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FrogLog

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

INSIDE

Plectrohyla dasypus. Photo by: Jonathan Kolby.

Amazing Amphibians

Stories of Success

FrogLog

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Editorial

Amphibian conservation is about more than just avoiding extinction or talking about what is going wrong—it is about motivating others. It is about shining the spotlight on projects that are working and thereby providing a model to copy and implement elsewhere. This edition of *FrogLog* is about these amphibian conservation success stories. These stories range from protecting, restoring or creating habitats to captive breeding and releases, as well as the implementation of conservation education and awareness programs.

This edition of *FrogLog* is also about the power of collaboration. Our ability to achieve these conservation successes depends on our willingness to collaborate with others. The issues facing amphibians are dynamic and interrelated. It is very rare that you have a single organization or individual with the resources and the ability to single-handedly and effectively tackle the combination of these issues.

Organizations with a vested interest in conservation are constantly understaffed and underfunded. The ability to pool these resources together through collaborative efforts becomes an extremely powerful and effective conservation tool. Each organization or individual that comes together on a project brings their own unique set of diverse skills and strengths to the table that when combined, can achieve great things.

George Bernard Shaw wrote, "If you have an apple and I have an apple and we exchange these apples then you and I will still each have one apple. But if you have an idea and I have an idea and we exchange these ideas, then each of us will have two ideas."

As the new IUCN quadrennium gets underway, it is my pleasure and honour to extend a warm welcome to the over 400 members of the Amphibian Specialist Group. Each of you brings to the group a diverse range of skills, approaches and backgrounds, which makes the ASG a truly global authority on amphibian research and conservation.

Over the course of the next four years we look forward to working together with all of you to develop even more amphibian conservation success stories. And we encourage you to share your success stories with us—we want to hear from you and we want to help share your stories with others.

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ASG Program Officer

FrogLog

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Membership Update

Thank you to all those who registered with the ASG during the 2013-2016 quadrennium registration period. In total we received 468 membership registration forms from members in approximately 80 countries. There are now 41 Regional Groups within the ASG with several new groups looking to be established in the next few months.

The information collected in the membership registration form will enable us to help identify the priorities for the ASG during the course of this quadrennium and an annual report to be published later this year will outline these priorities.

During the course of the coming months members of the ASG will be invited to join working groups designed to help tackle specific challenges relating to amphibian conservation.

If you are interested in joining the ASG or think your name has been accidentally omitted from the list below, or if you know of someone who is not on the list and should be, please feel free to contact ASG Program Officer, James Lewis (jplewis@amphibians.org).

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How To Engage In Policy Without Staff?

Dr. Jaime García-Moreno

The Amphibian Survival Alliance (ASA) is an initiative with rather limited human resources to oversee all the different issues that need to be considered when we are talking of Amphibian Conservation. It is therefore important that we make good use of the network and platforms provided by our partners.

In late February there were a couple of examples of how we try to approach this. On February 21st BirdLife and IUCN co-hosted the event "Biodiversity's Ticking Time Bomb" at the European Parliament in Brussels to highlight the issue of Invasive Alien Species ahead of a legislative EU proposal, and Dr. Piero Genovesi, chair of IUCN SSC Invasive Alien Species Specialist Group, was very supportive of including the case of the chytrid fungus and its impact on amphibians as part of his presentation during this event. This is important as a reminder to the EU authorities that the Bd fungus needs to be seen as an invasive alien species; such a denomination will guarantee a direct link to EU's Biodiversity Strategy, which in turn should allow support for continued research and monitoring of the disease. *Bd* is already present in Europe, and it is important to remember that ¾ of European amphibian species are endemic to the continent.

A few days later, also in Brussels, ASA participated in a meeting convened by the RACE project (Risk Assessment of Chytridiomycosis to European amphibian biodiversity). This was a much smaller meeting involving representatives from the EU Commission (Directive Generals of Environment and of Health and Consumers -SANCO) and the World Organisation for Animal Health, and was a space to dialogue with policy makers and for all of us present to understand what it takes for an issue to be established in the policy agenda at the EU level. In addition to the chytrid problem, this was a good forum to highlight the problem arising from the lack of a specific code for amphibians in the Harmonized System of the World Customs Organisation, which makes it very difficult to track amphibian trade around the world, let alone monitoring the chytrid fungus.

Together with RACE coordinators, we sent a message to Mr. Ladislav Miko, European Commission's Deputy Director General for the Food Chain at the Directive General for Health and Consumers, to inform him about the general amphibian declines throughout the world, the role of the chytrid fungus epidemic, the role of trade, and the opportunity that exists for the European Commission to help assist amphibian trade to adopt 'best principles' practice and to build further on the network that the RACE project has started in order to continue monitoring the impact of the chytrid in Europe.

We urge you to let us know whenever opportunities like the ones described here arise. We are happy to feed you then with additional information whenever possible in order to present the larger picture of a given issue. The more all the partners of the alliance present such issues to their different audiences, the more impact the ASA will have in the end.

Dr. Jaime García-Moreno, Amphibian Survival Alliance. Email: Jaime.GarciaMoreno@iucn.org

We Want To Help Your Conservation Project

Hispaniola Giant TreeFrog. Photo: Robin Moore

The ASG aims to promote amphibian conservation projects, either in place or needed, from around the world. We hope that by promoting these projects both awareness and funding will increase for amphibians around the world. If you would like to include a project on amphibians.org please follow the below guidelines and send details to ASG Program Officer James Lewis. These guidelines have been developed to help you write a profile that is concise yet containing enough information that potential supporters get a good sense of the project aims and outcomes.

Project summary (max 350 words – shorter is better): brief description of the project highlighting the area intended to be protected, some key features, including species (if the target species list is large please indicate number of taxa under each IUCN Red List Category, i.e. 4 CR, 7 EN amphibians), habitats, and communities, also include names of any implementation partners, and give a clear idea of the project goals.

Target Species: why the area is important for biodiversity, using the Red List as the ultimate metric – i.e., always include Threatened species in addition to other species that may be of interest. Mention the amphibians present and their level of threat, other animals and plants that may benefit from your project, and other benefits and services of the area (e.g. ecosystem services, religious value of the site, community resource).

Partners: who is doing the work? Why is each institution well positioned to do be involved in the project? And how will they be coming together in a coordinated fashion? Proposal reviewers often look for a strong collaborative project that demonstrates involvement from multiple local organizations, community groups and strong governmental support where possible so if this is reflected in your project please make this clear.

Activities: what exactly will be done (research land tenure, buy a piece of land, restore habitat, hire and train rangers, develop conservation strategy, establish a reserve, educate local communities, etc)? Try to mention specific activities, how they will help the project, and which partner(s) is/are likely to be involved. A clear indication of how these actions will positively impact the Red List status of the species is important.

Products and outcomes: what will be achieved at the end of the project? Try to provide a good idea of expected results, ideally linking to the activities mentioned and also synergies with other conservation activities taking place in the region.

Budget: what are the costs of the properties proposed for purchase? Please indicate a breakdown of all costs (including legal fees), any necessary consultancies regarding evaluation of land tenure or other issues, and a timeline including such details as deadline for downpayment, payment schedule, and proposed dates of legal transfer from current owner (please indicate current owner specifically and, if possible, copy of land title) to your organization. In order to clearly distinguish land purchase costs from other requested funds, please organize your budget under the following sub-headings: (1) Land Purchase and Reserve Designation, (2) Land Management and Restoration, (3) Research and Monitoring, (4) Ecotourism and Business Infrastructure, (5) Community Outreach and Training.

Images: Good photographs of the site and/or target species to illustrate the proposal are always welcome.

FrogLog and Alytes

By Franco Andreone

The International Society for the Study and Conservation of the Amphibians (ISSCA) is publishing *Alytes*, the only journal dedicated to all the aspects of the amphibian biology. Its aim is to devote more attention to the challenges regarding amphibian conservation, and for this Alytes' editorial board is seeking papers that report original observations or reviews. The journal is currently under a process of format change and in the forthcoming months it will be re-launched to gain a wider audience. More recently, a special issue on amphibian conservation edited by A. Angulo and F. Andreone, was published and entitled *Bridging the gap between science and policy in amphibian conservation*. In the process of making the content available to a wider public, we are now offering two contributions which can be downloaded for free. Those interested in contributing to *Alytes* can contact me at franco.andreone@gmail.com

You can download the most recent publications here.

Regional ASG Update—Peru

By Rudolf von May

Peru's Amphibian Specialist Group (ASG) currently comprises 20 members who are affiliated with several Peruvian universities, natural history museums and non-profit conservation organizations working throughout the country. The group recently held an election to select two of its members to serve as national Co-chairs for the next IUCN quadrennium (2013-2016). As the outgoing national Chair, and on behalf of the other members of Peru's ASG, I would like to welcome Ariadne Angulo, the new national Chair, and Giussepe Gagliardi-Urrutia, the new Co-chair.

It is an honor to have both Ariadne and Giussepe take the lead of Peru's ASG and guide the group's activities during this new IUCN quadrennium. Ariadne Angulo, Ph.D., has over 19 years of involvement with amphibians, including aspects related to amphibian conservation, bioacoustics, systematics, ecology and evolutionary biology. She is currently the IUCN SSC Amphibian Red List Authority Coordinator and is in charge of overseeing the maintenance of the global amphibian assessment database for the IUCN Red List of Threatened Species™. The elected Co-chair, Giussepe Gagliardi-Urrutia, works for the Amazon Biodiversity Research Program at the Instituto de Investigaciones de la Amazonia Peruana (IIAP), based in Iquitos, Peru. He has extensive experience conducting biodiversity inventories throughout the country, with a special emphasis on Amazonian amphibians and reptiles. In the past decade, both Ariadne and Giussepe have actively participated in numerous research projects in collaboration with other colleagues. Their combined expertise brings new perspectives, and will help improve the group's efforts to develop the scientific basis needed to understand and conserve amphibian species in Peru. One example of this commitment is their immediate response. Within a few days of being elected, Ariadne and Giussepe proposed that a Peru-focused assessment initiative be the pilot project to launch an on-line forum to facilitate amphibian assessments (http://www.amphibians.org/redlist/forum/peru/; http://www.inaturalist.org/projects/peru-amphibian-redlist-assessment-forum). This forum was developed as a collaboration between iNaturalist (www.inaturalist.org), the ASG and the Amphibian RLA, and will facilitate the exchange of information and provide feedback to the Amphibian RLA on IUCN Red List assessments. We encourage the herpetological community working in Peru to take a look at this site and, if available, contribute with species-specific information.

