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FrogLog

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Promoting Conservation, Research and Education for the World's Amphibians

REGIONAL EDITION: THE AMERICAS

In Memoriam: César Ramón Molina Rodriguez

Half a Million Dollars Directed Towards Saving Amphibians Around the World

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Diasporus aff. *diastema*. Photo: Twan Leenders.

FrogLog

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Editorial

Dear *FrogLoggers*,

When writing the editorials for *FrogLog*, I often try to find a unifying theme underpinning each edition. And as I read through the articles for this edition, it quickly became apparent to me that it is more than simply a regional edition focused on the Americas; it is an issue about *progress*.

At the end of 2013, the Amphibian Survival Alliance gave out its first grant. Since then, the Alliance has directed more than $500,000 to global amphibian conservation efforts worldwide, thanks to the support of Alliance Partners and readers like you. But the news gets even better. The Alliance is now on track to investing close to $1,000,000.00 to amphibian conservation, research and education projects around the world by June of this year.

Now that's progress!

As many of you will recall, in mid-2013, the IUCN SSC Amphibian Specialist Group, in partnership with the Alliance, formed 12 thematic working groups with the objective of reviewing the Amphibian Conservation Action Plan (ACAP), which was originally published in 2007. These working groups were asked to develop a clear strategy addressing today's conservation challenges worldwide. The first revision to ACAP has now been completed and is available on our website, where it will be regularly re-assessed and updated, making it a truly living document.

Now that's progress!

Still not impressed? I guarantee you will be when you discover the many other exciting signs of progress throughout the pages of this edition. From multidisciplinary thinking and solutions, to harnessing the power of partnerships and the fostering of new areas of research, this edition has it all.

From the Great White North of Canada to the southernmost tip of South America, we're seeing on-the-ground conservation successes and promising approaches that address the most pressing conservation issues amphibians face today.

Now that's progress!

Candace M. Hansen-Hendrikx
Editor-in-Chief

Photo: Robin Moore.

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FrogLog

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Photo: Robin Moore.

Success! Half a Million Dollars Directed Towards Saving Amphibians Around the World

By James P. Lewis & Candace M. Hansen-Hendrikx

Now that is progress! At the end of 2013, we gave out our very first grant. Since then we have supported 24 projects around the world, helping to direct a total of $551,586.99 to global amphibian conservation efforts.

This incredible achievement was made possible thanks to the support of Alliance Partners such as Global Wildlife Conservation, Rainforest Trust, Detroit Zoo, Synchronicity Earth, Andrew Sabin Family Foundation, International Conservation Fund of Canada and The British Herpetological Society and because of dedicated individuals like you. Together we're making a difference.

This has been a great start, and provides a solid foundation on which to start taking even bigger steps to protect the amphibians we all love. There are still more habitats to protect and more amphibians to be safeguarded against deadly infectious diseases.

For instance, we are actively raising support for a new 2,000 acre reserve in the remote Cuchumatanes mountains of Guatemala to protect the cloud forest home of several salamanders and frogs found nowhere else in the world.

By June we want to hit the $1,000,000.00 mark. By the end of this year we want to have invested $2,000,000.00 in amphibian conservation, research and education projects around the world.

Over 100 Alliance Partners are coming together to say, "Now is the time for action."

Now is the time for you to join us in making the future even brighter for the millions of amphibians that share this planet with us by visiting www.amphibians.org to learn more about our work and how you can get involved.

asa
amphibian survival alliance

Updating the Amphibian Conservation Action Plan (ACAP): Towards a Living Road Map

By Sally Wren, Ariadne Angulo, Helen Meredith, Jos Kielgast, Leida Dos Santos & Phil Bishop

In mid-2013 the IUCN SSC Amphibian Specialist Group (ASG), in partnership with the Amphibian Survival Alliance (ASA), began the process of establishing 12 thematic working groups comprising ASG members with interest and expertise on specific topics relevant to global amphibian conservation. The objective of the working groups is to review the Amphibian Conservation Action Plan (ACAP), published in 2007, and develop a clear strategy to address amphibian conservation challenges worldwide, as well as support its implementation.

The immediate aim of each working group has been to develop priorities for conservation action, building on the relevant section of the ACAP. Groups were asked to identify the major constraints to achieving conservation success within their subject area, and then to identify specific actions that would contribute to overcoming these challenges.

The first revision to ACAP has now been completed, with each working group having identified short- and medium-term actions (short-term actions should be achievable in 6–12 months, and medium-term actions achievable in 1–5 years in most cases). Due to pressing deadlines priority actions for some groups were developed by the ASG Secretariat and were reviewed by experts, however, since this is an ongoing process working groups will be formed for all thematic areas in the future. The outputs from the working groups are now available on the Amphibian Specialist Group's website, where they will be regularly re-assessed and updated as a living document. It is our hope that making ACAP a living document will allow it to be both dynamic and adaptive to achievements and changing needs.

Several common themes have emerged among each group's priority actions, including: a need to better understand species taxonomy, life history and distributions; development of effective responses to threats; improved communication between researchers and conservationists; better dissemination of data in several languages; the importance of building capacity in areas where it is most needed (both geographically and skills-based); and the identification of suitable funding mechanisms, especially for long-term projects. Furthermore, there are also several cases where groups identified a link between their priorities and those of other groups, emphasising how cooperation is essential for effective amphibian conservation outcomes.

Nyctanolis pernix. Photo: Robin Moore.

Implementation of the priority actions has now begun; the ASA is working on these priorities through its partners, and the ASG thematic working groups (WGs) will be further developing those identified priorities where ASG WGs are best placed to spearhead the process. Each thematic area has been allocated a Lead Contact from the ASG Secretariat, who will help co-ordinate the process in the long-term (see individual working group pages for details of ASG Lead Contacts).

Undoubtedly many of you will already be engaged in projects that contribute to the identified priority actions—please explore the working group outputs to find areas that are relevant to your current and future work. Where your work is in line with the priority actions, we would love to hear more about your projects, and to help raise awareness and assist you to connect with relevant working groups and the ASA. Please contact the ASG Lead Contact for each group for further information.

The Amphibian Specialist Group Secretariat would like to thank all the ASG members who were involved in developing these priority actions, in particular the working group facilitators. Without their passionate commitment and collaboration it would not have been possible, so we greatly appreciate the contribution of your time and expertise.

ASG

IUCN SSC Amphibian Specialist Group

Amphibian Survival Alliance Seed Grant Program: Small Investments with Huge Returns

By Candace M. Hansen-Hendrikx

The Amphibian Survival Alliance Seed Grant Program supports a variety of projects that may not otherwise receive the funding necessary to drive them onwards to success. These relatively small investments provide individuals the opportunity to pilot creative initiatives and research projects that we believe will have a positive impact on amphibians.

In many cases, these grants encourage the multidisciplinary thinking and solutions necessary to address the most pressing conservation issues amphibians face today. Some of the projects we support help build the leadership and local capacity urgently needed for on-the-ground conservation success, while others promote unique collaborations between different groups including academic institutions, governments, organizations, businesses and individuals. By fostering new areas of research while both encouraging and enhancing partnerships, these grants demonstrate that successful amphibian conservation outcomes can be achieved at a relatively low-cost.

When reviewing the projects, we look for those types of projects that are most likely to have on the ground conservation impact. Project don't need to be restricted to the tropics and the focal species don't need to be threatened but if they aren't, there needs to be a clear indication as to how the work can help threatened amphibians.

With that in mind, we are pleased to announce the next open call for seed grant applications. Full details and an online application form can be found at www.amphibians.org/seedgrants. Our seed grants are normally awarded in amounts ranging from USD $500–1,000. While we accept and review seed grants all year, grants are only announced four times per year in *FrogLog*:

- December 1st for seed grants announced in January
- March 1st for seed grants announced in April
- June 1st for seed grants announced in July
- September 1st for seed grants announced in October

As you can imagine, we unfortunately receive more seed grant applications than we are able to fund at this time. If you feel seed grants are important for the future of amphibian conservation and are interested in supporting a seed grant or establishing a fund to support these activities please email Candace M. Hansen-Hendrikx, director of communications and partnerships, to arrange a time to discuss options.

And with that, the Alliance is pleased to announce the latest round of Seed Grants awardees:

- Sara Costa: Ecotoxicological studies as a tool on amphibians' disease risk assessment.
- Dr. Paul A. Hoskisson: The use of eDNA to evaluate and monitor the Golden Tree Frog (*phytotriades auratus*) population.
- Thomas Doherty-Bone: Assessing the status of amphibians on Mountains Kupe and Nlonako in the wake of disease-associated declines in the Highlands of Cameroon.

Ecotoxicological Studies as a Tool on Amphibians' Disease Risk Assessment

By Sara Costa, Emanuele Fasola, Barbara Santos, Ariana Moutinho, Nuno Costa & Isabel Lopes

Amphibian natural populations are strongly affected by pathogenic agents (*1,2*). The risk of disease is dependent on the pathogen environmental conditions, capacity of infection and the host's susceptibility to disease. Changing the environment, in which amphibians thrive, modifies the distribution, increases the risk for diseases transmission and even affects the ecology of populations. Therefore, chemical contamination can act as stressor both for host and/or pathogens and can change the host-pathogen's dynamics (*3,4*). *Saprolegnia* spp. has been responsible for large economic losses in aquaculture industry. Furthermore, it frequently causes infections and population losses in amphibians (*2*). The interaction of environmental changes with pathogen has been assigned as a reliable explanation for the emergence of infections (in places where they were not previously reported). Hence, it is important to understand the effects that environmental changes can have on pathogens and clarify potential interactions between disease and environmental changes. To study this subject, generate new knowledge and promote a more accurate conservation of amphibians, ecotoxicological methodologies will be carried out. Alarming projections for sea levels rise foresee the salinization of low-lying coastal freshwater ecosystems (due to the intrusion of seawater), which will affect ecologically important biodiversity hotspot (which usually holds high amphibian's diversity). It is also aimed to study the effects of metal contamination because it is almost ubiquitous and correlated to anthropogenic activity. Therefore two chemical perturbation scenarios will be studied: increased

Ribeiro da Água Forte, metal contaminated effluent used on ecotoxicological assays. This water body drains an area of mining slug, which have for decades have been a source of acid waters impacting groundwater and soils, including *Pelophylax perezi* habitat in Aljustrel. Portugal. Photo: Sara Costa.

***Pelophylax perezi*. Photo: Cátia Santos.**

salinity (NaCl will be used as a surrogate of seawater) and metal contamination (a mining effluent contaminated with several metals and where *Pelophylax perezi* populations were observed will be used). Data from the ongoing projects, showed that potentially, metal contamination can drive genetic erosion and changes on *Pelophylax perezi* skin microbiome composition.

Lagoa das Braças, a shallow freshwater coastal lagoon, habitat of *Pelophylax perezi* and other amphibian species such Hyla arborea, *Discoglossus galganoi* and *Bufo bufo*. Quiaios, Portugal. Photo: Sara Costa.

The main objective of this project is to assess the role of environmental alterations (metal-contamination and salinization) driving amphibian populations to become more sensitive to infectious agents like *Saprolegnia* sp. To achieve this objective four specific objectives will be tackled:

a) evaluate the effects of metal contamination and salinization on the pathogen agent *Saprolegnia* and in early life stages of the amphibian species;

b) assess the existence of skin bacteria that potentially can inhibit *Saprolegnia* growth;

c) assess the effects of metal contamination and increased salinity on bacteria that inhibit *Saprolegnia* growth; and

d) assess the infection prevalence on eggs and tadpoles, by *Saprolegnia* molt under the specific scenarios of non-effect levels (to amphibians) of salinization and metal contamination.

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The use of eDNA to Evaluate and Monitor the Golden Tree Frog (*Phytotriades auratus*) Population

By Paul Hoskisson

The extraction and identification of DNA from environmental samples (eDNA) has recently shown great potential for the monitoring of endangered and elusive species. The endemic Trindad Golden Frog (*Phytotriades auratus*) is considered Critically Endangered based on its restricted geographical range and decline of its habitat. The distribution of this species is intimately linked to the endemic bromeliad *Glomeropitcainia erectiflor*a and the phytotelma of this species are the only known habitat of this species and its larvae. Current surveying methods for *P. auratu*s require the destructive sampling of the bromeliads and as a result there has been no systematic survey of *P. auratus* in over 20 years. *Phytotriades auratus* is found in only two locations in Trinidad: above 600 metres on the peaks of El Tucuche and Cerro del Aripo.

We have designed and developed a PCR based assay for *P. auratus* using eDNA as a template. This assay is a robust, discriminatory and species-specific assay for use with eDNA templates with a sensitivity of >100 ng of DNA per litre of water. We would therefore like to use the 2015 field season to non-destructively sample *G. erectiflora* phytotelmata on El Tucuche and Cerro del Aripo as a proof of concept study using *P. auratus*, before a more detailed and extensive analysis in 2016. If this approach is successful, it has great potential for surveying many species of rare/elusive phytotelmata-associated amphibians.

Dr. Paul A. Hoskisson and *Phyllomedusa trinitatis*. Photo: Hannah Davidson.

Golden Tree Frog, *Phylotriades auratus*. Photo : Mark Greener.

Meet Amphibian Hero CREA

By Candace M. Hansen-Hendrikx

I sat down with Michael S. Roy, founder and president of Conservation through Research, Education and Action (CREA)—one of our newest Amphibian Survival Alliance (ASA) partners—whose very vision is where every person is an advocate for the conservation of nature to get a first-hand account of what they're doing to help safeguard the future of amphibian sand how you can help as well.

Amphibians the world over are facing probably the world's most serious extinction crisis. What are your thoughts on the future prospects for amphibian conservation and preventing further extinctions?

Habitat destruction, *Bd* and climate change. Can it get any worse? It is the perfect storm and yes we are living in an extinction era. It is likely that many more species will go extinct, some before they are even discovered. But I do not think it is the end of amphibians. Crucial to this conversation is funding, not only for amphibians but also for conservation in general. There simply is not enough of it. Developing countries do not have the financial resources necessary to conserve their biodiversity and so it will require a large international effort to make the difference that is needed. International funding efforts for biodiversity conservation has not come close to what is needed. International commitments to halt human induced extinctions by 2020 is a noble objective but it has been estimated that it will cost approximately $80 billion (B) annually to achieve this, an order of magnitude more than what is currently being spent (*1*). Currently $7 B or so are spent on protected areas worldwide with only $1 B in developing countries where most biodiversity exists. A recent study found that the 40 most severely underfunded countries contain 32% of global biodiversity (*1*). Generally, the future does not look good for amphibians. However, although the conservation community is in a continual state of triage, there are some exciting success stories and efforts taking place to safeguard remaining amphibian species and document new species and ranges.

What do you think are some of the most promising developments in the fight to prevent further amphibian population declines?

Current efforts to breed endangered species *ex situ* has been met with success, however reintroduction efforts will face a large challenge. These efforts will rely on the existence of conservation areas free of disease and free of impacts, unless "immunization" can be shown to be effective. It will be interesting to see the results and lessons learned from recent attempts to reintroduce captive bred species. It is also surprising and humbling to know that we are rediscovering certain species that were caught up in the Central American *Bd* wave. Although their gene pools are likely to have been decimated, some species appear to be hanging

on in some surprising places.

Why did you join the Alliance and what are you doing to help protect amphibians?

The ASA and CREA have a similar philosophy that conservation can only be successful through collaboration and awareness building, all based on a platform of science. CREA aims to protect some of last regions on Earth that hold spectacular biodiversity through several approaches including managing the Cocobolo Nature Reserve in Panama, through which it undertakes research and education activities. Our amphibian research, in collaboration with a number of academic institutions, in addition to documenting species ranges, is looking at niche requirements of vulnerable highland frogs and attempts to understand how some species have shown resilience against *Bd*. We hope this research will support reintroduction efforts in the future. In addition, we are providing opportunities for school and university students to learn about tropical ecology and conservation issues right at the forest frontier in Panama. These courses aim to build awareness based on science and practical guidance on how each one of us can make a difference in any walk of life.

What can the average person, as well as the private sector, do in order to tangibly and actively participate in amphibian conservation?

The average person must seek to gain information on these issues. It was somewhat of a silver lining to hear the *Bd* outbreak in Madagascar be mentioned as top news on the British Broadcasting Corporation (BBC). I would hope that such news inspires people to learn about these issues. Once they have knowledge they will hopefully be inspired to tell others and to demand action from our governments and industry leaders.

Large corporations are now as rich or richer than entire governments and sometimes have disproportionate control over them and their policies. Right or wrong, they do however have the resources to do something about the global extinction crisis. Actively engaging with corporations and finding common ground is essential if we are to conserve global ecosystem function and biodiversity. Finding avenues to engage and educate the private sector in ecosystem service risks that endanger their profits and their reputation can, and has, shifted business models. Private sponsorship of flagship conservation projects has been show to be an efficient and new revenue stream (2). Current examples however are few and far between, but it is up to organizations like the ASA and its members, to continue building the science and the awareness so that incorporating ecosystem health and services into a company's business plan becomes as mainstream as forecasting profit and loss.

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The Yin Yang Frog's *(Leptobrachium leucops)* assessment is due to be published mid-year. Photo: Jodi Rowley.

"Red Listing" to Help Save Southeast Asia's Amphibians

By Tim Cutajar

Southeast Asia has an amazing diversity of amphibians, something we're becoming increasingly aware of due to recent discoveries. A combination of field expeditions and genetic investigation has revealed nearly 100 new amphibian species in the last 10 years alone.

Southeast Asian amphibians are also some of the most threatened in the world. With one of the highest deforestation rates globally and many species being harvested for the pet trade and traditional medicines, it seems clear that this region should be a priority for amphibian conservation.

Despite a large amount of scientific information being gathered about the amphibians of this region, a major obstacle to amphibian conservation in Southeast Asia is transforming this into something clearly actionable. Fortunately, there is a great tool that helps us translate scientific information and expert opinion into data useful for conservation—and that's where I come in.

The IUCN Red List of Threatened Species lets us compile all information relevant to a particular species' extinction risk and determine how threatened it is, which in turn helps us understand the best course of action to save it and which species need our limited conservation resources. This information is then made accessible to conservation managers, and indeed anyone with an internet connection. Multiply that by more than 6,000 frogs, newts, salamanders and caecilians already on the IUCN Red List, and you have the best starting point for holistic amphibian biodiversity conservation possible.

But nearly all "red listing" was being done by a handful of passionate biologists in their spare time, which meant getting those 100 new Southeast Asian amphibian species (plus some older ones) the conservation solutions many of them so desperately need quite late—maybe even too late. However, thanks to the generosity of the Australian Museum Foundation (AMF), a new project is underway to make sure that doesn't happen: the AMF Amphibian Conservation Internship. I'm the lucky intern, and this exciting project is equipping me to dedicate more time and resources than ever to this vital process to help ensure Southeast Asia's amazing amphibians can survive well into the future.

So far, it's going swimmingly; we've already assessed two frogs (the Vampire Flying Frog and Helen's Flying Frog) and a newt (the Lao Warty Newt), each listed as Endangered and now receiving significant attention—check them out at www.iucnredlist.org.

But that's just the start, and there's plenty more to do. I have one day a week for one year to figure out what I can about the distributions, populations, ecology, threats and conservation actions for nearly 100 invaluable Southeast Asian amphibians and get them on that list—wish me luck, and watch this space!

Acknowledgements

This is a super-collaborative project, and I'd like to thank the Australian Museum Foundation for their generous funding, Dr. Jodi Rowley for her amazing expertise and support offered throughout the process, the rest of the Amphibian Red List Authority Southeast Asia Working Group for their tireless work and passion and all of the amphibian experts throughout the region, whose continuous contributions we couldn't do without.

Tim and toad, El Fuerte, Mexico. Photo: Ubaldo Castillo.

The Vampire Flying Frog *(Rhacophorus vampyrus)* is already on the Red List. Photo: Jodi Rowley.

Building Partnerships for Amphibian Conservation: Peru's Amphibian Specialist Group and Yanachaga-Chemillén National Park

By Ariadne Angulo, Rudolf von May & Javier Icochea

Amphibian declines and extinctions are now a recognized global phenomenon, and this reality has not spared the tropical nation of Peru, which is home to more than 570 known amphibian species (*1*) and ranks as the third most amphibian-rich country in the world. Many of its species are range-restricted endemics that occur nowhere else and their survival is threatened by encroaching human activities. While herpetological expertise and knowledge of the country's amphibians have increased notably over the last few decades, the shortage of financial resources poses a continuous challenge to most research and conservation initiatives in Peru, including those that support field and museum-based studies as well as monitoring the status of the country's amphibian populations over time.

One promising alternative to this conundrum would be to establish strategic collaborations with local partners that share similar interests. Local partners that are committed and engaged in the protection of biodiversity and natural resources are a tremendous asset to amphibian conservation: first of all, because they work at the local level, they have a much better understanding of what is happening on the ground, and are in a better position to monitor populations and alert the amphibian conservation community of emerging issues. Second, it is more resource-efficient, as the cost of organizing regular survey and monitoring field trips with herpetologists based elsewhere make conducting these activities on an ongoing basis financially and logistically challenging. Third, local partners that are established in an area usually already have relationships with important stakeholders, in this way helping the amphibian conservation community to establish communications with these parties. On the other hand, local partners can benefit from the expertise and resources that the amphibian conservation community can bring in the form of taxonomic expertise, updated scientific information, and capacity building. It is in this spirit of collaboration that the Amphibian Specialist Group's Peru branch (ASG Perú) and Yanachaga-Chemillén National Park (PNYCH) have established a partnership for amphibian conservation in the Selva Central region of Peru.

Sticker distributed in Oxapampa, central Peru, to promote public awareness about amphibian conservation. Produced with support from the Mohamed bin Zayed Species Conservation Fund.

Park Director Salomé Antezano (left) showing an individual of *Ameerega planipaleae* to youth from the Yanachaga Ecological Brigade. Photo: Ariadne Angulo.

Yanachaga-Chemillén National Park is a 122,000 ha protected area that extends from 460 to 3,643 m asl (http://www.sernanp.gob.pe/sernanp/zonaturismoi.jsp?ID=36) and covers three major ecosystems: Andean grassland, cloud forest and lowland Amazon rainforest. This steep elevational, ecosystem and climatic variation gradient supports a very diverse amphibian fauna in the park and its buffer zone. The amphibians of the cloud forest ecosystem are noteworthy in that they tend to be range-restricted, endemic and potentially threatened by multiple stressors including habitat loss, pollution, disease and climate change.

Individual conversations with PNYCH's leadership in late 2012-early 2013 revealed that they were keen to develop a monitoring initiative of the park's biodiversity. According to the park's Master Plan, a major goal is to develop the knowledge of the status of all threatened and endemic species. In consultation and partnership with the park's leadership, ASG Perú applied for a Mohamed bin Zayed Species Conservation Fund (MBZ) grant to study the conservation status of two Critically Endangered (CR) species that are known from the park's buffer zone: *Ameerega planipaleae* and *Atelopus* cf. *reticulatus*. The ASG Perú-PNYCH project was selected and funded by MBZ in early 2014, and the initial field surveys were carried out in March 2014.

PNYCH has been an exemplary partner for ASG Perú in every way: it waived the project's research and scientific collection permit fee given how important this study is for conservation of the park's biodiversity, it is supporting ASG Perú's field work by providing access to their offices and transportation to field sites and providing park guard assistance for field work. Additionally, park staff coordinated an outreach campaign on amphibian conservation with local youth (the Park's Ecological Brigade, consisting of over 60 students between 11–18 years of age from six local schools) and the Park Director acted as a mediator with contacts with local authorities. ASG Perú, on the other hand, has provided taxonomic and technical expertise, capacity building for park guards and access

to various field resources acquired with the funds from the MBZ grant. Additional projected outputs from the project will include an identification guide to the amphibians of the park's cloud forest.

This experience has motivated both ASG Perú and PNYCH to further pursue development of an amphibian monitoring program tailored for park guards. Park guard patrols are part of PNYCH regular activities and provide a unique opportunity to survey and monitor threatened amphibians both within the park boundaries and within its buffer zone, which is often subject to various anthropogenic impacts such as deforestation. A PNYCH park guard, Dante Lopez, took a keen interest in amphibians as a result of the project and is willing to act as a focal point for amphibians in the park. In addition, thanks to PNYCH's initiative, other protected areas in the Selva Central region (Otishi National Park, Pampa Hermosa National Sanctuary, San Matías San Carlos Protected Forest, Pui Pui Protected Forest and Yanesha and Ashaninka Communal Reserves) have also expressed an interest in having their staff trained in amphibian identification, survey and monitoring techniques, which would allow to scale up efforts for amphibian conservation over much of central Peru's protected area system and respective buffer zones. ASG Perú is currently in the process of exploring funding options to carry out further training and continue to work in partnership with PNYCH to advance amphibian conservation in the threatened cloud forest ecosystem of central Peru.

Park guard Dante Lopez with the Critically Endangered *Ameerega planipaleae*. Photo: Ariadne Angulo.

Former Park Director Genaro Yarupaitán (right) and park ranger Werner Loechle (left) receiving materials from ASG Perú-PNYCH project. Photo: Rudolf von May.

Acknowledgements

We thank the Mohamed bin Zayed Species Conservation Fund for its generous support of our project, PNYCH staff for their support with permits and logistics in the field, Global Wildlife Conservation for assistance with grant management, Peru's Servicio Nacional de Areas Naturales Protegidas por el Estado (SERNANP) and Servicio Nacional Forestal y de Fauna Silvestre (SERFOR) for granting us scientific research and collection permits both within (SERNANP) and outside (SERFOR) of protected areas, respectively, and Edgar Curi and Leoncio Luna for assistance in the field. RvM also thanks the National Science Foundation Postdoctoral Research Fellowship in Biology (DBI-1103087) and the National Geographic Society Committee for Research and Exploration (Grant # 9191-12) for supporting his participation in this project.

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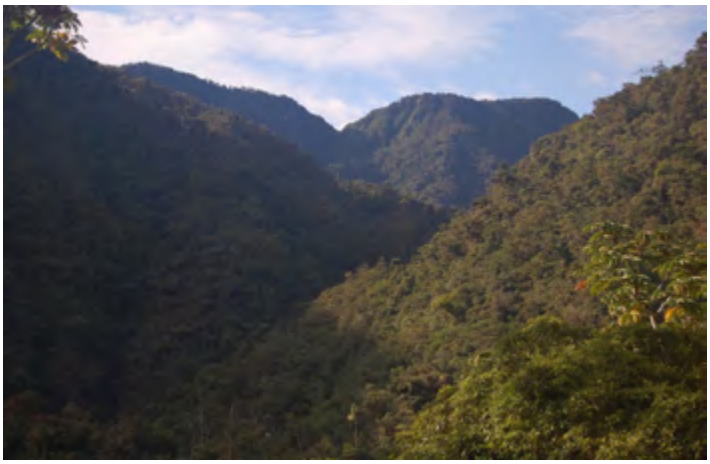


Cloud forests of central Peru, home to many endemic and threatened amphibian species. Photo: Ariadne Angulo.

Dynamic Biodiversity Assessment: Introduction to the Forum on "Amphibian Alliance for Zero Extinction Sites in Chiapas and Oaxaca"

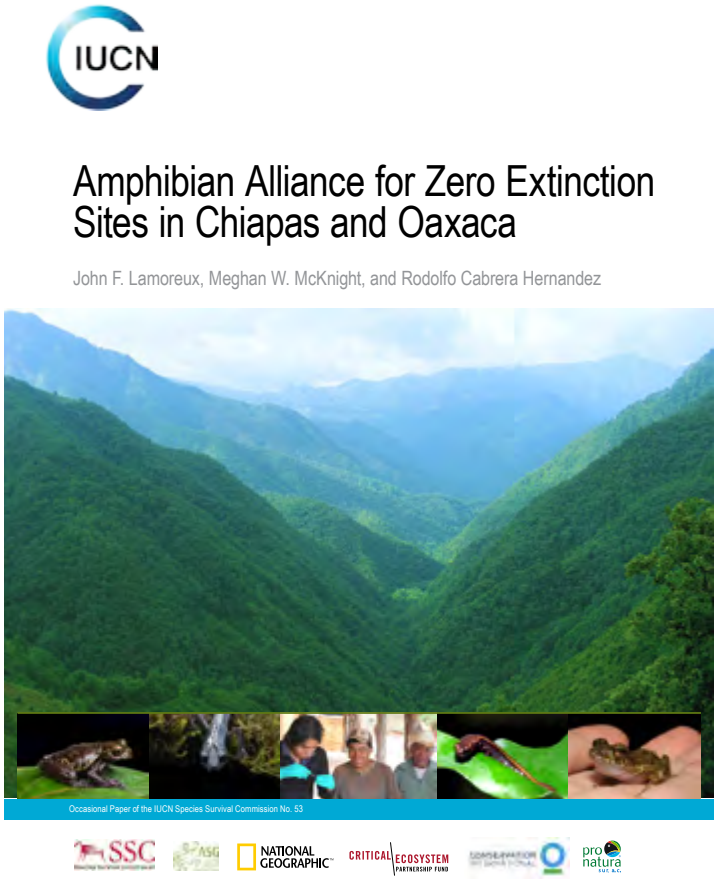
By [1]Thomas Brooks & [2]Simon Stuart

We would like to start by thanking Ariadne Angulo for convening this *FrogLog* forum on John Lamoreux, Meghan McKnight and Rodolfo Cabrera Hernandez's remarkable account of their 2008–2009 studies of "Amphibian Alliance for Zero Extinction Sites in Chiapas and Oaxaca," and for inviting us to introduce it. The commentaries of our co-contributors to the forum, Gabriela Parra Olea, Sean Rovito, Brian Gratwicke and Joseph Mendelson, clearly explain the importance of the book for amphibian conservation in Mexico. Here, we want to place it in the broader context of the critical importance of dynamism in five kinds of biodiversity assessment.

