing when the subject was of opposite sex. The subject-examinees narrated that they could learn from the errors done by their fellow examinee (11/18, 61.1%). 75% non-subject examinees expressed their willing to be subject so that they can learn from their fellows' error. **Conclusion:** Using fellow examinees as subjects is beneficial for the both the non-subject and subject examinees. **Funding sources:** Navana, Beximco, Unihealth, Square & Acme Pharma, Bangladesh Ltd.

11.6

EFFECTIVENESS OF FREQUENT DISCUSSION EMBEDDED INTERACTIVE LECTURE (FDEIL) IN LEARNING MEDICAL SCIENCE

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Background: In traditional lecturing system learners find less opportunity to make a critical point clear. Recent studies supported that students need interactive lectures. Frequent discussion embedded interactive lecture (FDEIL) has been preferred as a good method of learning medical science. However the effectiveness of FDEIL has not yet been tested. **Objective:** To compare the effectiveness of FDEIL with lecture followed by discussion (LFD) in learning medical science. **Method:** This comparative study was conducted in the Department of Physiology, Noakhali Medical College. Total 48 2nd year MBBS students were selected in the study. Among them 29 students were assigned in case group who were exposed to FDEIL and 19 students in control group who were exposed to LFD lecturing. The topic was 'the function of cerebellum', same for both the groups and taken by the same teacher. In FDEIL teacher delivered lecture for 15 minutes and gave a 5 minute's break for discussion.

Thus a one-hour lecture was divided into such 3 lecture-discussion cycles. In LFD, information was delivered by the teacher for 45 minutes and last 15 minutes were scheduled for discussion among students. At the end of the sessions both groups were asked to answer 20 objective questions each bearing 2 marks. **Result:** The case group obtained more mean \pm SD marks than control group but the difference did not reach the level of significance (25.43 \pm 6.32 vs 22.58 \pm 5.28, 95% CI, -6.77 to 6.381, $p=0.111$). More students of case group obtained $\geq 60\%$ marks than control group (72.4% vs 47.4%) but the difference was not statistically significant ($p=0.80$). **Conclusion:** FDEIL and LFD are similarly effective in learning medical science. Funding sources: Globe & Ziska, Pharma, Bangladesh.

11.7

READING THE PRIMARY LITERATURE: SKILLS OF FIRST-YEAR AND SENIOR UNDERGRADUATE BIOLOGY MAJORS

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The ability to find, evaluate, read and understand scientific literature is vital to trained scientists, but the integration of scientific literacy training in science curricula is inconsistent. To evaluate gains in the ability of undergraduate biology majors to read scientific literature, first-year and senior students were assigned to read a primary (i.e. experimental) article and write answers to a series of questions about key components of the article. A scorer who was blinded to the academic year of the subjects scored the answers to each question according to a rubric. Each subject's scores for each question were then aggregated to provide an overall score for the subject's understanding of the article. A crossover design involving two articles of similar complexity will enable longitudinal assessment of individual students and could also be used to evaluate specific interventions to improve information literacy training. In addition, the survey and scoring strategy can be adapted easily to other topics, providing a tool for quantitative assessment of information literacy across scientific fields. A parallel survey on the same subject population evaluates the ability to search scientific databases and identify relevant, peer-reviewed resources.

12.0: CARDIOVASCULAR AND RESPIRATORY PHYSIOLOGY

12.1

GETTING TO THE HEART OF PLASMA-ACCESSIBLE CARBONIC ANHYDRASE IN FISH

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Teleost fish possess a number of physiological adaptations that likely had a major role in their tremendous adaptive radiation and evolutionary success. One such adaptation is the Root-effect, whereby hemoglobin oxygen carrying capacity is reduced as red blood cell (rbc) pH decreases. Recent evidence suggests that the presence of plasma-accessible carbonic anhydrase (CA) – the enzyme that catalyzes $\text{CO}_2/\text{HCO}_3^-$ inter-conversion – can greatly enhance oxygen delivery to skeletal muscle by short-circuiting rbc pH regulation and engaging the Root-effect. We tested the hypothesis that plasma-accessible CA is present in the heart lumen of Coho salmon, an active teleost with spatially limited coronary circulation. The presence of a membrane-anchored CA isoform in the heart was confirmed by qPCR and Western blot. To test whether or not this isoform is accessible to plasma in the heart lumen, CA activity was measured in excised intact atria using a modified electrometric delta-pH assay. The atrial chambers were pre-treated with either saline or phosphatidylinositol-specific phospholipase C (PI-PLC) to cleave the membrane anchor of CA. In support of our hypothesis, we observed a significant 2.3-fold reduction in CA activity in atria pre-treated with PI-PLC relative to controls. We suggest that plasma-accessible CA in the salmon heart functions to safe-guard oxygen supply to regions of this vital organ devoid of coronary vessels. Supported by NSERC Canada and Canada Research Chair Program.

12.2

ELECTRICAL ACTIVATION AND REPOLARIZATION SEQUENCE IN THE RAINBOW TROUT HEART

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We investigated electrical activity of the ventricle of the isolated rainbow trout heart by optical mapping. We used 11, ~200 g rainbow trout acclimated to 13°C and conducted experiments at 13 °C using blebbistatin to prevent movement and Di-4-ANNEPES to visualize transmembrane voltage. Results show activation progressing from the AV junction with a base to apex excitation profile. Adrenaline slowed conduction across the ventricular surface of the heart. No transmural differences in ven-

tricular action potential (i.e. between spongy and compact layers) were observed. Adrenaline increased ventricular action potential duration and resulted in electrical alternans at fast stimulation frequencies. Conduction restitution was slowed and APD restitution was accelerated as contraction frequency was progressively increased. These results reveal interesting differences in ectotherm electrical activation and restitution compare with mammals particularly in relation to alternans and the effects of sympathetic stimulation on the electrical activity of the heart.

12.3

ANTIOXIDANT DEFENSE MECHANISMS IN THE RED BLOOD CELLS OF LONGHORN SCULPIN (*MYOXOCEPHALUS OCTODECEMSPINOSUS*) IN RESPONSE TO HYPOXIA AND RAPID RE-OXYGENATION

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Unlike their mammalian counterparts, teleost red blood cells (RBCs) retain a functioning nucleus and other organelles throughout their lifespan. Under natural conditions, all RBCs are continuously exposed to reactive oxygen species (ROS) as a consequence of their role as oxygen transporters. Teleost RBCs have an additional source of ROS due to their high rates of aerobic metabolism and functioning mitochondria. The current work aims to provide insight into the role of oxidative damage in the aging and/or removal of teleost RBCs from circulation. Levels of antioxidants (glutathione redox state, glutathione peroxidase (GPx) and catalase) were measured in RBCs from longhorn sculpin (*Myoxocephalus octodecemspinosus*) held under ambient conditions, during hypoxia (40% O_2 saturation) and after a rapid reoxygenation event. Based on comparisons within the literature, levels of antioxidants in the sculpin RBCs are higher than mammalian RBCs, suggesting these cells are better poised to defend against oxidative stress. Additionally, as both glutathione redox state (GSH:GSSG) and GPx activity were significantly elevated after rapid reoxygenation (no significant change in catalase activity) it suggests that the sculpin RBCs are capable of defending against increased production of free radicals as a result of fluctuating environmental conditions. Funding for this study was provided by Mount Desert Island Fellowship in Molecular Physiology and Georgia Southern University FRC seed funding.

12.4

HEART RATE AND BLOOD OXYGEN DEPLETION IN LOGGERHEAD TURTLES, *CARETTA CARETTA*

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In diving mammals and birds, heart rate plays a role in depletion of the blood oxygen store. To investigate whether a relationship between heart rate and blood oxygen depletion exists in marine reptiles, we implanted P_{O_2} electrodes and electrocardiogram leads in four loggerhead turtles (*Caretta caretta*) and attached a data logger to the turtles' carapaces. Time depth recorders and accelerometers were deployed to record dive profiles. During submerged periods of 20 to 40 mins, heart rate decreased from 6 to 10 beats min^{-1} to as low as 2-4 beats min^{-1} during the longest dives, much lower than previously reported in loggerhead turtles. Venous P_{O_2} values were between 50 to 70 mmHg at the surface and then declined monotonically to less than 20 mmHg during very long submergences. The observed low heart rates coincided with venous P_{O_2} depletion rates of 1 to 1.5 mmHg min^{-1} . Our data suggest that heart rate is the primary determinant of venous oxygen depletion during voluntary submergences in loggerhead turtles. This research was supported by NSF grant IOS-1121324. C. Williams was supported by an NIH Multidisciplinary Exercise Sciences Training Program Fellowship.

12.5

MECHANISMS FOR SUPPRESSION OF OXYGEN DELIVERY AND CONSUMPTION IN HIBERNATING BROWN BEARS

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Biochemical mechanisms underlying hibernation in mammals remain poorly understood. We measured blood metabolites of hydrogen sulfide (an ubiquitous signaling molecule inhibiting respiration rates in mitochondria) and oxygen affinity in red blood cell lysates from winter hibernating and summer active free-ranging brown bears (*Ursus arctos*) to determine their potential role in regulating oxygen consumption and delivery, respectively, during hibernation. We found that during hibernation plasma

hydrogen sulfide may be regenerated from its oxidation products (such as thiosulfate and polysulfides) and contribute to depress metabolism of perfused tissues, while cysteine is primarily used to enhance synthesis of glutathione, a major cellular antioxidant. Oxygen binding curves of red blood cell lysates were left-shifted in hibernating bears due to decreased temperature and 2,3-diphosphoglycerate, a major hemoglobin cofactor, thus limiting oxygen delivery to tissues. In conclusion, our results indicate that hydrogen sulfide and organic phosphates play concerted roles in the suppression of oxygen consumption and delivery, respectively, of hibernating brown bears in their natural environment. This study was approved by the Swedish Ethical Committee on animal research and was supported by grants from the Danish Council for Independent Research, Natural Sciences, the National Institute of Health and the Scandinavian Brown Bear Research Project.

12.6

RESPIRATORY PHYSIOLOGY IN CETACEANS

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August Krogh, the grandfather of comparative physiology, stated that 'For every defined physiological problem, there is an optimally suited animal that would most efficiently yield an answer'. Marine mammals are a prime example of this physiological principle. They thrive in the extreme environment of the sea, readily coping with atelectasis, hyperoxia, hypoxia, ischemia/reperfusion, and intravascular gas bubbles. Understanding cardiorespiratory physiology is vital to understanding the limitations that may affect survival. We study respiratory physiology in a large range of cetaceans, from harbor porpoise to beluga whale, to try to understand how pressure affects lung function. Cetaceans have compliant lungs and stiff conducting airways, in comparison to their terrestrial counterparts. These traits are assumed important for altering gas exchange during diving. However, recent work indicates great variability in the structural properties of the respiratory system between marine mammal species, possibly indicating species differences in diving ability. For all species studied, maximum expiratory flow rates exceed 140 L sec⁻¹, while maximum inspiratory flow rates range between 10 to 40 L sec⁻¹. The measured esophageal pressure suggests that odontocetes exhale passively during spontaneous respiration, but actively during maximal exhalations. Our results confirm an amazing respiratory capacity in cetaceans, and provide new data on chest compliance in odontocetes.

12.7

THE EVOLUTION OF UNIDIRECTIONAL PULMONARY AIRFLOW

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Bird lungs have conventionally been thought to be unique in having air flow through most of the conducting airways in the same direction during both inspiration and expiration. Aerodynamic valves cause unidirectional flow through a circular system of tubes, which are organized in an analogous manner to the arteries, capillaries and veins of the blood circulatory system. In contrast, the conducting airways of mammalian lungs arborize, with the branches ending in alveoli, and gases travel tidally in the bronchial tree. Unidirectional flow and cross-current gas exchange may improve the efficacy of the avian lung and may serve to enhance the ability of birds to sustain vigorous levels of exercise. Measurements and visualization of patterns of airflow in the lungs of a range of reptiles are revealing that birds are not unique in having unidirectional airflow. These data raise new questions about the functional underpinnings of unidirectional flow, the selective drivers for this trait, and the evolutionary history this system. This research was funded by NSF (IOS - 0818973 and IOS 1055080).

12.8

NATURE VERSUS NURTURE: HYPOXIC CARDIOVASCULAR AND RESPIRATORY RESPONSES IN BAR-HEADED GEESE (*ANSER INDICUS*) AND RELATED WATERFOWL

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The biannual high altitude migration of bar-headed geese (*Anser indicus*; BHG) across the Himalayas is a remarkable feat. While respiratory and hematological O₂ transport enhancements help facilitate this migration, little is known about the cardiovascular maintenance of O₂ transport during hypoxia-the hypoxic cardiovascular response (HCVR). We characterized the intra- and interspecific plasticity in HCVR across altitude, comparing domestic BHG (born at 0m), wild BHG (born at 3200m), and domestic barnacle geese, a related species (*Branta leucopsis*; born at 0m). All birds were exposed to stepwise hypoxia while measuring cardiovascular (heart rate,

stroke volume, cardiac output), respiratory (breathing frequency, tidal volume), and metabolic variables. Hypoxia exposure led to cardiac output increases driven by stroke volume. The HCVR of 0m BHG and barnacle geese did not differ. However, during normoxic recovery all variables in BHG returned to resting levels, whereas barnacle geese had an elevated heart rate and blood acidosis. This suggests that BHG matched O₂ supply and demand during hypoxia, while barnacle geese used anaerobic metabolism. There were significant differences between 0m and 3200m BHG. 3200m BHG were more hypoxia tolerant, responding at a lower P₅₀. Thus, altitude matters-rearing at 0m profoundly impacts the BHG HCVR. Funding: NSERC, Vanier Canada, and Killam Trusts.

13.0: EVOLUTIONARY PHYSIOLOGY

13.1

EVIDENCE OF COUNTERGRADIENT VARIATION AND ADAPTIVE SLOW GROWTH RATE IN A MARINE ISOPOD (*IDOTEA BALTHICA*) LOCALLY ADAPTED TO LOW SALINITY

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Local adaptation is crucial to the generation and maintenance of biodiversity, and is most likely to occur over strong environmental gradients in species with direct development. This study used a common garden approach to investigate the hypothesis that Baltic populations of the brooding marine isopod *Idotea balthica* (found in marine waters and in the low salinity Baltic Sea,) are locally adapted to low salinity. These data provide the first indications of countergradient variation (CnGV) and adaptive slow growth rate in Baltic *Idotea balthica* in comparison to fully marine counterparts; Evidence of CnGV found in metabolic rate analysis, while reduced growth rate independent of salinity treatment demonstrated adaptive slow growth. Increased oxidative stress in both populations with increased deviation from respective salinity of origin conditions, and differential mortality in the salinity treatments (salinity 32, 20 and 5) also validate the hypothesis of local adaptation within the Baltic. The suit of traits under selection in low salinity *Idotea balthica* have the common theme of metabolic budget management, a commonality with CnGV over temperature gradients but this is the first data with respect to salinity. Identifying and understanding the evolutionary significance of hidden genetic variation resulting from CnGV over environmental gradients is essential for conservation biology and predicting the effects of environmental change.

13.2

HYBRID BREAKDOWN AND PHYSIOLOGICAL COMPENSATION IN GENE EXPRESSION IN THE COPEPOD *TIGRIOPUS CALIFORNICUS*

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Allopatric populations diverge genetically as a result of mutation, selection and random drift. When divergent populations subsequently interbreed, interlocus interactions may lead to incompatibilities resulting in reduced hybrid fitness (hybrid breakdown). In the copepod *Tigriopus californicus*, hybrids between populations frequently suffer from hybrid breakdown that has been traced to mitochondrial dysfunction. To better understand the molecular mechanisms underlying hybrid breakdown, we used RNA-seq to compare patterns of gene expression between two divergent natural populations and their reciprocal hybrids. Unlike previous studies that found extensive underexpression of genes in hybrids between sister species, only 1.2% of genes in the transcriptome were misexpressed in *T. californicus* interpopulation hybrids, and nearly 80% of these were overexpressed rather than underexpressed. Moreover, the functional pathways encompassing the misexpressed genes (including OXPHOS and antioxidant response) are largely consistent with the known physiological consequences of mitonuclear incompatibilities in hybrid *T. californicus*. These results indicate that gene regulation in low-fitness interpopulation hybrids includes a signature of compensatory responses to mitochondrial dysfunction. Our results suggest that hybrid breakdown at early stages of speciation may result from initial incompatibilities amplified by the cost of compensatory physiological responses. Supported by NSF DEB1051057.

13.3

VARIATION BETWEEN STICKLEBACK POPULATIONS IN COLD TOLERANCE AND MECHANISMS OF FRESHWATER IONOREGULATION

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Most studies of adaptive divergence in physiological traits in different environments have examined single abiotic factors in isolation, yet environments seldom differ in only a single abiotic factor. Here, we use threespine stickleback (*Gasterosteus aculeatus*) to examine potential adaptive divergence in response to both low salinity and

cold temperatures by examining variation between populations in cold tolerance, growth at low temperature, and the effects of temperature on mechanisms of freshwater ionoregulation. We show that freshwater-resident stickleback have greater tolerance of acute exposure to low temperatures than do marine stickleback, but that this difference in cold tolerance is abolished with acclimation to low temperatures. Although acclimated marine stickleback could tolerate acute exposure to cold temperatures, they did not grow as well as freshwater stickleback at low temperature. Because size at first reproduction is strongly associated with fecundity in stickleback, these data suggest that winter temperatures during the first year of life could represent a barrier to colonization of freshwater habitats. There were substantial differences in ion transporter mRNA levels in the gill between populations and in response to low temperature exposure in fish in fresh water. These data suggest that challenges associated with ionoregulation in cold freshwater may, in part, explain the poor growth of marine stickleback in cold freshwater. Funded by NSERC.

13.4

TRANSGENERATIONAL INHERITANCE OF A STRESS PROTEOME IN THE LEAST KILLIFISH, *HETERANDRIA FORMOSA*, EXPOSED TO COPPER DURING EARLY LIFE STAGE.

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Nowadays, there is increasing evidence that pollutants can induce transgenerational impacts in a population. Assessing the toxicity of chemicals must take into consideration the possible inheritance of their effects through generations. In the present study, we have tested the hypothesis that an exposure to Cu in fish early life stage (ELS) can modify the protein expression profile in offspring. One week old viviparous least killifish, *Heterandria formosa*, were exposed to Cu at 15 µg/L for a period of one week, a condition inducing physiological adaptation. Fish were then held in clean water till maturity. After breeding, the expression profile of cytosolic proteins of 2 weeks old larvae was analyzed using 2D-DIGE followed by protein identification by nano-LC-ESI-MS/MS. We observed that Cu exposure in parents affects the expression of 50 protein spots in offspring, categorized into diverse functional classes related to protein turnover, chaperoning, metabolic process, ion transport or oxidative stress. Furthermore, we determined global DNA methylation in parents and in offspring using the LUMInometric Methylation Assay (LUMA). In conclusion, this study originally provides evidence that an exposure to a pollutant during ELS in a fish can affect the cellular phenotype in the offspring, assessed at the proteomic level. Ongoing researches will investigate the possible role played by epigenetics in this phenotypic inheritance.

13.5

RAPID EVOLUTION OF PHYSIOLOGY IN LABORATORY POPULATIONS OF *DROSOPHILA*

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We have examined a wide range of physiological characters in 30 outbred *Drosophila melanogaster* populations, all of which descend from one common ancestral population. These populations have well-known phylogenetic relationships and selection histories over the course of more than 30 years of sustained and replicated parallel selection. We have phenotypically compared these populations for the following characters: starvation resistance, desiccation resistance, flight endurance, and cardiac performance. The 30 populations in this study include long-established populations (ACO_{1.5}, B_{1.5}, and CO_{1.5}) and newly-established populations with corresponding recent selection regimes (AO_{1.5}, BO_{1.5}, and NCO_{1.5}). There was extensive physiological convergence between the long-established and newly-established populations subjected to the same selection regime. There was also rapid divergence among the populations newly subjected to the three different selection regimes. By conducting electric pacing and flight exhaustion assays with manipulative conditioning, we have started to unpack the physiological relationships between cardiac function, locomotor performance, and such other functional characters as fecundity, longevity and stress resistance. By conducting these manipulative experiments, we determined which particular stresses reduce or improve heart function, as well as the role of metabolic reserves in cardiac function.

13.6

PHYSIOLOGICAL SYNERGISM AND ANTAGONISM IN THE EVOLUTION OF LIFE HISTORIES

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Evolution acts on multiple traits simultaneously. We know little of how physiology enables or constrains the response to simultaneous selection on multiple traits such as body size and development time. From previous work, these two life history traits are

regulated by the interaction of three physiological factors: growth rate, the timing of the cessation of juvenile hormone secretion and the timing of secretion of the ecdysteroid hormones. Here we show how antagonism and synergism at the physiological level enables and constrains simultaneous selection at the phenotypic level of the two life history traits in the hawkmoth *Manduca sexta*. After 10 generations of selection on all four combinations of body size and development time the three physiological factors explained 93% of the response of development time to simultaneous selection and 99% of the response of body size. When the two life history traits were under synergistic selection, the response to selection was due largely to hormonal regulation and constrained by growth rate. When the life history traits were under antagonistic selection, the response to selection was due primarily to the change in growth rate and constrained by the two hormonal traits. Evidence suggests that the framework used here for the regulation of the response to simultaneous selection has broad applicability to a diverse range of taxa including green algae, plants, amphibians, mammals and other insects. Funding was provided by NSF-IOS.

13.7

DOES MMR REGULATE OF LIFE-HISTORY TRAITS? CORRELATED CHANGES IN MICE SELECTED FOR MASS-INDEPENDENT MMR

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Basal metabolic rate (BMR) is commonly used a measure of animal energetics in inquiries into potential trade-offs between metabolic expenditure and life-history traits. Maximal metabolic rate (MMR) measures the maximal aerobic capacity to metabolize energy, and might be involved in trade-offs. We tested how selection on different aspects of metabolic rates in laboratory mice (*Mus musculus*) affected induced immune responses, growth trajectories, and food consumption. Mice were bred for high mass-independent MMR, high mass-independent MMR plus low mass-independent BMR, or randomly with regard to metabolic rates. Selection for high mass-independent MMR suppressed inflammatory responses, but not antibody responses. Selection also altered overall growth patterns and increased maximal growth rates and food consumption. Changes in growth and immunology were correlated with evolved changes in mass-adjusted MMR, while changes in food consumption were correlated with evolved changes in mass-adjusted BMR. Consequently, MMR might be an important mediator of life-history traits, and measurements of MMR would provide insights when investigating associations between metabolism and life-history traits. Funded by NSF IOS-0344994 to JPH.

13.8

MOLECULAR EVOLUTION AND ADAPTATION OF INSULIN-LIKE/TOR SIGNALING ACROSS AMNIOTES

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Molecular evolution of the Insulin/Insulin-like Signaling (IIS) and Target of Rapamycin (TOR) network is central to modulating physiology, metabolic and life-history traits including growth, reproduction and aging. Nonetheless, it remains unclear which genes in the network are generally targets of positive versus purifying selection. Here, we focus on the divergence of the IIS/TOR network between amniote clades that are divergent in many aspects of their physiological: reptiles (including birds) and mammals. We examined 61 genes in the IIS/TOR network across 66 amniote species - the most comprehensive taxonomic depth to date; including 18 liver transcriptomes we generated from non-avian reptiles. We found that 1) members of the IIS/TOR network have exceptional evolutionary rates between reptiles and mammals, 2) extracellular and membrane genes of the network evolve faster and are under stronger positive selection than intracellular signaling genes, 3) genes with fewer interactions with other proteins/genes have faster evolutionary rates than genes with higher connectivity, and 4) the two primary ligands (*igf1* and *igf2*) have opposite patterns of selection in mammals vs. reptiles. The phylogenetic depth and breadth of our data, along with the patterns of divergence in this network suggests that IIS/TOR network may be one key to the diversification of physiological and life-history traits between reptiles and mammals. NSF IOS1253896, ISU CIAG, JSM Foundation.

14.0: METABOLISM, ENERGETICS, AND PERFORMANCE

14.1

ONTOGENETIC AND INTERSPECIFIC METABOLIC SCALING IN INSECTS

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Design constraints imposed by increasing size cause metabolic rate in animals to increase more slowly than mass. This ubiquitous biological phenomenon is referred to as metabolic scaling. However, mechanistic explanations for interspecific metabolic scaling do not apply for ontogenetic size changes within a species implying different mechanisms for scaling phenomena. We show that the Dynamic Energy Budget theory approach of compartmentalizing biomass into reserve and structural components provides a unified framework for understanding ontogenetic and interspecific metabolic scaling. We formulate the theory for the insects and show that it can account for ontogenetic metabolic scaling during the embryonic and larval phases, as well as the U-shaped respiration curve during pupation. After correcting for the predicted ontogenetic scaling effects, which we show to follow universal curves, the scaling of respiration between species is approximated by a $\frac{3}{4}$ power law, supporting past empirical studies on insect metabolic scaling and our theoretical predictions. The ability to explain ontogenetic and interspecific metabolic scaling effects under one consistent framework suggests that the partitioning of biomass into reserve and structure is a necessary foundation to a general metabolic theory. This work was supported by a Discovery Project (DP110101776) and Australian Research Fellowship (DP110102813) grant from the Australian Research Council.

14.2 COMBINING X-RAY SYNCHROTRON IMAGING OF AMBER FOSSILS WITH BODY SIZE CHANGES IN THE INSECT FOSSIL RECORD TO ELUCIDATE THE EFFECT OF ATMOSPHERIC OXYGEN ON PALEO-PHYSIOLOGY

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Changes in atmospheric oxygen levels have been hypothesized to have led to the evolution of insect gigantism. However, the fact that not all insects exhibited gigantism coupled with the paucity of the fossil record and the complex interactions between oxygen and organisms makes it difficult to definitively accept or reject the oxygen-size link. To address these questions, we examined changes in the fossil record in light of our understanding of how oxygen affects modern insect physiology. Here we carried out the first statistical study of average and maximum body size of insect fossils across geologic times of both high and low oxygen levels. The results of these studies support the link between fluctuations in oxygen and insect evolution: 1) the maximal and average size of *Protodonata* and *Paleodictyoptera* fossils correlate positively with modeled atmospheric oxygen, 2) *Blattodea* fossils showed little variation in maximum size, but average size was correlated with atmospheric oxygen, and 3) the Triassic hypoxic event appears to have a larger impact on insect body size than the Paleozoic hyperoxic event. Secondly, we have used x-ray synchrotron imaging to generate 2D images and 3D tomographic reconstructions of the tracheal system of both modern insects and amber fossils. The ability to measure fossil tracheae provides a unique look at the impact of oxygen on insect paleophysiology and a possible biological proxy for atmospheric oxygen. Supported by NSF EAR 0746352.

14.3 DIEL CHANGES IN CORAL METABOLISM: POTENTIAL REGULATION BY PHOSPHORYLATION

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Corals experience hyperoxic conditions during the day due to photosynthetic activity by their symbiotic algae and hypoxic conditions at night due to cellular respiration by the coral holobiont. Previous studies have revealed corals undergo daily oscillations in the expression of putative metabolic genes. However, the main anaerobic energy producing pathways in corals, as well as the mechanisms by which corals switch from aerobic to anaerobic metabolism, are unknown. To study if phosphorylation plays a role in regulating metabolism in corals, we compared metabolic enzyme activity and phosphorylation levels between day and night in the coral *Acropora yongei*. Branches of *A. yongei* were maintained under a 12h:12h light:dark cycle, and samples were taken one hour before the lights turned off and one hour before lights turned on to capture daytime and nighttime metabolism, respectively. Activities of the aerobic enzymes citrate synthase and malate dehydrogenase were significantly higher during the day than at night, suggesting higher metabolic flux during aerobic conditions. Lactate dehydrogenase activity was negligible both during the day and night, indicating that, unlike vertebrates, it is not a major anaerobic enzyme in *A. yongei*. The protein phosphorylation pattern during the day was different than at night; ongoing studies will identify if any of the differentially phosphorylated proteins are metabolic enzymes. Further studies relating changes in intracellular oxygen concentrations to metabolism and metabolic regulation will help reveal underlying mechanisms of daily metabolic rhythms in corals to inform predictions of their biological responses to future global change. NSF EF-1220641.

14.4 SPECIFIC DYNAMIC ACTION IN DECAPOD CRUSTACEANS

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The metabolic consequences of feeding and digestion (specific dynamic action) were investigated in a number of decapod species. The animals exhibited the typical pattern of postprandial oxygen uptake characterized by a rapid increase, followed by a more prolonged decrease to pre-feeding levels. There was an increase in the SDA with increasing meal size. However, unlike other taxa, this increase was primarily afforded by an increase in the duration oxygen uptake remained elevated, rather than an increase in peak oxygen consumption. The texture of the meal appeared to play a greater role than the actual nutrient content; harder meals required more energy to digest. While previous work on non-decapod crustaceans suggests that mechanical digestion only comprises a small part of the overall SDA budget, the current findings showed that 30% of the SDA budget could be due to mechanical digestion. This is not unexpected since the gastric mill, which processes food, is a complex apparatus controlled by over 40 muscles. In fish, some characteristics of the SDA response are closely related to food transit rates, whereas in crustaceans there was no relationship, probably due to differences in architecture and functioning of the gut. The metabolic changes associated with the SDA are discussed in relation to environmental perturbations and the ability of animals to balance the simultaneous demands of several physiological systems. Funding-NSERC.

14.5 REAL-TIME PHYSIOLOGY: CAN IT ASSIST AQUACULTURE PRODUCTIVITY?

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Aquaculture is a rapidly expanding, multi-billion dollar industry worldwide (~USD 119.4 billion in 2010) and sustained growth is essential for food security. Understanding how target aquaculture species respond to a variety of environmental conditions (e.g. temperature, oxygen concentration, emersion time) is essential for maximizing productivity. However, there are still persistent knowledge gaps in regards to the physiology of key intensively reared aquaculture species, and in how that knowledge is collected and applied to commercial farming. Oysters are cultured globally and are difficult to monitor due to their shell and grow-out systems. Recent advancements in biosensors have enabled long-term, non-invasive monitoring of a range of physiological and environmental variables that are relevant to animal health and productivity including heart rate, temperature, shell gape, water depth and light level. This study forms part of a larger body of work examining how the environment and farm stressors affect oyster physiology. We are exploiting the relationship between heart rate and metabolic rate in the laboratory and in the field (on farms) to build the predictive algorithms for real-time sensing to ultimately lead to decision support systems for the aquaculture industry.

14.6 TEMPERATURE AND ACIDIFICATION VARIABILITY REDUCE PHYSIOLOGICAL PERFORMANCE IN THE INTERTIDAL ZONE PORCELAIN CRAB *PETROLISTHES CINCTIPES*

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Under future climate scenarios, coastal ecosystems are projected to have increased extremes of low tide-associated thermal stress and ocean acidification-associated low pH, the individual or interactive effects of which have yet to be determined. To characterize energetic consequences of exposure to increased variability of pH and temperature, we exposed porcelain crabs, *Petrolisthes cinctipes*, to conditions that simulated current and future intertidal zone thermal and pH environments. During the daily low tide, emersed specimens were exposed to no, moderate, or extreme heating (11°C, 25°C, and 30°C, respectively), and during the daily high tide experienced no, moderate, or extreme acidification (pH 8.1, 7.6, and 7.15, respectively). Respiration rates and upper thermal limits of cardiac performance were assessed following 17 days of exposure. Thermal variation had a larger overall effect than pH variation, though there was an interactive effect between the two environmental drivers. Under the most extreme temperature and pH combination, respiration rate decreased 25% from controls while heat tolerance increased 2°C from controls, indicating a smaller overall energy budget of which a larger portion is devoted to basal maintenance (i.e. survival). These results suggest negative long-term ecological consequences, such as reduced energy for behavior and reproduction, for intertidal ectotherms exposed to increased extremes in pH and temperature. Funding by NSF MEB-1041225 to JHS.

14.7

WHAT LIMITS PERFORMANCE IN WILD PACIFIC BLUEFIN TUNA?

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Understanding the role of physiological limitations in changing seas requires models that integrate metabolism, cardiac physiology and thermal constraints. Measuring such limits in nature is difficult. We combine field and lab studies to understand the physiological limitations of Pacific bluefin tuna in the California Current Ecosystem (CCS). Tuna were archival tagged (n=520) in the CCS data recorded provided body temperature, ambient temperature, pressure and position. A lab validated model was used to calculate energy intake (kcal/day) in 144 fish using the heat increment of feeding (HIF). HIF events occurred on 91 % of tuna days at large (n= 39,000 HIFs). Daily energy intake estimates indicate a median value of 820 kcal and a maximum of 4106 kcal. An "energy utilization landscape" of foraging physiology in the CCS enabled evaluation of tuna movements in relationship to environmental conditions. Energy intake was correlated with temperature, thermocline depth, and chlorophyll a. As ambient temperatures increased at the southern end of the range, energy intake declined but energetic costs increased. Tuna move north to more favorable conditions off California where HIF events indicate large increases in energy intake. Fish move south as cooler conditions persist in winter. The integration of wild energetic data with lab obtained cardiac and blood data permits investigating hypotheses on the physiological limits and whole animal performance of Pacific bluefin tuna.

14.8

VALIDATION OF THE RELATIONSHIP BETWEEN 3-DIMENSIONAL BODY ACCELERATION AND OXYGEN CONSUMPTION IN TRAINED STELLER SEA LIONS (*EUMETOPIAS JUBATUS*) DIVING WITH INCREASED OXYGEN DEPLETION

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We tested the ability of overall dynamic body acceleration (ODBA) to predict the rate of oxygen consumption $\dot{V}O_2$ in freely diving Steller sea lions under a broad range of physiological conditions. The sea lions executed dives to 40m within three dive types (single dives, bouts of short dives with 4-6 dives per bout, or bouts of long dives with 10-12 dives per bout), leading to a range of oxygen debts and activity levels. We found that while ODBA predicted $\dot{V}O_2$ for animals when data from all dive types were combined, dive type was a significant model factor. The potential relationships between $\dot{V}O_2$ and ODBA were not clarified by dive duration, food consumed, proportion of dive cycle spent diving, or number of dives per bout. However, when data from each dive type was analyzed separately, there were no significant linear relationships within any dive types. It is not clear whether the lack of predictive power within dive type was a result of a lack of statistical power, or whether it reflected a true absence of a physiological relationship. The average percent error for all significant models was 6.8-10.4% and standard error of the estimated $\dot{V}O_2$ was 2.5-35.9%. Overall, the extensive range of dive behaviours and physiological conditions we tested in our study did not improve the ability of ODBA to predict $\dot{V}O_2$. Funding was provided by a grant from NPMSF to North Pacific Universities Marine Mammal Research Consortium, US NOAA, and Deakin University.

15.0: OSMOTIC AND ION REGULATION: JUNCTIONS AND TRANSPORTERS

15.1

A ROLE FOR SEPTATE JUNCTION PROTEINS IN THE REGULATION OF SALT AND WATER BALANCE IN LARVAL MOSQUITO (*Aedes Aegypti*)

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The septate junction (SJ) of invertebrate epithelia occludes the intercellular space between invertebrate epithelial cells. SJs are composed of transmembrane and cytosolic SJ proteins, some of which are reported to be essential for epithelium integrity. The contribution of SJ proteins to the maintenance of salt and water balance in aquatic invertebrates is not known. In this study, a role for SJ proteins in the ionoregulatory strategies of larval mosquito (*Aedes aegypti*) was considered. These animals reside in freshwater (FW), but are capable of tolerating saline environments. In either case, it can be hypothesized that SJ proteins are involved in regulating the ionic and osmotic gradient between internal and external environments. Genes encoding *A. aegypti* SJ proteins megatrachea, sinuous, coracle, scribble, discs large, neurexin IV, lethal giant

larva, gliotactin, mesh and snakeskin were identified. SJ proteins exhibited tissue specific differences in transcript abundance and SJ proteins immunolocalized to regions of cell-to-cell contact between epithelial cells. Tissue specific changes in SJ protein mRNA abundance were also observed when *A. aegypti* larvae were reared in either FW or brackish water, and this occurred in conjunction with changes in paracellular permeability. Data support the hypothesis that SJ proteins play an important role in the maintenance of salt and water balance in the aquatic larvae of *A. aegypti*. Funding: NSERC.

15.2

TRICELLULAR TIGHT JUNCTION PROTEINS AND THEIR CONTRIBUTION TO PARACELLULAR OCCLUSION IN THE FISH GILL EPITHELIUM

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The vertebrate tight junction (TJ) complex is an apico-lateral structure that occludes the paracellular space and largely controls the paracellular permeability of an epithelium. Around the periphery of epithelial cells bicellular TJs (bTJs) form the majority of the paracellular interface by linking two bordering cells. Where three epithelial cells interface tricellular TJs (tTJs) connect and run vertically in an apical-to-basal direction. In vertebrates, tTJ proteins described to date include tricellulin, lipolysis-stimulated lipoprotein receptor and immunoglobulin-like domain-containing receptor. Because tTJs are necessary for intercellular occlusion at barrier weak points, it can be hypothesized that these structures and their protein composite will play an important role in the maintenance of barrier integrity in tight epithelia that interface between extracellular fluid and the external environment of an animal. One such tissue is the gill epithelium of fishes which separates extracellular fluid from a surrounding medium that differs greatly in ionic composition. This presentation considers the role that tTJ proteins play in the gill epithelium of teleost fishes both in terms of how tTJ proteins contribute to the electroresistive and paracellular properties of an *in vitro* gill model as well as how the endocrine system and environment impact their apparent involvement in maintaining gill epithelium integrity. Funded by NSERC Canada.

15.3

ABUNDANCE AND LOCALIZATION OF BRANCHIAL CLAUDINS IN RAINBOW TROUT (*ONCORHYNCHUS MYKISS*) AND IMPLICATIONS IN HYPOSMOREGULATION

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Tight junction proteins of the claudin family are known to control paracellular permeability. In euryhaline fish, changes in the leak pathway across the gill epithelium are critical during salinity acclimation and are a result of altered claudin abundance and localization. The aim of this study was to evaluate the role of Claudin-10c, -d, -e and Claudin-30 in gill function in freshwater (FW) and seawater (SW) rainbow trout. We examined mRNA and protein abundance along with localization. A tissue distribution survey showed that the 4 claudins studied were predominantly expressed in gill tissue. Transcript and protein expression of all Claudin-10 isoforms were significantly up-regulated after SW transfer (p<0.001), while no difference in Claudin-30 expressions was observed. In accordance with these expression patterns, *in silico* prediction showed that Claudin-10c, d and e could form cation-selective pores and thus be critical to sodium secretion in SW. Claudin-30 is known as a resistance forming claudin and its unchanged abundance with salinity suggests an epithelial barrier function in both FW and SW gills. Immunofluorescence microscopy revealed that Claudin-10e is localized exclusively in association with ionocytes in SW. Ongoing investigation is focusing on the subcellular localization of Claudin-10 isoforms. This study was supported by the National Science Foundation (IBN 12-51616) and the Arkansas Bioscience Institute.

15.4

HYPOTONICITY STIMULATES K⁺ FLUX THROUGH THE WNK-SPAK/OSRI KINASE CASCADE AND THE NCC69 SODIUM-POTASSIUM-2-CHLORIDE COTRANSPORTER IN THE DROSOPHILA RENAL TUBULE

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Objective: Adult *Drosophila melanogaster* maintain hemolymph osmolarity within a narrow range. Hypotonicity increases fluid secretion in insect renal tubules, but the mechanisms are unknown. Here, we examined whether the fly NKCC, Ncc69, and its kinase regulators, with-no-lysine (WNK) and the Ste20-related proline alanine rich (SPAK)/oxidative stress response (OSR1) homolog Fray, mediates this response.

Methods: We used the Ramsay assay to measure main segment fluid secretion and K⁺ flux in *Drosophila* Malpighian tubules. *In vitro* kinase assays were performed

using proteins purified from *E. coli*. **Results:** Decreasing *Drosophila* WNK activity in the principal cell of the tubule caused a reduction in K⁺ flux. Similarly, knocking down the SPAK/OSR1 homolog *fray* also decreased K⁺ flux. A hierarchical WNK-Fray signaling cascade regulates K⁺ flux through Ncc69, since i) a constitutively active Fray mutant rescued the *wnk* knockdown phenotype; ii) Fray directly phosphorylates Ncc69 *in vitro*; and iii) the effect of *wnk* and *fray* knockdown was abolished in *Ncc69* mutants. The stimulatory effect of hypotonicity on K⁺ flux was absent in *wnk*, *fray*, or *Ncc69* mutant tubules. **Conclusion:** Hypotonic conditions stimulate K⁺ flux through the activation of the WNK-Fray-Ncc69 pathway in the principal cell of the fly renal tubule. Increased net fluid secretion provides a mechanism for enhanced clearance of a water load and the homeostatic maintenance of extracellular osmolarity.

15.5

ANURAN-SPECIFIC AQUAPORIN 2 HOMOLOG IN A URODELE, EVIDENCE FOR EARLY GENE DUPLICATION IN AMPHIBIAN EVOLUTION

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Amphibians are unique among living vertebrates in that their skin serves as a primary surface for water absorption from pond water and moist substrates. Cutaneous water absorption by anurans is regulated by arginine vasotocin-stimulated insertion of anuran-specific aquaporins (AQP2s) into the apical membrane of the outermost living cell layer. Synteny analysis suggests that AQP2s are syntenic with AQP2, and they arose from gene duplication in the course of amphibian evolution. The chronology for the divergence of urodeles (order caudata) from anurans is hotly debated but dates to at least the Permian era. Our hypothesis is that the gene duplication that gave rise to "anuran-specific" AQP2s occurred prior to this divergence. Here we characterize an aquaporin in the skin of the newt, *Notophthalmus viridescens*, that has sequence identity intermediate between AQP2s. Immunohistochemistry shows this aquaporin (AQP-nv3) to be translocated to the apical membrane when dehydrated newts rehydrate. These observations demonstrate the gene duplication giving rise to cutaneous expression of AQP2 homologs occurred prior to the divergence of anurans and urodeles. Supported by Grants-in Aid for Scientific Research (26440165) from the Ministry of Education, Culture, Sports, Science and Technology of Japan.

15.6

SEQUENCING AND GENE EXPRESSION OF AQUAPORIN-9 IN FREEZE TOLERANT COPE'S GRAY TREEFROGS

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Cope's gray treefrogs *Hyla chrysoscelis* accumulate glycerol in response to cold as part of their freeze tolerance mechanism. This glycerol is suspected to derive from glucose that is stored in the liver before it is exported through the bloodstream to other tissues. We therefore hypothesized that tissues from *Hyla chrysoscelis*, including hepatocytes, would express aquaporin 9 (AQP9), a transmembrane protein from the aquaglyceroporin family that facilitates glycerol transport. We also hypothesized that AQP9 would be up-regulated during cold acclimation to promote glycerol permeability. To test this hypothesis, we designed primers based on published AQP9 sequences, generated cDNA from mRNA extracted from liver, and identified a gene, HC-9, with high homology (69%) to human AQP9. Quantitative PCR analysis revealed HC-9 expression in liver, stomach, bladder, kidney, eye, lung, muscle, and skin (but not in intestine, brain, heart, fat, or red blood cells). In tissues from frogs that had been cold acclimated, there was no statistical up-regulation of HC-9, and a statistical down-regulation in muscle and stomach, contrary to our hypothesis. In frozen and thawed animals, expression of HC-9 mRNA was down-regulated in most tissues. Future assessment of HC-9 protein expression will help to identify the role of this protein in cold acclimation and freeze tolerance. Supported by NSF IOS-1121457.

15.7

GENE EXPRESSION OF THREE ISOFORMS OF UREA TRANSPORTER IN AN AESTIVATING AFRICAN LUNGFISH

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The African lungfish, *Protopterus annectens*, is amonotelic in water. However, it increases urea synthesis and accumulation during the induction and maintenance phases of aestivation and turns ureotelic upon arousal from aestivation. The drastic increase in urea excretion upon arousal necessitates the involvement of urea transporters (UTs). This study aimed to clone and sequence isoforms of *ut* from the liver of *Protopterus annectens* and examine their mRNA expression in various tissues/organs of during the three phases of aestivation. Three *ut* isoforms were expressed by *P.*

annectens and aestivation had different effects on their mRNA expression in various tissues/organs. Our results shed light on how urea transport was regulated in different tissues/organs in *P. annectens* during the transition between amonotely and urea accumulation during the induction phase of aestivation and the transition between urea accumulation and ureotelic during the arousal phase of aestivation. This study was approved by the NUS IACUC.

15.8

DEHYDRATION AND THIRST OF HYDROPHINE SEA SNAKES

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Secondarily marine vertebrates are challenged to obtain water in saline environments. Recent investigations have indicated that several species of marine snake dehydrate at sea and are dependent on environmental sources of fresh water, representing a shift of paradigm from previous "textbook" literature. However, freshwater requirements in the majority of sea snakes, represented by hydrophine species, are not known. Hence, we investigated the extent of dehydration and responses to fresh water in five species of hydrophine sea snakes collected during seasonal drought near Weipa, Cape York Peninsula, Australia. Very few snakes that we collected drank fresh water immediately following their capture, even though measurements of total body water and condition of feces indicated these snakes were dehydrated. None of these species drank sea water after dehydration in air and offered sea water or fresh water to drink. Dehydrated individuals of three species drank fresh water, but only at mean dehydration thresholds of -26 to -29 % of initial body mass. Two other species did not drink fresh water when similarly dehydrated. Available data indicate that sea snakes have relatively high levels of total body water (around 80% of body mass), are comparatively resistant to dehydration, and have diverse thresholds for thirst. New data indicate that some species possibly live independently of environmental fresh water. Funding was provided by NSF grant IOS-0926802 to HBL.

16.0: TRAINEE WORKSHOP: NON-TRADITIONAL CAREER PATHS FOR COMPARATIVE PHYSIOLOGISTS

16.1

THE SOFT MONEY RESEARCH POSITION: HOW DOES IT WORK?