***Rhinella yanachaga*. Photo: Ariadne Angulo.**

Another, exciting news is that 18 new amphibian species known from Peru were described during 2012. Of these, 10 belong to the terrestrial-breeding frog family Strabomantidae (*Oreobates amarakaeri*, *O. gemcare*, *O. machiguenga*; *Phrynopus badius*, *P. curator*, *P. interstictus*, *P. vestigiatus*; *Pristimantis bustamante*, *P. mariaelenae*, *P. stipa*), four are tree frogs (Hylidae: *Dendropsophus frosti*; *Osteocephalus cannatellai*, *O. germani*, *O. vilmae*), and each of the remaining species belongs to a different family (Allophrynidae: *Allophryne resplendens*; Bufonidae: *Rhaebo ecuatoriensis*; Centrolenidae: *Centrolene sabini*; Telmatobiidae: *Telmatobius mendelsoni*).

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Amphibian conservation husbandry

3rd - 7th June 2013

The ACH course immensely helped to improve my knowledge and skills of amphibian husbandry.
ACH course participant, 2009

The Amphibian Conservation Husbandry (ACH) course is a five day intensive course held at Durrell's headquarters in Jersey, designed to expose participants to the latest theory and practice of amphibian husbandry. Participants will be equipped with the skills and knowledge to establish and manage captive populations and breeding programmes for some of the world's most threatened amphibians.

Who is the course for?

The ACH course is designed specifically for curators, zoo keepers, private breeders and others interested in the captive management of amphibians.

What is the course content?

The course involves lectures, guided tours and plenty of practical sessions to try out newly learnt skills. The course is co-directed by leading amphibian experts from Durrell Wildlife Conservation Trust and additional external experts from around the world.

This course is officially endorsed by Amphibian Ark and Amphibian Survival Alliance.

The course will include the following topics:

- Understanding the natural history of your species
- Water quality, testing and filtration
- Temperature, lighting and UV
- Enclosure design and decoration
- Nutrition and breeding live foods
- Healthcare, disease and biosecurity
- Breeding difficult species
- Supporting *in-situ* conservation

What is the cost?

The course fee is £750 (discounted to £600 if paid at least 8 weeks in advance). Optional full board on-site accommodation is available for £210 for six nights.

amphibian ark

For further information please contact:
+44 (0)1534 860037 or **academy@durrell.org**

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durrell wildlife conservation trust durrell.org
an international charity saving species from extinction

2012 Amphibian Conservation Accomplishments in AZA Institutions

By Shelly Grow

Golden mantella tadpole. Photo: Brian Freiermuth - Steinhart Aquarium at The California Academy of Sciences.

Zoos and aquariums accredited by the Association of Zoos and Aquariums (AZA) have made a commitment to the conservation of amphibians and specifically to 1) increase the capacity of accredited zoos and aquariums to respond to threats facing amphibians, 2) create and sustain assurance populations of threatened amphibians, and 3) increase public awareness of, and engagement in, amphibian conservation. With the support of directors, curators, keepers and partners, AZA-accredited zoos and aquariums experienced conservation progress and successes in 2012, including in the areas of citizen science, assurance populations, conservation breeding, field work and research.

Citizen Science

FrogWatch USA™, AZA's frog and toad monitoring citizen science program, experienced another year of growth. Nine in-person chapter coordinator trainings were held throughout the country, providing instruction to over 120 prospective chapter coordinators, including staff from more than 30 AZA institutions, state biologists, parks and recreation department staff, college and university faculty, master naturalists and others. These trainings promoted growth of the FrogWatch USA chapter network which reached almost 70 chapters by the end of 2012. Local chapters recruit, train and support volunteers, and are the primary means of growing the public's participation.

Species Survival Plan® Programs

The Wyoming toad (*Anaxyrus baxteri*) and the Puerto Rican crested toad (*Peltophryne lemur*) Species Survival Plan® (SSP) programs celebrated active field conservation seasons. Ten Wyoming toad SSP breeding facilities produced over 13,000 tadpoles and toadlets for reintroduction at two locations. Field surveys, led by the U.S. Fish and Wildlife Service and including SSP participants, searched for undiscovered wild populations of toads and evaluated habitats as potential release sites. A soft-release pilot study placed SSP-bred tadpoles in protective enclosures at a lake shoreline site where a declining population had not been augmented since 2003, and moved the toadlets upland into corral structures after metamorphosis. Photo recognition software identified individual toads and documented their survival, growth, and movements at a Safe Harbor site, offering a new monitoring tool appropriate for toads too small for the injection of PIT tags.

Ten Puerto Rican crested toad SSP breeding facilities sent almost 27,850 tadpoles to Puerto Rico for release at three—including two new—sites, bringing the total number of reintroduction sites to five. Disney's Animal Kingdom (Florida) documented the recovery effort and engaged a film crew to record the breeding process at Disney, document the transport and release of tadpoles in Puerto Rico, interview recovery group partners on the island and film an educational workshop about the crested toad at an elementary school. The Fort Worth Zoo (Texas) brought two school teachers to Puerto Rico to learn about the crested toad, so they may incorporate conservation methods into their teaching.

Assurance Populations

AZA-accredited facilities contributed significant resources to creating and maintaining assurance populations for species of conservation concern. The Vancouver Aquarium (British Columbia, Canada) maintains an assurance population consisting of 25 genetic groups for the Northern leopard frog (*Lithobates pipiens*) Recovery Team in British Columbia where the species is listed as Endangered. Nine of these genetic groups were added in 2012, and the aquarium will breed and release frogs in coming years. An assurance population of Oregon spotted frogs (*Rana pretiosa*), a species present at only four wetlands in British Columbia, is also maintained at the Aquarium and currently contains 49 genetic strains.

Wyoming toadlets for release. Photo: Val Hornyack, Toledo Zoo.

The Philadelphia Zoo (Pennsylvania) maintains assurance populations of nine Endangered species of the genus *Eleutherodactylus* that are endemic to Haiti. The populations now number 1,500 individuals and in 2012, the Zoo celebrated the first successful hatch of the F2 generation of *Eleutherodactylus bakeri.* A Haitian delegation visited the Zoo to see the colony and incorporated it into a long-term conservation strategy for Haitian amphibians.

To support the establishment of assurance populations within amphibians' range countries, the Toledo Zoo (Ohio) created partnerships in the Dominican Republic and helped build a state-of-the-art amphibian conservation facility at the Dominican National Zoo which now houses several native threatened amphibian species. The Toledo Zoo also supported a workshop to help amphibian biologists from the Antilles and Caribbean, Central America and South America build capacity for breeding and maintaining endemic frog species.

The Saint Louis Zoo's Ron Goellner Center for Hellbender Conservation (Missouri), Missouri Department of Conservation and

***Eleutherodactylus inoptatus* juvenile. Photo: Carlos Martinez Rivera, Philadelphia Zoo.**

U.S. Fish and Wildlife Service announced that Ozark hellbenders (*Cryptobranchus alleganiensis bishopi*) had bred outside the wild for only the second time when eight female hellbenders laid eggs in artificial nest boxes located in simulated stream habitats at the Zoo. All three of the Zoo's hellbender populations have now produced eggs, including a population that had been maintained indoors for eight years. By late 2012, the eggs had hatched into more than 2,500 larvae. Five years of research at the Nashville Zoo (Tennessee) yielded results when two Eastern hellbenders (*Cryptobranchus alleganiensis alleganiensis*) hatched from eggs that were produced and artificially fertilized from Zoo animals. This assisted reproductive technology technique will be refined until the species is reliably reproduced.

Hellbender. Photo: Christian Sperka, Nashville Zoo.

Commitments have been made to breed several other species at AZA-accredited zoos and aquariums. The Toledo Zoo announced the first breeding of four species of woodland salamanders: the Green salamander (*Aneides aeneus*); the Midland mud salamander (*Pseudotriton montanus diastictus*); the Northern two-lined salamander (*Eurycea bislineata*); and the Long-tailed salamander (*Eurycea longicauda*), bringing the number of plethodontid salamanders that have reproduced in the Zoo's salamander program to ten. The National Mississippi River Museum and Aquarium (Iowa) successfully bred the Laotian newt (*Paramesotriton laoensis*) in both 2011 and 2012, making it the only AZA-accredited facility breeding this species. The San Antonio Zoo (Texas) became the first zoo outside Mexico to reproduce the Endangered Mexican burrowing frog (*Smilisca dentata*). The Steinhart Aquarium at the California Academy of Sciences (California) began developing techniques to raise Critically Endangered Golden mantellas (*Mantella aurantiaca*) on a large scale while managing for genetic diversity, reared over 300 individuals and sent more than 100 to other AZA institutions.

Reintroductions

Zoos and aquariums participated in both long-term assurance, as well as short-term head-starting, programs. In 2012, scientists from the Columbus Zoo and Aquarium (Ohio) and the Ohio Department of Natural Resources made the first release of zoo-reared Eastern hellbenders (*Cryptobranchus alleganiensis alleganiensis*) in an effort to re-establish multiple self-sustaining hellbender populations in Ohio. The repatriation sites were in a stream where hellbenders historically occurred but that became polluted. The stream has since recovered and is now one of the highest quality waterways in the state. Prior to the release, zoo veterinarians performed health screenings and surgically implanted radio-transmitters. The partnership includes the Oglebay Good Zoo (West Virginia), which collected the eggs in 2007 that matured into the released animals, the Toledo Zoo, who began housing wild-collected eggs in a biosecure room in 2012 and several others.

The Oregon spotted frog (*Rana pretiosa*) Reintroduction Project received AZA's 2012 Top Honors for North American Conservation. Since 2008, four facilities, Northwest Trek Wildlife Park, Woodland Park Zoo and Cedar Creek Correctional Center in Washington and the Oregon Zoo (Oregon), have reared and released over 5,400 Oregon spotted frogs with the support of multiple government and private organizations.

In partnership with the World Bank, the Tanzanian government, several Tanzanian universities, and international agencies, the Wildlife Conservation Society's Bronx Zoo (New York) and the Toledo Zoo each reintroduced 1,000 Kihansi spray toads (*Nectophrynoides asperginis*) they had bred to Tanzania's Kihansi Gorge in October 2012, along with 500 animals bred in Tanzania. Although listed as Extinct in the Wild, reintroductions will continue and project partners have expressed optimism about the future of the toad in the wild.

Research

Accredited zoos and aquariums participated in research to develop new conservation tools. Since 2007, the North Carolina Zoological Park, in partnership with the North Carolina Wildlife Resources Commission, has conducted Eastern hellbender (*Cryptobranchus alleganiensis alleganiensis*) surveys to determine the conservation status of this species. To increase the number of waterways that can be surveyed each year, the project team began utilizing a

Laotian newt. Photo; Abby Urban, National Mississippi River Museum and Aquarium.

new technique to determine the species' presence or absence in a waterway. Water samples are collected and pumped through filters designed to trap bits of DNA from sloughed off skin cells and excreted feces and urine, known as "aquatic environmental DNA", or "eDNA". Research will investigate: whether the method can provide information on population densities, the effects of stream flow rates on quantifying eDNA, and the effectiveness of using samples to assess the reproductive status of hellbenders. Another research team is developing methods to sample wetlands for the presence of amphibian species of conservation concern by isolating and amplifying DNA from water samples of enclosures holding Flatwood's salamanders (*Ambystoma bishopi*), Dusky gopher frogs (*Lithobates sevosus*), Striped newts (*Notophthalmus perstriatus*) and Gulf-coast waterdogs (*Necturus cf beyeri*) supplied by the Jacksonville Zoo and Gardens.