First, taxonomy. While Lamoreux *et al.* is not a taxonomic monograph, the authors' work repeatedly emphasises the conservation importance of ongoing taxonomic exploration and revision. The most obvious example is their discovery of an apparently new species of *Plectrohyla* frog (pages 206–210), of which a description is being prepared by Antonio Muñoz. Many other cases involve revision of species limits, for example, that proposed for *Thorius adelos* (pages 130–131). Conservation needs clarity regarding its underlying units.

Second, extinction risk. More than a decade has passed since the publication of the first Global Amphibian Assessment. The heroic efforts of the Amphibian Specialist Group Red List Authority towards a second global assessment are ongoing, and Lamoreux *et al.* will doubtless be an important contribution to this end. The authors consider the application of the IUCN Red List Categories and Criteria to all narrow-ranged amphibian species in Oaxaca and Chiapas which are actually or potentially Endangered and Critically Endangered. They make numerous recommendations as a result, *e.g.*, that *Pseudoeurycea aurantia*, currently considered Vulnerable, should be re-assessed as Critically Endangered (pages 97–99). Given the urgency of these re-assessments, it is critical that funding be mobilized for them as soon as possible.

Third, identification of Key Biodiversity Areas. Standards for the identification of sites contributing significantly to the global persistence of biodiversity are in the process of being consolidated, but will certainly include Alliance for Zero Extinction (AZE) sites—holding effectively the entire population of at least one Critically Endangered or Endangered species. On the basis of their fieldwork, Lamoreux *et al.* are able to propose comprehensive revisions to the region's AZE site identification (pages 221–273), most of which were available in time to inform the 2010 global revision of the initial 2005 AZE assessment. These revisions included the delineation of the astonishing Sierra de Juárez AZE site (pages 104–145), to which no less than 23 Endangered and Critically Endangered species are restricted. This is more than any other site in the world, although, terrifyingly, Lamoreux *et al.* found just two of these species, and six of them are Critically Endangered (Possibly Extinct) already. Much more work remains, both for AZE (*e.g.*, Lamoreux *et al.* make a strong case that Ayutla (pages 162–170) should be a separate AZE site), and more generally (*e.g.*, to reconcile delineation of the region's AZE sites with the five Important Bird & Biodiversity Areas so far identified in Chiapas).



Fourth, protected areas. It is striking both how few protected areas are documented in the region in Protected Planet (page 311), and how poorly these represent AZE species and sites (page 13). Protected Planet compiles the official UN List of Protected Areas, from national government submissions of protected area data. However, these official data often overlook community level actions to protect sites, including private reserves. The Convention on Biological Diversity's Aichi Target 11 reflects the importance of the full range of governance arrangements for "protected areas and other effective area-based conservation measures," and further to the World Parks Congress in November 2014, efforts are underway to allow documentation of such sites in Protected Planet. This will be critical for Oaxaca and Chiapas, where most site conservation is community-led, sometimes in partnership with Mexico's Comisión Nacional de Áreas Naturales Protegidas (page 14). The heart of Lamoreux *et al.*'s work documents their fieldwork at eight AZE

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sites (pages 82–219), including detailed descriptions of ongoing and recommended actions towards their protection. These draw not only from their biological fieldwork and literature review but also from extensive discussions with the communities living in and around each site. For only one site, the spectacular Cerro de las Flores (pages 212–219), do the authors identify an opportunity for establishment of a national park; in most other cases their recommendations focus on community initiatives.

Fifth, reintroductions. Lamoreux *et al*. make a compelling case (pages 280–285) that conservation of the region's amphibians will require more than captive breeding alone. There needs to be more risk taking. Moreover, information from experimental reintroductions and adaptive management, in response to the devastating threats of habitat loss, water use, fire, climate change and chytridiomycosis (pages 8–56) have to be shared more effectively among practitioners. The Reintroduction Specialist Group and Conservation Breeding Specialist Group will have important roles to play in advancing such ideas.

Lamoreux *et al*.'s work demonstrates powerfully the essential role of field science in continual revision and reassessment of all five of these biodiversity assessment systems. It is eminently feasible to get such boots on the ground: donor investment in this research totalled <$50k over a year-and-a-half; and it all took place within a day's drive of the Western hemisphere's second largest city. We'll therefore conclude with a call for the conservation community to replicate Lamoreux *et al*.'s fantastic example in the many hundreds of other regions where such work is desperately needed around the world.

COMMENTARIES

By Gabriela Parra Olea

The Alliance for Zero Extinction (AZE) is a consortium of 93 non-governmental organizations (NGO) working to protect species that are likely to go extinct without immediate conservation action. In 2005, the AZE published the first list of sites and species that met the criteria required to be considered an AZE site. Interestingly, amphibians make up the largest portion of the global AZE list with over half of the species (AZE 2010). Even though Mexico is only the fifth richest country worldwide in terms of amphibian species, it has by far the greatest number of AZE sites (68) and species (151). Sixty-five percent of Mexico's AZE species are amphibians (64.9%) and they trigger the majority of sites. Also, about one-third of Mexico's amphibian AZE sites and species are found in the southern states of Chiapas and Oaxaca.

This report focuses on sites and species within Chiapas and Oaxaca that qualify for AZE designation. The authors nicely summarize the findings of a project that began January 2008 and ended July 2009. During the course of this project, the authors found ten (possibly 11 if the identification of *Plectrohyla psarosema* is correct) of the 22 highly threatened species they were looking for and produced well-informed overviews of the sites where they occur. They also found a new species of *Plectrohyla* treefrog from the San Cristóbal de las Casas area and several species in the highest IUCN Red List categories. The report is the result of scholarly work both from the field and literature research, with very high quality text and illustrations. It is a complete treatise with the perfect mixture of scientific and anecdotal information and, to the best of my knowledge, it represents the first work that also incorporates the social views of the local communities about the AZE sites and species in Chiapas in Oaxaca, giving both aspects the same importance.

In part I of this report the authors focus on the factors that threaten the well-being and the preservation of Mexican amphibians. The list of factors affecting the biodiversity and conservation of Mexican amphibians is large; however, the authors chose five that in particular are the most important for the country. These threats are: habitat loss, water use, fire, climate change and chytridiomycosis. The authors present very up-to-date and thorough, yet concise, reviews of these topics.

Parts II and III of the report are a series of site profiles, which make up the heart of the book. The profiles discuss the sites and the species they contain in a very user-friendly manner. The authors also provide a delineation of the AZE sites, background of the area (geographic and political), conservation measures,and species accounts. This part of the book is very rich in information and it is presented in a very interesting and fun to read manner. They gathered all the scientific information available for the species, as well as unpublished data and anecdotes from researchers who have worked in these areas or have looked for the target species. They also incorporated all the information they could get from talking to

***Pseudoeurycea juarezi*, Critically Endangered (CR). Photo: Rodolfo Cabrera Hernandez.**

***Cerro de las Flores, Oaxaca.* Photo: John Lamoreux.**

the local people and their own observations and impressions of the area and its species.

In the fourth section of the report, the authors provide a series of recommendations to improve the AZE 2005 list. Many of these are already taken into account in the revised version of the 2010 AZE list. Reading this report and looking at the changes done from the 2005 list to the updated 2010 version, it becomes clear that projects such as these should be conducted for all of the AZE sites and species. Only with this kind of detailed work, can we evaluate and accept or recommend changes to the conservation status of any given species.

Part V, which was called "additional material," contains an up-to-date in-depth review of chytridiomycosis and its effect on amphibian diversity and conservation. The authors provide a series of measures to be undertaken by students and researchers working on this topic. This part is carefully studied, well thought out and the recommendations are based on all the known information about the chytrid fungus and its interactions with amphibians. In my opinion, this section of the report should be read by all students taking a general herpetology class or any organismal diseases class. It took all the known information and provided a concrete list of actions to be done in the laboratory and in the field.

At the beginning of the report, the authors "confessed" they did not consider themselves the best candidates to do the job. In my opinion, they proved to be the perfect candidate because they had one thing that everybody else lacked: TIME. From reading the report it clearly comes across that the authors really devoted themselves full time for a year and a half to this project. The result is a document that contains scholarly data presented in a very fun and easy to read manner with a clear interest and concern about amphibian conservation. I believe this report will be a necessary addition to the library of all biologists interested in Neotropical conservation. It sets an example of what really needs to be done after scientists gather together to propose conservation status categories for any taxon.

***Plectrohyla calthula* (CR). Photo: Meghan McKnight.**

By Brian Gratwicke

The mountainous tropical forests of southern Mexico have an extraordinary concentration and diversity of threatened amphibians. The Alliance for Zero Extinction (AZE) identified 16 small, unprotected sites in Chiapas and Oaxaca that contain virtually the entire known range of some species. While there are 588 AZE sites globally, the extraordinary concentration of sites in southern Mexico captured the attention of authors John Lamoreux, Meghan McKnight and Rodolfo Cabrera Hernandez. Virtually all of these AZE site designations were triggered by endemic amphibians, and this report details a systematic expedition to ground-truth the assessments.

***Plectrohyla acanthodes* (CR). Photo: John Lamoreux.**

The authors evaluate the potential for preventing further habitat degradation at these sites by creating detailed site profiles, surveying amphibians and reporting on conversations with herpetologists and locals. In the process, the authors found 10 of the 22 target amphibian species, and rediscovered four species flagged as Possibly Extinct by The IUCN Red List of Threatened Species. They used this information to help refine the AZE recommendations and consolidate the number of sites for the region from 16 to 12. The report also contains a well-researched and up-to-date review of threats to the amphibians in the area, including habitat modification, invasive species, climate change, pesticides and fire. A separate chapter reviews the incremental progress we have made to understand the amphibian-killing fungus that has severely impacted Neotropical amphibians. Based on a review of the fungus *Batrachochytrium dendrobatidis* (*Bd*) research, the authors present a hopeful outlook on the potential for relevant field-management actions to manage or reduce the threat of chytridiomycosis in the future. The major emphasis of this report, however, is the indisputable fact that if these amphibians are to stand a chance of persisting or recovering in the future we need proactive, creative engagement with locals to protect these special places. I hope that this document will be translated into Spanish and will be socialized in the communities surveyed, and among talented Mexican conservationists that could put the information to good use. In my experience, I have found that people like to be told why their part of the world is special. When people embrace their own defining natural treasures, these special amphibians have the potential to become part of the identity of the people that live there.

By Sean Rovito

Too often, a major disconnect exists between those with information regarding the status of wild populations of threatened species and those seeking to design conservation strategies to protect these species. Herpetologists may spend days, or sometimes just hours, at a site conducting surveys for frogs, salamanders and caecilians. Such surveys are usually conducted as part of other research projects; after all, looking for species that are hard to find (or possibly even extinct) fits poorly with the need to generate publications. Even under the best of conditions, many species (especially of the latter two groups) can be difficult to find, and the failure to locate a particular species on any given day could simply be the result of bad luck. Conservation decisions are often made on the basis of such limited information, sometimes without any input from the people who live in the communities surrounding the species' habitat. For this reason, the work by Lamoreux *et al*. represents a major contribution to amphibian conservation in southern Mexico. The authors dedicated a large amount of time not just to surveying the remaining habitat of threatened amphibian species in the region at Alliance for Zero Extinction (AZE) sites, but also to speaking with local communities about conservation measures they have implemented. As a result of their surveys, the authors found a number of species that had been seen only a few times in recent years, or not at all, and such observations are extremely valuable in conservation assessments. Their work is extensively researched and focuses not just on adding information from field surveys, but also on improving the quality of existing assessments and maps based on these results. It establishes a baseline that can be used for future amphibian monitoring in the region. Perhaps the most lasting impact of the work, however, will be their effort to link community-based conservation initiatives with broader, international efforts to protect the high amphibian biodiversity of the region at AZE sites (and beyond). While species assessments and regional-scale planning are essential, there is no substitute for on-the-ground efforts at habitat protection, which can only be carried out by the people living in these areas. Making connections with these communities requires time, effort and patience; fortunately, Lamoreux *et al*. appear to possess all three of these qualities.

John Lamoreux collecting with a local resident. Photo: Meghan McKnight.

San Cristóbal de las Casas, Chiapas. Photo: John Lamoreux.

By Joseph R. Mendelson III

Meghan McKnight and Rodolfo Cabrera Hernandez with local residents of Concepción Pápalo, Oaxaca. Photo: John Lamoreux.

This is a fascinating document. I honestly have never seen anything quite like this. Using the conceptual framework of the Alliance for Zero Extinction (AZE), the authors visited candidate or identified AZE sites across the states of Oaxaca and Chiapas, Mexico, to attempt to assess the continued existence of AZE amphibian species at those sites. The results are mostly distressing, given that many focal species were not found—or were found in very low numbers. Their field work catalogues the continued human assault on montane forest habitats, and also highlights the overwhelming spectre that chytridiomycosis has cast across this landscape. Tellingly, I found this sentence buried in the text, referring to a site known as Cerro de las Flores, Oaxaca: "*We stopped our vehicle at the water's edge and immediately saw numerous tadpoles of more than one species*" (page 216). This is the most optimistic sentence in the entire 344 pages, yet it reads like the daily observations recorded in the field notes of William Duellman, Janalee Caldwell or Jonathan Campbell from the region in the 1950s, 60s or 70s. The remainder of the text reads like my own field notes from the region in the 1990s or 2000s.

The work is exhaustively thorough in terms of literature cited and general attention to detail. The authors clearly tracked and checked every published piece of information and map, and they identify (and clarify, when possible) innumerable inconsistencies between various sources of information such as the online IUCN Red List accounts—and including some of my own publications. They kept track of the many species-level name changes that have occurred across decades of literature and maps, and heavily relied on published museum records, as well as those listed on HERPNET. My only criticism is that HERPNET is a starting point for reviews of species distributions and such, and anything substantive (such as this project) requires considerable additional work to verify identifications, clear up locality errors, errors in data entry or seek additional specimens. Two examples include the authors' discussion of additional records of the frog *Plectrohyla pycnochila* that demand verification, or two additional specimens of the salamander *Pseudoeurycea mystax* that are in Mexican (UNAM-MZFC) collections, but do not appear on HERPNET for some unknown reason. Such efforts were, understandably, beyond the scope of this work and, to their credit, the authors painstakingly point out many, many instances where there are issues of disagreement regarding some museum records. All conservationists and taxonomists working in this region in the future will want to become aware of all of the issues the authors have raised; this is a very productive aspect of this book.

Beyond the herpetological information, the book features detailed site descriptions (including some nicely recovering sites!) that will inform all future work in the region. The authors went to great lengths to interview local residents on various issues related to conservation efforts, local animals, and recent climatology. Most herpetologists lack the patience and diplomacy such conversations require. I learned a lot about sites that I have visited many times. The book includes extensive discussion of the AZE logical framework, informed discussion of the validity of relevant IUCN Red List rankings, and particularly well-conceived summaries of chytridiomycosis and general recommendations for conservation planning in the context of that disease. My favorite line of text here is: "*Unfortunately, practitioners sometimes regard the success of locating missing species and/or breeding them in captivity as the end goal. The ultimate goal should be the recovery of threatened species, and to accomplish this we suggest adopting a more process-oriented mindset with a greater emphasis on experimental reintroductions accompanied by monitoring and an open exchange of results*" (page 280). I could not agree more strongly.

In summary, this remarkable book functions like a monographic review of an endangered subset of Mexican amphibians, a current status report on those species and their native habitats and a primer for regional conservation planning. The authors' attention to detail and thoroughness is commendable. I will never travel to this region without carrying a copy of this book with me.

Plectrohyla acanthodes **(CR). Photo: Meghan McKnight.**

Agriculture on steep slopes in Totontepec, Oaxaca. Photo: Meghan McKnight.

Protecting Amphibians from Pesticides

By William Quarles

The latest publication from the Bio-Integral Resource Center reviews the connection between pesticides and amphibian decline. In some cases, amphibians are killed directly upon exposure. There are also sublethal effects such as delayed metamorphosis, increased predation, reduced size, reproductive problems, deformities and depressed immune systems that lead to destruction. We identify which pesticides are causing the worst damage.

Amphibians are exposed through direct overspray, pesticide drift, rainfall and runoff into water bodies. About 83% of urban streams and 57% of agricultural streams are contaminated with enough pesticide to be hazardous to aquatic life. Windblown pesticides from aerial applications are killing amphibians in remote mountain areas.

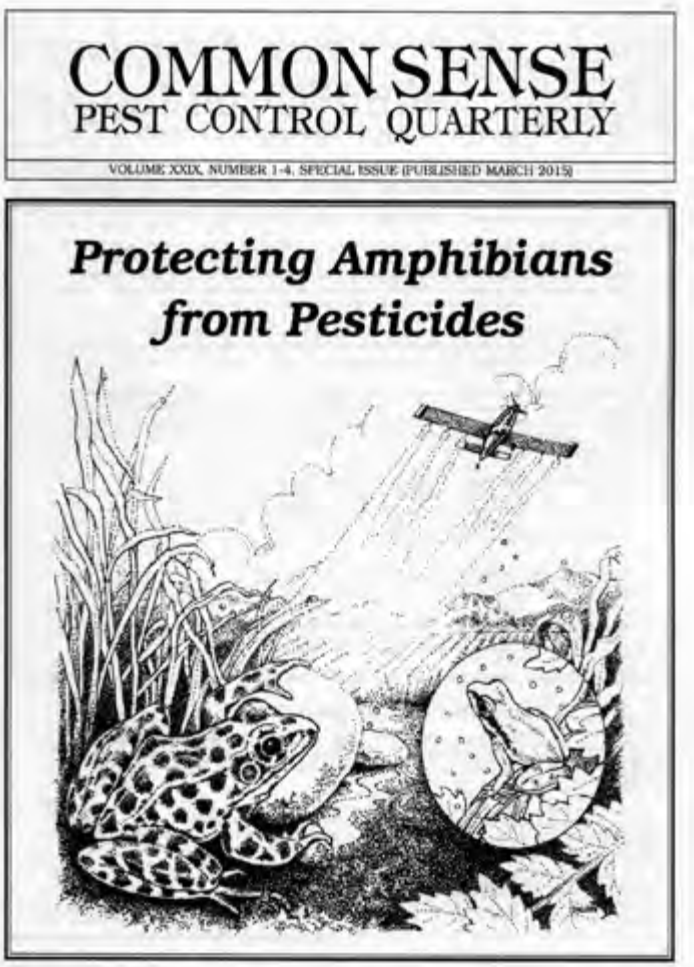
We also provide solutions to the problem. Some pesticides such as glyphosate, atrazine, endosulfan, chlorpyrifos and chlorthalonil should be used much less or restricted. Glyphosate and atrazine together represent 30% of all conventional pesticides used in the U.S.

We can also protect amphibians by using IPM methods in agriculture, homes, gardens and professional landscapes. A combination of cultural methods, resistant plants, conservation biocontrol and reduced risk pesticides can be effective.

Buying organic food, replacing lawns with native plants, creating backyard water gardens, helping with wetland restoration and using mechanical methods, mulches and competitive plantings for weed control can help protect amphibians from pesticides.

Backyard gardens with resources for amphibians, birds, butterflies, bees and beneficial insects are aesthetically pleasing and can help prevent the relentless slide of amphibians toward extinction.

You can download a copy of "Protecting Amphibians From Pesticides" here.



The Website for the Admirable Red-Belly Toad is Online

By Luis Fernando Marin da Fonte

A website devoted to the Admirable Red-Belly Toad is now available online. *Melanophryniscus admirabilis* is a microendemic and Critically Endangered species from the southern Brazilian Atlantic Forest. On the website you can find information about its natural history and the place where it occurs (just along 700 meters of a river); discover threatens to its small and only known population; read the latest news (especially, regarding conservation matters); and also find all the published scientific information about the species, including a special report published in *FrogLog* (*1*) telling how a multi-institutional effort stopped the construction of a hydroelectric power plant that recently threatened the species. Additionally, you can enjoy some videos, audios and pictures of this beautiful toad. Since it is a Brazilian species and our intention is to improve environmental awareness and education in the region, the main language of the website is Portuguese (but you can also find a special section and some scientific information in English). Furthermore, you can register for the Admirable Red-Belly Toad Friends Club ("Clube dos Amigos do Sapinho"), receive your Club Membership Card and thus help to protect it by your efforts. Take a look at the website and at our Facebook Fan Page. Share it with your friends and help us spread the world the story of this admirable species!

Melanophryniscus admirabilis. Photo: Luis F.M. Fonte.

More than 100 people attended the fourth ACRS, Cambridge, 2015. Photo: Evie Button.

ACRS 2015: Empowering the Future of Amphibian Conservation

By [1]Candace M. Hansen-Hendrikx, [1]James P. Lewis, [2]Helen Meredith, [3]Christopher Michaels, [4]Rachael Antwis, [5]Collin VanBuren & [4]Che Weldon

The Amphibian Conservation Research Symposium (ACRS) is the only international series of conferences devoted to sharing research and strategies to empower the future of amphibian conservation. In 2015 ACRS was adopted, through a joint initiative, by the Amphibian Survival Alliance (ASA), the world's largest partnership for amphibian conservation and the IUCN SSC Amphibian Specialist Group (ASG), a global network of experts building a foundation for conservation. ACRS has remained true to its roots dating back to 2012 by bringing together amphibian conservationists and researchers from around the world to gain experience, learn new ideas and make contacts.

Each year individuals present talks and posters detailing evidence-based approaches and management strategies that promote amphibian conservation and this year was no different. Held between between April 11th-12th at the University of Cambridge, ACRS 2015 was the biggest yet, with more than one hundred attendees from 55 institutions in 18 countries. The atmosphere over the two days was buzzing with enthusiasm and energy as amphibian conservation practitioners and scientists discussed projects and planned for the future. With 17 presentations over the course of the symposium, there were a diverse array of topics covered including amphibian ecology, taxonomy, disease, captive husbandry and *in situ* management. These included talks presented by researchers like Gemma Harding (University of Kent and DICE, UK) who asked the question if amphibian reintroductions conformed to IUCN protocols and Edward Netherlands (North West University, South Africa) who assessed the effectiveness of Passive Acoustic Monitoring for determining species diversity at a locality when compared to traditional methods. The abstract program book can be downloaded here.

Three key-note presentations were delivered covering three different, but equally important, areas of amphibian conservation. Sarah Thomas from the Zoological Society of London spoke on the importance of considering social aspects of conservation issues when establishing or assessing success of conservation interventions, such as what animals people are willing to help save or what social programs actually succeed in teaching people about

[1]Amphibian Survival Alliance. [2]IUCN SSC Amphibian Specialist Group. [3]Zoological Society London. [4]North-West University. [5]University of Cambridge.

conservation issues. Rebecca Smith from the University of Cambridge highlighted the need for the right kind of evidence (*i.e.*, data collected from controlled studies to show what the effect of an intervention is when compared to a scenario where no intervention was undertaken) required to make decisions about the best management strategies and showed where to find it on the Conservation Evidence website. Finally, Jennifer Luedtke, the Amphibian Red List Authority Deputy Coordinator, explained The IUCN Red List of Threatened Species in detail by showing what the different classifications mean and how easy it is for researchers to make much-needed assessments on their focal species by presenting a case study on the Red List status of Helen's Tree Frog (*Rhacophorus helenae*).

Talks were also given by three Future Leaders of Amphibian Conservation, the first cohort of a new program started at this year's ACRS, which aims to bring in up-and-coming leaders into the field of amphibian conservation who may not have otherwise been able to attend the event, enabling them to share their innovative work in the field. This year's Future Leaders were Arun Kanagavel from India, who presented his research as an EDGE fellow working on people's perceptions of amphibians in the Western Ghats; Esther Matthew from South Africa, who presented her research on the use of sniffer dogs as a new method for finding elusive species for monitoring; and Karla Pelz Serrano from Mexico, who presented both the social and biological work she is doing in Mexico on the Critically Endangered Lake Lerma Salamander (*Ambystoma lermaense*). The attendance of these individuals at ACRS 2015 was made possible through funding from Women in Wildlife, Nordens Ark and Prof. Richard Preziosi from the University of Manchester.

In keeping with the new international theme of the symposium, ACRS 2016 is likely to be held in Potchefstroom, South Africa, in late January (details to be confirmed). This university town is home to North-West University, where the meeting will be hosted, and is located about 1.5 hours drive from Johannesburg, making it relatively accessible for Europeans, Africans, and other attendees from around the world.

ACRS 2015 would not have been the success it was without the generous support of Chester Zoo, the British Herpetological Society, VetArk Professional, Nordens Ark, Peregrine Livefoods, Arcadia and Shepreth Wildlife Park.

As we look to the future, the ACRS Steering Committee would like to continue providing an annual award to early career conservationists as Future Leaders of Amphibian Conservation from around the world. This award not only provides the winners with an ability to present at the symposium, but, more importantly, it provides them with an opportunity to network with other attendees and become Fellows of this new initiative. In addition to providing the support necessary for the award recipients to attend the symposium, it is our intention to also work with them to build a support network of experts, organizations and funders that will help them achieve their conservation goals and, most importantly, remain in the field of amphibian conservation for the long-term. We encourage you to be part of our long-term vision for ACRS and welcome your support for this initiative and these Fellows in future years. We are happy to discuss these sponsorship opportunities with you further.

We would also welcome general funding support for the symposium itself, which helps to keep the registration cost low, which is particularly important for students. Please contact the ACRS Steering Committee to start the conversation.

The future of amphibian conservation, research and education depends on what we do now as a community. ACRS provides all of us with a chance to work together and support the amphibian conservation leaders of tomorrow, helping to develop them into the confident, experienced and well-connected leaders we need to make a real difference for amphibians.

We would like to thank everyone who participated in this years symposium and we hope you will join us in the years to come.

ACRS Future Leaders of Amphibian Conservation 2015. From left to right, Dr. Karla Pelz Serrano (Mexico), Arun Kanagavel (India) and Esther Matthews (South Africa). Photo: Andrew Stephenson.

ACRS

Amphibian Conservation Research Symposium

Complex Bacterial Challenge in Fight against Deadly Amphibian Disease

By Morwenna Grills

Chytrid fungus (*Batrachochytrium dendrobatidis*) is thought to be behind the decline or extinction of at least 200 species of frogs. It is also one of the reasons why 31% of amphibian species are currently listed as Threatened by the International Union for the Conservation of Nature.

This latest study used bacteria from frogs in Belize to test the limitations of probiotic treatments. This form of treatment aims to introduce bacteria cultivated from amphibians that aren't affected by the disease to those at risk of infection to boost their immunity.

Dr. Rachael Antwis who carried out the study while completing her Ph.D. at Manchester's Faculty of Life Sciences explains, "Using beneficial bacteria to act as 'probiotics' for disease mitigation is already common in agriculture and human health. In fact, many bacteria that reside on amphibian skin have been shown to inhibit the growth and survival of *B. dendrobatidis*. However, the reliability of the potential probiotics hasn't been tested against the shifting targets the disease presents."

To assess the effectiveness of probiotic treatments, the team used bacteria taken from frogs in Belize, where the species has shown resilience despite the long-term presence of the disease in the area. Fifty-six strains of bacteria were isolated and stored for use in the laboratory.

The team challenged the bacteria against different genetic strains of the disease and then looked at whether the bacteria had inhibited the growth of the disease in its various forms. They found the bacteria performed in a variety of ways with only a small number inhibiting all forms of the disease. The bacteria that had an impact on one strain of the disease didn't have the same impact on the other genetic variations.

Dr. Trenton Garner from The Zoological Society of London's Institute of Zoology explains the results: "Because only a small proportion of the bacteria that could be used as candidate probiotics showed broad-spectrum inhibition against the global pandemic *B. dendrobatidis* lineage, we believe probiotic treatments are unlikely to be consistently successful when confronting a variety of fungal genotypes. Because of the enormous genetic variability of the disease and its ability to rapidly evolve, it's vital that any treatment takes this into account."

***Agalychnis callidryas*. Photo: Rachael Antwis.**

***Agalychnis moreletii*. Photo: Rachael Antwis.**

He continues,"We suggest that a variety of bacteria be used when creating probiotic treatments as this is likely to offer more comprehensive protection of hosts from *B. dendrobatidis* and other threatening amphibian pathogens."

The results of the study have been published in the journal Applied and Environmental Microbiology.

Moving forwards the scientists say that further research is needed to fully understand how bacteria inhibit *B. dendrobatidis* growth and the ability to infect hosts. Looking at the *B. dendrobatidis* genome for virulence factors will be fraught with difficulty but this study demonstrates some hope for finding effective probiotic treatments from within the amphibian community.