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The number of students receiving doctorates in biology in the United States increased from 3,803 in 1981 to 8,135 in 2011. Meanwhile the number of biological-science Ph.D. recipients in tenure-track positions dropped precipitously from 55% in 1973 to 15% in 2006, and industry has not absorbed the slack. Soft money research positions, where funding comes from grants and contracts, are an alternative to maintaining a research career in the absence of available tenure-track positions. Although these positions can seem career limiting or demoralizing at times since the success rates on grants and contracts are low, a strong will to survive and a tough skin can make this career trajectory work. In addition, this pathway offers a solution for balancing family and career conflicts, such as providing a flexible schedule for child rearing and helping to solve the dual career problem. Being successful requires diversification of skills and experiences, working effectively in groups, and giving a good sales pitch. For over a decade I have maintained 100% funding on soft money to solve my dual career problem. In this presentation, I will provide my recipe for success and the pros and cons of this career alternative. References: M. Barinaga. 2000. Soft money's hard realities. Science 289:2024-2028. D Cyranoski, N Gilbert, H Ledford, A Nayar, M Yahia. 2011. The Ph.D. factory. The world is producing more Ph.D.s than ever before. Is it time to stop? Nature 472:276-279. AA Shea. 2013. For graduate programs, it's time to get real. The Chronicle of Higher Education.

16.2

SCIENCE FOR THE PUBLIC! CAREERS IN SCIENCE CENTERS AND MUSEUMS

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Intrigued by the possibility of bringing your knowledge and passion for science to non-specialists? Interested in working with teachers, K-12 students, and the general public? Curious about what it takes to be a scientist in a museum or science center? We'll discuss the joys and challenges of being a scientist doing "informal" science, and look at the variety of paths to that end.

16.3

TAKING THE ROAD LESS TRAVELED: NON-GOVERNMENTAL ORGANIZATIONS

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Life after graduate studies in comparative physiology often focuses on academic career paths, or to a lesser extent, industry and governmental paths. Less common is the research path associated with non-profit institutions. Affiliation with non-profit research institutions can often be similar to a life on soft money, yet the avenue enables means for acquiring research funds that are more expansive than can be pursued by government institutions or that are pursued by academia and industry. Non-profit status permits access to grants and contracts from federal funding sources as well as private foundations and direct donations from the public. The National Marine Mammal Foundation (NMMF) is a non-profit with a business model that involves stable income through veterinary, research, and training support it provides to the United States Navy Marine Mammal Program (MMP). This affiliation, which is supported via a federal contract, additionally provides access to dolphins and sea lions for physiological research, which is leveraged to address issues related to the NMMF mission and vision. Research on MMP dolphins is funded through grants from federal agencies and commercial institutions, and complements additional NMMF activities (e.g. conservation efforts) supported by private foundations and direct public funding. Comparative physiological research as a non-profit is not a path suited for those expecting to survive off of the occasional grant, but requires active pursuit of research opportunities and the cross-fertilization of ideas and fields, often requiring the individual to become knowledgeable in multiple fields. However, the career path opens opportunities that might otherwise be limited by institutional affiliation and can be a lucrative path to the motivated researcher.

16.4

THE UNDERGRADUATE UNIVERSITY PATH

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Undergraduate institutions present distinct opportunities in teaching and research and provide a rewarding career option. Broadly trained comparative physiologists are uniquely poised to succeed at such institutions. This discussion will present a brief overview of the range of undergraduate institutions and faculty positions. We will examine the day-to-day activities, expectations, benefits and challenges of faculty at undergraduate institutions. Finally, the remainder of the presentation will provide advice for how graduate students and post docs can best prepare to succeed when applying to undergraduate jobs, as well as tips for the application/interview process. Reference: Campbell, A. M., Quintero, O. A., and Frederick, J. (2012). How to get a teaching job at a primarily undergraduate institution. The American Society for Cell Biology.

16.5

THE TRANSITION TO INDUSTRY

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In this talk I will go over some of the considerations regarding a career move from academia to the private sector. For biologists, the private sector available is, in large part, composed of the pharmaceutical industry, the biotechnology industry, and contract research groups (both non-profit and for-profit). What is similar between working in academic labs and private-sector labs, and conversely what is different about these work environments? Is the move to the private sector irreversible? What are the types of jobs that are available to biologists in the private sector? What do you need to learn about prospective employers in the private sector before pursuing a job with them, and also what do you need to know about yourself before deciding what to pursue?

17.0: OVERCOMING A MAJOR PHYSIOLOGICAL BARRIER: ADAPTATION FROM SALINE TO FRESHWATER HABITATS

17.1

RAPID EVOLUTION OF IONIC REGULATION DURING SALINE TO FRESHWATER HABITAT INVASIONS

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Marine to freshwater transitions constitute major evolutionary events in the history of life, serving as stepping-stones for the colonization of land. Invasive species are often striking in their capacity to extend their ranges into novel habitats, providing valuable models for studying incipient adaptations during colonizations. Within the past century the copepod *Eurytemora affinis* has invaded freshwater habitats multiple times independently from saline sources. These invasions were accompanied by evolution of increased freshwater tolerance and hemolymph osmolality at low salinities and evolutionary shifts in activity and expression of ion transport enzymes (V-type H⁺ ATPase, Na, K-ATPase). These evolutionary changes occurred in parallel across independent invasions and in laboratory selection experiments. High-food concentration significantly increases low-salinity tolerance, allowing saline populations to overcome the freshwater barrier, and food x salinity response has evolved following

freshwater invasions. The copepod microbiome also shows parallel shifts across independent invasions. We are exploring interactions among key factors underlying freshwater adaptation and are probing transcriptional responses of the copepod to these factors, as well as shifts in its microbial metagenome as it invades. NSF OCE-1046372, DEB-0745828, DEB-0448827. Lee *et al.* 2013. Feasting in Fresh Water: Impacts of food concentration on freshwater tolerance and the evolution of food x salinity response during the expansion from saline into freshwater habitats. *Evol. Appl.* 6:673-689. Lee *et al.* 2012. Rapid evolution of body fluid regulation following independent invasions into freshwater habitats. *J. Evol. Biol.* 25:625-633. Lee *et al.* 2011. Pumping ions: Rapid parallel evolution of ionic regulation following habitat invasions. *Evolution*. 65:2229-2244.

17.2

COLONIZATION OF FRESHWATER HABITATS FROM THE MARINE ENVIRONMENT: LESSONS FROM STICKLEBACK AND KILLIFISH

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Colonization of freshwater from the marine environment requires a fundamental shift in the physiology of ionoregulatory tissues from ion secretion to ion absorption. To understand the mechanisms associated with this shift, we have taken a comparative approach using populations of fish living in different salinity environments. Here, I summarize our recent work related to this question using threespine sticklebacks (*Gasterosteus aculeatus*) and killifish (*Fundulus heteroclitus*). The distribution patterns of both of these species have been profoundly affected by the history of glacial cycles. With the retreat of the glaciers following the last glaciation, anadromous stickleback repeatedly colonized newly emerging freshwater habitats. In killifish, which are found in estuaries along the Atlantic coast, northern populations exhibit substantially greater tolerance of freshwater than do the southern populations that were not directly associated with the glacial margins. In both killifish and stickleback there are points in the species range where populations that differ in salinity tolerance come into contact and interbreed. This has resulted in sharp genetic clines in both species that are centered at the transition between the freshwater and saltwater parts of estuaries. The maintenance of these clines in the face of ongoing gene flow is consistent with the action of natural selection, and can provide insight into the mechanisms of ionoregulation in freshwater fishes.

17.3

OSMOREGULATION IN THE ANADROMOUS LAMPREY *PETROMYZON MARINUS*

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The lamprey gill shares many morphological similarities with its teleost counterpart and it has generally been predicted that similar ion transport mechanisms also occur at the molecular level. Thus although the lamprey lineage diverged over 550 MYA, the story of ion regulation is likely equivalent. Using the anadromous marine lamprey, *Petromyzon marinus*, we test this hypothesis using a combination of molecular genetics and immunohistochemical approaches to elucidate the repertoire of branchial ion transporters. We find that CFTR Cl⁻ channel (Abcc7), and Na⁺: K⁺: 2Cl⁻ cotransporter 1 (Slc12a1) orthologues are transcribed in gill which supports homology with the teleost seawater ionocyte secondary active Cl⁻ secretory mechanism during the marine phase of the lamprey life cycle. These Chloride Cells have also been demonstrated to be also rich in Na⁺/K⁺-ATPase (NKA, Atp1a1). In freshwater, hypertonic regulation is facilitated by the vacuolar-type proton ATPase (e.g. Atp6v1b and Atp6v1e) and Na⁺: Cl⁻ cotransporter (Slc12a3) as has also been shown in some teleost fishes. However, in addition, lamprey express a salinity sensitive epithelial Na⁺ channel (ENaC, Scnn1) and H⁺/K⁺-ATPase (Atp12a) which are absent in teleosts but present in sarcopterygii. To summarize, we have found that although there are many similarities with more extensively studied teleost species, there are also some striking differences which can be traced back to a lineage specific gene loss event in the teleost ancestor.

17.4

EVOLUTIONARY TRANSITION TO FRESHWATER IN THE SHRIMP *MACROBRACHIUM AMAZONICUM*: ECOPHYSIOLOGICAL ADAPTATIONS

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Osmoregulation was compared in two separate populations, recently separated into two species from the Amazon delta (A: *Macrobrachium amazonicum*) and from the Pantanal (P: *M. pantanalense*). In A, all stages are hyper-hypo-osmoregulators but only zoea 1 and postlarval stages also hyper-regulate in FW. Thus, larvae of A can

hatch in riverine FW but must then develop in brackish estuarine waters. The P shrimps are land-locked in FW. All stages hyper-regulate in FW but are osmoconformers at salinities ≥ 17 ppt. A and P shrimps in FW produce hypotonic urine. Gill develop faster in P vs A larvae, so that Na⁺/K⁺-ATPase (NKA) is localized in early larval gills of P, but only in branchiostegites of A. In juvenile A and P, V-H⁺-ATPase (VHA) and NKA are localized in a single branchiostegite cell type. In FW gills, VHA is localized apically in pillar cells, while NKA is restricted to septal cells. These cells likely cooperate in ion uptake. In A shrimps, VHA mRNA expression is higher in FW. In P shrimps, the loss of hypo-osmoregulation may be related to the lack of ion transport in the branchiostegites, and the ability of all stages to hyper-regulate in FW may be due to the early development of functional gills. These results illustrate evolutionary adaptations (loss and gain of functions) underlying the invasion of FW habitats. Reference: Boudour-Boucheker N, Boulo V, Lorin-Nebel C, Elguero C, Grousset E, Anger K, Charmantier-Daures M, Charmantier G. Adaptation to freshwater in the palaemonid shrimp *Macrobrachium amazonicum*: comparative ontogeny of osmoregulatory organs. *Cell Tissue Research* 353, 87-98, 2013.

18.0: CHALLENGES FROM THE VERY BEGINNING: DEVELOPMENTAL PHYSIOLOGY, EPIGENETICS, AND CRITICAL WINDOWS

18.1 CRITICAL WINDOWS IN ANIMAL DEVELOPMENT: STRESSOR DOSE, EFFECT SIZE AND EXPERIMENTAL DESIGN

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An animal's developmental trajectory is a result of interactions between genome and the environment. The ability to modify phenotype via phenotypic plasticity allows animals to cope with challenges during development, including altered environmental conditions. Critical windows are periods during embryonic development and/or early life when phenotypes are particularly plastic and responsive to intrinsic or extrinsic (environmental) factors. Using embryonic Lake whitefish (*Coregonus clupeaformis*), I demonstrate how critical windows can be viewed as 1-dimensional - discrete periods during development in which phenotypic modification occurs. However, a 3-dimensional construct in which the relationship between time during development, dose of the stressor applied, and the resultant phenotypic modification can define a critical window in much more detail. Using the example of survival and morphology of larval *Artemia* raised in different salinity levels during development, I demonstrate how the interaction between exposure time and stressor dose causes a measured phenotypic change, which itself may vary within a critical window. This 3-dimensional experimental design potentially reveals much more about interactions between environment and phenotype during development than traditional chronic exposure studies. Reference: Burggren WW & Mueller CA 2014. A 3-dimensional, system approach for developmental critical windows and sensitive periods: Is a "window with sharp edges" too simplistic a view? *Physiol Biochem Zool*, in review.

18.2 NOISY EMBRYOS? THE POTENTIAL EVOLUTIONARY IMPORTANCE OF VARIATION IN THE TIMING OF DEVELOPMENTAL EVENTS

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Heterochrony - differences in the timing of developmental events between species - is considered by some as a key link between ontogeny (development) and phylogeny (evolution). However, evidence for the mechanistic basis of heterochrony is limited. Here I suggest how variation in the timing of developmental events within species could act as the raw material for the evolution of heterochrony. *In vivo* measurements from developing embryos have demonstrated levels of heterochrony within a clade of freshwater gastropods similar to those observed in vertebrates. High levels of variation in the timing of developmental events have also been demonstrated in the freshwater pulmonate, *Radix balthica*, and this variation is increased in stressed embryos. Moreover, a parent-offspring comparison of development showed that the timing of some events such as shell formation and the initiation of crawling are heritable; the fitness benefits of such variation are currently being explored. Together these findings suggest that intraspecific variation in developmental event timing could provide the raw material for selection to produce heterochrony. References: Tills, O., Rundle, S.D. & Spicer, J. (2013) Parent-offspring similarity in the timing of developmental events: a potential link between ontogeny and phylogeny. *Proceedings of the Royal Society B* 280: 1769 doi:10.1098/rspb.2013.1479. Rundle, S.D., Smirthwaite, J.J., Colbert, M.W. & Spicer, J.J. (2011) Predator cues alter the sequence of developmental events in gastropod embryos. *Biology Letters* 7: 285-287.

18.3 MITIGATING THE RISKS ASSOCIATED WITH ACCELERATED OR DEFICIENT PERINATAL GROWTH

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Across species, variation in intrauterine and neonatal development promotes physiologic diversity, but individuals at the extremes of perinatal growth are at an increased risk of adult disease. Independent of genetic susceptibility, strong correlations are present between perinatal weight gain, central leptin resistance and adult metabolic syndrome. In contrast, perinatal growth restriction increases hypothalamic leptin sensitivity and decreases the risk of adult obesity, but heightens the risk of neurodevelopmental impairment and hypertension. Global "catch-up growth", after the critical perinatal window has closed, elicits adult obesity and exacerbates the programmed hypertensive phenotype. Targeted pathway-specific interventions are thus needed to optimize neurodevelopmental outcome without compromising cardiometabolic health. As a key neurotrophic and homeostatic hormone, leptin has emerged as a potential mediator of programmed adult cardiovascular and metabolic disease. We will discuss the latest research into the developmental origins of hypertension and obesity with an emphasis on emerging interactions between leptin and the central renin-angiotensin system. References: Erkonen GE, Hermann GM, Miller RL, Thedens DL, Nopoulos PC, Wemmie JA, & Roghair RD. (2011) Neonatal leptin administration alters regional brain volumes and blocks neonatal growth restriction-induced behavioral and cardiovascular dysfunction in male mice. *Pediatric Research* 69: 406-12. Hermann GM, Dallas LM, Haskell SE, & Roghair RD (2010) Neonatal macrosomia is an independent risk factor for adult metabolic syndrome. *Neonatology* 98: 238-44.

18.4 EPIGENETIC INFLUENCES IN DEVELOPMENTAL COMPARATIVE ANATOMY AND PHYSIOLOGY

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Epigenetics has long been of interest to the medical field because of its influence in human health (e.g. the epigenetics of cancer), but recently the potentially profound impact of epigenetics on comparative physiology and morphology is making itself known, particularly in the epigenetic inheritance of molecular, cellular, and physiological phenotypes that may be either adaptive or maladaptive. Such epigenetic inheritance can take two general forms. Context-dependent epigenetic inheritance occurs as a result of direct exposure of the developing animal to a stressor (e.g. hypoxia, hypothermia), and may persist across multiple generations as long as the stressor persists. This contrasts with germ-line dependent inheritance, which occurs as a result of the experiences of the germline - the eggs and sperm and even the stem cells that produce them, potentially persisting across generations even in the absence of the original causative stressor. For those epigenetically inherited traits that cross multiple generations, or even across multiple *F_i* broods produced by the *F₀* generation, the effect is not necessarily digital (off-on-off), but may more accurately be portrayed as analog, where the effect can wash out (or even wash in) across multiple generations or broods. This presentation discusses epigenetic inheritance of physiological and morphological characteristics, drawing from the very limited data sets currently available from the comparative physiological and morphological literature to show examples in both vertebrates and invertebrates of both context-dependent and germ-line dependent epigenetic inheritance.

19.0: COMPARATIVE GASTROINTESTINAL PHYSIOLOGY: FROM GENES TO ANIMAL PERFORMANCE

19.1 HOW THE GUT LIMITS NUTRITION, AND THE INFLUENCE OF THIS ON THE ECOLOGY AND EVOLUTION OF AN INSECT HERBIVORE, THE GRASSHOPPER

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Models of foraging and digestion are typically based on the assumption that animals need to maximize the uptake of nitrogen or energy, given costs. However, using chemically defined diets has shown growth, development and reproduction are maximized when animals are able to obtain an *optimal blend* of nutrients, rather than by maximizing the intake of any one or all of these. For insect herbivores, the macronutrients protein and carbohydrate, most influence life history and fitness. While the specific blend of nutrients for a herbivore can be ascertained in the laboratory, this information cannot simply be transferred to natural foods by grinding up plant tissues and measuring their chemical composition. The ability of an insect herbivore that chews plant leaves to absorb nutrients depends on how these nutrients have been packaged within the plant, interacting with the insect's food processing tools (typically the mandibles and gastrointestinal tract) and plasticity of its behavioural and physiological responses. We present evidence from a model insect herbivore, the grasshopper that demonstrates the role the gastrointestinal tract (GIT) exerts over nutrient uptake through controlling gut emptying and intermeal intervals, and the degree to which plasticity of the GIT can improve both the rate and ratio of nutrients absorbed.

Evidence suggests nitrogen may be limiting, not through being in short supply, but rather its excess relative to carbohydrate and the control it exerts over nutrient uptake. We discuss the potential ecological and evolutionary consequences of how the GIT functions. Reference: Clissold FJ, Tedder BJ, Conigrave AD, Simpson SJ. 2010. The gastrointestinal tract as a nutrient-balancing organ. *Proc B* 277:1751-9.

19.2

EVOLUTIONARY AND MOLECULAR MECHANISMS UNDERLYING INTESTINAL FLEXIBILITY FOR SNAKES

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Snakes possess an adaptive interplay between feeding ecology and digestive physiology. Frequently feeding snakes modestly regulate intestinal performance whereas infrequently feeding species up and down regulate intestinal form and function with each meal. The selective benefit of down regulation for infrequent feeders is a reduction in energy expenditure while fasting, whereas maintaining intestinal readiness is more beneficial for frequent feeders. Cellular mechanisms underlying modest and wide regulation of intestinal function reside in the modulation of microvillus length and hence brushborder surface area. Frequently feeding snakes experience no post-prandial change in microvillus length and no change in function. Wide regulation of intestinal function for infrequently feeders is explained by a 5-fold modulation of microvillus length. These modes of intestinal regulation stem from distinct gene expression regimes with greater expression changes predicted for infrequently feeding snakes. Infrequently feeding Burmese pythons experience massive shifts in expression of regulatory, trafficking, structural, and transporter-associated genes that direct intestinal flexibility. The link between snake feeding habits and digestive physiology is founded in the integrated evolution of cellular and molecular programs that underlie adaptive phenotypic responses (NSF IOB0466139). Secor, SM 2005 Evolutionary and cellular mechanisms regulating intestinal performance of amphibians and reptiles. *Integr Comp Biol* 45:66-78. Castoe TA et al 2013 The Burmese python genome reveals the molecular basis for extreme adaptation in snakes. *PNAS* 110:20645-20650.

19.3

THE ROLE OF GUT MICROFLORA IN THE NUTRITION OF MARINE HERBIVOROUS FISHES

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Hindgut microbes contribute to host energy supply by converting dietary carbohydrates to short-chain fatty acids, and can also supply amino acids in some animals. We examined three aspects of symbiotic protein supply to marine herbivorous fishes: (i) gut histology and ultrastructure; (ii) nitrogen metabolism of gut microbiota, principally nitrogen fixation; and (iii) stable isotope analysis of nitrogen uptake by fish from dietary algae and hindgut microbes. Fish species varied in gut structure and symbiont distribution. Algae were more degraded in species where bacteria attached to algal fragments. The dinitrogen reductase gene *nifH* was amplified from hindgut samples of all fishes surveyed, and expression was demonstrated by RT-Q-PCR. Nitrogen fixation was detected by acetylene reduction and ¹⁵N₂ labelling methods; the first demonstration of this in vertebrates. Rates of fixation in some fish species compared to those in higher termites. Compound Specific Stable Isotope Analysis showed variation in the proportions of amino acids contributed by algae and microbes, but microbes contributed >70% δ¹⁵N in some cases. Our results show that hindgut microbes contribute amino acids to fish tissues, and some microbial amino acids incorporated by fish include nitrogen fixed in the gut. Support: Marsden Fund UOA0908. References: Clements KD, Angert ER, Montgomery WL, Choat JH (2014) Intestinal microbiota in fishes: what's known, and what's not. *Mol Ecol* 23: 1891-1898.

19.4

AMYLASE GENETICS AND BIOCHEMISTRY UNDERLIE A DIGESTIVE SPECIALIZATION IN PRICKLEBACK FISHES

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Many herbivorous vertebrates have evolved elevated amylase activities in their guts, which usually arises via increased amylase gene copy number. I compared amylase gene sequences, amylase gene copy number, promoter region variation, and amylase biochemistry in prickleback fishes (family Stichaeidae) with different diets to better understand the underpinnings of dietary amylase activity variation. The herbivorous *Cebidichthys violaceus* expresses at least two amylase isoforms, and has 14 copies of amylase in its genome with two different promoter regions associated with each isoform. The herbivorous *Xiphister mucosus*, as well as four other omnivorous or carnivorous pricklebacks, express one amylase isoform, and have four to six copies of amylase in their genomes with a single promoter region. Thus, elevated amylase activity in *X. mucosus* is largely explained by elevated expression of one gene with relatively low copy number, a novel result for vertebrate amylases. However, despite their

genetic differences, and independent evolution of herbivory, assays on intestinal homogenates suggest that *C. violaceus* and *X. mucosus* digest different starches with similar efficiencies. These data have implications for the digestion of algal starches, as green algae use amylose and red algae use amylopectin as their storage polysaccharides, respectively. Therefore, convergent evolution of elevated amylase activity in *C. violaceus* and *X. mucosus* has different genetic underpinnings, but potential functional consequences remain to be revealed. UCI Startup Funds.

20.0: RESPONSES TO GLOBAL CHANGE: ACCLIMATIZE, ADAPT OR DIE

20.1

TRAIT-BASED APPROACHES TO PREDICTING THE RESPONSES OF SPECIES TO GLOBAL CHANGE

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Changes in climate and land-use are occurring rapidly, and at a global scale. Because millions of species are impacted, we cannot develop predictions for how each individual species might respond. Rather, we need a predictive framework that reduces the dimensionality of this task by identifying key characteristics of those taxa and regions that are most at risk. Here, we explore two such trait-based approaches to predicting responses. First, we focus on the predictive ability of physiological tolerance of high temperatures in ants. Globally, we find that ants inhabiting lower latitudes tend to be at the greatest risk under climate change owing to environmental temperatures being close to their thermal limits. Similarly, we find that among two large-scale experimental warming arrays, positioned at the northern and southern boundaries of temperate hardwood forests in eastern North America, ant thermal tolerance was strongly predictive of ant density at the low latitude site where temperatures routinely exceed ant thermal limits, but not the high latitude site where temperatures remain below ant thermal limits. Second, we focus on the predictive ability of species' resource-use and demographic traits in context of butterfly phenological responses to urbanization-based changes in land-use against a naturally occurring geographic temperature gradient. We find that opportunistic species avoid the phenological delays characteristic of many species inhabiting highly urbanized and geographically warm areas. These case studies suggest trait-based approaches may be useful for generalizing responses to other systems.

20.2

OCEAN ACIDIFICATION EFFECTS ON TEMPERATE ROCKFISHES

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The North American west coast provides a natural experimental system for testing the effects of ocean acidification on marine organisms. Congeners with varying exposures to acidic waters during spring upwelling may have different physiological tolerances. Rockfish (*Sebastes* spp.) provide an excellent comparative study system comprising >100 species with diverse life history traits and niches. We compared physiological and gene expression differences between juvenile winter- and spring-spawning rockfish, blue (*Sebastes mystinus*) and copper rockfish (*S. caurinus*), respectively, after chronic exposure to four pCO₂ treatments. Both species exhibited decreased critical swimming speeds and copper rockfish also had reduced aerobic scopes at the highest pCO₂ treatment. RNAseq transcriptome analyses of muscle tissue from the same individuals showed that the species exhibit divergent gene regulation strategies for coping with high pCO₂. Among pCO₂ treatments, each species had 100s of differentially expressed genes, but fewer than 20 were common to both species. Transcriptome profiles of physiologically tolerant rockfish may help explain the mechanistic basis underlying their increased ability to compensate against the effects of elevated pCO₂. Our study highlights the need for integrative comparative studies for assessing the adaptive capacity of fishes in responding to ocean acidification. Hofmann GE, Todgham AE (2010). Living in the now: physiological mechanisms to tolerate a rapidly changing environment. *Annu. Rev. Physiol.*, 72, 127-145.

20.3

MECHANISTIC OVERLAP BETWEEN PLASTIC AND EVOLVED RESPONSES TO HEAT STRESS

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Organisms and populations may respond to changing environments through plasticity or evolutionary adaptation. While these have historically been viewed as separate processes, the genetic assimilation hypothesis holds that environmentally induced (plastic) phenotypes are often the first step in adaptation to environmental change, and later become genetically "assimilated," such that the original environmental stimulus is no longer required to produce the phenotype. We tested this hypothesis by com-

binning artificial selection and heat shock experiments with RNA sequencing of the crustacean *Tigriopus californicus*. Transcriptomic responses to heat shock and artificial selection suggest substantial mechanistic overlap between plastic and evolved response to heat stress, and provide some of the first experimental support for the genetic assimilation hypothesis. To the extent that adaptation to changing environments tends to occur through genetic assimilation, our results suggest that plasticity and evolution may provide overlapping, rather than additive benefits to species responding to global change.

20.4

COMPARING PHYSIOLOGICAL PLASTICITY VS EVOLUTIONARY ADAPTATION VS PHYLOGENETIC CONSTRAINT ON SPECIES DISTRIBUTIONS; *DROSOPHILA* AND BEYOND

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Insects including *Drosophila* species have proven to be good experimental subjects for comparing the relative importance of plastic changes versus inherent genetic variation in physiological adaptation because the experimental environment can be tightly controlled across generations. Here we report on a unique comparison of *Drosophila* species from eastern Australia and beyond. Variation in physiological traits was closely tied to species distributions; in particular, responses to extreme conditions of cold, aridity and heat (under dry conditions) correlated closely with the ecological niche of the species. Species had similar levels of plasticity to thermal extremes but differed markedly in resistance levels. There was little variation in species performance across constant culture temperatures, highlighting the importance of genetically-based resistance to extremes in dictating distributions. There was strong phylogenetic signal in responses to extremes likely to be tied to genome differences including multi-copy gene families. Mid-latitude species/populations are more vulnerable to climate change than those at latitudinal extremes. Comparative genomics is providing an unprecedented picture of the genetic architecture of species confined to particular climatic zones. ARC 120100916 and ARC FL100100066. Overgaard, J., Keamey, M.R., & Hoffmann, A.A. 2014. *Glob. Change Biol.* **20**, 1738. Kellermann, V., Overgaard, J., Loeschcke, V., Kristensen, T.N., & Hoffmann, A.A. (2013). *PLoS One* **8**, e72072.

21.0: EVOLUTIONARY AND DEVELOPMENTAL ORIGINS OF ENDOTHERMY

21.1

THERMOREGULATION AND THERMOGENESIS IN REPTILES

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A standard assumption of reptilian metabolism and thermoregulation has centred on their strong reliance on behavioural control over body temperature, due in part to their dependence on the environment for body heat (e.g. ectothermy). Numerous large reptiles, however, are known to exhibit thermal inertia sufficient to bestow prolonged elevation of body temperature above ambient. In some cases, homeothermy is made possible via constitutive muscle recruitment. More recently, however, we have discovered that tegu lizards exhibit a form of reproductive endothermy, with elevated nightly body temperatures up to 6°C higher than ambient temperatures. This reproductive endothermy is seasonal, and cannot be explained entirely by thermal inertia since tegus brought indoors for 7 days continue to exhibit body temperatures up to 3°C above ambient temperatures. To sustain these elevated body temperatures, tegus show seasonal changes in metabolism, as reflected in sustained increases in heart rate throughout the day. The origins of this increased metabolism is unknown and may simply reflect an overall increase in metabolism that accompanies reproductive changes in both sexes. Although female lizards show a slightly greater temperature differential, males also exhibit this response, suggesting a global rise in metabolism throughout the body rather than a phenomenon related to oogenesis. The fact that tegu lizards as small as 2 kg in size, similar in size to the earliest ancestral mammals, can exhibit a prolonged endothermic response may provide fuel for theories attempting to explain the context for the evolution of endothermy.

21.2

DEVELOPMENT OF ENDOTHERMY IN MARSUPIAL AND PLACENTAL MAMMALS

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In contrast to eutherians the newborn marsupial joey is ectothermic, relying entirely on the thermally stable pouch of its mother for thermal homeostasis. The development of endothermy occurs as a continuum of events rather than a step-wise change with the transition beginning, depending on the species, usually by halfway through pouch life. The transition zone between ectothermy and endothermy can be clearly defined by examining the resting mass-specific rate of oxygen consumption at pouch body temperature (Tb), which gradually changes from values close to the predicted metabolic rate for an adult ectotherm (e.g., lizard) of the same mass to values pre-

dicted for an adult marsupial. A myriad of morphological, physiological, and homeostatic changes that allow for endothermy and the maintenance of independent Tb occurs across this time period; these are discussed.

21.3

TESTING COMPETING HYPOTHESES OF THE EVOLUTION OF ENDOTHERMY

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Hypotheses of the evolution of endothermy posit that it evolved concurrently with high levels of basal metabolic rate (BMR) as a correlated response to either selection for high rates of maximum aerobic metabolism (MMR), or an increased capacity for elevated, sustained metabolic rates associated with parental care (SusMR). I review studies aimed at testing predictions of these hypotheses, i.e. an inexorable link between (i) BMR and MMR, or (ii) BMR and SusMR, as well as their underlying physiological mechanisms. There is mounting evidence of ample genetic variation in BMR and MMR, a pre-requisite for their concerted evolution. On the other hand, failure to demonstrate a positive association between BMR and SusMR mostly stems from the mismatch in the timing of variation in BMR and SusMR, rather than from lack of a functional link between the two. I discuss the discrepancies between conclusions drawn from intra- and inter-specific studies on the significant factors affecting variation in traits underlying BMR, MMR and SusMR, such as the contribution of skeletal muscles and the fatty acid composition of cell membranes. Inconsistencies in the results of studies testing the hypotheses of the evolution of endothermy can be resolved by (i) artificial selection experiments emulating mechanisms of evolution of metabolic rates and correlated traits, and (ii) focused comparative studies on animals from slow-fast life history continuum represented by species from tropical and temperate geographical zones. Reference: Konarzewski, M. Książek, A. 2013. Determinants of intra-specific variation in basal metabolic rate. *J. Comp Physiol. B*, **83**, 27-41.

21.4

DEVELOPMENT OF ENDOTHERMY IN ALTRICIAL AND PRECOCIAL BIRDS

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All birds begin life with an ectothermic phenotype and develop endothermy some point after hatching. Precocial species attain an endothermic phenotype rapidly upon hatching which requires high metabolic rate, thermal insulation, and an internal thermostat to stimulate heat production upon cooling. An ectothermic phenotype persists in altricial species for an extended period. To better understand development of endothermy in a number of species, we characterized changes in aerobic capacity, from mitochondria to whole animal, along with concomitant changes in cardio-respiratory development and thyroid hormone level. A common pattern in precocial species is a rapid increase in skeletal muscle mitochondrial oxidative respiration associated with an endothermic metabolic response to cooling immediately after hatching. Altricial species show greater variability in the timing of peak skeletal mitochondrial respiration compared with their endothermic metabolic response. Thyroid hormone levels rise rapidly upon hatching in precocial species and gradually in altricial species. Manipulation of thyroid hormones in altricial species influences development of resting metabolism, organ growth, insulation, and timing of obtaining endothermy. Future focus of research into development of endothermy should include development of muscle metabolism and cardio-respiratory function and their association with hormonal regulation of endothermic metamorphosis. Supported by NSF Grant IOS-1146758.

22.0: DETERMINANTS OF SKELETAL MUSCLE DIVERSITY

22.1

EVOLUTIONARY SELECTION OF MYOFIBRILLAR PROTEIN ISOFORMS FOR SPECIFIC MUSCLE FUNCTIONS

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Understanding the role of myofibrillar protein isoforms in the regulation of muscle contractile properties has greatly advanced over the past three decades. This is due, in large part, to the ability to study contractile properties of single muscle fibers and the determination of their myofibrillar protein isoform composition. This approach has been applied to multiple vertebrate species with natural variations in isoform expression patterns and to transgenic models in which the expression of a specific isoform was altered. The results from many laboratories have led to several well-accepted generalizations, including the pivotal role of myosin heavy chains in determining shortening velocity and power generation, the fundamental role of myosin light chains in modulating the same properties, and the key roles of isoforms of tropomyosin and of troponin subunits in regulating calcium activation of contraction. With this solid knowledge base, we can now ask which specific patterns of contractile

protein isoform expression have been selected to serve the incredibly diverse range of motor functions in different muscles, especially among vertebrate craniofacial muscles, where isoform expression patterns are highly divergent among different species. Addressing this will provide an understanding of how myofibrillar protein isoforms contribute to the fascinating and elaborate comparative physiology of muscle contraction. I will provide illustrations of specific patterns of contractile protein isoform expression and their association with contractile properties across a broad range of vertebrate motor functions.

22.2 COMPARATIVE PHYSIOLOGY OF BODY WEIGHT-SENSITIVE SKELETAL MUSCLE PLASTICITY

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Efficient animal locomotion requires skeletal muscle function to be finely tuned to variation in body weight. We know very little about how animals sense their weight quantitatively nor do we know how such fine tuning occurs exactly. The main focus of research addressing such questions has been on skeletal muscle mass plasticity with less attention being paid to plasticity and body weight-dependency of skeletal muscle properties that occurs in parallel. Here I will present a growing body of work that demonstrates the existence of an evolutionarily conserved body weight sensing mechanism that controls plasticity in skeletal muscle properties at the level of sarcomere gene expression patterns. In addition, I will discuss how dietary quality and disease can affect this mechanism in both insect and mammalian study systems.

22.3 CARDIAC MYOSIN ALPHA AND VENTRICULAR HYPERTROPHY PROTECT GROUND SQUIRRELS IN HIBERNATION.

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As deep hibernators, golden-mantled ground squirrels (*Callospermophilus lateralis*) have multiple challenges to cardiac function during torpor and subsequent arousals. At dramatically slower heart rates, chamber dilation and reduced cardiac output could lead to congestive myopathy. We performed echocardiography on squirrels before and after hibernation. The left ventricle hypertrophied 30% during hibernation. Left atrial ejection generally was absent during hibernation, but returned weakly with arousals. We analyzed cardiac myosin heavy-chain (MyHC) isoforms in squirrels before and during hibernation, and prior to emergence. Relative abundance of cardiac MyHC alpha increased during hibernation, at hibernacula temperatures of 20 and 4 °C. An increase in contractile speed and power from MyHC alpha may aid force generation at low temperature and low heart rates. Unlike cardiomyopathies where MyHC alpha is replaced by MyHC beta to reduce oxygen consumption, squirrels demonstrate a cardioprotective mechanism to maintain cardiac output during torpor. Squirrels thus prevent cardiac dilation and reduced cardiac output at body temperatures and heart rates far below what non-hibernators can tolerate. (NIH MBRS S06 GM063119 (BCR) Reference: Nelson, O.L. and Rourke, B.C. 2013 Increase in cardiac myosin heavy-chain (MyHC) alpha protein isoform in hibernating ground squirrels, with echocardiographic visualization of ventricular wall hypertrophy and prolonged contraction. *J Exp Biol* 216, 4678-4690.

22.4 THE DIVERSITY AND EVOLUTION OF LOCOMOTOR MUSCLE PROPERTIES IN ANURANS

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Anuran jumping is a model system for linking muscle physiology to organismal performance. Additionally, anuran species display substantial diversity in their locomotion and morphology, reflecting their habitats (including aquatic, terrestrial, arboreal, and fossorial environments) as well as other anatomical or life history traits (such as protective toxins). Some anurans are renowned for performing powerful leaps from riverbanks or tree branches, but other species move predominantly via burrowing, swimming, short hops, or even diagonal-sequence gaits. Many anurans with similar locomotion and morphology are actually convergent, (e.g. multiple independent evolutions of "tree frogs"), while closely related species may differ drastically, as with the bullfrog-like river toad compared to other Bufonid toads. These multiple independent evolutionary changes in locomotion allow us to test the hypothesis that evolutionary increases in locomotor performance will be linked to the evolution of faster, high-power muscles. We tested the jumping, swimming and running performance of fourteen species of anurans and one salamander, followed by measurement of the contractile properties of the semimembranosus and plantaris muscles and anatomical measurements, then analyzed these data using both traditional statistics and phylogenetic independent contrasts. Contrary to our hypothesis, we found that locomotor performance showed little correlation to muscle properties, but was tightly linked to anatomical properties. This suggests that locomotor per-

formance is more dependent upon anatomy than upon muscle contractile properties, and the evolution of increased locomotor performance is not linked to the evolution of faster, higher-power muscles.

23.0: CARDIOVASCULAR AND RESPIRATORY PHYSIOLOGY

23.1

DO *DROSOPHILA* LARVAE EXPERIENCE FUNCTIONAL OXYGEN LIMITATION LATE IN THE INSTAR?

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Arthropods gain considerable mass between molts, increasing oxygen demand with some aspects of the respiratory system fixed in size until the next molt. We hypothesized that internal hypoxia could be one of several cues that could trigger molting in insects and other arthropods. In support of this hypothesis, hypoxic rearing (10 kPa) triggered an ecdysone peak and molting to adulthood at a smaller body size in *Drosophila* (JEB 216:4334). Here we tested for evidence that late-third instar *Drosophila* larvae show signs of functional oxygen-limitation. Lactate levels did not increase later in the third instar, and were unaffected by rearing in hyperoxia. However late-instar larvae had stronger capacities to generate lactate in response anoxia, suggesting past selection for coping with late-instar oxygen limitation. Metabolic rates rose linearly with mass early in the instar, but showed no further increase with mass later in the instar. Hyperoxia (30 kPa) increased late-third instar larval metabolic rates, though hypoxia (10 kPa) did not lower them. Together these data support the conclusions that *Drosophila*, and likely other insects, can experience functional oxygen limitations later in the instar, that internal hypoxia is one of several factors capable of inducing molting, and that molting occurs before internal P_{O_2} decreases sufficiently to trigger anaerobiosis. This research was supported by NSF IOS 1256745.

23.2

A STUDY ON EMBRYONIC DEVELOPMENT RATE AND CARDIO-RESPIRATORY PERFORMANCE OF THE NORWAY LOBSTER, DURING ACUTE EXPOSURE TO ELEVATED PCO₂, MANGANESE AND HYPOXIA

Hannah Styf¹, Helen Nilsson Sköld¹, Hannah L Wood¹, Anna-Sara Krång¹, and Susanne Eriksson¹

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As a consequence of the anthropogenic carbon dioxide (CO₂) emissions since mid-18th century, our oceans have gradually become more acidic, a process named ocean acidification (OA). Aquatic organisms experience a multitude of simultaneous environmental factors and combined stressors have the potential to be synergistic. Thus, experiments should combine OA with other ecologically relevant stressors, in order to fully understand future impact on species, populations and ecosystems. The marine decapod crustacean *Nephrops norvegicus* is of immense importance to both ecology and economy. It lives in sediment burrows and is seasonally exposed to periods of oxygen deficiency (hypoxia, <30% O₂ saturation), as thermoclines build up and oxygen is consumed. These events are exacerbated by anthropogenic eutrophication and are predicted to increase both spatially and temporally, as well as in severity, due to thermal elevation caused by climate change. Hypoxia causes reduction of MnSO₄ in sediments and release manganese into the overlying water, where it becomes bio-available to benthic organisms. In this study, berried *Nephrops norvegicus* were exposed to ocean acidification (1600 ppm) for a period of seven weeks and an additional stressor the final week, i.e. hypoxia (~25% O₂) or manganese (8 mg/l). The aim was to investigate development rate (yolk consumption), cardiorespiratory performance (heart rate and rate of oxygen consumption) as well as wet/dry weight during embryonic development.

23.3

REPEATABILITY AND MORPHOLOGICAL CORRELATES OF FISH BEHAVIOR IN HYPOXIA

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The habitat of the gulf killifish, *Fundulus grandis*, is characterized by variable oxygen levels. In this study, we evaluated the repeatability and morphological correlates of two behaviors of fish in hypoxia: aquatic surface respiration (ASR), where fish ventilate their gills with oxygen-rich surface waters, and loss of equilibrium (LOE), a non-lethal measure of tolerance to extreme hypoxia. We exposed fish to gradually lowered oxygen levels and recorded the time and oxygen levels when fish conducted ASR and LOE. We also measured total filament length, total filament number, and average filament length of gills from these fish. Results from 14 fish used in 2 to 4 trials, each spaced by approximately 2 weeks, show that the time when fish conduct ASR and the oxygen levels at ASR, are significantly dependent upon the individual ($P < 0.05$) and have a repeatability of about 50%. The time to LOE approached statistical significance ($P = 0.09$) with a repeatability of 22%. Mass-adjusted average filament length was significantly correlated with time at ASR ($r = 0.53$, $P < 0.05$) and showed

weak negative relationship with the oxygen levels at ASR ($r = -0.44$, $P = 0.11$), suggesting that fish with longer gill filaments do not conduct ASR until oxygen levels reach lower values. Thus, fish behaviors during hypoxia, ASR in particular, vary in a repeatable fashion among fish, which may be explained, in part, by intraspecific variation in gill morphology. Funded by NSF (DBI: 1040996).

23.4 EFFECTS OF HYPERCAPNIA ON GILL VENTILATION, STANDARD METABOLIC RATE, OXYGEN SUPPLY CAPACITY AND SWIMMING PERFORMANCE IN RED DRUM (*SCIAENOPS OCELLATUS*)

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Oceanic CO₂ levels are predicted to reach 1000 µatm by the end of this century. In fish, exposure to elevated water CO₂ levels (hypercapnia) reduces the CO₂ gradient over the gills, causing CO₂ accumulation in the blood and respiratory acidosis. During metabolic acidosis fish partially restore blood pH by increasing CO₂ excretion via hyperventilation. A hyperventilatory response is also seen during hypercapnia exposure and is assumed to alleviate acid-base disturbances; however, this likely comes at an osmoregulatory cost owing to the osmoregulatory compromise of fish. As such, hypercapnia induced hyperventilation could potentially carry energetic trade-offs tied to an increase in cost of osmoregulation. We show that gill ventilation in red drum (*Sciaenops ocellatus*) increases after acute exposure to 1000 and 4000 µatm hypercapnia, respectively. Standard metabolic rate, however, was unchanged indicating that the hyperventilatory response to ocean acidification places, at most, only a minor energetic load on this species. Furthermore, we examine the effects of acute and chronic hypercapnia exposure on critical oxygen tension, aerobic scope, critical swimming speed and cost of transport. Our results show that ocean acidification has little effect on swim performance or oxygen supply capacity. All experiments were conducted in accordance with the University of Texas at Austin Institutional Animal Care and Use Committee. This work was funded by NSF (EF 1315290).

23.5 OPTIMIZATION OF *IN VITRO* INCUBATION OF GILLS FROM *FUNDULUS GRANDIS*

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Low dissolved oxygen, hypoxia, is a prevalent feature of many aquatic habitats, and fish that occur in these habitats employ a suite of behavioral, morphological, and physiological mechanisms to deal with low oxygen. As the site of gas exchange, fish gills are directly subject to variation in ambient oxygen levels. The purpose of this study was to optimize *in vitro* culture conditions for intact gills from the Gulf killifish, *Fundulus grandis*, with the ultimate goal of using these preparations to study the cellular and molecular responses of this tissue to hypoxia. Gills were dissected and incubated in two media, using two apparatus, and assayed for viability by trypan blue staining and lactate dehydrogenase (LDH) leakage at 2 and 24 h of incubation. Trypan blue staining was uniformly low (< 2%) and did not differ between incubation media, apparatus, or incubation time. The percentage of gill LDH that leaked into the medium (about 5-20%) was consistently higher than trypan blue staining and significantly influenced by incubation medium: lower LDH leakage occurred when gills were incubated in sterile one-third strength sea water than when incubated in physiological saline. Although there were trends toward higher trypan blue staining and LDH leakage at 24 h incubation compared to 2 h, neither was significant, partly due to high variability in both measures of tissue viability. Current work continues to optimize gill culture conditions. Funded by the Louisiana Board of Regents.

23.6 PURIFICATION AND CHARACTERIZATION OF ANTIBODIES AGAINST KILLIFISH HIF-1A

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The hypoxia-inducible factor (HIF) is a transcription factor that regulates gene expression in animals exposed to low oxygen concentrations, or hypoxia. Previously, we generated polyclonal antibodies in chickens against HIF-1α from the killifish *Fundulus heteroclitus*. The objectives of the current project were to clone, express, and purify a fragment of HIF-1α and use it to affinity purify these antibodies. A 300 bp region of *F. heteroclitus* HIF-1α cDNA was cloned and used to express a recombinant HIF-1α protein fragment of approximately 13 kDa. The protein fragment was purified by affinity, ion-exchange, and size-exclusion chromatography. The purified protein was coupled to AminoLink Plus resin and used to affinity purify the chicken anti-killifish HIF-1α antibodies. In western blots, the affinity purified antibodies specifically recognized full length killifish HIF-1α (ca. 95 kDa) made by *in vitro* transcription and translation (IVTT). Next, we used these antibodies to immunoprecipitate IVTT HIF-1α, optimizing the buffer composition; amounts of primary antibody and the immunoprecipitating resin; incubation times; and the recovery of input HIF-1α.