Other field based research focused on species distribution and declines. The Calgary Zoological Society's Centre for Conservation Research (Alberta, Canada) continued participating in a long term Northern leopard frog (*Lithobates pipiens*) population monitoring project, with the goals of improving monitoring techniques, determining if, or to what extent, leopard frogs are declining in the province, identifying key habitats and determining the factors influencing occupancy dynamics. The Smithsonian National Zoological Park (DC and Virginia) partnered with the U.S. Geological Survey to investigate habitat suitability and microhabitat preferences of the federally-listed Endangered Shenandoah salamander (*Plethodon shenandoah*) in response to shrinking habitat and climate change. The Philadelphia Zoo initiated a program with Haitian partners to understand how frogs in Haiti and the Dominican Republic cope with deforestation and habitat degradation. This information provides government and private stakeholders with

Photo: Carlos Martinez Rivera, Philadelphia Zoo.

the tools necessary to protect amphibian habitat and will inform plans to reintroduce amphibians from the Zoo's *ex-situ* conservation program. The Denver Zoo (Colorado) broadened their Lake Titicaca frog (*Telmatobius culeus*) conservation effort in Peru to include pilot studies for obtaining population estimates using distance sampling methodology, while opportunistically collecting DNA samples and swabbing for *Batrachochytrium dendrobatidis* (*Bd*). The Zoo offered a workshop on snorkeling, husbandry and surveying for staff members of the Lake Titicaca National Reserve and the General Directorate of Forestry and Wildlife and continued its community outreach by sponsoring Frog Day Celebrations in Lima and at the lake. Community awareness is one goal of a long-term monitoring project by the Milwaukee County and Racine Zoos (Wisconsin) on behalf of the Grenada frog (*Pristimantis euphronides*) and a cinematographer accompanied project partners to Grenada to produce short films about the island's reptiles and amphibians for the Grenada Information Service (GIS) television station.

Eleutherodactylus audanti. **Photo: Carlos Martinez Rivera, Philadelphia Zoo.**

Animals housed at zoos and aquariums may help generate knowledge applicable to threats in the wild. The Smithsonian National Zoological Park (DC) supported research on the genetic response of Lowland leopard frogs (*Lithobates yavapaiensis*) to *Bd*, with the goal of clarifying host responses to the fungus. The Zoo opened an Appalachian salamander research lab to host a post-doctoral candidate studying how temperature and water quality influence stress indicators, metabolism and immune function of Eastern hellbenders (*Cryptobranchus alleganiensis alleganiensis*. The Zoo continued to support Panamanian golden frog (*Atelopus zeteki*) conservation with an assurance population and worked with a PhD candidate from Panama investigating isolation and cryopreservation of *Atelopus* sperm. The Zoo also provided support to the Panama Amphibian Rescue and Conservation (PARC) project and assurance colonies in El Valle and Gamboa.

Conclusion

AZA's Amphibian Taxon Advisory Group (ATAG) developed resources to support engagement in amphibian conservation and released the second edition of the *Amphibian Husbandry Resource Guide*. This expanded edition includes chapters that have been significantly updated, as well as new chapters on Assisted Reproduction Technology and Amphibian Data Entry. Both this document, a Spanish translation of the first edition, and other ATAG-developed amphibian conservation resources are available on AZA's website (www.aza.org) to promote further engagement in amphibian conservation. AZA congratulates its accredited zoos and aquariums for their amphibian conservation efforts and dedication, while recognizing that conservation challenges continue to grow.

Acknowledgements

This summary is adapted from contributions submitted to the Association of Zoos and Aquariums for inclusion in the *Amphibian Conservation 2012—Highlights and Accomplishments* report, available on AZA's website (www.aza.org/amphibian-news/).

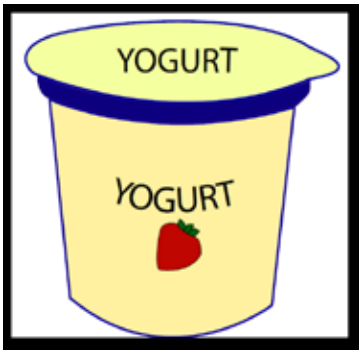
Lake Titcaca frog. Photo: Tom Weaver, Denver Zoo.

Proactive Conservation of Malagasy Frogs: Development of Probiotic Conservation Strategies

By Molly Bletz & Reid Harris

Amphibians are threatened by the fungal disease chytridiomycosis, which has already caused declines and extinctions around the world. Chytridiomycosis is caused by a pathogenic fungus *Batrachochytrium dendrobatidis* (*Bd*) and is the largest disease threat to biodiversity at the present time (*1*). *Bd* is currently absent from Madagascar, but it will likely arrive at any time (*2, 3*). The map shows the areas where *Bd* will thrive, which is also where most of the frogs live. *Bd* has spread rapidly around the world, so we predict that it is only a matter of time before the pathogen reaches Madagascar where it will likely decimate the diverse frog fauna. It is imperative to consider a prevention and mitigation strategy now in order to prevent catastrophic declines and extinctions in Madagascar like those seen other tropical areas. In Central America, species like the Panamanian Golden Frog can no longer be found due to *Bd*.

Here is a frog (*Rana muscosa*) in a probiotic bath. Bath treatment prevented population extinction as a result of Bd arrival in California and it can work in Madagascar. Photo by: Vance Vredenburg, San Francisco State University.



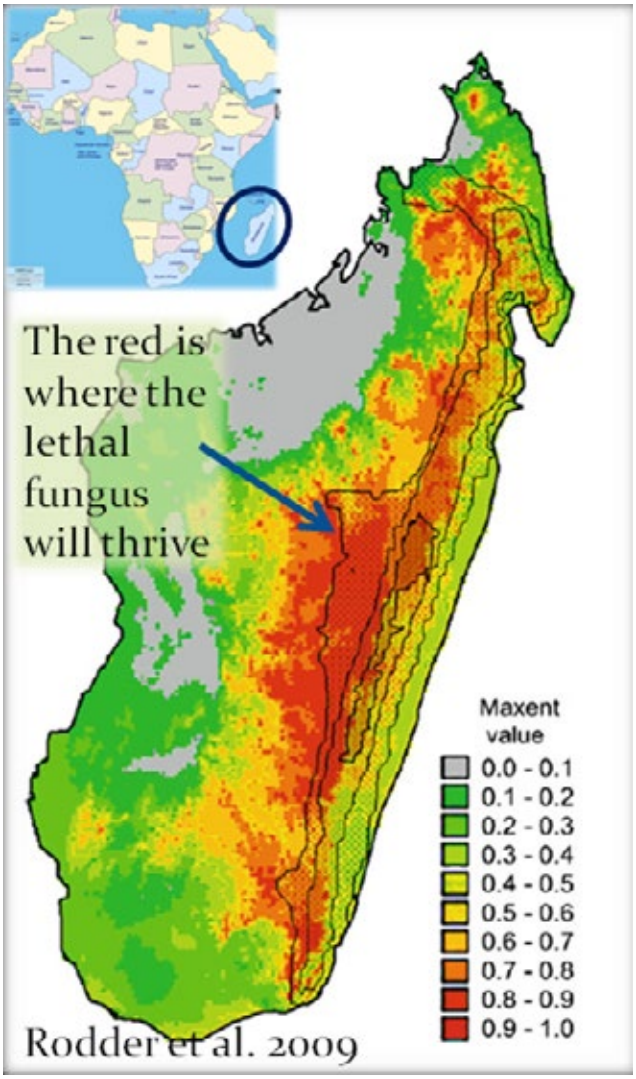
What can be done?

We consume yogurt to restore a beneficial microbial community in our bodies and protect us from disease. The same principle works for frogs. The addition of beneficial bacteria to a frog's skin is a promising disease mitigation strategy based on growing evidence that microbes are an important defense for plants and animals. The addition of *locally-occurring* protective bacteria to amphibians has effectively prevented disease in laboratory trials and two recent field trials (*4-7*).

Our goal is to find effective probiotics in order to implement probiotic conservation strategies that will help preserve the remarkable amphibian biodiversity in Madagascar. We will use our recently developed six stage filtering protocol to identify effective probiotics (*8*). We have developed two strategies:

- Strategy One (Species-specific): find probiotics for Critically Endangered species in Madagascar, such as *Mantella aurantiaca*, which can be administered by individual probiotic baths.
- Strategy Two (Community-based): find probiotics that can protect amphibian communities by treating breeding ponds.

**We stress that bioaugmentation approaches must use bacteria found on amphibians in the local environment to improve success and minimize biosafety concerns.*



What will it take to conserve these frogs?

Filtering Protocol: Six Phases

- **Microbe sampling:** Collect bacteria from phylogenetically diverse amphibian species.
- ***Bd*-Inhibition Assays:** Filter out bacterial candidates that do not inhibit *Bd*.
- **Environmental Persistence Trials:** Determine which candidates can persist in the environment to enable environmental treatment for community-based strategies.
  **Only for community-based treatment strategies.*
- **Host Colonization & Persistence Trials:** Determine which of the remaining candidates are able to establish effectively on the host amphibian.
- **Clinical Trials:** Ensure that bacterial candidates can inhibit Bd and reduce the effects of chytridiomycosis on amphibians.
- **Small-scale Field Trials:** Measure the ability of a probiotic candidate to be successful in nature.

EFFECTIVE PROBIOTICS

**For more information see Bletz et al., 2013.*

Frogs of Madagascar. Top row (left to right): *Platypelis spp.*, *Boophis madagascariensis*, *Gephyromantis redimitus*; Bottom row (left to right): *Boophis luteus*, *Mantella aurantiaca*, *Mantella crocea*. Photos by: Devin Edmonds, Association Mitsinjo.

What are the conservation implications?

Probiotic disease mitigation for wildlife is a new conservation frontier. Significant progress has been made in the field of amphibian probiotics as a possible tool for mitigating amphibian chytridiomycosis both in the laboratory and field (*8*). Applying these techniques before the arrival of *Bd* is still a novel concept but is well worth investigating in Madagascar. Probiotic conservation strategies offer the possibility of conserving species while keeping them in their native habitats. This is important because the capacity does not exist to house all the species that need to be protected from *Bd*. Finding effective frog probiotics has the potential to prevent catastrophic declines of frogs in Madagascar. Once *Bd* is detected in a region of Madagascar, our goal is to have a bank of local probiotics in place that can stem the devastation of the spreading epidemic. This is important for the ecosystem and for the people of Madagascar.

