Covid-19 and the ISWE Community

Board Statement on Covid-19
The ISWE board members would like to extend their sympathies to all who have been affected by the covid-19 pandemic by illness, loss of friends or family, or economic or other hardship. Please stay safe and healthy.

ISWE members share their experiences during Covid-19
Covid-19 has impacted all of us, both personally and professionally. A few members share their experiences in the field and in the lab navigating this unprecedented event.

Report from the elephant forest during the pandemic
Sanjeeta Sharma Pokharel, Ph.D., of the Indian Institute of Science, has been spending a lot of time with elephants during the pandemic. “I had to postpone my lab-related work as I was locked in our field research station, far away from my institute, in the Mudumalai National Park, Tamil Nadu, India. I had a great time observing wild Asian elephants and the landscape.”

During her time in the field, Dr. Pokharel decided that Bhunte, a baby elephant, could offer some advice on how to be safe during a pandemic, and she created a cartoon and blog campaign. Dr. Pokharel developed the Bhunte character in 2016 as a means to spread awareness about the conservation plight of elephants. Since then, Bhunte has even been featured in the national press in India.

Lab start-up in the time of Covid-19
By: Kathleen E. Hunt, Ph.D., Assistant Professor, George Mason University, USA

In spring 2020 I started teaching at my first tenure-track academic job at George Mason University in Virginia. I’m a seasoned career scientist with more than two decades of full-time research under my belt, but I’ve been based at AZA institutions for most of my career. It just so happened that in 2019 I decided to shift to a traditional academic position. I was thrilled to get an offer from GMU, especially since GMU has strong ties to the Smithsonian’s famed research facility in Front Royal. So, fresh from the invigorating ISWE conference in South Africa and a wonderful November spent teaching endocrinology in Brazil, I was excited to start setting up my brand-new lab at GMU and really get to work.

[cont’d pg 5]
**Arbor Assays Launches New ISWE Mini-kits**

Arbor Assays has launched two new ISWE-mini kits for corticosterone and 17β-estradiol! Kits feature ISWE-generated and owned antibodies and conjugate, and standard stock solutions. Each kit is enough for 50 plates.

**Corticosterone ISWE Mini-Kit (ISWE007)** includes corticosterone sheep antisera concentrate, a 3-CMO-cortiosterone - HRP conjugate concentrate, and a stock solution of corticosterone for use as a standard.

**17β-Estradiol ISWE Mini-Kit (ISWE008)** comprises 17β-Estradiol sheep antisera concentrate, a 6-CMO-17β-Estradiol - HRP conjugate, and a stock solution of 17β-Estradiol for use as a standard.

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**Call for Papers**

The open access journal Animals (ISSN 2076-2615) is compiling papers for a special issue: "Perspectives on Physiological Measures of Animal Welfare in Chronic Conditions"

Original research papers or short reviews focusing on the use of physiological measures to assess positive or negative animal welfare during living conditions, chronic interventions or disease are invited.

**Papers are due: 31 March 2021**  
Detailed information can be found [HERE](#)

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**Photos From the Field**

Weekly urofeca samples from male-female African penguin pairs housed at National Zoological Garden, South Africa were collected by Chantel Holdstock over one year to measure urofeca glucocorticoid metabolite concentrations. *This study by Scheun, Gulson, & Ganswindt (2020)* confirms that non-invasive steroid monitoring can be an effective tool for defining and assessing environmental stressors for African penguins and potentially other captive seabirds.

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**Guess Whose Poo?**

Can you “guess whose poo”? Check out the picture below, and see if you can correctly identify the poo. [Answer page 5]

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Reminder:
Arbor Assays is offering ISWE members an extra 5% off list price for products over $50 through the end of 2020—a total discount of 30%! (*excludes mini-kits*)
Dr. Nei Moreira, Professor and Animal Physiology Lab Coordinator, shares with us the lab’s exciting projects focused on South American species!

**What makes your lab unique?**
We study endangered South American species, especially from Southern Brazil.

**Are you interested in pursuing collaborations within the ISWE community?**
Yes, we are interested in collaborations to study endocrinology of South American species. We want to improve our endocrine lab, with new equipment and techniques. We offer the possibility of doing collaborative research in South American species, together with captive breeding centers and zoos located in Brazil and also in Paraguay, where we already have research partnerships.

**What are some of your current projects?**
- Use of the kisspeptin analog C6 to trigger ovulation in the Southern tiger cat (*Leopardus guttulus*) as a new strategy to ameliorate reproductive technology in felids
- Reproduction in tapirs
- Semen cryopreservation in felids

**Any recent publications to highlight from your lab?**
- Erdmann, RH; Blank, MH; Ribeiro, RN; Oliveira, MJ; Cubas, ZS; Pradiee, J; Goularte, KL; Moreira, N. 2020. Cryopreservation of margay (*Leopardus wiedii*) spermatozoa: Effects of different extenders and frozen protocols. *Theriogenology* 143: 27-34.