The next phase of this project is to immunoprecipitate and measure endogenous HIF-1α from multiple tissues harvested from killifish exposed to various durations and levels of hypoxia. Funded by the Louisiana Board of Regents and the National Science Foundation (DBI: 1040996).

23.7 ACCLIMATION TO OVERNIGHT HYPOXIA AND INCREASED TEMPERATURE IMPROVE AEROBIC PERFORMANCE IN SALMON (*SALMO SALAR*) AND CHARR (*SALVELINUS ALPINUS*)

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Fish are increasingly influenced by rising temperatures and decreasing oxygen levels in aquatic ecosystems. However, the capacity of fish to tolerate the combination of these stressors is not well understood. The purpose of the current study was to analyze the effects of warming and overnight hypoxia on capacity of fish to tolerate hypoxia. One-year old salmon and charr were acclimated to cold (8 °C) and warm (14 °C) temperatures in either normoxic conditions or with an overnight hypoxia for one month, after which their heart tissue was collected and hypoxia tolerance measured. Both warm acclimation and overnight hypoxia more than doubled the hypoxia tolerance of salmon and also increased the tolerance of charr by 25%. Charr were significantly more tolerant to hypoxia than salmon. Charr also had larger hearts and thicker compact layers than salmon. The relative ventricle mass was higher in cold acclimated fish, but the thickness of the compact layer of the ventricle increased with both warm and hypoxia acclimation. This may contribute to improved hypoxia tolerance. The results show significant species differences in the tolerance of multiple environmental stressors in salmonid fish. The experiments were done following APS guiding principles for the care and use of animals (animal testing permit nr. ESAVI/4068/04.10.07/2013). The study was funded by Kone Foundation, BIOINT Doctoral program and the Academy of Finland.

23.8 LONG-TERM ACCLIMATION TO HYPOXIA DOES NOT CONFER CROSS-TOLERANCE TO HIGH TEMPERATURE IN STEELHEAD TROUT (*ONCORHYNCHUS MYKISS*)

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It has been suggested that acclimation to high temperatures or hypoxia confers tolerance against the other oxygen-limited stressor. Thus, we investigated if acclimation to hypoxia (>3 months at 40% air sat.) improved the trout's critical thermal maximum (CTM), or affected primary physiological variables that determine upper thermal tolerance [oxygen consumption rate (\dot{M}_{O_2}), hematocrit (Hct), and cardiovascular function and morphology]. \dot{M}_{O_2} (resting and max.), CTM (24.7 vs. 25.3°C), and Hct did not differ between hypoxic- and normoxic-acclimated trout when measured in normoxia. However, cardiac output (\dot{Q}) plateaued in hypoxic trout >20°C, despite similar increases in heart rate as compared to normoxic fish, and this resulted in a lower \dot{Q}_{max} . This limited pumping capacity was not associated with changes in cardiac morphology or *in vitro* maximum stroke volume, suggesting that alterations in ventricular filling dynamics or myocardial contractility constrain cardiac function in hypoxic-acclimated fish at high temperatures. Our finding that hypoxic-acclimated trout consumed more oxygen for a given increase in \dot{Q} is consistent with data on Atlantic cod, and suggests that long-term hypoxia improves tissue oxygen extraction or utilisation, and that this compensates for diminished heart performance. In summary, our data do not support the concept of 'cross-tolerance' with respect to these two environmental variables, but offer additional insights into fish physiological plasticity.

23.9 HYPOXIA-INDUCED APOPTOSIS IN THE HEARTS OF HYPOXIA-TOLERANT TILAPIA (*OREOCHROMIS HYBRID* SP.) AND HYPOXIA-SENSITIVE STRIPED BASS (*MORONE SAXATILIS*)

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Oxygen deprivation (hypoxia) is a common stressor affecting cardiomyocytes when blood flow is reduced or cut off from the heart. In response, cardiomyocytes have been shown to undergo apoptosis which is suggested to have a role in the pathogenesis of cardiovascular diseases, the leading cause of mortality in developed nations. Since many species of fish can survive low oxygen levels that would be fatal to mammals, fish are an ideal model system to study changes at the cellular and molecular level that prevent or repair hypoxia-induced damage in cardiomyocytes. Evidence of apoptosis at the cellular level (caspase-3/7 activity) will be measured in a hypoxia-sensitive species, striped bass (*Morone saxatilis*) and a hypoxia-tolerant species, tilapia (*Oreochromis hybrid* sp.) at four key time points: 1) prior to hypoxia exposure (normoxic control), 2) after four hours at the species' specific P_{crit} (hypoxia), 3) immediately upon return to normoxia (reperfusion), and 4) after four hours at normoxia (recovery). Additionally, RT-qPCR will be used to quantify changes in the tran-

scriptome of cardiomyocytes of pro-apoptotic (*BAX* and *FAS*), anti-apoptotic (*BCL2* and *FASL*) and repair (*Hsp70*) genes. We hypothesize pro-apoptotic genes will have higher gene expression and caspase activity will be higher in striped bass while anti-apoptotic and repair genes will have higher expression in tilapia. Funding was provided by Georgia Southern University via a GSO grant (AR) and FRC seed funding (JML).

23.10

ACUTE THERMAL CHALLENGES OF CARDIAC FUNCTION IN PACIFIC BLUEFIN TUNA

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Understanding the physiology of vertebrate thermal tolerance is critical for predicting how animals respond to climate change. Pacific bluefin tuna experience a wide range of ambient temperatures and occupy the largest geographic niche of all tunas. Their capacity to endure thermal change is due in part to cardiac specializations that improve performance across a range of temperatures. To better understand the cellular mechanisms that enable bluefin hearts to maintain function across thermal gradients we combined confocal microscopy and electrophysiology to define interactions between temperature, adrenergic stimulation and contraction frequency on the electrical activity and Ca^{2+} dynamics of Pacific bluefin ventricular myocytes. We demonstrate that acute cooling and acute warming modulate the excitability of the cardiomyocyte by altering ion flux during the action potential. When we included sympathetic stimulation (500 nM adrenaline) we found that compensatory changes in the action potential and ion influx resulted in fairly constant Ca^{2+} cycling across a 20 °C acute temperature gradient. The results indicate the tuna heart maintains consistent contraction and relaxation cycles during acute temperature changes. We hypothesize that the capacity to operate across larger thermal gradients plays a key role in the bluefins capacity for broad thermal niche utilization. Study supported by NOAA, Monterey Bay Aquarium Foundation and the University of Manchester.

23.11

TEMPERATURE DEPENDENCE OF BLOOD-OXYGENATION IN JUVENILE SANDBAR SHARKS (*CARCHARHINUS PLUMBEUS*)

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Juvenile sandbar sharks (*Carcharhinus plumbeus*) use shallow inshore areas on the east coast of the United States (e.g., Chesapeake Bay) as summer nursery grounds before emigrating to southern coastal waters or into deep adjacent waters as adults, and thus experience a great range of environmental temperatures. In the nursery grounds water temperatures typically range from 20-26°C, but sharks are known to experience water temperatures between 15-30°C throughout their lives. Sandbar sharks are ectothermic and thus their blood temperature equilibrates with that of the ambient water. Consequently, water temperature could have a profound influence on blood-oxygenation due to the generally exothermic nature of oxygen (O_2) binding to the heme groups of hemoglobin. We hypothesized that the temperature dependence of blood- O_2 binding would be minimal in juvenile sandbar sharks, permitting sufficient O_2 uptake in the warm waters of their nursery grounds. To test the effects of temperature changes on blood- O_2 binding in the sandbar shark we constructed O_2 equilibrium curves (OECs) on whole blood at a range of temperatures that juvenile sandbar sharks typically encounter throughout the year. Representative OECs and blood O_2 tensions at 50% Hb- O_2 saturation (P_{50}) are reported for two carbon dioxide tensions over the experimental temperature range, and interpreted with respect to the thermal niche of juvenile sandbar sharks. Funding provided by NSERC.

23.12

EFFECT OF TEMPERATURE ACCLIMATION ON HEMOGLOBIN-OXYGEN BINDING PROPERTIES IN PACIFIC BLUEFIN TUNA (*THUNNUS ORIENTALIS*) AND YELLOWFIN TUNA (*THUNNUS ALBACARES*)

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Current climate change trends may threaten the survival of many marine species, so understanding responses to increasing ocean temperatures and CO_2 is critical for predicting future impacts. This study investigates thermal acclimation effects on hemoglobin-oxygen (Hb- O_2) binding properties in Pacific bluefin tuna (*Thunnus orientalis*) and yellowfin tuna (*Thunnus albacares*) maintained in captive tanks at 17°, 20° and

24°C acclimation temperatures. Oxygen binding properties of acclimated hemoglobin were examined under experimental temperatures (15°-35°C) and CO_2 levels (0%, 0.5% and 1.5%). Bluefin tuna demonstrated a reverse temperature-dependent trend between 15°-30°C at each acclimation and all CO_2 levels. In contrast, yellowfin tuna produced a normal temperature-dependent effect at each acclimation at 0% CO_2 , but temperature-independent and reverse temperature-dependent effects at 0.5% and 1.5% CO_2 . Both species demonstrated a normal Bohr Effect. Thermal acclimation in bluefin tuna produced increased O_2 affinity at 17°C-acclimation, and a significantly steeper oxygen equilibrium curve slope at 24°C-acclimation compared to the other acclimations. These findings indicate that Pacific bluefin tuna possess a reverse temperature-dependent effect, consistent with other bluefin species, while yellowfin tuna utilize a mix of effects. While temperature acclimation produced changes in Hb- O_2 binding properties, more work is needed to clarify the functional significance of such changes.

23.13

AERIAL RESPIRATION IN POLYPTERIDS

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Polypterids are a basal family of actinopterygian fishes (ray-finned) that share similarities with extant sarcopterygians (lungfishes and tetrapods) including the ability to breathe air using paired ventral lungs. Two polypterid genera, *Erpetoichthys* and *Polypterus*, are able to completely satisfy O_2 demand breathing air, however, only *Erpetoichthys* is known to be amphibious. To investigate whether gas exchange capacity was the limiting constraint in the evolution of amphibious behavior, we measured O_2 consumption, CO_2 production, and respiratory partitioning between pulmonary and cutaneous gas exchange in *Erpetoichthys* and *Polypterus* over a three-hour period in air. Pulmonary gas exchange accounted for 80% and 57% of total VO_2 and VCO_2 in *Erpetoichthys* and 82% and 60% in *Polypterus*. Differences in respiratory partitioning of O_2 and CO_2 resulted in reduced respiratory exchange ratios (RER) at the lungs and elevated RERs at the skin in *Erpetoichthys* (0.6-lungs and 1.7-skin) and *Polypterus* (0.6-lungs and 1.9-skin). Other amphibious fishes when removed from water typically show low air-breathing organ RERs (<0.7) indicating an inability to fully excrete CO_2 into air with this organ. Our data show similar findings in *Polypterus* where pulmonary gas exchange participates in CO_2 excretion, yet cutaneous gas exchange continues to play a significant role. *Erpetoichthys*, on the other hand, shows an elevated ability for pulmonary CO_2 excretion. Regardless, the total RERs of both genera were 0.7-0.9 indicating that *Polypterus* did not differ in its overall aerial respiratory capacity to process CO_2 from its amphibious relative *Erpetoichthys*. Our data suggest that in polypterids, gas exchange ability was not the limiting constraint in the evolution of amphibious behavior. Instead, factors such as, lung ventilation mechanics, locomotive ability, and ecological factors may have been important influences.

23.14

THE EFFECTS OF *UMBELLULARIA CALIFORNICA* ESSENTIAL OIL ON THE CUTANEOUS VASCULATURE OF FROGS

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Most plant species produce chemical compounds called secondary metabolites that enhance fitness in a variety of ways. Many of these compounds are also physiologically active in vertebrates and have widespread medicinal uses. The most ubiquitous secondary metabolites are the terpenoids, many of which cause vasodilation of the aorta and mesenteric arteries. In this study, we examined the vasoactive effects of the essential oil of *Umbellularia californica*, which contains the terpenoid umbellulone. Oil obtained via steam distillation using aerial portions of *U. californica* was applied directly to cutaneous arterioles of frogs. Arteriole diameter was monitored both before and after oil application by video microscopy. Within seconds of application, the oil caused significant vasoconstriction that persisted until the oil was washed off. Our control, medical grade sesame oil, caused no observable effects when applied using the same protocols. These results are opposite to the vasodilatory effects of terpenoids on aortic rings and mesenteric arteries. This suggests that the vasoactive effects of umbellulone are different from other terpenoids, that the vasoactive effects of terpenoids differ depending on blood vessel type, or that application of the complete essential oil affects vasculature differently than application of the isolated terpenoid. This research was approved by SUU-IACUC and funded by a grant from SUU Undergraduate Research and Scholarship Program.

23.15

BAROREFLEX CHARACTERISTICS OF ANURAN AMPHIBIANS FROM DIFFERENT ENVIRONMENTS

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Anurans from terrestrial environments have an enhanced ability to maintain arterial blood pressure through lymph mobilization in response to desiccation or hemorrhage.

We compared baroreflex function in three species of anurans that span a range of environments, dehydration tolerance and an ability to maintain arterial blood pressure with dehydration and hemorrhage. The cardiac limb of the baroreflex loop was studied using pharmacological manipulation of blood pressure and the resulting changes in heart rate were quantitatively analyzed using a four-parameter sigmoidal logistic function. Resting mean arterial blood pressure (P_m) in the aquatic species, *Xenopus laevis*, was 3.6 ± 0.3 kPa and was less ($P < 0.005$) than the semiaquatic species, *Lithobates catesbeianus* (4.1 ± 0.2 kPa), or the terrestrial species, *Rhinella marina* (4.7 ± 0.2 kPa). The maximal baroreflex gain was not different among the three species and ranged from 12.1 to 14.3 beats min^{-1} kPa^{-1} and occurred at P_m ranging from 3.0 to 3.8 kPa, which were slightly below the resting P_m for each species. Resting P_m in the three species were near the upper saturation point of the baroreflex curve which would enable animals to respond primarily to hypotensive, rather than hypertensive, changes in blood pressure. This is consistent with the hypothesis that the baroreflex is a key sensory component that allows anurans to maintain arterial blood pressure by mobilization of lymphatic return in response to hypotension.

23.16

DOES THE RIGHT-TO-LEFT SHUNT AFFECT ASSIMILATION EFFICIENCY, DIGESTA TRANSIT, AND POSTPRANDIAL METABOLISM IN ALLIGATORS?

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Multiple hypotheses exist regarding the functional significance of the right-to-left ("pulmonary bypass") shunt utilized by non-avian reptiles. The unique cardiovascular arrangement of crocodilians, including the complete anatomic separation of right and left ventricles and presence of right and left aortae, provides a useful model for testing these hypotheses, as the shunt can be ablated by surgical occlusion of the left aorta. This study aims to clarify the role of the right-to-left shunt in digestion. The shunt is hypothesized to aid digestion by delivering hypercapnic blood to the oxyntic cells of the stomach, increasing the rate of acid secretion and, consequently, digestion of bone contained therein. Additional experiments, however, demonstrated that animals with chronic occlusion of the left aorta were not remarkably smaller than sham-operated controls and would have likely reached sexual maturity during the same breeding season. In this study, we use flow through respirometry to determine postprandial metabolic rate (specific dynamic action, SDA), and total fecal collection to measure transit time of digesta along with apparent assimilation efficiency. The results may clarify whether there is a functional or adaptive role of the reptilian right-to-left shunt.

23.17

HYPERCARBIC VENTILATORY RESPONSE IN LIZARDS (*TROPIDURUS TORQUATUS*) ACCLIMATED TO DIFFERENT TEMPERATURES

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Temperature is a factor that influences physiological processes and may cause changes in breathing patterns of some species. We evaluated the ventilatory responses to CO_2 of the lizard population, *Tropidurus torquatus*, from the South of Brazil (biome Pampa), acclimated to three different temperatures. To this study, animals (0.040 ± 0.005 kg, males) were collected in Alegrete, (Rio Grande do Sul, Brazil) during the summer season and were acclimated to 20 ($n=2$), 25 ($n=2$) or 30°C ($n=2$) during 30 days. Pulmonary ventilation was measured by pneumotacographic method at 25°C during normocarbica, hypercarbia (5% CO_2) and post-hypercarbia. Lizards acclimated to 25°C showed a higher ventilatory response to CO_2 compared to animals acclimated at 20 and 30°C, due to an increase in tidal volume. Regarding post hypercarbic response, no difference was observed between groups. Therefore, our preliminary data suggest that animals acclimated to 25°C appear to be more sensitive to CO_2 . Financial Support: FAPESP and CNPq.

23.18

THE AVIAN PARADOX: AVIAN RESISTANCE TO PROTEIN GLYCATION

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Avian plasma glucose (P_{Glu}) concentrations are naturally 1.5-2 times those of mammals of similar body mass. Avian tolerance of exceptionally high P_{Glu} is enigmatic considering that mammals with similar values would suffer severe pathologies stemming from hyperglycemia-induced protein glycation. What is not currently

known is how birds protect circulating proteins and the vasculature from high P_{Glu} . The purpose of this study was to explore this "avian paradox." Because protein glycation is an exothermic chemical process, we hypothesized that the unique physiology of birds (higher body temperatures than mammals) would help protect them from protein glycation. Since P_{Glu} concentrations are inversely correlated with body mass, we measured percent glycated albumin in the plasma of three species of birds with varying body masses and body temperatures: house finches (*Haemorrhous mexicanus*, 16-27g, 42.5°C), mourning doves (*Zenaidura macroura*, 90-130g, 41.5°C) and mallard ducks (*Anas platyrhynchos domesticus*, 981-1492g, 40.2°C). Using top down mass spectrometry we discerned that P_{Glu} does not predict albumin glycation demonstrating the generic, species agnostic nature of the avian paradox. In addition, birds with the highest body temperature (finches) have the lowest albumin glycation in comparison to doves and ducks ($6.17 \pm 0.95\%$, $11.7 \pm 1.8\%$, $9.70 \pm 0.7\%$, respectively; $p=0.05$). These data support the hypothesis that higher body temperatures in birds may protect from protein glycation.

23.19

NATURE HIBERNATION PROTECTS MYOCARDIAL ISCHEMIA

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True mammalian hibernation is a unique strategy for surviving extreme environmental stress during winter months, and therefore might induce cardioprotection, even though that is not a stress ever encountered in hibernating animals. The goal of this investigation was to test this hypothesis in a true mammalian hibernator, woodchucks. Myocardial infarction was induced by coronary artery occlusion (CAO) followed by coronary artery reperfusion (CAR) and the myocardial infarcts were quantitated in summer, when not hibernating and in winter, when hibernating. The ischemic area at risk (AAR) was similar in both groups (30 ± 1.9 vs. $32 \pm 5.1\%$). Myocardial infarction expressed as a fraction of the AAR was $49 \pm 5.4\%$ in woodchucks in summer, and significantly less ($p < 0.01$) in woodchucks in winter ($13.2 \pm 4.5\%$). The cardioprotection in woodchucks in winter was mediated by eNOS (endothelial NOS), in contrast to classical preconditioning, which is mediated by iNOS. Whereas there were numerous genes up or downregulated in hibernating woodchuck hearts compared with ischemic preconditioning, one mechanism was unique in the woodchuck, upregulation of CREB, which when upregulated in the heart in summer induced protection similar to that observed in the woodchuck heart in winter. The hibernating woodchuck heart is a novel model to study cardioprotection, since powerful cardioprotection occurs naturally in winter months, even in the absence of ischemic preconditioning.

23.20

CHARACTERIZING RESPIRATORY PARAMETERS IN CAPTIVE BELUGAS AND FROM EXCISED LUNGS

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Little data currently exists on the beluga respiratory system. Our objective was to characterize and compare respiratory parameters in belugas using two methods: (1) pulmonary mechanic tests (PMT) on resting, captive belugas at the Vancouver Aquarium and L'Océanographique, and (2) mechanics of excised lungs obtained from the Inuit harvest in the western Arctic. For PMTs, a pneumotachometer was placed over the blowhole and an esophageal balloon catheter was inserted to obtain simultaneous measurements of transpulmonary pressures and flow for each breath. For excised lungs, both static and quasi-static pressure-volume curves were generated. Respiratory parameters characterized include flow rates (L/s); resting tidal, maximum lung, and minimum air volumes (mL/kg); specific lung compliances ($\text{cmH}_2\text{O}^{-1}$); opening airway and esophageal pressures (cmH_2O); and expiratory CO_2 and O_2 concentrations (%). These tests can be used to better understand normal respiratory physiology in cetaceans, and PMTs can be used clinically to assess the pathophysiology of respiratory disease and in health assessments of wild populations.

23.21

GRAND PARADIGM SHIFT IN DIVING PHYSIOLOGY: THE LIKELIHOOD OF THE BENDS IN MARINE VERTEBRATES

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Decompression sickness (DCS) is a collection of symptoms caused by bubbles forming due to gas phase separation in the body. Anatomical, physiological, and behavioral traits are thought to protect breath-hold diving marine vertebrates against DCS. However, increasing evidence suggest that under certain circumstances marine mammals and sea turtles appear to experience gas bubbles following diving. The abundance and gas composition of gas bubbles found after severe decompression versus decomposition was experimentally established in New Zealand White Rabbits. Using our methods we found that 85% of stranded marine mammals, 100% of bycaught marine mammals, and 43% of bycaught sea turtles, presented with gas bubbles. Deep divers presented with larger amounts of gas bubbles than non-deep divers. The gas composition of bubbles found in cetaceans, pinnipeds, and sea turtles was similar to the gas composition of bubbles in compressed and decompressed rabbits. The results suggest that diving air-breathing vertebrates deal daily with gas management and potential gas bubble formation. In extreme cases, animals seem to exhibit DCS lesions and symptoms similar to human divers.

23.22

ADAPTATION TO HIGH ALTITUDE IS LINKED TO GROWTH OF THE LUNGS AND HIGHER ALVEOLAR SURFACE IN MICE BUT NOT IN RATS

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We tested the hypothesis that rats and mice living for >30 generations at high altitude (HA) have different adaptation of their lung structure. We used adult mice and rats raised under laboratory conditions in La Paz, Bolivia (3600m - HA) or in Quebec City (Sea Level - SL). After anesthesia, the lungs were inflated and fixed with PFA, lung weight and lung volume were measured, lungs were then embedded in paraffin and relative and total alveolar surfaces were calculated by morphometric analysis on lung slices (5µm thick). We performed weight independent comparisons using allometric corrections. Compared to rats, HA mice have higher lung weight and volume (2 x higher, p<0.0001). The relative alveolar surface was $0.34 \pm 0.01 \text{ m}^2/\text{cm}^3/\text{g}^{0.13}$ in rats and $0.45 \pm 0.02 \text{ m}^2/\text{cm}^3/\text{g}^{0.13}$ in mice (p<0.0001), while the total alveolar surface was $0.009 \pm 0.001 \text{ m}^2/\text{g}^{0.88}$ in rats and $0.031 \pm 0.002 \text{ m}^2/\text{g}^{0.88}$ in mice (3.4 x higher, p<0.0001). Lung volume and weight of the HA and SL rats were similar, but were respectively 2.1 and 1.8 x higher in HA mice compared to SL mice. At SL, exposure to hypoxia during postnatal days 4-14 (alveolar formation) slightly increases (1.25 x) lung volume in mice, but decreases the relative alveolar surface. We conclude that in mice living at HA genetic selection might increase the volume of the lungs and favor the expansion of the alveolar surface. Differences between rats and mice explain their divergent success at colonization of HA regions. Funded by NSERC.

23.23

AEROBIC PHYSICAL TRAINING INCREASES CONTRACTILE RESPONSE AND REDUCES THE CARDIAC FIBROSIS IN RATS SUBMITTED TO EARLY ESTROGEN DEPRIVATION

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Introduction: We investigated the effects of early estrogen deprivation on the heart and the role of physical training in this condition using different approaches: contractility, morphology and function, and cardiac fibrosis. **Methods:** Female Wistar rats (N=48) were assigned into two groups: ovariectomized (OVX; 22 weeks-old) and control rats (SHAM; 22 weeks-old). Each group was subdivided into two subgroups, sedentary and trained (aerobic physical training by swimming for 10 weeks). **Results:** The contractile responses to cardiac β -agonists were similar, including an increased response to a β_1 -agonist (dobutamine) observed after physical training. The OVX sedentary group presented changes in cardiac morphology, which resulted in a decreased ejection fraction, fractional shortening and cardiac index in relation to the SHAM sedentary group. Physical training did little to alter these findings. Moreover, the histology analysis showed a significant increase in cardiac fibrosis in the sedentary OVX group, which was not observed in the trained OVX group. **Conclusions:** Early estrogen deprivation impairs cardiac morphology and cardiac function. This condition also increases cardiac fibrosis; however, it does not affect contractility induced by dobutamine and salbutamol (β_2 -agonist). Furthermore, this model of physical training prevented increased fibrosis and promoted an increased cardiac contractile response but had little effect on the morphological and functional parameters. Funding: FAPESP (2014/01937-9); CNPQ (135531/2011-0).

23.24

DYNAMICS OF OXYGEN UTILIZATION DURING PASSIVE AND ACTIVE CYCLING EXERCISE

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¹Dept. of Bio-sys. Eng., Yamagata Univ., 4-3-16 Jonan, Yonezawa, 992-8510, Japan. Few studies have compared oxygen utilization during passive exercise with that during active exercise. In this study, we investigated the dynamics of oxygen utilization during passive and active cycling exercise. Eight healthy young men participated in this study. All exercise tests were performed in a seated position on an electronically braked cycle ergometer. After a 2-minute rest, the subjects received passive cycling exercise or performed active cycling exercise at an output intensity of 26 W for 3 min, followed by a 2-minute rest. Throughout the exercise tests, pulmonary gas exchange rates, ventilation level, and heart rate were measured breath by breath, and hemoglobin concentrations of the vastus lateralis muscles were measured at 10 Hz. Oxygen uptake linearly increased by approximately 250 ml/min during passive exercise compared with the rest level, whereas it exponentially increased by approximately 500 ml/min during active exercise. During both exercises, the change in deoxyhemoglobin concentration exponentially decreased (passive: 0.12 a.u., active: 0.10 a.u.). However, during active exercise, it increased by 0.02 a.u. at 50 sec after starting exercise, and then it was steady. These results showed that oxygen extraction of contracted muscles was not accelerated during passive exercise, whereas it was accelerated during active exercise.

23.25

THE EFFECTS OF MODULATING ENDOTHELIAL NITRIC OXIDE SYNTHASE (ENOS) ACTIVITY AND COUPLING IN CORONARY, HINDLIMB, RENAL, AND MESENTERIC VASCULAR INFLAMMATION MODELS

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Ischemia/reperfusion (I/R) injury is initiated in part by vascular endothelial dysfunction, which is characterized by reduced endothelial-derived nitric oxide (NO) and/or increased oxidative stress, followed by inflammation. When the tetrahydrobiopterin (BH₄) to dihydrobiopterin (BH₂, oxidized form of BH₄) ratio is reduced, eNOS can become uncoupled shifting production of NO to superoxide (SO). Protein kinase C epsilon activator (PKCε+) enhances eNOS activity while PKCε inhibitor (PKCε-) reduces eNOS activity. The effects of PKCε+ or PKCε- combined with BH₄ or BH₂ were studied in rat myocardial and hindlimb I/R, rat renal lithotripsy, and rat mesenteric inflammation models. Promoting eNOS coupling using PKCε+ with BH₄ or inhibiting uncoupled eNOS activity using PKCε- significantly increased blood NO and decreased blood H₂O₂ levels in reperfused femoral and renal veins, reduced BH₂-induced leukocyte-endothelial interactions in mesenteric postcapillary venules, and improved post-reperfused cardiac function associated with reduced leukocyte heart tissue infiltration when compared to controls. In contrast, PKCε+ with BH₂ had opposite effects. These results suggest that enhancing coupled eNOS or inhibiting uncoupled eNOS activities can attenuate the I/R-induced vascular endothelial dysfunction, inflammation, and organ damage. This study was supported by NHLBI Grant 2R15HL76235-02 and the CCDA at PCOM.

23.26

THE EFFECT OF STREPTOKINASE INFUSION ON CARDIAC BIOMARKERS & ST SEGMENT OF ELECTROCARDIOGRAM POST MYOCARDIAL INFARCTION IN HUMANS

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Acute STEMI (ST-segment elevation myocardial infarction) is one of the most common diagnoses in admitted patient's world widely with significant mortality. Thrombolytic therapy remains the most common reperfusion strategy for STEMI, particularly in smaller centers. Efficacy of treatment is dependent on various factors specially type and time to treatment. Ability to decrease symptom onset to door time is limited, national organization have placed substantial emphasis on decreasing door to needle time (DTN time). Guidelines recommended DTN time is 30 minutes or less. This study was designed for elevation of STEMI treatment in hospital, especially for determining of SK injection time. Methods and Materials were including all STEMI patients eligible for thrombolytic therapy in Nikan hospital included in this study. The most common damaged wall was anterior wall with greatest mortality. ST segment in patients who received SK in 30 min was in optimal condition also in this cases decrease of CPK was according to the guidelines and Troponin I concentration respectively. Finally, the result was that we should emphasize to limitation of controllable risk factor such as smoking and Hyperlipidemia to decrease cardiovascular events. As the people medical knowledge level and good 115 Emergency services have significant effects on decreasing pre-hospital delay but the ability to decrease this delay is limited. So we should decide to decrease in-hospital delay & DTN time from mean 57 minutes to recommended min. Isolation of cardiac Emergency room with highly educated physician and personal should be used to achieving this goal.

23.27

PROTEIN KINASE C (PKC) DELTA (Δ) ACTIVATOR ATTENUATES N^{G} -NITRO-L-ARGININE-METHYL-ESTER (L-NAME) INDUCED LEUKOCYTE-ENDOTHELIAL INTERACTIONS IN RAT MESENTERIC POST-CAPILLARY VENULES

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Vascular endothelial dysfunction has been reported as the initial and critical step leading to inflammation-related diseases. It is characterized by reduced endothelial-derived nitric oxide (NO) and/or increased oxidative stress (i.e., superoxide (SO)), which leads to increased leukocyte-endothelial interactions. Endothelial dysfunction is attenuated by enhancing NO generation via endothelial NO synthase (eNOS) or by reducing SO release via inhibition of NADPH oxidase. PKC δ negatively regulates NADPH oxidase and reduces SO release, which will attenuate quenching of NO. The role of PKC δ on leukocyte-endothelial interactions is unclear. This study examined these interactions, which are characterized by leukocyte rolling, adherence, and transmigration in rat mesenteric postcapillary venules via intravital microscopy. We found that superfusion of L-NAME (MW=270, 50 μM , n=5) significantly augmented inflammation by inhibiting NO production via eNOS compared to Krebs' buffer control (p<0.01, n=5). Conversely, PKC δ activator (Myr-MRAEDPM, MW=1130, 10 μM , n=4), a cell-permeable peptide, significantly attenuated L-NAME induced leukocyte-endothelial interactions (p<0.05). The preliminary data suggest that PKC δ activation may be an important mechanism in attenuating leukocyte-endothelial interactions induced by endothelial dysfunction. This study was supported by the Center for Chronic Disorders of Aging at PCOM.

24.0: DEVELOPMENTAL PHYSIOLOGY

24.1

GENETICALLY DETERMINED VARIATION IN METABOLIC ALLOCATION-POTENTIAL FOR ADAPTATION TO ENVIRONMENTAL CHANGE

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Defining the genetic basis of physiological variance is key for predicting adaptive potential of populations to climate change. For most organisms of interest to comparative physiologists – in particular, for marine animals – this goal has been elusive, owing to a lack of genetically enabled models. We crossed pedigreed lines of the Pacific oyster *Crassostrea gigas* to produce >20 families that were screened for physiological contrasts. Larvae were assayed for variation in physiology of Na^+ , K^+ -ATPase, at three biological levels: gene expression, total protein, and *in vivo* ion transport. Further, the proportion of metabolic rate allocated to the physiological activity of Na^+ , K^+ -ATPase was determined. Similar-sized larvae from different families had up to 3-fold variation in ion transport that was not detectable by changes in gene expression or total protein. Significant variation also occurred in the proportion of metabolic rate allocated to ion transport, ranging from 10 to 39%, depending on family. Protein synthesis which accounted for an even greater proportion of metabolic rate, varied substantially in rate and hence ATP demand between families. Further, variation exists among families in how ATP was reallocated to synthesis and transport under simulated ocean acidification. This variation reflects adaptive potential within populations and suggests that while many individuals may suffer under climate change, others may be resilient. Supported by NSF grant EF1220587.

24.2

EXPOSURE TO LOWERED PH AND ACUTE THERMAL STRESS INCREASES MORTALITY IN EMBRYONIC PORCELAIN CRABS

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Increased atmospheric $p\text{CO}_2$ is expected to lead to decreased oceanic pH and increased frequency and severity of extreme heat events, both of which are likely to be deleterious for intertidal ectotherms. However, responses to these forcing agents can vary greatly even within a single species, and sensitive early life history stages may pose an ecological weak link in population persistence. We investigated the effects of reduced pH and acute thermal stress on growth and survival of embryos of the porcelain crab *Porcellana platycheles*. Early stage embryos were removed from field-collected females (n = 6; 96 embryos/female) and reared until hatching (~27 days) under one of two pH treatment conditions (pH=8.0, pH=7.6). Embryos were exposed to one of four temperature regimes: (1) constant ambient 20 °C, or 1 h exposure to 31 °C on (2) Day 1, (3) at start of heart beat, or (4) both Day 1 and start of heart beat. Photos of

embryos were taken every other day for the duration of the experiment. Embryo lengths and volumes were estimated from photos using the program ImageJ and growth rates were calculated as change in length/time. Embryo mortality was ~25 times higher with heat shock in early development (Day 1) under low pH when compared to controls, suggesting that *P. platycheles* embryos are likely to be strongly negatively impacted by acute heating events predicted under future climate scenarios. This project was funded by NSF grant MCB-1041225 to JHS, and PU funding to PC.

24.3

REALLOCATION OF ATP IN RESPONSE TO THE STRESS OF OCEAN ACIDIFICATION

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Defining the potential for resilience is essential for understanding and predicting the biological impact of climate change. Organisms withstand sublethal stress through physiological adjustments to maintain homeostasis, resulting in tradeoffs in use of metabolic energy. We have studied tradeoffs by quantifying ATP allocation to protein synthesis and ion regulation in the sea urchin, *Strongylocentrotus purpuratus*, developing under present-day (control) and near-future $p\text{CO}_2$ conditions (~800 μatm). Seawater acidification had minimal effect on size, growth and metabolic rate. However, substantial biochemical rate compensation occurred to sustain this resilience at the organismal level. Larvae of similar size and metabolic rate responded to acidification with ~50% increases in protein synthesis rate and ion transport by Na^+ , K^+ -ATPase. Up to 86% of the ATP pool was allocated to these two processes. In terms of the hierarchy of ATP allocation, protein synthesis is more significant than the cost of supporting changes in ion transport. We are currently manipulating $p\text{CO}_2$ and pH independently to determine which component of the seawater carbonate chemistry is responsible for the observed biochemical changes. We conclude that the apparent minimal responses to acidification at the organismal level actually involve major changes at the cellular level, with substantial impacts on energy allocation within fixed ATP pools. Supported by NSF EF1220587.

24.4

CHARACTERIZING METAMORPHOSIS OF THE ALFALFA LEAF-CUTTING BEE USING MICRO-COMPUTED TOMOGRAPHY

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Pupal development is particularly sensitive to environmental perturbations, because the insect is immobile while physiological systems essential for the adult are constructed from larval and imaginal tissues. Severe environmental fluctuations during the pupal period could disrupt development, resulting in a negative impact on adult performance. In the alfalfa leaf-cutting bee, *Megachile rotundata*, exposure to cold temperatures during pupal development causes sub-lethal effects on adult performance, including flight deficiencies, altered metabolic rates, behavioral changes, and decreased longevity; however, the mechanisms underlying these effects are not known. Disruption of one or more developing systems may be responsible for the deleterious effects seen in adults after pupae were exposed to cold temperatures; however the internal changes associated with pupation are a "black box." We used μCT to investigate the development of flight structures, metabolic reserves, and the digestive tract during *M. rotundata* pupal development. Pupal development occurs over three weeks, during which time we were able to clearly see development of these structures. Gut and flight structures appear to change continuously during the pupal period. However dense metabolic reserves may decrease only slightly throughout pupal development. These data will allow us to better understand what happens during metamorphosis, and to make comparisons about how these systems are altered after cold exposure.

24.5

SHIFTING INCUBATION TEMPERATURES ALTER HEART RATE AND OXYGEN CONSUMPTION OF LAKE WHITEFISH EMBRYOS AND HATCHLINGS

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Critical windows are periods of developmental susceptibility when an animal's phenotype may be vulnerable to environmental fluctuations. We incubated whitefish embryos at control temperatures of 2°C, 5°C, or 8°C, and shifted embryos into another of those same temperatures at the end of gastrulation or organogenesis to de-

termine if gastrulation or organogenesis represent critical windows. Heart rate (f_H) and oxygen consumption (VO_2) were measured across embryonic development, and VO_2 was measured in 1d larvae. Thermal shifts up or down from initial incubation temperatures caused f_H and VO_2 to differ from control embryos measured at the same temperature (2°C, 5°C, 8°C). Most prominently, when embryos were measured at organogenesis compared to controls, exposure to 2°C or 5°C through gastrulation resulted in lower VO_2 and f_H at 8°C, exposure to 2°C resulted in lower VO_2 and f_H at 5°C, and exposure to 5°C and 8°C resulted in lower VO_2 at 2°C. Through the latter half of development, VO_2 and f_H trended toward recovery to control values for thermally shifted treatments. However, in hatchling larvae measured at 2°C, VO_2 was higher in groups exposed to 8°C or 5°C through organogenesis, compared to 2°C controls (43-65% increase), which further indicates that development through organogenesis represents a critical window of physiological plasticity. This study presents a unique experimental design that identified periods of thermal sensitivity in fish embryonic development.

24.6 PHYSIOLOGICAL CONSEQUENCES OF COMPENSATORY GROWTH IN THE CHECKERED GARTER SNAKE, *THAMNOPHIS MARCIANUS* Kaitlyn Holden¹, Anne Bronikowski¹, and Neil Ford²

¹Ecology, Evolution and Organismal Biol., Iowa State Univ., Bessey Hall, Ames, IA, 50011, ²Biology, Univ. of Texas at Tyler, 3900 University Blvd., Tyler, TX, 75799. Variation in physical and developmental environments can influence life-history traits through phenotypic plasticity. Poor natal nutrition can lead to subsequent energy intake being diverted between compensatory growth or delayed maturation. If an organism does allocate energy to compensatory growth and "on-time" maturation, there may be consequences across physiological axes. For example, immune defense is an energetically expensive biological process, which can trade-off with traits such as growth. Here we test how poor natal nutrition impacts growth, immune function and glucocorticoid production both during a natal phase of poor nutrition and a subsequent phase of rich nutrition in the checkered garter snake, *Thamnophis marcianus*. We found accelerated growth rates after the switch to high-quality food, with no impact on innate immune function and glucocorticoid production. However, we found negative impacts on subsequent reproduction, with those individuals experiencing poor natal nutrition producing smaller offspring. This suggests that innate immune function is not compromised when excess energy is allocated towards growth compensation. Thus immune function may be maintained during periods of stress at the expense of other attributes.

24.7 HYPOXIA DURING CRITICAL WINDOWS OF ONTOGENY ALTERS ORGAN MASS AND CARDIOVASCULAR FUNCTION IN THE AMERICAN ALLIGATOR (*ALLIGATOR MISSISSIPPIENSIS*)

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Hypoxic incubation represents an important and relevant challenge to the maturation of embryonic reptiles. We have previously reported that chronic hypoxic (10% O₂; H) incubation of American alligator eggs results in a decrease in embryonic mass, increase in relative heart mass, decrease in heart rate and decrease in arterial pressure. Further investigation has identified a "critical window" for hypoxic application is within the 20-70% of incubation period. Our goal was to further isolate the critical period by narrowing the windows of hypoxic incubation as well as reciprocal shifts between normoxic (N) conditions and 10% O₂ conditions at 50 and 70% with measurements taken at 90% of incubation. Shifting of hypoxic embryos from 10% O₂ to normoxia (H to N) at 50% or 70% of incubation resulted in a relative enlargement of the heart compared to control values. Further while embryonic mass was decreased in eggs moved back to normoxia at 70%, embryonic mass of eggs that were returned to normoxia at 50% did not significantly differ from the control groups. Functional cardiovascular phenotypes produced as a result of hypoxia occur within a critical window spanning a 20% segment of incubation. Currently we are investigating changes in gene expression that may provide insight into the basis for the morphology and functional difference between these groups. NSF CAREER IBN IOS-0845741 to DAC.

24.8 TEMPERATURE EFFECTS ON HEART RATE AND BAROREFLEX FUNCTION OF EMBRYONIC AMERICAN ALLIGATOR (*ALLIGATOR MISSISSIPPIENSIS*)

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Nest temperatures of American alligators are known to fluctuate on a diurnal cycle. While these fluctuations have been previously documented, the impact on embryonic cardiovascular physiology is poorly understood. Our prior studies have demonstrated that cardiovascular regulatory capacity in alligator embryos is limited compared to adults and the embryo relies heavily on endocrine control of function. Further, embryonic alligators possess a hypertensive baroreflex response only when studied at 30°C. A recent study suggested that cardiovascular homeostatic mechanisms are temperature dependent in embryonic alligators, with vagal tone activated both above and below 30°C. Based on these findings we hypothesized that a hypotensive baroreflex would be functional when embryos are studied temperatures above and below the 30°C threshold. Further, the baroreflex gain would increase in response to the elevation in resting heart rate. We assessed heart rate and arterial pressure responses of embryonic alligators at 90% of incubation to increasing and decreasing temperatures. In addition the Oxford method was used to assess the cardiac limb of the baroreflex. The project is ongoing and the findings of these investigations will be presented. Project funded by NSF CAREER IBN IOS-0845741 to DAC.

24.9 THE EFFECTS OF CHRONIC AND ACUTE HYPOXIA ON CARDIAC FUNCTION IN EMBRYONIC CHICKENS

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Chronic hypoxia is a known developmental insult, and studied extensively for its hemodynamic effects in early chicken development. However the effects of chronic and acute hypoxia on cardiac function and arterial coupling in the late-stage embryonic chickens. Our goal was to quantify how chronic and acute hypoxia affects cardiac function. Eggs incubated at 21% or 15% O₂. At 19 days, prior to lung ventilation, embryos were anesthetized and surgically instrumented to study left ventricular (LV) pressure-volume loops. Hearts were collected for mRNA analysis. Hypoxia-incubation led to growth restriction (-20%) increased heart/body ratio (+17%) and LV/body ratio (+40%). The maximal rate of LV pressure generation, dp/dt_{max} , was lower in hypoxia-incubation (-20%), as were end systolic LV elastance (E_{LV} ; +30%), arterial elastance (E_A ; +120%), and LV output (-46%). Both hypoxia-incubation and acute hypoxia (10% O₂) lengthened tau, the half-time of relaxation (+20%). Acute hypoxia reduced heart rate (-8%) and increased end diastolic pressure (+20%) without changing dp/dt_{max} , tau, E_{LV} or E_A . These were accompanied by reduced mRNA for intracellular calcium handling genes. Hypoxia-incubation reduces LV function slowing pressure generation and relaxation, possibly driven by altered intracellular excitation-contraction coupling. The ratio of E_A/E_{LV} is much higher with hypoxia-incubation, indicating decreased cardiac efficiency.

24.10 DEVELOPMENTAL PHYSIOLOGY OF THE PEKIN DUCK (*ANAS PEKIN*) DUCTUS ARTERIOSUS

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In developing avian embryos, the left and right ductus arteriosus shunt pulmonary blood away from the lungs to the systemic circuit and chorioallantois. In mammals and birds studied to date, the ductus arteriosus (DA) are oxygen sensitive vessels that constrict in response to increasing oxygen levels. We examined physiology of the DA from Pekin duck, *Anas pekin* in internally and externally piped (IP/EP) embryos. The *in vitro* contractile response of the left DA was measured using wire myography. Duck DA from both internally and externally piped stages contracted in response to increasing levels of oxygen. The DA relaxed in response to high levels of NaS only in the presence of high oxygen levels. Under low oxygen levels, the relaxing response was muted. The DA contracted in response to stepwise increasing phenylephrine. At low concentrations, norepinephrine produced a weak contraction, followed by relaxation at higher concentrations. In the presence of oxygen, sodium nitroprusside produced strong DA relaxation. In the presence of acetylcholine, the DA initially relaxed at low concentrations followed by contraction at higher concentrations. The EP ductus relaxed in response to the Rho-kinase inhibitor fasudil hydrochloride under high oxygen levels. These results suggest the physiology of the duck DA is similar to that of chicken DA. This research was funded in part by an HHMI grant to Lee Hughes and NSF IOS 1146758 (EMD).

24.11 DEVELOPMENTAL CHANGES IN MRNA LEVELS OF AVANT AND PGC-1A IN DUCK LIVER AND HEART

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atures. After hatching, a neonate bird must increase aerobic capacity to obtain endothermic capacity. A number of transcription factors and transporters, including PGC-1 α and avian-ANT (avANT), are thought to play a role in development of endothermy by regulating mitochondrial biogenesis and function. To further understand the molecular regulation of endothermic development, we isolated mRNA from heart and liver of a precocial species, the domestic Pekin Duck (*Anas pekin*), at embryonic and neonatal stages of development. On the day of hatching, cardiac PGC-1 α mRNA levels increased significantly compared to externally piped (EP) embryos. Liver PGC-1 α mRNA levels significantly declined from embryonic day 24 (E24) to EP. Both cardiac and liver avANT significantly increased from E24 to 1dph. A better understanding of the roles of transcription factors play in mitochondrial biogenesis during development of endothermy will provide a better understanding of the regulation of physiological development occurring in neonate avians during early development. Supported by NSF IOS 1146758 (EMD).