Dr. Antwis concludes, "A lot more work is definitely needed before we can identify an effective cure for this devastating disease. But as a scientist I believe we not only have a moral obligation to keep searching, but an ecological one too. Amphibians inhabit the middle of food chain, making up a vital part of our ecosystem. If they go then that could spell disaster for many more species."

Mantidactylus pauliani is a Critically Endangered species threatened by habitat destruction. The species can be found only in the massif of Ankaratra, where the prevalence of chytrid in 2013 reached up to 100%. Photo: Gonçalo M. Rosa.

Chytrid Fungus Found in Madagascar: An Update of Planned Conservation Strategies

By Angelica Crottini, Ché Weldon, Dirk S. Schmeller, Eric Robsomanitrandrasana, Falitiana C. E. Rabemananjara, Franco Andreone, Gonçalo M. Rosa, Hiarinirina Randrianizahana, Jeff Dawson, Matthew C. Fisher, Molly Bletz, Nirhy H. C. Rabibisoa, René Rabenarivahiny, Sahondra Rabesihanaka, Tsanta Fiderana Rakotonanahary

The amphibian pathogenic fungus known as *Batrachochytrium dendrobatidis* (*Bd*), has now been detected in Madagascar (*6*). This pathogen has already caused dramatic declines of frog populations in Central America, Australia, the western United States, Europe and east Africa (*5, 10, 11*). Using molecular diagnostic assays, over 4,100 amphibian specimens have been screened for *Bd* between 2005–2014 at many locations across the country. The first record of *Bd* surprisingly occurred in 2010 in the Makay Massif, a very remote area in western Madagascar. In the following years, *Bd* was confirmed at the same place and detected in four other locations: Ankaratra, Antoetra, Ankarafantsika and Ranomafana. Most notably in Ankaratra and Ranomafana, *Bd*'s prevalence reached between 50% and 100% (*6*).

The detection patterns of *Bd* appear to be following trends similar to other tropical regions of the world, where the likelihood to detect *Bd* seems to be higher in the dry, cooler season and in association with mid-high elevation locations (*6*). Furthermore, some of the *Bd*-positive samples indicated that they belong to the highly virulent Global Panzootic Lineage (*Bd*GPL). This lineage has been found on every continent where amphibians live and is associated with almost all known *Bd*-mediated amphibian declines (*8*). However, fortunately to date, the detected infection intensities have been low and no die-off events related to the chytrid fungus have been reported in Madagascar. Thus, it is also possible that the *Bd* present is a hypovirulent introduced lineage, or an endemic lineage that has always been present but has evaded detection until now.

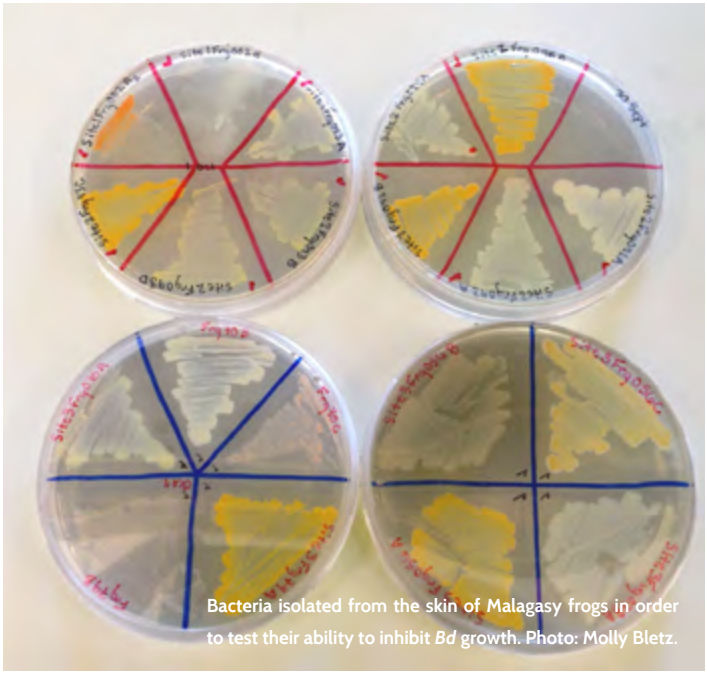
Finding *Bd* in Madagascar poses a crucial conservation risk to the World's amphibian diversity, as Madagascar is home to about 500 frog species, of which more than 99% are endemic to the island. Importantly in November 2014, a conservation meeting, ACSAM2 (A Conservation Strategy for the Amphibians of Madagascar 2) brought together a national and international panel of experts to confront the threats to Madagascar's amphibians. During this meeting, the potential crisis of *Bd*'s presence in Madagascar was highlighted and a new Action Plan for the conservation of Malagasy amphibians will be released soon, including a section on emerging infectious diseases (*1*). The consensus of the group was that continued research to elucidate the "unknowns" surrounding the detection of *Bd* in Madagascar alongside concerted efforts to develop and implement conservation actions, are critical. Among the major outputs of this meeting is a plan to continue the National Monitoring Program (see *15*) in order to gather robust data on the patterns of prevalence, intensity and distribution of *Bd* across the island; to develop an emergency response strategy for the amphib-

ians of Madagascar including planning and building captive assurance populations of priority species; to streamline permitting issues in order to prevent rapid responses from being held up in bureaucratic holes.

Another important response is to identify the *Bd* lineage(s) and to characterize its/their virulence(s). Researchers are currently trying to isolate and culture *Bd* from infected frogs in the field. With a living culture of the Malagasy *Bd*(s) it will be possible to determine the evolutionary history of the Malagasy *Bd* within the context of the global genomic diversity of *Bd*, its origin and it's effects on Malagasy frogs upon exposure.

Development and implementation of mitigation strategies in the event that populations begin to be negatively affected by *Bd* is another key approach. Among the available mitigation strategies is the use of skin probiotics. That approach is based on the principle that beneficial bacteria that inhabit amphibians' skin can inhibit and kill the chytrid fungus (*7*). Laboratory and field trials have shown that when one of these beneficial anti-*Bd* bacteria is added to amphibian skin through a probiotic bath, the amphibians are able to survive *Bd* infection (*e.g.*, *9*, *14*; but also see *4* in press). *In vitro* assays of Malagasy amphibian skin bacteria against *Bd* have shown that some of the collected bacteria can strongly inhibit *Bd* growth, suggesting that probiotics may help to prevent infection. On another front, the characterization of planktonic microorganism communities in relation to *Bd* presence and absence may also lead to an effective landscape level mitigation strategy (*13*). Bioaugmentation of these communities with *Bd*-consuming microfauna may inhibit the colonization of *Bd* in new habitats. Conservationists are working in order to better understand the dynamics of these systems and quickly develop an effective strategy.

Over the last few years, the amphibian research community succeeded in creating a dynamic group of experts working in synergy to mitigate several treats affecting the amphibians of Madagascar (*2*, *6*, *12*, *15*). The Chytrid Emergency Cell in Madagascar is working closely with the Amphibian Specialist Group-Madagascar to coordinate all initiatives and mitigation actions against chytrid at the national level (*12*). Supporting the coordinated initiatives of this network will hopefully result in successful actions to fight this new challenge for amphibian conservation in Madagascar.



Bacteria isolated from the skin of Malagasy frogs in order to test their ability to inhibit *Bd* growth. Photo: Molly Bletz.

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Researchers sampling frogs for chytrid detection at Isalo National Park. Photo: Franco Andreone.

César with *Crocodylus intermedius* juveniles in Santos Luzardo National Park, Apure state, April 2011. Photo: Omar Hernández.

In Memoriam: César Ramón Molina Rodriguez (1960-2015)

By Omar Hernández

César was born May 31, 1960, son of Julio Ramón and Ezequiela Luisa. He was always very proud of his parents given that his father, a labourer at Cementos La Vega and his mother, a housewife, made every effort to give him everything to achieve and education and graduate at the university level. His childhood and adolescence took place at Parroquia La Vega in Caracas, and from what he recounted to me about that period, it was during his walks as a child in the few remaining wild areas that still existed in the Haciendas La Vega and Montalban, that he discovered his vocation as a wildlife biologist; times where he always scuffled with other children hunting birds(luckily for me, we had not met each other at that time).

He graduated as a biologist from the Central University of Venezuela in 1991. Always with an affinity for herpetology, his undergraduate degree thesis evaluated the diet of the Green Iguana in the Llanos, specifically in the Fundo Masaguaral, a known conservation cattle ranch.

His first job as a biologist was as head of the Unit of Fauna of the "Project for the Inventory of Natural Resources of the Guayana Region" for the Venezuelan Corporation of Guayana, where he participated in numerous scientific expeditions to the rainforests of Bolivar and Amazonas states. It was during one of these expeditions to the upper Orinoco where he spent time with the local indigenous people and befriended a little Yanomami girl named Jaema—a name that years later he would give to his own daughter, today a beautiful teenager of whom he always spoke very proudly.

He participated in over twenty national expeditions to the rainforests of southern Venezuela, he worked in ten environmental assessments for the petroleum and petrochemical industry, was Curator of Herpetology at the Science Museum Foundation in Caracas, working later as researcher of the Herpetology Section in the Museum of Natural History of La Salle Foundation of Natural Sciences in Caracas. He was also a fauna consultant specialist in various environmental impact studies.

In 2003 he graduated as Doctor in Biology, with a specialization in Ecology, at the Central University of Venezuela , where he received honours for the quality of his thesis "Ecology of *Mannophryne herminae* (BOETTGER, 1893) (ANURA: DENDROBATIDAE) in the Cordillera de la Costa, Venezuela" an honorific mention was granted to him.

In the savannas of Capanaparo, Apure state. To the right of César, his student Alejandro Moreno; to his left, local expert Elin Bastida and Director of Inparques Apure, Jesús Rodríguez, January 2011. Photo: Omar Hernández.

In 2004, he was appointed Director of Biological Trade and Biosafety in the National Office of Biological Diversity of the Ministry of the Environment, where I can still remember his frustration trying to minimize the red tape that is so detrimental to scientific research in the country.

Later in 2007 he won the academic contest as Professor for the School of Biology of the Central University of Venezuela (UCV) ,and was appointed as researcher at the Laboratory of Biology and Conservation of Amphibians and Reptiles of the Institute of Zoology and Tropical Ecology of this university.

His contribution as a person in search of a better country was also extended to a variety of volunteer roles including as Regional Chair for Venezuela for the IUCN SSC Amphibian Specialist Group, Treasurer of the Board Directors of the Crocodile Specialist Group in Venezuela, Secretary of the Association of Biologists of Venezuela, Member of the Directive Board of the Asociación Civil Provita, and referee for various journals and instructor in many courses.

César was a tenacious researcher and much of his research was financed with his own money and using his own vehicle. Devotee of hard work in the field, throughout his short 15 year career in research institutes he left an impressive and extensive contribution to the knowledge of Venezuelan wildlife, with 90 scientific publications in journals, books and 56 scientific abstracts or papers presented at conferences and workshops. Obviously his productivity as a scientist was constant throughout his short life, including publishing during the years he occupied management and administrative positions.

We were friends back in the first semester at university and for over 30 years, but especially in the last years we became even closer friends, sharing long conversations on Friday nights, where we talked about every imaginable subject. It is now in these gatherings when we, his friends, miss him the most. Few people are as clear and direct as César, and although for some this trait was difficult to

In Ranch Santa Luisa, Apure state. César with his student Ingrid Márquez, January 2013. Photo: Omar Hernández..

deal with, many of us appreciated his honest and direct approach, straight forward, without detours or half-truths, always sincere and eager to help. He earned great appreciation from his students, as can be seen from the fact that in his short seven years as professor at the university he supervised 16 undergraduate and graduate theses, and at the time of his departure, many students were about to begin their theses with him. He motivated UCV students to research amphibians, a taxonomic group with many species on the verge of extinction, but under-appreciated at the country level by some.

With his premature passing a lot of conservation projects and actions remain in the inkstand; some of those plans and activities which we would so often discuss, but we always thought that we still had a lot of time to develop them. When he passed away, we not only lost a friend and a colleague, but someone we could rely on to continue with the arduous and on occasions, discouraging task of preserving the biodiversity of our country.

But my most cherished memories of César are when we were students and our major worry was not to miss a party where we could dance "salsa," and now, thinking of him and his legacy, Célia Cruz's version of the song "I will Survive" comes to my mind:

"Listen to my sound, my old sound
It has the key of any generation
In the soul of my people, in the leather of the drum
In the hands of the "conguero," in the feet of the dancer
I will live, there I will be
While a dance troupe passes with my rumba I will sing
I will always be what I was, with my sugar for you
I will live, I will live"

In Río Capanaparo, Santos Luzardo National Park, Apure state. César assessing *Podocnemis unifilis* and *Podocnemis vogli* turtle shells, both species consumed by the local population, January 2011. Photo: Omar Hernández.

In Ranch La Fe, Guárico state. César giving a course during the IV Ecology and Conservation Course of Crocodylia of Venezuela, February 2012. Photo: Omar Hernández.

In Ranch Santa Luisa, Apure state. César assessing research site with two of his students, Ingrid Márquez and Carlos Amilibia, January 2013. Photo: Omar Hernández.

In Ranch Masaguaral, Guárico state. César giving a course during the IV Ecology and Conservation Course of Crocodylia of Venezuela, February 2012. Photo: Omar Hernández.

Spanish translation can be found here.

The EDGE Fellowship Programme

Building capacity to save the world's most extraordinary species

The Zoological Society of London's EDGE of Existence programme (www.edgeofexistence.org) is the only global conservation initiative focusing specifically on threatened species that represent a significant amount of unique evolutionary history. One of the most effective ways in which the programme is working to secure the future of EDGE species is by awarding two-year Fellowships to future conservation leaders ("EDGE Fellows") working on poorly-known EDGE mammal, amphibian, bird or coral species. We now seek applications for the 2016-18 cohort.

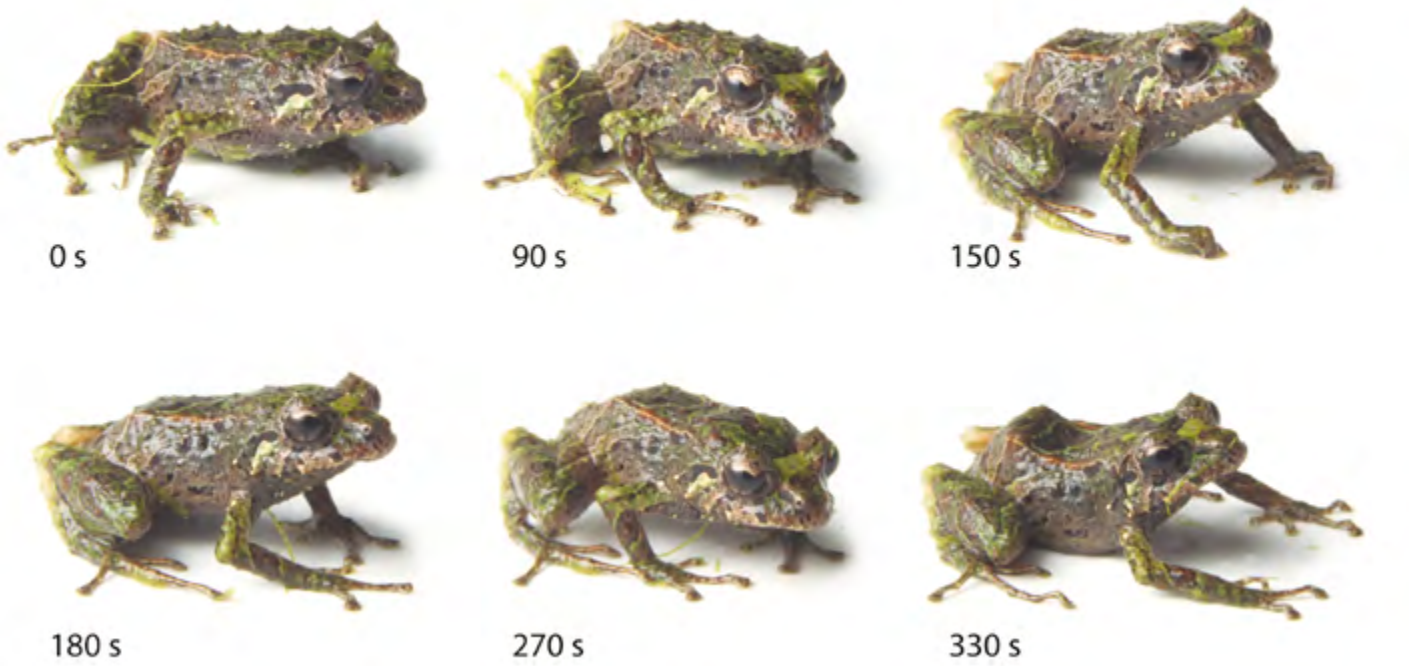
EDGE Fellows follow a comprehensive two-year training programme comprising of:

- A grant of £8,000 to undertake a 2-year project on a top-100 EDGE species
- A 4-week Conservation Tools training course
- Ongoing technical support and mentoring
- A 2-week Conservation Leadership training course in London

The application form, guidelines, and eligibility criteria are available to download on the EDGE of Existence website (http://goo.gl/YtI9VX) or can be requested from the EDGE Fellows co-ordinator (olivia.couchman@zsl.org). We strongly encourage applicants to discuss their projects with us before applying, and are happy to give feedback on a draft of the application, providing they are sent far in advance of the deadline.

The EDGE Fellowship application deadline is **15th June 2015**. All applications will be reviewed by a panel of experts. Successful applicants will be announced by the end of August 2015.

This figure shows the variation in the Mutable Rainfrog's skin texture, as shown in the Zoological Journal of the Linnean Society. Photo courtesy of the Zoological Journal of the Linnean Society.



Shape-Shifting "Punk Rocker" Frog Discovered in Cloud Forest of Ecuador

By Lindsay Renick Mayer & Candace M. Hansen-Hendrikx

When Katherine Krynak, a PhD student at Case Western Reserve University, pulled the frog out of its container and placed it on a smooth white board for her husband Tim Krynak to take a photo, she was overwhelmed by disappointment. She and Tim, project manager at Cleveland Metroparks Natural Resources Division, had spent three years looking for what they believed was a new frog species, one they had found in the cloud forests of Ecuador and named the "punk rocker" frog because of its thorn-like spines.

But this frog, the one now in front of them, was decidedly smooth.

So Katherine put the frog back in the container, setting it gently on moss. It was then that something extraordinary happened: within moments the frog changed from smooth to spiky. Not only had the pair discovered a new species, they had discovered a new species that could do something no other vertebrate had been known to do.

"Discovering a new species is incredible enough," Katherine says. "You wouldn't think anything could top that. And then you discover that it also changes shape, suddenly growing spines that then disappear. I just kept asking 'did that really happen?'"

Katherine, Tim and researchers from Reserva Las Gralarias, Universided Indoamerica and Tropical Herping in Ecuador revealed their findings in the Zoological Journal of the Linnean Society, challenging traditional scientific knowledge and underscoring the critical need for habitat protection in the Ecuadorian Andes. Farming practices, urban sprawl and mining continue to put pressure on the Ecuadorian cloud forests.

"These types of new discoveries, those that defy—and improve—our understanding of the natural world, are only possible as long as we protect the most biodiverse corners of our planet," says Robin Moore, conservation officer at the Amphibian Survival Alliance. "If we close the door to new discoveries, we may never be able to fully understand the extent of what we've lost."

The Krynaks discovered the new species, called the Mutable Rainfrog (*Pristimantis mutabilis*) in 2006 at nature preserve Reserva Las Gralarias. Researchers believe the species' secret shape-shifting skills may help the marble-size frog be better camouflaged in its mossy surroundings.

The papers' authors worked to confirm that the Mutable Frog is a new species by conducting morphological and genetic tests and studying the frogs' calls. They found that the Sobetes Robber Frog (*Pristimantis sobetes*), a related species, can change its skin texture, too. According to paper co-author Carl R. Hutter, from the University of Kansas, the next steps in the research are to determine if additional species share this ability and then to look for the physiological mechanisms that allow the species to do what scientists once thought only invertebrates such as octopuses and cuttlefish could do. The finding underscores the importance of thorough scientific observation, Hutter says.

"Take good photographs of the frog in the wild when it is found and again observe the frog later to see if anything has changed," Hutter says. "The individual should be carefully observed several times."

The Mutable rainfrog was discovered in the humid, mossy cloud forest habitat in Ecuador's Reserva Las Gralarias, an Amphibian Survival Alliance partner: Photo: Timothy J. Krynak.

The Krynaks helped form Las Gralarias Foundation, an Amphibian Survival Alliance partner, to support the reserve's conservation efforts. They plan to continue surveying Mutable Rainfrogs and to work with fellow researchers to document their behavior, lifecycle, and to estimate their population, all in effort to better conserve both this paradigm-shifting species and its habitat.

"We need to know its natural history characteristics," Katherine says. "When it breeds, how its eggs develop, when they hatch, what do these guys eat, what's their preferred habitat, what's their home range, how much can they possibly move, are populations isolated because of this ability or inability to disperse? We don't know any of these things. In order to protect them, we need to learn these life history characteristics so we can protect habitat in a way that is conducive to their long-term survival."

Photo: Timothy J. Krynak, courtesy of the Zoological Journal of the Linnean Society.

Photo: Lucas Bustamante, courtesy of the Zoological Journal of the Linnean Society.

Incilius campbelli. Photo: FUNDAECO.

Protecting Amphibian Habitats Now: A Canadian International Conservation Organization Weighs In

By Anne Lambert

When we founded the International Conservation Fund of Canada (ICFC) in 2007, we didn't need convincing that amphibians were a taxonomic group of particular concern. We knew of widespread declines in amphibian populations and that a much higher proportion of amphibian species were threatened with extinction than birds or mammals. We understood something of the special vulnerability of amphibians and the problems of habitat loss and fragmentation, deteriorating water quality, the spread of diseases and rapid climate change.

ICFC aims broadly to protect the world's most threatened and diverse ecosystems and species, with most of our work in the biodiverse tropics where the funding gap for conservation needs is greatest. Amphibians have been a prime focus of several of our projects, as described below, and amphibians also benefit from our work in protecting landscape-scale reserves such as the Kayapo Indigenous Territories in Brazil (which span 11 million hectares—30% larger than the island of Ireland) and Peru's Los Amigos Conservation Concession, which spans 146,000 hectares of old growth Amazonian forest and indirectly protects an additional million hectares of state lands including Manu National Park and a large Territorial Reserve for Indigenous People in Voluntary Isolation.

MORNINGSIDE, SRI LANKA

One of our first two projects, with Conservation International and the Wildlife Heritage Trust (Sri Lanka), aimed to achieve protected status for the Morningside cloud forest in Sri Lanka. These government-owned lands, which lie east of the Sinharaja World Heritage Site, were known to be home to five amphibian, three lizard and three freshwater crab threatened species. (The project's biodiversity surveys also led to the discovery of three new species—a shrew, a gecko, and a frog.) With less than 5% of Sri Lanka's cloud forest remaining, with Morningside's importance as a center of endemism, and with the threat of timber production leases and conversion to agriculture, it was a clear conservation priority.

The joint initiative resulted in the land being turned over (at no cost) by the Land Reform Commission to the Forest Department. Thus, 1,000 hectares, including all of the area in which Morningside endemics occur, is now under the Forest Department's jurisdiction and in better conservation shape than before the initiative took place.

SIERRA CARAL, GUATEMALA

ICFC was proud to be one of the organizations involved in acquiring land to create the 1,900-hectare Sierra Caral Amphibian Conservation Reserve in Guatemala, an achievement many *FrogLog*

Bradytriton silus. Photo: Robin Moore.

readers will have heard of (see *FrogLog* issues 95 and 111). Our able local partner is Fundación para el Ecodesarrollo y la Conservación (FUNDAECO), whose amphibian coordinator, Carlos Vasquez Almazán, Curator of Herpetology at the National University of San Carlos Museum of Natural History, drew international scientific attention to the conservation importance of Sierra Caral.

And its importance is indisputable. The 2012 list of threatened species at Sierra Caral included five Critically Endangered, four Endangered and two Vulnerable amphibian species. Sierra Caral is also important habitat for 120 Neotropical migratory bird species and it provides clean water and watershed protection, which benefits surrounding communities.

The conservation triumph at Sierra Caral was greatly magnified in 2014 when Guatemala formally established the encompassing 19,000-hectare Sierra Caral Water and Forest Reserve.

SAN ISIDRO, GUATEMALA

ICFC and others, including the Amphibian Survival Alliance (ASA), are now pursuing another important opportunity in Guatemala.

The Sierra de los Cuchumatanes in northwestern Guatemala is considered a "hotbed of unique and threatened amphibians" and is also an Important Bird Area, supporting populations of three globally threatened species. The remoteness of the region's humid subtropical forest has protected it to date, but there are increasing pressures to clear forests for coffee plantations. Once again, an expedition led by Carlos Vasquez was key in finding a prime piece of land, which happens to be in urgent need of protection (the present owners plan to clear it for coffee production within a year). The Vasquez team recorded 40 individuals (including egg-laying females) of the Critically Endangered Black-Eyed Leaf Frog (*Agalychnis moreletii*). In total seven threatened amphibian species and seven endemic amphibians have been recorded there. ICFC hopes to contribute US$75,000 toward the $690,000 acquisition cost for the 2,000-acre (809 ha) property.

DOING MORE FOR AMPHIBIAN CONSERVATION

At ICFC, we're always thinking about how we can increase our impact. The equation is simple: increase revenues and spend each dollar well. But how?

On the revenue side, we are not experts! But I would comment that the general public is much less aware of biodiversity loss than biologists and conservationists seem to realize. Nor does biodiversity loss receive much media attention these days. Climate change gets top coverage among green issues and even that is under-covered. We need to get the facts of biodiversity loss out there but always accompanied by the good news that conservation is a superb investment, that it's happening all over and that it can be easily ramped up with further support. As conservation advocates, we need to make an emotional as well as an intellectual connection with the general public.

Although their appeal is nowhere as broad as that of birds, amphibians are nonetheless a relatively "charismatic" and photogenic taxonomic group with fascinating life histories. We should perhaps move beyond the "canary in a coalmine" cliché to proclaim that this uniquely vulnerable group of organisms merits our attention

Sierra Caral waterfall. Photo: Robin Moore.

for a host of reasons: because they are marvelous creatures facing difficulties because of human actions; because they have an important ecological role as part of the food chain; and because protecting amphibians protects myriad other species sharing their habitats as well as the important ecosystem services that wetlands provide to humans, such as water storage and filtration and climate regulation.

As to effectiveness, we look for good local partners and particularly good opportunities in terms of the long-term conservation benefits in relation to cost and risk. We now have relationships with some great conservation organizations in the areas where we work and they (and our conservation colleagues) bring us their best ideas. We also value groups like the IUCN SSC Amphibian Specialist Group, the Amphibian Survival Alliance and the Alliance for Zero Extinction that provide good information on current conservation priorities and opportunities. And the human component is vital. We look for ways to involve local communities directly in conservation efforts, and we've found that public outreach can be amazingly effective in boosting awareness of conservation needs and spurring conservation action.

A few tough questions that we ponder when considering support for reserves (and welcome input on):

When is it too late to save a species or ecosystem? Is there really the potential for a viable population of a species to be maintained long term? How do we assess that? Will the ecosystem survive future expected climate change?

What size of reserve, in combination with similar connected habitats, is needed for long-term viability of an entire ecosystem (including sparsely distributed species)?

Are legal protections reasonably secure? Do our actions help or harm the interests of local communities? What are the best ways to address human threats? How can we best involve local communities? How can we get society to care about preserving nature?

Conservation is not an all-or-nothing, win-or-lose proposition. Extinctions will continue, but every durable conservation gain we make today will result in more biodiversity enduring for future generations. It all matters. And there is no longer-term impact than preventing extinctions.

For more information on ICFC and its projects please visit http://icfcanada.org/.

ICFC
International Conservation Fund *of* Canada

Globally considered Vulnerable by The IUCN Red List, the Oregon Spotted Frog (*Rana pretiosa*) is one of the most endangered amphibians in Canada. Photo: Oregon Spotted Frog Recovery Team.

Amphibian Conservation in "The Great White North:" Working Towards Evidence-Based Best Practices

By [1]Jessica Steiner

Canadian amphibian conservation stories rarely appear in *FrogLog*, and seldom make the international stage. There is some justification for this; most Canadian species are considered relatively secure from a global perspective and represent peripheral populations at the northern extent of their geographic range. Of the 46 amphibian species occurring in Canada, all but two are considered of Least Concern according to the IUCN Red List, the notable exceptions being the Oregon Spotted Frog (*Rana pretiosa*, Vulnerable) and Western Toad (*Anaxyrus boreas*, Near Threatened). However, in-line with global trends, amphibians are one of Canada's most threatened terrestrial vertebrate groups. At a national level, over half (54%) of our amphibian species are currently identified as At-Risk by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC).

Wildlife Preservation Canada (*1*) is a charitable non-government organization working to save endangered wildlife species from disappearing from Canada. We focus on species whose numbers are so critically low that direct human intervention (*e.g.*, conservation breeding, translocations, headstarting) is required for recovery. Our priority target list identifies those species in need of these intensive efforts, as recommended in recovery strategies and by species experts, and is updated annually. Unfortunately there is a growing need for these tools, and especially so with herpetofauna; about a third of those identified as At-Risk by COSEWIC have made our list. Amongst the species with which we currently work, amphibians and reptiles represent over 40%, with projects in multiple provinces. Among these projects is a conservation-breeding and headstarting program for the recovery of the Oregon Spotted Frog in the Fraser Valley of British Columbia, it's only Canadian location. Combined with habitat restoration, invasive American Bullfrog (*Lithobates catesbeiana*) control, long-term monitoring and community outreach, the program has seen some recent early success in re-establishing populations to historic sites.