People Involved: Molly Bletz & Reid Harris, *James Madison University, Amphibian microbial ecologists*

Local Collaborators: Falitiana Rabemananjara, *Chytrid Emergency Cell, Coordinator*

Franco Andreone & Ché Weldon *ASG-Madagascar & Chytrid Emergency Cell, Member*

Devin Edmonds *Association Mitsinjo, Amphibian Conservation Director*

For more information about this project or if you are interested in donating to this conservation effort please visit: frogprobiotics.org

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Amazing Amphibians

Anura

The Spiny Giant Frog

The Spike-thumb Frog

The Dusky Gopher Frog

The Hellbender

Amazing Amphibians is an initiative born out of a joint desire to share the incredible diversity of amphibians with the world. Biodiversity is the backbone of all life on earth and through this initiative we hope to celebrate some of the amazing amphibians around us, promote the fantastic work taking place to protect them and highlight the areas we still need to work on.

Each Monday a new Amazing Amphibian will be posted online for you to share on your website, social media network and through email. We hope that you will enjoy this initiative and help spread the word about just how Amazing Amphibians really are!

If your organization would like to be involved with the Amazing Amphibians program please email: amazing@amphibians.org.

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The Spiny Giant Frog

The Spiny Giant Frog, *Eleutherodactylus nortoni*, is listed as 'Critically Endangered' on the IUCN Red List of Threatened Species™. It is found in the montane forests of south-western Haiti and the Dominican Republic.

The primary threat to this frog throughout its range is habitat destruction due to extraction of wood for charcoal production and agriculture, which is ongoing even in the protected areas of the Tiburon Peninsula of Haiti and Massif de la Selle. Similarly, deforestation is taking place at the only known locality of the Spiny Giant Frog in the Dominican Republic as a result of mining, charcoaling and agriculture.

Although the Spiny Giant Frog occurs in two Haitian national parks, Macaya and Morne La Visite, there is no management of these areas for conservation and the habitat continues to be destroyed. This species is also known from the Dominican national park Sierra de Bahoruco, which is better managed than those in Haiti; however, degradation of the habitat within this park's limits also continues. Strengthening the management of these protected areas is essential for the survival of this frog, as is maintenance of the remaining habitat.

Submit your observations of this species to the Global Amphibian BioBlitz on iNaturalist and they will appear on this map.

Learn more about this species on Amphibiaweb.

The Hellbender

The Hellbender, *Cryptobranchus alleganiensis*, is listed as 'Near Threatened' on the IUCN Red List of Threatened Species™. It is found in the United States in large areas of Kentucky, Tennessee, and Pennsylvania, as well as in smaller areas of the surrounding states. Its distribution seems to follow the Appalachian Mountains until Eastern New York. There are two subspecies of Hellbenders, the Ozarks Hellbender (*Cryptobranchus alleganiensis bishopi*) and the Eastern Hellbender (*Cryptobranchus alleganiensis alleganiensis*). Both can grow to be almost a meter long and are known for being the largest salamander in North America.

The primary threat to Hellbenders is degradation of habitat, as it has little tolerance for environmental change in their preferred habitat of clear, fast-flowing rocky rivers and streams. These include chemical and thermal pollution of water, construction of dams, and activities that cause sedimentation in nest sites. Some fishermen believe the Hellbender destroys their game fish, and thus kill them. Over-collecting of Hellbenders as live animals has also been a serious threat in some areas.

The Hellbender is well protected in certain areas, such as national forests, national parks, and other public lands. There are captive breeding programs for both subspecies with the Saint Louis Zoo successfully hatching over 1,000 Ozarks Hellbender this past winter and the Buffalo Zoo working with State Fish and Game to breed and release Eastern Hellbender larvae. Possible future conservation efforts could include educating fishermen about the species, as well as cessation of activities that cause sedimentation in their native rivers. A recent study of Hellbender genetics showed low genetic diversity within populations possibly due to recent reductions in population sizes, so conservation should also focus on preserving the remaining genetic diversity that does exist in each population.

Submit your observations of this species to iNaturalist and they will appear on this map.

Learn more about this species on Amphibiaweb.

Endangered Species Recovery course

8th - 19th July 2013

The training covers a specialised, complex area, which can only be delivered by an organisation that has proven success in this field, such as Durrell.
Course participant, 2007

Our famous Endangered Species Recovery (ESR) course provides a thorough introduction to the issues and practical skills involved in saving species from extinction. You will be encouraged to develop a critical understanding of biodiversity conservation and the issues it raises.

Who is the course for?
This course is perfectly suited for those wanting an up to date introduction to wildlife conservation and has also proved invaluable to those considering a career move into conservation, and for students or graduates wishing to improve their knowledge.

Previous ESR course participants have gone on to run field conservation programmes in places such as Sumatra and Hawaii and to obtain senior positions in the zoo community, to name just a few.

What is the course content?
The course gives an introduction to issues such as the value of and threats to biodiversity, planning and implementing species recovery programmes, conservation genetics, captive species management, and community conservation. Participants will also be introduced to a wide range of practical research skills. Further details are available upon request.

Rather than just providing theory, this course draws upon Durrell's proven practical experience of restoring endangered species. The course will include current case studies of conservation work by Durrell on some of the most critically endangered primates, birds, reptiles and amphibians.

Who is running the course?
The course is delivered by many of Durrell's own conservation specialists and a number of additional internationally renowned external conservation experts.

Where will the course be run?
The course will run at Durrell Conservation Academy, Durrell Wildlife Conservation Trust, Jersey, British Channel Islands.

What is the cost?
The course fee is £1400 (discounted to £1120 if paid at least 8 weeks in advance). Optional full board on-site accommodation can be provided at an additional cost of £420.

For further information please contact:
+44 (0)1534 860037 or **academy@durrell.org**

Durrell Conservation Academy
Durrell Wildlife Conservation Trust
Les Augrès Manor, La Profonde Rue
Trinity, Jersey, JE3 5BP

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an international charity saving species from extinction

Evidence for Amphibian Conservation: A Global Synopsis

By Rebecca Smith

If you're involved in practical amphibian conservation, you probably have to make frequent decisions about how to manage the habitat or animals you are trying to conserve. For example is it better to control fish in ponds or to create new breeding sites? Is it worth trying to boost population numbers with captive bred animals? Conservationists ought to be sharing experience and information on a global scale to help others make such decisions. But research has shown that more often they use advice from immediate colleagues, local experts or their own experience to inform decisions.

Substantial amounts of scientific knowledge and experience about conservation actions are being overlooked, either because they are within the scientific literature, which can be hard to get hold of or to understand, or because they are retained within small local networks.

The Conservation Evidence project at the University of Cambridge is part of a global effort to change this. It aims to make scientific knowledge about what works and what doesn't work in conservation freely available and accessible. It also provides a way for conservationists to document and globally disseminate the effects of actions they have tested themselves.

Our website, www.ConservationEvidence.com, summarises and brings together evidence about the effects of conservation management for both habitats and species. It is a reliable information resource designed to support decisions about how to maintain and restore global biodiversity. Evidence is freely available through a searchable database, an open access journal and a series of synopses.

We have already produced synopses of evidence for bird conservation, wild bee conservation and wildlife conservation in European farmland. Nine other synopses are currently underway. One is for amphibian conservation, funded by Synchronicity Earth.

Over four hundred studies included

We have listed over 90 possible conservation interventions for amphibians, anywhere in the world, and we are now summarising evidence for the effectiveness of each. The synopsis will include summaries of around 400 studies, all of which have monitored the outcomes of one or more specific conservation actions. Most look directly at amphibian numbers, diversity, breeding success or health. For three of the interventions, we have found over 50 studies monitoring their effects.

Over 200 of the studies summarised for the amphibian synopsis were based in the Americas. For example, the effectiveness of the restoration or creation of wetlands was monitored in 26 studies, culverts in 14 and captive breeding or rearing in 40 studies. Forest management practices such as selective logging or leaving woody debris were investigated in 32 studies and prescribed fires in 18. Awareness raising projects are also included in the synopsis. For example a project in Mexico found that training local boatmen about native amphibian biology and conservation increased their knowledge and income and increased visitor awareness of axolotl conservation (Bride et al. 2008)

For a number of interventions we have found no evidence of whether or not they are effective for amphibians. Despite our search effort it is possible that some evidence has been missed. However, it is also likely that the effects of many conservation projects have not been monitored, or if they have that results have not been made widely available. This highlights the need for conservation projects to include monitoring and the dissemination of results.

Have you written a paper or report describing the effects of a management intervention to conserve amphibians? Have you tried a novel technique for the conservation of an amphibian species? It could be anything from protecting, restoring or creating habitat to translocating species or educating the public. If the intervention was directly tested and its effects monitored quantitatively, we would like to include your evidence in our synopsis.

Are your results not published yet, or have you tested conservation interventions for something other than amphibians? If so the Conservation Evidence Journal is an open access journal that publishes research, monitoring results and case studies on the effects of conservation interventions. There are no publication fees. A paper needs to have a detailed description of the intervention and a clear quantification of the consequences, but simple, concise papers are welcomed.

Please contact Rebecca Smith r.k.smith@zoo.cam.ac.uk if:

- You have evidence of the effectiveness of conservation interventions for amphibians.
- You would like an electronic copy of the amphibian synopsis once it is available.

We aim to complete the amphibian synopsis by July 2013. It will be available as a searchable database, a free pdf and a book on our website **www.conservationevidence.com**.

Details for submitting a case study to the Conservation Evidence Journal can also be found on our website.

ConservationEvidence.com

Providing evidence to support decisions about nature conservation

The SPLAT Project: Mitigating Amphibian Road Mortality in the Clayoquot Sound UNESCO Biosphere Reserve

By Barbara A. Beasley

On warm rainy nights, hundreds of amphibians are killed along the highway that leads to the heart of the Clayoquot Sound UNESCO Biosphere Reserve on the west coast of Vancouver Island, Canada. Twelve years ago, a small group of volunteers joined me in instigating a series of activities to address the problem. We began by counting the remnants of amphibian carcasses, or "splats", along 40 km of Highway 4 to identify which species were killed and where the density was highest (*1*). The term "splat" originally represented only a disheartening point in a survey, but as time went on and more people showed an interest, SPLAT became an acronym for the "Society for the Prevention of Little Amphibian Tragedies". Good fortune and hard work has led to some positive outcomes for the SPLAT Project. I want to share our story with others who are involved in similar projects around the world.

We found the highest density of amphibian mortality along a 1.6 km stretch of the highway within half a kilometer of a 4 ha wetland, called Swan Lake (Fig. 1). We counted egg masses at Swan Lake over several years and discovered one of the largest breeding populations of Northern red-legged frogs (*Rana aurora*) recorded in Canada. The Northern red-legged frog is listed as Special Concern by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) (2) and, thus, under the provisions of British Columbia's legislation to conserve habitat for listed species, Swan Lake and the forest surrounding it (18 ha) became protected as a Red-legged frog Wildlife Habitat Area (WHA) in 2010. This small piece of protected land is just outside the Pacific Rim National Park Reserve. The complete protection of the wetland and adjacent forest habitat at Swan Lake is important, not only to the Red-legged frog, but also to Northwestern salamanders (*Ambystoma gracile*), Rough-skinned newts (*Taricha granulosa*), Pacific treefrogs (*Pseudacris regilla*), Western red-backed salamanders (*Plethodon vehiculum*), Wandering salamanders (*Aneides vagrans*) and other wildlife. The assurance that habitat beside the highway will never be degraded by development prompted us to proceed with plans to mitigate the threat of traffic.