24.12

THYROID HORMONE AND DEVELOPMENT OF ENDOTHERMY IN KING QUAIL

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Thyroid hormones (TH) are key regulators of vertebrate metabolism and are thought to regulate development of endothermy in mammals and birds. To better understand TH effects on development of metabolic physiology in a small, precocial avian, we treated embryonic and neonate King Quail (*Coturnix chinensis*) with triiodothyronine (T3) or the thyroperoxidase inhibitor, methimazole to induce hyper and hypothyroidism, respectively. Preliminary results suggest King Quail hatchlings (up to 1dph) maintain constant metabolic rate as temperature decreases from 35°C to 20°C. Metabolic rate of older birds (>1dph) began to increase as ambient temperature cooled below 20°C. Previous work with other avian species (Red-winged Blackbird (*Agelaius phoeniceus*) and Double-crested Cormorant (*Phalacrocorax auritus*)) suggests hypothyroid treatment will delay maturation of endothermic capacity by affecting organ mass, such as heart and liver, as well as oxygen consumption rates and mitochondrial respiration of permeabilized skeletal muscle fibers. We expect T3 treated animals to obtain endothermic capacity more quickly after hatching than control, while the embryonic ectothermic phenotype typical of birds will persist for a longer duration after hatching in hypothyroid animals. Supported by NSF IOS 1146758 (EMD).

24.13

HOW TO BUILD A FURNACE: THE ROLE OF T3 IN DEVELOPMENT OF ENDOTHERMY IN THE ALTRICIAL RED-WINGED BLACKBIRD (*AGELAIUS PHOENICEUS*)

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Thyroid hormones (TH) are key regulators of metabolism and are thought to regulate development of endothermy in birds. To better understand effect of TH on development of an altricial species, we treated neonate Red-winged blackbirds with methimazole to induce hypothyroidism. We measured whole animal O₂ consumption (VO₂) and ventilation, followed by mitochondrial respiration of permeabilized fibers from breast and thigh muscle. We examined organ mass and characters such as wing chord and femur length. Whole animal VO₂ of hypothyroid subjects differed significantly from control: treated animals exhibited lower VO₂ on 5dph and 7dph. Hypothyroid animals showed significantly lower body mass than control. Heart mass of treated animals was significantly lower than control, while liver mass was higher. Wing chord and femur length were significantly lower in hypothyroid animals. Mitochondrial respiration from permeabilized breast muscle was significantly lower in 7dph hypothyroid animals than control. Thigh mitochondrial respiration showed no difference between control and treated animals at any age, though data suggest a trend of lower hypothyroid respiration. Our data suggest TH plays an active role in the systemic attainment of endothermic capacity. In the neonate avian, multiple systems develop in concert to produce an endothermic phenotype, but reduced TH can delay maturation of endothermic capacity. Supported by NSF Grant IOS-1146758.

24.14

CONDITIONAL PULMONARY OVEREXPRESSION OF CLAUDIN 6 (CLDN6) DURING EMBRYOGENESIS DELAYS LUNG MORPHOGENESIS

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Claudin 6 (Cldn6), a tetraspanin tight junctional protein, is a participant in cell junction assembly and compromised membrane permeability results when Cldn6 expression is abnormal. In order to assess the effects of persistent tight junctions involving Cldn6

during lung development *in vivo*, a doxycycline (dox)-inducible conditional transgenic mouse was generated that up-regulates Cldn6 in the distal lung. Pups had access to dox from before conception until sacrifice date at embryonic day (E) 18.5. Transgenic pups that express Cldn6 were removed by c-section and compared to non-transgenic littermates. Quantitative real time PCR, immunoblotting, and Cldn6 immunohistochemistry revealed elevated Cldn6 in transgenic mice compared to controls. There were no differences between the groups in terms of lung size or weight. Histological evaluations led to the discovery that E18.5 Cldn6 transgenic pups appeared to be in the canalicular stage of development wherein respiratory airspaces were thickened and fewer in number. In contrast, controls appropriately appeared to have entered the sacular stage coincident with thin airspace walls and spherical architecture. Immunostaining for transcriptional regulators including TTF-1 and FoxA2 was conducted to assess cell differentiation programs and specific lung cells were identified via staining for surfactant protein C (Type II cells), T1alpha (Type I cells), CCSP (Clara cells), and FoxJ1 (ciliated epithelium). These data suggest that Cldn6 is an important junctional protein potentially involved in the programming of epithelial cells during lung development. Furthermore, normal downregulation of Cldn6 as development proceeds may influence differentiation associated with the transition from the canalicular to the sacular lung.

25.0: ENDOCRINOLOGY AND REPRODUCTION

25.1

HYPERGLYCEMIC AND PUTATIVE HYPERLIPIDEMIC ACTIVITIES OF THE RECOMBINANT CRUSTACEAN HYPERGLYCEMIC HORMONE CHH-B1 ISOFORM IN THE PACIFIC WHITE SHRIMP *LITOPENAEUS VANNAMEI*

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Crustacean hyperglycemic hormone (CHH) is the most abundant neuropeptide of the sinus gland in crustacean's eyestalks. The main function of CHH is to stimulate hyperglycemia in hemolymph. Studies suggested that CHH also promotes hyperlipidemia under a high energy demand. Lipids are structural body components that serve as alternative metabolic substrates to glucose (Glc). Triglycerides (Tg) are the main reserve lipids in crustaceans, serving as a source of fatty acids for energy fuel, and glycerol as substrate for gluconeogenesis. CHH-B1 is one of the three CHH isoforms found in the Pacific white shrimp *Litopenaeus vannamei*. Recombinant CHH-B1 with C-terminal tags (C-myc, 6XHis) was previously cloned and expressed in yeast showing hyperglycemic activity *in vivo*. Since CHH-family peptides are often multifunctional, we study the effect of the recombinant CHH-B1 on hyperglycemic triglyceride metabolism. Recombinant CHH-B1₁₋₁₆ was expressed in *Pichia pastoris* and purified by affinity chromatography. Shrimps on intermolt were injected with 2 µg of rCHH-B1₁₋₁₆ and hemolymph was sampled at 0.5, 1, 2, 4, and 24 h post-injection to measure plasma Tg and Glc levels. Results showed that rCHH-B1₁₋₁₆ caused hyperlipidemia by significant raising (P < 0.05) Tg levels at 2 h, matching with a hyperglycemia depletion maybe due a switching to lipid oxidation as energy fuel and also to regenerate consumed glucose reserves by a rapid Tg uptake by tissues.

25.2

BIASED SIGNALING BY TWO ENDOGENOUS GnRH ISOFORMS DIFFERENTIALLY REGULATES TOTAL LH AND GH AVAILABILITY IN GOLDFISH PITUITARY CELLS

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In goldfish (*Carassius auratus*), both endogenous gonadotropin-releasing hormones, GnRH3 and GnRH2, stimulate luteinizing hormone (LH) and growth hormone (GH) release through the activation of multiple signal transduction cascades including isoforms of phosphoinositide 3-kinase, protein kinase C, and mitogen-activated protein kinase, as well as via the differential use of Ca²⁺-dependent intracellular signaling mechanisms. In this study, we examined the interactions between these signaling pathways in acute and long-term control of basal and GnRH-stimulated LH and GH release using primary cultures of dispersed goldfish pituitary cells. Our results indicate that distinct signal transduction networks selectively control basal and GnRH-stimulated hormone release in a time-, pituitary cell type-, and GnRH isoform-specific manner and that changes in total LH and GH availability are often dissociated from their known mRNA expression profiles. These findings provide important details into the molecular mechanisms that couple biased GnRH activation to signal transduction responses, in general, while also adding to our understanding of how intracellular signaling dynamics within the neuroendocrine system ultimately contribute to the regulation of whole-organism physiology. (Supported by NSERC, AIHS, and the Killam Trusts)

25.3

PERCHLORATE EXPOSURE DOES NOT AFFECT HORMONE CYCLING OVER DIET AND REPRODUCTIVE SEASON SCHEDULES IN THREESPINE STICKLEBACK

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Photoperiod drives many hormonally-mediated physiological processes. For example, thyroid hormones (thyroxine (T₄) and triiodothyronine (T₃)) affect growth, metabolism, and reproduction, and in some species, exhibit daily and seasonal variations. Previously we showed that exposure of threespine stickleback (*Gasterosteus aculeatus*) to the endocrine disruptor perchlorate results in pronounced structural changes in thyroid and gonad, while T₃ and T₄ concentrations remain stable. The current study evaluated the interactive effects of time (diel, reproductive season (RS)) and perchlorate treatment on thyroid and androgen hormone regulation in Alaskan *G. aculeatus*. Wild-caught adult stickleback were exposed to 100 ppm perchlorate and sampled over the 24 hour day and across the RS (May-July). Whole body T₃ and T₄ concentrations showed no significant differences within a given day in response to perchlorate. Across the RS, whole body T₃ concentration remained stable while T₄ significantly increased, but neither was significantly affected by perchlorate. Similarly, the level of 11-ketotestosterone (11-KT), a major fish androgen, was not influenced by perchlorate exposure. The concentration of whole body 11-KT in males steadily declined across the RS. The observed increase in T₄ across the RS may have implications for energetic investment in reproduction near the end of the stickleback life cycle, although perchlorate does not modify this response. Funding: NIEHS # 1RO1ES017039-01A1.

25.4

EXOGENOUS AVT SUPPRESSES COURTSHIP BEHAVIOR IN *XENOPUS LAEVIS*

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Arginine vasotocin (AVT) is a neurohypophysial peptide known to influence social and reproductive behavior in non-mammalian vertebrates. In anurans, increased AVT in males is associated with increased courtship behaviors and decreased aggression; however it is increasingly apparent that the effects of AVT may depend on the social structure of a given species. *X. laevis*, a fully aquatic frog species with a prolonged breeding season, possess a rich repertoire of vocalizations used for inter- and intra-sexual signaling. They do not appear to be territorial, but do show signs of a male social hierarchy and flexible alternative reproductive tactics. We examined how exogenous AVT altered vocal communication and clasping behavior in male *X. laevis*. When injected males were paired with sexually receptive females, advertisement calling was significantly decreased during the first hour after injection as compared with a saline control. When two AVT-injected males were paired, there was no difference in advertisement calling from control, but there was an increase in male chirps and growls, calls associated with male-male competition. Furthermore, we found that AVT administration led to decreased clasping behavior; male-male amplexus was eliminated and male-female amplexus was reduced and delayed. Thus, unlike in other frog species, courtship behaviors were decreased immediately following AVT administration and male aggression may be increased. These differences may be due to underlying differences in social structure from other anuran species tested, or they may stem from the different physiological consequences of altering osmotic regulation in this aquatic animal. Funded by Denison University Research Fund and Anderson-Bowen Endowments.

25.5

TRH INCREASES GH, BUT NO THYROID HORMONES RELEASE DURING COLD EXPOSURE IN GREEN IGUANA

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Thyrotropin-releasing hormone (TRH) has dual actions in controlling secretion of hypophyseal hormones among vertebrates. In non-mammalian species, TRH is able to stimulate GH secretion in addition to the regulation of thyroid hormones (THs). However, little is known about its role on the somatotrophic and thyroid systems in reptiles as a consequence of fluctuations on environmental temperatures. Here we describe the effect of 48 h cold exposure (25 and 18 °C, respectively, being the control 35 °C) upon the GH-IGF1 and thyroid systems on green iguana. Serum GH increased slightly at 25 °C, and by 41 % at 18 °C. In turn, IGF-1 concentration remained unchanged although its liver gene expression increased 3-fold. In contrast, thyroid activity diminished in both experimental conditions, as shown by the decrease of TSH hypophyseal gene expression (~90 %), and the serum concentration of T₄ and T₃ (from 11.6 to 5.3-7.8 ng/ml and from 0.87 to 0.05-0.25 ng/ml), and the iodothyronine

deiodinase 2 (D2) activity reduction (from 992.5 to 22 fmol⁻¹254nm⁻¹h⁻¹). However, TRH hypothalamic gene expression and content increased at both temperatures, by 1 and 4.1-fold, respectively. Also, GH increased after 2 h of 1 µg TRH intravenous injection in iguanas, whereas its hypophyseal gene expression was reduced. In contrast, there was no response on THs concentration. Our results suggest an effect of TRH over GH modulation, independent of its role as a regulator of THs. Funded by CONACYT & PAPIIT-UNAM.

25.6

OVERWINTER CHANGES IN WEDDELL SEAL BODY CONDITION AND HORMONE PROFILES: IMPLICATIONS FOR PREGNANCY?

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Weddell seals (*Leptonychotes weddellii*) actively forage from Jan/Feb after completing their molt, until they haul-out for pupping the next Oct/Nov. In years 2010-2012, body composition was determined for post-molt (Jan/Feb; 53 non-reproductive) and pre-breeding (Oct/Nov; 31 non-reproductive, 17 reproductive) adult female Weddell seals. Overwinter changes in physiology varied by study year; however, animals were overall larger and in better condition (greater lipid stores, P<0.001) after the winter, in Oct/Nov. Twenty females were handled in both seasons, and exhibited the same increases in body mass and condition, regardless of their reproductive status the following year. Because changes in body mass are mediated by numerous endocrine factors, cortisol, thyroid hormones (T₃ and T₄), growth hormone (GH), and insulin-like growth factor (IGF)-1 levels were measured. Of these, T₄ and GH were significantly higher during the post-molt period in Jan/Feb (P<0.001), likely assisting with hair regeneration and protein sparing, respectively. Animals with higher T₄ levels in Jan/Feb were also significantly more likely to have a pup the following year (χ²=6.348, P=0.012), indicative of its role in embryo attachment and maintenance of early pregnancy. This study shows that the overwinter foraging period is critical for Weddell seals to gain condition, and that some hormones influencing fuel use during the molt may also impact reproduction the next year. Funding source: NSF0838892.

25.7

AMELIORATIVE CAPACITY OF QUERCETIN ON ALCOHOL AND NICOTINE INDUCED INFERTILITY IN EXPERIMENTAL RATS

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Over the past decades the male gonad has been exposed to various substances; among these substances are alcohol and nicotine. This study was designed to investigate the effect of quercetin on alcohol and nicotine induced infertility in male albino rats. Male rats (180-200g), randomly divided into nine groups of five rats each as follow. Group 1: control, group 2: corn oil 2ml/kg bw, group 3: quercetin (30mg/kg bw), group 4: alcohol (3g/kg bw as 25%v/v), group 5: nicotine (1.0 mg/kg bw), group 6: alcohol (3g/kg bw as 25%v/v) + nicotine (1.0 mg/kg bw), group 7: alcohol (3g/kg bw as 25%v/v) + quercetin (30mg/kg bw), group 8: alcohol (1.0mg/kg bw) + quercetin (30mg/kg bw), group 9: alcohol (3g/kg bw as 25%v/v) + nicotine (1.0mg/kg bw) + quercetin (30mg/kg bw). A marked significant decrease (P<0.05) in sperm profile (motility, count, mature sperm and morphology) was observed in sperm collected from the epididymis of the alcohol, nicotine and alcohol plus nicotine treated animals. Histological examination of testis sections in male albino rats treated with alcohol and/or nicotine for 52 days respectively revealed degrees of alteration when compared to control. However, lack of offspring after mating affirms the outcome of this study and this suggests that both alcohol and nicotine have antifertility activities with probable site of action as the testis. **Keywords:** Alcohol, nicotine, reproductive functions, oxidative stress, infertility.

25.8

WITHDRAWN

26.0: THERMAL PHYSIOLOGY

26.1

WATER BEFORE IONS? EARLY CHILL COMA ION BALANCE CHALLENGES THE CURRENT MECHANISTIC MODEL OF CHILL COMA

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At their critical thermal minimum, insects enter reversible paralysis known as chill coma during which water and ion balance is lost. It is currently thought that during coma, migration of Na⁺ from the hemolymph to the gut is followed by water, causing hemolymph ion concentrations (such as K⁺) to increase. Muscle depolarization due to increased hemolymph [K⁺] is suggested to explain chill coma paralysis. Chill coma

onset is a rapid process—however until now, water and ion balance had been assessed no earlier than six hours after coma onset. Using fall and spring field crickets (which differ in relative cold tolerance), we aimed to characterize water and ion balance in early chill coma to 1) verify whether hemolymph $[K^+]$ disruption explains paralysis, and 2) to generate hypotheses about mechanisms underlying loss of ion and water balance. Surprisingly, hemolymph K^+ imbalance in early chill coma did not account for muscular paralysis in either species. Furthermore, hemolymph $[Na^+]$ actually increased over the first hour of chill coma, before returning to control levels by six hours. Hemolymph water migrated to the gut in fall field crickets but not in spring field crickets, the latter of which maintained lower hemolymph $[Na^+]$ in general. We hypothesize that temperature-induced changes in epithelial permeability to water explains loss of water balance and subsequent ion balance during chill coma. Funding source: Natural Sciences and Engineering Research Council.

26.2

THERMAL SENSITIVITY OF MUSCLE PERFORMANCE IN THE CHILL SUSCEPTIBLE LOCUST, *LOCUSTA MIGRATORIA*

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Many insect species enter a state of neuromuscular paralysis when their body temperature is lowered to a critical limit but the physiological and cellular processes underlying this chill coma are largely unknown. Previous studies on locusts show that muscle force production is highly depressed at low temperature implicating impairment in cellular mechanism in the muscle per se. Aiming to determine these mechanisms we examined the thermal sensitivity of several events in the excitation-contraction-coupling process including: i) Passive membrane properties and propagation of electrical signals; ii) Intracellular Ca^{2+} regulation during muscle stimulation and iii) Ca^{2+} -affinity/sensitivity and maximum force of the contractile proteins. Thus far the data show that low temperature resulted in a marked depolarization of resting membrane potential, but had negligible effects on the passive membrane properties and muscle excitability, suggesting intact ability to excite and trigger action potentials in muscles at low temperature. Using skinned muscles fibers we show that similar contractile force can be obtained at high and low temperature provided that the muscle is stimulated with saturating doses of Ca^{2+} . However, preliminary results suggest that Ca^{2+} affinity is markedly decreased at low temperature. A reduction in Ca^{2+} sensitivity of the contractile filaments at low temperature could therefore explain loss of muscle function during chill coma. **Funding sources:** The research was funded by a Sapere Aude DFF-Starting grant (to J.O.) from The Danish Council for Independent Research | Natural Sciences and by grant (to A.F.) from the Faculty of Science and Technology of Aarhus University.

26.3

THE POTENTIAL TO RESIST CHILL COMA: HOW TEMPERATURE AFFECTS FLIGHT MUSCLE RESTING MEMBRANE POTENTIAL AND HEART RATE IN DROSOPHILIDS

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¹Biosci., Aarhus Univ., C.F. Moellers Alle 3, Bldg. 1131, Aarhus C, 8000, Denmark. Most insects enter a reversible coma at low temperatures. The critical temperature causing this chill coma (CT_{min}) is strongly correlated to species distribution. Chill coma is caused by loss of neuromuscular excitability, which is correlated with a depolarization of resting muscle membrane potential (V_m). Here, we examined the thermal dependence of muscle V_m in five *Drosophila* species with markedly different cold tolerances (CT_{min} range of 7.3 to -2°C). The V_m of the flight muscle depolarized distinctly when temperature was ramped from 20 to -3°C and, as hypothesized, this depolarization occurred at lower temperatures in the most cold tolerant species. This finding supports the notion that inter-specific differences in CT_{min} are related to a loss of membrane potential and muscle excitability. However, the estimated V_m at the CT_{min} varied among the species (~-54 to -30 mV), such that species-specific sensitivities to depolarization may also contribute to cold tolerance variation. We also examined the thermal dependence of the heart muscle, by examining heart rate (HR), using an Arrhenius break point analysis. HR declined with temperature in all species, but a breakpoint in the temperature/HR relationship and cessation of HR was found to occur at lower temperatures in more chill-tolerant species. Thus, inter-specific variability in cold tolerance is strongly associated with the ability to defend cardiac and skeletal muscle function. Funded by Sapere Aude, DFF.

26.4

VARIATION IN THERMAL TOLERANCE, HYPOXIA TOLERANCE AND METABOLIC RATE IN THE ATLANTIC KILLIFISH, *FUNDULUS HETEROCLITUS*

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According to the oxygen and capacity limited thermal tolerance (OCLTT) hypothesis, as temperature increases, aerobic scope declines, causing hypoxemia, decreased

organismal performance, and eventual loss of tolerance. Consequently, differences in thermal tolerance should be associated with differences in metabolic rate and hypoxia tolerance among individuals. We have addressed this relationship using Atlantic killifish, *Fundulus heteroclitus*, a species found along the east coast of North America. The southern subspecies, (*F. h. heteroclitus*) is more tolerant of high temperatures, has lower routine metabolic rate and better tolerance of hypoxia than does the northern subspecies (*F. h. macrolepidotus*), consistent with the predictions of the OCLTT hypothesis. To further examine these relationships, we tested thermal tolerance, hypoxia tolerance and metabolic rate in several populations of *F. heteroclitus* from along the coast. In general, higher thermal tolerance was associated with higher hypoxia tolerance and lower metabolic rate. We also collected four hundred individuals from an admixed population and examined trait associations within this population. There were no significant correlations between thermal tolerance, hypoxia tolerance and metabolic rate within this population, suggesting that at the individual level the expected relationships between the three measurements are not well supported, despite the patterns observed for between-population comparisons. Funded by NSERC.

26.5

SUB-LETHAL HEAT STRESS CAUSES APOPTOSIS IN COLD-ADAPTED ANTARCTIC FISHES

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The extremely cold-adapted fishes of the Southern Ocean possess some of the lowest upper thermal thresholds of any species. These fishes have evolved in the sub-zero waters of Antarctica for millions of years and possess profoundly cold-dependent physiologies. Some species have lost the traditional heat shock response but they nevertheless can respond to heat via modification of the transcriptome. Many of the genes that vary in expression during thermal stress in these fishes are related to the broad cellular stress response, a set of stress-related processes potentially including cell cycle arrest and/or apoptosis. Here, we used flow cytometry to examine the effect of sub-lethal heat stress on cell cycle progression. We also quantified apoptotic cells. Finally, levels of pro-apoptotic proteins and markers of cell cycle progression were also measured. Taken together, these studies indicate that even relatively mild heat exposures (2°C) can cause apoptosis in these cold-adapted and environmentally sensitive species. To our knowledge, this is the lowest upper thermal threshold for the initiation of apoptosis in any known species. The research was funded by the National Science Foundation's Office of Polar Programs.

26.6

BEHAVIOURAL REGULATION OF WATER LOSS IN FOUR AUSTRALIAN SKINKS

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As global climates warm, temperature induced activity constraints may not be the primary stressor facing ectotherms. An upward trend in air temperature will be accompanied by dramatic changes in spatial and temporal variability of precipitation. However, observational data are confounded by the correlation between high temperatures and high aridity, making it difficult to distinguish thermoregulatory from hydro-regulatory behaviour in the field. Recent evidence suggests that hydric condition may strongly influence activity patterns of lizards and that desiccation risk may be as (or more) important than temperature in restricting range and activity patterns. Here we use experimental manipulation to decouple temperature- and water-based constraints on activity, isolating the hydroregulatory behaviors of four species of *Egernia* skink (nocturnal and diurnal). Lizards are assigned to either a control or water restricted group and introduced to enclosures providing basking and retreat sites within a finely-controlled weather room which runs a daily temperature cycle ranging from 15C to 30C and alternating daily between 20% and 80% RH. Each lizard experiences a dry and a wet day while activity patterns and body temperatures are recorded. This study will better our understanding of the mechanisms of water regulation and the significance of hydric condition, allowing more robust predictions of the effect of changing climates on ectotherms.

26.7

THERMAL PREFERENCE DURING METABOLIC RECOVERY FROM ANOXIC HIBERNATION IN PAINTED TURTLES

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Spring emergence from anoxic hibernation is a critical time in which western painted turtles (*Chrysemys picta bellii*) in northern latitudes must undergo metabolic recovery from severe lactic acidosis accumulated during winter. Little is known about the physiology of metabolic recovery from overwintering, especially the potential role of temperature. To gain insight into the interaction between thermoregulation and metabolic recovery, cannulated turtles were implanted with iButtons to log internal body

temperature, submerged in anoxic water for 45 days at 3°C, and then placed in a thermal gradient ranging 3°C to 24°C to measure thermal preference until plasma lactate returned to control levels (<5 mM). Mean plasma lactate at the start of recovery was 67.0 mM (SEM=5.14, N=4). During recovery, turtles chose temperatures ranging 11–22°C with a mean of 19°C (SEM=2.52, N=4). The total time to recovery ranged from 49–265 hours with a mean of 111 h. There was an inverse relationship between peak lactate and temperature preference, which meant that turtles with higher lactates preferred colder temperatures. This study shows that hibernating turtles can tolerate large increases in environmental temperature, despite being held at 3°C for several months. A colder thermal preference with severe lactic acidosis is consistent with previous studies of reptiles and amphibians. This study was funded by the National Science Foundation.

26.8

THE ROLE OF AMBIENT TEMPERATURE ON TOXIN INGESTION BY A MAMMALIAN HERBIVORE

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Intrinsic factors like nutrients and plant toxins are known to influence feeding behavior in mammalian herbivores; however, far less is known about extrinsic factors like ambient temperature. There is growing evidence that plant secondary compounds (PSCs) become more toxic at higher temperatures due to decreased liver function. This phenomenon, known as temperature-dependent toxicity, could have critical implications for mammalian herbivores that must balance homeothermy with PSC detoxification in a warming environment. Here, we investigated the effect of ambient temperature on ingestion of PSCs from creosote bush (*Larrea tridentata*) by the desert woodrat, *Neotoma lepida*. We evaluated the maximum tolerable dose of creosote resin at warm (28–29°C), intermediate (25°C) and cool (21–22°C) temperatures. The maximum dose (g resin/day) for woodrats at 28°C was 32% less than woodrats at 22°C. We also tested the ability of woodrats to maintain body mass on a constant dose of creosote resin set below the maximum dose at the warm temperature. On this dose, woodrats at 29°C and 25°C lost >10% of their body mass compared to woodrats at 21°C. Our results demonstrate that increased temperatures limit PSC ingestion in an herbivorous rodent. Studying these interactions will advance the field of herbivore ecology and may enable more accurate predictions of herbivores' response to climate change. University of Utah IACUC 12-12010, NSF ISO to MDD 0817527, ASM & SICB GIAR to PK.

26.9

FROM FUR TO BLUBBER: EVOLUTIONARY AND ONTOGENETIC TRANSITIONS IN MAMMALIAN INSULATION

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One of the biggest challenges for endotherms in cold environments is thermoregulation—the maintenance of a high and relatively stable core body temperature. Here we describe consistent trends in the morphological and thermal properties of insulation in marine mammals, across both evolutionary and ontogenetic time scales. We compared morphological characteristics and thermal function of fur and blubber among pinniped groups (otariids, phocids, odobenids) in an evolutionary context, and examined the same traits across ontogeny in harp seals. As pinnipeds shift their thermoregulatory strategy from fur to blubber, hairs become shorter, flatter, and less dense, while the blubber layer increases in thickness and lipid content. The ontogenetic changes in thermoregulatory strategy among neonatal phocids mimic the evolutionary patterns observed among marine carnivores. Across species, differences in total insulation are influenced substantially by taxonomy, body size, and habitat. Within families and across ontogeny, however, animals exhibit a balance between fur and blubber that results in similar values for total insulation, even as thermal strategy shifts. Overall, we observed convergent evolutionary trends in thermoregulatory strategy that were recapitulated with ontogeny.

27.0: CONSERVATION PHYSIOLOGY

27.1

EXPLORING LOCAL ADAPTATION TO OCEAN ACIDIFICATION IN *MYTILUS CALIFORNIANUS* DURING SIMULATED UPWELLING EVENTS

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Local adaptation has been posited to play a large role in population successes in the future. As an upwelling region that experiences episodically high levels of pCO₂, the US West coast may be particularly impacted by ocean acidification. Our team at UCSB, as part of Ocean Margin Ecosystem Group for Acidification Studies (OMEGAS), has investigated this by integrating oceanographic data and physiological experiments on *Mytilus californianus* juveniles grown at nine sites in OR and CA. Mussels were collected live and placed into two newly designed multi-sample experimentation apparatus named Acidification Respirometry Chambers (ARCs) to measure the comparative respirometric effects of simulated upwelling events. Using a CO₂ gas-mixing system, we simulated four relevant ocean conditions based on oceanographic data (CA Relaxed – 18°C, 300 pCO₂; CA Upwelling – 12°C, 650 pCO₂; OR Relaxed – 14°C, 300 pCO₂; OR Upwelling – 8°C, 1200 pCO₂). When normalized for size, no significant site-specific differences in respiration were identified. Conversely, mussel respiration significantly decreases during periods of upwelling, independent of origin site, suggesting that *M. californianus* react as one population to the pressures of upwelling. However, the largest mussels came from high upwelling sites in OR, meaning that food availability may play a compensatory role and ultimately dictate survival whether the mussels originate in OR or CA. This work supported by NSF Award# 1220359.

27.2

HIGH PRESSURE NEUROLOGICAL SYNDROME IN SHALLOW-WATER MARINE INVERTEBRATES: IMPLICATIONS ON CLIMATE DRIVEN BATHYMETRIC RANGE SHIFTS AND ACCLIMATIZATION TO DEPTH

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In response to current rapid ocean surface warming, many marine ectotherms are shifting their latitudinal ranges to remain within their thermal scope. Bathymetric range shifts are also occurring in order to seek refuge from warming surface waters. Little is known about the ecological and physiological processes governing depth distribution limits in species: temperature and hydrostatic pressure are considered to be two dominant factors. Our study quantified differential gene expression in response to acute and long-term combined pressure and temperature exposures in the shrimp *Palaemonetes varians*: a model organism for thermal and pressure physiology. Increases in the transcription of genes coding for an NMDA receptor-regulated protein, an ADP ribosylation factor, β-actin, two heat shock protein 70kDa isoforms (HSP70), and glyceraldehyde-3-phosphate dehydrogenase (GAPDH) were found in response to elevated pressure. NMDA receptors have been implicated in pathways of excitotoxic damage to neurons and the onset of high pressure neurological syndrome (HPNS) in mammals. These data indicate that the sub-lethal effects of barotrauma are associated with transcriptional disturbances within the nervous tissue of crustaceans, and cellular macromolecular damage. Such transcriptional changes lead to the onset of symptoms similar to that described as HPNS, which may act as a limit to prolonged survival at depth. Further, we provide evidence of the synergistic effects of hydrostatic pressure and temperature. The interplay between these co-varying factors may be key to understanding the potential for shallow-water invertebrates to acclimatize to depth, and seek refuge from rapidly warming surface waters.

27.3

ADAPTIVE VARIABILITY IN SALINITY TOLERANCE EXPLAINS HABITAT VARIABILITY BETWEEN GENETICALLY DISTINCT POPULATIONS OF SACRAMENTO SPLITTAIL

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Sacramento Splittail (*Pogonichthys macrolepidotus*) are classified as a species of concern in California. Understanding the status of Splittail is complicated by the existence of two genetically distinct populations: the San Pablo (SP) population, a warm, brackish water spawner; and the Central Valley (CV) population, a cooler, freshwater spawner. To test the hypothesis that this habitat variability is driven by adaptive physiological differences in salinity tolerance between populations, we investigated the physiological response to salinity exposure of wild-caught adult and juvenile Splittail of both populations. Juveniles were exposed to salinities from <1 to 14 ppt for up to 14 days, and assessed for plasma osmolality (osmo) and ion levels, muscle moisture (MM) and gill Na⁺ K⁺ ATPase. At 11 ppt, plasma osmo did not differ between SP and CV populations. At 14 ppt, osmo remained elevated until 72 hours for the SP population and 7 days for the CV population. For wild adult Splittail, 14 ppt salinity exposure resulted in mortalities, and no differences were seen between the populations exposed to 11 ppt for 24 hours. The delayed recovery of mm and osmo for the juvenile CV relative to the SP population, suggests impaired water and ion balance in juvenile CV Splittail facing a salinity challenge. Such impairment suggests juvenile CV Splittail are less tolerant of saline waters, and supports our hypothesis of adaptive physiological differences in salinity tolerance.

27.4

TEMPERATURE AND HYPOXIA AFFECT SWIMMING ENERGETICS AND KINEMATICS OF BROWN TROUT (*SALMO TRUTTA*)

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The brown trout (*Salmo trutta*) is an economically and ecologically important fish native to the British Isles. In an era of accelerated climate change the cold and highly oxygenated conditions favoured by this species are being threatened. While many studies have described the effects of a single stressor on fish performance, few have examined the interacting effects of multiple stressors on multiple measures of performance. The aim of our study was to investigate the effects of temperature and oxygen on fish swimming kinematics and energetics. Rate of oxygen consumption (MO_2) was measured at three temperatures (14, 18 and 22°C) and two dissolved oxygen (DO) levels (100% and 70% air saturation) at slow and fast sustained swimming speeds (0.6 and 0.8 ms^{-1}). Our data show an increase in MO_2 with increased swimming speed and temperature. DO has no effect on MO_2 at low swimming speed. At higher swimming speed, however, MO_2 is lower in hypoxic conditions suggesting a switch to anaerobic swimming. Swimming kinematics also change, with both tail beat frequency and amplitude differing between hypoxic and normoxic conditions suggesting alternative, but potentially unsustainable locomotor muscle recruitment. These results suggest that the physiology of adult brown trout lacks the plasticity to accommodate changes in water temperature, velocity and oxygen levels in line with climate change predictions. This study was funded by the Natural Environment Research Council (NERC).

27.5

BEHAVIOURAL RESPONSES OF BLACK PERCH TO MARINE SYNECHOCOCCUS CYANOBACTERIA

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Black perch (*Embiotoca jacksoni*) are native to Coastal California where large-scale blooms of *Synechococcus* cyanobacteria are predicted to become more prevalent by the end of the 21st century. To date, the effects of prolonged exposure to this abundant genus of cyanobacteria on fish behaviour is unknown. However, it has recently been demonstrated that *Synechococcus* strain CC9311 can inhibit the growth of other cyanobacteria by the release of toxic compounds. In this study we investigated whether exposure to bloom-like concentrations of two *Synechococcus* strains, CC9311 and CC9902, alters behaviour of black perch using the light/dark test. Fish were exposed to *Synechococcus* strain CC9311 or CC9902 ($1.5 \times 10^6 \text{ cells ml}^{-1}$) or to control seawater in experimental aquaria for 3 days. Motion tracking software was used to track fish movement inside of the light/dark arena. We found significant effects of CC9311 on dark preference, velocity, immobility, and meandering compared to control fish. After testing we placed all groups into normal seawater and retested 3 days later. There were no longer any significant differences between groups, indicating that the effects of CC9311 are reversible. Funding Sources: NSERC (TH), Alfred P. Sloan Research Fellowship (BR2013-103) (MT).

27.6

BREVETOXIN METABOLISM AND PHYSIOLOGY USING FRESH-WATER TURTLES AS A MODEL TO MEASURE MORBIDITY IN ENDANGERED SEATURTLES

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Karenia brevis is a key dinoflagellate in harmful algal blooms (Red tides) which are increasing in frequency and duration worldwide. *K. brevis* produces neurotoxins known as brevetoxins (PbTx). PbTx binds to voltage-gated sodium channels (VGSC) which depolarizes cells and triggers apoptosis. PbTx also alters neurological and immune function and induces inflammation. Brevetoxicosis is difficult to treat in endangered sea turtles, however, as the physiological impacts have not been investigated and the magnitude and duration of PbTx exposure are unknown. Freshwater turtles (*Trachemys scripta*) are being used as a model system to determine toxin dynamics and physiological impacts. PbTx distributes to all organ systems in turtles, with higher levels in liver, bile and feces suggesting excretion via bile salts. Despite evident clinical impacts no significant lesions are found in animals exposed over 2 weeks to PbTx. As in mammals, PbTx in the turtle brain binds to VGSC, triggering Ca^{2+} influx that can be abrogated by the VGSC antagonist tetrodotoxin or the NMDA antagonist MK-801. However, turtle neurons are surprisingly resistant to PbTx. Cell viability

decreased in a dose dependent manner from 100-1000nM PbTx; the LC50 was significantly higher than in mammalian neurons. Understanding distribution, clearance, and effects of PbTx in these model turtles will allow us to design treatment strategies for sea turtles exposed to red tides. Funded by NOAA ECOHAB grant NA11NOS4780031.

27.7

AVIAN THERMOREGULATION IN THE HEAT: TOLERANCE TO HEAT STRESS VARIES GREATLY AMONG SPECIES

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We studied thermoregulatory performance of summer-acclimatized wild birds to heat stress in the deserts of Australia, North America and South Africa. We measured evaporative water loss (EWL), resting metabolic rate (RMR) and body temperature (T_b) continuously using ramped temperature profiles with increasing air temperatures in 40+ species, which included 10 orders with body size ranging from 7- 450g. We estimated the upper critical thermal limit (CTM) for each species by tracking T_b , EWL and RMR and activity when exposed to air temperatures ranging from 30-64°C. We found that birds from the orders Columbiformes (pigeons and doves) and Caprimulgiformes (nighthawks and nightjars) had the highest CTMs and were able to effectively thermoregulate at air temperatures as high as 60°C. Passerine birds, in contrast, showed a much more limited capacity for thermoregulation at high air temperatures and exhibited CTMs near 50°C. We found that thermal tolerance is primarily driven by the predominant pathway of evaporative heat loss, where birds that evaporate water from the skin or have a well-developed gular apparatus were most effective thermoregulators at high air temperatures. Body size was also a critical factor in determining the capacity of a species to tolerate high temperatures. This work greatly expands our knowledge of avian tolerance to heat and provides insights into how rapid warming and more intense heat waves may change avian distributions and community structure.

27.8

PHYSIOLOGICAL AND BEHAVIORAL RESPONSES TO ENVIRONMENTAL CHALLENGES IN THE WESTERN TERRESTRIAL GARTER SNAKE, *THAMNOPHIS ELEGANS*

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Populations of the western terrestrial garter snake, *Thamnophis elegans*, in northern California exhibit divergent ecotypes on the pace-of-life continuum and thus provide a natural laboratory to study the factors that influence responses to variable environments. Understanding intraspecific variation in this response is of central importance in predicting species' ability to survive in altered landscapes. Here, we present data from a number of recent experiments to quantify these differences: standard metabolic rate measured across a range of active temperatures, physiological indicators of a stress response following a capture/restraint protocol, and behavioral differences in lab-raised offspring under simulated predator attack. Through studies examining multiple aspects of the phenotype, we hope to illuminate the complex interplay between physiology and behavior that form an integrated response to environmental challenges. Funding for these studies was provided by the Iowa Science Foundation, PrairieBiotic Research, Inc., the Society for Integrative and Comparative Biology, and the Iowa Department of Natural Resources.

28.0: DEVELOPMENTAL PHYSIOLOGY

28.1

MOLECULAR BIOLOGICAL AND GENOMIC ANALYSIS OF THE DEVELOPMENTAL PHYSIOLOGY OF INTEGUMENTAL TRANSPORT

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For over a century, biologists have speculated that the highly dilute and complex mixture of dissolved organic matter in seawater might serve as an important nutritional resource for animals. Frequently, the metabolic demands of non-feeding stages of marine invertebrates cannot be accounted for solely by use of endogenous energy reserves; exogenous sources are required to balance energy budgets, even for developmental stages lacking digestive systems. Recently, a gene family for sodium/amino acid co-transport was identified in ectoderm of sea urchin embryos. Further comparative studies revealed that this family of genes is present in multiple species, representing many phyla. Using larval families generated in crosses of pedigreed lines of a bivalve, we have shown that amino acid transport capacity is genetically determined and related to differential growth. The developmental complexity of the regulation of

nutrient flux in these larvae was revealed by a genomic analysis that identified 23 putative amino acid transporter genes (*cf.* 4 in the same gene family in humans). The characterization of transporter gene families and their physiological functions have important implications for the study of integumental transport and for understanding the relationships between nutrition and other physiological processes that depend on the same gene families (e.g., chemoreception, osmoregulation, neurotransmission).

28.2 ONTOGENETIC CHANGES IN THE OSMOTIC STRESS RESPONSE OF BLUE MUSSELS

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Blue mussels, *Mytilus edulis*, are important members of intertidal communities and their distributions depend on the ability to respond to osmotic stress. Changes in global climate are predicted to alter near shore salinity, so tolerance throughout development will affect recruitment of mussels and the resilience of intertidal community structure. While the osmotic stress response (OSR) in adult mussels has been well studied, the larval stages—which are thought to be more sensitive to environmental stressors—are often ignored. Marine mussels are osmoconformers; during osmotic shock, they regulate intracellular free amino acid (FAA) pools to remain isosmotic to the environment. Taurine is an important osmolyte utilized by adult mussels during osmotic stress and may play a role in the larval OSR. Our aim is to correlate changes in the FAA pools of larval and juvenile mussels to variation in expression of a taurine transporter gene (*muTAUT*) during the course of a 48 h exposure to low salinity. Results from NMR spectroscopy show compositions of the FAA pool vary throughout ontogeny, though the fluctuations of taurine cannot be explained by variation in expression of *muTAUT*. Overall, our NMR and quantitative PCR results indicate that mussel larvae differ from their post-metamorphic counterparts. More attention should be focused on physiological responses to stress throughout development, as the larval OSR may play an important role in the stability of adult communities.

28.3 TRACHEAL SYSTEM STRUCTURE AND FUNCTION CHANGES WITHIN AN INSTAR IN THE CATERPILLAR, *MANDUCA SEXTA*

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As juvenile insects grow, body mass increases continuously, while sclerotized structures increase in size only after molting. Between molts, within an instar, hypoxia tolerance decreases. One hypothesis is that the tracheal system is sclerotized and increases in size only upon molting, leading to decreased O₂ supply. To test this hypothesis, we used synchrotron x-ray imaging to record tracheal system structure and function. Caterpillars at the beginning and end of each instar were placed in 21% and 1% O₂. Maximum and minimum diameters of major tracheae in the head and hind segments were measured, along with body size and mass. Body mass and length increased logarithmically from hatching to 5th instar, while head width increased only after molting. In support of our hypothesis, maximum tracheal diameter increased after molting, most significantly from the 4th to the 5th instar. These data demonstrate that the decreased hypoxia tolerance within an instar is due to the fixed size of major tracheae. To determine if the tracheal compression results in lower O₂ availability, we measured gene expression of hypoxia inducible factor 1- α and 1- β in caterpillars at the beginning and end of each instar. Together these results will provide a comprehensive view of how the insect respiratory system changes in structure and function within an instar. Funding provided in part by NSF IOS-0953297 and NIH NCRR P20RR015566.

28.4 EFFECTS OF CALCIUM AVAILABILITY ON GROWTH AND SURVIVAL OF *ACIPENSER FULVESCENS* IN EARLY LIFE STAGES

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Lake sturgeon (*Acipenser fulvescens*) have undergone population declines due in large part to anthropogenic habitat changes. To offset these impacts, stock enhancement with hatchery-raised juveniles is often utilized to restore natural populations. Therefore, determining optimal growth conditions in the early life stages is critical for this species. In this study, wild-caught adult lake sturgeon were induced to spawn in captivity, and fertilized eggs were incubated, hatched, and the larvae reared in four environments differing in calcium concentration (0.1, 0.2, 0.4, and 1.5 mM Ca²⁺). The impact of this altered water chemistry on hatching success, survival, and growth were measured at distinct developmental stages until initiation of exogenous feeding. Neither hatching success nor sustained larval survival to the feeding stage differed significantly due to environmental [Ca²⁺]. Protolavæ had similar mass and growth rates in all environmental calcium concentrations over the course of development. In contrast, larval total length was consistently higher in lower environmental

[Ca²⁺]. Surprisingly, lake sturgeon in calcium-limited environments maintained a higher condition index ($K = \text{mass} \times \text{total length}^{-3}$) beginning at 11 days post-hatch. These results suggest that environmental calcium availability may influence early life development and growth of *A. fulvescens*.

28.5 GENE EXPRESSION PATTERNS OF ALTERNATIVE DEVELOPMENTAL TRAJECTORIES IN EMBRYOS OF AN ANNUAL KILLIFISH

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Alternative developmental pathways are powerful adaptations for a species to survive unpredictable extreme environmental conditions. Embryonic diapause in the annual killifish, *Austrofundulus limnaeus*, is a state of arrested development and severe metabolic dormancy that may last for several months and thus allows embryos to survive in ephemeral habitats. However, development may also follow an alternative trajectory where individuals can instead "escape" entry into diapause and develop continuously until the completion of development. Early development along the two trajectories is morphologically indistinguishable, and the regulatory mechanisms that control phenotype are currently unknown. Incubation temperatures can induce cohorts of embryos to exclusively develop along either the diapause (20°C) or escape (30°C) trajectory. Using RNAseq, we have generated complementary transcriptomic profiles of mRNAs and small non-coding RNAs during embryonic development in *A. limnaeus*. Embryos committed to either the escape or diapause trajectories have unique gene expression patterns well before entry into diapause and before the phenotypes are morphologically distinct. Our findings suggest the existence of unique mRNAs and small RNAs that regulate the development of these alternative phenotypes and that support the drastically different metabolic demands of diapausing and developing embryos. This work is supported by a grant from the National Science Foundation.

28.6 EVIDENCE OF HYPOXIC METABOLIC PROGRAMMING IN DEVELOPING ALLIGATOR HEARTS

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Hypoxic stress during development can cause changes in tissue structure/function that persist into adulthood (developmental programming). Changes in mitochondrial function are likely to play a key role in this process, but very few investigations have explored this possibility. To this end, we investigated the effects of *in ovo* hypoxia on cardiac mitochondrial function in developing alligators. American alligator (*Alligator mississippiensis*) eggs were maintained at 30°C in either normoxia (21% O₂) or chronic hypoxia (10% O₂) and experiments were conducted on embryos at 90% development and in post-hatchlings from both incubation conditions raised in normoxia to 2 years of age (juveniles). Cardiac mitochondria were isolated using differential centrifugation and maximal aerobic capacity was assessed using an Oroboros microrespirometer. Mitochondria from embryonic alligators had a lower aerobic respiratory capacity than their juvenile counterparts, reflecting a developmental switch from glycolytic to aerobic energy production after hatching. Hypoxia did not affect mitochondrial function in embryonic alligators, but juveniles previously subjected to *in ovo* hypoxia had a greater aerobic capacity and mitochondria were more tightly coupled (more efficient) compared to normoxic juveniles. These results provide evidence of adaptive metabolic programming in developing alligators and highlight this animal as a novel alternative to mammalian models of fetal programming. This work was funded by The National Science Foundation (NSF) and The Wellcome Trust.