Although exposed to the same worldwide suite of threats, conserving amphibian and reptile populations at their northernmost range limit, in a country as vast and ecologically diverse as Canada, presents unique challenges and opportunities (*2*). Peripheral populations, such as those found in Canada, may in fact play an important role in conserving declining species in terms of persistence and genetic diversity (*3*). In the context of climate change in particular, Canadian populations may be more successful at adapting given the large geographic distribution and breadth of seasonal variation currently tolerated by many species within our borders (*2*). Fungal *Batrachochytrium* spp. and *Ranavirus* pathogens are amongst the major threats that have been linked to global amphibian declines and both have been detected in Canada, even in remote northern regions of the country (*4, 5, 6*). However their history in Canada and risk to northern amphibian populations remains largely unknown.

[1]Conservation Programs Director, Wildlife Preservation Canada.

A conservation-breeding program, combined with headstarting of wild egg masses has seen some early success in re-establishing populations of *Rana pretiosa* to historic sites; all released frogs are marked with injectable elastomer as part of long-term monitoring and evaluation. Photo: Oregon Spotted Frog Recovery Team.

Eastern Tiger Salamander (*Ambystoma tigrinum*). There are two distinct populations in Canada, one (Carolinian) was assessed as Extirpated in 2012, the other (Prairie) is considered Endangered. IUCN Red List Status is Least Concern. Photo: Shutterstock.

Snake Fungal Disease is also an emerging threat in eastern Canada, with similar knowledge gaps. Information on the distribution of these pathogens, and their effect on host populations is important for understanding whether climate change will increase the threat of disease.

The predicted increases in average temperatures as a result of climate change could see potential northward shifts in species distribution (*7*). However, that potential will depend on the ability of species to disperse and shift their ranges as required and assuming habitat availability elsewhere. Given the loss and fragmentation of habitat, expanding road networks and general low dispersal ability of herptiles, it is likely that the persistence of many isolated populations will require or at least benefit from some degree of human intervention. In light of this, tools for direct species management, such as translocations and conservation-breeding, are increasingly being considered as practical climate-change adaptation strategies (*8*).

In most cases, the long-term conservation value of these strategies has not been scientifically proven, and their use remains controversial (*9*). With limited conservation dollars, there is a real need to evaluate these techniques in order to determine their effectiveness and ensure their appropriate use. Given the proportion of our work devoted to amphibians and reptiles, and the growing need for the types of intervention programs in which we specialize, Wildlife Preservation Canada is developing a national research program focused on rigorously evaluating the consequences of conservation interventions on species management. Working collaboratively with partners across Canada, this initiative will ensure Canadian content is added to global initiatives (*10*) to summarize science-based evidence on the effects of such tools, providing important national and regional context. We will be reaching out to those involved in applied conservation of herpetofauna in Canada to prioritize needs, and identify opportunities for partnership—if you are such a person, please contact us!

It is this recent undertaking which brought us to the Amphibian Survival Alliance, and we are proud to be a new Associate Partner. Our endeavors coincide with recent priorities identified by the IUCN Amphibian Specialist Group such as helping to refine the protocols for amphibian reintroductions and developing a database of amphibian conservation strategies. We are looking forward to making connections with the broader network in pursuing our goals, while contributing to similar global initiatives. Just as we can certainly learn from the international community, and the work we are undertaking will no doubt serve to inform best practices elsewhere.

Acknowledgements

We'd like to thank the Oregon Spotted Frog Recovery Team, including the Vancouver Aquarium and Greater Vancouver Zoo, and numerous volunteers, for their commitment to the preservation and recovery of the Oregon Spotted Frog in Canada.

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Wildlife Preservation Canada

RECOVERY ▪ CONSERVATION ▪ KNOWLEDGE

Photo: Vancouver Aquarium.

Vancouver Aquarium Breeds Tadpoles of Endangered Oregon Spotted Frog and Finds Juveniles Surviving in the Wild

By Dr. Dennis Thoney

As part of the global effort called Amphibian Ark (AArk), the Oregon Spotted Frog Recovery Team, which includes the Vancouver Aquarium Marine Science Centre, maintains an assurance population of this highly endangered species. Populations have rapidly declined in British Columbia and along the Western coast of North America. In Canada, there are only six small extant populations of Oregon spotted frogs.

This is the fourth year the Aquarium has successfully bred this species. Although the program has produced over 16,000 tadpoles for release since 2010, this year the number of tadpoles produced was lower than in previous years which is thought to be due in part to an unseasonably warm winter in the Lower Mainland.

For the first time since the program began, B.C.'s Oregon Spotted Frog Recovery Team recently found juvenile frogs in the wild, indicating that the tadpoles released in recent years are surviving and growing. The juveniles were found near Chilliwack, B.C., where they are released.

Two of the six established wild populations also were recently discovered in areas where the species was not previously known to exist. Egg masses were also found that are thought to have come from juvenile frogs raised by the Aquarium and released into the wild, a sign of the program's success in supplementing and conserving the wild population.

Finding these egg masses and juvenile frogs in the wild is an encouraging sign that our efforts are having a positive impact on this endangered species. These are important indicators of a healthier population, although the road to recovery is a long one.

Early last century, there were hundreds of thousands of Oregon spotted frogs, ranging from northern California up into British Columbia's Fraser Valley. Because of habitat destruction and the introduction of non-native species such as Eastern Canada's Bullfrog and the Reed Canary Grass, their numbers have declined over 90 per cent from past numbers in British Columbia. Historically, Oregon Spotted Frog populations had occurred throughout the Fraser Valley, from South Surrey to Hope. This species is more aquatic than closely related species in the area and require connected wetlands and floodplains for habitat.

In an effort to protect this endangered species, the Aquarium joined B.C.'s Oregon Spotted Frog Recovery Team in 2000. A diverse group of biologists and land managers in B.C. are coordinating efforts to conserve, manage and recover the Oregon Spotted Frog in Canada. Since 2007, Aquarium staff has been collecting Or-

Head of Animal Operations, Vancouver Aquarium Marine Science Centre.

egon Spotted Frog eggs to establish an aquarium-based assurance population. In 2010, the Aquarium became the first aquarium in the world to breed this species.

The Oregon Spotted Frog Recovery Program includes habitat management, monitoring, research, and restoration that is conducted in partnership with the B.C. Ministry of Environment; B.C. Ministry of Forests, Lands, and Natural Resource Operations; Canadian Wildlife Service; Department of National Defense; Seabird Island Band; Stó:lo Tribal Council; District of Kent; Fraser Valley Regional District; Greater Vancouver Zoo; Toronto Zoo; Vancouver Aquarium; Mountain View Conservation Centre; Grouse Mountain Refuge for Endangered Wildlife; Wildlife Preservation Trust Canada; Simon Fraser University; University of Ottawa, University of British Columbia; B.C. Conservation Foundation; and Fraser Valley Watersheds Coalition.


Photo: Vancouver Aquarium.


Photo: Vancouver Aquarium.


Photo: Vancouver Aquarium.

Jefferson Salamander. Photo: Conservation Halton.

City Closes Road to Protect Endangered Jefferson Salamander

By Sharon Will

King Road in Burlington, Ontario, Canada was closed from the base of the Niagara Escarpment to Mountain Brow Road from March 25 to April 15 to allow the Endangered Jefferson Salamander safe passage during its annual migration to lay eggs.

The Jefferson Salamander is a national and provincially protected Endangered species. The City of Burlington has closed the same section of road for a three-week period since 2012.

"The Jefferson Salamanders are a unique part of Burlington's biodiversity," said Mayor Rick Goldring. "Closing the road each year allows us to contribute towards the protection of this Endangered species."

"The annual road closure ensures the salamanders can cross safely from one side of the road to the other without the threat of traffic," said Bruce Zvaniga, the city's director of transportation services. "The closure is a significant conservation measure that assists in the protection and recovery of the Jefferson Salamander population in Canada."

In Canada, the Jefferson Salamander is found in Southern Ontario in select areas of deciduous forest, mostly along the Niagara Escarpment. Forested areas in Burlington provide the necessary breeding habitat required by this species.

Jefferson Salamanders spend the winter underground. As the weather warms up and the spring rains begin, the salamanders emerge and migrate to breed in temporary ponds formed by runoff, laying their eggs in clumps attached to underwater vegetation. By late summer, the larvae lose their gills and leave the pond to head into the surrounding forests.

"Conservation Halton is pleased the City of Burlington recognizes the importance of a full closure of King Road to help protect the Jefferson Salamander," said Ken Phillips, Conservation Halton chief administrative officer. "Our studies show the road closure has had a positive effect and is helping the salamanders safely cross the roadway on their way to the breeding ponds."

Adult salamanders migrate to their breeding ponds in mid-March or early April during wet rainy nights. They show strong affinity for their birth pond and can be very determined to reach it, often crossing busy roads.

To learn more about the Jefferson Salamander, visit the Ministry of Natural Resources website.

Winter Salamandering: The Eastern Newt

By Matt Ellerbeck

For individuals who like to observe, watch or record herptiles (reptile and amphibians) in the wild, the winter can often be a long and dismal season. Fortunately for me, my favorite group of herptiles are the salamanders and these amphibians can be found year round if you know where to look. In a previous piece, I wrote about my excursions to observe Mudpuppies (large neotenic salamanders). However, the Mudpuppy is not the only salamander species that can be seen in the winter. The Eastern Newt (*Notophthalmus viridescens*) is another type of ''winter salamander.''

Eastern newts are semi-aquatic salamanders belonging to the Salamandridae family. Most individuals who encounter this species do so when the newt is in its Red eft stage (terrestrial juvenile). At this point in its life, the newt is a vibrant orange color with red spots. When the newt matures after several years, its color changes to mostly olive green (dorsal surface), with bright yellow/orange bellies that are often mottled with spots. The tail also flattens and becomes more paddle-like.

In this semi-aquatic stage the newts spend less time on land and are primarily found in the water, either in permanent or semi-permanent bodies. Habitat includes well-vegetated ponds, marshes, swamps, canals and even ditches. Nature seldom holds absolutes, and the Eastern Newt is no exception. Therefore, under certain circumstances adult newts may become more land bound. Such factors that contribute to newts becoming more terrestrial include decreasing water levels and higher temperatures. Extended land stays can result in tail fins decreasing and more granular skin.

Unlike the Red eft stage, adult newts are not frequently seen due to their more aquatic life styles. Especially since the newts blend in well to aquatic vegetation. However, under special circumstances a myriad of these salamanders can be viewed at once!

It has been well documented that in ponds that do not freeze to the bottom, Eastern Newts can often be seen active under the layer of ice. Pitkin and Tilley (*1*) observed an unusual aggregate of adult newts in a Massachusetts pond in the winter. Large numbers of adults had gathered together in ice-free areas of the pond. The exact functions of these aggregates are unknown (*1*).

In the beginning of February 2015, I was very lucky to observe one of these newt gatherings in person. I travelled nearly two hours from my home to reach a pond to see the newts. A small ice-free section, only a few meters across, allowed for viewing. Between 60 and 100 newts were seen here! Some could be seen resting on the bottom, while others would periodically swim for short bursts. Several were temporarily placed in a container to take a photo of them with ease, after which they were returned to the pond.

What a wonderful experience to see so many salamanders in the dead of winter! However, despite the numerous observations of these newts, we should not become complacent in regards to their conservation. Many newts are killed every Spring on roads when they migrate to breeding sites. If someone sees a newt on the road and it is safe to do so, please stop (wet the hands if possible), scoop up the newt and gently take it across, in the direction it was headed. To avoid running over salamanders, if possible do not drive during dawn and dusk hours on wet spring nights, or during or right after rain storms. Taking alternate routes that do not cut through forests or wetlands can also reduce the risk of hitting salamanders. If you must drive, be sure to travel slowly and keep an eye out for salamanders. Be alert to listen for Spring Peeper Frogs and Wood Frogs. The calls of Spring Peepers and Wood Frogs means that there are amphibian breeding pools in the area. In these areas be extra watchful for salamanders.

Several adult Eastern Newts from the pond aggregate. Photo: Matt Ellerbeck.

As Eastern Newts live primarily in aquatic habitats, they need pristine water to survive. Degradation to their habitat often occurs via siltation, pollutants and even agricultural runoff. Landowners can help by allowing buffers of tall grass, trees, saplings, shrubs, ferns and other natural plants to grow around the edge of ponds and wetlands. Such buffers help protect against erosion and various forms of runoff. Land owners are also encouraged to take measures to prevent soil erosion/siltation. The addition of silt and clay into aquatic areas can severely degrade the salamander's habitat. Silt and clay also fill in important depressions under rocks and sunken debris which are used by the salamanders as cover for themselves, their eggs and are used as areas to find prey. Use silt fences or sediment traps when doing construction or landscaping to stop sediment from reaching the water. Planting cover crops, native plants or shrubs can also prevent erosion. Dense crop stands physically slow down the velocity of rainfall before it contacts the soil surface, preventing soil splashing and erosive surface runoff.

By taking these simple actions, individuals can help ensure that our Eastern Newts stay numerous!

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Yosemite toad. Photo: Lucas Wilkinson, U.S. Forest Service.

Amphibians Get Life-Saving Protections under the Endangered Species Act

By Collette Adkins

Almost four years have passed since the Center for Biological Diversity struck a historic legal settlement with the U.S. Fish and Wildlife Service, requiring the agency to make initial or final decisions on whether to add hundreds of imperiled plants and animals to the endangered species list by 2017. The settlement caps a decade-long effort by the Center's scientists, attorneys and activists to secure federal help for some of the country's least protected, but most imperiled, species. Under that settlement, dozens of amphibians across the United States are now on track to receive life-saving protections under the Endangered Species Act.

The purpose of the Endangered Species Act is to prevent the extinction of the most at-risk plants and animals, increase their numbers, spur their full recovery and, eventually, their removal from the endangered list. Two elements give the Act its "teeth:" the citizen-suit provision allows public-interest groups and individuals to petition and sue the government to make sure the Act protects species as it was intended to, while the "critical habitat" provision provides a crucial tool for protecting the lands and waters that species need to survive and recover.

The Center's landmark agreement has already yielded important results by securing proposed and finalized listing and critical habitat protections for numerous amphibians in the United States. Here are the amphibians that received protections in 2014:

- **Oregon Spotted Frog** listed as Threatened with proposed critical habitat covering more than 68,000 acres and 24 stream miles. Once abundant from British Columbia to California, spotted frogs have disappeared from 90 percent of their former range, mostly because their wetland habitats are being destroyed.
- **Sierra Nevada Yellow-Legged Frog** and **Mountain Yellow-Legged Frog** (Northern population) listed as Endangered with 1,105,400 acres of critical habitat proposed for the Sierra Nevada Yellow-Legged Frog and 221,498 acres for the northern population of the Mountain Yellow-Legged Frog. Yellow-Legged Frogs throughout the Sierra Nevada have suffered dramatic declines in range and numbers due to habitat destruction and degradation, disease, predation by nonnative trout, pesticides and climate change.
- **Yosemite Toad** listed as Threatened with 750,926 acres proposed as critical habitat. Yosemite Toads have disappeared from many areas and suffered population losses, including in Yosemite National Park, where these toads were first discovered and given their name. Yosemite Toads are threatened primarily by livestock grazing, climate change and pesticides.

Senior Amphibian and Reptile Attorney, Center for Biological Diversity.

- **Georgetown Salamander** and **Salado Salamander** listed as Threatened with 1,031 acres proposed as critical habitat for the Georgetown Salamander. These fully aquatic animals require clean, well-oxygenated water and are threatened by activities that disturb their surface springs, pollute their water or reduce its flow to their underground aquatic habitats. The Salado Salamander is extremely rare and has been observed just a few times over the past several decades, despite intensive survey efforts. The Service determined in 2001 that the Georgetown Salamander deserves federal protection; the Georgetown Salamander waited more than a decade for the Service to finalize the listing.

All in all, in the past four years, more than 140 species have been added to the endangered species list, but hundreds are still waiting for protection. And recovery dollars are now spread ever thinner on a per-species basis. Yet the Obama administration's proposed budget for 2016 includes only slight increases in overall funding for the U.S. Fish and Wildlife Service, the agency whose mission it is to stave off the wildlife extinction crisis in the United States. When adjusted for inflation, the proposed budget is essentially flat, and actually provides fewer dollars on a per-species basis than was budgeted at the start of the Obama administration. The agency's proposed budget for endangered species is less than the government spends on a single F-35c fighter jet.

Approximately $23 million was requested to list and protect highly imperiled species under the Endangered Species Act—a small increase from last year, but roughly the same amount of funding received in 2010, even though hundreds of species are waiting for decisions about their protection.

Recovery efforts are funded separately from listing, but as with listing, the Administration has requested far less than is needed. Indeed, most of the more than 1,500 species protected under the Endangered Species Act receive far from adequate funding for recovery. The most recent expenditure report produced by the Fish and Wildlife Service covering 2013 shows that nearly 60 percent received less than $50,000 and about 21 percent received less than $10,000—including 48 species that received $1,000 or less. Yet the Obama administration is only asking for a marginal increase. Progress toward recovery is directly correlated with the availability of federal resources and dollars for recovery, and perpetually inadequate funding is limiting species recovery.

The Endangered Coqui Llanero, for example, received zero funding in 2013. This tiny, gorgeous frog was listed in 2012 and faces extinction because its entire population lives in just one wetland system, under dire threat from urban development and water contamination from nearby landfills.

Scientists estimate that approximately one-third of amphibians in the U.S. are at risk of extinction. Surprisingly, though, just 32 of the approximately 1,500 species protected under the U.S. Endangered Species Act are amphibians. Because the Endangered Species Act is America's strongest environmental law and surest way to save species threatened with extinction, the Center works to gain these needed protections through scientific petitions, advocacy and lawsuits. The Center is also working to ensure that Congress designates the funding for endangered species recovery that the Fish and Wildlife Service desperately needs to accomplish its goals.

More information on the Center's campaign to address the amphibian extinction crisis is available here: BiologicalDiversity.org/herp.

Texas Blind Salamander. Photo: USFWS.

Coqui Llanero. Photo: USFWS.

Oregon Spotted Frog, *Rana pretiosa*. Photo: Kelly McAllister, Washington Department of Fish and Wildlife.

Fig. 1: The Eastern Hellbender (*Cryptobranchus alleganiensis*). Photo: Kevin

Environmental DNA and Amphibian Monitoring: Insights from the Eastern Hellbender

By [1]Stephen Spear

In 2008, a study was published that demonstrated the ability to extract American Bullfrog (*Lithobates catesbeianus*) DNA directly from lake water (*1*). This intriguing result has led to an explosion of research using this approach, which is referred to as environmental DNA or eDNA. The interest in eDNA is demonstrated by the publication of a special issue on the topic in the March 2015 issue of *Biological Conservation*. This issue highlights the diversity of eDNA work with aquatic organisms such as amphibians and fish. This issue also included a paper that I, along with colleagues John Groves (North Carolina Zoo), Lori Williams (North Carolina Wildlife Resources Commission) and Dr. Lisette Waits (University of Idaho), authored on the applications of eDNA to North America's largest salamander, the hellbender (*Cryptobranchus alleganiensis*) (Fig. 1) (*2*). Our work with hellbenders has revealed the utility of eDNA for amphibian conservation programs, while at the same time highlighting future challenges for the role of eDNA in the monitoring of imperiled species.

I began to develop a hellbender conservation program for The Orianne Society at the same time as many of the first eDNA papers, inspired by the 2008 bullfrog study, began to be published. In particular, I was very intrigued by the eDNA work that Dr. Caren Goldberg and colleagues were doing with Rocky Mountain Tailed Frogs (*Ascaphus montanus*) and Idaho Giant Salamanders (*Dicamptodon aterrimus*). They found that eDNA reliably detected both these species in rivers and streams of the Idaho mountains (*3*). Perhaps the same technique could help us monitor hellbender populations.

The reason I was interested in hellbenders in the first place is because they appear to be one of the most rapidly declining salamanders in North America. There are two subspecies of hellbenders. In Missouri and Arkansas, there is the Ozark Hellbender (*Cryptobranchus alleganiensis bishopi*), which is already listed as Endangered under the U.S. Endangered Species Act (ESA) and reduced to a handful of known localities. The subspecies I am working with is the Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*), and it is currently being evaluated as a possible candidate for the ESA list. Eastern Hellbenders historically had a very wide range in the United States, from New York to Georgia and as far west as Missouri. Yet currently there are relatively few rivers where even this subspecies can easily be found.

The overarching cause behind this decline seems to be sedimentation due to erosion from activities such as agriculture, development and mining, as well as a reduction in water quality due to pollution and fragmentation of river systems by dams. Hellbenders require spaces under rocks that fill in with sedimentation, and they receive 90 percent of their oxygen through their skin underwater, making them especially susceptible to pollution (Fig. 2). In addition, most populations of any species are going to be negatively affected by barriers, resulting in both habitat loss and the disruption of gene

[1]The Orianne Society.

Fig. 2: Eastern Hellbender. Hellbenders are threatened by siltation that fills in spaces under rocks as well as pollution that reduces oxygen available to be taken in through its skin. Photo: Pete Oxford.

flow. The good news is that states have recognized these problems. Hellbenders currently receive some level of protection in each state where the species exist, and each state has some current level of ongoing hellbender conservation programs and actions.

The use of eDNA is appealing for hellbender conservation efforts for multiple reasons. The most obvious is that searching for hellbenders is hard work! Hellbenders spend the majority of their time under large rocks in cold streams. Therefore, researchers, usually donning wetsuits, must snorkel or scuba dive to the bottom of streams. Furthermore, because hellbenders tend to prefer larger rocks, a single individual needs assistance in lifting the rock, often with log peaveys and multiple individuals. Once a rock is lifted, the snorkeler swims underneath, trusts the strength of the individual(s) lifting the rock to not crush them, and looks for a hellbender (Fig. 3). This survey method can be quite effective but is time consuming, requires a team of workers and despite the best efforts of surveyors, disrupts hellbender microhabitats. This level of effort is necessary if we are trying to get demographic information or count individuals, but many state agencies don't even know where hellbenders exist in their state anymore, and so simple presence/absence surveys is often a goal. If eDNA works for detecting hellbenders, we could classify presence/absence through a visit by a single individual filtering a liter of water (Fig. 4).

In the summer of 2011, I collected water samples at known and historic sites in eastern Tennessee working with Dr. Michael Freake at Lee University. These first samples clearly demonstrated that eDNA could detect hellbenders, even at low density (at least based on snorkel survey data). The success of these early trials has led to an integration of eDNA into inventory work that was already being conducted by partners in Tennessee, North Carolina and Georgia. To date we have collected hundreds of samples, confirming presence at known sites, determining which historic sites still contain hellbenders, and even adding new occurrences at sites that had either been unsampled or where previous surveys had not found them. On the negative side, we have had very few positives in some regions, such as central Tennessee, that support the observations of severe declines. Although we have not rigorously estimated detectability, we have found that a single sample is almost always sufficient to find hellbender DNA with only a small hand-

Fig. 3: Technique for capturing hellbenders by snorkel survey. Large rocks are lifted up with log peaveys, while other surveyors search underneath for hellbenders. Photo: North Carolina Wildlife Resources Commission.

ful of exceptions. Because of this, I would argue that for simple presence/absence surveys, eDNA may be the most useful tool for hellbender monitoring.

There is an important caveat to the success of eDNA in our hellbender work. The truth is, basing conclusions about hellbender status simply on presence/absence can give us a very biased view. Hellbenders can live a very long time (at least 30 years) and so a few older individuals could potentially persist in a stream for a long time, even if they are not successfully reproducing. Unfortunately, the above scenario seems to be common, and simple presence/absence can't distinguish between a viable population and a geriatric one.

There is hope for eDNA to help in this regard as well, however. We use a type of genetic analysis known as quantitative PCR (qPCR), which allows us to not only see if the species' DNA is there, but also to quantify the amount of eDNA in the sample. Some studies (*4*,*5*) have demonstrated that in some situations, this estimated eDNA concentration could be used to predict the number of individuals present. Furthermore, for species with a discrete breeding season (such as hellbenders), we may expect to see an increase in eDNA during the breeding season if reproduction is taking place.

We evaluated these two possibilities in our recent *Biological Conservation* article using some of our North Carolina work as a case study (*2*). First, we collected eDNA samples at 23 sites where snorkel surveys were occurring at the same time and tested the correlation of eDNA concentration with survey numbers. Second, we sampled nine sites across three river systems at six time points throughout the active season to see how the levels of eDNA changed through the season. We found there was no correlation between eDNA amount and survey numbers. Part of the issue was that eDNA detected hellbenders at 90 percent of the sites, whereas snorkel surveys only observed hellbenders 40 percent of the time! This demonstrates the efficacy of eDNA, but also provides a pretty strong indication that snorkel surveys are less effective for determining actual numbers or density of hellbenders, and thus it makes it really hard to test if eDNA predicts hellbender abundance in a field setting.

Fig. 4: Filtering water for eDNA. Water is collected and poured into a cup with a filter on the bottom, and then pushed through the filter using vacuum filtration. Photo: Pete Oxford.

Fig. 5: Folding filter after filtration to store in tube with ethanol for later extraction and qPCR analysis. Photo: Pete Oxford.

However, we did see a big difference when looking at changes in eDNA amount across time. Specifically, we saw a large increase during September, which coincides with the breeding season of hellbenders. This was an exciting development for the prospect of using eDNA to evaluate reproductive success, but it is only a first step. We actually saw this same pattern in a tank with a captive male hellbender where clearly no reproduction took place. We need to learn more to see if there are patterns associated with sites that do successfully reproduce compared to sites that do not. We are currently considering such research questions.

In conclusion, our work so far with hellbender eDNA has helped fill gaps in our knowledge of where the species still occurs while providing glimpses into how we might assess population status. Certainly eDNA does not replace traditional survey techniques (nor is it meant to), but it provides a powerful complementary tool. While I have focused on hellbenders for this article, there are countless aquatic eDNA projects emerging, and it may not be very long until eDNA is part of standard aquatic biology survey methods. I am excited to both follow and contribute to the further development of this field as we unlock its full potential for amphibian conservation.

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Ambystoma maculatum eggs found at a greenway in Forsyth County, GA,, USA. The eggs grow symbiotically with green algae. Photo: Seth McWhorter.

Adopt-A-Stream: Citizens Undertake Amphibian Conservation Efforts in the Southeastern United States

By Seth McWhorter & Norman Leonard

During the 1990s, citizen science in Georgia began in earnest when Georgia Adopt-A-Stream (AAS) started monitoring water quality. The plan was simple: raise awareness about the importance of stream ecosystems while simultaneously gathering data on streams and organisms within them. To help monitor the condition of streams, professionals encouraged trained citizens to help count macroinvertebrates, test for bacteria, and run several chemical assays. Twenty-five years later the program has grown to include more than 500 sites in Georgia and, recently, has begun to extend into surrounding states.

While all aspects of Adopt-a-Stream deliver insight into stream health, amphibian monitoring, which was added to the AAS arsenal in 2005, provides a plethora of data about the abundance and biodiversity of salamanders and frogs at AAS monitoring sites. The collection of these data provides deeper insight into local environmental conditions, in addition to keeping a finger on the pulse of taxa in the midst of worldwide decline. The Southeastern United States includes some of the most diverse amphibian assemblages. The Appalachian Mountains alone house 55 species of salamanders, 21 of which occur only in the mountain chain. Unfortunately, amphibians respond drastically to pollution and disappear quickly from polluted streams. Thus, the work of the public to maintain and monitor amphibians' stream habitats stabilizes populations of salamanders and frogs in the Southeastern United States.

A group of students at Clemson University in South Carolina started an Adopt-a-Stream amphibian monitoring site. The stream they chose runs through a nature preserve on campus. Cathleen Reas Foster, a Natural Resources agent for Clemson University who helped kick start the AAS site, contacted Dr. Norman Leonard of the University of North Georgia and Kevin Smith of Keep Forsyth County Beautiful, asking for advice for the students working under her. To help raise awareness and knowledge, Adopt-a-Stream holds amphibian workshops in counties that run the AAS program, so the three decided to hold an amphibian workshop on Clemson University's campus. On March 27, 2015, Dr. Leonard, five of his salamander research students, and Kevin Smith traveled to Clem-

Students from Clemson University check the PVC pipes at the amphibian monitoring site. Photo: Cathleen Foster.

The beaver pond where the University of North Georgia group caught amphibians in Lake Zwerner Dahlonega, GA. Photo: Dr. Norman Leonard.

son University to hold the amphibian workshop. Before heading to Clemson, the research group searched for animals near their home campus to use during the presentation. Together with his research students, Dr. Leonard collected 11 species, including examples of frogs, toads and newts. Dr. Leonard assigned each student a species to research to help teach the amphibian workshop.