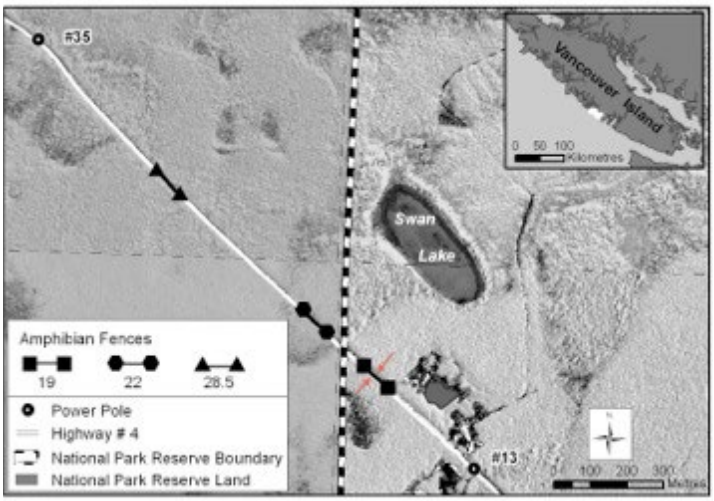

Fig. 1: Map of Swan Lake and Highway 4 on the west coast of Vancouver Island. Red Arrow shows the location of the concrete box culvert at roadside fence 19. Road mortality surveys were done from hydro pole # 13 to # 35.

Association of Wetland Stewards for Clayoquot and Barkley Sounds, Ucluelet, B.C., Canada, V0R 3A0

Fig. 2: Roadside trapping to monitor movement patterns of amphibians along Highway 4, Vancouver Island. Photo: Jacqueline Windh.

We started with a temporary measure. We installed 90 m sections of plastic fencing along the verge of the highway in three places where mortality densities were highest. We placed pitfall traps along these fences and opened the traps in the autumn and spring of 2005 and 2006, and in the autumn of 2008 and 2009. We opened the traps on warm rainy nights and checked them the following morning (Fig. 2). After recording the species and size, we released each captured amphibian on the opposite side of the highway. We continued to survey for "splats" in unfenced sections of the highway on the same nights that we trapped. We estimated that, on average, 20% of those trying to cross were being killed by traffic. The trapping program also helped us fine-tune our knowledge of amphibian spatial movements. We pinpointed the 90 m section of fenced highway where most amphibians were caught trying to cross. We used this information to propose the location of a permanent mitigation measure.

Mitigation to reduce road mortality is not new. Europeans have been installing roadside barriers and culverts since the 1960s. But in North America, and especially Canada, we have lagged behind. Until recently, we have tried very few mitigation structures and done little follow-up monitoring to test their effectiveness in connecting habitats and reducing road mortality (3). Given the uncertainty, it was progressive for the British Columbia Ministry of Transportation and Infrastructure (MoT) to decide to invest in an underpass system at the site. It helped that we had collected ample background data, we were trying to safeguard a species-at-risk within a UNESCO Biosphere Reserve, and that non-government organizations agreed to commit support for monitoring the effectiveness of the system after it was installed.

We reviewed the literature for projects done in Europe (*4*, *5*) and the U.S. (*6*, *7*) to learn what sort of tunnel system would function well for the amphibian families being killed on our highway—ranidae, ambystomatidae, salamandridae and plethodontidae. We also visited sites where culverts had been installed in British Columbia and looked at available data on how well they functioned. Despite mixed results, three critical structural features were recommended for all amphibians: large size, natural substrate, and fencing to lead animals to the tunnel entrance.

Fig. 3: Installation of a pre-fabricated concrete box culvert. Photo: Barbara Beasley.

After procuring an elevation survey of the road and drainage ditches at the Highway 4 site, MoT helped us choose a culvert that was as large as possible to fit the size of the roadbed. It also had to be affordable and match MoT's safety specifications. We selected a pre-fabricated concrete box culvert (1.8 m wide by 0.9 m deep) and MoT oversaw the installation by local contractors in 2011 (*8*). The structure was half-filled with dirt and downed wood to provide a natural substrate like the forest floor (Fig. 3). We secured plastic fences to the entranceways and angled them away from the highway up to 60 m into the forest.

We used remote time-lapse photography to monitor amphibian movements through the culvert. We mounted a RECONYX camera from the ceiling of the culvert at the west end facing east on an angle toward the ground. We programmed it to take one photo per minute and up to three photos per second if triggered by a motion sensor. We assessed directional movements by following individuals through a series of images (Fig. 4 a, b, c). We photographed 229 amphibians moving through the box culvert over 175 nights of surveillance in autumn and spring of 2011 and 2012. Adult and juvenile Red-legged frogs comprised 60% of detections. The rest were mainly Northwestern salamanders and a few Rough-skinned newts and Western red-backed salamanders (*Plethodon vehiculum*). The numbers moving through the culvert per night in autumn were lower than we expected from earlier trapping sessions along the 90 m fence, but not in the spring. One explanation is that our ability to detect the smaller-sized bodies of juveniles (mainly moving in autumn) in the photos was poorer than our ability to detect adults (mainly moving in spring). In addition to the individuals moving in a clear direction through the tunnel, we photographed 42 individuals (a mix of age classes and species) that turned around or sat still for a long period of time. This behavior could indicate reluctance, or a different type of habitat use.

Our success at this stage is in showing that a box culvert, when carefully placed with guiding fences, can increase the permeability of a road for amphibians in coastal British Columbia. Similar structures have been effective in Europe, but not all species respond the same way (*9*). This is the first successful trial for Red-legged frogs and Northwestern salamanders in Canada. We know that one culvert will have a minor effect in diminishing the road mortality on the highway but it is a start. We are investigating whether amphibians will move through existing drainage culverts when they are fitted with guiding fences. The results to-date indicate that some drainage culverts could serve as amphibian passageways during dry periods. A model examining the effect of traffic on the persistence of the Swan Lake population will help us determine how much more mitigation is needed.

We hope the SPLAT Project will help others restore amphibian dispersal and migration across existing roads. We do not recommend culverts for mitigating the negative impacts of building new roads through wetland complexes or high value habitats. In those cases, it is best to relocate the new road to a less sensitive site, or if that is not possible, then construct bridges or raised roads. Funneling dispersing animals through a series of narrow culverts should be seen as making the best of a bad situation rather than an ideal solution.

Acknowledgments

The Association of Wetland Stewards for Clayoquot and Barkley Sounds coordinated the SPLAT Project with financial support from the B.C. Ministry of Transportation, Habitat Conservation Trust Foundation, Clayoquot Biosphere Trust, and Environment Canada. Parks Canada and Simon Fraser University donated time and materials to the project. Many community members volunteered to help collect data and carry amphibians across the highway.

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Fig. 4 a, b, c.: A series of time-lapse photographs of two adult Red-legged frogs (*Rana aurora*). Photos: Barbara Beasley.

Salamander Watch

By Matt Ellerbeck

Amphibians species from all over the world are experiencing significant population declines and these includes salamanders. As such, I am committed to do all I can to help contribute to the preservation of these amazing creatures. This is primarily done through raising awareness and promoting stewardship actions and initiatives that can have a positive impact on salamander populations. This is achieved through outreach education, articles and media appearances.

Red-backed salamander (Lead phased)-*Plethodon cinereus.* Photo: Matt Ellerbeck.

However, one of the efforts that I enjoy the most is observing salamanders out in their natural habitats. Given that salamanders are in such terrible decline, it is an immense pleasure and privilege to encounter these magisterial animals. This is something that I have been doing since I was a young child. It is also how I first became interested in salamanders. My first memory of a wild salamander was a vibrant Red eft (*Notophthalmus viridescens*) that I found under some shingles behind my grandparents cottage. It's beautiful orange background color and stunning red spots instantly intrigued me. Shortly thereafter, I encountered a lovely Blue-spotted salamander (*Ambystoma laterale*) in the backyard after a heavy rain storm. These early salamander encounters have stuck with me, engraved into my memory. They serve as a testament of my interest and passion for salamanders. There have since been many more salamander encounters and this continues today.

However, now all of my salamander observations are recorded. This includes the species seen, number of individuals, habitat type, date, time and temperature. Photos of all encounters are also taken to confirm identification. All of the observations I collect are sent to Ontario's Reptile and Amphibian Atlas to help contribute to the understanding of salamander populations, habitats and behaviors in the province of Ontario.

The Ontario Reptile and Amphibian Atlas is a volunteer based project that uses such information to monitor changes in the ranges of reptile and amphibian species and fluctuations in their population numbers. Data are also used to determine if a species is at risk (Endangered, Threatened or Special Concern Status) and to identify and manage important habitat for rare reptiles and amphibians. The objective of the atlas project is to improve knowledge of the distribution and status of Ontario's reptiles and amphibians by collecting observation submissions from the public, carrying out field surveys and amalgamating existing databases.

Although, I am consistently active in regards to my outreach efforts I always make time to go out and collect salamander observations for the Atlas. However, striving to find the time is easy when I am motivated in part by wanting to help contribute to such a worthwhile and important project and secondly because I am beyond elated to see salamanders out in the wild!

Such observations are also especially important as salamanders are not commonly observed. As generally fossorial creatures, salamanders usually only emerge late at night or during periods of rain—not the times most people are out hiking or visiting woodlands. Other forms of herpetofauna in Ontario (i.e turtles, snakes and frogs) are more commonly encountered by individuals as such species will often bask in sunny areas where viewing is easy. Salamanders are seen less often and so I feel it is essential that I collect much information on them for the Atlas as caudata observations are likely to be less than those of other herptiles.

Blue-spotted salamander-*Ambystoma laterale*. Photo: Matt Ellerbeck.

Photos and recorded videos from such encounters are also useful not just for the Atlas project and understanding populations, but also in promoting a general better understanding of salamanders and their conservation in Ontario. During my presentations and educational displays I have had many conversations with the general public. From these discussions I have learned that many people were previously unaware that salamanders were found in the region. Individuals cannot be expected to get active and involved with stewardship and conservation actions for animals that they are not even aware are present! Making people more aware of salamanders and Ontario's biodiversity as a whole is of great importance when fostering an interest in conservation.

This is how my favorite childhood past time, watching salamanders, has evolved from a hobby into a way for me to help advance the betterment of my favorite animal.

Matt Ellerbeck - Salamander Advocate & Conservationist, Save The Salamanders Project. www.savethesalamanders.com

"A Small but Beautiful Bit of Light:" Citizen Science for Amphibian Conservation in Southwest New Hampshire

A Spotted salamander crossing a road in Keene, NH during the spring migration. Photo: Zach Schneider.