28.7 EFFECT OF THYROID HORMONE MANIPULATION ON ENDO-THERMAL DEVELOPMENT IN DOUBLE-CRESTED CORMORANTS (*PHALACROCORAX AURITUS*)

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In endotherms, thyroid hormones (TH) regulate development, metabolism and body temperature. In this study, we treated neonatal altricial Double-crested Cormorants with 3,5,3'-triiodothyronine (T₃) or the thyroperoxidase inhibitor, methimazole (MMI), to induce hyperthyroidism and hypothyroidism, respectively. We measured whole animal O₂ consumption (VO₂) and ventilation by flow through respirometry followed by measures of growth and size on days 4, 8, and 14 post hatching (dph). The VO₂ of T₃ MMI and control hatchlings on 8dph were higher than in 4dph hatchlings. Hatchlings of 4dph treated with MMI had significantly lower VO₂ than 4dph control, independent of body mass. Within treatment groups, there was a significant age effect on body mass, cardiac ventricle mass, liver mass, spleen mass, hematocrit, femur length, tarsus length, wing chord, and head and beak length from 4 to 14dph. Body mass of 14dph control hatchlings was significantly higher than in 14dph MMI

hatchlings. On 14dph, cardiac ventricle mass, femur length, tarsus length, wing chord and head and beak of MMI treated animals was significantly smaller than from the other two groups. Our data suggest that TH have a critical effect on growth and maturation of endothermy. We also provided evidence that decrease in TH delays attainment of endothermic capacity and growth in developing altricial avian neonates. Supported by NSF Grant IOS-1146758.

28.8

QUANTIFICATION OF LEFT VENTRICULAR FUNCTION IN EMBRYONIC CHICKENS (*GALLUS GALLUS DOMESTICUS*) AT 70% OF INCUBATION USING ULTRASONIC IMAGING

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Quantification of ventricular function in developing animals has been dependent on the transparency of the body wall and heart during ontogeny, as in amphibian, fish, and early stage bird embryos. The chicken embryo is a classic model for investigating features of cardiovascular maturation in endothermic vertebrates. A fundamental feature of the final two-thirds of incubation is that embryonic mass and arterial pressure rise continuously. Peripheral resistance also declines during this time, a consequence of increasing vascular collateralization and cross-sectional area during development. Our goal was to quantify ventricular function at 70% of incubation, a time-point when the chorioallantoic membrane vasculature and blood volume are maximal in embryonic development. We used the non-selective β -adrenergic receptor agonist isoproterenol to induce vasodilation and antagonist propranolol to decrease heart rate. Left ventricular output at rest and during pharmacological manipulation was quantified with ultrasound imaging of aortic diameter and blood flow velocity. These measures were coupled with chorioallantoic membrane arterial pressure measurements to establish the simultaneous relationship between afterload pressure and ventricular function. Our data suggest that pharmacologically increasing or decreasing vascular resistance alters ventricular function and output in the late stage embryonic chicken. Funded by NSF Career IBN IOS-0845741 to DAC.

29.0: METABOLISM: HYPOXIA AND ANOXIA

29.1

ERYTHROPOIETIN THROUGH EVOLUTION: A SPECULATIVE VIEW

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Reduced oxygen availability induces erythropoietin (Epo) gene expression that in turn elevates erythrocyte production and hence the blood's oxygen content. Interestingly and in contrast to Andean-Tibetans only moderately increase their hematocrit at high altitude. Obviously, evolution has selected a blunted erythropoietic response for Tibetans. At sea level, however, the hematocrit of men and mammals is set well below the maximal possible oxygen uptake capacity. We found that the optimal hematocrit in acutely and chronically Epo-treated mice is 0.58 and 0.68, respectively. However, these high hematocrit values are seldom reached in men and mammals keeping blood viscosity low, thereby reducing potential cardiovascular risks. As previously hypothesized by Carlos Monge, most men and mammals are sea level design and if so, one might speculate that oxygen-dependent Epo signal did not originally evolved to increase erythrocyte production. Indeed Epo exerts a neuro-protective function when the CNS is challenged with reduced oxygen supply (e.g. stroke). The fact that Epo/Epo receptor-like proteins are expressed in very low organisms including insects provides further (speculative) evidence that Epo might have evolved as a factor that influences/protects the CNS. We observed that increased Epo levels in the mouse brain augmented exercise performance in a non-erythroid manner. In other words: a single dose of recombinant human Epo demonstrated an unexpected improvement in maximal exercise performance that was independent of total hemoglobin mass, whole blood volume and cardiovascular parameters. We are testing if this observation translates into human volunteers.

29.2

EVOLUTION OF CYTOCHROME OXIDASE SUBUNIT 4-2 AS A HYPOXIA RESPONSIVE GENE

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The mammalian COX4-2 gene appears to have subfunctionalized into a role in hypoxic metabolism. Human and rodent COX4-2 protein possesses structural features that preclude allosteric repression of COX by ATP, characteristic of COX4-1, and the COX4-2 gene itself is hypoxia responsive. In other vertebrates, COX4-2 is more ubiquitous based upon mRNA levels in fish and reptiles. Fish COX4-2 expression appears to be restricted to specific cell types. The COX4-2 gene of lower vertebrates also lacks hypoxia responsiveness, based upon reporter genes constructed

from fish, amphibian and reptile orthologs. COX4-2 protein sequences suggest the disulfide bridge seen in the human and rodent orthologs would be precluded in other mammalian lineages and lower vertebrates, each of which lack the requisite CYS pair. The coordinating ligands of the ATP binding site are largely conserved across mammals and reptiles, but in *Xenopus* and fish, mutations may disrupt the ability of the protein to bind ATP at this site. Collectively, these results suggest that many of the genetic and structural features of COX4-2 that impart responsiveness and benefits in hypoxia may be restricted to the Euarhontoglires lineage that includes primates, lagomorphs and rodents. In other taxa the structure/function of COX4-2 may differ from the pattern seen in humans and rodents. Funded by NSERC-Canada.

29.3

CYTOCHROME C OXIDASE OXYGEN BINDING AFFINITY VARIES WITH HYPOXIA TOLERANCE IN INTERTIDAL FISHES

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The ability of mitochondria to generate ATP is heavily affected by oxygen (O_2) supply. In the marine intertidal, fish from the family Cottidae (sculpins) experience daily fluctuations in O_2 with species located in the higher intertidal oscillating between ~0 to 400% air saturation and show higher hypoxia tolerance, whereas subtidal species rarely experience hypoxic events and show lower hypoxia tolerance. This difference in their ability to tolerate low O_2 is reflected in modifications to various steps in the oxygen transport cascade, where hypoxia tolerant sculpins show higher gill surface area, better hemoglobin O_2 binding affinity, and an overall lower O_2 consumption rate. In this study, we focus on the mitochondria and the terminal protein of the oxygen transport cascade, cytochrome c oxidase (COX), to determine if O_2 use by mitochondria differ among sculpin species that vary in hypoxia tolerance. Hypoxia tolerant species showed higher COX oxygen binding affinity (measured as lower K_m for O_2) in both brain and liver, with brain tissue in all species overall having lower K_m O_2 than liver tissue. There were no differences in COX enzyme activity (V_{max}) between species. In an attempt to explain differences in COX K_m O_2 , we further investigate nucleotide and amino acid differences of the mitochondrial DNA-encoded catalytic subunit of COX. (Funding: NSERC).

29.4

CHARACTERIZING THE INFLUENCE OF ANOXIA EXPOSURE ON THE ISOLATED HAGFISH HEART

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Pacific hagfish, *Eptatretus stoutii*, can survive more than 36 hours of anoxia. Recent work indicates that these fish enter a metabolic suppression after 6 hours of anoxia. In the current study we integrated whole animal experiments with studies of isolated hearts to examine the response of the heart to anoxia exposure. We found that 36 hours of anoxia causes blood pH to decrease from 7.8 ± 0.03 to 7.0 ± 0.07 , hemoglobin O_2 saturation to decrease from $68.6 \pm 2.4\%$ to 0, total CO_2 to increase from 7.0 ± 0.8 mM to 7.8 ± 0.1 mM and hematocrit to increase from $10.4 \pm 0.8\%$ to $17.8 \pm 1.2\%$. We are currently utilizing calorimetry to determine how anoxia exposure influences metabolic heat production by the isolated, perfused heart and if this response is affected by acidosis. Preliminary results indicate that anoxia exposure at pH 7.8 causes an initial reduction in heat production and that death occurs at approximately 12 hours of anoxia. In addition, exposure of the anoxic heart at pH 7.8 to pH 7.0 causes an increase in heat production. Finally, heat production by the isolated heart increases to initial levels upon re-oxygenation. Together these results demonstrate that the hagfish heart is able to withstand extended periods of anoxia and that the metabolic suppression is blunted at low pHs. Supported by NSERC Canada and Canada Research Chair Program.

29.5

DEEP SEQUENCING OF THE HEPATOPANCREAS TRANSCRIPTOME REVEALS NEW ISOFORMS OF HEMOCYANIN AND THEIR REGULATION IN RESPONSE TO LOW O_2 /HIGH CO_2 IN THE PACIFIC WHITELEG SHRIMP, *LITOPENAEUS VANNAMEI*

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Acclimation to low O_2 in many organisms involves regulation at the level of the transcriptome. Previous microarray results suggest that the hypoxia-specific transcriptomic signature of the *L. vannamei* hepatopancreas is reduced or reversed with the addition of environmental CO_2 . Here we used high throughput RNA sequencing to explore the involvement of new isoforms of hemocyanin (Hc) in the CO_2 response. Hepatopancreas mRNA of juvenile *L. vannamei* exposed to air-saturated water (normoxia),

low O₂, or low O₂/high CO₂ for 4 or 24 h, was pooled, sequenced (HiSeq 2500) and assembled (Trinity: 46,049 contigs) to create a deep reference transcriptome (1642X coverage). Annotation of the assembly revealed sequences encoding the single large and small Hc subunits, two previously undescribed full-length isoforms of the large subunit, and 12 partial sequences. mRNA of individual shrimp was sequenced (6/treatment); resulting reads were quantified (eXpress) and regulated genes identified from pairwise comparisons at each time (DESeq2). This analysis confirmed that CO₂ had an antagonistic effect on the transcriptomic response to low O₂. Only 1 Hc (partial c143731) was significantly upregulated in low O₂ (24 h) with fold change (FC) = 8.1 compared to normoxia; the response was blunted (FC = 6.2) by high CO₂. We are exploring the importance of these novel full length and partial isoforms to the structural and functional response of Hc in low O₂ alone and with high CO₂ (NSF IOS-1147008).

29.6

ANOXIA-RESPONSIVE SMALL RNA GENE EXPRESSION IN ANNUAL KILLFISH EMBRYOS

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Embryos of the annual killifish *Austrofundulus limnaeus* survive without oxygen for over 100 days at their most tolerant stage, but only survive a matter of hours at their least tolerant stage. Studying the changes in their cellular response to anoxia may help build the foundation for human heart attack and stroke treatment. Recently, small non-coding RNAs (such as microRNAs) have been found to play a role in metabolic depression and cellular response to hypoxia, by altering gene expression. In this study, we examined small RNA expression profiles in *A. limnaeus* embryos exposed to anoxia and recovery at various developmental stages. We identified 1000s of highly differentially expressed RNA transcripts. Many of these match sequences of hypoxia-responsive miRNAs described in other organisms, while many transcripts may be miRNAs unique to *A. limnaeus*, or other small RNAs with important cellular function. At certain stages of development, we have discovered highly differentially expressed and abundant small RNAs that originate from the mitochondrial genome, particularly from tRNA genes. Recent literature indicates tRNA-derived fragments can perform microRNA-like function as gene silencers. Understanding the expression patterns of these fragments over development and in response to anoxia, as well as their cellular location and function, may alter and clarify our understanding of the cellular stress response. Funding: NIH R01 HL095454, NSF IOS-1354549, NSF DGE-1057604, Sigma Xi GIAR award.

29.7

IS ANOXIA AND ROS-MEDIATED GABA RECEPTOR INHIBITORY SHUNTING IN TURTLE CORTICAL NEURONS MEDIATED BY TONIC, FAST OR SLOW PHASIC CURRENTS?

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The freshwater painted turtle brain is a working model of a vertebrate anoxia-tolerant brain that can be exploited to elucidate the underlying protective mechanisms. We have previously shown that excitatory glutamatergic signaling in the cerebral cortex is inhibited by 50% and GABAergic signaling increases by 100% in response to anoxia. The increased GABA levels activate pyramidal neuron GABA-A receptors (G-AR) forming a shunt that prevents depolarization and generation of an action potentials. While the source of GABA is likely stellate interneurons it is unclear which type of stellate neuron it is, or which pool of G-AR is activated. Here we use a pharmacological approach to deduce the relative contributions of phasic fast synaptic, slow peri-synaptic and tonic extrasynaptic G-ARs. Since reactive oxygen species (ROS) levels decrease with anoxia we also investigate the impact of ROS scavengers on G-AR activation. Using whole-cell and perforated patch clamp techniques we determined that both anoxia and ROS scavenging causes: whole cell conductance to increase from 4.5 to 6.6nS; G-AR reversal potential to move to membrane potential; and slow peri-synaptic G-AR mediated currents to double in amplitude from 238 to 450pA. Bicuculline was used to quantify tonic G-AR currents which doubled with anoxia and ROS scavenging from 17 to 40 pA and phasic fast currents also doubled from 30 to 60pA. We conclude that all three G-ARs are responsible for the anoxia-mediated shunting current.

29.8

CAN KETONE BODIES PROTECT THE HEART AGAINST THE EFFECTS OF CHRONIC HYPOXIA?

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Sustained exposure to hypobaric hypoxia is associated with loss of skeletal muscle but also fat mass. Consequent generation of ketone bodies has been hypothesized to offer metabolic protection for vital organs, acting as both metabolic substrates and modulators. In this regard, they modulate the hypoxic response via HIF1 α , decrease ROS accumulation and increase mitochondrial efficiency in the brain. Exploration of such effects in other tissues has, however, been limited. We hypothesized that hypoxic rats would exhibit elevated ketone body production in the liver and enhanced use of ketone bodies by the heart, whilst their metabolism in skeletal muscle would decline. To test this hypothesis we exposed rats to 10% O₂ for 4 weeks and compared them with normoxic controls fed *ad libitum* and normoxic control pair-fed to account for the anorexic effects of hypoxia. Our results indicate an increase in the production of ketones in the liver of hypoxic rats and subsequent oxidation of ketones in the heart with no change in skeletal muscle. Lipid oxidation was not affected in the liver but declined in both cardiac and skeletal muscles compared to controls. Cardiac tissue from hypoxic rats also had higher oxidative phosphorylation rates when using ketone bodies and had higher glutathione peroxidase activity. Our results support the hypothesis that ketones utilisation is enhanced in cardiac tissue during chronic hypoxia, and may exert protective effects.

30.0: OSMOTIC AND ION REGULATION: SALINITY, OSMOLYTES, AND pH

30.1

PHYSIOLOGICAL AND FUNCTIONAL GENOMIC MECHANISMS OF SEAWATER TO FRESHWATER TRANSITIONS IN THE ALEWIFE

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Colonization of freshwater by seawater organisms has led to substantial adaptive diversification and radiation, especially among fishes. Landlocked, freshwater-only populations of Alewife (*Alosa pseudoharengus*) have independently evolved multiple times from anadromous populations that migrate from seawater to freshwater to breed. This system enables us to test whether predictable changes in osmoregulatory physiology and its genetic bases have accompanied the transition to freshwater. Through a series of salinity challenge experiments, we show that landlocked forms have predictably evolved increased tolerance of freshwater, which has traded off for reduced tolerance of and osmotic balance in seawater. To characterize divergence in gene expression that underlies these different physiological responses to salinity challenges, we sequenced and *de novo* assembled gill transcriptomes. The expression patterns of several genes that function in osmoregulation have similarly differentiated in multiple landlocked populations relative to anadromous expression patterns. However, most genes showing parallel divergence were not known to be osmoregulatory, suggesting that a variety of pathways have been under selection in the transition to freshwater. This analysis reveals multiple dimensions of adaptation to exclusively freshwater habitats. Funding provided by MVAO foundation.

30.2

FROM COMPARATIVE INTESTINAL TRANSCRIPTOME ANALYSIS TO CHARACTERIZATION OF TRANSPORTERS LINKING NUTRIENT ABSORPTION WITH ION AND ACID-BASE REGULATION

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Tilapias are a group of freshwater fishes, however, some of these species are tolerant to high salinities. Differences in salinity tolerance were observed between two closely related species, the highly tolerant *O. mossambicus* and the more sensitive *O. niloticus*. The anterior- and posterior-intestine transcriptome of those two species, acclimated to seawater and freshwater, was sequenced using the Illumina Hi-Seq, following by gene expression and gene-ontology (GO) analyses. The results indicate a species-specific salinity-dependent gene expression patterns in the anterior intestine. Overall, between 182 and 404 genes were significantly up-regulated in either seawater or freshwater, including 70 genes with reversed salinity response, up-regulated in one species and down-regulated in the other. From these genes we have focused on proton- and sodium-dependent peptide and amino acid transporters. Seawater and freshwater-acclimated fish were sampled at three time points after feeding (6, 24, 72 h), representing different digestive stages. The PepT1a, PepT1b, PepT2, B⁰AT1, and B⁰AT1 genes were analyzed for their expression and protein localization along the intestine, and their association with sodium and proton transporters. We found differential expression and localization of these genes along the intestine, correlated them with the different digestive stages, and with salinity adaptation physiology. This research was funded by BARD.

30.3

THE REGULATION AND FUNCTION OF POLYAMINES IN *FUNDULUS* SPECIES DURING SALINITY STRESS

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¹Biological Sci., Louisiana State Univ., 216 Life Sci. Bldg., Baton Rouge, LA, 70803. Polyamines are a family of low molecular weight organic cations produced in part by the coordinated actions of arginase II (Arg II) and ornithine decarboxylase (Odc). Although little is known of their function in fish, polyamines have been implicated in diverse physiological processes. We describe a possible role of polyamines in hypo-osmotic tolerance in the Gulf killifish, *Fundulus grandis*, and the mummichog, *F. heteroclitus*, two species known to tolerate large fluctuations in environmental salinity. Adult fish were reared in 5 ppt water for at least 1 month and acutely transferred to 0.1, 0.5, 1, 2, and 5 ppt water. Fish were randomly sampled at 6 h, 1 d, 3 d and 7 d post-transfer. We have shown that relative Arg II and Odc mRNA levels and Odc protein activity are highly up-regulated in the gills after hypo-osmotic exposure. Hypo-osmotic exposure also increases the concentrations of polyamines, including putrescine, spermidine, and spermine. Although gill putrescine levels remain elevated throughout the first 7 days post-transfer to fresh water, concentrations of spermidine and spermine decrease with time, suggesting these compounds are regulated catabolically. This presentation will discuss the role of polyamines apoptosis during salinity-induced gill remodeling and osmoregulation during salinity stress. This research was supported by funding from the National Science Foundation (EF-0723771).

30.4

REGULATION OF ORGANIC OSMOLYTE CONCENTRATION IN TISSUES OF EURYHALINE TELEOSTS

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¹Animal Sci., Univ. of California, Davis, One Shields Ave., Davis, CA, 95616. To cope with hyperosmotic stress cells accumulate compatible organic osmolytes, which replace harmful inorganic electrolytes while maintaining homeostasis of cell volume and intracellular ionic strength. Previous work has established that myo-inositol is an important compatible organic osmolyte in several tissues of adult Mozambique tilapia (*Oreochromis mossambicus*). Here we investigate the regulation of the biochemical pathway that is responsible for myo-inositol biosynthesis. We find that the activity of the key enzymes of this pathway (myo-inositol phosphate synthase -MIPS and inositol monophosphatase - IMPase) is directly regulated by the ionic milieu, in addition to mRNA and protein abundance regulation. Furthermore, we show that two enzymes, MIPS and IMPase, are regulated in multiple tissues of euryhaline fish (brain, gills), in multiple species of euryhaline fish (including three-spined sticklebacks, *Gasterosteus aculeatus*), and in multiple developmental stages of euryhaline tilapia. Of interest, immunohistochemistry reveals that major target tissues for accumulation of myo-inositol during salinity stress differ in larvae from those in adults. We conclude that myo-inositol is a key (but not the only) compatible organic osmolyte that enables euryhaline teleosts to cope with salinity stress. Supported by NSF grant IOS-1355098.

30.5

CELLULAR MECHANISMS FOR ACID/BASE SENSING AND REGULATION IN ELASMOBRANCH GILLS

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The functional specificity of the elasmobranch gill makes it an excellent model for the study of acid/base (A/B) regulation at the cellular and systemic levels without interference from other ion-transporting processes. We immunolocalized the anion-exchanger pendrin to base-secreting V-H⁺-ATPase (VHA)-rich cells in gills from leopard shark (*Triakis semifasciata*). In starved sharks, VHA signal was mostly cytoplasmic and pendrin was present at the apical pole. However, in fed sharks VHA translocated to the basolateral cell membrane and pendrin to the apical membrane, respectively, indicating upregulation of acid absorption and bicarbonate secretion. To further characterize this mechanism, we added a cell-permeable cAMP analogue to isolated and cultured gill cells, which induced the translocation of VHA to the cell membrane. The bicarbonate-sensitive A/B sensor soluble adenylyl cyclase (sAC) was highly abundant in both base-secreting and Na⁺/K⁺-ATPase-rich acid-secreting cells. Enzymatic assays revealed bicarbonate-stimulated and KH7-sensitive cAMP production in gill homogenates, indicative of sAC activity. In addition, sAC activity was present in nuclei isolated from gill tissue. Finally, we found that both acid- and base-secreting cells express high amounts of transmembrane adenylyl cyclases (tmACs). We propose a model whereby blood/acid base status is sensed by the combined action of sAC and G-Protein Coupled Receptors/tmAC, with a potential role for nuclear sAC in the regulation of gene expression. This research was partially supported by a fellowship from the NIH Marine Biotechnology Program Grant (GM067550) to JNR.

30.6

OCEAN ACIDIFICATION STIMULATES RESPIRATORY PLASTICITY IN THE ESTUARINE RED DRUM (*SCIAENOPS OCELLATUS*)

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Anthropogenic CO₂ release is causing unprecedented changes in the oceanic carbonate system. These environmental changes are too rapid for many species to adapt through evolutionary processes; however, adaptive capacity may be enhanced by phenotypic plasticity. This study examined the effects of ocean acidification (OA) on red blood cell and branchial gas exchange parameters using a combination of molecular, biochemical and morphological methods. OA had no impact on red blood cell parameters after 24 and 72 h versus control, while *slc4a1* expression was significantly decreased by 14 days of exposure. Follow up experiments using anemia supported the finding that red blood cells do not show plasticity in response to respiratory stress. In contrast, branchial diffusion distance was reduced by 30% after 14 days of exposure and RhAG and RhCG1 expression doubled after 72 h, both of which could increase CO₂ excretion rates at the expense of salt and water balance. Interestingly, branchial Na⁺, K⁺ ATPase activity doubled after 14 days of exposure to OA; no change was observed in CFTR or NKCC1 expression. These results suggest that respiratory plasticity may enhance the adaptive capacity of fish in response to OA; however, the osmoregulatory trade-offs may increase baseline metabolic costs. All experiments were performed in accordance with the University of Texas at Austin Institutional Animal Care and Use Committee. Funding for this project was provided by NSF (EF 1315290).

30.7

CHANGES TO INTESTINAL TRANSPORT PHYSIOLOGY AT VARYING LEVELS OF HYPERCAPNIA IN THE GULF TOADFISH (*OPSANUS BETA*)

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Marine teleosts defend blood pH during hypercapnia with a well-documented response, which includes the elevation of HCO₃⁻. In contrast to regulatory responses at the gill to achieve net base retention, hypercapnia leads to increased base secretion in the intestine. This base loss is thought to occur through pathways previously implicated in osmoregulatory processes, where chloride is exchanged for bicarbonate to aid in water absorption. The objective of this study was to expose gulf toadfish to various CO₂ levels (0.25, 0.5, 1, 2% CO₂) to examine changes and identify trends that occur in intestinal transport physiology, intestinal calcium carbonate production rates, and carbonate cation composition during hypercapnia. Results of this study indicate a close relationship between the increase in intestinal bicarbonate secretion and the rate of chloride uptake, suggesting increased anion exchange with increasing hypercapnia. Reduced fluid Mg²⁺ (mM) and increased carbonate production appear to be characteristic at high CO₂ levels, changes that could impact carbonate composition and thus solubility. Impacts of CO₂ on other ions and the relationships between ion concentrations in plasma, rectal fluid, and calcium carbonates will also be discussed. This research was support by an NSF grant through Dr. Martin Grosell (PI) and through a NSF Graduate Fellowship awarded to Rachael Heuer.

30.8

CRAB PROTEOMICS: RESPONSES TO SIMULTANEOUS LOW PH AND TEMPERATURE STRESS

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We investigated the proteomic responses of gill tissue of the spider crab (*Hyas araneus*) and the intertidal porcelain crab (*Petrolisthes cinctipes*) to simultaneous temperature and pH stress. Both species showed a decrease in tyrosine metabolism, while simultaneously changing the abundance of lectins and serine proteases, which are thought to lead to the conversion of pro-phenoloxidas (including hemocyanin) to phenoloxidas. These catalyze the synthesis of quinones and melanin from tyrosine. Quinones are involved in the sclerotization of the arthropod cuticle and both species showed abundance changes in a number of cuticle proteins. The porcelain crab also showed changes in the abundance of proteins that are involved in the excretion of ammonium, and thus the excretion of proton equivalents, across the gill tissue. This response to low pH was dependent on the immersion/emersion and temperature conditions animals experienced. Changes in the abundance of proteins involved in the urea cycle might indicate that it too is involved in the excretion of bicarbonate ions and thus protons. Low pH also decreased the abundance of chaperones of the endoplasmic reticulum. In general, crustacean gill tissue changed a number of proteins involved in cuticle structure and possibly the passive ion transport properties of the api-

cal side of the gill epithelium. These changes were accompanied by changes in the active transport of ammonium and the urea cycle (funded by NSF EF-1041227).

31.0: THE CHALLENGE OF TEACHING PHYSIOLOGY IN A CHANGING ENVIRONMENT: INNOVATION AND RESOURCES

31.1

VISION AND CHANGE UPDATE: PROGRESS IN IMPLEMENTING REPORT GOALS IN UNDERGRADUATE BIOLOGY EDUCATION

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The 2011 AAAS Vision and Change report serves as a useful platform for discussions about how life science departments at diverse institutions across the country can improve life science education for their undergraduate students. This talk will explore the impact of national conversations about life science education reform on curricula, educators and plant science education, and examine novel institutional strategies for implementing the recommendations of the Vision and Change report. AAAS, Vision and Change in Undergraduate Biology Education: A Call to Action. Eds. Carol Brewer and Diane Smith. AAAS, Washington, D.C. Available at: <http://visionandchange.org/files/2011/03/Revised-Vision-and-Change-Final-Report.pdf>.

31.2

TEACHING AND LEARNING BY INQUIRY

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In this readily available information age, student centered learning is known to help students develop competency in required disciplines and be better able to manage their lifelong learning goals. Fortunately, new and revised pedagogy is available for teachers (even with large enrollment classes) to help their students develop critical thinking skills and construct meaningful knowledge and competency in areas. This presentation will define inquiry and describe why building new knowledge should be the goal of teaching. Ideas will be shared for beginning to change didactic lectures into active learning experiences and for incorporating various student centered activities into laboratory and lecture sessions. Specific examples of learning by experimentation, team-based learning, problem-based learning, case-based learning, project-based learning and discussion-based learning will be described. Layman, JW, Inquiry and Learning. New York: The College Board, 1996. Sutman, FX, Paper presented at annual meeting of AAAS, Philadelphia, PA, 1998. Michaelsen, LK, AB Knight, and LD Fink, Team-based Learning: A Transformative Use of Small Groups in College Teaching. Stylus, Sterling, VA, 2002. *Advances in Physiology Education* (advan.physiology.org) and Life Science Teaching Resources Community (www.lifescitrc.org).

31.3

ASSESSING STUDENT LEARNING AFTER CONVERTING TO INQUIRY

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We compared gains in student content learning over a 10-yr period in which an introductory biology laboratory curriculum was changed to inquiry. As the level of inquiry increased, student learning gains on content exams trended upward. Students who participated in 14-wk-long inquiry laboratories outscored their peers in 7-wk inquiry and 1-wk traditional labs on MCAT exam questions (scores of 64.73%, 61.97%, and 53.48%, respectively, $p < 0.01$). End of semester surveys conducted when traditional 1-wk laboratories ($n=167$ students) were used had low response rates and predominately negative opinions (only 20% of responses were positive), whereas those conducted after 7-wk ($n=543$) or 14-wk ($n=308$) inquiry laboratories had high response rates and 71% and 96% positive reviews, respectively. In an assessment of traditional content coverage in courses, three indexes were averaged to calculate traditional forms of coverage and showed a decrease by 44% over the study period. We believe that the quantitative and qualitative data support greater student-driven inquiry in the classroom laboratory, which leads to deeper learning in fewer topic areas (less teaching) and can reap gains in scientific thinking and fundamental understanding applicable to a broader range of topic areas (more learning) in introductory biology.

31.4

CONSTRUCTING CONCEPT INVENTORIES AND ASSESSING THEIR VALUE

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Teaching and learning of physiology involves the development of enduring understandings and ability to apply core concepts. Backwards design can guide course and curricular design by first identifying the core concepts that will shape learning goals. Second, backwards design requires one to determine what evidence (assessment) can demonstrate achievement of goals. Finally learning activities are designed that align with goals and assessments (Dirks et al. 2014). Learning for conceptual understanding requires conceptual assessment tools, including concept inventories (CIs). Concept inventories are validated instruments that assess student understanding and application of conceptual knowledge. CIs been used to assess student learning gains, diagnose particular student misconceptions, and to evaluate instructional innovations and curricular reform. CI development requires an understanding of the underlying conceptual framework that experts apply implicitly and knowledge of common student misconceptions that are used in the construction of a CI assessment tool. Homeostasis is one of the fundamental core concepts in physiology (Michael & McFarland 2011). A physiology CI for homeostasis, based on a conceptual framework and student misconceptions, is in development. Comparative physiology provides a particularly effective opportunity for teaching, learning and assessment of conceptual understanding of homeostasis. *NSF DUE-1043443*. Dirks, Wenderoth & Withers. 2014. *Assessment in the College Science Classroom*. WH Freeman. Michael, J. & McFarland, J. 2011. The core principles ("big ideas") of physiology: results of faculty surveys. *Adv Physiol Ed*. 25:336-341.

32.0: MOLECULAR AND PHYSIOLOGICAL FEATURES OF ANIMAL DIAPOUSE

32.1

AMPK BUFFERS ADVERSE EPIGENETIC CHANGE AND CONSEQUENT TRANSGENERATIONAL REPRODUCTIVE DEFECTS FOLLOWING ACUTE ENERGY/NUTRIENT STRESS

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Environmental change is a significant factor in the evolution of selective adaptations and thus probably exerts a major influence on much of our present day morphology and physiology. Stresses that arise due to adverse environmental conditions have been shown to affect chromatin modifiers that can in turn cause adaptive or maladaptive changes in gene expression. These chromatin modifications can be transient, they can persist throughout the lifetime of the organism, or even span multiple generations. Following embryogenesis emergent *C. elegans* larvae will arrest development in a quiescent diapause-like state until they encounter an food source that will permit the onset of postembryonic development. In the absence of an adequate energy/nutrient source the L1 larvae can survive two weeks in this quiescent state. We have found that AMP-activated protein kinase (AMPK) is critical for survival during this period. In addition, AMPK is required to buffer modifications to the chromatin landscape to ensure that gene expression remains inactive in the primordial germ cells during the quiescence. In its absence, chromatin marks are changed and consequently compromise the reproductive fitness of the starved animals following recovery, in addition to affecting the subsequent generations that never experienced the initial energy/nutrient stress. Our data suggest that AMPK impinges on critical targets that affect gene expression to coordinate the initiation of germ line development with environmental and physiological constraints.

32.2

INSULIN SIGNALING AS A KEY REGULATOR OF INSECT DIAPOUSE

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The insulin signaling pathway was first suggested as a key player in regulation of the dauer state in the nematode *Caenorhabditis elegans*, and it now appears to be equally important in regulating the equivalent dormant state, diapause, in insects. In experiments with *Culex pipiens*, the mosquito that vectors West Nile virus, we have shown that the arrested ovarian development of adult females, a characteristic of the overwintering diapause in adult insects, is the consequence of a shut-down in the insulin signaling/FOXO pathway. Insects produce numerous insulin-like peptides (ILPs), but not all are involved in the diapause response. ILP-1 appears to be the peptide in this family that is most closely linked to diapause in *C. pipiens*. ILP-1 acts upstream of juvenile hormone (JH) and contributes to shutting down the synthesis of JH that is essential for ovarian development. This mediation of JH synthesis is linked to suppression of allatotropin, the neuropeptide that promotes JH synthesis. Diapause can be simulated in nondiapausing mosquitoes by using RNAi to knock down expression of the genes encoding either ILP-1 or the Insulin Receptor, and the knock down can be rescued by application of exogenous JH. Fat accumulation and enhanced stress tolerance, features that also characterize the diapause state, appear to be linked to the insulin pathway through the action of a key transcription factor, FOXO. This pathway thus appears to generate many features that characterize the diapause phenotype. Similar results observed in *Drosophila melanogaster* and the flesh fly *Sarcophaga*

crassipalpis suggest a common reliance upon insulin signaling for regulation of diapause in insects. Reference: Sim C. and Denlinger DL (2013) Insulin signaling and the regulation of insect diapause. *Frontiers in Physiology* 4:189 (doi:10.3389/fp-hys.2013.00189).

32.3

THE ROLE OF MATERNAL PROVISIONING AND MICRORNA REGULATION OF DIAPAUSE IN THE ANNUAL KILLFISH *AUSTROFUNDULUS LIMNAEUS*

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Annual killifish survive in ephemeral ponds by producing embryos that may enter diapause at three distinct developmental stages (diapause I, II, and III). Diapause II is a profound state of dormancy and is likely key to survival of embryos through the dry season. However, escape embryos skip diapause II and develop directly to diapause III. Entrance into diapause II is an alternate developmental pathway controlled by both maternal inputs and the temperature experienced by free-living embryos. Embryos developing at 20°C enter into diapause II while those developing at 30°C develop as escape embryos. Fertilized embryos destined to develop along the escape trajectory are provisioned with a unique gene expression signature for both small and large RNA molecules, and have increased levels of insulin-like growth factor II protein. Incubation at 30°C induces an increase in the expression of small RNA transcripts that are known to block expression of maternal mRNA transcripts that control early development. It appears that entrance into diapause is controlled maternally, but can be altered by zygotic small RNAs in response to environments conducive to direct development. This work identifies genetic mechanisms supporting diapause and bet-hedging strategies in annual killifish development. Reference: Podrabsky JE, Garrett IDG, Kohl ZF. 2010. Alternative developmental pathways associated with diapause regulated by temperature and maternal influences in embryos of the annual killifish *Austrofundulus limnaeus*. *J Exp Biol* 213:3280-3288.

32.3

BIOENERGETICS OF DIAPAUSE IN A CRUSTACEAN EXTREMOPHILE

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Gastrula-stage embryos of *Artemia franciscana* (brine shrimp) display developmental arrest and a respiratory depression of over 99% upon release from the adult female as they enter diapause. A substantial contributor to inhibition is a restriction of oxidative substrate to the mitochondrion that involves an orchestrated interplay at multiple enzymatic sites including trehalase, hexokinase, pyruvate kinase and pyruvate dehydrogenase. Proton conductance across the inner membrane is not diminished in mitochondria from diapause embryos versus post-diapause, and thus mitochondrial membrane potential ($\Delta\psi$) is likely compromised because respiration of diapause embryos is depressed below that required to compensate for proton leak. Under such conditions, the F_1F_0 -ATP synthase could reverse and deplete all cellular ATP. ATP decreases during diapause, but significant amounts remain (ATP:ADP ratio = 1.3). We predict that the hydrolysis activity of F_1F_0 -ATP synthase is blocked during diapause by the inhibitor protein IF1, which as previously shown binds to the catalytic domain when $\Delta\psi$ is low and matrix pH is acidic. Inhibition of affinity-purified ATP synthase from *A. franciscana* with bovine IF1 (residues 1-60) shows a strong pH-dependency. Work with IF1 from *A. franciscana* is underway to clarify mitochondrial stasis during diapause. (NSF grant IOS-0920254, NIH grant 2 RO1 DK046270-14A1). References: Patil Y, Marden B, Brand MD, Hand SC (2013) Metabolic downregulation and inhibition of carbohydrate catabolism during diapause in embryos of *Artemia franciscana*. *Physiol Biochem Zool* 86: 106-118. Bason JV, Runswick MJ, Feamley IM, Walker JE (2011) Binding of the inhibitor protein IF1 to bovine F_1 -ATPase. *J Mol Biol* 406: 443-453.

33.0: NEW PERSPECTIVES ON THE ECOLOGY AND EVOLUTION OF HOMEOSTASIS

33.1

INTERGRATING PHYSIOLOGICAL ASSESSMENTS OF ANIMAL HEALTH TO POPULATION MODELS

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One of the main challenges in conservation biology is to detect when populations are at risk of decline before an extinction event takes place. Towards this aim, environmental physiologists have been assessing animal health by measuring indicators of homeostatic breakdown or allostatic load, such as circulating glucocorticosteroid hormones, within and among populations. Yet, these studies fall short of predicting population dynamics because they are not designed to derive quantitative relationships between glucocorticoids and survival probabilities or reproductive output. We propose to use demographic projection models in which we include functions relating population-level distributions of glucocorticoid levels with transition probabilities and fecundities. These models also determine the most sensitive transition probabili-

ties/fecundities for population dynamics, thus directing us to which life history stage we should focus more extensive assessments of homeostatic stability. Here we show examples of this approach in frog and bird models. This work was funded by National Science Foundation grants to EJC (GSS-1134687) and to RG (DMS-1029482). References: Crespi, E.J., Williams, T.D., Jessop, T.S., Delehanty, B. 2013. Life history and the ecology of stress: How do glucocorticoid hormones influence life-history variation in animals? *Functional Ecology* 27:93;106.

33.2

THE REACTIVE SCOPE MODEL: PREDICTING THE EFFECTS OF CHALLENGES TO HOMEOSTASIS

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Homeostasis - a balancing act requiring a range of physiological counter-balances. How do such counter-balances maintain homeostasis when faced with challenges, predictable, unpredictable or extreme? The *Reactive Scope Model* provides a conceptual framework for applying homeostasis theory to assess and make predictions about stress. The model considers the balance of physiological mediators operating within four distinct ranges. 1) *Predictive Homeostasis* includes circadian and seasonal variation of mediators at baseline levels. 2) *Reactive Homeostasis* includes the range required to respond to unpredictable challenges (i.e. acute stressors). These two ranges encompass the *Reactive Scope*. Physiological mediators at levels outside of the *Reactive Scope* increase potential for pathological effects. 3) *Homeostatic Overload* occurs when the response to a challenge exceeds the *Reactive Scope* and the mediators become detrimental. 4) *Homeostatic Failure* occurs when levels of physiological mediators fall below that required to maintain basic function. The specifics of what defines each of the four ranges can change with individual, species, and/or population, allowing the model to be adapted to include effects of early life stress, dominance hierarchies, seasonality etc. Applying the dynamic nature of homeostasis theory, the *Reactive Scope Model* allows for a versatile set of principles to consider the complexities and consequences of maintaining physiological balance in a challenging world.

33.3

PHYSIOLOGICAL REGULATORY NETWORKS: THE ORCHESTRA OF LIFE?

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Linear thinking and reductionism have been valuable in physiology, but much in biology depends upon complex interactions. Subsequently, the conventional single-system approach might benefit from more attention to interactions among systems and their components. Already, such efforts are paying off; cross talk among the immune, endocrine and nervous systems, for example, are integral to successful defense against parasites and other insults. Also, at other levels of organization (i.e., genes, communities, etc.), there are already useful frameworks for the study of complex interactions, namely networks. Here, we present a conceptual model of physiological regulatory networks (PRNs), the full suite of molecules and their regulatory relationships that mediate individual homeostasis. Although systems biology has embraced the study of networks, networks at the level of whole-individuals have received little attention. We argue that studies of PRN connectivity, modularity and hierarchy could transform how we think about and manage health and disease. PRN theory might also inform many ecological and evolutionary processes such as disease cycles and spread and species range expansions and contractions. Some have recently claimed that physiology is rocking the foundations of biology; we offer the PRN framework as one component of a new foundation to help unify heretofore disjunct (proximate and ultimate) biological realms. **References:** Cohen, Martin, McWilliams, Wingfield, and Dunne. 2012. Physiological regulatory networks: ecological roles and evolutionary constraints *TREE* 27: 428-435; Martin, LB and AA Cohen. Physiological regulatory networks: the orchestra of life? *In Integrative Organismal Biology*, Martin, Woods and Ghalambor, Wiley Press, in press (December 2014).

33.4

INFORMATION THEORY, HOMEOSTASIS, AND EVOLUTION

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Physiologists commonly view homeostasis as a set of mechanisms evolved to protect organisms from damaging extremes. Here I propose that homeostasis also evolves to minimize noise in physiological channels. Fluctuations in physiological factors constitute inescapable sources of noise that corrupt the transfer of information through biological systems. Drawing on information theory, I develop the idea that homeostatic regulation creates quiet physiological backgrounds for the transmission of all kinds of physiological information. This view leads to two additional ideas about the roles of homeostasis. First, homeostatic systems act as coupled pairs of transmitters and receivers, such that the performance of any one system influences information pro-

cessing in others. This dependence implies that multiple homeostatic systems, embedded within individual organisms, should show strongly synergistic or emergent effects. The second, related idea is that homeostatic systems should in general act as evolutionary capacitors. In typical or benign conditions, homeostasis hides physiological variation, such that variation accumulates over generations. In rare, extreme conditions, such variation may suddenly become visible to selection, giving rise to rapid but intermittent evolutionary shifts in homeostatic systems. This work was supported by the National Science Foundation (IOS-0844916). References: Woods, H. A., & Wilson, J. K. (2013). An information hypothesis for the evolution of homeostasis. *Trends in Ecology & Evolution* 28, 283-289. Woods, H. A. (2009) Evolution of homeostatic physiological systems. In *Phenotypic Plasticity of Insects: Mechanisms and Consequences*. Eds. DW Whitman & TN Ananthakrishnan, Science Publishers, Enfield, NH.

34.0: LINKING BEHAVIOR AND PHYSIOLOGY IN ANIMAL NAVIGATION AND ORIENTATION

34.1 USING GENETICS TO REVEAL MIGRATORY FLIGHT ORIENTATION MECHANISMS IN THE MONARCH BUTTERFLY

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The eastern North American monarch butterfly (*Danais plexippus*) has emerged as a powerful model system to study animal clocks, and navigational and migratory mechanisms. The navigational capabilities used by monarchs to accomplish their yearly long-distance migration rely primarily on a time-compensated sun compass orientation mechanism, in which circadian clocks in the antennae time compensate the sun compass output in the brain (1). The molecular and cellular mechanisms involved in the generation of the migratory syndrome remain however largely unknown. To further our understanding of the genetic and neuronal basis of migration, we have recently sequenced the monarch genome and developed genetic tools to knockout clock genes using nuclease-mediated gene targeting approaches (2). Our progress on the use of TALENs and CRISPR to genetically manipulate the monarch will be presented. This will allow us to eventually knock-in fluorescent reporter tags into clock gene loci to facilitate the dissection of the neuronal circuits underlying the flight orientation of migratory monarchs. References: 1) Reppert, S.M., Gegear, R.J. & Merlin, C. 2010. Navigational mechanisms of migrating monarch butterflies. *TINS* 33, 399-406. 2) Merlin, C., Beaver, L.E., Taylor, O.R., Wolfe, S.A. 7 Reppert, S.M. 2013. Efficient targeted mutagenesis in the monarch butterfly using zinc-finger nucleases. *Gen Res* 23, 159-168.

34.2 3D NEURAL COMPASS IN THE BAT BRAIN

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Head-direction cells are neurons that become active whenever the animal's head points to a specific direction (azimuth) in the horizontal plane, in a manner similar to a compass. Although the survival of the animal may depend on successful orientation in three-dimensional (3D) space, it is unclear how 3D head-direction is represented in the brain. Here we recorded from neurons in the dorsal presubiculum of Egyptian fruit bats, mammals well-adapted to 3D spatial behaviors, and found head-direction cells tuned to azimuth, pitch or roll – of which 30% were conjunctively tuned to multiple combinations of Euler angles. 3D head-direction tuning was observed in both crawling and flying bats. Head-direction cells were organized according to a functional-anatomical gradient, suggesting a gradual transition from 2D to 3D representations along the transverse axis of the presubiculum. When bats were in the upside-down position, most neurons remained directionally-tuned; surprisingly, however, their azimuth-tuning was shifted by 180° relative to the upright position—suggesting that 3D head-direction is represented on a toroidal manifold, rather than in standard spherical coordinates. The toroidal model was further supported by experiments in which bats crawled on a vertical ring – revealing that the tuning of pitch cells was circular, continuous and unimodal within the available 360° of pitch. Taken together, these results demonstrate a 3D head-direction mechanism in mammals, which could support navigation in 3D space. *These authors contributed equally to this work.