The workshop began with Smith presenting details about the mission of Georgia Adopt-a-Stream and efforts in Forsyth County to incorporate amphibian monitoring in its AAS program. Smith is an Adopt-a-Stream trainer in Forsyth County, Georgia, United States. After the overview of AAS, Dr. Leonard spoke about the importance of amphibians in stream ecosystems, emphasizing the animals' physiology as well as their roles as top predators. He noted that amphibians may be indicators of water quality thanks to their permeable skin and also that stream-dwelling salamanders provide information about the invertebrate community, the primary prey items of their larvae. So if an area experiences a reduction in salamander or frog abundance, it may be a sign of poor water quality or other habitat alteration. After talking about amphibians as a group, Dr. Leonard detailed identification information for common species found in Clemson, South Carolina so that workshop participants could include specific species information on the amphibians encountered at monitoring sites.

Once the presentation ended, the attendees broke up into groups and visited stations manned by the students from the University of North Georgia. Each group stayed at a station for five minutes to learn about various species of amphibians and even a few snakes, which were included simply because they're interesting. At this

An adult Seal Salamander (*Desmognathus monticola*) captured and released by Dr. Norman Leonard. Photo: Dr. Norman Leonard.

Dr. Norman Leonard holds a Red Salamander (*Pseudotriton ruber*). Photo: Dr. Norman Leonard.

Two Southern Cricket Frogs (*Acris gryllus*) caught between feet of each other exemplify the phenotypic plasticity present within amphibian species. Photo: Seth McWhorter.

A Red-Bellied Snake (*Storeria occipitomaculata*) from the Clemson Outdoor Lab. Photo: Dr. Norman Leonard.

point, the attendees handled the specimens and learned how to hold each animal without harming it.

After the groups visited every station, Barbara Foster, the reptile curator and FrogWatch regional coordinator at the Greenville, South Carolina Zoo, explained how AAS amphibian monitoring can fit into the FrogWatch framework. FrogWatch is a nationwide citizen science program that allows citizens to study wetlands in their area and report information on frog calls and captures. At the end of her presentation, she played several frog calls to help those attending distinguish between different calls and report the data.

Though the classroom experience provided knowledge on the capture and study of amphibians, nothing raises awareness and knowledge like dirtying your hands and capturing a few herps. So, once the classroom portion ended, a field expedition headed to the stream and wetlands being used for the Clemson University's Adopt-a-Stream site. The group first searched a section of the stream that had PVC pipes and cover boards used to passively trap frogs and salamanders, following Adopt-a-Stream protocol. Further up the trail, several beaver ponds provided an excellent area to capture and release reptiles and amphibians. A recent cold front decreased the frog activity at the site, but still the group found several frogs and salamanders in and around the ponds. The gentle handling and release of the frogs and salamanders in their native habitats helped establish a connection between the classroom material and the actual ecosystems. The group also captured several small snakes, including a Red-Bellied Snake (*Storeria occipitomaculata*). Once the sun went down, the workshop officially ended, but many participants stayed to keep searching for more specimens. The devotion of many of the young volunteers foretells a bright future in the field of amphibian conservation.

While the workshop targeted general citizen scientists, the attendees mainly came from academia in the form of students and professors. The group from Georgia accommodated for attendees present, however, and helped raise the knowledge of those looking to be leaders in the field of conservation biology. The more information the professionals of the field have in hand, the more knowledge that can be passed on to citizens seeking to cure the ecosystems of which they are a part. Without citizen scientists, the progress made toward conserving the world's various ecosystems would significantly slow down. Citizen science allows professionals to allocate the field work load and analyze data more efficiently. Through AAS and other programs, the conservation movement can spread like fire throughout the world in professionals and citizens alike.

The growth of Adopt-a-Stream coincides with an increased effort in conservation. With continued efforts, the program will increase the health of streams and amphibians throughout the southeastern United States. As long as we work together with a passion and love for the natural world, Adopt-a-Stream and similar programs around the world will raise the awareness of the impact of humans on the ecosystems we live in and around, and hopefully save many of our amphibian friends from extinction.

Lindsey True displays an American Toad (*Anaxyrus americanus*) for the class. Photo: Cathleen Foster.

Annie Rae Carlton teaches workshop participants about Queen Snakes (*Regina septemvittata*). Photo: Cathleen Foster.

Competitive Eaters: The Influence of Density on Cannibalism among Wood Frog Tadpoles

By Dale M. Jefferson

Wood Frogs (*Lithobates sylvaticus*) are explosive breeders that use highly variable temporary wetlands as natal habitats for their developing offspring. Where environmental conditions facilitate pond drying, tadpole densities can rapidly increase, intensifying competition. Deteriorating conditions can also lead to reductions in quality and/or quantity of dietary resources (*1*). Wood Frog tadpoles are opportunistic predators of larval amphibians including conspecifics (*2*). Cannibalism is a significant important interaction among tadpoles, and has even been suggested as the evolutionary pressure that led to the development of synchronous breeding in wood frogs (*2*).

Cannibalism is both an intraspecific predator-prey interaction and a form of interference competition, "limiting population size before the resource itself becomes limiting" (*3*). Conspecific tissues represent a potentially ideal diet satisfying all nutritional requirements for growth, development and reproduction because all nutrients should be available in the appropriate proportions (*4*). Cannibalism may sustain individuals to metamorphosis under conditions of dietary stress (*5*). The purpose of this research was to characterize the proximal mechanisms that commonly influence the expression of cannibalistic behaviour among wood frog tadpoles.

CONDITIONAL BENEFITS OF CANNIBALISM

Initial feeding experiments (*6*) indicated that growth and development was significantly lower in tadpoles fed conspecific tissues (*Fig. 1*) relative to a high quality diet of similar protein content (brine shrimp). However, the diet of conspecific tissues did offer conditional benefits; tadpoles fed this diet exhibited significantly faster development relative to those fed a low quality, low protein diet (corn meal) but exhibit no improved growth relative to those fed the low quality diet. Where tadpoles were housed individually they preferentially avoided consuming conspecific tissues, and subsisted on their lipid stores until the risk of starvation instigated the initiation of feeding. Tadpoles raised with competitors initiated feeding upon conspecific tissues immediately. Short-term exposure to a low quality diet does not illicit a significant hormonal stress response suggesting that poor diet alone is unlikely to influence cannibalistic behavior. Additionally, we found that tadpoles fed conspecific tissues exhibited the greatest variation in size among individuals relative to all other feeding treatments. This suggests that tadpoles consuming conspecific tissues exhibited variation in feeding behavior, with some individuals potentially adapting a bolder personality and/or aggressive behavior providing them a competitive advantage over individuals with a shy personality and/or cautious behavior allowing them to grow larger.

RESPONSE TO CONSPECIFIC CUES

A subsequent experiment (*7*) on individually housed tadpoles found that when chemical alarm cues released from injured conspecifics were added to their diet (a mix of cornmeal and brine shrimp) they failed to exhibit the expected anti-predator aversion behaviour. Rather than avoiding the food that had been soaked in alarm cues, and thus being smaller and slower to develop than the control group (fed the same diet soaked in water) they were of similar mass, developed at the same rate, but had a larger body length and body width. This body morphology did not conform to the characteristics associated with a defensive morphology, but appeared related to an elongated intestine (*8*), which is a competitive response previously described in wood frog tadpoles. Tadpole intestines are coiled resulting in the longer and wider body, and improves digestion and assimilation of consumed foods providing a competitive advantage. This response may have resulted from tadpoles perceiving chemical cues from injured conspecifics as a sign of increasing competition. Chemical alarm cues rapidly degrade in water, which may cause a short-term avoidance response in tadpoles exposed to them. Conversely, it is believed that tadpoles also release chemical cues indicative of competition and that these are released in tadpole faeces and nitrogenous waste. Thus, as alarm cues degraded competitive cues would increase as the chemical cues added to the food would contain them and further excretion from the subject would increase the concentration over time. This means that tadpoles were potentially able interpret potential risks and adjust their responses to efficiently adapt to the overall ecological risks.

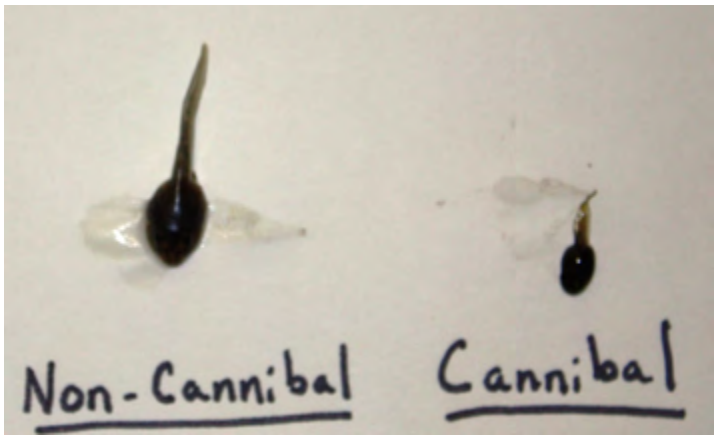

Fig. 1: Differences in size of tadpoles fed brine shrimp (non-cannibal) and those fed conspecific tissues (cannibal). Photo: Dale Jefferson.

FEEDING INITIATION

Finally, the feeding response of tadpoles was tested for various diets in the presence or absence of competition, and at three different time periods (*9*). Tadpoles exhibited adaptive response to diets over time. However, the presence of a competitor reduced response times to all diets including conspecific tissues. Similarly, the presence of dietary cues (regardless of dietary quality) reduced tadpole feeding initiation times on conspecific tissues (*Fig. 2*). These results suggest competition among feeding tadpoles could result in aggressive behavior leading to indiscriminate predation and cannibalism.

SUMMARY

The results of this research suggest tadpoles are not necessarily becoming cannibalistic in order to exploit a profitable food source, but rather in response to competition. It appears that cannibalism is the result of escalating aggression in response to the crowded

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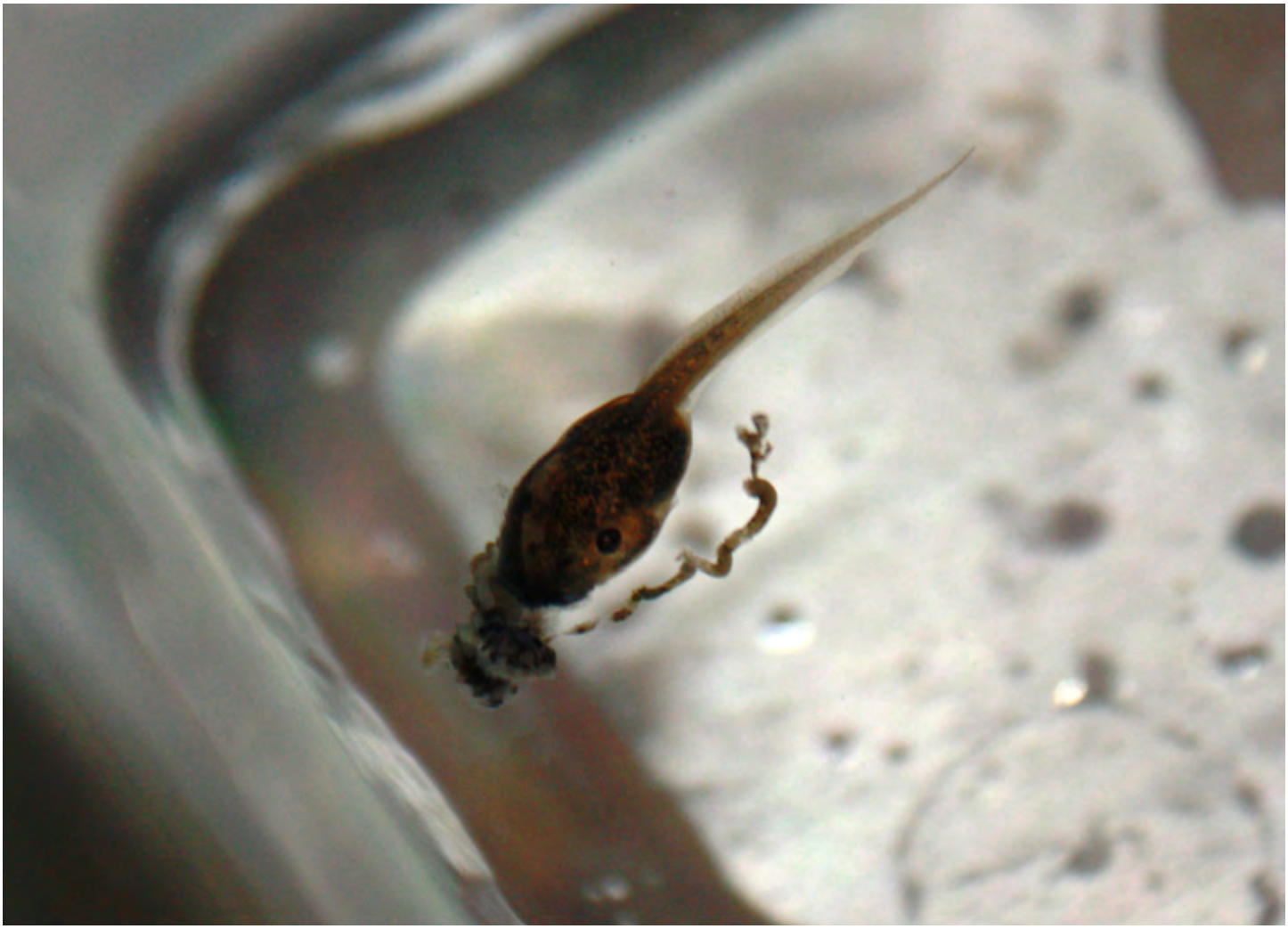

Fig. 2: Wood frog tadpole eating conspecific tissues. Photo:Dale Jefferson.

conditions that can arise within the temporary wetlands in which these tadpoles develop. Where wetlands may quickly evaporate and tadpole densities rapidly increase it may be necessary for individuals to adapt more aggressive competitive behaviors, which could lead to cannibalism.

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Fig. 3: Tadpoles forming feeding aggregations while foraging. Photo: Dale Jefferson.

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Aerial Dispersal of Amphibian Chytrid Fungus: Detection in Rainwater from Honduras

By Jonathan E. Kolby

Global spread of the amphibian chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) is considered to be a driving force in many recently observed amphibian declines and extinctions (*1*, *2*). For this reason, understanding the variety of pathways of *Bd* dispersal is necessary to accurately predict emerging patterns of spread and direct conservation resources towards species and regions likely to become most affected. *Bd* can be spread by the physical movement of infected amphibians, either naturally (*3*), or by the international trade in live amphibians (*4*–*6*). In the absence of amphibian hosts, this pathogen can be transported in contaminated water (*7*), by crayfish (*8*), or possibly on the feet of waterfowl (*9*). Still, records of *Bd* presence in extremely remote locations far removed from amphibian trade and instances of *Bd* in strictly terrestrial amphibians suggests that additional uncharacterized pathways of *Bd* dispersal may exist.

From 2010–2013, I investigated the potential influence of weather events on the spread of *Bd* at research sites in Cusuco National Park, Honduras (CNP). *Bd* is widespread in this cloud forest (*10*) and infected tadpoles are abundant in most rivers sampled. The landscape is steep, mountainous and adorned with towering waterfalls where clouds of aerosolized river water particles are produced by the crashing forces of water upon impact with the plunge pool and rocks below. Since river water contains *Bd* shed from infected tadpoles, it seemed plausible that *Bd* could sometimes become incorporated into this river water mist, which is subject to crosswinds winds before falling from suspension. Since no sampling methods currently exist to detect the presence of *Bd* suspended in air moisture alone, I instead collected rainwater and employed established protocols for water filtration to investigate this possible phenomenon.

Bd was present in rainwater collected from one of 20 storm events sampled (*11*). The density of *Bd* detected was low, and we cannot ascertain its viability from molecular presence alone, both of which apply to most instances of environmental *Bd* detection. This rainwater sample was captured at a hillside previously cleared of trees by a landslide, providing an area where we could collect rainwater that did not drip directly from overhead vegetation. Nonetheless, it is likewise possible that this *Bd* did not originate in a river as I surmised, but instead may have been dislodged by rain splash from the skin of an infected frog perched elsewhere within the forest, and then carried some distance by the wind before precipitating at this collection site.

The spread of *Bd* in rainwater is currently unpredictable based on our results, but may affect patterns of observed *Bd* distribution. Although we detected low *Bd* density in rainwater, it was enough to cause *Bd*-positive results, as the commonly used qPCR diagnostic technique is highly sensitive and can detect material from as little as one *Bd* zoospore in a sample, regardless of its condition (*12*). Therefore, while it is unknown whether *Bd* is commonly viable following aerial spread, even contact with dead *Bd* zoospores can produce *Bd*-positive skin swab results in the absence of infection or disease.

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At the first crack of thunder, Jonathan Kolby and Sara Ramirez hop into action to collect rainwater samples for *Batrachochytrium dendrobatidis* (*Bd*) presence in Cusuco National Park, Honduras. Photo: Jonathan Kolby.

Further, if occasionally viable in rainwater, similarly low exposure dosages can cause infection in susceptible amphibians. For example, exposure to an estimated single *Bd* zoospore (isolate JEL#275, from Colorado, USA) caused infection in 38% of boreal toadlets (*Bufo boreas*) when the exposure duration lasted 24 hrs, and in 100% of toadlets when exposure to one zoospore persisted for three days (*13*). Although many of these toadlets did succumb to chytridomycosis following the treatments, I only wish to emphasize that the low *Bd* density observed in rainwater does not alone suggest the inability to cause infection and/or *Bd*-positive skin swab results.

When *Bd* presence is detected in unexpected species and locations, aerial *Bd* transport provides one potential explanation. Recently, *Bd* was detected in wild amphibians in Madagascar (*14*), which is especially curious given the absence of commercial amphibian importation, often regarded to be the most common and obvious mechanism of international *Bd* spread. While trade does appear to be a significant current pathway of dispersal, transoceanic *Bd* spread did also occur prior to the emergence of significant amphibian trade during the mid-20th century. Although the global origin of *Bd* remains unknown, recent analyses of specimens in museum collections have now revealed *Bd* presence in the USA in 1888 (*15*), in Brazil in 1894 (*16*), North Korea in 1911 (*17*) and Cameroon in 1933 (*18*). Thus, the absence of intentional amphibian importa-

One of many cascading waterfalls present in Cusuco National Park, Honduras. Photo: Jonathan Kolby.

tion into a region such as Madagascar should not be considered isolation from *Bd* exposure, and introduction via aerial dispersal is possible, especially given the island's proximity to *Bd*-positive regions in nearby continental Africa. Likewise, a growing number of terrestrial amphibian species have tested positive for *Bd*, contrary to early beliefs that *Bd* is an aquatic pathogen of significance only to amphibians associated with permanent water bodies. Not only have terrestrial direct-developing species with limited home ranges somehow become exposed to *Bd*, but some also appear to have suffered dramatic *Bd*-associated population decline, most notably Archey's Frog in New Zealand, *Leiopelma archeyi*, (*19*). Aerial spread of *Bd*, whether by birds, flying insects or meteorological events could help explain some of these unusual and unexpected observations.

Identification of the spectrum of local and global *Bd* dispersal pathways is needed to improve our understanding of this pandemic and help guide the development of potential management interventions to control future spread. Although global amphibian declines appear to be a relatively a recent phenomenon, the "virulent" BdGPL clade has existed for approximately 1,000 + years (*20*) and thus its presence alone does not simply explain the recent emergence of disease. The international trade in live amphibians is now spreading *Bd* at alarming rates and warrants immediate biosecurity intervention, but *Bd* may still spread internationally in the absence of intentional amphibian trade. Understanding the differences between the past and present frequent avenues of dispersal, rates of global *Bd* spread, patterns of distribution and outcomes of exposure may each provide deeper insight into the catalyst(s) for this contemporary amphibian global disease event.

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Tadpoles of *Duellmanohyla soralia* at the base of this waterfall tested positive for *Bd*. Photo: Jonathan Kolby.

Duellmanohyla soralia. Photo: Jonathan Kolby.

Recent Progress for the Honduras Amphibian Rescue and Conservation Center (HARCC)

By [1*,2]Jonathan E. Kolby & [3]Brandon L. Greaves

Fundraising efforts continue to pave the way towards opening day for the Honduras Amphibian Rescue and Conservation Center (HARCC), based at Lancetilla Botanical Garden and Research Institute in Tela, Honduras. Meanwhile, the biosecure HARCC location at which amphibians will be housed has now entered construction. With assistance from the Omaha Zoo Foundation to locate additional project support, a generous donation has now been granted by Werner Enterprises, Inc., an American freight carrier and transportation and logistics company headquartered in Omaha, Nebraska. Werner has donated two 20-foot insulated ocean shipping containers to HARCC, which now represents a crucial thrust forward in facility development. Initial HARCC blueprints relied upon a building that had been provided onsite at Lancetilla, inside which we would fabricate a series of isolated amphibian rooms. Unfortunately, upon closer inspection of this facility, we determined it would be too difficult and costly to make the series of structural repairs that would have been required to raise the integrity of the building to meet our biosecure standards. We have been pursuing our "Plan B"—modified ocean shipping containers that will be stationed right outside this building which will instead be converted into the HARCC headquarters office and a feeder-insect rearing facility. With this recent donation from Werner Enterprises, Inc., we are now in the fabrication stage during which these two containers will be transformed into the mobile biosecure amphibian rescue laboratories that will be used to protect three species of Critically Endangered Honduran amphibians from Cusuco National Park: *Plectrohyla dasypus*, *Plectrohyla exquisita* and *Duellmanohyla soralia*. After construction of these laboratories is completed at Omaha's Henry Doorly Zoo & Aquarium in the USA, they will then be shipped to Honduras and installed onsite at Lancetilla.

You can now see HARCC amphibian rescue updates as they happen by following our brand new Facebook page! Simply click here and request notifications: HARCC-Honduras Amphibian Rescue and Conservation Center

For more information about HARCC or to inquire about opportunities for involvement, please contact us at Hondurasarcc@gmail.com

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Plectrohyla exquisita. Photo: Jonathan Kolby.

Plectrohyla dasypus. Photo: Jonathan Kolby.

I Colloquium about Amphibian Research and Conservation in Universidad Nacional, Heredia, Costa Rica

By Víctor J. Acosta-Chaves

Approximately 200 amphibians occur in Costa Rica making it one of the Neotropical amphibian hotspots despite the country's small area size (*1*). Despite being one of the most studied tropical countries (*2*) a myriad of threats put as much as 10% of the country's amphibian diversity at risk (*3*). Research on how to counter and mitigate this decline is urgently needed. Much of the past and current research has focused on health population status, taxonomy, behavior, habitat selection and natural history of previously unstudied or threatened species and success of *in situ* conservation and management strategies (Fig. 1). Most of the results of these studies are limited to selected scientific spheres. However, the public is also interested in these topics as posts in social networks (*e.g.*, "Herpetólogos de Costa Rica" on Facebook) dedicated to herpetology in Costa Rica suggests. Even when the Universidad Nacional has been related with wildlife conservation and environmental education programs, amphibians have not been the main topics of those efforts until now, in contrast with similar institutions as in Universidad de Costa Rica (UCR).

Because of this need, on February 23, 2015, a short colloquium was organized at the Universidad Nacional (UNA) by Asociación de Estudiantes de Ciencias Biológicas (UNA), members of Red MesoHerp in Costa Rica and the SAVE THE FROGS organization. Goals of this afternoon were: A) share (ongoing) research on amphibians of Costa Rica (and Panama) primarily thorough M.Sc. and Ph.D. students at the meeting; B) to create an open and free source of amphibian information for the public which are normally not invited to scientific workshops or symposiums about amphibians; C) motivate young undergraduate students to develop projects in amphibian conservation or research; and D) open discussion about the presented experiences in the field, laboratory and else where.

Fig. 1: Blue-Sided Leaf Frog (*Agalychnis annae*: Hylidae), an Endangered species in Costa Rica that was treated at the colloquium. Photo: V. Acosta.

Special guest of the colloquium was Dr. Kerry Kriger whom presented a lecture about the work of SAVE THE FROGS and its activities around the globe (Fig. 2). In total, 12 oral presentations (20 minutes each) were given on conservation and research experiences (Fig. 3). Topics varied and included research on phyllogeography, amphibians skin bacteria and its inhibitory properties against chytrid, pesticides and frog population health, habitat selection and occupancy of cloud forest anurans and more. The colloquium was a great success due to approximately 90 attendees and the clear need to create open spaces for extension and discussion of scientific research, conservation experiences and discussion on the mentioned research in and between academics and non academics. The goal for the future is to continue to organize similar activities on herpetological topics and include more institutions and people in order to facilitate connection and collaboration between researchers, policy makers and novices.

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Fig. 2: Kerry Kriger lecturing about his experiences in amphibian conservation. Photo: V. Acosta.

Fig. 3: Batrachologists joining the colloquium (left to right): Ángel Sosa, Cesar Barrio, Joseph Vargas, Juan Abarca, Kerry Kriger, Víctor Acosta, Josimar Estrella, Erick Ballestero and José Salazar. Missing are: Adrián García and Héctor Zumbado. Photo by V. Acosta.

A captive bred *Nototriton abscondens* at four months old. This is the first time any species from the genus *Nototriton* has bred in captivity. Photo: Brian Kubicki.

Amphibian Ark Announces Seventh Annual Call for Proposals for its Seed Grant Program

By [1]Kevin Johnson

This $5,000 competitive grant is designed to fund small start-up *ex situ* rescue projects for species that cannot currently be saved in the wild. Successful proposals will reflect AArk values—please pay careful attention to the grant guidelines below for details on the types of projects that are favored. This includes those that:

- Focus on species whose threats cannot be mitigated in nature in time to prevent their extinction and who therefore require *ex situ* intervention to persist;
- Work with species within their native range country;
- Involve range-country biologists;
- Adhere to recommended biosecurity standards for *ex situ* programs;
- Link *ex situ* programs to *in situ* conservation;
- Involve partnerships to maximize the likelihood of the program's long-term sustainability.

Interested participants can download grant guidelines as a pdf file. **Applications are due no later than May 15, 2015.**

Since 2009, AArk has awarded 16 seed grants, with 11 of these being within the Americas, in Argentina, Bolivia, Brazil, Chile, Colombia, Costa Rica, Ecuador, Honduras and Puerto Rico. Some of these programs are highlighted below. All of these new projects have progressed well, with most securing additional funding, and successful captive breeding programs now underway. Like many new conservation projects, though, they are seeking additional resources to expand and improve their programs. If you would like to support any of these programs, please contact the program managers directly, or contact AArk staff at info@amphibianark.org

COSTA RICAN AMPHIBIAN RESEARCH CENTER SUCCESSFULLY BREEDS MOSS SALAMANDERS FOR THE FIRST TIME

Costa Rica is home to 200 species of amphibians within a tiny national territory of approximately 51,000 km^2. Nearly a quarter of Costa Rica's amphibian diversity is made up of three genera and 49 species of salamanders. Unfortunately most of Costa Rica's salamanders are still surrounded by mystery in regards to their general biology and actual distributions.

Brian Kubicki from the Costa Rican Amphibian Research Center (CRARC) is hoping to obtain a better understanding of Costa Rican salamanders through research efforts both in the wild and in captivity. Since 2012 Brian Kubicki and Aura Reyes of the CRARC have invested hundreds of hours working with all eight Costa Rican species of Moss Salamanders (*Nototriton*) at numerous sites throughout the country in order to take a closer look at their taxonomy, natural history and distribution. During 2013 the CRARC collected several

[1]Taxon Officer, Amphibian Ark.

species of Costa Rican moss salamanders to form captive colonies. These salamanders adapted very well to their specially designed terrariums, yet the researchers did not observe breeding until 2013.

From June to December of 2014, researchers discovered several egg masses in the terrariums at the CRARC, resulting in the first-known captive breeding events for any species of moss salamander. In total, three species of moss salamander have been successfully bred in the CRARC lab (*Nototriton abscondens*, *N. gamezi* and *N. major*). Raising the tiny captive-bred moss salamander neonates presented some initial difficulties, but modifications to their food items and enclosures have shown positive results so far, with several captive-bred individuals currently nearing adult sizes.

CRARC is currently seeking additional resources to help with electrical consumption in the lab for the chiller, pumps for the salamander systems, materials for modifying or creating new enclosures and costs involved in continued field observations and collections. CRARC plans to continue conducting *in situ* and *ex situ* studies not only on Costa Rican moss salamanders, but on additional species of *Bolitoglossa* and *Oedipina*, which are the two other genera of salamanders native to Costa Rica.

CRARC's web site, www.cramphibian.com, includes more details about the group's research and conservation work and the species it is working with.

DEVELOPING THE BOLIVIAN AMPHIBIAN INITIATIVE

The Bolivian Amphibian Initiative was established in 2010 in Cochabamba, Bolivia, by Arturo Muñoz Saravia, and it focuses primarily on poorly known aquatic frogs of the genus *Telmatobius*, such as the Critically Endangered Titicaca Water Frog (*Telmatobius culeus*) and the Vulnerable-classified Water Frog (*Telmatobius hintoni*). The Bolivian Amphibian Initiative is using these two species as model species to develop husbandry guidelines that can be used with the other 12 Bolivian species of *Telmatobius* classified as Endangered by the IUCN Red List of Threatened Species. During the Conservation Needs Assessment for Bolivian amphibians in 2014, 13 of the 14 species of *Telmatobius* were recommended for urgent *ex situ* rescue.