By Brett Amy Thelen

Every spring, as the earth thaws and spring rains drench New England, many thousands of Spotted salamanders (*Ambystoma maculatum*), Jefferson-blue spotted salamanders (*Ambystoma laterale-jeffersonianum*), Wood frogs (*Lithobates sylvaticus*) and spring peepers (*Pseudocris crucifer*) make their way to vernal pools in warm, evening rains to breed. In some years, when weather conditions align just so, the migration happens in just one or two "Big Nights." Some of these species are quite long-lived and return, year after year, to the same vernal pool to breed; in many cases, it's the very pool where they themselves were born.

These days, their journeys often take them across busy roads and roadkill has become a significant conservation concern. One study in western and central Massachusetts found that roadkill rates commonly found on even relatively quiet roads could be high enough to lead to local extirpation of Spotted salamander populations in as few as 25 years (*1*). Another study reported that 50-100% of salamanders attempting to cross a paved rural road in upstate New York didn't survive the trek (*2*).

The best citizen science projects are approachable, meaningful, and *fun!* Photo: Mary Kate Sheridan.

Salamander tunnels have effectively reduced amphibian road mortality in some communities, but are costly to design and construct. In the absence of such infrastructure, Ashuelot Valley Environmental Observatory (AVEO) trains volunteers to count migrating amphibians and to safely usher the critters across roads during one or several Big Nights each spring.

Over the years, AVEO—the citizen science arm of the Harris Center for Conservation Education, a regional land trust and conservation education organization in the Monadnock Region of southwestern New Hampshire—has coordinated a broad spectrum of community-based conservation research and monitoring projects, but we're probably best known for these Salamander Crossing Brigades.

How it Works

AVEO coordinates Salamander Brigades at eight known sites in southwest New Hampshire with high concentrations of road-crossing, spring-migrating amphibians. Each formal crossing site has one or two volunteer Site Coordinators, who are responsible for

compiling data, managing field equipment (temporary *Salamander Crossing* road signs, field identification sheets, data forms, and extra flashlights), and assisting other Crossing Brigade volunteers in the field. All volunteers are trained in species identification, road safety and basic data collection. As the time nears, AVEO staff watch the weather closely; when conditions seem right for an evening migration, we notify volunteers via email and social media. We also post a five-day "Salamander Forecast" on our website. Volunteers arrive at their sites around sunset and stay until car or amphibian traffic slows, typically before midnight. As they move amphibians across the road, volunteers keep count of how many individuals they find, by species; dead amphibians are counted separately and removed from the road to prevent duplicate counts of the same individual.

These citizen scientists also help find new-to-us amphibian road crossings, which we add to an online map of amphibian crossing sites in the Monadnock Region. Our goal is to identify road crossings in every area town, so that enthusiastic volunteers do not need to drive far, or even at all, to participate. We also strive to empower people to become stewards of their neighborhood road crossings, whether or not the crossing is staffed by a Site Coordinator or demarcated by *Salamander Crossing* signs. Although AVEO trains volunteers and keeps interested folks apprised of potential migrations, the true power of this program lies in its potential to inspire decentralized, grassroots, do-it-yourself amphibian crossing efforts among groups of friends and neighbors. Because of the highly-localized and weather-dependent nature of amphibian migrations, local coordination is essential; as of this writing, amphibian crossing efforts of various scales (from single road crossings to regional initiatives) exist in at least eight Northeastern and mid-Atlantic states, including Vermont, Massachusetts, Connecticut, New Jersey, New York, Pennsylvania and Virginia.

A Salamander Crossing Brigade volunteer admires a spotted salamander as he helps it across the road during the spring amphibian migration in southwest New Hampshire. Photo: Myra Rebillard.

A Salamander Crossing Brigade volunteer helps a Jefferson/Blue-spotted complex salamander across a road in Keene, NH during the spring migration. Photo: Sigrid Scholz.

In the last five years, AVEO's Salamander Crossing Brigade volunteers have crossed nearly 12,000 amphibians in southwest New Hampshire alone.

"A Small but Beautiful Bit of Light"

The Salamander Crossing Brigades aren't just about reducing road mortality; they're also a thrilling opportunity for wildlife observation and for increasing awareness of roadkill and amphibian conservation. For many folks, a road crossing is the first time they've ever laid eyes on the amphibian species that live in their neck of the woods. The Spotted salamander is a highly charismatic creature with a smile that could melt an iceberg, so it should come as no surprise that these encounters have a lasting impact:

"I was thrilled to go out last night and see my first spotties! AMAZING! Last night was one I'll remember forever."

"[Your message about the salamander crossing] got to the heart of why we keep doing field work in sometimes miserable conditions: It's important for the earth; it feels really good to be doing something positive; and it's a hoot to know there are other slimy-fingered loonies out there in the middle of the night sharing the thrills and the heartaches."

"I had an absolute blast last night out on River Road. I rounded up a posse consisting of two other graduate students and a FDNY firefighter visiting from Brooklyn. I'm sure he wasn't expecting to be out saving salamanders. Anyway, thank you so much for letting me be a part of all of this; it is a truly humbling experience. We were all pretty cold and soaked to the bone last night and we went home, put water on for tea, changed into dry clothing, and talked about the evening. We will always have fond memories of the migration."

"My son and I wouldn't miss it…It is a wonderful night. It's like stepping into another world."

"I hear a gazillion peepers and think that some of those out there would not be singing if it weren't for people like you and the volunteers who helped them in their passage. In a world filled with negativity and sad stories, it is a small but beautiful bit of light."

The Vernal Pool Project

The Salamander Crossing Brigades make a great entry point for new citizen scientists—data collection is simple, volunteer commitment is minimal and participation is rewarded with a high likelihood of personal interaction with charismatic amphibians. As a result, in any given year, 75 to 150 people will actively participate

in AVEO's Salamander Brigades, and another hundred or so will follow along from home. A fraction will then move on to volunteer with our Vernal Pool Project, a citizen science initiative aimed at identifying and documenting small, seasonal wetlands that serve as critical breeding habitat for several amphibian species of conservation concern in New England.

Vernal pools are largely unprotected by existing New Hampshire wetlands regulations—which are geared toward larger, more permanent wetlands—and are thus especially vulnerable to development, logging and road impacts. They are also notoriously difficult to predict via remote sensing data. For these reasons, vernal pool identification and conservation is most effective at the local level. However, town planners, conservation commissions, forest managers and landowners can only protect these vital breeding habitats if we know where they are.

AVEO's Vernal Pool Project volunteers have now documented **nearly 80 vernal pools in the Monadnock Region, with trainings and fieldwork slated to continue for at least several more springs.** Along the way, our intrepid vernal pool citizen scientists have also discovered three previously-unknown Jefferson salamander occurrences (a state-listed Species of Special Concern in New Hampshire) and identified a slew of potential vernal pools to scope out in future years. Vernal Pool Project data have been utilized for land use planning and site review by local land trusts and conservation commissions, and data from pools on public land are shared via an online, illustrated, interactive Google Map.

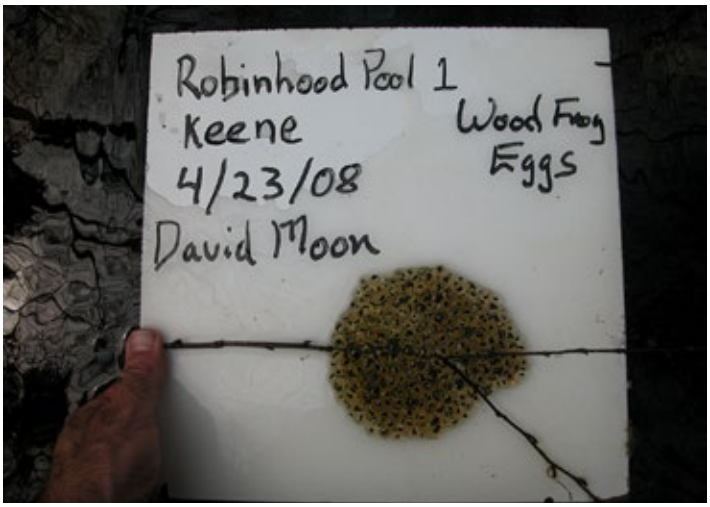
Vernal pool field inventories require a higher level of volunteer commitment, more intensive training, and greater scientific rigor than the Salamander Brigades. They also require more imagination on the part of volunteers, since the charismatic megafauna have for the most part left the pools by the time our citizen scientists arrive to suss out egg masses and larvae. Still, the ephemeral nature of vernal ponds—brimming with life in spring, dry by fall—is enchanting, and the "treasure hunt" aspect of searching for isolated pools amid a matrix of forested uplands and of searching for salamander and frog egg masses beneath the surface of the pools' dark waters, is a draw. As one volunteer described, "When we found the first milky white glob [a spotted salamander egg mass] submerged in dark, tannic waters, we could have been Forty-niners discovering gold or Mayflower passengers sighting land. Eureka!"

Local Citizen Science for Local Conservation

There's a reason we're all hearing more and more about citizen science and that's because it's growing in popularity. Traditional scientific institutions recognize the value of building support for their work by inviting the public to participate in it and nature enthusiasts get a lot out of knowing that someone actually cares about their observations. But citizen science projects require careful planning to be both good education and good conservation science.

The best projects are approachable, meaningful and *fun* for volunteers. They employ field methods than can be taught in one or two short training sessions, have concretely defined goals and objectives, and are undertaken in reasonably pleasant field conditions. (If you want your volunteers to collect completely unreliable data, send them out to a field site swarming with distracting, bloodthirsty mosquitoes!) In areas with a strong sense of local identity, like small-town New England, the most effective citizen science projects also involve close collaboration with community members, as volunteers, as supporters and equally as importantly,

Wood frogs hitch a ride across the road with a Crossing Brigade volunteer on Big Night. Photo: Lou Kaletsky.



Wood frog egg mass documented in a vernal pool in Keene, NH. Photo: David Moon.

as local leaders—land trusts and landowners, members of conservation commissions and planning boards—who can use the data to inform local land use planning and conservation efforts.

When these pieces are in place and a citizen science project comes together, its conservation impact can move through a community in unexpected ways. As described by one mother whose daughter has grown up counting and crossing critters with her as part of the Salamander Brigades, "Yes I go [out to the road crossings] and I love doing it, but then, I love to be able to share it out from me, too. I often do Girl Scout leader trainings in the spring, and it's almost always raining. I will take leaders out at night and we go off and try to find things moving in the night. I then become a secondary source of information and it spreads out like rippling waters. Like AVEO is the center of information and we are the ripples."

Brett Amy Thelen is the Program Director of Ashuelot Valley Environmental Observatory, the citizen science arm of the Harris Center for Conservation Education. All photos were taken by AVEO citizen scientists. For more information on the Salamander Crossing Brigades or Vernal Pool Projects, visit www.aveo.org or contact Brett at thelen@harriscenter.org.