34.3 NEURAL REPRESENTATION OF THE HIERARCHY OF CELESTIAL CUES IN THE DUNG BEETLE BRAIN

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Some diurnal (*Scarabaeus lamarki*) and nocturnal (*S. satyrus*) dung beetles have developed a unique orientation behavior to avoid competition for food at a dung pat.

They cut off a piece of dung, form it into a ball and roll it away along straight paths. To keep a straight path, both species rely on skylight cues such as the sun, moon or the polarized skylight. We analyzed the hierarchy of these signals in both species in the field by setting the polarized light in conflict with the sun or the moon. We found that the sun is the main reference for the internal compass in diurnal beetles, whereas in nocturnal beetles polarized light is the dominant cue. However, what does this internal compass look like in the beetle brain and how is the hierarchy of compass signals represented at the neural level? The central complex (CX) is a brain area that likely serves as the insects' internal compass. Immunohistochemistry and 3D reconstructions were used to characterize the beetles' CX-network. In addition, CX-cells were analyzed through intracellular recordings while mimicking the sun or the moon by light spots that move on a circular path around the beetle's head and skylight polarization by a dorsally rotating polarizer. All cells responded to both stimuli, and thus encoded both celestial cues. Interestingly, when presented both cues simultaneously, neurons in the nocturnal species responded only to the polarizer, while cells were tuned only to the light spot in the diurnal species. This shows that in the beetle CX single neurons encode a hierarchy of different celestial cues, accurately reflecting our behavioral findings.

34.4 POLARIZED LIGHT NAVIGATION IN *DROSOPHILA*

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When normalized to body length, the migratory flights of drosophilid flies are among the longest aerial excursions in the animal kingdom. The means by which these tiny flies maintain a straight course over many kilometers is not known, but evidence suggests that pattern of polarized light in the dawn or dusk sky provides an important navigation cue. Recently, our laboratory has shown that flies will maintain a roughly constant course heading using the pattern of polarized light in a natural sky. To study navigation under more controlled conditions, we developed an indoor flight simulator in which a rigidly tethered fly can orient itself under an ersatz sky via a closed-loop feedback system. The results indicate that flies vary with respect to the azimuthal orientation of polarized light they prefer. In other words, each fly appears to choose a random heading, but then maintains that heading over time. We are currently using the genetically-encoded calcium indicator GCaMP to study circuitry that underlies polarization navigation in tethered, flying flies. Our initial results indicate that, as in other insects, the detection of polarized light starts within specialized central photoreceptors in the dorsal rim of the compound eye. Support: NSF 0623527, NIH 5-T32-MH019138, Paul G. Allen Family Foundation. References: Dickinson, M.H. (2014). Death Valley, *Drosophila*, & the Devonian Toolkit. *Ann. Rev. Entomol.* 59:51–72. Weir, P. T. and Dickinson, M.H. (2012). Flying *Drosophila* orient to sky polarization. *Current Biology* 22, 12-20.

35.0: THERMAL PHYSIOLOGY

35.1 AN *IN VIVO* INVESTIGATION OF LOW TEMPERATURE ENERGETICS IN *DROSOPHILA MELANOGASTER* USING ³¹P NMR

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Low temperatures lead to energetic failure in aquatic ectotherms, but the extent to which this occurs in terrestrial ectotherms remains unclear. Here, we use ³¹P nuclear magnetic resonance (NMR) spectroscopy to dynamically monitor levels of energetically important phosphorus-based molecules, including ATP and phosphoarginine, during the course of cold exposure and recovery, using populations of *Drosophila melanogaster* selected for either fast (hardy) or slow (susceptible) recovery from cold-coma. We hypothesized that flies enter a lower energy state when lowered below the cold-coma threshold, and that cold-adaptation reduces the extent of the energetic disruption or facilitates a faster recovery. Phosphoarginine and sugar phosphates increased abruptly when flies were cooled and then steadily decreased over the course of the cold exposure. An unknown metabolite decreased sharply upon cold exposure and recovered upon rewarming. Hardy flies had markedly lower levels of this unknown metabolite during both cold exposure and recovery. We found little evidence for changes in ATP during the course of cold exposure or recovery. We conclude that energy limitation does not appear to underlie evolved differences in cold-coma recovery times, and suggest future work be done to identify this interesting metabolite that responds to cold, and whose dynamics *in vivo* are strongly altered by cold adaptation. This work was funded by NSF grant 1051890 to DAH, ASE, and TJM.

35.2

NEW APPROACHES TO UNDERSTANDING INSECT FREEZE TOLERANCE

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The ability of some insects to survive internal ice formation was first described over 200 years ago, yet we still do not understand the underlying mechanisms, nor are we able to induce robust freeze tolerance in non-freeze tolerant insects. I will briefly discuss what is known about insect freeze tolerance, and how the approaches we have taken in the past may have led to dead-ends. I will then describe new approaches being taken in my laboratory to elucidate the mechanisms underlying freeze tolerance in insects. These are 1) the development of new comparative models that allow variation in freeze tolerance to be studied on seasonal, geographic, and evolutionary scales; 2) the utilisation of omics technologies (primarily RNA-Seq) to identify new candidate molecules and pathways; 3) an hypothesis-testing approach to critically examine the existing models of insect freeze tolerance. I will present a selection of data derived from each of these approaches, particularly focusing on whether freeze tolerance has an underlying set of common mechanisms in spite of having evolved independently on multiple occasions. I will also discuss new data about the processes associated with thawing in insects that survive freezing, and discuss the identity, function, and synthesis of acetylated lipids, which may function as intracellular cryoprotectants. I conclude that these new approaches signal an exciting time in the study of freeze tolerance, and the likelihood of identifying novel biochemical and physiological mechanisms of cold tolerance in ectotherms.

35.3

MITOCHONDRIAL AND NUCLEAR GENETIC VARIATION RELATE TO HEAT AND COLD TOLERANCE IN A MONTANE LEAF BEETLE

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Montane ectotherms are often confronted with dangerously high and low temperatures in summer when individuals are active and completing reproduction and development to adulthood. We investigated effects of exposure to stressfully hot and cold temperatures for adults and larvae of the willow beetle *Chrysomela aeneicollis* collected in California's Sierra Nevada Mountains. To investigate relationships between genetic variation and thermal phenotype, we sampled beetles from an introgressed population where southern and northern nuclear and mitochondrial genomes recombine freely. We measured running speed after heat or cold stress, cold tolerance, and activity of Cytochrome oxidase, then obtained genotypes at COII and the nuclear gene phosphoglucose isomerase. Heat tolerance was greatest for beetles with a southern mitochondrial and nuclear genotype but lowest for beetles with the southern mitochondrial genotype and northern nuclear genotype. In contrast, beetles with the southern mitochondrial genotype showed the greatest cold tolerance if they possessed the northern nuclear genotype, but lowest cold tolerance when they possessed southern nuclear and mitochondrial alleles. We discuss the significance of these results for understanding of the role of mitonuclear interactions in evolution of thermal tolerance. This research was supported by NSF (DEB 0844404/06).

35.4

NEVER MIND THERMAL PERFORMANCE CURVES-IT'S ALL ABOUT TOLERATING THE EXTREMES! SENSITIVITY TO THERMAL EXTREMES PREDICT CURRENT (AND FUTURE?) DISTRIBUTION OF *DROSOPHILA* SPECIES

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Climatic factors influence the distribution of ectotherms, raising the possibility that distributions of many species will shift rapidly under climate change. Recent studies comparing performance curves of species from different climate zones have suggested that tropical species are more susceptible to climate change than temperate species. However, comparisons involving responses to thermal extremes have found that mid-latitude species are more susceptible. Using a group of 10 related *Drosophila* species with tropical or widespread distribution, we undertook a detailed investigation of their growth performance curves (fecundity, developmental success, and developmental time) and tolerance to thermal extremes (adult heat and cold resistance). Using these data we modeled distribution patterns and compared these models to current observations of species distribution. Thermal performance curves proved to be a poor predictor of current species distributions while adult tolerance to thermal extremes provides a good correlate of current distributions. Thus, in their current distribution range, most of the examined species experience heat exposure close to, but rarely above, the functional heat resistance limit. Similarly, adult cold resistance proved a good predictor of species distribution in cooler climates. Given these findings we pre-

dict that both tropical and widespread *Drosophila* species will face a similar proportional reduction in distribution range under future warming.

35.5

THERMAL PERFORMANCE, AEROBIC SCOPE, AND RELEVANCE OF THE OCLTT HYPOTHESIS

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It has long been debated how temperature determines physiological performance and field distribution in ectothermal animals. The oxygen- and capacity-limited thermal tolerance (OCLTT) hypothesis states that thermal performance in aquatic ectotherms is limited by aerobic scope ($MO_{2max} - MO_{2min}$). This hypothesis has gained popularity despite relatively little experimental evidence. Here we examine the evidence for the core assumption, that oxygen supply to the tissues is the causal mechanism for reduction in other performance metrics. The available literature demonstrates that aerobic scope often continues to increase with temperature up to lethal temperatures, while other performances such as growth or reproductive output can be limited at much lower temperatures. We argue, in accordance with the results of several recent experiments, that while oxygen supply can limit thermal tolerance it is not the main cause of performance decline during slow warming in fish. We advocate the multiple performances - multiple optima (MPMO) view of thermal performance, which states that any physiological function can be limiting and that this can differ between species, life stages and rates of heating. In order to advance our understanding of thermal physiology, field distribution and climate change sensitivity, we should identify these limiting functions in fish.

35.6

THERMAL TOLERANCE AND MOLT CYCLE-DEPENDENT GENE EXPRESSION IN JUVENILE DUNGENESS CRABS

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We hypothesize that the mTOR pathway and AMP-dependent protein kinase (AMPK) are involved in the regulation of the molt cycle in response to temperature in juvenile Dungeness crabs *Metacarcinus magister*. We used PCR cloning to obtain partial cDNA sequences of mTOR, Rheb, AKT, S6K and AMPK. We incubated crabs at 12 d, 18 d and 26 d postmolt at 5, 10, 15, 20, 25 and 30°C for up to 14 d to study survival and progression of the molt cycle. We quantified gene expression in the molting gland of AKT, upstream of mTOR, and S6K, downstream of mTOR, of the possible housekeeping gene RbS3, and of Na⁺/K⁺-ATPase in crabs that had been held at 10, 15 or 20°C. Survival was 97-100% at temperatures from 5-20°C, and was time-dependent, but 0% after 14 d at 25°C. All animals had died after 24 h at 30°C. Significant progression of the molt cycle was observed at 15 and 20°C, but not at 5 and 10°C. An overall two-way ANOVA indicated an effect of temperature on expression of all four genes, whereas only AKT, RbS3 and Na⁺/K⁺-ATPase were significantly affected by molt stage. The *post hoc* Tukey multiple comparisons test revealed significant effects of temperature at either 26 d, 32 d or at both time points, gene expression being significantly lower at 20°C compared to 10°C. Molt-stage specific effects were prominent at 20°C, with increased gene expression in premolt. We conclude that AKT, an activator of mTOR, may be involved in the regulation of the molt cycle in a temperature-dependent manner. Funding: EU FP7 Marie Curie International Outgoing Fellowship PIOF-GA-2012-326483 to ACW, NSF grant IOS-1257732 to DLM.

35.7

VARIATION IN TRANSCRIPTOMIC SIGNATURES OF THERMAL ACCLIMATION IN FOUR KEY AQUATIC INSECTS IN CALIFORNIA RIVERINE FOOD WEBS

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Predicting changes in trophic ecology of riverine systems in the face of future climate warming requires an understanding of the thermal performance of aquatic insect larvae. Larvae that differ in key trophic traits (i.e., grazer vs. predator) may also differ in the efficiency with which they use energy under various thermal regimes. We tested the hypothesis that metabolic energy efficiency is maximized at optimal temperatures and declines at higher temperatures due to an increase in fermentative metabolism and an induction of stress responses to cope with thermal or oxidative damage by examination of transcriptomic shifts with temperature, with the presumption that physiological changes involve gene expression. We compared thermal effects on gene expression of four aquatic insects (*Pteronarcys californica*, *Calinuria*

californica, *Hesperoperla pacifica* and *Dicosmoecus gilvipes*) across temperatures of the spring-summer range during late larval instar phase (~12-30°C). Illumina RNA-seq, *de novo* transcriptome assembly using Trinity, and analysis using Bowtie2/edgeR and EdgeR were used to identify differentially expressed genes within each species. Heat sensitive predator taxa exhibited a reduced induction of cellular stress response genes. Biomarkers of thermal acclimation from one species, *D. gilvipes*, were used to compare across a fine-scale range of temperatures, and across populations from sites with differing thermal characteristics (Mendocino, Sierra Nevada, Big Sur). These data allow a fine-scale analysis of local physiological adaptation to temperature and shifts in metabolic ecology of streams across much of the central to northern California landscape.

35.8

SEASONALLY INDUCED HEPATOTRANSCRIPTOMIC CHANGES IN THE FREEZE TOLERANT NORTH AMERICAN WOOD FROG *RANA SYLVATICA*

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The North American wood frog *Rana sylvatica* uses an overwintering strategy that includes metabolic depression and freeze tolerance. Summer-acclimated animals will not survive freeze events, suggesting that there is seasonally induced acclimation in wood frogs. Previous hepatoproteome analysis indicated a distinct shift in metabolism between summer and winter acclimated animals. The current study evaluated the hepatic gene expression differences between seasonally acclimated wood frogs. Using frogs just thawed and four weeks post-thaw, we isolated total RNA (4 frogs – 2 per condition, in triplicate) and constructed Illumina TruSeq RNA v2 libraries. All twelve samples were sequenced (100 bp PE) on a HiSeq2000. FASTQ files were processed for quality and adaptor trimming in CLC Genomics Workbench 7.04. *De novo* assembly of the transcriptome was performed after duplicate reads removed – minimum contig length of 400 bp. The assembled wood frog transcriptome generated 55,789 contigs, which were annotated (Blast, GO, KEGG) via Blast2GO. The annotated transcriptome served as the reference for RNA-Seq based differential gene expression. Initial analyses have indicated at least 61 cDNAs exhibiting significant ($p < 0.05$) differential expression between just thawed and four week post-thawed frogs. Comparisons to other freeze-tolerant animals are being analysed for similarity of differential hepatic gene expression during seasonal acclimation.

36.0: ENDOCRINOLOGY AND REPRODUCTIVE PHYSIOLOGY

36.1

ELUCIDATING THE REGULATORY MECHANISMS OF CIRCADIAN CONTROL OF OVARIAN ECDYSTEROID PRODUCTION AND RELEASE DURING EGG DEVELOPMENT IN ADULT FEMALE *RHODNIUS PROLIXUS*

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The significance of endocrine rhythms is recently being appreciated for their role as important 'messengers of time'. Endocrine rhythms distribute timing information to tissues and cells to coordinate the timing of physiological events in an organism. Ecdysteroids, a predominant molting hormone in insects, are released with a circadian rhythm to regulate larval circadian organization. Prothoracicotropic hormone (PTTH) is a neuropeptide released rhythmically from the brain that stimulates the prothoracic glands (PGs) in *Rhodnius prolixus* larvae to produce ecdysteroids. While the PGs are absent in *R. prolixus* adults, this PTTH-ecdysteroid axis still persists with the ovaries replacing the PGs as the primary producers of ecdysteroids in female insects. A clear circadian rhythm in hemolymph ecdysteroid titer released from the ovaries *ex vivo* during egg development has been reported. From this, the possible existence of an endogenous ovarian circadian clock is discussed. We examine the role of neuro-hormonal control (i.e. PTTH rhythms) of ecdysteroid production by the ovaries in adult female *R. prolixus* by stimulating ecdysteroid release with PTTH (crude brain extract and recombinant PTTH peptide) and quantifying using an ecdysteroid radioimmunoassay. In addition, removal of rhythmic PTTH input from the brain during egg production was also considered. Funding: Natural Sciences and Engineering Research Council of Canada.

36.2

ONE CELL OR TWO? DIRECT VISUALIZATION OF LIGAND-RECEPTOR INTERACTION PROVIDES NOVEL INSIGHTS INTO THE EVOLUTION OF INSECT RENAL FUNCTION

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Insects are under unique constraints due to their high surface area to volume ratio. Nevertheless, the insect renal (Malpighian) tubules secrete primary urine faster (on a per-cell basis) than any other known epithelium. In the classic insect model *Drosophila melanogaster*, ion and water transport are controlled by two main secretory cell types; principal and stellate cells; and evidence suggests that this arrangement is general within the large Order of Diptera (true flies). However, is this two-cell model general across the insects? To address this, we developed a novel technique for comparative endocrinology by directly visualizing kinin receptors (diagnostic of the stellate cell identity) in live tissue with a fluorescently labeled kinin peptide. This technique allowed us to rapidly analyze strategically selected representatives of every major insect Order; hereby covering nearly 400 million years of evolution and >90% of insect diversity; thus providing an unprecedented overview of the evolution of insect renal function. Our data indicate that the two-cell model is a derived trait that evolved approximately 350 million years ago in the endopterygote insects, whereas a single cell model is basal to the more ancestral Exopterygota. Funding: BBSRC, NIH & Danish Council for Independent Research (FNU).

36.3

BRAIN MONOAMINES AND BEHAVIOR-RELATIONSHIP TO PERSONALITY TRAITS AND THE EFFECTS OF SOCIAL INTERACTION

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Fights for social dominance are stressful and results in an activation of the brain serotonergic system. Subordinate animals in an established dominance hierarchy are characterized by a chronic elevation of brain serotonergic activity, an effect that seems to mediate the behavioral inhibition observed in these animals. By contrast, social dominance has been associated with elevated dopaminergic activity, and dopamine (DA) has behavioral effects to some extent opposing those of serotonin (5HT). In addition to effects of the social environment, brain monoaminergic functions are controlled by genetic factors. For instance, zebrafish (*Danio rerio*) classified as bold, showing a propensity for being social dominant, display higher expression of DA2b receptor mRNA than shy conspecifics. In rainbow trout (*Oncorhynchus mykiss*) and Baltic salmon (*Salmo salar*) bold and shy individuals differ in the expression of 5HT1A receptor mRNA. In salmonids, bold and shy fish also show divergent effects of stress on brain 5HT neurotransmission. Personality traits are to a large extent genetically controlled. However, teleost fish are extremely plastic and it is not clear to what extent; personality related; differences in monoaminergic systems is related to heritable factors or early social interaction.

36.4 WITHDRAWN

36.5

CORTICOSTERONE RESPONSES AND THE ABILITY OF BIRDS TO COPE WITH ENVIRONMENTAL CHANGE

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Birds, like other animals, live in complex environments that can change at any time. When stimuli from the external environment are perceived to be a threat or potential threat then a stress response is initiated and corticosterone is secreted. There is considerable individual variation in corticosterone responses, and a stimulus that initiates a large response in one bird may initiate a small response in another bird. For example, peak corticosterone responses to capture and handling were 15 times higher in some birds than others in a study of little penguins (*Eudyptula minor*) in New Zealand. Corticosterone responses and behavioural responses to environmental stimuli are together determined by individual characteristics called personality. Birds with low corticosterone responses and proactive personalities are likely to be more successful (have greater fitness) in constant or predictable conditions, whilst birds with reactive personalities and high corticosterone responses will be more successful in changing or unpredictable conditions. It is proposed that birds with reactive personalities and high corticosterone responses will be better able to cope with environmental changes due to climate change than birds with proactive personalities and relatively low corticosterone responses. Phenotypic plasticity in corticosterone responses can be quantified using a reaction norm approach, and reaction norms can be used to determine the degree of plasticity in corticosterone responses of individual birds, and mean levels of plasticity in responses of species of birds. Reaction norms for corticosterone responses can in future be used to help predict the ability of birds to cope with environmental changes due to climate change.

36.6

INFLUENCE OF CORTICOSTERONE ON GROWTH, HOME-CAGE ACTIVITY, WHEEL RUNNING, AND MAXIMAL OXYGEN CONSUMPTION IN REPLICATE LINES OF HOUSE MICE SELECTIVELY BRED FOR HIGH VOLUNTARY WHEEL-RUNNING BEHAVIOR

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Corticosterone, chiefly referred to as a "stress hormone," impacts a surprising variety of traits, including skeletal growth, cognition, and motivation. Hence, changes in corticosterone levels may impact multiple aspects of locomotor behavior, including both motivation and physical abilities. Previous rodent studies have found that altering circulating corticosterone levels can affect activity levels or induce a depressive state, depending on dosage and other factors. We have used a long-term artificial selection experiment to examine the evolution of high levels of voluntary wheel-running behavior in laboratory house mice. As compared with four non-selected control (C) lines, the four replicate High Runner (HR) lines are smaller in body size, run ~3-fold more on wheels on a daily basis, have higher home-cage activity when deprived of wheels, and have higher maximal oxygen consumption (VO₂max) and basal circulating corticosterone levels. To examine further the role of corticosterone in locomotion, we administered 50 µg/ml corticosterone hemisuccinate in the drinking water of HR and C male mice from weaning to seven weeks of age. Mice were then tested for wheel running and VO₂max. Corticosterone reduced growth rate and body mass-adjusted VO₂max of both HR and C mice, and decreased wheel running in HR lines. The present results suggest that corticosterone can impact wheel running via changes in VO₂max when animals run near their aerobic limits. Supported by NSF IOS-11212732.

36.7 MOLECULAR RESOLUTION OF AN ACUTE STRESS RESPONSE IN A FREE-RANGING MARINE MAMMAL

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Environmental stress negatively affects reproduction and survival of marine mammals, threatening ecosystem stability. However, our understanding of the stress response in these organisms is currently limited to circulating hormone measurements and is largely based on terrestrial model systems. To identify marine mammal-specific markers of stress, we examined the molecular signature of an acute stress response in free-ranging northern elephant seals. We stimulated the hypothalamic-pituitary-adrenal (HPA) axis of juvenile seals by administering adrenocorticotropic hormone (ACTH) and analyzed the downstream response in target tissue by transcriptomics. We sequenced RNA isolated from muscle of animals before and after ACTH injection and generated the first reference sequence for the northern elephant seal using a user-friendly standalone protocol for transcriptome assembly. We identified a number of genes and pathways altered in response to acute stress, which include immune system markers, muscle stem cell factors, and metabolic pathways, among others. Genes upregulated in response to HPA axis challenge can be used as robust molecular markers of acute stress in elephant seals and other marine mammals.

36.8 BREAKING DIAPAUSE: SUCCESSFUL USE OF ULTRASONOGRAPHY SHOWS INTRA-SPECIFIC VARIATION IN THE PROBABILITY AND TIMING OF EMBRYO IMPLANTATION IN WEDDELL SEALS

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Weddell seals (*Leptonychotes weddellii*) give birth in Oct/Nov each year, followed by the breeding season (Dec). In Jan/Feb 2014, we handled prime-age multiparous female Weddell seals that did not have a pup (N=6) a few months previously, as well as females that had given birth early (Oct; N=6) or late (Nov; N=5) in the pupping season. All reproductive females were handled between 92-103 days post-partum. We successfully applied transrectal ultrasonographic techniques to characterize the reproductive tract (uterine body and horns, ovaries with corpora lutea), detect early pregnancy (embryonic vesicles < 3mm), and estimate the length of diapause in Weddell seals. In older embryos, crown-rump-length, heartbeat, and limb bud development were also observed. Of these animals, 83% of skip-breeders, 100% of early moms, and 60% of late moms were detectably pregnant. The embryonic vesicle and crown-rump-length were larger in skip-breeding females relative to post-partum females (vesicle: 3.763±1.329 vs. 923±307mm²; embryo proper: 30±9.0 vs. 12±4.7mm). A fetal growth curve constructed from ultrasound measurements revealed sufficient variation in fetal size among reproductive groups to suggest that diapause does not synchronize the time of embryo attachment in Weddell seals. This study demonstrates that ultrasonography can be used to address important biological questions regarding intra-specific variation in reproductive rates and phenology in a wild animal. Funding source: NSF1246463.

37.0: HIBERNATION, FLIGHT, AND SUBSTRATE METABOLISM

37.1

SEASONAL METABOLISM OF BROWN ADIPOSE TISSUE IN HIBERNATING THIRTEEN-LINED GROUND SQUIRRELS

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Brown adipose tissue (BAT) is a vital organ for mammalian hibernation, dissipating energy and producing heat via mitochondrial uncoupling protein 1 (UCP1) to rapidly enable rewarming from torpor. However, BAT's unique ability is not necessary outside the hibernation season, suggesting a potential seasonal change in mitochondrial content, respiratory capacity, or both. To address this question, we examined the mitochondrial content and bioenergetics of BAT in thirteen-lined ground squirrels across four time points: Summer (SU), Fall (FA), Hibernation (HIB) and Spring (SP). Respiration rates of isolated BAT mitochondria were measured using an oxygen electrode with three substrates: Succinate (SUC), Glycerol-3-Phosphate (G3P), and Ascorbate/TMPD (Asc/TMPD). G3P-fueled and Asc/TMPD-fueled respiration were both significantly higher in HIB than in both SU and FA ($p < 0.05$). SUC-fueled respiration was significantly lower ($p < 0.05$) in SP than in HIB. Relative mitochondrial content of BAT was estimated by measuring mitochondrial DNA (mtDNA) copy numbers via qPCR. BAT mtDNA copies were significantly lower in SP ($p < 0.05$). These data imply that BAT bioenergetics and mitochondrial content is not static across the year, even though UCP1 is present year-round. The active suppression of BAT metabolism outside of hibernation may be playing a vital role in the overall energetic savings of a hibernator. This project was funded by the University of Minnesota McKnight Presidential Endowment.

37.2

IS SAPONIN-PERMEABILIZATION APPROPRIATE FOR CHARACTERIZING MITOCHONDRIAL METABOLISM? ASSESSING MITOCHONDRIAL RESPIRATION IN HIBERNATING AND EUTHERMIC GROUND SQUIRRELS

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Saponin-permeabilization of small tissue slices is increasingly popular for characterizing mitochondrial metabolic functions including respiration rates and ROS production. This technique is more convenient and faster than traditional mitochondrial isolation, and is well described for muscle and brain, but not for liver. The goal of this study was to assess the suitability of this method by comparing mitochondrial metabolism between saponin-permeabilized liver slices and isolated mitochondria from the same livers of a hibernator, the 13-lined ground squirrel. As we have demonstrated in several previous studies, state 3 respiration, fuelled by succinate, was suppressed by 65% in mitochondria isolated from torpid ground squirrels when compared with aroused, euthermic animals. Permeabilized liver slices showed respiratory control comparable to isolated mitochondria, but no suppression of state 3 respiration. These results held regardless of the method used to standardize respiration rate (protein, citrate synthase activity and cytochrome A content for mitochondria, wet weight and citrate synthase activity for permeabilized slices). These results suggest that the mechanisms underlying mitochondrial metabolic suppression in torpor were reversed by the permeabilization procedure. We cannot recommend this method for characterizing mitochondrial metabolism, at least for conditions where acute metabolic changes are known to occur. Funding: NSERC Canada.

37.3

BROWN FAT TRANSCRIPTOME DYNAMICS: PRESERVATION OF SELECTED MRNAS ACROSS A TORPOR BOUT SUPPORTS RAPID THERMOGENESIS DURING AROUSAL

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Brown fat (BAT) undergoes an annual cycle of recruitment and atrophy in 13-lined ground squirrels. Throughout winter hibernation, BAT cycles between quiescence and activity to match the whole body metabolism of torpor and arousal. We examined the BAT transcriptome in 10 distinct physiological and seasonal states of the hibernator's year to gain molecular insight into this dynamic phenotype. EDGE tags were collected, mapped to the genome then analyzed for sample clustering, differential gene expression among states, and hierarchical clustering to reveal gene expression patterns. Most gene expression changes were seasonal; transcripts involved in lipid transport and metabolism dominate the period of hibernation, whereas those involved in apoptosis and RNA-processing dominate the period of spring homeothermy. Significantly, many transcripts increased during torpor at low body temper-

ature when transcription is depressed. Additional experiments using rt-qPCR validate the high-throughput sequencing results and reveal that some of these transcripts uniquely gain a polyA tail during torpor, whereas others are preserved relative to bulk transcripts which slowly degrade across the torpor bout by an apparently robust mechanism based on mathematical modeling. Because the preserved transcripts are dominated by genes involved in BAT metabolic activity, we hypothesize that this selected subset is protected for immediate translation upon arousal, thereby assuring rapid thermogenesis. Ack: NIH1R21DK095180.

37.4

CHARACTERIZING CARDIAC MOLECULAR MECHANISMS OF MAMMALIAN HIBERNATION VIA QUANTITATIVE PROTEOMICS

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This study uses cutting-edge proteomic approaches to elucidate cardioprotective mechanisms used during mammalian hibernation. Mammalian hibernation is characterized by dramatic reductions in body temperature, heart rate, metabolism and oxygen consumption, which poses significant challenges to the physiology of hibernators, especially for the heart, which maintains function throughout extreme conditions, resembling hemorrhagic shock. To identify novel cardioadaptive strategies we merged large-scale RNA-seq data with large-scale iTRAQ-based proteomic data in heart tissue from thirteen-lined ground squirrels (*Citellus tridecemlineatus*), throughout the circannual cycle. Protein identification and data analysis were run through Galaxy-P, a new multi-omic data analysis platform enabling effective integration of RNA-seq and MS/MS proteomic data. Galaxy-P uses flexible, modular workflows that combine customized sequence database searching and iTRAQ quantification to identify novel ground squirrel-specific protein sequences and provide insight into molecular mechanisms of hibernation. This study allowed for the quantified identification of 2007 cardiac proteins, including 447 novel peptides. Identification of novel peptides allows for improved genomic annotation of this non-model organism, as well as splice variants, mutations, or genome re-organization that underly novel cardio-protective mechanisms used during hibernation. NSF grant 1147079 and USARMC contract W81XWH-11-1-0409.

37.5

DO POLYUNSATURATED FATTY ACIDS IMPROVE MIGRATORY FLIGHT PERFORMANCE?

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The natural doping hypothesis states that long chain n-3 polyunsaturated fatty acids (PUFA) help prime the flight muscles of birds for endurance flights, enabling long distance migration. Proposed mechanisms include the alteration of membrane composition and oxidative enzymes increasing aerobic and endurance capacity. However, evidence also suggests that n-6 PUFA may be beneficial to migratory performance. It is unclear if the effect is due to PUFA in general or the type of PUFA. We tested directly if long chain n-3 and n-6 PUFA improve endurance flight performance in a migratory songbird. Yellow rump warblers (*Setophaga coronata*) were fed diets high in monounsaturated fatty acids (MUFA), long chain n-3 or n-6 PUFA for 6 weeks. Flight performance was assessed by voluntarily flying birds in a wind tunnel for up to 6 hours. Quantitative magnetic resonance was used to measure the disappearance of wet lean and fat mass for the estimation of metabolic flight costs and fuel mixture composition. Feeding the n-6 PUFA diet decreased flight cost by ~20% compared to the MUFA diet. The n-3 PUFA diet was intermediate in flight cost and did not differ statistically from the n-6 PUFA or MUFA diets. No difference in the proportional contribution of fat and lean mass to the fuel mixture was found. This study supports the capacity of long chain PUFA to increase the metabolic efficiency of flight, but this effect was attributed to n-6 PUFA and not n-3 PUFA. Research supported by NSERC.

37.6

COLD AND EXERCISE TRAINING PRODUCE SIMILAR INCREASES IN MAXIMAL METABOLIC OUTPUT IN HOUSE SPARROWS

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Maximal metabolic outputs for exercise and thermogenesis in birds presumably influence fitness through effects on flight and shivering performance. Because both summit (M_{sum} = maximum thermoregulatory metabolic rate) and maximum (MMR = maximum exercise metabolic rate) metabolic rates are functions of skeletal muscle, correlations between these measurements might occur. We measured the effects of 3-week experimental cold and exercise

training protocols on body (Mb) and muscle masses, basal metabolic rate (BMR), M_{sum} , and MMR in house sparrows (*Passer domesticus*). We also measured citrate synthase (CS), β -hydroxyacyl CoA-dehydrogenase (HOAD), and carnitine palmitoyl transferase (CPT) activities, and mRNA expression for plasma membrane fatty acid binding protein, fatty acid translocase, the muscle growth inhibitor myostatin, and its two proteinase activators (TLL-1, TLL-2) in pectoralis (PEC). Both training protocols increased M_{sum} , MMR, Mb, PEC mass, CPT and CS activities in PEC above control levels. BMR, however, increased with cold but decreased with exercise training. Cold-trained birds had lower expression of myostatin, but higher TLL-1 and TLL-2 levels than controls. However, no significant differences in gene expression occurred for exercise-trained birds. These data indicate that cross-training effects between cold and exercise may occur for birds, but mechanisms underlying the changing phenotypes are not identical. This research was funded by NSF IOS-1021218.

37.7

HYDROGEN ISOTOPE (δ^2H) DISCRIMINATION IN TILAPIA

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Hydrogen isotope (δ^2H) analysis has become as a valuable tool in the study of avian migration, however, the biochemical framework required for ecologists to confidently apply δ^2H in natural settings has yet to be adequately revealed. Two fundamental questions that need to be addressed are (1) what proportion of hydrogen in consumer tissues is derived from food versus drinking or environmental water, and (2) what is the magnitude and degree of variation in δ^2H trophic discrimination factors (Δ^2H_{nd}) between consumers and these resources? We completed a 3x3 controlled feeding experiment on fingerling Nile tilapia (*Oreochromis niloticus*) to address these questions. We found that the proportion of hydrogen in tilapia tissue derived from environmental (tank) water ranged from 21-24% for muscle and 24-29% for liver. We then used a linear mixing model to estimate the proportion of hydrogen derived from dietary protein (47%), carbohydrates (11%), and lipids (19%) that was utilized by tilapia for tissue synthesis. With this information and the δ^2H values of water and food in each treatment, we could calculate mean (\pm SD) Δ^2H_{nd} values, which were $-26\pm3\%$ for muscle and $-14\pm4\%$ for liver. In addition to providing one of the first experimental glimpses into how animals assimilate hydrogen for tissue synthesis, we anticipate that our results can be used to quantify the contribution of autochthonous (algae) versus allochthonous (riparian vegetation) resources in freshwater aquatic ecosystems.

37.8

THE BREATH BECOMES ISOTOPICALLY HEAVIER AS THE BODY BURNS MORE CARBOHYDRATES DURING INTENSE EXERCISE: EXPLOITING THE NATURAL DIFFERENCES IN ^{13}C BETWEEN LEAN AND LIPID TISSUES

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The lipids in the body contain measurably lower concentrations of ^{13}C than nonlipids because of the isotopic discrimination of ^{13}C -containing precursor molecules during lipid synthesis. We examined whether this naturally occurring difference in ^{13}C distribution is sufficiently large to study the changes in the $\delta^{13}C$ of the exhaled breath during different levels of exercise. We predicted that $\delta^{13}C$ of the breath would increase with the increased reliance on carbohydrate oxidation known to occur during high intensity exercise. We measured ventilatory and metabolic variables before (28 min), during (56 min), and after (28 min) exercise in 48 healthy young adults assigned to either a resting treatment or one of three exercising treatments where they maintained a heart rate of 130, 150, or 170 BPM. We found that the mean metabolic rates of the exercising groups increased 4.4-fold, 6.1-fold, and 7.7-fold above resting values, respectively. Exercise caused mildly increased respiratory exchange ratios indicative of increased carbohydrate oxidation, but these changes were too variable to be correlated with exercise intensity. In contrast, the $\delta^{13}C$ of the exhaled breath increased by 0.6‰, 1.1‰, and 1.8‰, respectively for the three groups and was significantly correlated with the intensity of exercise, suggesting that ^{13}C of exhaled breath can provide a complementary, noninvasive approach to studying shifts in substrate oxidation *in vivo*.

38.0: BIOMECHANICS, LOCOMOTION, AND FUNCTIONAL MORPHOLOGY

38.1

MUSCULOSKELETAL PLASTICITY OF THE HINDLIMB/TAILO MOTOR MODULE IN THE AMERICAN ALLIGATOR (*ALLIGATOR MISSISSIPPIENSIS*)

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Archosaur posture during terrestrial locomotion is, to some degree, a function of the caudofemoralis longus muscle (CFL). In extant reptiles, the CFL originates on the transverse processes and chevrons of the caudal vertebrae, and inserts on the fourth trochanter of the femur. This allows the CFL to act as a major retractor/medial rotator of the thigh. We tested whether the loss of CFL function elicits downstream effects on skeletal morphology of the hindlimb/tail module of archosaurs using bilateral tenotomy to deactivate the CFL in juvenile alligators ($n=12$ per group, $Mb=180\pm30g$, snout-vent length= $213\pm28mm$). After eight months, experimental animals exhibited no significant differences in femur length, external diameter at the fourth trochanter, or average chevron length. Polar moment of inertia and cross-sectional area at the fourth trochanter were similar in experimental and sham-operated control groups. In contrast, CFL wet mass and fiber length were significantly reduced, by 23% and 13% respectively, in experimental animals. CFL tenotomy thus elicited changes at the muscular level, but not at the skeletal level, suggesting a potential decoupling of muscular and skeletal plasticity in alligators. Given that no significant changes were observed in terrestrial locomotor performance (determined by 2D walking kinematics) following tenotomy, our results may also point to compensatory mechanisms that maintain stance and gait in alligators regardless of CFL deactivation.

38.2 SKELETAL MUSCLE PERFORMANCE UNDER SUB-MAXIMALLY ACTIVATED CONDITIONS

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¹Ecology & Evolutionary Biology, Univ. of California, Irvine, McGaugh Hall, Irvine, CA, 92617, ²Biomedical Physiology & Kinesiology, Simon Fraser Univ., 8888 University Dr., Burnaby, V5A 1S6, Canada, ³Organismal & Evolutionary Biology, Harvard Univ., Concord Field Station, 100 Old Causeway Rd., Bedford, MA, 01730. Skeletal muscles are rarely maximally recruited; yet our understanding of muscle physiology is based largely on studies using maximal activation. Here we explore the effects of activation level on whole muscle force-velocity and force-length properties. Force-velocity properties were determined in rat plantaris muscles with all, mainly fast or mainly slow motor units activated. Maximum shortening velocity declined at lower activation levels, but was independent of motor unit type activated. We propose that the effect of muscle resistance to shortening predominates over the effect of active motor unit type in determining force-velocity properties at low activation levels. Force-length properties were determined in frog plantaris muscles using stimulation conditions which elicited high force-high Ca^{2+} , low force-high Ca^{2+} and low force-low Ca^{2+} contractions. Optimum muscle length increased with decreasing force, irrespective of Ca^{2+} levels. Hence, we propose that the varying effect of internal mechanics underpin the shift in optimum length at low activation levels. These findings suggest that the interaction of contractile elements and the physical properties of muscle (mass, viscosity, compliance etc) result in the non-linear scaling of muscle performance with activation level. Such mechanistic explanations of sub-maximal muscle performance will improve our understanding of *in vivo* muscle function and will inform new muscle models. Supported by NIH AR055295 and AR055648.

38.3 SIZE MATTERS: THE IMPACT OF BODY MASS ON BIOCHEMICAL AND STRUCTURAL PROPERTIES IN HARBOR SEAL MUSCLES

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The physiological attributes of muscle fibers (e.g., fiber type, size, enzymes profiles) influence muscle performance capacity, and are known to vary inter- and intra-specifically due to changes in age, diet and activity patterns. Muscle phenotype has been closely linked to underwater foraging ability of adult marine mammals, but its resilience to reductions in diving and foraging activities that accompany reproduction and molt has not yet been assessed. Seasonal differences in muscle metabolic profiles and structure were compared in 39 harbor seals (*Phoca vitulina*) captured in Glacier Bay National Park, Alaska; 12 prior to the pupping season, and 27 after the molt. Seals were in better condition (% lipid) in spring, but there were no seasonal differences in any other measured parameter, indicating that harbor seals maintain muscle performance capacity year-round. However, heavier seals had significantly larger muscle fibers, and this effect was greatest in Type IIA and IID/X fibers. In addition, mass was negatively correlated with citrate synthase and β -hydroxyacyl CoA dehydrogenase activity, but not with lactate dehydrogenase activity or myoglobin concentration. Findings suggest that muscle properties reflect underlying metabolic scaling constraints and indicate that mass should be considered when interpreting species or population-specific differences. Funded by NSF EPS-0346770.

38.4

STRUCTURE AND MECHANICS OF THE CETACEAN DIAPHRAGM AND ITS CONTRIBUTION TO THORAX PRESSURIZATION DURING A DIVE

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Over the first 100 m depth of a dive, the thorax of a cetacean is substantially compressed by the ambient pressure, reducing the air volume in the lungs and raising thoracic pressures. This depth is often traversed within 30 s, so the instantaneous thoracic pressures depend on how quickly the chest wall and diaphragm can deform. To better understand how thoracic pressures might be controlled, we examined diaphragm morphology and mechanics from several cetacean species. We hypothesized that the oblique angle of cetacean diaphragms affords large reductions of thorax volume under dorso-ventral compression of the body. However, its larger radius of curvature exposes the diaphragm to larger stresses from trans-diaphragmatic pressures, making diaphragm deformation and thoracic pressures more difficult to control. We found that cetacean diaphragms lack the muscle-free central tendon of terrestrial mammals. Instead the muscle is continuous over the entire structure and is reinforced with superficial collagen fibres. Mechanical modeling based on geometry from CT scans revealed the collagen fibres were oriented to resist the predicted dominant stresses. The continuity of the muscle may allow adjustment of the diaphragm curvature and so better control of the thoracic volume than could be effected with a central tendon. Supported by NSERC to RES.

38.5

SHRIMP EXOSKELETON MORPHOLOGY, MINERALIZATION, AND BIOPHOTONICS UNDER OCEAN ACIDIFICATION CONDITIONS

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While most marine calcifiers exhibit increased dissolution and decreased calcification rates under ocean acidification conditions, such effects have rarely been investigated in crustaceans. These common effects would, however, be deleterious for crustaceans, which depend on their calcified exoskeleton for many critical functions. The objective here was to determine the effect of CO₂-driven reduction in seawater pH on the structure, mineralization, and biophotonics of the exoskeleton of a common caridean shrimp, *Lysmata californica*. Shrimps were exposed to ambient (7.99 ± 0.04) and reduced pH (7.53 ± 0.06) for 21d, after which the exoskeleton morphology and mineralization were examined using SEM-EDX, and its transparency and reflectance analyzed with hyperspectral imaging. Data showed that the percent calcium content of the exoskeleton doubled in reduced pH conditions, but with no change in exoskeleton thickness, indicating an increase in the mineral/matrix ratio. There was also a 5- to 7-fold decrease in exoskeleton transparency, and a slight increase of reflectance intensity in some red areas of the exoskeleton in shrimp exposed to reduced pH. These results suggest that even short-term exposure to moderate reductions in pH can significantly alter the mineralization and biophotonic properties of the shrimp exoskeleton, with potential impacts on crypsis and survival. Funded by Scripps Institution of Oceanography and AFOSR MURI BIOPAINTS FA9550-10-1-0555 (to DDD).

38.6

EXERFLYZER: A HIGH-THROUGHPUT SYSTEM FOR INDUCING AND QUANTIFYING FLIGHT BEHAVIOR IN *DROSOPHILA* OVER EXTENDED TIME PERIODS

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Drosophila melanogaster has emerged as an important model for the role of muscle in metabolic homeostasis and systemic aging due to ease of genetic manipulation, and a high degree of evolutionary conservation among relevant pathways. In mammals, pathways implicated in sarcopenia, and metabolic diseases such as type II diabetes, are strongly affected by exercise. In *Drosophila*, flight requires a roughly tenfold increase in body-wide metabolic rate vs. rest; fivefold vs. walking. Therefore, to exploit the potential of *Drosophila* to uncover mechanisms of exercise effects, we have developed the exerFlyzer - an environmental chamber and imaging system to continuously quantify aggregate flight and walking behavior (spontaneous or induced) in large cohorts (hundreds) of flies for periods of days or weeks. With code developed using Matlab image acquisition and processing toolboxes, the exerFlyzer intermittently captures short sequences of images and identifies objects in motion between sequential frames, using velocity to differentiate between flight and walking. This method simplifies the data processing and eliminates the need for the sophisticated camera arrays used in tracking studies. Mechanical and visual stimuli are used to induce flight. The exerFlyzer enables one to vary and to control for muscle-driven metabolic load - an important effector of muscular signaling, and a behavioral metric relevant to a broad range of physiological processes conserved across multiple taxa.

38.7

AGE RELATED CHANGES IN FLIGHT MUSCLE ULTRA-STRUCTURES OF THE HAWK MOTH, *MANDUCA SEXTA*: A NOVEL NON-VERTEBRATE ANIMAL MODEL FOR INVESTIGATING VERTEBRATE SKELETAL MUSCLE FUNCTION, DISEASE, DEGENERATION, AND AGING

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Murine animal models are commonly utilized to gain insight into the pathogenesis of vertebrate diseases, or to evaluate the efficacy of potential therapeutic regimens. The need for novel non-vertebrate animal models for investigating vertebrate skeletal muscle physiology, disease, and aging is growing. More importantly, non-vertebrate animal models are facile, ethically acceptable, and inexpensive surrogates to model a variety of human skeletal muscle diseases. The flight muscle of the hawk moth, *Manduca sexta*, is metabolically similar to endothermic vertebrate skeletal muscle anatomy and physiology. We report the advantages of using *Manduca sexta* as a non-vertebrate animal model to study skeletal muscle physiology, disease, and aging. We compared ultrastructural changes of young (day 1), middle aged (day 3), and older (day 6) flight muscles of *Manduca sexta*. The flight muscle of older moths exhibited a degenerative process similar to that observed in skeletal muscle of aging murine models. The results suggest that the flight muscle of *Manduca sexta* is an ideal non-vertebrate animal model for investigating vertebrate skeletal muscle function, disease, degeneration, and aging.