In 2010, the initiative collected Water Frog (*Telmatobius hintoni*) tadpoles from the wild, and although early reproductive efforts were not successful, in 2012 the species was bred successfully for the first time in captivity. Later in 2012, pairs of Titicaca Water Frogs were established, and researchers observed amplexus soon after. Unfortunately, after 300 eggs were laid, only about a third of these hatched.

The Bolivian Amphibian Initiative is also carrying out field work in Lake Titicaca to study the Titicaca Water Frog. The group has found some populations in the lake that are still in good condition, but in some areas almost 50 percent of the population was dead and other individuals were in very poor condition and looked very sick. There are also several areas where it was very difficult to find frogs and these populations are facing a variety of challenges. The initiative plans on conducting additional field research to determine what it is happening there, and the causes for these significant losses.

More information can be found on the website at www.bolivian-amphibianinitiative.org

The Bolivian Amphibian Initiative plans to carry out more field research on the Endangered Titicaca Water fFog (*Telmatobius culeus*) to try to determine the causes of the significant population declines. Photo: Arturo Muñoz Saravia.

DEVELOPING A CONSERVATION PLAN FOR THE UPLAND COQUÍ

The Upland Coquí (*Eleutherodactylus portoricensis*), or "Coquí de la Montaña," from Puerto Rico, is a high-elevation montane species classified as Endangered by the IUCN Red List of Threatened Species, and was recommended for *ex situ* rescue during the 2011 AArk Conservation Needs Assessment for Caribbean amphibians. This species has a dramatically declining distribution throughout most Central and Western mountain ranges (locally extinct), and it is only found in eastern and southeastern forest areas above 650 meters in elevation.

Neftalí Ríos from the University of Puerto Rico in Humacao was granted collection and Institutional and Animal Care and Use Committee (IACUC) permits in December of 2013. Ríos initiated field work, collecting 28 male and female adults, and captive efforts for this species to gain a better understanding of factors influencing its population dynamics and its poorly understood reproductive biology.

In May 2014, the first-ever captive reproduction occurred and researchers recorded amplexus in the captive breeding facilities at UPR-Humacao. Since then, 15 egg clutches have been laid in breeding areas, including on artificial bromeliads, but 57 percent have been lost to oophagy and abandonment by the guarding male. The female was removed after detecting oviposition in a terrarium to reduce oophagy. Abandoned egg clutches are being raised in petri dishes until eclosion. Hatchlings took collembolans immediately until juveniles were three to five months old and ready to feed on fruit flies (dusted with multivitamins and calcium). Juveniles were then placed into one liter deli cups with sphagnum moss as bedding and are doing fine so far. Approximately 60 juveniles about

Captive breeding facility and terrariums for the Upland Coquí (*Eleutherodactylus portoricensis*) at the University of Puerto Rico in Humacao. Photo: Neftalí Ríos.

1/3 the size of adults are now being raised on a diet based on multivitamin-dusted fruit flies and pin-size crickets. Researchers do not know the time required to reach adulthood, but the main goal is raising them until the individuals are reproductively mature and to breed them accordingly.

Future plans include continuing *ex situ* captive efforts and *in situ* population monitoring with the Mountain Coquí to develop captive-breeding protocols for similar montane *Eleutherodactylus* in need of conservation. Researchers have initiated a breeding ecology project using artificial breeding houses (a modified version of the 1970s and 80s "bamboo houses" frequently used by visiting herpetologists in Puerto Rico) to better understand the species' breeding biology in the wild. The researchers seek additional support to help with food and supplements; materials in the laboratory for the water filtering system, electrical fixtures, and materials for creating new mini-terrariums and costs involved in field work.

For additional information, please visit www.uprh.edu/labneftali (Spanish version only, English version under construction).

ARGENTINA GETS ITS FIRST AMPHIBIAN RESCUE CENTER

With the support of an AArk seed grant in 2014, the first amphibian rescue center in Argentina has been established at La Plata University in Buenos Aires, and is being managed by Federico Kacoliris and Jorge Williams. This center aims to protect endemic and endangered amphibians from Argentina with the goal of promoting the long-lasting viability of their wild populations.

The long-term plan for the center is to work in three stages: 1) establishing assurance colonies of highly threatened species and breeding them; 2) working *in situ* to alleviate the main threats and restoring their natural habitats, and 3) establishing a reintroduction program aimed at stabilizing wild populations.

Last year the facilities for amphibians' assurance colonies at La Plata University—were built and permits were sought to collect founder animals of two species, the Critically Endangered Valcheta's Frog (*Pleurodema somuncurensis*), which is endemic to the Somuncura plateau, Patagonia; and Calchaquíes Valleys' Frog (*Telmatobius pisanoi*), which is classified as Endangered and endemic to Valles Calchaquies in northern Argentina. The primary threats for these two species are habitat loss, predation by exotic fishes, alteration of watersheds and agricultural and potential mining activities. Chytridiomycosis is also a potential threat for both species.

Earlier this year the permits to work with Valcheta's frogs were received and during March a field trip resulted in the collection of 40 individuals (20 males and 20 females) of this species. The individuals were brought to the rescue center and they are now stabilized within the terrariums, which recreate their native habitat. No individuals died during the trip.

During the next few months the rescue center will provide these individuals with the necessary conditions to encourage reproduction. Although a lot of work needs to be done, conservationists hope that the species will breed in captivity, so that the second and third stages of the plan can be initiated. In the meantime the rescue center is seeking additional funds to expand the *ex situ* facilities to other endangered species and to start with the habitat recovery plan.

Early in 2015, 20 pairs of Endangered Valcheta's Frog (*Pleurodema somuncurensis*) were collected as founder animals for a new assurance colony at Argentina's first amphibian rescue center in Buenos Aires. Photo: Federico Kacoliris.

New Rescue Lab for Endangered Amphibians Opens in Panama

By Devin Murphy

Smithsonian Conservation Biology Institute (SCBI) and Smithsonian Tropical Research Institute (STRI) scientists working together as part of the Panama Amphibian Rescue and Conservation Project (PARC) opened a new safe haven for endangered amphibians today, April 8. The state-of-the-art, $1.2 million amphibian center at STRI's Gamboa field station is the largest amphibian conservation facility of its kind in the world. The new center expands on the capacity of the El Valle amphibian conservation center to implement a national strategy to conserve Panama's amphibian biodiversity by creating captive assurance populations.

Panama is a biodiversity hotspot for amphibians with more than 200 species of frogs, salamanders and caecilians. For the past 20 years, however, many of Panama's unique and endemic amphibian species have declined or disappeared as a result of the deadly chytrid fungus that has spread throughout Latin America and the Caribbean. In fact, a third of amphibian species in Panama are considered threatened or endangered. Amphibian conservationists around the world have been working to establish captive populations of the world's most vulnerable amphibian species to safeguard them from extinction. Since 1980, 122 amphibian species are thought to have gone extinct worldwide, compared to just five bird species and no mammals during the same period.

"Our biggest challenge in the race to save tropical amphibians has been the lack of capacity," said Brian Gratwicke, amphibian scientist at SCBI and international coordinator of PARC. "This facility will allow us to do so much more. We now have the space needed to safeguard some of Panama's most vulnerable and beautiful amphibians and to conduct the research needed to reintroduce them back to the wild."

The center features a working lab for scientists, a quarantine space for frogs collected from the wild and amphibian rescue pods capable of holding up to 10 species of frogs. In the working lab, SCBI scientists will continue research focusing on things like a cure for chytrid. They published findings last month in *Proceedings of the Royal Society* showing that certain Panamanian Golden Frogs were able to survive infection with chytrid as a result of a unique skin-microbe community already living on their skin. Seven amphibian rescue pods house the amphibian collection and colonies of insects needed to feed them. Amphibian rescue pods are constructed from recycled shipping containers that were once used to move frozen goods around the world and through the Panama Canal; they have been retrofitted to become mini-ecosystems with customized terrariums for each frog species.

New Rescue Lab at the Gamboa Amphibian Research and Conservation Center Photo: Smithsonian Conservation Biology Institute.

"Our project is helping implement the action plan for amphibian conservation in Panama, authored by Panama's National Environmental Authority, Environment Ministry, in 2011," said Roberto Ibantilde, STRI project director for PARC. "This is only possible thanks to the interest in conservation of amphibian biodiversity by the government of Panama and the support we have received from businesses in Panama."

The new rescue lab will be crucial to ongoing breeding efforts and breakthroughs, such as the successful hatching of an *Andinobates geminisae* froglet. SCBI and STRI scientists hatched the first *A. geminisae* froglet in human care in one of the amphibian rescue pods at the existing Gamboa amphibian conservation center. The tiny poison frog species, smaller than a dime, was discovered and described for the first time in Panama in 2014. They simulated breeding conditions in a rescue pod. The new facility will provide much-needed space to grow and expand, allowing them to build assurance populations for many more species. A small exhibition niche provides a window directly into an active rescue pod, where visitors can see rescued frogs and scientists as they work to conserve these endangered frogs.

PARC is a partnership between the Houston Zoo, Cheyenne Mountain Zoo, Zoo New England, SCBI and STRI. Funding for the new facilities was provided by Defenders of Wildlife, Frank and Susan Mars, Minera Panama, the National Science Foundation and USAID.

As a research facility, PARC is not open to the public. However, there are interpretive panels and a window into the research pod where visitors can get a glimpse of the project in action. To learn more, the public is welcome to visit the new Fabulous Frogs of Panama exhibit at the Smithsonian's Punta Culebra Nature Center, located on the Amador Causeway.

This is the first captive-bred *Adinobates geminisa*e frog at the Gamboa Amphibian Research and Conservation Center. The species is endemic to Panama, was described to science in 2014, and has a very restricted distribution. Photo: Smithsonian Conservation Biology Institute.

Life in the Time of *Bd*: Long-Term Monitoring of Wild *Atelopus* in Eastern Panama

Atelopus limosus in habitat. Photo: Twan Leenders.

By [1]Twan Leenders, [2]Michael Roy & [3]Alex Shepack

They are tiny and barely visible on the shaded, moss-covered rocks they inhabit. However, in spite of their miniscule size their continued presence represents something huge: metamorphs of the Limosa Harlequin Toad (*Atelopus limosus*) indicate that there is still hope for some *Atelopus* species. A seemingly thriving wild population in eastern Panama now provides unique opportunities for research that can benefit conservation efforts worldwide.

The Limosa Harlequin Toad is classified as "Endangered" by the IUCN because of its small geographic range and impacts of habitat fragmentation and degradation throughout its area of occurrence. It is a species of lower elevations and theoretically less vulnerable to *Bd*-related declines than the middle and high elevation species in its genus (*1*). Nevertheless the spread of *Bd* in eastern Panama is taking a toll on this species even though parts of its range include areas of pristine habitat within and outside of protected areas (*e.g.*, Chagres National Park) (Brian Gratwicke, pers. comm.).

Amphibian Survival Alliance (ASA) Partner organizations Conservation through Research Education and Action (CREA) and the Roger Tory Peterson Institute of Natural History (RTPI) have implemented a long-term monitoring and research initiative in Cocobolo Nature Reserve (which is managed by CREA) that sets out to: 1) address the ecology and population dynamics of wild *Atelopus*; 2) assess the amphibian assemblage in the reserve; and, 3) investigate how certain species or populations persist in spite of *Bd*.

Initial surveys of the *Atelopus limosus* in Cocobolo Nature Reserve, in cooperation with the Smithsonian Tropical Research Institute (STRI), indicated that *Bd* may not have been present in 2011 and that the population appeared stable. Recent field work has provided additional reason for cautious optimism. In March 2013 we were able to document the first wild breeding event of *Atelopus limosus*; subsequent field work in 2014 revealed the presence of about two dozen metamorphs and young juveniles in suitable habitat. Concerted efforts to document recruitment and population dynamics during field work in February and March of this year, just prior to the publication of this article, led to the discovery of more than 120 juvenile *Atelopus limosus* in two separate watersheds contained within the Cocobolo Nature Reserve. Standardized photographs of the unique dorsal pattern of each frog were taken to facilitate future analysis using pattern recognition software, and all were swabbed for *Bd*. In spite of the initial indication that *Bd* was not present in the reserve, very recent findings of (non-*Atelopus*) dead frogs in the habitat and the knowledge that nearby *A. limosus* populations have declined in recent years due to *Bd* (Roberto Ibañez, pers. comm.) may indicate that the situation is more complex. Analysis of the swabs and data gathered during the most recent field work will provide more insight soon.

Located on the continental divide in eastern Panama, Cocobolo Nature Reserve encompasses a significant range of elevations,

[1]Dr. Twan Leenders is a conservation biologist with over 20 years of herpetofaunal research experience in the Neotropics; he is president of the Roger Tory Peterson Institute of Natural History and an Affiliate at Yale University's Peabody Museum of Natural History. [2] Dr. Michael Roy is a population geneticist with extensive field experience in Panama and abroad; he is president of the Panamanian NGO "Conservation Research Education Action" (CREA) which manages the Cocobolo Nature Reserve [3] Alex Shepack, M. Sc., is a doctoral student at Southern Illinois University (SIU) and a research associate at RTPI, his studies focus on how amphibian communities respond to continued *Bd* exposure by studying select recovering amphibian populations.

***Atelopus limosus* habitat. Photo: Twan Leenders.**

habitats and historic land uses. Within its boundaries, Caribbean, Pacific, Central and South American faunas mix with influences from a still poorly understood zone of endemism in the Chagres Highlands (2). The resulting amphibian assemblage heightens the conservation value of Cocobolo Nature Reserve, where other species of conservation concern, such as *Craugastor tabasarae* (IUCN Critically Endangered) and a recently discovered undescribed species of minute salamander (*Bolitoglossa* sp.) have been found. Thus far, 49 amphibian species have been documented in Cocobolo Nature Reserve, but herpetofaunal surveys have only just begun and have been limited to the dry season, suggesting that many additional species can be expected to occur there. The reserve is part of the Mesoamerican Biodiversity Corridor and bordered by vast areas of uninterrupted primary forest in Kuna Yala territory and Chagres National Park. Based on climate and habitat characteristics, Cocobolo Nature Reserve has significant potential to harbor additional conservation priority amphibians.

The continued presence of sensitive species in a region where *Bd* is prevalent warrants further investigation. A significant amount of research has been dedicated to documenting declines associated with *Bd* throughout the world but far less effort has been dedicated to understanding how certain species or populations persist in spite of *Bd.* Numerous theories have been proposed, including innate resistance, changes in *Bd* pathogenicity, evolved resistance, climactic refuges, behavioral fevers and several others. Cocobolo Nature Reserve and the local *Atelopus* provide unique opportunities to study possible causes for *Bd* resistant populations. Furthermore, the enigmatic abundance of the dendrobatid frog *Silverstoneia flotator* and several other amphibians that occur syntopically with the *Atelopus* population begs further research. In areas where *Atelopus* populations were extirpated, *S. flotator* persists in seemingly healthy numbers (Twan Leenders, pers. obs.) suggesting that it may play a role as a vector or reservoir for *Bd,* or possibly display elevated resistance to the pathogen.

Surveying and examining amphibian populations within Cocobolo Nature Reserve in the context of *Bd* prevalence and pathogen infection dynamics is a research priority for the coming years. However, a different angle to our studies involves the spatial analysis of amphibian population changes in light of historic land use patterns and ongoing forest regeneration in sections of the reserve. In conjunction, these studies will hopefully elucidate how the area's amphibians have managed to persist and seemingly increase in a

***Craugastor tabasarae*. Photo: Twan Leenders.**

Bolitoglossa sp. Photo: Twan Leenders.

Diasporus aff. *diastema.* Photo: Twan Leenders.

Silverstoneia flotator carrying tadpoles. Photo: Twan Leenders.

Dendrobates auratus. Photo: Twan Leenders.

Atelopus limosus eggs and larvae *in situ*. Photo: Twan Leenders.

Students working in transect. Photo: Twan Leenders.

time when other populations and species are declining precipitously. The conservation implications of these studies are numerous: this project will benefit applied conservation programming and increase knowledge about how populations respond to stressors like disease and habitat change. Testing observed individuals for *Bd* will allow us to produce maps of infection throughout the reserve to analyze for possible climactic refugia and understand how infection prevalence varies by season. Analysis of the genetic structure of this population of *A. limosus* will determine whether there has ever been an impact by *Bd* or elucidate how this population has persisted in spite of *Bd.* If found to be present, samples of the local *Bd* strain will be cultured and analyzed for its phylogenetic position and relatedness to known pathogenic strains of the fungus.

Detailed habitat use data on the *A. limosus* population is being gathered along a 1,000 m transect and correlated with environmental data collected by a series of automated loggers. It is our hope that the information gathered from this study will allow for predictive modeling and identification of other potentially functional *Atelopus* habitat. In addition, one of the potential future applications of this study is the restoration of riparian corridors in collaboration with neighboring farmers to create controlled release sites for *A. limosus* originating from *ex situ* reassurance populations that are currently "stranded" in carefully controlled facilities. Although these programs have been successful in boosting captive populations of *A. limosus*, they have not yet been able to release individuals back into the wild due to the continued presence of *Bd.* Initial conversations on this topic have begun with the Panama Amphibian Rescue and Conservation Project and will evolve as the project continues.

Juvenile ***Atelopus limosus***, photographed on a standardized background. Their unique dorsal pattern can be used to identify individual frogs. Photo: Twan Leenders.

Alex Shepack swabbing for ***Bd***. Photo: Twan Leenders.

The combination of fundamental research, applied conservation and experiential education and outreach is at the core of CREA and RTPI's mission. Opportunities for researchers, volunteers or student groups to get involved in this project, or other ongoing research and conservation initiatives, are available through both organizations. For more information, please contact Michael Roy (michael@crea-panama.org) or Twan Leenders (tleenders@rtpi.org).

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Unexpected Results in Probiotics Study Leads to new Questions for Scientists Seeking to Protect Golden Frogs

By Lindsay Renick Mayer

In the Panamanian Golden Frog Lab at the Smithsonian Conservation Biology Institute, one fact has remained steadfast: there is as much to learn from life as death, from the expected as the unexpected.

SCBI biologist Matt Becker knows this well. For his PhD dissertation at Virginia Tech, Becker took four probiotics—or beneficial bacteria—from frog species in areas of Panama where fungal disease chytridiomycosis was endemic and gave 32 Golden Frogs a probiotics bath before infecting them with *Bd*. In the end, Becker was disappointed to find that the beneficial bacteria didn't successfully stay on the frogs' skin to protect them from *Bd*. And yet somehow, seemingly impossibly, 30 percent of the frogs survived exposure to the disease. Only one Golden Frog in past studies had survived, which left Becker wondering what had happened.

"At first I wasn't even certain that those frogs had actually been infected," Becker says. "You basically take a very concentrated *Bd* stock and put less than a milliliter in a large container of water. But it always works. We had all but two frogs became infected."

The search for what the frogs that cleared infection had in common led Becker to an interesting and unexpected finding. Those frogs, it turned out, did have one thing in common from the start that the frogs that died didn't share: a high abundance of six bacteria completely unrelated to the probiotics Becker had tried. This surprising result is the focus of Becker's current study as a postdoctoral fellow at the Smithsonian Conservation Biology Institute: Did the high abundance of these six bacteria actually protect the individual frogs from the deadly pathogen or was this merely a correlation, while something else was at play? And if these bacteria did save the frogs from otherwise certain death, can researchers figure out how to get these bacteria to stick to the skin of frogs that don't already have that defense?

The next phase of the study will also try to determine why some of the Panamanian Golden Frogs had high abundances of these six bacteria, while others did not, even though they all came from the Maryland Zoo in Baltimore. His most recent work was published in Proceedings of the Royal Society in March.

"The findings in this paper are very encouraging," says Reid Harris, director of international disease mitigation for the Amphibian Survival Alliance and co-author on the paper. "If these bacteria do lead to survival, then we have the exciting possibility of adding these bacteria to frogs that lack them, or that have them in low numbers. If these bacteria colonize and persist, then this probiotic solution will allow us to get protected frogs into the wild. It is exciting to imagine getting a species that is probably extinct in the wild back into nature."

Cori Richards-Zawacki, an assistant professor at Tulane University, is grappling with similar questions, though with a different set of seemingly unlikely survivors. What Richards-Zawacki and fellow researcher Jamie Voyles found in the wild after *Bd* swept through is encouraging. At first they found just a variable harlequin frog (*Atelopus varius*)—the sister species to the Panamanian Golden Frog—here or there at a handful of sites where they were once abundant. Most recently, however, they discovered a population of about 40 frogs at one site and are working to understand how this species is making a small comeback.

"We're looking into whether the pathogen has become less pathogenic, or if the frogs have developed some sort of resistance," Richards-Zawacki says. "This ties really well into what Matt Becker has found, that there seems to be some natural variation in how susceptible individuals are to this disease."

Neither Richards-Zawacki nor Becker are certain that the beneficial bacteria is actually the direct cause of survival, but they're both determined to discover the difference between the survivors and those that succumb to *Bd*. They're looking at the possible role of genetics—both of the frogs and the fungus—in varied responses to the pathogen. They're talking to scientists who are investigating how the frogs' environment can affect individuals' ability to fight off disease. And they're meeting with scientists doing immunology work in search of a potential vaccine.

"I'm definitely hopeful," Richards-Zawacki says. "I'd love to someday see either a re-introduction take place or for us as a community to do everything we can to foster the recovery of the populations that we have."

Dr. Cori Richards-Zawacki searching for Panamanian Golden Frogs at a site in Central Panama where they were once abundant. Photo: Brian Gratwicke.

First Isolation of Chytrid from French Guiana

By Jennifer Shelton, Pria Ghosh & Matthew Fisher

Since its discovery in the late 1990s, the amphibian parasitic fungus *Batrachochytrium dendrobatidis* (*Bd*) has been reported in captive and wild amphibian populations in Central, South and North America, Europe, Africa and Australia. This distribution most likely reflects ancient patterns of endemism but also more contemporary movements as *Bd* is vectored between countries via international trade. Most recently the fungus has been identified in Madagascar (*1*)—a hotspot for amphibian biodiversity. This finding has serious implications for the health of Malagasy amphibians, 99% of which are not found anywhere else in the world. Where *Bd* has been found it generally exists in two capacities: either as a long-term endemic infection with no obvious effects on host fitness, or as an epizootic associated with clinical disease and population declines. Examples of endemic *Bd* lineages are those occurring in Southern Africa (*Bd*CAPE), Switzerland (*Bd*CH) and Brazil (*Bd*BRAZIL). A hypervirulent, globally-distributed lineage is known as *Bd*GPL (Global Panzootic Lineage) and is responsible for almost all known chytrid-driven declines to date.

*Bd*GPL has been found to share geographical distributions with endemic lineages, such as *Bd*BRAZIL in the Atlantic Coastal Forest of Brazil (*2*). If coinfections were to occur between different lineages then ecological theory shows that one lineage will inevitably outcompete the other when two similar lineages occupy the same niche. With its successful global spread *Bd*GPL would likely be the aggressor in this situation; a notion supported by laboratory findings that *Bd*GPL displaces *Bd*CAPE in experimental coinfections and by field observations suggesting that *Bd*CH has been replaced by *Bd*GPL. On the other hand, *Bd*GPL is rare to non-existent in some regions, which could be due to amphibians having developed an intrinsic resistance to endemic *Bd*s, or due to infection with an endemic, non-virulent lineage that protects against *Bd*GPL.

Prof. Mat Fisher's group, collaborator on the Global Bd Mapping Project (www.bd-maps.net), has recently received funding from the Leverhulme Trust for a project aimed at characterising the worldwide biodiversity of amphibian chytrids and exploring the fitness of chytrid lineages in competitive interactions. The first step will be to develop a universal DNA barcoding pipeline that captures all chytrid fungal lineages, which will be applied to skin-DNA samples in order to determine the fungal communities existing on amphibian skins. Frog toe-clippings will then be used to isolate and culture

A) *Allobates femoralis*. Photo: Pria Ghosh. B) *Atelopus flavescens.* Photo: Matthew Fisher. C) *Dendrobatidis tinctorius*. Photo: Matthew Fisher.

infecting chytrids for genome sequencing in order to identify novel chytrid genomes and lineage-specific qPCR will be used to survey the prevalence and intensity of infection of known *Bd* lineages.

Recently, Mat Fisher and research assistant Pria Ghosh visited a research station in La Réserve Naturelle Nouragues, French Guiana. The reserve covers 150,000 ha, with 9,000 ha designated exclusively for scientific research, and presents a fantastic opportunity to study a pristine, remote rainforest where human impact has so far been extremely limited. Along with local researchers Elodie Courtois from CRNS and Claude Miaud from CEFE/CNRS, they collected 280 samples (toe-clippings and skin-swabs) from six frog species: *Allobates femoralis*, *Anomaglossus baeobatrachus*, *Atelopus flavescens*, *Dendrobatidis tinctorius*, *Leptodactylus myersi* and *Pristimantis espedeu*s. The samples were collected across a 400 m altitudinal gradient, from the Arataye river valley to the top of the Inselberg.

The aim was to survey chytrid diversity in an area that is, as far as is reasonably possible, unlikely to harbour introduced lineages of chytrids. The work was funded by CNRS, and it is hoped that this field trip will kickstart an ongoing collaboration where the diagnostic capabilities of Imperial College London will be paired with the data and extensive monitoring systems put in place by the Reserve Management teams across French Guiana. The widespread surveillance of the reserve has so far resulted in detection of *Bd* via qPCR from skin swabs (*3*,*4*). However, limited diagnostic capabilities in the country mean that it is not currently known what type of *Bd* is present, nor the prevalence or infection intensity as many of the samples have yet to be analysed. The skin swabs will be tested with lineage-specific qPCRs, the results of which will be fed back into the reserve management authorities. Backdated collections of swabs from 2014 and 2013 will also be analysed. A key objective of the trip was to attempt to isolate and culture a French Guiana *Bd* which, if successful, will be the first isolation from the country. It is hoped that lineage-specific qPCRs will discover novel chytrid diversity that may be missed by the traditional qPCR analysis. Excitingly, Pria has been able to isolate *Bd* zoospores and sporangia from two of the samples collected and work can now begin on trying to establish pure cell cultures of these isolates prior to genome sequencing.

It is hoped that this study will act as a springboard for improved *Bd* surveillance and monitoring in reserves across the country through collaboration between the reserve management authorities and the Fisher Group at Imperial College London. It is essential for future chytrid management to develop an increased understanding of the distribution and characteristics of *Bd* lineages. This study should be a step in the right direction for management of amphibian biodiversity in French Guiana.

Researchers collecting samples in the rainforests of La Réserve Naturelle Nouragues, French Guiana. Photo: Pria Ghosh.

Time to return after a successful trip; Prof. Claude Miaud loads the helicopter for the ride home from Nouragues Reserve. Photo: Matthew Fisher.

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Photo: CEDIA.

Amazon Survey Finds Potentially New Frog Species

By Joe Lowe

A rapid biological inventory conducted in the Peruvian Amazon has confirmed the region's herpetological importance with the discovery of four frog species believed to be new to science.

Conducted by Chicago's Field Museum, the study sought to create an initial, detailed inventory of the flora and fauna found within the Tapiche-Blanco watershed in Peru's Loreto Department.

Sampling terrestrial and aquatic habitats, herpetologists found well-preserved communities of amphibians and reptiles throughout the area. The team recorded 64 amphibian and 48 reptile species. They estimate total regional herpetofauna to include at least 124 amphibians and 100 reptiles in total.

Among noteworthy species found during the survey was the Poison Dart Frog (*Ranitomeya yavaricola*), which is restricted to southern Loreto. The four frog species considered potentially new to science include *Hypsiboas* aff. *cinerascens*, *Osteocephalus* aff. *planiceps*, *Chiasmocleis* sp. nov. and *Pristimantis* sp. nov. (lacrimosus group).

Of the reptiles confirmed present, two are globally Vulnerable species, the Yellow-Footed Tortoise (*Chelonoidis denticulata*) and the Yellow-Spotted River Turtle (*Podocnemis unifilis*).

The Tapiche-Blanco watershed forms the core of a proposed 740,000-acre reserve, which is part of a major conservation project implemented by the Center for the Development of an Indigenous Amazon (CEDIA) with Rainforest Trust's support. Upon establishment, the White Sands National Reserve will help consolidate a 10-million-acre wildlife corridor.

Despite its impressive biodiversity, the Tapiche-Blanco watershed remains vulnerable to destruction by extractive industries. The area is threatened by logging, road building and oil drilling.

"This study is a great benefit for our protection efforts. We now have a wealth of evidence about the biological importance of the region and can make an even stronger case as to the merits of its conservation," said Christine Hodgdon, International Conservation Manager for Rainforest Trust. "At the same time, the extent of what we stand to lose and the urgent necessity of preserving it has never been clearer."

RAINFOREST TRUST

Conservation and Ecology

Geotrypetes seraphini. Photo: Benjamin Tapley.