**Tell us a little about your lab.**
Dr. Moreira: The Animal Physiology and Immunology Laboratory performs wildlife endocrinology research, as well as conducting Animal Physiology and Immunology Practical classes for veterinary students.

Our laboratory staff consists of myself; Katherinne Maria Spercoski, Professor, Immunology Lab Coordinator; Marina Giombelli Rosenberger, Lab Technician; and graduate students. We often collaborate with other institutions (including: Smithsonian Institution; University of São Paulo; University of Veterinary Medicine, Vienna-Austria; and UNESP—Jaboticabal.

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Recent Publication Highlights

The “Publication Highlights” section offers brief summaries of recent publications by ISWE members. If you would like to see your article in an upcoming newsletter, send us the citation and a photo showing your work in action. All submissions welcome! (email: ISWE.socialmedia@gmail.com)

Blubber cortisol levels in humpback whales (Megaptera novaeangliae): A measure of physiological stress without effects from sampling

One challenge in studying the health and well-being of wildlife is discovering the best matrix for hormone analyses—one that is minimally invasive, readily available, and provides consistent, longitudinal results. Fecal glucocorticoid metabolite monitoring has become the standard for monitoring the physiology of mammals, but it is not always feasible, especially for aquatic species that defecate at unpredicted locations or times. In smaller aquatic mammals, analyses of blubber biopsies provide reliable endocrine results that are not affected by the acute stressors of sampling. However, measuring blubber glucocorticoids had not been validated previously in baleen whales such as humpbacks.

Mingramm et al. (2020) collected blubber samples from Australian humpback whales (Megaptera novaeangliae) to assess the efficacy of their use in measuring cortisol concentrations and physiological stress. Over multiple migrations they darted and collected 187 blubber samples from individually identified whales. Humpbacks were divided into two groups: adult and juvenile live whales (deemed “healthy”) and banked biopsies from deceased whales (n=35; deemed “stressed” due to traumatic mortality events). EIA of blubber extracts with cortisol antibody (R4866, C. Munro, UC Davis) detected quantifiable concentrations in live and dead humpbacks, with deceased whales averaging ~10x higher concentrations. Furthermore, blubber cortisol varied seasonally; whales migrating to breeding grounds (north migration June/July) had higher concentrations than those migrating south to feeding grounds (Sept/Oct). Mingramm et al. did not find strong evidence that sampling affected the blubber cortisol concentrations even though several whales exhibited extended, atypical behaviors in response to sampling efforts. Thus, similar to other species, there is a time delay of several hours before hormone concentrations are expressed in blubber of humpbacks. This study provides evidence that blubber cortisol is an appropriate measure of chronic physiological stress in Australian humpback whales. Additionally, it establishes a foundation to explore how biological and/or environmental factors affect the well-being of humpback populations. [Written by: Elizabeth Freeman & Morgan Bragg]


Exposure to artificial light at night during the larval stage has delayed effects on juvenile corticosterone concentration in American toads, Anaxyrus americanus

A handful of studies have examined the effects of either predation or artificial light at night (ALAN) on stress-related hormones, survival, and other factors in amphibians. However, few studies have examined the interaction between predation and ALAN or tracked effects across life stages. K. Cope and collaborators from Case Western Reserve University (Cleveland, OH) and Cleveland Metroparks Zoo examined how ALAN and predator presence affected survival, growth, development, and hormone concentrations of American toads (Anaxyrus americanus). They hypothesized that both ALAN and predator presence would act as stressors on developing larvae and that the effect would be exacerbated in larvae exposed to both. Toad larvae experimentally reared in outdoor mesocosms were exposed to ALAN comparable to typical conditions in regional wetlands (white LED light around 3 lux), dragonfly larvae as predators, or both. They sacrificed a subset of larval and metamorphosing animals and housed the remaining animals as juveniles in a laboratory setting for later examination of carryover effects. They conducted hormone analyses at Cleveland Metroparks Zoo by homogenizing whole-animal samples, extracting hormones using a liquid-liquid (methanol:dichloromethane) extraction, and analyzing corticosterone concentrations using a RIA from MP Biomedicals. Although the dragonfly predators significantly reduced tadpole survival and mass at metamorphosis, neither ALAN nor the interaction of predation and ALAN affected corticosterone levels in toad larvae and metamorphs. The effects of ALAN on corticosterone were evident only when survival was taken into account, and the highest corticosterone concentrations were found in groups exposed to ALAN that had the lowest survival in the absence of predators. Additionally, toads reared with exposure to ALAN had significantly higher corticosterone concentrations as juveniles, showing a carryover effect. Importantly, this study shows that environmental stressors during development can have potentially negative effects on fitness that are not evident until later life stages, providing important information to conservationists addressing the amphibian crisis. [Written by: Grace Fuller]

Covid-19 experiences (continued...)