38.8

EFFECT OF GRAVIDITY ON JUMP PERFORMANCE AND MUSCLE PHYSIOLOGY IN THE AMERICAN LOCUST

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Trade-offs exist between different physiological characteristics. In terrestrial animals, the trade-off between gravity (or load) on striding locomotion has been well documented. However, the effect of load on jumping performance is relatively unknown. In lizards, an increased load equal to 30% of body mass produced shorter and less frequent jumps. However in *Schistocerca americana* grasshoppers, 30% heavier gravid females have a similar jump performance to non-gravid females. We tested the hypothesis that grasshoppers vary their jumping muscle contraction duration to increase force produced when gravid (or loaded). We used high-speed video analysis to compare jump performance and electromyography to measure jumping muscle contraction times. Controls indicated that a 30-minute rest after electrode implantation was sufficient for the grasshoppers to regain their jumping ability. Results indicate that muscle contraction duration increases with load. Gravid grasshoppers show a 30% increase in contraction time compared to non-gravid females. We examined the effect of added mass on jump performance by attaching weights (20% or 40% of body mass) to non-gravid females and gravid females. When weights equal to 40% of body mass are added, muscle contraction duration increased over 50% in non-gravid females but did not change in gravid females. These results suggest that gravid females are already at their limit for muscle contraction duration. Unlike lizards, grasshoppers may show no effect of gravity on jump performance because they use a catapult mechanism to store energy prior to jumping. Support was provided by Union College Student Research Grants.

39.0: OSMOTIC AND IONIC REGULATION

39.1

POTENTIAL SOLUBLE ADENYLYL CYCLASE ISOFORMS IN CORAL

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Soluble adenylyl cyclase (sAC) is a bicarbonate-stimulated source of the ubiquitous signaling molecule cAMP. sAC genes have been identified in diverse phyla, suggesting it is an evolutionarily conserved CO₂/pH/HCO₃⁻ sensor. We are investigating the role of sAC in corals, where physiological processes such as photosynthesis and calcification depend on and alter internal acid/base conditions. The *Acropora digitifera* genome contains a sAC gene encoding for a ~190 kDa protein. However, using RT-PCR, we have amplified several sAC transcripts from the related coral species, *Acropora yongei*. These sequences indicate the presence of at least one splice variant, encoding for a truncated 68kDa protein that includes both catalytic domains, and several other insertions/deletions/polymorphisms. Whether such differences represent unique sAC genes or variations of a single gene (alleles or splice variants) will be investigated using Rapid Amplification of cDNA Ends (RACE) PCR. Alternative splicing of the coral sAC gene, known to occur in mammalian systems, suggests multiple physiological roles of sAC in corals. Interestingly, immunostaining using coral sAC-specific antibodies reveals that sAC is present in multiple cell types throughout coral epithelia; we hypothesize that splice variants could be related to cell-

type specific functions and may contribute to distinct cellular processes. This research was funded by an NSF GRFP fellowship.

39.2

DIFFERENTIAL LOCALIZATION OF ION-TRANSPORTING PROTEINS SUGGEST SPECIES-SPECIFIC PHYSIOLOGICAL MECHANISMS IN CORALS

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The movement of ions across a cell membrane is critical to a wide variety of physiological functions across all phyla, however, little research has been conducted on ion transport in Cnidarians. Understanding coral physiological mechanisms is of increasing interest, however, due to mounting environmental stress. Two key ion transport dependent processes are calcification, and photosynthesis by the endosymbiotic algae. Here, we utilized fluorescent immunostaining to localize three key ion transport proteins in two species of scleractinian corals: *Acropora yongei* and *Sylopora pistillata*. Proteins investigated included sodium-potassium ATPase (NKA), plasma membrane calcium ATPase (PMCA), and sodium-bicarbonate cotransporter (NBC). PMCA, NKA and NBC were each observed in the calcifying epithelium of both species. However, other coral tissues demonstrated species-specific localization patterns: both NKA and NBC were found in the oral ectoderm of *A. yongei* only, NKA being on the apical membrane, while PMCA was found in the endoderm of *S. pistillata*, only. The similar localization of these proteins in the calcifying tissue suggests they play a conserved role in calcification, while the differences in location between other tissues indicates that corals have evolved diverse ion transport mechanisms for other functions. These differences may lead to differential responses to environmental stressors. National Science Foundation grants EF-1220641 to MT and OCE-1226396 to KB.

39.3

EXTREME STRESS TOLERANCE OF THE INTERTIDAL TARDIGRADE *ECHINISCOIDES SIGISMUNDI*

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Tardigrades may likely be the toughest animals on Earth. These microscopic ecdysozoans endure prolonged periods of environmental extremes by entering dormant states, such as cryptobiosis (latent life). The aim of this study was to investigate stress tolerance in the species *Echiniscoides sigismundi*, which lives in marine tidal zones across the globe. Tardigrades were monitored in groups of 20 animals using stereomicroscopes and considered active and alive if they showed muscle movements. Freeze tolerance experiments revealed that *E. sigismundi* tolerates freezing to -30°C, -78°C and -196 °C (liquid nitrogen) for 2 weeks with a mean±sd survival of 95±5%, 100±0% and 64±15% (n≥3). Notably, tardigrades transferred to -30°C prior to submersion in liquid nitrogen had a survival of 99±3% (n=5), suggesting a clear effect of cooling rate. Experiments on osmotic stress revealed an extraordinary high tolerance to changes in external salinity. *E. sigismundi* tolerates exposure to distilled water for at least 4 days, as well as 48 hours exposures to salinities ranging from 1.6‰ to 245‰. The tardigrade further tolerates desiccation for 48 hours from seawater as well as distilled water, with a survival of 92±5% and 90±7% (n≥5). Collectively, our results show that *E. sigismundi* is highly tolerant of environmental perturbations, and as such, provides a unique model to gain further insights into latent life phenomena and adaptations to extreme environments. Supported by the Carlsberg Foundation.

39.4

BRANCHIAL CHAMBER OF *LITOPENAEUS VANNAMEI* DURING POST-EMBRYONIC DEVELOPMENT

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The crustaceans have a branchial chamber (BrCh) that includes gills, branquioestegite, epipodites and pleura. All these structures are necessary to perform osmoregulatory activity during shrimp's life. In order to understand the transition and physiological implications of these structures during *Litopenaeus vannamei* ontogenesis, we examined the cell topography of BrCh and the morphogenesis of gills and epipodites through histology (either H & E staining and direct optical visualization, 1-100X) in different stages of development (n=3-10). Adult *L. vannamei* has 18 gills, 5 epipodites, and branquioestegites. BrCh is continuously improving from larvae to PL13 and is fully developed in PL15. The first epipodite was observed in Mysis 1, while the others were present only as buds of epipodites through PL6. The gills were appeared until PL1 like a simple tube. Cellular differences were observed between structures, because the function can be variable between developmental stages.

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39.5 ALLATOSTATIN A-LIKE FACTORS IN THE AQUATIC LARVAE OF *CHIRONOMUS RIPARIUS*: REGULATION OF HINDGUT MOTILITY, ION REABSORPTION AND IMPLICATIONS FOR SALINITY EXPOSURE

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The aquatic larvae of the non-biting midge *Chironomus riparius* are ubiquitous benthic inhabitants of the northern hemisphere. Larval *C. riparius* exhibit tolerance to waters with a wide range of ionic composition from ion-poor fresh-water to salinated brackish-water. We recently discovered that decreased activity of ionomotive pumps in the hindgut epithelium are at least in part responsible for long-term salinity tolerance where ion transport (absorption) is down regulated. In this study we provide evidence for the presence of Allatostatin A-like neuropeptides in the larvae of *Chironomus riparius* and demonstrate their physiological activity on the hindgut. Allatostatin A-like immunoreactive (AST-IR) cells and processes were present in the nervous system and gut. The terminal ganglion contained 4 AST-IR cells which gave rise to axons that projected onto the hindgut and posterior midgut. Application of a cockroach AST to the semi-isolated hindgut of larval *C. riparius* led to dose-dependent inhibition of muscle contractions with an EC₅₀ of ~10 nM. AST also decreased ion transport at the hindgut measured as K⁺ flux which coincided with an AST induced reduction in the activity of ionomotive pumps. The effects of AST on hindgut ion transport are similar to those observed over long term exposure to salinity. Funding: Natural Sciences and Engineering Research Council of Canada, Ontario Ministry of Research and Innovation.

39.6 SODIUM AND AMMONIA TRANSPORT IN *AEDES AEGYPTI* LARVAE REARED IN VARYING WATER NA LEVELS.

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Larval mosquitoes can survive and complete development over an extremely wide range of water chemistries indicating impressive phenotypic plasticity in their ion transport capabilities. *Aedes aegypti* mosquito larvae can maintain hemolymph ion balance in water varying from 0 to ~150mM Na. Previous studies have identified anal papillae as the main site of Na uptake and ammonia excretion with possible coupling of the two processes. In two Research Methods in Physiology classes (Biol 309), undergraduate students characterized Na uptake and ammonia excretion *in vivo* in freshwater larval *A. aegypti* hatched and reared in 60 and 1000 µM Na media. Na uptake kinetics revealed that larvae reared in 60 µM Na possess a higher maximum capacity transporter with a higher affinity for Na compared to larvae reared in 1000 µM Na medium. In their respective rearing conditions, Na uptake rates were significantly lower in the 60 versus 1000 µM Na, suggesting a greatly reduced diffusive Na efflux and hence permeability. Protein expression studies confirmed the presence of a basolateral NaK ATPase and apically located V-type H ATPase and NHE3 on the anal papillae. Ammonia excretion was inhibited by ouabain in both groups, however, Na uptake was inhibited only in the 1000 µM Na reared larvae and only by ouabain plus taurocholate, indicating the presence of an organic anion transporting protein (oatp) that can transport ouabain away from NaK ATPase and induce ouabain resistance.

39.7 RNAI KNOCKDOWN OF AMMONIA TRANSPORTER AND OSMOREGULATORY TRANSCRIPTS IN LARVAL MOSQUITOES, *AEDES AEGYPTI*

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Our understanding of ammonia excretion and osmoregulatory mechanisms in aquatic insects has been greatly enhanced by pharmacological inhibition of transporters and channels that are involved in ammonia, water and ion movement. Characterization by pharmacological means however may not account for the functional contributions of multiple protein isoforms. RNA interference (RNAi) techniques, which use double-stranded RNA (dsRNA) to mediate gene silencing, can help circumvent this concern by generating a loss-of-function phenotype that can be linked to a specific gene transcript. In this regard, we will use RNAi in the present study to pinpoint potential non-redundant functions of transport and channel protein isoforms that are known to be involved in the osmoregulation and ammonia excretion of larval *A. aegypti*. dsRNA designed to target ammonia transporter and aquaporin transcripts will be delivered to larvae orally and knockdown efficiency of each gene will be determined by quantitative real-time PCR. Scanning ion-selective electrode technique (SIET) and

gut permeability assays will be used to evaluate changes in transport as a result of gene silencing. In addition to providing valuable insight into gene function, we anticipate that this research will provide supporting evidence for current models of ammonia excretion and osmoregulation in aquatic larval insects. Funding: Natural Sciences and Engineering Research Council of Canada, Ontario Ministry of Research and Innovation.

39.8 FUNCTIONAL CHARACTERIZATION OF HETEROLOGOUSLY EXPRESSED *DROSOPHILA MELANOGASTER* ORGANIC CATION TRANSPORTER ORCT IN *XENOPUS LAEVIS* OOCYTES

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Organic cations (OCs) include endogenous metabolites and xenobiotics that must be eliminated from the organism to ensure survival. The midgut and Malpighian tubules of insects have been shown to be involved with the active transport of OCs. Two putative organic cation-like transporters (OCTs), named *orct* and *orct2*, have previously been cloned from adult *Drosophila melanogaster*. qPCR revealed the mRNA transcripts of *orct* and *orct2* were differentially expressed in the midgut and Malpighian tubules. Moreover, mRNA expression of these transcripts increased following exposure to prototypical type I OC, tetraethylammonium (TEA). Gene expression patterns coincide with previous physiological research conducted, which showed the capacity of insect excretory tissues to transport TEA. *Drosophila* ORCT was expressed in *Xenopus* oocytes, where ORCT transport kinetics were analyzed using [¹⁴C]-labeled TEA. ORCT-mediated TEA uptake was saturable. Maximal transport capacity (J_{max}) and transport affinity for mediated TEA uptake (K_i) were 5 µM per oocyte and 0.33 mM, respectively. TEA uptake was inhibited by type I and type II OCs. Quinine and verapamil inhibited TEA influx by 33 and 43%, respectively, whereas cimetidine and vinblastine did not reduce TEA uptake. This is the first study to provide the functional characterization of a heterologously expressed *insect orct* in *Xenopus* oocytes. This work was supported by an NSERC grant to MRR.

39.9 EFFECTS OF DIURETICS ON RENAL CALCIUM OXALATE CRYSTALLIZATION IN A *DROSOPHILA* (FLY) MODEL OF OXALATE NEPHROLITHIASIS

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Nephrolithiasis is a painful, expensive health care problem with complex genetic and environmental etiologies. Calcium oxalate (CaOx) accounts for 70% of human cases, yet details of stone formation are poorly understood. As dilute urine appears to lessen stone occurrence, diuretics are used clinically as treatment. Using comparative approaches, we are dissecting this process. A GWAS analysis of miniature schnauzers (40% CaOx stone incidence v. 1% in canines/humans) revealed genes associated with CaOx stone formation. Zn is present in Randall Plaques, associated with human CaOx stones, and Slc39a10 (Zip10, Zn transporter) was one dog gene identified. Our group has established a fly model of CaOx crystal formation, providing simple/fast approaches for this complex condition. In this study, Malpighian tubules (MT) were incubated with NaOx±Zn to quantify CaOx crystallization. NaOx alone and NaOx+Zn increase average crystal size in wild-type (WT) and Zip10 knock-down flies. However in the presence of NaOx+Zn, knock-downs exhibit an increase in crystal number and total crystal formation compared to NaOx alone indicating a role for Zip10 in CaOx crystallization. We sought to determine if furosemide (F) and hydrochlorothiazide (HTC) affect CaOx crystal formation in the fly MT. The data imply that HTC decreases crystal number, but increases total crystal area. Future experiments will address if this effect is associated with MT water transport or effects on oxalate transporters.

39.10 CONSERVATION OF THE OSMOSENSITIVE AND THERMOSENSITIVE AN-TRPV1 ION CHANNEL IN OSMOREGULATING ANIMALS

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Transient receptor potential (TRP) channels have been involved with the complex ways in which metazoans sense stimuli around them. They are implicated in vision, thermosensation, hearing and mechanosensation among many others. Several are activated by more than one different stimulus, which are then integrated in the gating step that allows the ions to pass through the channel's pore. The superfamily is divided in 6 subcategories, and even though one member has been described in yeasts, all the

others appear in more complex multicellular organisms, such as nematodes, insects and all kind of vertebrates. Recently, we have cloned from rat Supraoptic Nucleus (SON) a variant form of TRPV1, a member of the Vanilloid subfamily. This variant lacks part of the N-terminus (Δ N-TRPV1). Heterologous cells transfected with Δ N-TRPV1 expressed a capsaicin-insensitive channel that was activated by cell shrinking or physiological increases in temperature. Based in multiple alignments of TRPV1 sequences, we have predicted the conservation through vertebrates of Δ N-TRPV1. This hypothesis has been corroborated by the detection of five variant homologs, in fish, amphibians, birds and mammals including human. The high conservation of Δ N-TRPV1 on osmoregulating animals and the evidence pointing for this specific role detecting extracellular fluid osmolality changes, suggests that this variant is a key piece in the machinery that has allowed animals to adapt to and conquest dry land.

39.11

THE ROLE OF PULSATILE UREA EXCRETION IN CHEMICAL COMMUNICATION AND PREDATOR AVOIDANCE IN GULF TOADFISH (*OPSANUS BETA*)

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In response to bottlenose dolphin (*Tursiops truncatus*) foraging vocalizations, Gulf toadfish (*Opsanus beta*) have been shown to have elevated levels of plasma cortisol and decreased vocal activity, suggesting a stress response and a predator avoidance strategy (2). Toadfish have the unique ability to pulse urea across their gills, and we hypothesize that this pulsatile urea excretion may also play a role in the predator avoidance strategy. Urea has been found to serve as a cloaking molecule when co-excreted with ammonia (1) and is suggested to be involved with chemical communication (3). Neuroendocrine responses that control urea production and excretion will be compared in toadfish exposed to dolphin vocalization playbacks and control fish exposed to snapping shrimp sounds. Expression of the toadfish serotonin 5-HT_{2A} receptor, the toadfish urea transporter (tUT) and urea cycle enzymes will also be measured. Future experiments will concurrently measure urea excretion and vocal activity to determine, ultimately, if toadfish switch from vocal to chemical communication to avoid predation. References: 1. Barimo JF et al. (2006) J Exp Biol 209, 4254-4261 2. Remage-Healey L et al. (2006) J Exp Biol 209, 4444-4451 3. Sloman KA et al. (2005) Physiol Biochem Zool 78, 724-735. Supported by NSF Grant #IOS-0920547.

39.12

CHARACTERIZATION OF THE SEROTONIN TRANSPORTER (SERT) AND EFFECTS OF FLUOXETINE TREATMENT ON SERT IN THE GULF TOADFISH, *OPSANUS BETA*

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Recent studies have demonstrated that fluoxetine (a popular selective serotonin reuptake inhibitor and the active ingredient in Prozac) can be detected in surface waters near wastewater effluent sites, and therefore has the potential to affect aquatic organisms. The objectives of the current study were to characterize the toadfish serotonin transporter (SERT) on the molecular level and elucidate the effects of fluoxetine treatment on SERT mRNA expression and function *in vivo*. The full-length sequence of toadfish SERT (2kb) was confirmed via PCR using primers designed from available sequence data and shows 75% identity to a known SERT sequence of another teleost. Current work is investigating SERT mRNA expression patterns in toadfish tissues under control conditions and fluoxetine treatment. Interestingly, fluoxetine treatment appears to affect metabolic control of oxygen consumption under conditions of hypoxia, as there is a significant ($p < 0.05$) difference in the regulation index (RI), or degree of metabolic regulation, between fluoxetine-treated (mean RI = 0.2013, SD = ± 0.24 , n = 8) and untreated fish (mean RI = 0.5241, SD = ± 0.27 , n = 7). Whether this difference can be attributed to changes in SERT upon fluoxetine exposure will be discussed. Ongoing work seeks to examine the transport kinetics and pharmacological sensitivity of toadfish SERT using *Xenopus laevis* oocytes to express isolated toadfish SERT cRNA. Supported by the UM Fellowship.

39.13

THE ROLES OF GLUTAMATE AND PUTRESCINE IN γ -AMINO-BUTYRIC ACID (GABA) SYNTHESIS IN *FUNDULUS HETEROCLITUS* DURING OSMOTIC STRESS

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The inhibitory neurotransmitter γ -aminobutyric acid (GABA) has been implicated in cell volume regulation during osmotic stress via its action on the GABA_A receptor. Although GABA is typically produced from glutamate by glutamate decarboxylase 1

(GAD1), the polyamine putrescine (PUT) can potentially serve as an important precursor of GABA during abiotic stress. Interestingly, euryhaline species of *Fundulus* accumulate PUT in the gill during acute freshwater exposure, suggesting that PUT may become increasingly important as a source of GABA during osmotic stress. The current study aims to compare the relative importance of glutamate and PUT in GABA synthesis in the mummichog, *Fundulus heteroclitus*, during acute hypo-osmotic exposure. *F. heteroclitus* acclimated to 12 ppt water were acutely transferred to 0.1 ppt water, and the gill, liver, intestine, kidney, muscle and brain were sampled after 6 h, 1 d and 7 d. Aminoxyacetic acid, a known inhibitor of GAD1, was administered to one control and one treatment group. The mRNA levels and enzymatic activities of two key enzymes in GABA synthesis; aldehyde dehydrogenase 9 family, member 1 and GAD1; were measured. In addition, the levels of GABA, glutamate, and the polyamines PUT, spermidine, and spermine were quantified. This study will assess whether GABA, in addition to its role in the central nervous system, plays an osmoregulatory role in the periphery of fishes. This research was supported by funding from the National Science Foundation (EF-0723771).

39.14

TIGHT JUNCTION PROTEINS AND THE REGULATION OF SALT AND WATER BALANCE IN AN EXTANT AGNATHAN, *PETROMYZON MARINUS*

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The sea lamprey (*Petromyzon marinus*) is an extant jawless vertebrate with a multi-staged life cycle comprising a freshwater (FW) dwelling filter-feeding larval (ammocoete) phase, followed by a complex metamorphosis which results in a parasitic juvenile that resides in seawater (SW) and feeds on the blood of fishes. While the larvae are acknowledged to be stenohaline FW animals, post-metamorphic sea lamprey are euryhaline, allowing for SW entry and an eventual return to FW for the terminal spawning phase of their life cycle. Basic strategies of lamprey osmoregulation appear to largely mirror those seen in more derived aquatic vertebrates such as teleost fishes and in this regard, ultrastructural observations of the lamprey tight junction (TJ) complex in ionoregulatory tissues strongly support the idea that this structure significantly contributes to overall salt and water balance. However, the molecular architecture of the lamprey TJ complex and its response to life stage related alterations in environmental ion levels is unknown. This study reports on the trans-membrane and cytosolic proteins that make up the TJ complex of the lamprey and how the molecular 'machinery' of the larval and post-metamorphic lamprey TJ complex responds to changes in the ionic strength of its environment. Funded by NSERC Canada

39.15

HOMOLOGOUS SERUM AND HEPARIN ALTER TIGHT JUNCTION PROTEIN ABUNDANCE AND THE PERMEABILITY OF A PRIMARY CULTURED SALMONID GILL MODEL

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Due to the architectural complexity and cellular heterogeneity of fish gills, primary cultured model gill epithelia are useful tools to investigate factors that contribute to salt and water balance in fishes. Specifically, culture models are helpful for experimentally dissecting the role of tight junction (TJ) proteins in the regulation of gill epithelium integrity. However, despite model suitability it is prudent to consider refining techniques so as to better mimic *in vivo* conditions. For example, replacing a typically used media supplement such as fetal bovine serum (FBS) with native fish serum could produce a more authentic model by eliminating foreign proteins and introducing homologous growth factors. But native serum supplementation in salmonid cell culture has been reported with mixed success and in some cases has been described as cytotoxic. While fish plasma can be a replacement for serum in primary gill cell culture, convention suggests that plasma may lack some of the mitogenic properties of serum. Furthermore, the effect anti-coagulating agents (required for plasma preparation) on the properties of primary cultured epithelia have not been explored. This study considered the use of homologous serum as an FBS replacement in a culture trout gill epithelium model and its subsequent effect on permeability and the molecular biology of the gill TJ complex. In addition, the effect of heparin on the physiology of the gill model was also considered. Funded by NSERC Canada

39.16

REGULATION OF GILL CLAUDIN ISOFORMS IN MOZAMBIQUE TILAPIA (*OREOCHROMIS MOSSAMBICUS*) BY SALINITY AND CORTISOL

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¹Biological Sci., Univ. of Arkansas, SCEN 601, Fayetteville, AR, 72701, ²Hawaii Inst. of Marine Biology, Univ. of Hawaii, 46-007 Lilipuna Rd., Kaneohe, HI, 96744, ³Biology, Skidmore Coll., 815 N. Broadway, Saratoga Springs, NY, 12866. In euryhaline teleosts, reorganization of gill tight junctions during salinity acclimation results in part from dynamic expression of specific claudin isoforms. We identified five transcripts encoding tight junction proteins in the tilapia gill transcriptome: *occludin* (*occl*) and *claudin* (*cldn*) -10c, -10e, -28a and -30. Gill expression of these genes in Mozambique tilapia was examined during acclimation to freshwater (FW), seawater (SW) and in response to hormone treatment. Transfer of tilapia to SW elevated *cldn-10c* and *cldn-10e*, while *cldn-28a*, *cldn-30* and *occl* were stimulated during FW acclimation. In hypophysectomized tilapia transferred to FW, cortisol replacement stimulated *cldn-10c*, *cldn-10e* and *cldn-28a* expression, whereas no effect of cortisol was seen on *cldn-30* and *occl*. *In vitro* experiments with gill filaments showed that cortisol stimulated expression of all four examined *cldns*, suggesting a direct action of cortisol *in situ*. The data indicates that elevated *cldn-10c* and -10e expression is important during acclimation of tilapia to SW possibly by conferring ion specific paracellular permeability. On the other hand, expression of *occl*, *cldn-28a* and -30 appears to contribute to reorganization of branchial epithelium during FW acclimation. Hormone treatment experiments showed that both FW- and SW-induced *cldns* are under stimulatory cortisol control. Funding sources: NSF (IBN: 11-19693 and 12-51616), USDA (2008-35206-18785 and 18787), ABI.

39.17

PARACELLULAR PATHWAY REGULATION IN RESPONSE TO SALINITY CHANGES IN THE JAPANESE MEDAKA (*ORYZIAS LATIPES*)

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Claudins are tight junction proteins responsible for ion selectivity of the paracellular pathway in transport epithelia and thus essential in teleost osmoregulation. This study investigates responses of 13 claudins (*cldn*) to salinity challenges in the euryhaline Japanese medaka. Short- or long-term effects of freshwater (FW) and seawater (SW) transfer were examined. In the gill, *cldn-27a*, -28a, *b* and -30c did not vary with salinity which is consistent with a barrier-forming role. Putative cation-selective pore-forming *cldn-10a*, *c*, *d* and *e*, were all more abundant in SW than in FW gills. This is in agreement with the need of SW fish to secrete NaCl via trans- and para-cellular ions movements. Two *cldn-10b* splice-variants were predominantly expressed in the kidney, with higher expression in SW- than in FW-acclimated fish. This suggests a role in monovalent ion reabsorption, particularly essential in SW to produce isotonic urine enriched with divalent ions. *Cldn-15a*, *b* and -25 isoforms were mainly expressed in the intestine. *Cldn-15a* and -25 did not display any variation during salinity challenges. However, *cldn-15b* was highest in FW which suggests a specific role of this putative pore-forming tight junction protein in hyper-osmoregulatory mechanisms in the intestine of medaka. This work was approved by the Animal Care and Use Committee of the University of Arkansas (IACUC #14042) and supported by a NSF grant (IBN 12-51616) and the Arkansas Biosciences Institute.

39.18

EXPRESSION OF GILL Na⁺/K⁺-ATPASE α -SUBUNIT ISOFORMS IN EURYHALINE JAPANESE MEDAKA (*ORYZIAS LATIPES*) DURING SALINITY CHALLENGES

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Some euryhaline teleosts exhibit a switch in the Na⁺/K⁺-ATPase (NKA) α -isoform when migrating between/exposed to (FW) and seawater (SW). The objective of the present study was to identify NKA α -isoforms in medaka and define their role in gill osmoregulation. We classified 6 NKA α -isoforms (*ala*, *alb*, *alc*, *a2*, *a3a* and *a3b*) and investigated salinity dependent expression. In a tissue distribution screening we found that *ala*, *alb* and *alc* were highly expressed in osmoregulatory organs. Long-term acclimation of medaka to FW and SW induced no change in gill expression of *a2*, *a3a* and *a3b*. Thus only *al* isoforms were further investigated. In both short-term and long-term salinity challenges, NKA *ala* and *alb* were up-regulated in SW gill while no effect of salinity was observed for *alc*. For reference some additional transport proteins were examined. Gill *ncc* and *nhe3* expression were elevated in FW, while *cfr* and *nkcc1a* were up-regulated in SW. This supports findings in some other teleosts and is in accordance with putative roles in ion-uptake and -secretion, respectively. Specific FW and SW *al* isoforms have been described in teleosts such as trout and tilapia. The present study suggests that in medaka no such NKA switch is involved in salinity acclimation. The enzyme kinetics of gill NKA in FW- and SW-acclimated medaka are currently under investigation. This study was supported by Arkansas Biosciences Institute.

39.19

FOUR Na⁺/K⁺-ATPASE α -SUBUNIT ISOFORMS IN THE THREE ELECTRIC ORGANS AND THE SKELETAL MUSCLE OF THE ELECTRIC EEL

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The electric eel, *Electrophorus electricus*, possesses three different electric organs (EOs) and is capable of generating both high and low voltage discharges. While many studies have focused on channels/transporters involved in bioelectrogenesis, few studies have examined mechanisms involved in the restoration of resting potential and the related recharging process. Na⁺/K⁺-ATPase (*nka*) is one of the key transporters involved in restoring the resting potential. We have identified for the first time four different *nka* α -subunit isoforms (*nkaa1b.1*, *nkaa1b.2*, *nkaa2* and *nkaa3*) from the main EO, Hunter's EO, Sach's EO and skeletal muscle (SM) of *E. electricus*. Molecular characterization of these isoforms indicated that they might have different Na⁺ binding affinities. Studies on the mRNA expression levels of the isoforms showed that the main and Hunter's EOs had significantly higher levels of *nkaa2* than the SM, while the SM had significantly higher mRNA expression of *nkaa1b.1* and *nkaa3*. Overall, our results indicate that *E. electricus* expressed EO-specific Nkaa and SM-specific Nkaa, and the protein abundance of Nkaa and the kinetic properties of Nka varied among its three EOs. This study was approved by the NUS IACUC, and funded by MINDEF through TMSI.

39.20

CHARACTERIZING SODIUM UPTAKE IN THE BLACKSKIRT TETRA (*GYMNOCORYMBUS TERNETZI*)

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The blackskirt tetra, (*Gymnocorymbus ternetzi*) is a native fish of the Rio Negro, a tributary of the Amazon River, which is ion-poor and very acidic (pH<5). This species possesses a sodium uptake mechanism that is quickly upregulated during low pH exposure so as to prevent net loss of ions from their blood to the water due a stimulated efflux. Immunolocalization studies confirm the presence of both Na⁺/K⁺ ATPase and V type H⁺ ATPase transporter proteins in the blackskirt tetra gills and that their location shifts to a more protected region of the gill when fish are exposed to low pH water. Western blotting indicates that the quantity of both ATPase proteins is unchanged yet activity assays show that both ATPase activities are low and drop to zero by 24 h at low pH. Isotopically measured sodium uptake was insensitive to both Ouabain and Bafilomycin exposure. Protein expression assays indicate the presence of an epithelial sodium channel (eNaC) and that a large component of the low pH-stimulated Na⁺ influx is phenamil sensitive. We propose a novel coupling mechanism of the basolateral ouabain-insensitive isoform of Na⁺/K⁺ ATPase with an apical (water facing) eNaC transporter. Funding was provided by the ALSAM and Doheny foundations and the USD Office of Undergraduate Research.

39.21

THE EFFECTS OF WATER IONIC COMPOSITION ON THE RATE AND DEGREE OF ACID-BASE REGULATION IN RAINBOW TROUT, *O. MYKISS*, DURING HYPERCARBIA AT REST AND SUSTAINED EXERCISE

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Hypercarbia (elevated environmental CO₂) is an environmental factor that can have significant physiological consequences in animals, particularly those in aquatic habitats. Exposure to hypercarbia can elicit a potentially lethal blood acidosis to which an animal must compensate in order to maintain homeostasis. Although not fully understood, the exchange of environmental Na⁺ and Cl⁻ with their acidic and basic counterparts (H⁺ and HCO₃⁻, respectively) plays an important role in pH compensation. The rate and degree of pH recovery during hypercarbia has been linked to several factors including ambient ion availability and possibly an animals activity level. However no study to date has directly quantified these relationships within a single species. Blood parameters of softwater, artificial hardwater and saltwater acclimated rainbow trout exposed to hypercarbia were used to elucidate the rate and degree of pH recovery among the aforementioned conditions. Both hardwater and saltwater trout were able to compensate for the hypercarbic disturbance faster than their softwater counterparts, and exercising fish demonstrated faster pH recovery compared to resting fish. Funding: NSERC

39.22

GUT CARBONATE EXCRETION BY FISH INCREASES EXPONENTIALLY WITHIN NATURAL OCEAN SALINITY RANGE (25-40 PSU)

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Marine teleost fish precipitate ingested Ca^{2+} ions in their intestine as calcium carbonate crystals. These are then excreted into the environment where they have the potential to impact global ocean chemistry and carbonate sediment formation. It has previously been estimated that up to 45% of all marine calcium carbonate production could be from fish (Wilson et al., 2009; *Science*, 323: 359-362). One environmental variable not previously considered in this estimate is salinity. This study aims to assess how salinities across the natural oceanic range could affect the intestinal precipitation of calcium carbonate by fish. Euryhaline shanny (*Lipophrys pholis*) and stenohaline turbot (*Scophthalmus maximus*) were maintained at salinities of 25, 30, 35 and 40 psu. For both species the carbonate production rates were relatively stable between 25 and 35 psu (the global average for ocean salinity) but increased 3-fold at 40 psu. For both species this was unrelated to metabolic rate changes across the salinities. The mechanism behind such a large increase in carbonate production rate over a small salinity change is uncertain, but is likely explained by relative changes in drinking rate (for osmoregulation) with salinity and intestinal Ca^{2+} absorbed for skeletal growth (rather than precipitated and excreted). If this is true for other marine fish it will have a major influence on estimates of global carbonate production by fish. Supported by the Natural Environment Research Council, UK.

39.23

PHENOTYPIC PLASTICITY IN RESPONSE TO HYPERCAPNIA INDUCED ACID-BASE DISTURBANCES IN RED DRUM (*SCIAENOPS OCELLATUS*)

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Anthropogenic CO_2 emissions since the pre-industrial era have raised oceanic CO_2 by 40%, reducing pH by 0.1 units. This can induce acid-base disturbances in marine organisms that are compensated through regulatory pathways. We assessed the plasticity of regulatory pathways in red drum (*Sciaenops ocellatus*) gills in response to hypercapnia induced acid-base disturbances. Red drum were exposed to ocean acidification (OA) levels (1000 μatm) for 24h, 72h, and 14d with concurrent controls as well as 5000 μatm for 0h, 4h, 8h, 24h, and 48h to assess plasticity in response to strong acid-base disturbances. Gill samples were analyzed for 7 genes (NHE1, NHE2, NHE3, CAc, NBC, slc26a3, slc26a5) thought to be involved in acid-base regulation via qPCR. No significant changes in gene expression were observed with the exceptions of NHE1, which transiently down-regulated in OA, and NHE3, which down-regulated after 48h in 5000 μatm . Results show that red drum do not show phenotypic plasticity in response to acid-base disturbances. As an estuarine species that regularly undergoes environmental fluxes in CO_2 , red drum may naturally maintain high baseline levels of machinery to compensate for hypercapnia induced acid-base disturbances. Further work is being conducted to identify a comprehensive physiological mechanism for hypercapnia compensation in red drum. All experiments were performed in accordance with the UT Institutional Animal Care and Use Committee. Funding provided by NSF (EF 1315290).

39.24

CHANGES IN EXPRESSION OF AQUAPORIN ISOFORMS IN AN AESTIVATING AFRICAN LUNGFISH

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African lungfishes can undergo long periods of aestivation on land during drought. During aestivation, they are confronted with desiccation and the impediment of ammonia excretion leading to increased urea synthesis and accumulation. Aquaporins (AQP) are a superfamily of integral membrane proteins that generally function for the selective passage of water or glycerol, and some AQP could facilitate ammonia transport. This study aimed to clone and sequence *aqp* from various tissues/organs of *Protopterus annectens* and examine their mRNA expression in these tissues/organs during the induction, maintenance and arousal phases of aestivation. *P. annectens* expressed six isoforms of *aqp* and aestivation led to changes in the mRNA expression of some *aqp* isoforms in certain tissues/organs. These results provided insight into how aestivating *P. annectens* regulated water channels to cope with desiccation during the induction and maintenance phases and rehydration during the arousal phase. They also shed light on the possible role of *aqp* in facilitating ammonia transport in *P. annectens*. This study was approved by the NUS IACUC.

39.25

EFFECTS OF TEMPERATURE ON HYPOTONIC SWELLING INDUCED BY WATER AND GLYCEROL IN HEPATOCYTES FROM COPE'S GRAY TREEFROG

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The freeze tolerant treefrog *Hyla chrysoscelis* accumulates glycerol during cold acclimation. When extracellular water freezes or thaws, cells must survive volume changes induced by transmembrane gradients of glycerol and water. We predicted that cold acclimation would induce enhanced permeability to glycerol (to facilitate its uptake) and to water (to facilitate osmotic equilibration between intra- and extra-cellular fluids). To test this, we measured cell volume change in isolated hepatocytes exposed to inward directed gradients of glycerol or water at either 5°C or 20°C. For cells from both warm-acclimated (WA) and cold-acclimated (CA) animals, cell swelling induced by a water or glycerol gradient was faster at 20°C than 5°C. We were not able to detect an effect of acclimation temperature on permeability (swelling rate) or on maximum cell size induced by swelling. However, WA animals better survived hypo-osmotic swelling, whereas CA animals better survived glycerol-induced swelling. 0.3 mM HgCl_2 , which blocks most aquaporins that are likely to facilitate water and glycerol permeability, was more effective at suppressing cell swelling induced by glycerol than by water. Our results suggest that properties of both membrane lipids and integral proteins contribute to the long- and short-term effects of temperature on water and glycerol permeability, and that water and glycerol transport occur at least in part via separate pathways. Supported by NSF IOS-1121457.

39.26

CARDIAC PH REGULATION AND BUFFERING OF THE WESTERN PAINTED TURTLE (*CHRYSEMYS PICTABELLI*)

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The Western painted turtle (*Chrysemys picta bellii*) spends its winters below the ice where oxygen sources are depleted, during which they rely on anaerobic metabolism. During anoxia, the heart accumulates the highest lactates and has the lowest intracellular pH of any organ, while cardiac function, itself, is depressed. To determine whether the buffering power of the painted turtle ventricle is unique from other vertebrates, we used fluorescence microscopy and ammonium chloride pre-pulsing to measure the pH-dependence of total and intrinsic buffering power in isolated ventricular myocytes. Our data show that painted turtle ventricular myocyte intrinsic buffering is lowest (9-2 mequiv L^{-1}) at the pH ranges that occur during anoxia (pH 7.2-6.55) and is highest (62 mequiv L^{-1}) at normal and alkaline pH's (greater than 7.2), which differs from mammals, where the opposite is observed. The same trend is seen with bicarbonate dependent buffering, where buffering power drops from 40 mequiv L^{-1} at pH 7.2 to 0.5 mequiv L^{-1} at pH 6.8. A decrease in bicarbonate dependent buffering has been shown in mammals, but is substantially lower in the turtle. This leads to a total buffering of between 49 mequiv L^{-1} and 2.5 mequiv L^{-1} during anoxia. We suggest that a decrease in buffering power at a low pH could be an advantage for the turtle, as H^+ are allowed to accumulate more quickly, which will have the effect of preventing cross-bridge cycling and preserving ATP. This study was funded by the National Science Foundation.

39.27

CONTROL OF THE OSMOTIC FUNCTION OF THE AVIAN LOWER GASTROINTESTINAL TRACT

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Unlike mammals, the urine of birds is not "stored" in a bladder until it can be voided. The urine of birds enters the terminal portion of the gastrointestinal tract (GI), the cloaca. It does not remain here to be excreted, but is moved by reverse peristalsis into the colon of the GI tract. Once in the colon, the composition of the urine is significantly modified. Previous experiments in our laboratory demonstrated that the reverse peristaltic movements are modulated by sensors with the cloaca and are sensitive to the composition of the urine. Urine with an osmolality 100 mOsm/ kgH_2O greater than the concurrent plasma is not refluxed into the colon. In attempts to identify the sensory elements in the cloaca that detect the osmolality of urine, Western blots and immunocytochemistry experiments were carried out on tissue from Mourning doves (*Zenaidura macroura*). Western blots were run on homogenates of the cloaca and the colon and immunocytochemistry was done on colon and cloaca that was separated in three segments (proctodeum, urodeum and coprodeum). The information from these experiments strongly suggested that the osmoreceptor was the vanilloid type receptor TRPV4, a moderately selective calcium channel. Moreover, it appeared that the greatest density of this receptor was located within the urodeum, the region of the cloaca that receives the urine from the kidney. A model is proposed that suggests the function of this system in the control of the reverse peristalsis.

39.28

EVIDENCE OF RAPID PHYSIOLOGICAL EVOLUTION IN AN INTRODUCED LAKE POPULATION OF THREESPINE STICKLEBACK

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Post-glacial diversification of Threespine Stickleback (*Gasterosteus aculeatus*) offers unique insight into how changes in halohabitat shapes physiological evolution in aquatic organisms. The presence of both ancestral marine/anadromous and derived freshwater-resident populations allow us to examine rates and direction of evolution following habitat transitions. Anadromous sticklebacks have also been recently introduced into several lakes in Alaska, facilitating study of divergence in osmoregulatory traits in real time. Our common-environment experiments between an anadromous population and landlocked descendant revealed that salinity tolerance has changed after only two generations of freshwater occupancy. We then examined ion transporters in the gill to clarify osmoregulatory mechanisms underlying differences in halotolerance. Na⁺/K⁺ ATPase activity was higher in seawater-challenged fish, but did not differ between populations. Preliminary immunoblotting results indicate that abundance of the Na⁺/K⁺-2Cl⁻ cotransporter increased in seawater, but the response was diminished in the lake population. This result suggests that freshwater invasion can result in rapid evolutionary change in the osmoregulatory system, including altered phenotypic plasticity. Funding provided by the John Rankin Scholarship Fund, Sigma Xi, and MVAO.

40.0: "OMICS" IN COMPARATIVE PHYSIOLOGY

40.1

ENHANCING THE RESTORATION OF CALIFORNIA'S ESTUARIES BY EXPLORING THE GENETIC BASIS OF ENVIRONMENTAL TOLERANCE IN OLYMPIA OYSTERS (*OSTREA LURIDA*)

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The Olympia oyster, *Ostrea lurida*, is the only oyster native to the west coast of N. America and a foundation species in estuarine habitats. Once abundant, *O. lurida* is now functionally extinct in the wild. As part of a plan to restore California's degraded estuary ecosystems, *O. lurida* is being considered for reintroduction. However, the ability of oysters to tolerate climate change may influence the success of restoration efforts. Oyster restoration should use genotypes capable of surviving future conditions, but which *O. lurida* populations will be most tolerant of climate change is uncertain. In San Francisco Bay, climate change will increase the frequency of freshwater flooding events that can cause mass mortality in oyster beds. In this study, tolerance of low salinity was explored in two *O. lurida* populations within San Francisco Bay and one population in nearby Tomales Bay. Oysters from Loch Lomond had significantly higher survival rates during freshwater challenge than Oyster Point or Tomales Bay populations. We are presently exploring mechanisms of differential salinity tolerance in *O. lurida* using RNA sequencing. Shifts in gene expression following exposure to reduced salinity are being compared among the three populations to determine physiological changes that underlie enhanced freshwater tolerance. Changes in allele frequency among oysters surviving freshwater challenge and controls are also being identified to understand evolutionary bases of salinity tolerance.

40.2

PROTEOMIC RESPONSES OF THE MUSSEL *MYTILUS GALLO-PROVINCIALIS* TO AERIAL EXPOSURE INDUCED HYPOXIA AND SIRTUIN INHIBITION

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Low tide in the rocky intertidal exposes marine organisms to environmental stressors including aerial emersion induced hypoxia (AEIH). Mussels in particular use alternative anaerobic pathways to combat hypoxia. The Mediterranean blue mussel *Mytilus galloprovincialis* has successfully invaded and displaced the native congener along the Pacific coast by being more physiologically tolerant to stressors. However, little is known in regard to this species' physiological responses to AEIH or how sirtuins, an NAD-dependent deacetylase, may influence their stress tolerance. The purpose of this study was to use a proteomics approach to assay global protein changes in mussels acclimated to two tidal regimes (constant immersion or immersion/emersion) and exposed to 24 h of one of three treatments: normoxia (constant immersion), AEIH, or AEIH and sirtuin inhibition (SI). We found abundance changes in proteins related to molecular chaperoning, the cytoskeleton, oxidative stress, energy metabolism, and protein degradation. In general, after 24 h of AEIH we found a decrease in abundance of non-essential proteins and an increase in abundance of molecular chaperones. SI alone and exposure to both AEIH and SI resulted in an increase in the proteins involved in proteolysis. This study highlights the need to not only study proteomic re-

sponses to stressors but to also identify possible mechanisms underlying stress tolerance, such as that by sirtuins (funded by NSF IOS 1146840).

40.3

THE PROTEOMIC RESPONSE OF SUBTIDALLY AND TIDALLY-ENTRAINED CALIFORNIA RIBBED MUSSEL *MYTILUS CALIFORNIANUS* TO ANOXIA STRESS

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Rocky intertidal organisms experience extreme shifts in abiotic factors, such as anoxia stress, due to tidal fluxes. During low tides, the California ribbed mussel (*Mytilus Californianus*) normally closes its shell to avoid desiccation instead of gaping to augment gas exchange, and thus faces anoxia stress and anaerobic metabolism. The energetic expense is partially met through a simultaneous down-regulation of metabolism. To characterize how entrainment to a tidal and subtidal rhythm affects protein synthesis in *M. californianus*, mussels were acclimated to tidal and subtidal conditions with a photoperiod (12h:12h) to mimic natural circadian rhythms. There were five groups including normoxic controls at 0h, 6h, and 72h and anoxic treatment groups treated with 100% nitrogen gas at 6h and 72h. Gill tissue was extracted and subjected to proteomic analysis so proteins could be identified using mass spectrometry. Of the constantly immersed mussels, only about 12% of proteins were significantly changing in contrast to the 17% of proteins for mussels acclimated to a tidal regime. Tidally-entrained mussels responded to anoxia by producing oxidative stress proteins after exposure to low oxygen conditions, especially at the 72h time point. The proteomic changes indicate that tidally-entrained mussels have a greater capacity to cope with acute anoxia than constantly submerged mussels. NSF IOS 1145840.