Towards evidence-based husbandry for caecilian amphibians: Substrate preference in *Geotrypetes seraphini* (Amphibia: Gymnophiona: Dermophiidae)

Benjamin Tapley, Zoe Bryant, Sebastian Grant, Grant Kother, Yedra Feltrer, Nic Masters, Taina Strike, Iri Gill, Mark Wilkinson & David J. Gower

Maintaining caecilians in captivity provides opportunities to study life-history, behavior and reproductive biology and to investigate and to develop treatment protocols for amphibian chytridiomycosis. Few species of caecilians are maintained in captivity and little has been published on their husbandry. We present data on substrate preference in a group of eight Central African *Geotrypetes seraphini* (Duméril, 1859). Two substrates were trialled; coir and Megazorb (a waste product from the paper making industry). *G. seraphini* showed a strong preference for the Megazorb. We anticipate this finding will improve the captive management of this and perhaps also other species of fossorial caecilians, and stimulate evidence-based husbandry practices.

B. Tapley, Z. Bryant, S. Grant, G. Kother, Y. Feltrer *et al. Herp. Bul.* **129**, 15–8 (2014).

Amphibian community responses to playa restoration in the rainwater basin

Benjamin J. Beas & Loren M. Smith

Alterations to wetland hydrology associated with conversion of grassland watersheds to croplands can alter functional components that drive many wetland ecosystem services. Biodiversity provisioning is one of many services that can be negatively impacted by grassland conversions. Our objective was to compare amphibian communities of playa wetlands in native grassland versus cropland watersheds and to determine if restoration of cropland wetlands via sediment removal improved these habitats for amphibians. We collected amphibian species richness, water depth, wetland area, and emergent plant cover data from 34 playa wetlands in 2008 and 32 playa wetlands in 2009. Playas were selected to include those within natural grassland watersheds (*i.e.*, reference playas), cropland watersheds, and restored watersheds within the Rainwater Basin (RWB) of Nebraska. While we did not detect a difference in amphibian species richness among land-use types in 2008, in 2009 (a dry year within the RWB) amphibian species richness averaged nearly two times greater in restored playas (3.18) compared to cropland playas (1.70). Average water depth (2008 and 2009) and playa area (2008 only) were best predictors of amphibian species richness. Restored playas provided the most reliable water availability for amphibians in the RWB where 90% of the wetlands have been drained.

B. J. Beas, L. M. Smith, *Wetlands* **34**, 1247–1253 (2014).

***Pristimantis mutabilis* sp. nov. in life. Sub-adult male, MZUTI 2191, photographed in its natural habitat during the night (left; photo by J. M. Guayasamin) and under laboratory conditions during the day (right; photo by Lucas Bustamante).**

Phenotypic plasticity raises questions for taxonomically important traits: A remarkable new Andean rainfrog (Pristimantis) with the ability to change skin texture

Juan M. Guayasamin, Tim Krynak, Katherine Krynak, Jaime Culebras & Carl R. Hutter

We describe a new frog, *Pristimantis mutabilis* sp. nov., from the Andes of Ecuador. Individuals of the new species are remarkable for their ability to change skin texture from tuberculate to almost smooth in a few minutes, being the first documented amphibian species to show such dramatic phenotypic plasticity. The new taxon is assigned to the *P. myersi* group. It differs from other members of its group by body size (adult males 17.2–17.4 mm; adult females 20.9–23.2 mm), arboreal habitat and red flash coloration in females. We document three call types for the new species, which differ through their number of notes and amplitude peaks. The three types are pulsed calls that share a dominant frequency of 3186.9–3445.3 Hz. Surprisingly, we also document similar skin texture plasticity in species (*P. sobetes*) from a different species group, suggesting that this ability might be more common than previously thought. The discovery of these variable species poses challenges to amphibian taxonomists and field biologists, who have traditionally used skin texture and presence/absence of tubercles as important discrete traits in diagnosing and identifying species. Reciprocal monophyly and genetic distances also support the validity of the new species, as it has distances of 15.1–16.3% (12S) and 16.4–18.6% (16S) from the most similar species, *Pristimantis verecundus*. Additionally, each of the two known populations of *Pristimantis mutabilis* are reciprocally monophyletic and exhibit a high genetic distance between them (5.0–6.5%). This pattern is best explained by the presence of a dry valley (Guayllabamba River) that seems to be acting as a dispersal barrier.

J. M. Guayasamin, T. Krynak, K. Krynak, J. Culebras, C. R. Hutter, *Zool. J. Linnean Soc.* **173**, 913–928 (2015). doi: 10.1111/zoj.12222

Various pipe placement in Everglades National Park habitats: (A) Close up of paired tree-placed pipes on pine trees; (B) Single tree-placed pipes on adjacent pine trees in a recently burned forest; (C) Ground-placed pipes in an array in a prairie habitat; (D) Ground-placed pipes in a cypress swamp. Photo: J. H. Waddle.

A review of pipe and bamboo artificial refugia as sampling tools in anuran studies

Brad M. Glorioso & J. Hardin Waddle

The need to capture a sufficient and representative sample of a target population has long been a primary consideration in organismal field studies. The diverse behaviors and life histories of anurans have given rise to a diverse array of capture techniques for sampling frogs and toads, including visual encounter surveys, acoustic detection of calling males, coverboards, pitfall traps, funnel traps and dipnet or minnow traps for tadpoles. Another anuran sampling method, the bamboo or pipe trap, takes advantage of the need of arboreal frogs to find refuge from weather extremes and predators. Artificial refugia created by pipe traps capture treefrogs and other agile climbers often underrepresented in samples using other methods.

Artificial pipe-like refugia have been used

for more than 40 years in anuran studies, and have captured 28 species, primarily (82%) hylid treefrogs. Early pipe-like refugia were made using cut pieces of bamboo in the tropical forests of Puerto Rico, but most recent studies have used synthetic pipes and have occurred primarily in the southeastern United States.

Here, we summarize and evaluate different pipe designs and placement, and discuss the types of studies that have used this technique, including outreach and citizen science applications. We address potential concerns, such as equal capture probability, when using artificial pipe-like refugia, and suggest studies necessary to better interpret the data gained from this technique in anuran studies. Lastly, we provide guidance on best practices when implementing this technique.

B. M. Glorioso, J. H. Waddle. *Herp. Cons. Bio.* **9**(3), 609–625 (2014).

Translocation site for the population of Growling Grass Frog on the northern outskirts of Melbourne, Victoria. Photo: Sally Koehler.

Translocation of the threatened Growling grass frog *Litoria raniformis*: a case study

Sally L. Koehler, Daniel C. Gilmore & David A. Newell

Translocation is occasionally suggested as a last resort strategy for dealing with "unavoidable" loss of Growling Grass Frog *Litoria raniformis* habitat in urbanizing landscapes. However, examples of attempts to translocate an entire population of *L. raniformis* are rare and their success (or lack of success) is poorly documented in the literature. In this study, we detail the translocation of a population of *L. raniformis* from a farm dam being destroyed for residential development to a purpose-built wetland 480 m away.

The population was translocated between November 2010 and May 2011. We used mark-recapture to estimate the number of frogs in the population prior to translocation. Visual counts of *L. raniformis* at the dam indicated a maximum of 39 adult frogs to be present while 355 frogs were marked over the course of a single season (November 2010 to March 2011).

Translocation of 156 frogs and unassisted colonization by 32 frogs resulted in an estimated 70% of adults marked at the dam moving to the wetland and 91% of those remained there for the duration of the translocation study period.

The population and two measures of habitat quality (aquatic vegetation cover and water quality) were monitored for three active seasons post-translocation. Successful breeding was demonstrated for the first year only. A decline in breeding success was attributed to a reduction in habitat quality at the wetland, particularly the loss of submergent and floating vegetation due to the presence of Common Yabby *Cherax destructor*, a species that did not occur originally in the dam. We believe that colonization of the wetland by this crustacean was due to the wetland being constructed on-line. An attempt to control the *C. destructor* population and re-establish the aquatic vegetation was unsuccessful.

We encourage the publication of all successes and failures in future attempts to establish translocated Bell Frog populations. If further experimental translocations have low success rates, then translocations should be reconsidered as a conservation strategy for *L. raniformis* in urbanising landscapes and greater emphasis placed on *in situ* habitat protection.

S. Koehler, D. Gilmore, D. Newell, *Australian Zoologist online* doi: http://dx.doi.org/10.7882/AZ.2014.043 (2014).

A Mountain Coqui *(Eleutherodactylus portoricensis)* at El Yunque National Forest, Puerto Rico. Photo: Alejandro Ríos-Franceschi.

Population declines of Mountain coqui (*Eleutherodactylus portoricensis*) in the Cordillera Central of Puerto Rico

Brittany S. Barker & Alejandro Ríos-Franceschi

The Mountain Coqui (*Eleutherodactylus portoricensis*) is a frog endemic to montane rainforests in the Cordillera Central and Luquillo Mountains of Puerto Rico. Classified as Endangered by the IUCN Red List and as Vulnerable by the Department of Natural and Environmental Resources of Puerto Rico, this species has undergone considerable decline in the Luquillo Mountains. To evaluate the population status of *E. portoricensis* across its entire range, we conducted about 87 hours of surveys at 18 historical localities and 25 additional localities that we considered suitable for this species. We generated occupancy models to estimate the probability of occurrence at surveyed sites and to identify geographic and climatic factors affecting site occupancy. We also constructed a suitability map to visualize population status in relation to the presence of land cover at elevations where the species has been documented, and determined the dates when populations were last detected at historical localities. *Eleutherodactylus portoricensis* was detected at 14 of 43 localities, including 10 of 18 historical localities, but it was not detected at any localities west of Aibonito (western Cordillera Central). Occupancy models estimated the probability of occurrence for localities in the western Cordillera Central as zero. Site occupancy was positively associated with montane cloud forest, and negatively associated with the western Cordillera Central, maximum temperature, and precipitation seasonality. The suitability map suggests that declines have occurred despite the presence of suitable habitat. We suggest upgrading the extinction risk of *E. portoricensis* and potentially developing a captive breeding program for this species.

B. S. Barker, A. Ríos-Franceschi, *Herpetol Conserv Biol.* **9(3)**, 578–589 (2014).

Habitats and amphbians representative of multiple continents. Clockwise, from the upper-left: *Ensatina eschscholtzii, Hyla arborea, Scutiger nyingchiensis, Ptychadena mascareniensis, Atelopus flavescens.* Photos: G. F. Ficetola.

Habitat availability for amphibians and extinction threat: A global analysis

Gentile Francesco Ficetola

Habitat loss and degradation are the factors threatening the largest number of amphibian species, but quantitative measures of habitat availability only exist for a small subset of them. In our study, we evaluated the relationships between habitat availability, extinction risk and drivers of threat for the world's amphibians. We developed deductive habitat suitability models to estimate the extent of suitable

habitat and the proportion of suitable habitat inside the geographic range of amphibians at the global scale, covering species and areas for which little or no high-resolution distribution data are available.

We used information on habitat preferences to develop habitat suitability models at 300 m resolution for 5,363 amphibians species. We built deductive models, by integrating range maps with information on the elevation range and land-cover typologies suitable for each species, obtained from the literature. Model performance was assessed by comparing model output with point localities where species were recorded.

Validation success of models was high (94%), being better for forest specialists and generalists than for open habitat specialists. Generalists had proportionally more habitat than forest or open habitat specialists. The proportion of suitable habitat was lower for species having small geographical ranges, currently listed as Threatened, and for which habitat loss is recognized as a threat. Differences in habitat availability among biogeographical realms were strong, with less habitat available for species from the Saharo-Arabian and Oriental realms.

We then used habitat availability as a surrogate of area of occupancy. Using the IUCN criteria B2 and D2, we identified species having narrow area of occupancy, for which extinction risk is likely underestimated in the current IUCN Red List. Our analysis identified 61 forest species for which the extinction risk may be higher that currently assessed in the Red List, due to limited extent of suitable habitat.

Habitat models can accurately predict amphibian distribution at fine scale, and allow describing biogeographical patterns of habitat availability. The strong relationship between amount of suitable habitat and extinction threat may help the to help the assessment of conservation status in species for which limited information is currently available.

G. F. Ficetola, C. Rondinini, A. Bonardi, D. Baisero, E. Padoa-Schioppa, *Divers. Distrib.* **21**, 302–311 (2015).

Too wet for frogs: Changes in a tropical leaf litter community coincide with La Niña

Mason J. Ryan, Norman J. Scott, Joseph A. Cook, Beatriz Willink, Gerardo Chaves, Federico Bolaños, Adrián García-Rodríguez, Ian M. Latella & Sally E. Koerner

Extreme climatic events such as the El Niño Southern Oscillation profoundly affect many plants and animals, including amphibians, which are strongly negatively affected by drought conditions. How amphibians respond to exceptionally high precipitation as observed in La Niña events, however, remains unclear. We document the correlation between the exceedingly wet 2010–2012 La Niña and community-level changes in a leaf litter frog assemblage in Costa Rica. Relative abundances of species shifted, diversity and plot occupancy decreased, and community composition became homogenized with the onset of La Niña. These aspects remained altered for over 20-months but rebounded to pre-La Niña levels after approximately 12-months. We hypothesize that complex ecological cascades associated with excess moisture caused short-term declines in abundances of species and associated changes in community structure. If additional stressors such as disease or habitat loss are not co-occurring, frog communities can rapidly recover to pre-disturbance levels following severe climatic events.

M. J. Ryan, N. J. Scott, J. A. Cook, B. Willink, *et al.*, *Ecosphere*, **6**(1), 4 (2015). http://dx.doi.org/10.1890/ES14–00352.1

Male of *Psychrophrynella teqta* sp. nov. guarding eggs. Photo: Ignacio De la Riva.

A new species of *Psychrophrynella* (Anura: Craugastoridae) from the Cordillera Real, Department La Paz, Bolivia

Ignacio J. De la Riva & Patricia A. Burrowes

This publication provides a detailed description of a new species of frog in Bolivia, while discussing observations of parental care and conservation concerns associated to disease risk and climate change, in a diverse group of highly endemic amphibians. In 2012, while conducting fieldwork in the Cordillera Real, in the Andes of Department of La Paz, Bolivia, we found a new species of terrestrial, direct-developing frog in the genus *Psychrophrynella*. Individuals of *Psychrophrynella teqta* sp. nov. show a variety of color patterns, but they are distinguished from nearby conspecifics by having bright yellow or red blotches in the dorsum, that are absent in other species. In addition, the new species differs from all species in the genus because the males have a pulsed, versus tonal call. We found males guarding terrestrial egg clutches within refugia consisting of small ditches dug under rocks. The high number of eggs per nest, suggest that perhaps a single male might attend more than one clutch. This small frog, with its unique call and color pattern was found in a remote area of wet sub-paramo, adjacent to a small isolated village. The fact that as other species in the genus, the new species is infected by *Batrachochytrium dendrobatidis* and that its habitat in the Andean highlands is threatened by climate warming and drought, pose a conservation threat upon its discovery.

I. De la Riva, P. A. Burrowes, *Zootaxa* **3887**(3), 459–470 (2014).

***Leptodactylus fallax*. Photo: Benjamin Tapley.**

Meeting ultraviolet B radiation requirements of amphibians in captivity: A case study with mountain chicken frogs (*Leptodactylus fallax*) and general recommendations for pre-release health screening

Benjamin Tapley, Matthew Rendle, Frances M. Baines, Matthias Goetz, Kay Bradfield, David Rood, Javier Lopez, Gerardo Garcia & Andrew Routh

Conservation breeding programs are a tool used to prevent amphibian extinctions. The husbandry requirements of amphibians are complex. Ongoing research is needed to ensure optimal management of those captive-bred animals destined, in particular, for reintroduction. The UV-B and vitamin D_3 requirements of amphibians are largely unknown. Metabolic bone disease has been reported in a number of species. These include the Critically Endangered mountain chicken frog (*Leptodactylus fallax*) reared in captivity on diets supplemented with a high-calcium multivitamin and mineral supplement containing vitamin D_3 but without UV-B provision. Captive-bred *L. fallax* being reared for reintroduction to Montserrat were provided with UV-B radiation from metamorphosis and were fed on insects supplemented with vitamins and minerals. Overlapping heat, light and UV-B gradients were provided, mimicking what we believe best represents the natural situation and thereby facilitated self-

regulation of UV-B exposure. A subset of 10 frogs was periodically radiographed to assess skeletal health. Radiographic bone density and anatomical integrity appeared unremarkable when compared with a wild caught *L. fallax*. In addition to other routine health-screening, we recommend that radiography be performed to a structured schedule on a subset of all captive-bred and reared amphibians to assess skeletal health and to gauge the appropriateness of captive husbandry. We demonstrate here that, through the appropriate provision of a combination of both UV-B radiation and dietary supplementation, *L. fallax* can be bred and reared in captivity with healthy skeletal development.

B. Tapley, M. Endle, F. Baines, M. Goetz, K. Bradfield *et al. Zoo Biol.* **134**, 1,46–52 (2015).

Photo: Pete Oxford.

Using environmental DNA methods to improve detectability in a hellbender (*Cryptobranchus alleganiensis*) monitoring program

Stephen F. Spear, John D. Groves, Lori A. Williams & Lisette P. Waits

Isolation of environmental DNA (eDNA) is becoming a valuable tool for detecting presence of rare or secretive aquatic species. The recent use of quantitative PCR (qPCR) with eDNA sampling presents the possibility of using this method to infer population abundance and status. This approach would be especially useful for species such as the Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*), a declining, secretive, aquatic salamander that requires intense field survey effort to study. In 2012, we conducted eDNA sampling at sites across the range of the species in North Carolina. Our objectives were to assess presence across 61 sites, test for a correlation of abundance and biomass with eDNA estimates at a subset of 23 sites, and sample at multiple spatial and temporal scales in three river systems. Overall, we detected Hellbender eDNA at 33 sites, including all sites with 2012 Hellbender records, 71% of all recent or historic sites with hellbender presence, and at nine sites that lack species occurrence records. We did not find a correlation between eDNA estimates and field survey counts of individuals or biomass. We detected a strong temporal increase in eDNA during the September breeding period, but no consistent evidence of a spatial relationship with eDNA. Overall, our results demonstrate the efficacy of eDNA for detecting Hellbender populations. Furthermore, the potential utility of qPCR to assess population status in hellbenders requires further study and testing, although it may be promising for determining population reproductive status.

S. F. Spear, J. D. Groves, L. A. Williams, L. P. Waits, *Biol. Conserv.* **183**, 38–45 (2015).

Limited influence of local and landscape factors on finescale gene flow in two pond-breeding amphibians

Stephanie S. Coster, Kimberly J. Babbitt, Andrew Cooper & Adrienne I. Kovach

Dispersal and gene flow within animal populations are influenced by the composition and configuration of the landscape. In this study, we evaluated hypotheses about the impact of natural and anthropogenic factors on genetic differentiation in two amphibian species, the Spotted Salamander (*Ambystoma maculatum*) and the Wood Frog (*Lithobates sylvaticus*) in a commercial forest in central Maine. We conducted this analysis at two scales: a local level, focused on factors measured at each breeding pond, and a landscape level, focused on factors measured between ponds. We investigated the effects of a number of environmental factors in six categories including Productivity, Physical, Land Composition, Land Configuration, Isolation and Location. Embryos were sampled from 56 Spotted Salamander breeding ponds and 39 Wood Frog breeding ponds. We used a hierarchical Bayesian approach in the program GESTE at each breeding pond and a random forest algorithm in conjunction with a network analysis between the ponds. We found overall high genetic connectivity across distances up to 17 km for both species and a limited effect of natural and anthropogenic factors on gene flow. We found the null models best-explained patterns of genetic differentiation at a local level and found several factors at the landscape level that weakly influenced gene flow. This research indicates multiscale investigations that incorporate local and landscape factors are valuable for understanding patterns of gene flow. Our findings suggest that dispersal rates in this system are high enough to minimize genetic structuring and that current forestry practices do not significantly impede dispersal.

S. S. Coster, K. J. Babbitt, A. Cooper, A. I. Kovach, *Mol. Ecol.* **24(4),** 742–758 (2015).

Diseases and Toxicology

Batrachochytrium dendrobatidis exposure effects on foraging efficiencies and body size in anuran tadpoles

Shane M. Hanlon, Kyle J. Lynch, Jake Kerby & Matthew J. Parris

Chytridiomycosis, the amphibian disease caused by the pathogenic fungus *Batrachochytrium dendrobatidis* (*Bd*), is fatal to adults of many species. *Bd* is largely sublethal to amphibian larvae; however, it is known to reduce larval, or tadpole, growth rates, with possible long-term effects on population dynamics and fitness. We conducted an experiment to test how *Bd* altered southern Leopard Frog (*Lithobates sphenocephalus*) tadpole mouthpart damage, percentage of food ingested, and subsequent body size. We examined our results using path analyses. We hypothesized that *Bd* would increase mouthpart damage, causing less food to be ingested, and ultimately reduce body size. In our model, *Bd* exposure and increased mouthpart damage significantly reduced food ingested and subsequent body size. However, our study provides evidence against the long-standing hypothesis of mouthpart damage as a pathway for *Bd*-induced reductions in larval group. Here we provide evidence for reduced foraging efficiency (percentage of food ingested) as a mechanism for *Bd*-induced reductions in body size. This work highlights the importance of studying the sublethal effects of Bd on larval amphibians.

S. M. Hanlon, K. J. Lynch, J. Kerby, M. J. Parris. *Dis. Aqua. Org.* **112**, 237–242 (2015).

Phylogeny and differentiation of reptilian and amphibian ranaviruses detected in Europe

Anke C. Stöhr, Alberto López-Bueno, Silvia Blahak, Maria F. Caeiro, Gonçalo M. Rosa, António Pedro Alves de Matos, An Martel, Alí Alejo & Rachel E. Marschang

Ranaviruses in amphibians and fish are considered emerging pathogens and several isolates have been extensively characterized in different studies. Ranaviruses have also been detected in reptiles with increasing frequency, but the role of reptilian hosts is still unclear and only limited sequence data has been provided. In this study, we characterized a number of ranaviruses detected in wild and captive animals in Europe based on sequence data from six genomic regions (major capsid protein (MCP), DNA polymerase (DNApol), ribonucleoside diphosphate reductase alpha and beta subunit-like proteins (RNR-α and -β), viral homolog of the alpha subunit of eukaryotic initiation factor 2, eIF-2α (vIF-2α) genes and microsatellite region). A total of

ten different isolates from reptiles (tortoises, lizards, and a snake) and four ranaviruses from amphibians (anurans, urodeles) were included in the study. Furthermore, the complete genome sequences of three reptilian isolates were determined and a new PCR for rapid classification of the different variants of the genomic arrangement was developed. All ranaviruses showed slight variations on the partial nucleotide sequences from the different genomic regions (92.6–100%). Some very similar isolates could be distinguished by the size of the band from the microsatellite region. Three of the lizard isolates had a truncated vIF-2α gene; the other ranaviruses had full-length genes. In the phylogenetic analyses of concatenated sequences from different genes (3223 nt/10287 aa), the reptilian ranaviruses were often more closely related to amphibian ranaviruses than to each other, and most clustered together with previously detected ranaviruses from the same geographic region of origin. Comparative analyses show that among the closely related amphibian-like ranaviruses (ALRVs) described to date, three recently split and independently evolving distinct genetic groups can be distinguished. These findings underline the wide host range of ranaviruses and the emergence of pathogen pollution via animal trade of ectothermic vertebrates.

A. C. Stöhr, A. López-Bueno, S. Blahak, M. F. Caeiro, G. M. Rosa *et al.*, *PLOS ONE* **10(2)**, e0118633 (2015). doi:10.1371/journal.pone.0118633

Boophis luteus **from Ranomafana National Park. Photo: Molly Bletz.**

Widespread presence of the pathogenic fungus B*atrachochytrium dendrobatidis* in wild amphibian communities in Madagascar

Molly C. Bletz, Gonçalo M. Rosa, Franco Andreone, Elodie A. Courtois, Dirk S. Schmeller, Nirhy H. C. Rabibisoa, Falitiana C. E. Rabemananjara, Liliane Raharivololoniaina, Miguel Vences, Ché Weldon, Devin Edmonds, Christopher J. Raxworthy, Reid N. Harris, Matthew C. Fisher & Angelica Crottini

Amphibian chytridiomycosis, an emerging infectious disease caused by the fungus *Batrachochytrium dendrobatidis* (*Bd*), has been a significant driver of amphibian declines. While globally widespread, *Bd* had not yet been reported from within Madagascar. We document surveys conducted across the country between 2005 and 2014, showing *Bd's* first record in 2010. Subsequently, *Bd* was detected in multiple areas, with prevalence reaching up to 100%. Detection of *Bd* appears to be associated with mid to high elevation sites and to have a seasonal pattern, with greater detectability during the dry season. Lineage-based PCR was performed on a subset of samples. While some did not amplify with any lineage probe, when a positive signal was observed, samples were most similar to the Global Panzootic Lineage (BdGPL). These results may suggest that *Bd* arrived recently, but do not exclude the existence of a previously undetected endemic *Bd* genotype. Representatives of all native anuran families have tested *Bd*-positive, and exposure trials confirm infection by *Bd* is possible. *Bd*'s presence could pose significant threats to Madagascar's unique "megadiverse" amphibians.

Bletz *et al. Sci. Rep.* 5, 8633 (2015).

Effects of salinity and pesticide on sperm activity and oviposition site selection in Green treefrogs, *Hyla cinerea*

Anneke E. Wilder & Allison M. Welch

Increasing salinity in freshwater habitats is an emerging threat in both coastal and inland environments. In addition, pesticides can be harmful to aquatic organisms and have been detected worldwide in aquatic systems. Both of these changes can strongly affect amphibians, as they typically rely on aquatic environments. With external fertilization, the selection of an oviposition site can be crucial for ensuring fertilization success and for providing offspring with a hospitable environment for development. We examined the effects of increased salinity and a commonly applied insecticide, carbaryl, on sperm activity and oviposition site selection of the Green Treefrog (*Hyla cinerea*). Sperm motility and velocity both decreased as salinity concentration increased but showed no effect of carbaryl at concentrations at and above those expected in the environment. Artificial pools with weakly brackish water received less oviposition than did freshwater pools. Carbaryl also inhibited oviposition, but only when pools had been freshly dosed. These findings suggest that increased salinity may negatively affect reproductive success in *H. cinerea* through its effect on sperm function, but that individuals may be able to avoid these costs by selectively ovipositing in freshwater. Although carbaryl did not influence two measures of sperm movement, selective oviposition may help adults to avoid habitats that could be harmful to their developing offspring. Thus, selective oviposition may increase the ability of amphibian populations to persist in degraded habitats, as long as some suitable sites are still available.

A. E. Wilder, A. M. Welch, *Copeia* 2014, 659–667 (2014).

Wood Frog, *Rana sylvatica*, recent metamorph (froglet) raised in captivity from wild-caught tadpoles. Photo: María Forzán, Canadian Wildlife Health Centre. Bar = 1 cm.

Clinical signs, pathology and dose-dependent survival of adult wood frogs, *Rana sylvatica*, inoculated orally with Frog Virus 3 (*Ranavirus* sp, Iridoviridae)

María J. Forzán, Kathleen M. Jones, Raphaël V. Vanderstichel, John Wood, Frederick S. B. Kibenge, Thijs Kuiken, Wytamma Wirth, Ellen Ariel & Pierre-Yves Daoust

Frog virus 3 (FV3) is a pathogen responsible for severe mortalities in amphibian populations in most continents. FV3 is the type species of the genus Ranavirus, in the family *Iridoviridae*, and can cause severe disease and mortalities in fish and reptiles. Our objective was to experimentally infect captive-raised adult Wood Frogs, *Rana sylvatica* [*Lithobates sylvaticus*], with various doses of FV3, and evaluate the clinical signs, gross and microscopic lesions, survival time and possible excretion of virus through skin sheds and feces. The wood frog was chosen as an experimental subject because it is widely spread in North America and can be used as a representative of the so-called true frogs (Ranids), one of the groups most commonly affected by FV3 outbreaks in the wild. Oral dosing with FV3 was successful; median lethal dose (LD_{50}) was $10^{2.93\,(2.42–3.44)}$ pfu for frogs averaging 35 mm in length. Onset of clinical signs occurred 6–14 days post-infection (dpi) (median 11 dpi) and time-to-death 10–14 dpi (median 12 dpi). Each ten-fold increase in virus dose increased the odds of dying 23-fold and accelerated onset of clinical signs and death by approximately 15%. Ranavirus DNA was demonstrated in skin and liver of all frogs that died or were euthanized because of severe clinical signs. Shedding of virus occurred in feces (7–10

dpi; 3–4.5 d before death) and skin sheds (10 dpi; 0–1.5 d before death) of some frogs dead from infection. Most common lesions were dermal erosion and hemorrhages, hematopoietic necrosis in bone marrow, kidney, spleen and liver, necrosis in renal glomeruli and in tongue, gastrointestinal tract and urinary bladder mucosa. Presence of ranavirus in lesions was confirmed by immunohistochemistry. Intracytoplasmic inclusion bodies (probably viral) were present in the bone marrow and the epithelia of the oral cavity, gastrointestinal tract, renal tubules and urinary bladder. Our work describes a Ranavirus-Wood Frog model and provides estimates that can be incorporated into ranavirus disease ecology models.