My plan, which in retrospect looks so innocent and naive, was that I would spend January and February trying to get my new classes under control, and then when spring break rolled around in March 2020, I would submit my first batch of orders for all my lab equipment, and start two new research projects - one on elephants at the National Zoo and the Maryland Zoo in Baltimore, the other on a treasure trove of old WWII-era baleen at the Smithsonian National Museum of Natural History.

Spring break rolled around, but of course things didn't go as planned. I think none of us will ever forget that week in March 2020, when with each passing day the once-distant threat of the pandemic became shockingly more real and urgent. At the beginning of spring break everything was still more or less on track. Seven days later my university had shuttered all labs, halted all research, cancelled all fieldwork, froze purchasing and cancelled all pending orders (so much for lab set-up), sent everyone home, and swung all classes online. The National Zoo, the Maryland Zoo and the Smithsonian museum all closed. The shutdown had begun.

Looking back, I'm grateful that the teaching gave me something to focus on. Many of my friends at non-teaching institutions had nothing to do but sit at home racked with anxiety and compulsively checking their toilet paper stocks, but at least I had something else to think about. Teaching new classes in a new position is always overwhelming, but having to abruptly switch all lectures to be online, rewrite all exams, and somehow put the labs online too, along with hand-holding the 65 stressed and panic-y undergraduates, required absolute focus. For the next two and a half months I worked fourteen hours a day, seven days a week, with no breaks. But it gave me a mission and a focus, and it helped me bond with all my new colleagues - even if we couldn't see each other in person, we could help each other online.

But one thing I couldn't do at all was research. With spring teaching finally over, I am only now facing the reality that all my endocrine and endangered-species research is completely halted. My lab is still barren, literally just empty stretches of countertop populated by few lonely Rainins and a single Eppendorf Repeater Plus. I haven't been able to even order so much as a freezer, so I don't have any samples or reagents. The nearby museum samples that I thought would always be accessible are still locked away behind the closed doors of the Smithsonian. Zoos are still completely closed to outside researchers. The dramatic halt to research productivity has been a little dizzying - I haven't had a week without any research since 1990! I keep getting taken by surprise by unexpected repercussions, some tiny and some not so tiny. I've had to turn down several applicants for graduate students and post-docs, since I have no lab to offer them, and don't yet know when my lab can be operational. I can't fulfill my obligations to federal funders or to collaborators. Long-term datasets in Alaska and elsewhere now will have a gaping hole. My grant reports look pathetic, and even the Facilities statements of new proposals look a bit unconvincing ("I promise I'll have a lab soon.")

It ought to be very unsettling, but instead I mostly feel fortunate to still have my health and my job. How lucky is it that I happened to land the first, and only, stable-salaried job of my life just a few months before the pandemic hit? I'm also fortunate to be at an institution that is taking the pandemic quite seriously. Like most of my colleagues, I've turned my time instead to analyzing old, long-delayed datasets (I'm deep into a valuable, long-stalled 2012-2017 sea turtle dataset as I write this), and writing up new papers, and putting in new proposals. I'll even confess that it's been rather a relief to finally get a chance to work on the endangered species that I'd been studying before? The Black Lives Matter movement has also drawn my attention to unexpected repercussions, some tiny and some not so tiny. I've had to turn down several applicants for graduate students and post-docs, since I have no lab to offer them, and don't yet know when my lab can be operational. I can't fulfill my obligations to federal funders or to collaborators. Long-term datasets in Alaska and elsewhere now will have a gaping hole. My grant reports look pathetic, and even the Facilities statements of new proposals look a bit unconvincing ("I promise I'll have a lab soon.")

What is most worth studying now - should I shift to studying novel viruses of bats, as some of my colleagues have, or continue to work on the endangered species that I'd been studying before? The Black Lives Matter movement has also drawn my attention to questions I should have been paying more attention to all along: how can we best tackle the systemic racism that still exists in science, and how can we best recruit and support young scientists from underrepresented groups? And also overlooked in the news - I'm still worried about the continued erosion of environmental protections in the USA, and I still hear the steady drumbeat of climate change, which probably will ultimately be a worse catastrophe than the entire pandemic.

Alongside such concerns, the fact that my lab is empty and my research halted seems totally insignificant. The lab will open when it opens. Someday I will have more than a handful of pipettors, someday I will get my Smithsonian samples, someday I'll have grad students again, and will be able to place my Arbor Assays orders again. But in the meantime I'm just hoping that all my friends, family, all my favorite species, my nation, and the whole planet manage to get through this year intact.

Guess Whose Poo? (And what’s so funny...)

Answer: King penguin (*Aptenodytes patagonicus*)

The photo, taken in a zoo, shows urofeal droppings of a king penguin. Interestingly, Wang et al. (2019) found areas inhabited by king penguins produce tremendous amounts of N₂O—more commonly known as nitrous oxide or ‘laughing gas.’ On a more serious note, the study sheds light on how seabird colonization in areas of glacial retreat affects production of N₂O and other greenhouse gases.