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Temporary Salamander Crossing signs mark the site of amphibian road crossings on Big Nights.

Wetlands Restoration: Crafting Successful Messages for the Public

By Jeff Holmes

Communicating complex ecological concepts to the general public in a manner that inspires them to take action can be challenging for members of the conservation biology community. This is especially true when dealing with frequently overlooked or, worse yet, commonly reviled taxa such as herpetofauna. In general, amphibian and reptile conservation organizations face two major challenges when raising funds:

1. Passionate but poor: Those most likely to understand and care about amphibians, reptiles and their habitats are other herpetologists, many of whom are too poor or too busy raising their own funds to make a donation.

2. Financially qualified but disinterested: The greater general public often lacks sufficient awareness, understanding of and concern for the intrinsic value of amphibians, reptiles and their habitats to make a donation.

Jeff Holmes, Executive Director, The Amphibian And Reptile Conservancy.

Since we cannot make members of the herpetology community rich, our only option is to elevate the awareness, understanding and concern of the greater general public to the point that some will feel compelled to take action by making a donation.

Some members of the general public can be persuaded to act on a predominantly emotional basis by viewing cute, anthropomorphic images of salamander, turtle or frog faces accompanied by passionately compelling captions; these supporters often learn to appreciate amphibians and reptiles for their own intrinsic value. Many other members of the general public, however, need to know "What's in it for me?" For this sector of the population, it is important to establish links between issues in their everyday lives and herpetofaunal conservation.

On December 13, 2012, The Amphibian and Reptile Conservancy (ARC) launched our first fundraising campaign in support of our *17 by 17 Wetlands Creation and Restoration* program. In our most successful campaign action to date, we joined forces with nature photographer and graphic designer Dave Huth to create a series

of images and accompanying messages that combined both emotional appeal and informative factoids about how herpetofaunal conservation impacts our everyday lives.

About the 17 by 17 Wetlands Creation and Restoration program

Before we present our successful messaging collaboration with Dave Huth, it is important to provide information about the program the collaboration was designed to support.

ARC, in partnership with the Center for Wetland and Stream Restoration (CWSR), will create or restore 17 critically important wetlands across the United States by the year 2017. Each wetland will be custom-designed and constructed to meet the needs of the local associated herpetofauna and blend seamlessly into the surrounding ecological context. Each wetland project will have both direct and indirect conservation results.

Direct resutls

Each wetland will provide vitally important breeding and core habitat for wetland-obligate taxa, with a special emphasis on rare species and communities of amphibians.

Indirect results

Each wetland project will serve as either A) A hands-on training opportunity for professionals who can then create or restore their own wetlands and train others to do so, exponentially multiplying our overall impact on conservation by creating an ever-expanding talent pool of qualified wetlands management specialists, or B) A hands-on educational opportunity for students who will create a "living laboratory" that will continue to educate subsequent generations of students for years or even decades to come.

For those of us in the conservation community, this is a straightforward, easily understood and clearly important program. For the general public however, it is complicated at best and unappealing at worst.

The Collaboration: A Successful Messaging Campaign within a Campaign

ARC opened the overall campaign December 13, 2012 with a series of email solicitations, a fundraising drive using a social media application called Causes, and an introductory video on YouTube and Vimeo. ARC also continued our ongoing daily content stream on social media with a series of popular galleries showcasing the work of various contributing photographers accompanied by relatively generic messaging, most of which was not directly related to the wetlands campaign.

By early January, the email solicitations and online fundraising drives were temporarily parked to avoid donor fatigue. In their place, we decided to create a tightly focused "campaign within a campaign" about the importance wetlands creation and restoration. To accomplish this, we crafted a series of seven messages to be posted in our social media content stream at a rate of one per day over the course of one week. The messages were sequenced "countdown style," progressing from more informative to more emotionally appealing messages throughout the week, culminating in a broad, general declaration intended to appeal to virtually everyone. This finale is also ARC's slogan and is intended to establish common ground with everyone who loves the outdoors for whatever reason (hikers, backpackers, hunters, fishermen, mountain bikers, bird watchers, etc.).

In this age of short attention spans, however, messaging via text alone is often ignored on social media, no matter how compelling the content. The messages needed to be accompanied by eye-catching images or, better yet, embedded in the images themselves where the text is least likely to be ignored.

After reviewing the work of our contributing photographers, ARC decided to approach Dave Huth and ask if he would be willing to comb through his vast library of nature photography and match the perfect image with each message. As it turned out, Huth is also an outstanding graphic designer and offered to not only provide the images, but personally handle the layout and design to include the messages.

The result was a photo gallery that premiered on social media January 14, 2013 and ran through January 20, with a new image added daily. The gallery was titled "Seven Reasons to Take Action by Dave Huth" and the following text was used in the album description:

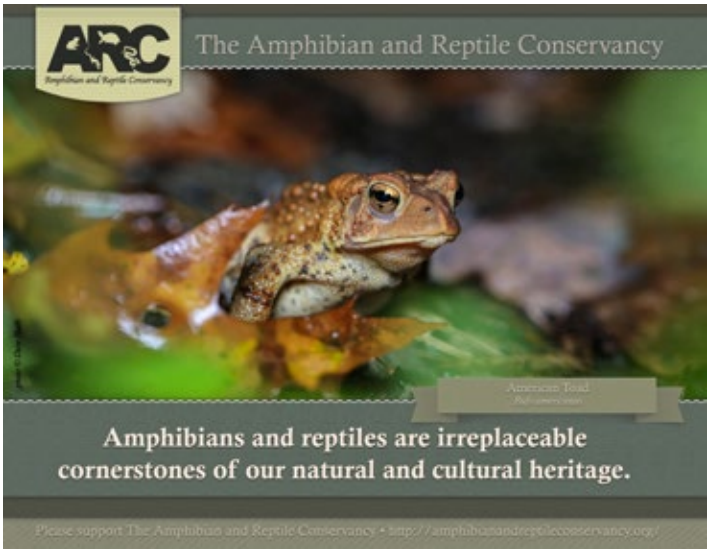
"A week's worth of images and messages about why amphibian and reptile conservation and wetlands restoration are so critically important to everyone.

These are wonderful images and messages to LIKE and SHARE with your friends both within and outside the conservation community. In plain English for everyone who enjoys the outdoors including hikers, backpackers, hunters, fishermen, bird watchers, mountain bikers… Or people who simply want clean air to breathe, clean water to drink and affordable, healthy food on their table (and who might take these necessities for granted).

When we approached contributing photographer Dave Huth with this concept, he ran with it. Dave personally selected the images for each message from his vast library of nature photography. He also personally did all the design and layout work. Make sure you reward him for this outstanding contribution to conservation by visiting his website: http://davehuth.com/

And please take a moment to make a contribution of $17 or more to wetlands restoration at http://amphibianandreptileconservancy.org/donate. If all of our fans gave $17, we would raise nearly $24,000! That amount will restore a lot of habitat."

The photos in this gallery have been viewed by thousands and have been posted on Facebook, Twitter, Flickr (by both ARC and Huth) and in Huth's blog, generating both donations and elevated notoriety to ARC.



Promoting Amphibian Conservation Through the College Classroom

By Mizuki Takahashi

Fifteen years ago, when I was a master's student at the University of Tokyo, I encountered one of the most influential books in my life, "Tracking the vanishing frogs" by Kathryn Phillips. I was shocked and disturbed by the story unfolding page after page; amphibians were rapidly and enigmatically disappearing worldwide. These same emotions, on the other hand, encouraged my dream of studying amphibians in the US, one of the leading countries in the field. Of course, my ultimate goal was to be part of global amphibian conservation efforts. After many years of graduate and postgraduate training in the US, I finally seized an opportunity to teach an upper level Amphibian Biology course at Bucknell University in 2011. A few things were certain to me by then. First, education is essential in promoting conservation. Second, the current generation college students in general lack direct experiences with nature. And finally, cookbook labs are boring not only to students but also to teachers. And when we are bored, we are careless, not engaged and thus do not learn well.

The first field trip to forest habitat in the fall of 2011. Photo by: Mizuki Takahashi.

With these three points in mind, it didn't take me long to decide to bring detection of chytrid fungus (*Batrachochytrium dendrobatidis*) into the laboratory section of the Amphibian Biology course. This fungus has been linked with many amphibian declines worldwide. Thus, examining the prevalence of chytrid fungus among local amphibians provides important information for conservation management. However, this kind of project is not appealing or rewarding to many university researchers because it is so simple, lacks novelty and does not often result in high impact publications. Yet, unlike frog calling surveys, it is difficult for layperson volunteers to conduct such studies because of the funding, equipment and techniques required. This may in part explain the lack of our knowledge on the distribution and prevalence of chytrid fungus in many areas of the world, including central Pennsylvania. I realized that this class project has a potential to contribute to the scientific community by expanding the chytrid fungus database (for example, www.Bd-maps.net), in addition to its educational value in amphibian conservation and providing an opportunity for undergraduates to experience amphibians in their natural habitats and also to learn science by doing science.

It is very difficult to diagnose fungal infection based on visible symptoms. So I spent a few weeks during the summer of 2011 teaching myself its detection methods through molecular analyses. I ordered primers (pieces of DNA sequences used to amplify specific parts of a target organism's genome) that are specific to chytrid fungus, obtained chytrid fungus DNA (for use as a positive control) from my friend at the University of South Florida and conducted a preliminary experiment to establish the methods for students to follow. It went well and gave me confidence that I could develop the lab section around this project.

During the 2011 fall semester, each of four groups of students in Amphibian Biology course (in total 13 students) conducted an independent research project examining chytrid fungus prevalence

Students collecting amphibians in a vernal pool surrounded by a conifer-deciduous mix forest. Photo by: Mizuki Takahashi.

Collected eastern newts (*Notophthalmus viridescens*) were individually contained to avoid cross contamination. Photo by: Mizuki Takahashi.

among local amphibians in one of three habitats: stream, forest or pond. Students were very excited about the fact that nobody knew how their results would turn out and they would be the first to know. I was very curious, too. There were some students who had never seen or touched wild amphibians. In addition, one student was afraid of them. Our first field trip was to forest habitat and it was a rainy, chilly day, not a perfect day for introducing students to amphibians in the field. Despite my concern, I was delighted to see the students' vivid excitement in finding Red-backed salamander (*Plethodon cinereus*), Slimy salamander (*Plethodon glutinosus*), and American toad (*Anaxyrus americanus*). We swabbed their skins, clipped one of their toes and released them at the site of capture. During the first four weeks of the semester, we visited three pond habitats, one forest, and one stream. The collected samples were brought back to the lab and students performed all of their work: DNA extraction, amplification of target DNA sequences using polymerase chain reaction, and agarose gel electrophoresis. In total, they examined 59 individuals from six species: 15 Dusky salamander (*Desmognathus fuscus*), two Bullfrog (*Lithobates catesbeianus*), eight Green frog (*L. clamitans*), one Wood frog (*L. sylvaticus*), 18 Eastern newt (*Notophthalmus viridescens*) and 15 Red-backed salamander. Of these 59, only one sample, Eastern newt, was found to be infected with chytrid fungus.