M. J. Forzán, K. M. Jones, R. V. Vanderstichel, J. Wood, F. S. B. Kibenge *et al.*, *J. General Virology* (in press)

Leukocyte profiles and helminths of Northern Leopard Frogs, *Rana pipiens*: Location, location, location

Dave Shutler, Andrée D. Gendron, Myriam Rondeau & David J. Marcogliese

Globally, amphibians face a variety of anthropogenic stresses that include exposure to contaminants such as agricultural pesticides. Pesticides may negatively affect amphibian immune systems, concomitantly increasing susceptibility to parasitism. We quantified nematodes and evaluated leukocyte profiles of Northern Leopard Frogs (*Rana pipiens* Schreber, 1782) collected from five wetlands in southwestern Quebec, Canada, that spanned a gradient of pesticide exposure. Three taxa of nematode parasites (*Rhabdias ranae* Walton, 1929, genus *Oswaldocruzia* Travassos, 1917 and genus *Strongyloides* Grassi, 1879) were sufficiently numerous for detailed evaluation. When all frogs were pooled, frog size was negatively correlated with nematode species richness, abundances of each of the three nematode species and densities of three different leukocytes. When all frogs were pooled, there was strong evidence of both negative and positive associations between pairs of parasite species. However, none of the previous relationships was significant within wetlands. Our results reveal strong spatial organization of amphibian–parasite communities and illustrate the importance of controlling for sampling locale in evaluating host–parasite associations. Finally, although several response variables varied significantly among wetlands, causes of this variation did not appear to be related to variation in nematode parasitism or pesticide exposure.

D. Shutler, A. D. Gendron, M. Rondeau, D. J. Marcogliese, *Can. J. Zool.* **93**, 41–49 (2015).

Acute toxic effects of the herbicide formulation and the active ingredient used in cycloxydim-tolerant maize cultivation on embryos and larvae of the African clawed frog, *Xenopus laevis*

Norman Wagner, Stefan Lötters, Michael Veith & Bruno Viertel

Because most genetically engineered crops are still awaiting approval in Europe, herbicide-resistant maize cultivation is here mainly restricted on cycloxydim-tolerant maize hybrids. We conducted the first amphibian-toxicological trails with the complementary herbicide formulation Focus® Ultra and its active ingredient cycloxydim. We used embryos and early-stage larvae of the African Clawed Frog (*Xenopus laevis*) as anuran model organism. Our results indicate that the herbicide formulation is significantly more toxic than cycloxydim alone. Therefore, it is suggested that the added substances either solely or in a synergistic action with the active ingredient are responsible for adverse effects. Furthermore, the formulation was found to be moderately toxic to embryos but highly toxic to early larvae, *i.e.*, the effects were life-stage-specific. Our results suggest that tests with the fish species Rainbow Trout (*Oncorhynchus mykiss*) are not in all cases appropriate to assess the risk in aquatically developing anurans because LC50 values are for rainbow trout more than 50- to 20-fold higher than for early *X. laevis* larvae. However, based on expected environmental concentrations for surface waters, there is apparently a large margin of safety in field use if buffer strips between the farm land and the breeding sites are regarded.

N. Wagner, S. Lötters, M. Veith, and B. Viertel, *Bull. Environ. Contam. Toxicol.* 94, 412–418 (2015).

Prevalence of the amphibian pathogen *Batrachochytrium dendrobatidis* in Eastern hellbenders (*Cryptobranchus a. alleganiensis*) in Western North Carolina, USA

Lori A. Williams & John D. Groves

Due to recent documented declines of Eastern Hellbender (*Cryptobranchus a. alleganiensis*) populations, in 2008–2012 we surveyed both wild-caught and captive hellbenders in Western North Carolina for the prevalence of a common amphibian pathogen, *Batrachochytrium dendrobatidis* (*Bd*). We sampled 165 wild and 15 captive animals of North Carolina origin. We collected morphometric data from hellbenders, noted the presence of any physical anomalies or injuries, and recorded water temperature and site elevation. We found *Bd* to be widespread in western North Carolina waterways and in all river basins and sub-basins sampled. *Batrachochytrium dendrobatidis* prevalence was 27.9% for wild-caught animals and 26.7% for captives. Adult female hellbenders had a significantly higher prevalence of *Bd* (38.8%) than adult males (19.7%; $P = 0.0207$). All age classes tested positive for *Bd*, including gilled larvae. From examining hellbender body condition, we found no physical evidence of acute infection or compromised immunity due to disease. There was no significant relationship between the presence of *Bd* and physical injuries or anomalies ($P = 0.1196$). We also found that water temperature ($P = 0.5038$) and elevation ($P = 0.5100$) were not significant for predicting the presence of *Bd*. Ours is the first report of *Bd* for Eastern hellbenders in North Carolina. Although it does not appear that hellbenders in North Carolina are in crisis due to disease, future monitoring and surveillance efforts within populations should continue, particularly across a variety of habitat disturbance regimes.

L. A. Williams and J. D. Groves, *Herpetol. Conserv. Biol.* **9(3)**, 454–467 (2014).

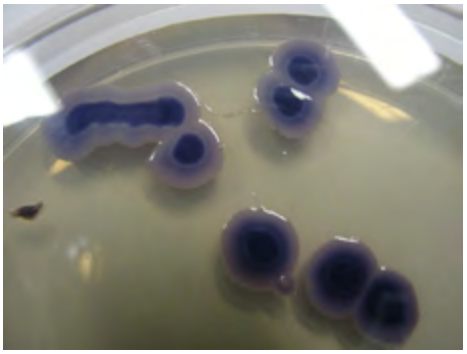
Differential efficiency among DNA extraction methods influence detection of the amphibian pathogen *Batrachochytrium dendrobatidis*

Molly C. Bletz, Eria A. Rebollar & Reid N. Harris

Chytridiomycosis, caused by the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*), is responsible for massive declines and extinctions of amphibians worldwide. The most common method for detecting *Bd* is quantitative polymerase chain reaction (qPCR). qPCR is a highly sensitive detection technique, but its ability to determine the presence and accurately quantify the amount of Bd is also contingent on the efficiency of the DNA extraction method used prior to PCR. Using qPCR, we compared the extraction efficiency of three different extraction methods commonly used for *Bd* detection across a range of zoospore quantities: PrepMan Ultra Reagent, Qiagen DNeasy Blood and Tissue Kit, and Mobio PowerSoil DNA Isolation Kit. We show that not all extraction methods led to successful detection of *Bd* for the low zoospore quantities and that there was variation in the estimated zoospore equivalents among the methods, which demonstrates that these methods have different extraction efficiencies. These results highlight the importance of considering the extraction method when comparing across studies. The Qiagen DNeasy kit had the highest efficiency. We also show that replicated estimates of less than one zoospore can result from known zoospore concentrations; therefore, such

results should be considered when obtained from field data. Additionally, we discuss the implications of our findings for interpreting previous studies and for conducting future *Bd* surveys. It is imperative to use the most efficient DNA extraction method in tandem with the highly sensitive qPCR technique in order to accurately diagnose the presence of Bd as well as other pathogens.

M. C. Bletz, E. A. Rebollar, R. N Harris. *Dis Aquat Org*. 113, 1–8 (2015).



Isolate culture of bacteria *Janthinobacterium lividum* from the skin of *Gastrotheca riobambae*, adult and metamorphs stages. Photo: Jorge C. Bresciano.

Variation in the presence of anti-*Batrachochytrium dendrobatidis* bacteria of amphibians across life stages and elevations in Ecuador

Jorge C. Bresciano, Carmen A. Salvador, Cesar Paz-y-Mino, Angela M. Parody-Merino, Jaime Bosch & Douglas C. Woodhams.

Amphibian populations are decreasing worldwide due to a variety of factors. In South America, the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) is linked to many population declines. The pathogenic effect of *Bd* on amphibians can be inhibited by specific bacteria present on host skin. This symbiotic association allows some amphibians to resist development of the disease chytridiomycosis. Here we aimed (1) to determine for the first time if specific anti-*Bd* bacteria are present on amphibians in the Andes of Ecuador, (2) to monitor anti-*Bd* bacteria across developmental stages in a focal amphibian, the Andean Marsupial Tree Frog, *Gastrotheca riobambae*, that deposits larvae in aquatic habitats, and (3) to compare *Bd* presence associated with host assemblages including 10 species at sites ranging in biogeography from Amazonian rainforest (450 masl) to Andes montane rainforest (3200 masl). We sampled and identified skin-associated bacteria of frogs in the field using swabs and a novel methodology of aerobic counting plates, and a combination of morphological, biochemical, and molecular identification techniques. The following anti-*Bd* bacteria were identified and found to be shared among several hosts at high-elevation sites where *Bd* was present at a prevalence of 32.5%: *Janthinobacterium lividum*, *Pseudomonas fluorescens* and *Serratia* sp. *Bd* was detected in *Gastrotheca spp*. and not detected in the lowlands (sites below 1,000 m asl). In *G. riobambae*, recognized *Bd*-resistant bacteria start to be present at the metamorphic stage. Overall bacterial abundance was significantly higher post-metamorphosis and on species sampled at lower elevations.

Bresciano *et al*., *EcoHealth*. 11 Feb. (2015). [Epub ahead of print]. doi: 10.1007/s10393-015-1010-y

Presence of Amphibian Chytrid Fungus (*Batrachochytrium dendrobatidis*) in Rainwater Suggests Aerial Dispersal is Possible

Jonathan E. Kolby, Sara D. Ramirez, Lee Berger, Dale W. Griffin, Merlijn Jocque, & Lee F. Skerratt

Global spread of the pathogenic amphibian chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) may involve dispersal mechanisms not previously explored. Weather systems accompanied by strong wind and rainfall have been known to assist the dispersal of microbes pathogenic to plants and animals, and we considered a similar phenomenon might occur with *Bd*. We investigated this concept by sampling rainwater from 20 precipitation events for the presence of *Bd* in Cusuco National Park, Honduras: a site where high *Bd* prevalence was previously detected in stream-associated amphibians. Quantitative PCR analysis confirmed the presence of *Bd* in rainwater in one (5%) of the weather events sampled, although viability cannot be ascertained from molecular presence alone. The source of the *Bd* and distance that the contaminated rainwater traveled could not be determined; however, this collection site was located approximately 600 m from the nearest observed perennial river by straight-line aerial distance. Although our results suggest atmospheric *Bd* dispersal is uncommon and unpredictable, even occasional short-distance aerial transport could considerably expand the taxonomic diversity of amphibians vulnerable to exposure and at risk of decline, including terrestrial and arboreal species that are not associated with permanent water bodies.

J. E. Kolby, S. D. Ramirez, L. Berger, D. W. Griffin, M. Jocque, L. F. Skerratt, *Aerobiologia*. doi: 10.1007/s10453-015-9374–6 (2015).

Call for recent publication abstracts

If you would like to include an abstract summary from a recent publication in this section of *FrogLog* (*FL*), please follow proper formatting conventions and style for your submission (*e.g.*, note the most recent issue of *FL* available); email: froglog@amphibians.org. We also encourage all authors of recent publications to inform Professor Tim Halliday (formerly DAPTF International Director at: tim.r.halliday@gmail.com) of their publication in order for it to be referenced on the AmpibiaWeb latest papers page. The full list of latest papers from AmphibiaWeb is also included in every edition of *FrogLog* following the Recent Publications abstract section.

General Announcements

Events

The following information can be found at: http://www.amphibians.org/meetings

April 2014

11–12 Amphibian Conservation Research Symposium 2015 University of Cambridge, Cambridge, UK

May 2014

30–1 June The Third International Symposium on Ranaviruses University of Florida in Gainesville, FL, USA

July 2014

13–6 The Association for Tropical Biology and Conservation – International, Honolulu, Hawaii

August 2014

2–6 The Society for Conservation Biology (and the International Congress for Conservation Biology), Monteplier, France

September 2014

7–12 Brazilian Herpetological Congress 2015, Fundacao de apoio da Universidade do Rio Grande Do Sul

Internships & Employment

Aquatic Field Technician, Sequoia Ecological Consulting, Inc.

Monterey, CA (Posted to PARC 03/26/15, Open Until Filled)

Arthropod / Poison Frog Volunteer field/ lab assistant needed

La Selva Biological Research Station in northeastern Costa Rica (Posted to PARC 03/18/15, Closing May 1)

FT Seasonal Field Technician (Amphibians/Fish)

Trask River in northwestern Oregon, Oregon State University (Posted to PARC 03/18/15, Closing April 6)

Salamander Field Technician

Ohio - Vinton County: Vinton Furnace Experimental Forest and Zaleski State Forest (Posted to PARC 03/13/15, Open Until Filled)

Penn State Academic Assistants, Desert Tortoise, Mojave Desert

Henderson, NV / Barstow, CA (Posted to PARC 01/18/15, No Closing Date Provided)

Native and Invasive Reptile Data Management Intern (Multiple)

Department of Wildlife Ecology and Conservation at the University of Florida Fort Lauderdale Research and Education Center, Fort Lauderdale, FL (Posted to PARC 01/08/15, Deadline for applications is 6 weeks before corresponding start date (Multiple))

Rock Iguana Volunteer Field Assistants

Hispaniola (Posted to PARC 03/01/15, Open Until Filled)

Full-Time Amphibian Department Zookeeper

Detroit, MI (Posted to PARC 12/02/14, No Closing Date Provided)

MS Graduate Opportunity: SUNY-ESF

Syracuse, NY (Posted to PARC 11/17/14, 12/15/14)

Chief of the Grand Canyon Monitoring and Research Center

Flagstaff, AZ (Posted to PARC 11/17/14, 12/3/14)

Graduate Assistantship on Bog Turtle Conservation Planning, Virginia Tech Department of Fisheries and Wildlife Sciences

Blacksburg, VA (Posted to PARC 11/10/14, No Closing Date Provided)

Ph.D. graduate assistantship available in population dynamics, ecology, and behavior of amphibians in a fire-dependent system

Blacksburg, VA (Posted to PARC 11/10/14, No Closing Date Provided)

Research Manager, Davidson College Herpetology Laboratory

Davidson, NC (Posted to PARC 11/10/14, Open Until Filled)

Zilla Marketing Brand Manager

Franklin, WI (Posted to PARC 11/04/14, Open Until Filled)

M.S. Assistantship, Amphibian Genetics

Western Kentucky University, Bowling Green, KY (Posted to PARC 10/03/14, Open Until Filled)

Research Manager, Brown Treesnake Research Project

Guam, Mariana Islands (Posted to PARC 06/05/14, Open Until Filled)

Funding Opportunities

The Amphibian Survival Alliance is pleased to announce an open call for seed grant applications. Seed grants are normally provided in amounts ranging from USD $500-$1,000 and are designed to help kick start projects or allow teams to try new innovative approaches to address conservation, research and education challenges. **Link**

The Leapfrog Conservation Fund has been created specifically to support the creation of new reserves for important amphibian habitat, or the expansion of existing reserves through local organizations. If your organization is working toward the protection of critical habitat for threatened amphibian species, we would love to hear from you. **Link**

The following information is kindly provided by the Terra Viva Grants Directory, for more information please visit: http://www.terravivagrants.org/

April

Zoological Society of London (ZSL) -- Animal Photography Prize 2015. The ZSL's Animal Photography Prize encourages entries from professional, amateur, and young photographers worldwide. The competition seeks to promote conservation, science, and education through photographs that inspire, challenge, examine, and capture the essence of animals. There is a registration fee of £15 for adults, and £5 for young persons under age 18. The contest will award cash prizes. The closing date for entries is 01 April 2015. Link

Netherlands Organization for International Cooperation in Higher Education (NUFFIC) -- MENA Scholarship Program. The Middle East and North Africa (MENA) Scholarship Program offers scholarships to professionals from ten MENA countries for short courses of two to twelve weeks in the Netherlands. Subject areas include agriculture, environment, and many others. The program invites applications from professionals up to age 45 who are nationals of and working in one of the following countries: Algeria, Egypt, Iraq, Jordan, Lebanon, Libya, Morocco, Oman,

Syria, and Tunisia. The application period closes 07 April 2015. **Link**

Energy and Environment Partnership for Southern and East Africa -- 11th Call for Proposals. The EEP for Southern and East Africa announces a new call for projects in renewable energy. The focus is on promising local private projects that are close to commercial maturity, but need a limited amount of bridging finance. Projects should be implemented in one or more of the following countries: Botswana, Burundi, Kenya, Lesotho, Mozambique, Namibia, Rwanda, Seychelles, South Africa, Swaziland, Tanzania, Uganda, or Zambia. Grants range from €200 thousand to €1 million, depending on co-financing shares. The submission deadline for concept notes is 13 April 2015. **Link**

African Leadership Academy -- Anzisha Prize for Young Innovators 2015. The Anzisha Prize is awarded to young innovators ages 15-22 in Africa who develop and implement innovative solutions to challenges facing their communities. Past awards include innovations in energy, agriculture, and ecological consumer products. The grand prize winners will share prizes worth US$75 thousand. The closing date for nominations and applications (English, French, Arabic) is 15 April 2015. **Link**

U.S. Fish and Wildlife Service -- Conservation in Central America. The USFWS "Wildlife Without Borders" includes a regional program for Central America. The aims are to reduce agricultural encroachment; conserve jaguar populations; improve wildlife security; and develop conservation capacity in the region. The announcement identifies the key landscapes of Central America for conservation activities. Regional (transnational) projects may include activities in the Dominican Republic and Colombia if they are clearly linked to work carried out in Central America. Grants will range from US$10 thousand to US$50 thousand. The deadline for applications is 15 April 2015. **Link**

Lawrence Foundation -- Grants for Environment. The Lawrence Foundation makes grants to U.S. nonprofit organizations for projects in environment, education, and other themes. There is no restriction on the geographical area where grant activities can be implemented. Average grant size is over US$10 thousand. The two deadlines for applications are 30 April and 01 November of each year. **Link**

May

Conservation Action Research Network -- Congo Basin Grant Program. The Congo Basin Grant Program provides competitive research grants of up to US$5 thousand for African graduate students and early-career professionals working in the areas of biodiversity, conservation, and environmental sustainability in the Congo Basin. Applicants should be 35 years of age or younger, and reside in one of the following countries: Cameroon, Central African Republic, Congo, Dem Rep Congo, Equatorial Guinea, Gabon, or Uganda. The deadline for applications (English, French) is 01 May 2015. **Link**

EUROPARC Federation -- Study Visits at Protected Areas in Europe and Eurasia. The EUROPARC Federation partners with the Alfred Toepfer Stiftung to award the Alfred Toepfer Natural Heritage Scholarships promoting international cooperation within the European dimension of protected area management. Grants of €3 thousand allow young conservationists to make study visits to one or more protected areas external to their home countries. In addition to western Europe, participating countries include Albania, Armenia, Azerbaijan, Bosnia & Herzegovina, Georgia, Macedonia, Moldova, Montenegro, Russia, Serbia, Turkey, and Ukraine. The deadline for applications is 08 May 2015. **Link**

Kurita Water and Environment Foundation – Water Research Grants 2015. The KWEF makes grants in Asian countries for water research at universities, colleges, and research institutes. Research topics include technologies to conserve and restore rivers, lakes, and other water resources – among other themes. Grants are up to 300 thousand yen per project per year. Applicants should be under age 40. Each applicant needs to provide a recommendation letter by a Japanese researcher who resides in Japan or in the researcher's home country. The application period is 01 April through 15 May 2015. **Link**

European Commission (EC) -- Biotechnology for Bioenergy in MERCOSUR. The EC will fund BIOTECH II to support the MERCOSUR countries of Argentina, Brazil, Paraguay, and Uruguay to develop innovative biotechnology in the areas of health and bioenergy. Grants are €200 thousand to €300 thousand, varying with cost shares. The program is open to nonprofit organizations in the EU, the MERCOSUR countries, and qualified international organizations. Proposals should include at least one participating organization or contractor in each of the MERCOSUR countries, and at least one in the EU. Reference EuropeAid/136457/ID/ACT/Multi. The deadline for applications is 22 May 2015. **Link**

European Commission (EC) -- Conservation and Climate Action in Turkey. The EC calls for proposals to promote cooperation between civil society in Turkey and the EU Member States through civil society dialogue in the areas of nature conservation and climate action, where civil society dialogue is particularly valuable for Turkey's successful accession to the EU. The program is open to nonprofit NGOs in Turkey and the EU countries. Applicants in Turkey must have at least one eligible co-applicant from the EU Member States, and applicants in EU Member States must have at least one eligible co-applicant from Turkey. Grants will range from €50 thousand to €150 thousand, varying with cost shares. Reference EuropeAid/136801/ID/ACT/TR. The deadline for applications is 25 May 2015. **Link**

University for Peace -- Masters Programs in Environment, Development, and Peace 2015-2016. The masters programs at UPEACE are open to applicants worldwide. Themes include environmental security and governance (including a specialization in climate change), sustainable natural resources management, and several others. The university offers partial tuition waivers in some cases. The application deadline is 30 May 2015. **Link**

June

Riverbanks Zoo and Garden -- International Wildlife Conservation. The Riverbanks Zoo and Garden (USA) makes grants for field conservation; habitat management; conservation education; ex situ captive breeding; animal health and welfare; and other themes in wildlife research and conservation. Grants generally range from US$1 thousand to US$5 thousand. The application deadlines are 01 June and 01 December. **Link**

July

NextWorldNow Community Investments-- Call for Proposals 2015. NextWorldNow is committed to working with community leaders who are solving difficult social problems. This includes social problems related to water, sanitation, deforestation, smallholder productivity, and food security -- among other subject areas. For 2015, NextWorldNow will invest up to US$10 thousand in grants or interest-free loans in one or more projects. The application deadline is 01 July 2015. **Link**

August

Western Bird Banding Association (WBBA) -- Small Grants for Bird Research and Monitoring in the Americas. The WBBA makes two grants each year, one in bird research and the other in bird monitoring, for projects in the New World (i.e., the Americas). Applications are open to individuals and organizations. Grants are US$1 thousand. Applications are due by 01 August of each year. **Link**

Fulbright Scholar Program -- Core Program for U.S. Faculty and Professionals 2016-2017. The Fulbright Scholar Program invites applications from U.S. scholars for research, teaching, and creative arts in an international context. Eligibility criteria include U.S. citizenship and a PhD or equivalent professional degree. The available openings include several in the developing world in subjects related to agriculture, environmental and biological sciences, geography, and others. The deadline for applications is 03 August 2015. **Link**

U.S. Agency for International Development (USAID) -- Conservation Partnerships in Brazil. USAID aims to create new partnerships in Brazil's Amazon region to support biodiversity conservation; protection of indigenous lands and natural resources; and applied biodiversity science, technology, and innovation. Potential partners in Brazil and the U.S. include foundations, NGOs, faith-based organizations, private businesses, business and trade associations, international organizations, colleges and universities, and government agencies. USAID anticipates making two or three awards with total funding of US$2.5 million over five years. Funding Opportunity APS-512-15-000001. The deadline for concept papers (Portuguese, English) is 19 August 2015. **Link**

September

American Orchid Society -- Annual Conservation Awards. The American Orchid Society sponsors the Conservation Recognition Awards to honor outstanding work in orchid conservation. One prize focuses on conservation in North America; the other has no geographical limitations. Prizes are US$500. The closing date for nominations is 01 September each year. **Link**

U.S. National Science Foundation -- Geography and Spatial Sciences Program 2015. The NSF funds an annual competition for research on the geographic distributions and interactions of human, physical, and biotic systems on the Earth's surface. Projects on a variety of topics (U.S. and international) qualify for support if they offer promise of enhancing geographical knowledge, concepts, theories, methods, and their application to societal problems and concerns. Past projects include several in the world's developing countries related to environmental disasters, waste management, marine spatial planning, wildlife conservation, and others. The deadline for full proposals is 03 September 2015. **Link**

Danish Ministry of Foreign Affairs -- Grants for Development Research 2016. Through the Danish Fellowship Center, Denmark's Ministry of Foreign Affairs announces funding in support of development research. Applicants in Denmark choose "North Driven" for research and capacity building in the priority countries of Danish international assistance. Applicants in Ghana, Nepal, and Tanzania choose "South Driven." Themes in "South Driven" include climate-smart agriculture and environmentally sustainable solutions, among others. In both programs, the deadline for Phase I applications is 04 September 2015. **Link**

Philip Henman Trust -- Grants for Development Projects. The Philip Henman Trust makes grants to UK-based development organizations, with few restrictions on thematic areas. Support is normally between £3 thousand and £5 thousand per year, with a maximum total of £25 thousand. The closing date for applications is 10 September each year. Link

Zayed International Prize for the Environment -- 7th Cycle. The Zayed International Prize for the Environment offers cash prizes to recognize and promote pioneering contributions in environment and sustainable development worldwide. The prize categories are: (i) Global leadership in environment and sustainable development, US$500 thousand; (ii) Scientific/ technological achievements in environment, US$250 thousand; (iii) Environmental action leading to positive change in society, US$200 thousand; and (iv) Young scientists award for environmental sustainability, US$50 thousand. All four categories are open to individuals, organizations, partnerships, or projects. Organizations can be governmental, non-governmental, community based, or private enterprise. The deadline for nominations is 30 September 2015. Link

FrogLog Schedule

January — Special Topical Edition
April — The Americas
July — Africa, West Asia, Madagascar, Mediterranean and Europe
October — Asia, Russia and Oceania

Instructions to Authors

Background

FrogLog has been one of the leading amphibian conservation community newsletters since the early 1990's. Over the years it has been affiliated with different groups but has always strived to help inform the community. In 2005 *FrogLog* became the official newsletter of the IUCN SSC Amphibian Specialist Group and is produced on a quarterly basis.

As the ASG's newsletter members are encouraged to contribute to *FrogLog*'s content and direction. To aid in this process each edition of *FrogLog* focuses on one of the six broad geographical zones identified by the ASG. The publication schedule is as follows:

- January—Special Topical Edition
- April—The Americas
- July—Africa, West Asia, Madagascar, Mediterranean and Europe
- October—Asia, Russia and Oceania

FrogLog invites contributions of research, reviews on current management and conservation issues, methods or techniques papers and, editorials. We also actively encourage submissions describing the current activities relating to projects and academic institutions in order to help inform the community as to the general state of current research and conservation activities.

Publication

FrogLog is published online at: www.amphibians.org and is Open Access.

Review

All contributions should ideally be channeled through Regional ASG Chairs, the details for which can be found at http://www.amphibians.org/asg/members/. If for some reason this cannot be done, contributions will be reviewed by at least one individual within the ASG. *FrogLog* is not a peer-reviewed publication and the onus for submitting accurate information remains with the authors.

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Candace M. Hansen-Hendrikx: cmhansen@amphibians.org

Editorial Committee

Candace M. Hansen-Hendrikx
Craig Hassapakis
Lindsay Renick Mayer

Additional reviewers will be requested as required.

Submission of Manuscripts

Manuscripts can only be received as electronic files. Text should be submitted in MS Word format and may contain tables, but figures should be sent as a separate attachment where possible. All documents should be sent to froglog@amphibians.org. Each file should be labeled in a style that illustrates clear association, i.e., authors_name_ms and authors_name _figure1.

Guidelines for Authors

All manuscripts must be written in Standard US English. For example, "colour" should be spelled "color."

Title

Titles should ideally be no more than 15 words.

Authors

Authors names should be written in full as follows: By James P. Lewis & Robin D. Moore

Main body of text

Use Georgia 11-point font. Genus and species names should be in italics as should the abbreviation for *Batrachochytrium dendrobatidis*, *Bd*. Suggested headings include Acknowledgements, Author Details and References and Notes.

Author details

Author details may be provided, including affiliations and contact details.

Figures

Figures should be numbered and include brief, concise legends. Where photographs or illustrations are used please state whom the image should be credited to, e.g., Photo: James P. Lewis. Graphics should preferably be submitted in tiff or jpeg format in the highest possible quality. Resolution should be at least 300 dpi at the final size.

Tables

Tables may be included within the text file and should be numbered and include brief, precise legends.

Citation of literature

FrogLog uses a numbering system for references and notes. This allows explanatory or more detailed notes to be included with the references. Journal names are abbreviated using common abbreviations to save space.

Journals/Periodicals

1. E. Recuero, J. Cruzado-Cortés, G. Parra-Olea, K. R. Zamundio, *Ann. Zool. Fenn.* **47**, 223 (2010).

Books

2. J. Gupta, N. van der Grijp, Eds., Mainstreaming Climate Change in Development Cooperation (Cambrdige Univ. Press, Cambridge, UK, 2010).

Technical reports

3. G.B. Shaw, Practical uses of litmus paper in Möbius strips (Tech. Rep. CUCS-29-82, Columbia Univ., New York, 1982).

Paper presented at a meeting

4. M. Konishi, paper presented at the 14th Annual Meeting of the Society for Neuroscience, Anaheim, CA, 10 October 1984.

Published Online Only

5. N. H. Sleep, Geochem. *Geophys. Geosyst.*, 10, Q11010 (2009): DOI:10.1029/2009GC002702.

Web site

6. National Oceanic and Atmospheric Administration, Beaufort Wind Scale, http://www.spc.noaa.gov/faq/tornado/beaufort.html (2012).

SPECIAL NOTE: Use only one space after all punctuation marks (this includes only one space after "periods" at the end of sentences).

Further examples and details can be found on our web site at: www.amphibians.org/froglog/guidelines/

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Boophis viridis, Madagascar. Photo: Robin Moore.