40.4

TRANSCRIPTOMIC ANALYSIS OF *DAPHNIA PULEX* RESPONSE TO INTERACTIVE EFFECTS OF TEMPERATURE AND SALINITY VARIABILITY

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Future climate scenarios are predicted to increase estuarine temperatures and salinities which will pose challenges for estuarine organisms. *Daphnia pulex*, a key primary consumer in estuarine foodwebs, have decreased survival and reproduction at temperatures above 15°C and salinities above 4. Multigenerational acclimation of *D. pulex* to variable temperature and salinity regimes conferred cross-tolerance to temperature and salinity stresses in low and moderate variation treatments (15°C, 0 and 15-30°C, 0). However, when acclimated to extreme variation temperature and salinity (15-30°C, 0-5), *D. pulex* exhibited cross-susceptibility and was less capable of responding to temperature or salinity stress. The mechanisms underlying these tolerance patterns were elucidated by employing a transcriptomics approach with these *D. pulex* specimens. This study examines cellular-level responses following acute temperature and salinity stress exposure (15°C, 0; 32°C, 7.4; 34°C, 8.8) in *D. pulex* acclimated to daily temperature and salinity spikes for 15 generations. We expect to see changes in gene expression between treatments that confer abilities to cope with heat and osmotic stress including differential expression of heat shock proteins and Na⁺/K⁺ ATPase pumps. This project was funded by the Berkeley Initiative for Global Change Biology.

40.5

TAKING A COMPARATIVE APPROACH TO UNDERSTANDING MICRORNA EXPRESSION AND ITS FUNCTIONAL CONSEQUENCES AFTER FLUOXETINE EXPOSURE IN TWO RELATED SPECIES, *CARASSIUS AURATUS* AND *DANIO RERIO*

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MicroRNAs (miRNA) are small non-coding RNAs that negatively regulate mRNA abundance and translation and contribute to the epigenetic control of gene expression. Fluoxetine (FLX), a selective serotonin re-uptake inhibitor and active ingredient in Prozac, is found in the environment and disrupts feeding, stress, and reproduction in fish by acting on the serotonin reuptake transporter. Female goldfish & zebrafish were exposed to environmentally relevant concentrations of waterborne FLX (540 ng/L) for 7 days. A custom designed miRNA microarray was used to assess the significantly regulated hepatic miRNA in zebrafish, which identified 6 specific miRNAs: let-7d, miR-22b, miR-140, miR-210, miR-301a and miR-457b. Using pathway analysis, miRNA were associated with the negative regulation of anabolic metabolism in zebrafish including adipogenesis, cholesterol biosynthesis, triacylglycerol synthesis, and insulin signaling. These specific miRNA were profiled in goldfish, a phylogenetic relative of the zebrafish, and results indicated a significant upregulation in all 6 miRNA under the same experimental conditions. Although miRNA sequences are

well conserved across species, their mRNA targets may be less conserved. To accomplish this comparison, RNASeq (Illumina) is used to obtaining the required 3'UTR information to generate predicted miRNA targets in goldfish. Using this strategy we will be able to determine conservation of target pathways and functional response between species.

40.6

WHOLE-GENOME METHYLATION PROFILING OF THREESPIN STICKLEBACK REARED IN HIGH AND LOW SALINITIES

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Epigenetic modifications, such as changes in DNA methylation, provide a unique connection between an organism's environment and the structure and function of its genome. Changes in DNA methylation patterns can result in phenotypic heterogeneity in genetically homogenous populations, and thus have the potential to be an important component of an organism's ability to colonize a novel habitat. While many aspects of DNA methylation mechanisms are well understood, there is limited understanding of the role of DNA methylation in natural populations, particularly in fishes.

Here, we use the threespine stickleback, *Gasterosteus aculeatus*, as a model system to explore the role of DNA methylation in the colonization of novel habitats. Populations of stickleback range from marine, to anadromous, to freshwater resident, and are thus exposed to differing environmental salinities. The association between repeated freshwater colonization events and conserved phenotypic changes among populations of stickleback presents an ideal system to study genotype/phenotype interactions. In this study, whole genome bisulfite sequencing was used to generate the first whole genome methylation profile of a stickleback. To characterize how DNA methylation patterns are impacted by salinity, clutches from marine sticklebacks were divided and fertilized in a brackish salinity representing their natural spawning environment, and a low salinity intended to represent colonization of a freshwater environment. From this data we identify differentially methylated genomic regions that may be important in the transition from marine to freshwater environments. Funding: NSERC.

40.7

DETERMINATION OF THE GILL TRANSCRIPTOME OF THE FAT-HEAD MINNOW (*PIMEPHALES PROMELAS*)

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RNASeq is a practical and accurate method for exploring the transcriptome and expressed proteins in non-model organisms. The fathead minnow (*Pimephales promelas*) is a widely used toxicology model and knowledge about its genetics will enhance further study into the interactions between this organism and its environment. We present the gill transcriptome of the rosy red strain of *P. promelas*. An Illumina HiSeq was used to produce 470,183,940 raw reads of 101 bp in length from prepared mRNA. After using the Broad Institute's Trinity software to perform a *de novo* assembly, 153,118 contigs were obtained with an average length of 997 bp and an N50 of 2081 bp. Annotation was performed with the Broad Institute's Trinotate package resulting in 72,334 unique blastx hits identified from the assembled contigs or 47.2% of the contigs. These transcripts were further classified by Gene Ontology (GO) terms, resulting in 53,845 (35.1% of contigs) unique transcripts mapping to 153 GO terms as well as the Kyoto Encyclopedia of Genes and Genomes annotation where 16,191 transcripts mapped to 6,948 unique KEGG orthology terms in 344 pathways. The gill transcriptome of the fathead minnow has been deeply sequenced and annotated allowing for further comparative genomic study as well as tentatively identifying expressed proteins. These experiments were conducted in accordance with the principles and practices set forth in the Declaration of Helsinki and the APS Guiding Principles in the care and Use of Animals.

40.8

HYPOXIA INDUCES GLOBAL REPROGRAMMING OF THE ACETYLOME IN THE ZEBRAFISH BRAIN

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In recent years it has become clear that changes in cellular energy status have major implications for the protein acetylome. Despite the relationship between hypoxia and redox status, little is known regarding the effects of low environmental oxygen on reversible lysine acetylation, and how a reprogrammed acetylome can modify mitochondrial metabolism. The objectives of this study were to elucidate the relationship between acetylation and redox status during hypoxia and examine their implications for mitochondrial function. To address these aims, we exposed zebrafish to 16 h of hypoxia and quantified acetylation levels in 1220 proteins in the brains of normoxic

and hypoxic zebrafish. Next, we analyzed redox status throughout the hypoxia time course and related this to activity of Sirt3, an NAD⁺-dependent deacetylase found in mitochondria. Finally, electron transport chain complex activities were assessed to determine the relationship between acetyl modifications and electron transport chain function. Our results (1) reveal large-scale reprogramming in the brain acetylome, with major modifications to mitochondrial targets; (2) show significant changes to the redox status within the brain during hypoxia; and (3) provide evidence that lysine acetylation may be related to large changes in complex activities and adenylate status in zebrafish exposed to hypoxic stress. Together, these data provide new insights into the role of protein modifications in mitochondrial metabolism. Funding provided by NSERC

40.9

EVOLUTION OF EMBRYONIC DIAPAUSE IN THE ANNUAL KILLIFISH *AUSTROFUNDULUS LIMNAEUS*: SEARCHING FOR CLUES IN THE GENOME AND EPIGENOME

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The annual killifish *Austrofundulus limnaeus* inhabits ephemeral ponds in regions of northern Venezuela, South America. Seasonal periods of drought kill all adult fish and permanent populations are maintained only through the production of stress-resistant embryos able to survive while buried in dried sediments. While the physiological changes necessary to promote survival in these harsh conditions have become clearer over the past two decades, little is known about the genetic and epigenetic changes that may have occurred in the killifish lineage to promote their unique life history. Here, we present the first draft genome and complete mitochondrial genome of a male *A. limnaeus* individual based on Illumina sequencing technology at >50X coverage. Four different DNA libraries were constructed and sequenced: one 180 bp fragment library, and three mate-pair libraries with mean insert sizes of 4, 8 and 12 kb. We compare *de novo* assembly results of the 1.5 billion short reads between different genome assemblers and determine assembly quality through mapping of RNA-seq data. In addition, we compare our assembly to genomes of other teleosts and explore heterozygosity and mitochondrial heteroplasmy in our lab population. Finally, we determine global changes in 5-methylcytosine content throughout *A. limnaeus* embryonic development by ELISA. These data will provide the framework for future genomic and epigenomic studies in annual killifish. Funding: NSF IOS-1354549.

40.10 Withdrawn.

40.11

EFFECTS OF OCEAN ACIDIFICATION ON JUVENILE ROCKFISH (*SEBASTES SPP*) GENE EXPRESSION

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Despite teleosts' high capacity for acid-base regulation, recent ocean acidification (OA) studies on tropical marine fish have documented negative physiological effects on growth and reproduction. Impacts on temperate fish, however, remain understudied. Our previous work showed that temperate rockfish reared under chronic OA conditions have reduced swimming performance, with juvenile blue rockfish (*Sebastes mystinus*) being more OA-tolerant than juvenile copper rockfish (*S. caurinus*). To investigate potential underlying mechanisms leading to differences in OA susceptibility, we compared rockfish transcriptomes after chronic exposure to predicted end-of-century pH levels (7.2, 7.5, 7.8, 8.0). We extracted total RNA from white muscle tissue and prepared cDNA libraries for RNAseq. We assembled a copper *de novo* transcriptome using Trinity, mapped sequences using RSEM, and examined differential gene expression (DE) using edgeR (FDR < 0.05). We identified 100s of significant DE genes among pH treatments for each species, with fewer than 20 genes in common between them, suggesting differential acclimation responses to chronic OA exposure. Our study is the first to use high-throughput sequencing to examine gene expression of OA-tolerant versus susceptible teleosts, providing important information about sublethal changes associated with OA resistance in marine fishes. Funded by COAST, CSUPERB

40.12

PROTEOMIC PROFILE AND PROTEOGENOMIC ANALYSIS OF SKELETAL MUSCLE IN A MAMMALIAN HIBERNATOR

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This study uses a proteogenomic approach that couples high-throughput proteomics with RNA-seq data to study skeletal muscle function during mammalian hibernation. Cessation of feeding, prolonged immobilization, and altered fuel availability are hallmarks of the hibernation phenotype that have major implication for tonality and functionality of skeletal muscle. Yet these animals exhibit limited changes to muscle function and mass through 5-6 months of hibernation. Mass spectral analysis using iTRAQ labeling produced quantitative proteomic data from the thirteen-lined ground squirrel (*Urocyon v. tridecemlineatus*) during the circannual cycle. These data agree with many of the physiological phenomenon known to occur in hibernator skeletal muscle, and the depth of coverage makes it possible to support both previously described and novel mechanistic hypotheses for these phenomenon. Using the customizable data analysis platform GalaxyP to merge the proteome data with Illumina HiSeq2000 transcriptome data from the same animals, we were able to identify protein sequences unique to the ground squirrel. Novel peptide sequence identification allowed for improved annotation of the ground squirrel genome, identification of alternative splice sites, mutations, and genomic organization, all of which may facilitate the altered physiology of hibernator skeletal muscle. This work was supported by NSF grant 1147079.

40.13

A FUNCTIONAL GENOMIC ANALYSIS OF WEDDELL SEAL DIVING ADAPTATIONS: VASCULAR BIOLOGY

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To examine the functional genomics of mammalian diving physiology, we sequenced genomic DNA extracted from Weddell seal liver, yielding a 90X Illumina/ALLPATHS-LG assembly. Our draft genome annotation was analyzed against the dog genome, identifying 16,839 protein-coding seal genes (versus ~21,000 in dog) and 2,660 non-coding genes. To understand tissue-specific vasoregulation during diving, we initially focused on the role of nitric oxide (NO) and its downstream messenger cGMP, generated by NO activation of soluble guanylate cyclase (sGC). We will test the hypothesis that NO-mediated local hypoxic vasodilation is blunted to permit central control of the diving response. In preliminary data, NO-induced sGC activation was lower in Weddell seal pup lung protein extracts than mouse lung. In seal lung, kidney and skeletal muscle, sGC activation induced by an NO donor was also less than NO-independent activation by the drug cinaciguat. Reduced sGC activity upon stimulation with NO may be due to amino acid sequence dissimilarities between species—the sGC $\alpha 1$ subunit differed between Weddell seal and mouse by 11%. These findings may imply impaired NO-sGC signaling in pulmonary, peripheral and splanchnic tissues of Weddell seals. Funded by NHGRI and MGH Dept. Anesthesia.

41.0: CONSERVATION PHYSIOLOGY

41.1

OCEAN ACIDIFICATION: EFFECTS OF CO₂ ON BEHAVIOR AND GABA FUNCTIONS IN TELEOST FISH

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Elevated levels of ocean CO₂, to be expected around the year 2100, have been shown to cause abnormal behavior in marine fish. Behavioral changes include increased activity levels, altered auditory and olfactory preferences and loss of lateralization. The normal behavior can be restored upon administration of a GABA-A antagonist, demonstrating that the function of GABA as the major inhibitory neurotransmitter is lost under acidosis. GABA-agonists on the other hand theoretically have an inhibitory effect on behavior of control fish, whereas CO₂ exposed fish should become even more affected. GABA-agonists have been studied less extensively in the context of ocean acidification, and the few studies available have provided contrasting results. Our results show that exposure to end-of-the-century CO₂ levels makes stickleback (*Gasterosteus aculeatus*) hyperactive, with no effect on lateralization. Muscimol did not aggravate the behavioral disruptions caused by CO₂, suggesting that elevated CO₂ is only affecting a small subpopulation of GABAergic synapses. GABA-A receptor subunit expression has been mapped in the zebrafish brain, in an attempt to utilize zebrafish (*Danio rerio*) as a model for studies on the effects of elevated CO₂ on fish behavior and GABAergic function.

41.2

CONTRASTING SENSITIVITY TO OCEAN ACIDIFICATION-INDUCED BEHAVIOURAL CHANGE IN ATLANTIC COD (*GADUS MORHUA*) AND THREE-SPINED STICKLEBACK (*GASTEROSTEUS ACULEATUS*)

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The rising atmospheric CO₂ levels are driving dissolution of CO₂ into surface waters. Coral reef fish show disturbed behaviour when exposed to elevated pCO₂, but how temperate fishes may respond to ocean acidification has been unknown. We exposed juvenile Atlantic cod and adult sticklebacks for one month to CO₂ levels possible year 2100 (1000 μ atm), and tested a range of behaviours. We show that sticklebacks display behavioural disturbances similar to coral reef fish, demonstrating that the effect is global. However, the behaviour of Atlantic cod was unaffected, which is surprising given that both species are tolerant to a large range of environmental conditions. This demonstrates that the impact of elevated pCO₂ on fish behaviour is diverse and unpredictable. Atlantic cod also showed strong avoidance behaviour of CO₂ when given a choice between control (390 μ atm), and elevated pCO₂ (1000 μ atm), indicating that they consider elevated pCO₂ suboptimal. This could potentially influence distribution and migration of fish in a future high-CO₂ ocean.

41.3

PACE OF LIFE EFFECTS CRITICAL WINDOWS FOR DISEASE EMERGENCE AND TRANSMISSION IN AMPHIBIANS

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To our ability to predict disease emergence and spread is the identification of factors that may be predictive of individual heterogeneity in disease susceptibility and capacity to transmit an infection. Here I detail a series of studies that show how a suite of physiological and behavioral traits, underlying the pace of life paradigm are predictive for Ranavirus transmission and epizootic dynamics in larval amphibians. Ranaviruses are globally distributed, double-stranded DNA viruses that are among the most non-specific, and generalist of emerging pathogens, which often cause lethal multi-host epizootics in amphibian communities. In amphibians susceptibility to this virus varies significantly among individuals, and increases dramatically during the critical developmental window of metamorphosis. We have found that differences in slow versus fast phenotypes of larvae along the pace of life continuum in growth and development rates trade-off with immunity causing variance in disease susceptibility and critical window patterns. In addition, these phenotypic differences also influence disease transmission through variation in behavior and host virulence (i.e. pathogen shedding). Through integrating these results into a pace of life explicit susceptible-infected epidemiological model, I detail how this model host-pathogen system can provide insight into the physiological, behavioral, and developmental factors influencing disease dynamics in animals and their populations.

41.4

ECOSYSTEM HEALTH AND ENVIRONMENTAL INFLUENCES ON INNATE IMMUNE FUNCTION IN THE LOGGERHEAD (*CARETTA CARETTA*) AND GREEN (*CHELONIA MYDAS*) SEA TURTLE

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Sea turtles are long-lived poikilotherms that migrate to near shore habitats that pose numerous anthropogenic threats. Of increasing concern is how anthropogenic impacts and global climate change may affect the emergence of infectious disease within Florida sea turtle populations as our understanding of their immune function is limited. As non-specific immune defenses evolved relatively early, it is likely that the sea turtle as a "living fossil" relies heavily on this modality. The objective of this research was to quantify phagocytosis in loggerhead (*Caretta caretta*) and green (*Chelonia mydas*) sea turtles. We isolated peripheral blood mononuclear cells (PBMCs) from polymorphonuclear granular leukocytes (PMNs) and utilized flow cytometry to quantify phagocytosis across a variety of temperatures in both healthy turtles and those from declining habitats where disease is prevalent. Results indicate that sea turtles that resided in a degraded habitat mounted a far less robust immune response than those from the more pristine site ($p < 0.05$). By interpreting how aspects of their habitat influence the immune function of sea turtles, we will be better equipped to develop recovery plans for these endangered species. This research was graciously funded by the Morris Animal Foundation.

41.5

DECOUPLING THE RELATIONSHIP BETWEEN IMMUNE RESPONSE AND STRESS HORMONES: AN IMMUNOLOGIC PROFILE OF THE NORTHERN ELEPHANT SEAL

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Female northern elephant seals utilize finite resources during terrestrial haul-outs that they must allocate to competing metabolic processes. In addition to fasting, the breeding haul-out is characterized by energy-demanding and immunologic challenges, such as parturition, lactation, and aggressive interactions with conspecifics. Northern elephant seals elevate cortisol, a hormone known for its immunosuppressive effects, to mobilize lipid resources for energy while fasting. Recent work has shown that cortisol levels at the beginning of fasting vary widely with long-term foraging success at sea. Although elephant seals experience sustained elevations in serum cortisol, they retain immune function as indicated by the presence of various markers. We measured a suite of antibodies and inflammatory proteins to analyze how these parameters vary with life history stage and stress hormone levels. These markers were higher during the breeding haul-out in comparison to the molt haul-out. Most markers showed no relationship to cortisol except for IgE, an immunoglobulin associated with parasite exposure, which decreased with increasing cortisol. This is the first time a suite of immune and inflammatory markers have been measured in northern elephant seals, showing a potential decoupling of stress hormones and immunosuppression in an animal that exhibits adaptive variation in cortisol. Funding was provided by the Office of Naval Research

41.6 WITHDRAWN

41.7

HEAT TOLERANCE OF AUSTRALIAN BIRDS: THE IMPORTANCE OF BODY SIZE, PHYLOGENY AND EVAPORATIVE PATHWAY

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Heat waves in Australia have resulted in a number of heat related avian mortality events where large numbers of birds die from dehydration or heat stroke. Due to the apparent differential susceptibility to heat among species, these mortality events have great potential to reshape avian communities as temperatures rise due to anthropogenic climate change. The goal of this investigation was to determine the role of body size, evaporative pathway, and phylogeny in the heat tolerance of Australian desert birds. Here we quantify metabolic rate, rates of evaporative water loss, and body temperature in eight species of Australian bird representing four orders and ranging in body size from 7g - 300g. Among these birds, large birds benefited from heat storage due to mild-hyperthermia, and had lower mass specific rates of water loss relative to metabolic heat gain. Small birds utilized hyperthermia to a greater degree and had higher rates of metabolism relative to rates of water loss, a product of active respiratory evaporation. Columbiformes outperformed most other species, due to their large body size and high rates of largely passive cutaneous evaporation, which resulted in low rates of metabolism and mild-hyperthermia. These data provide insight into the importance of phylogeny, evaporative pathway at high temperature and body size, which can be used in future studies to parameterize models to aid in the prediction and potential mitigation of heat related avian mortality events.

42.0: BIOMECHANICS, LOCOMOTION, AND FUNCTIONAL MORPHOLOGY

42.1

THE MECHANISTIC BASIS OF UNRELIABLE SIGNALS OF STRENGTH IN MALES OF THE TWO-TONED FIDDLER CRAB, *UCA VOMERIS*

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Signals used during aggressive contests between males of many species are thought to be reliable indicators of underlying quality. In some species, however, unreliable signals develop as a consequence of mismatches between signal size and the information being conveyed. We investigated physiological mechanisms that potentially govern signal reliability in the fiddler crab, *Uca vomeris*. Male *U. vomeris* exhibit both reliable and unreliable signals of strength via the expression of original and regenerated claw morphs. Regeneration of the major claw occurs after damage or loss of the original claw, and regenerated claws are always poorer in quality than the claws they replace. We examined morphological, biomechanical and biochemical characteristics of original and regenerated claws, to establish the best predictors of the variation seen in claw strength. We found that for a given claw size, regenerated claws have less muscle mass than original claws, and that for a given muscle mass regenerated claws were significantly weaker than original claws. Mechanical advantage was also lower in regenerated claws. The activity of three catabolic enzymes did not

differ between claw types. Poor strength in these regenerated claws resulted from a combination of physiological factors and thus the development of other unreliable signals should also be constrained by them.

42.2

THE EFFECTS OF SELECTION FOR DESICCATION OR STARVATION RESISTANCE ON TAKEOFF FLIGHT PERFORMANCE IN *DROSOPHILA MELANOGASTER*

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Flight requires a suite of highly derived physiological and morphological characteristics that combine to generate the required flight performance of a species. Small deviations in either of these parameters can result in substantial changes in flight performance. Environmental stressors, like those imposed during droughts, can generate directional selection that favors resistance to desiccation and/or starvation over the maintenance of flight performance characteristics. This research studied multiple populations of *Drosophila melanogaster* that were selected for resistance to desiccation or starvation. Using high-speed videography, takeoff flights of individual flies were tracked. Using custom software written in MATLAB, velocities and flight angles (relative to the point of takeoff) were compared between the selected lines and their corresponding control groups. The results showed that both the desiccation and starvation resistant lines were significantly heavier than their controls but were not different from each other. The additional glycogen stored in the abdomen of the desiccation selected flies did not result in any measurable reduction in flight performance. In contrast, starvation resistance flies exhibited a significant reduction in flight angle. The difference in the flight trajectories of the starvation resistance flies is likely due to cardiac impediment caused by additional fat stored during larval development.

42.3

MATERIAL AND STRUCTURAL CHARACTERIZATION OF MINERALIZED ELASMOBRANCH CARTILAGE: LESSONS IN REPEATED TILING PATTERNS IN MECHANICALLY LOADED 3D OBJECTS

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The 'tessellated' skeleton of elasmobranchs (sharks and rays) is a composite of mineralized tiles (tesserae), collagen, and unmineralized cartilage, organized in discrete phases. The constituents' material and structural properties (e.g. tiling geometries) are vital to skeletal growth and mechanics, but have been little investigated due to the technical challenges involved. We use high-resolution materials characterization and visualization techniques to examine a developmental series of tesserae, outlining the development of unique structural features that likely function in load bearing and energy dissipation, with some regions exceeding cortical bone's mineral content and stiffness. To examine interactions among tesserae, we developed an advanced tiling-recognition-algorithm to semi-automatically detect and isolate individual tiles in microCT scans of tesseral mats. The method allows quantification of shape variation across a wide area, allowing localization of regions of high/low reinforcement or flexibility in the skeleton. The combination of our material characterization and visualization techniques allows the first quantitative 3d description of anatomy and material properties of tesserae and the organization of tesseral networks in elasmobranch mineralized cartilage, providing insight into form-function relationships of the repeating tiled pattern, as well as fundamental tiling laws important for complex, mechanically loaded 3d objects. Funding: HFSP grant to MND & JW.

42.4

THE ELASMOBRANCH HEART DOESN'T TWIST: A SPECKLE-TRACKING ECHOCARDIOGRAPHY STUDY.

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Background: Compact myocardial fibers are arranged to form a helical architecture in the mammalian heart. This fiber arrangement generates rotation of the cardiac base and apex in the clockwise and counterclockwise directions as observed from the apex, respectively, resulting in left ventricular twist. It is unknown if ventricular rotation/twist occur in species with a univentricular heart. **Methods:** We investigated myocardial strain, a novel parameter of myocardial deformation in the circumferential (C) and longitudinal (L) directions, and ventricular rotation by 2D speckle-tracking echocardiography (Vivid E9, GE Healthcare) in 7 elasmobranchs (2 bullhead sharks, 2 brownbanded bamboo sharks, 3 whipsnakes) under sedation with tricaine methanesulfonate. The study conformed to the Guiding Principles in the Care and Use of Animals and was approved by the institution's ethics committee. **Results:** Speckle-

tracking was feasible in all cases. Peak global C- and L-strain was $-21.5 \pm 7.4\%$ ($-23 \pm 4\%$ in humans) and $-23.8 \pm 3.7\%$ ($-20 \pm 3\%$ in humans), respectively. Mean basal and apical rotation was -2.9 ± 3.8 (-9.6 ± 2.5) and -2.4 ± 3.8 (-11.1 ± 4.0), respectively. Rotational direction at the cardiac base was the same as that at the apex. **Conclusion:** Elasmobranch and human hearts have similar ventricular strain. In elasmobranchs, however, ventricular rotation was not prominent and ventricular twist was not observed, suggesting that the fiber arrangement differs between the elasmobranch heart and human heart. **Reference:** 1. Sun JP, Lam YY, Wi CQ, Yang XS, Guo R, Kwong JS, Merlino JD, Yu CM. Effect of age and gender on left ventricular rotation and twist in a large group of normal adults – A multicenter study. *Int J Cardiol* 167: 2215-2221, 2013.

42.5

TRENDS IN MORPHOLOGY AND BIOMECHANICS OF THE AQUATIC GILL VENTILATORY SYSTEM OF RAY-FINNED FISHES (ACTINOPTERYGII)

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To ventilate their gills, ray-finned fishes (Actinopterygii) use pumps in their oral and gill chambers to alternate between suction (inspiration) and compression (expiration). These mechanics are largely conserved across Actinopterygii, but there is considerable morphological and functional variation of the pumps. We used comparative approaches to investigate the evolution of ventilatory morphology and function across actinopterygians. Using recently published molecular data, we reconstructed the evolutionary history of restricted gill openings across 433 actinopterygian families. Restricted gill openings have evolved at least 11 times among ray-finned fishes with diverse morphology and ecology. We also studied ventilatory biomechanics among four benthic sculpins and found considerable variation in oral and gill chamber pressures. Using phylogenetically independent contrasts, we linked variation in these pressures to morphology of the ventilatory pumps. Variation in function correlated closely with the size of the branchiostegal apparatus, specifically the branchiostegal rays that form the floor of the gill chamber. We propose that adding a third pump to the traditional two-pump model, in which the branchiostegal rays work in parallel with the operculum, is a useful framework for comparative gill ventilatory studies. Funding sources include NSF (DEB-1310812), Sigma Xi Cornell University Chapter, Friday Harbor Laboratories, and the Stephen and Ruth Wainwright Fellowship.

42.6

PAINTED TURTLES LOSE BONE STRENGTH BUT MAINTAIN BONE TOUGHNESS DURING ANOXIC HIBERNATION

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Western painted turtles (*Chrysemys picta bellii*) can endure more than 170 days of anoxia at 3°C, in part because ions are released from the skeletal system that help to buffer lactic acidosis. We evaluated the effects of anoxic submergence at 3°C on painted turtle bone after 60, 130, and 170 days, and compared them to normoxic turtles held at the same temperature. To assess changes in the material properties of bone, beams (4x25 mm) were fashioned from plastron, broken in a three-point flexure test, and analyzed using DXA to determine bone mineral density. CO₂ and lactate concentrations were also measured in the beams by CO₂²⁻ titration and colorimetric lactate oxidase analysis, respectively. Anoxic plastron accumulated more lactate (143±16 vs. 14±1 mmol/kg; $p < 0.01$), lost more CO₂ (1306±53 vs. 1560±34 mmol/kg; $p < 0.05$), and had decreased bone mineral density (0.115±0.002 vs. 0.123±0.002 g/cm³; $p < 0.05$) compared to normoxic plastron. Anoxic plastron sustained a lighter maximum load (30±2 vs. 35±1 N; $p < 0.05$) and greater displacement (1.22±0.06 vs. 1.05±0.05 mm; $p < 0.05$) before fracture, but absorbed the same amount of energy before fracture as normoxic plastron (27±2 mJ). These results indicate that anoxia-induced demineralization decreases shell strength, but also increases shell ductility so that the energy absorbed before fracture is not diminished and the protection the shell provides is not compromised. *This study was supported by the National Science Foundation.*

42.7

TEMPERATURE DEPENDENCE OF MUSCLE FUNCTION ALTERS THE EFFECTIVE UTILIZATION OF TENDONS

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Tendons can protect muscles from stretch-induced damage during energy dissipating tasks. During such behaviors, mechanical energy is initially stored in tendons and as

muscle force declines, tendons recoil and stretch muscle fascicles. This mechanism is thought to reduce the rate of stretch directly applied to muscle fascicles. We examined how changes in muscle contractile properties alter the effective utilization of this elastic mechanism. We used temperature manipulation in an *in vitro* muscle-tendon unit (MTU) to examine how changes to the rate of force development and relaxation alter the rate at which tendons recoil and subsequently stretch muscle fascicles. We measured muscle fascicle lengths by instrumenting frog plantaris muscles with sonomicrometry crystals. MTU force and length were measured by a servomotor. We characterized the rate of force development and relaxation during twitches at 10, 20 and 30°C. The MTU was also actively lengthened at the various temperatures with a 50ms tetanic stimulation and a simultaneous 100ms stretch. As expected, the rates of force development and relaxation increased with temperature. We also observed that the rate of tendon recoil correlated positively with rising temperature during eccentric contractions. These data imply that factors that alter muscle kinetics can influence the rate of fascicle stretch and thus alter the likelihood of muscle damage. Supported by NSF grant 1051691.

42.8

THREE-DIMENSIONAL NEUROANATOMY OF THE KILLER WHALE (*ORCINUS ORCA*) FROM MAGNETIC RESONANCE IMAGES

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Whales, dolphins, and porpoises possess large brains characterized by extensive cortical complexity and distinctive morphologies. The killer whale (*Orcinus orca*) has one of the largest brains of all animals and evinces complex cognitive, sensory, and social capabilities. Therefore, we generated the first three-dimensional (3D) atlas of the *O. orca* brain to further understand the neurobiological underpinnings of these abilities. We acquired magnetic resonance (MR) images of an *in situ* postmortem brain of an adult *O. orca* using a 3 Tesla General Electric (GE) Scanner. Voxel-based morphometry was applied to derive 3D reconstructions and volumes of the gray matter (GM), white matter (WM), cerebrum, cerebellum, corpus callosum (CC), hippocampus, and inferior colliculi (IC). Consistent with findings in other delphinids, GM is expansive and convoluted, yet thin, while WM is extensively branched and pronounced. CC volume is greatly reduced in relation to the volume of the cerebral hemispheres suggesting increased hemispheric independence. Auditory structures (e.g. IC) are hypertrophied in *O. orca* signaling the importance of rapid integration and processing of acoustic stimuli for this echolocating species. Hippocampal volumes are hypertrophied suggesting transfer of learning, memory, and spatial orientation abilities to alternate neural structures. A.K. Wright was supported by an NSF Graduate Research Fellowship.

42.9

IMPROVING EXERCISE ADHERENCE AND PHYSICAL MEASURES IN HISPANIC WOMEN

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Purpose: Epidemiological data show that lack of physical activity increases risk factors for chronic diseases. Data also suggest that physical activity participation is lowest in minority women, particularly Hispanic women, and that exercise modality and attitudes toward exercise influence exercise adherence. The purpose of this study was to examine the effect of *Evaluative Conditioning* (EC) on physical activity adherence as well as on other psychosocial variables such as body image, self-esteem, and exercise self-efficacy. **Methods:** 142 Hispanic women (mean ± SD, age 36.8±15.9 yrs.) were randomly assigned to a physical activity either receiving EC or NC. **Results:** For HT, EC produced significantly greater exercise adherence than NC. All body composition variables improved regardless of training or conditioning except for LBM, which declined with NC ($p < 0.05$). Resistance training positively impacted all strength and functional variables with the exception of leg extension and usual gait speed ($p < 0.05$). Both self-esteem and body esteem improved with training ($p < 0.05$). **Conclusion:** EC can positively increase exercise adherence during HT in Hispanic women. Funding for the current study was provided by a University of Miami Barbara Marks/Katy Dean Research Award and a Transformative Research Grant from the Association for Consumer Research.

43.0: SCHOLANDER AWARD FINALISTS

43.1

DOES LOCAL ADAPTATION OF EXERCISE PHYSIOLOGY LIMIT ACCLIMATION CAPACITY AMONG LAKE WHITEFISH (*COREGONUS CLUPEAFORMIS*) ECOTYPES?

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Organisms can cope with environmental changes within their lifetime via acclimation and over generations populations may also undergo local adaptation. It has been hypothesized that if these processes have similar mechanistic bases, past local adaptation may constrain acclimation capacity. To address this question we compared populations of dwarf and normal Lake Whitefish (*Coregonus clupeaformis*). These ecotypes have undergone adaptive divergence in energy metabolism and swimming behavior such that the derived, dwarf fish are more active swimmers and have a slower growth rate than the ancestral, normal fish. We examined the extent of local adaptation in exercise physiology and compared the capacity for acclimation among ecotypes by conducting a common garden, swim training experiment on lab-bred and raised, size-matched fish. We found that dwarf whitefish have evolved a higher muscle mitochondrial and glycolytic enzyme content and higher levels of many oxygen transport related traits (ventricle size, ventricle mitochondrial content and percentage of aerobic skeletal muscle), as predicted by prior behavioral and gene expression studies. We found little plasticity in exercise physiology, and of the traits that did respond to exercise training and also varied among ecotypes (hematocrit, ventricle malate dehydrogenase activity), most had similar levels of plasticity in dwarf and normal fish, indicating that local adaptation does not constrain acclimation. However, dwarf whitefish had large ventricles that did not respond to training, while normal whitefish had smaller hearts that increased in size with training, suggesting that plasticity in this trait may be constrained by prior local adaptation. Funded by NSERC.

43.2

CORAL HOST CELLS ACIDIFY SYMBIOTIC ALGAL MICROENVIRONMENT TO PROMOTE PHOTOSYNTHESIS

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Symbiotic zooxanthellae algae residing inside coral tissues supply the host with the majority of their energy requirements through the translocation of photosynthetically fixed carbon. The zooxanthellae, in turn, rely on the host for the supply of inorganic carbon. Carbon must be concentrated as CO₂ in order for photosynthesis to proceed, and here we show that the coral host plays an active role in this process. The host-derived symbiosome membrane surrounding the zooxanthellae abundantly expresses vacuolar H⁺-ATPase (VHA), which acidifies the symbiosome space down to pH=4. Inhibition of VHA results in a significant decrease in average H⁺ activity in the symbiosome of up to 75% and a significant reduction in O₂ production rate, a measure of photosynthetic activity. These results suggest that host VHA is part of a novel carbon concentrating mechanism for zooxanthellae photosynthesis, and provide mechanistic evidence that coral host cells can actively modulate the physiology of their endosymbionts. This work was supported by National Science Foundation grants EF-1220641 and OCE-1226396, and an Alfred P. Sloan Research Fellowship (BR2013-103).

43.3

GNRH-SELECTIVE SIGNAL TRANSDUCTION NETWORKS AND THE CONTROL OF PITUITARY CELL FUNCTION

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Among its many functions, gonadotropin-releasing hormone (GnRH) stimulates luteinizing hormone (LH) and growth hormone (GH) release through direct actions on pituitary cell-types called gonadotropes and somatotropes, respectively. The presence of multiple GnRH isoforms is a common feature among chordates and our ongoing research examines how GnRH-selective signal transduction mechanisms activated by two native GnRH isoforms, GnRH3 and GnRH2, participate in the differential control of hormone release using primary cultures of dispersed goldfish (*Carassius auratus*) pituitary cells. In present study, we provide the first evidence for the involvement of class I phosphoinositide 3-kinase (PI3K) isoforms in GnRH actions and demonstrate that GnRH3 and GnRH2 binding to GnRHRs can bias the activation of class I PI3K-dependent signal transduction. These novel findings reveal the complexity of GnRH-stimulated PI3K activation and adds to the understanding of biased GnRH-selective signaling. Additionally, by studying naturally biased GnRH isoforms in a lower vertebrate model, our results help to elucidate how functional selectivity in ligand-receptor signal transduction networks has evolved and how this phenomenon ultimately impacts the neuroendocrine control of growth and reproduction. (Supported by NSERC, AIHS, and the Killam Trusts).

43.4

CONVERGENT AND DIVERGENT PATTERNS OF GENE EXPRESSION IN SCULPINS THAT VARY IN HYPOXIA TOLERANCE

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Sculpins, a diverse group of fishes, exhibit variation in hypoxia tolerance that is related to species distribution along the marine nearshore environment. To identify potential candidate traits involved in hypoxia tolerance, we assessed gene expression patterns over a 72 hour hypoxia time-course using microarrays. In the first set of experiments, we quantified divergent expression patterns in species with different hypoxia tolerance. We found that the hypoxia tolerant species down-regulated genes associated with a number of energetically costly biological processes while the hypoxia intolerant species did not. Additionally, gene expression did not change in the first 24 hours in the tolerant species of sculpin while in the intolerant species the greatest change occurred during the initial stages of hypoxia exposure. Together, these transcriptional patterns may, in part, help define hypoxia tolerance in these species of fish. In the second set of experiments, we examined 3 species with similar and intermediate levels of hypoxia tolerance for evidence of convergence of gene expression or the repeated evolution of the same traits. While we found some convergent gene expression patterns among the 3 species, a greater majority of genes exhibited non-convergent changes in gene expression. Therefore, convergent evolution of intermediate hypoxia tolerance is for the most part not a result of convergence of gene expression patterns in these species. (Funding NSERC)

43.5

OCEAN ACIDIFICATION DIRECTLY IMPAIRS OLFACTORY SENSITIVITY IN A MARINE TELEOST

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Coral reef fish exposed to ocean acidification (OA) predicted for year 2100 respond inappropriately to olfactory cues that are vital for their survival. A brain mechanism has been proposed based on GABA_A receptor dysfunction, but other potential causes and OA effects on non-coral reef species remain largely unknown. We hypothesized that OA has a direct effect on the olfactory sensitivity of fish and used the economically important European sea bass to test our hypothesis. We measured the response of the olfactory system to various odorants in control (CO₂ ~500 µatm) and OA (CO₂ ~1000 µatm) conditions by multi-unit recording from the olfactory nerve. The responses to several amino acids, bile acids, conspecific bile, and alarm cue, but not intestinal contents were reduced by up to 88% in OA seawater. Thus, OA reduces the olfactory sensitivity of bass directly. We propose this decrease in olfactory sensitivity is due, at least in part, to a reduction in odorant-receptor affinity at lower pH, providing a new mechanism for this impairment. Additionally, the detection threshold of some odorants increased by 2- to 4-fold in OA. Based on these changes we estimated that under OA conditions bass would need to be up to 48% closer to the odorant source to allow detection and this could have detrimental consequences on their survival in the ocean. Funding: ASSEMBLE (Grant no. 227799); Royal Society Newton International Fellowship to CP; CP is a beneficiary of a Starting Grant from AXA.

43.6

WHAT HAS K⁺ GOT TO DO WITH IT? THE DIFFERING ROLES OF EXTRACELLULAR K⁺ IN ONSET AND RECOVERY OF INSECT CHILL COMA

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Insects enter chill coma, a reversible state of paralysis, at temperatures below their critical thermal minimum (CT_{min}), and the time required for an insect to recover after a cold exposure is chill coma recovery time (CCRT). The CT_{min} and CCRT are important metrics of insect cold tolerance and are often used interchangeably, but these two traits are not necessarily physiologically linked. Here we test the hypothesis that depolarization of muscle membrane potential (V_m) at chill coma onset and its repolarization during chill coma recovery are driven by changes in extracellular [K⁺] and/or directly by low temperature. Using *Locusta migratoria* we measured *in vivo* muscle resting potentials during cooling, following prolonged exposure to -2°C and during chill coma recovery, and related changes in V_m to transmembrane [K⁺] balance and temperature. Although cooling rapidly depolarized V_m, hemolymph [K⁺] did not rise until locusts had spent considerable time in the cold. Nonetheless, a rise in hemolymph [K⁺] during prolonged cold exposure further depressed muscle resting potential and slowed recovery from chill coma upon rewarming. Thus, a disruption of extracellular [K⁺] does depolarize muscle resting potential and slow CCRT, but it is unrelated to the onset of coma. Because the mechanisms underlying these two traits are distinct, haphazard trait choice could lead to conflicting candidate genes under selection for cold tolerance.

43.7

CHANGES IN MO_2 , ANAEROBIC GLYCOLYSIS AND METABOLIC HEAT WITH DECREASING WATER PO_2 IN GOLDFISH

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Hypoxic survival in fish is dependent on the interactions between three mechanisms: improved O_2 uptake and utilization, anaerobic ATP production, and metabolic rate suppression (MRS). Many animals have been shown to employ all three mechanisms, but how these mechanisms interact with one another as environmental O_2 tensions (PO_2) decrease is unknown. The goal of this study was to characterize these changes in goldfish, a species of exceptional hypoxia tolerance, and it was hypothesized that the three hypoxia defense mechanisms would reach critical points at water PO_{2S} at or around the critical PO_2 of MO_2 (P_{aM}) where aerobic metabolism is first compromised. Calorimetry was used to simultaneously measure O_2 uptake/utilization (via MO_2) and metabolic heat, while anaerobic ATP production was determined through measurements of glycolytic metabolites (glycogen, lactate and ethanol). Preliminary results suggest that these critical points occur at different water PO_{2S} . Oxygen consumption rate shows a P_{aM} of ~35 mmHg O_2 , while anaerobic ATP production begins to increase at ~50 mmHg O_2 , and MRS is not evident until PO_2 reaches ~2 mmHg. This suggests that the relatively active lifestyle of goldfish in moderately hypoxic environments is maintained by anaerobically-produced ATP, while MRS is a strategy of tolerance reserved only for severe hypoxia or anoxia. (Funding provided by NSERC and UBC.)

43.8

INTEGRATING THE EFFECTS OF REPEATED COLD EXPOSURE FROM TRANSCRIPTOME TO WHOLE-ORGANISM IN THE EASTERN SPRUCE BUDWORM

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Organisms live in complex worlds where environmental stresses can be more or less intense, occur for longer or shorter periods, and repeat more or less frequently. Yet while single stress events have been well-studied, the physiological and fitness effects of these more complex patterns of stress are not well understood. The eastern spruce budworm, *Choristoneura fumiferana*, is an important forest pest of the boreal forest and previous studies have shown the importance of low temperature stress in regulating its population and range. In this study we manipulated the number of low temperature exposures budworm received, while controlling the intensity and total length of time of exposure. We found that while budworm that received repeated low temperature events had significantly greater cryoprotectant content (at a cost to glycogen reserves), survival to eclosion was significantly impacted. We also examined transcriptomic responses to low temperature stress, and found that while only 16 transcripts were significantly differentially regulated following a single low temperature stress event, 644 were differentially regulated following repeated low temperature stresses. These included transcripts for antifreeze proteins, several heat shock proteins, and electron transport chain proteins. These results suggest that current studies that focus on single stress events may be missing the full range of potential stress responses.

43.9

THE ROLE OF TRANSCRIPTION FACTOR GLIAL CELL MISSING 2 (GCM2) IN Ca^{2+} BALANCE IN ZEBRAFISH LARVAE.

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The present study investigated the role of the transcription factor, glial cell missing 2 (gcm2) in Ca^{2+} regulation in zebrafish larvae. Translational gene knockdown of gcm2 decreased Ca^{2+} uptake and the density of ionocytes expressing the epithelial Ca^{2+} channel (*zecac*), and disrupted overall Ca^{2+} balance. Acclimation to either low Ca^{2+} (25 μM [Ca^{2+}]) or acidic water (pH ~4.0) significantly increased the mRNA expression of gcm2. When measured in control water following acclimation to these conditions, Ca^{2+} uptake was significantly elevated. However, in fish experiencing gcm2 knockdown, no such stimulation of Ca^{2+} uptake was observed. Over-expression of gcm2 mRNA resulted in a significant increase in Ca^{2+} uptake and in the numbers of *zecac*-expressing ionocytes. When fish experiencing gcm2 knockdown was treated with waterborne cortisol, a well-known hypercalcemic hormone in fish, the treatment inhibited Ca^{2+} uptake in those larvae. These observations demonstrate a critical role for gcm2 in Ca^{2+} homeostasis in zebrafish larvae. All experiments were conducted in accordance with Guiding Principles in the care and Use of Animals and after the approval of the University of Ottawa Animal Care Committee (Protocol BL-226).

44.0: PLENARY LECTURE

44.1

MACROPHYSIOLOGICAL FORECASTING FOR POLICY IN A CHANGING WORLD

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The 2012 World Energy Outlook projected more than a 30% growth in global energy demand over the next several decades, most of it to be met by fossil fuel consumption. The net result will be an on-going rise in global temperatures and extreme high temperature events. Environmental change of other forms is likewise here to stay. At the heart of many policy interventions for both mitigating and adapting to these changes, lies forecasting, which is heavily dependent on physiology. Here we illustrate how investigations of physiological variation across space, time and phylogenies is improving forecasts of the impacts of the major environmental change drivers. In particular, the focus is on how enhanced understanding of variation in performance curves is improving forecasts of the impacts of changing temperatures and water availability, biological invasions, and interactions among the environmental change drivers, and so directly influencing policy. A further focus is on the simultaneous change in body size and its broader implications. Macrophysiology has done much to enhance ecophysiological forecasting. Nonetheless, several significant problems remain unresolved, despite ongoing implementation of policies both to mitigate and adapt to change. These are among the greatest modern challenges to physiology. (ARC DP140102815 and DP140101240). References: Chown, S.L. & Hoffmann, A.A. 2013. Ecophysiological forecasting for environmental change adaptation. *Funct. Ecol.* **27**, 930-933. Chown, S.L. & Gaston, K.J. 2008. Macrophysiology for a changing world. *Proc. R. Soc. B* **275**, 1469-1478.

NOTES

Booth #1

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