Our results of one positive sample out of 59 total specimens tested suggest low chytrid fungus prevalence among our sampled amphibians in central Pennsylvania. Other Pennsylvania studies have reported higher occurrences of chytrid fungus. For example, Groner and Relyea (2010) reported an infection rate of 34.8% (16 of 46 individuals tested) in Eastern newt populations of northwest Pennsylvania and Raffel *et al.* (2010) found an infection rate of 39.2% (60 of 153 individuals tested) in the same species in an adjacent county to our sampling sites. However, another study of eastern newt populations in eastern Pennsylvania along the Delaware Water Gap National Recreation Area found no infected individuals with 41 tested for chytrid fungus (Glenney et al. 2010). A similar case of spatial heterogeneity in this pathogen has been reported in Wise and Warren counties, Virginia, USA. When only comparing salamander species surveyed in both studies, the prevalence in Wise County was 14.8% (4/27; Davidson and Chambers 2011), while it was only 0.6% (1/171) in Warren County (Gratwicke et al. 2011). These variations in infection rates exemplify our limited understanding of spatial and possibly temporal heterogeneity of chytrid fungus prevalence and highlight the need for more data regarding the pathogen's distribution.

Students swabbing amphibian skins and taking field notes. Photo by: Mizuki Takahashi.

Our project through the college classroom showed that it is feasible for an undergraduate class to complete a chytrid fungus detection project within a 14-week semester. Upon the completion of the project, each group prepared a manuscript following the format of a peer-reviewed journal, Herpetological Conservation and Biology (HCB hereafter) and submitted it to me. Each group also gave a formal presentation to the rest of the class at the end of the semester. After the semester ended, one student synthesized a single manuscript by combining the manuscripts submitted by each of the four groups. The exciting news was that this manuscript was published in HCB in December 2012 with all students included as co-authors (Wunder et al. 2012). Another student presented their findings at a campus-wide research symposium in March 2012 at Bucknell University. I gave a poster presentation about our project at the 7th World Herpetology Congress in Vancouver, Canada, in August 2012. It was a fruitful project for the students, for me, and for the scientific community.

What is the future direction? I plan to teach Amphibian Biology every year and continue to implement similar projects. It would be interesting to monitor local chytrid fungus prevalence over years. We can also include new species and new localities. But it would be more exciting to diversify the project options from which each student group gets to choose their own project that is more unique from one another. During the summer 2012, I contacted the owner of a local zoo, who kindly consented to the idea of Bucknell students examining chytrid fungus prevalence among their amphibian collection. As a result, in the fall 2012 I added two more options to the previous year and laid out five possible projects to the students at the beginning of the semester: detection of chytrid fungus among forest, pond, stream, zoo, or pet shop amphibians. Here's part of what students found most recently: three frogs to be positive among 30 specimens tested from the zoo collection and one positive out of 20 pet shop amphibians. These results are valuable because published reports of chytrid fungus among zoo and pet shop amphibians are rare in the US. Recent studies suggest that global amphibian trade has likely resulted in the creation of the hypervirulent lineage of the fungus through the genome exchange between the previously isolated fungal lineages (James et al. 2009, Farrer et al. 2011). The devastating news is that this hypervirulent lineage is associated with mass die-offs of amphibians worldwide. Once again, it is likely us humans that triggered the global amphibian declines. I am currently helping one of the students formulate a manuscript for another publication (Winters et al. in prep). This example of examining traded amphibians shows another direction we can take to promote both amphibian conservation education and research through the college classroom. Finally, we may be able to make an even larger impact if a group of professors develop a network to share the progress and results of their independent classroom projects among different institutions.

Students and I are doing something I never imagined fifteen years ago when I read "Tracking the vanishing frogs" in the busy Tokyo metro. I ardently hope that some of the students involved in the research projects become significant contributors to the global amphibian conservation efforts in the near future.

Students conducting DNA extraction in the ecology laboratory at Bucknell University. Photo by: Mizuki Takahashi.

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Bucknell students visiting a local zoo, learning the animal care facility, and swabbing their amphibians. Photo by: Mizuki Takahashi.

Anuran Monitoring in the Sierra Nevada, California, U.S.

Fig. 1: *Rana sierrae.* Photo by: Kimberly Peterson.

By Cathy Brown, Lucas Wilkinson & Kathryn Kiehl

Long-term, bioregional monitoring of amphibians can provide information on population trends and emerging threats. Faced with uncertainty regarding two at-risk taxa, the mountain yellow-legged frog species complex (*Rana sierrae* and *R. muscosa*, referred to here as *Rana sp.*, Fig. 1) and the Yosemite toad (*Anaxyrus* [*Bufo*] *canorus*, Fig. 2), the U.S. Forest Service implemented multi-scale, probability-based monitoring to estimate population status and change on national forest lands within the taxa's ranges in the Sierra Nevada, California (*1-4*). Both taxa are considered warranted but precluded for listing under the U.S. Endangered Species Act due to the precedence given to higher priority actions (*5, 6*). Monitoring also included the sympatric Pacific treefrog species complex (*Pseudacris sierra and P. hypochondriaca*, referred to here as *Pseudacris sp.*, Fig. 3). By utilizing a unique long-term, multi-scale monitoring design, we provided quantitative and qualitative information on the status and ecology of these taxa which may benefit their conservation.

For population monitoring, the types of data that are logistically feasible to collect vary with scale and species ecology. For example, *Rana sp.* have a multiple-year tadpole stage requiring permanent water for breeding and generally are found close to water. Thus for this taxon, counts of animals are feasible to collect during visual encounter surveys, and can be used to evaluate changes in both distribution and relative abundance at large bioregional scales. In contrast, *A. canorus* and *Pseudacris sp.* breed in ephemeral water habitats, adults and subadults are terrestrial after the breeding period, and tadpoles transform and disperse in one season so that even basic abundance counts are seasonally dependent. In cases like this where obtaining demographic information (e.g., abundance, survival) at large scales is logistically impractical, occupancy can be used as a relatively affordable metric to evaluate distributional changes bioregionally (7) and more detailed demographic information can be collected at a smaller subset of locations (2). These constraints led us to develop a multi-scale monitoring approach where, at the scale of the species' Sierra Nevada range (on national forest lands), we estimated the proportion of occupied watersheds (2-4 km^2) for each taxon, the number of occupied sites (lakes, meadows, stream reaches) per watershed for each taxon, and relative abundance for *Rana sp.*, and then in two watersheds, abundance of adult breeding males and egg masses for *A. canorus* (1-3). Only occupancy was estimated for *Pseudacris sp.*

At the bioregional scale, opportunities and constraints that were considered during project design included 1) overlapping ranges and habitat use among taxa which allowed us to develop one integrated design; 2) patchy distribution of the taxa and their habitats which led to an unequal probability design; and 3) tradeoffs between sufficient sample size to estimate both status and change, ability to estimate change over a reasonable time period, and cost which led to the choice of a panel temporal design (1). We selected a spatially balanced, unequal probability sample using a generalized random tessellation stratified survey design (GRTS) for a finite resource (8) from three categories based on existing locality data for *Rana sp.* and *A. canorus*: *Recent* (taxon found between 1990 and 2001), *Historical* (taxon only found prior to 1990), and *Unknown*

Stanislaus National Forest, U.S. Forest Service, Sonora, California, U.S.

(taxon not found or occupancy was unknown). A larger portion of the sample was selected from the *Recent* category where taxa occurrence was expected to be higher, and smaller proportions from the other two categories with lower likelihood of occurrence to ensure representation across the entire monitoring area. Temporally, we used an augmented serially alternating panel design (9) where a subset of watersheds were to be visited every year and the remaining watersheds were to be visited once every five years on a rotating schedule. Visual encounter surveys were conducted in all lentic sites (lakes, meadows) and a sample of stream sites, and detection and count data were recorded. In a more intensive design component for *A. canorus*, we conducted capture-mark-recapture surveys for adult breeding males using the Robust Design (*10*) and egg mass counts in six meadows in two watersheds during spring breeding.

From 2002 to 2009, we completed the first cycle of bioregional monitoring which assessed population status. Our results corroborate other assessments that *Rana sp*. have declined in both distribution and abundance (*3*, *11*). Evidence of reproduction (eggs, tadpoles, metamorphs) was found in less than half of watersheds where animals were detected in the prior decade (*Recent* watersheds), only rarely in watersheds where the taxa was found prior to 1990 (*Historical* watersheds), and in few watersheds rangewide (Fig. 4). Relative abundance was generally low compared to numbers recorded in the literature. Only about 10% of occupied watersheds had > 500 tadpoles or > 100 adults or subadults and just over half of occupied watersheds had fewer than 20 animals (*3*).

Anaxyrus canorus is still fairly well-distributed relative to post-1990 records. However, it appears to have declined from levels observed historically and abundances in the two intensive watersheds were low compared with other reported population sizes (*2*, *12*). Evidence of reproduction was found in over 80% of *Recent* watersheds but in only about 10% of *Historical* watersheds (occupied prior to 1990, Fig. 4). Reproduction was found in about 20% of watersheds rangewide. The total number of unique individuals captured per intensively surveyed meadow ranged from 12–72. Annual population estimates were small; the largest population had only 16 to 21 males each year and egg mass counts were low. Count data from the bioregional surveys suggest that small populations may be reflective of a broader rangewide pattern.

By including the intensive component, we gained insights on other aspects of *A. canorus* demography including annual survival estimates and inter-annual breeding activities (*2*). For example, of the 130 males captured in the two intensively studied watersheds, half were captured in only one year and of the remainder, only 5% skipped one or more years. These proportions were similar to those reported from another long term study on *A. canorus* (*13*). Only a few animals moved among meadows for breeding, suggesting that Yosemite Toads have high site fidelity at the scale of individual meadows.

Fig. 2: *Anaxyrus canorus*. Photo by: Lucas Wilkinson.

Fig. 3: *Pseudacris sp.* Photo by: U.S. Forest Service Sierra Nevada Amphibian Monitoring Program.

Pseudacris sp. were widely distributed across the monitoring area (3). Because the monitoring was not designed for this taxon, we used *Rana sp.* and *A. canorus* locality data to infer prior occupancy. Evidence of *Pseudacris sp.* reproduction was found in most of the watersheds with prior locality data for *Rana sp.* or *A. canorus*, and about a quarter of watersheds in the bioregion (Fig. 4). No information on abundance is available for *Pseudacris.*

Quantitative, statistically rigorous monitoring data can inform management and conservation decisions. The results from this first probabilistic bioregional status assessment of this Sierra Nevada anuran community can be useful in environmental documents (e.g., biological evaluations under the U.S. National Environmental Policy Act), can help to inform management decisions, can contribute to the evaluation of management effectiveness, and can provide insights on the status and ecology of these taxa which may benefit their conservation.

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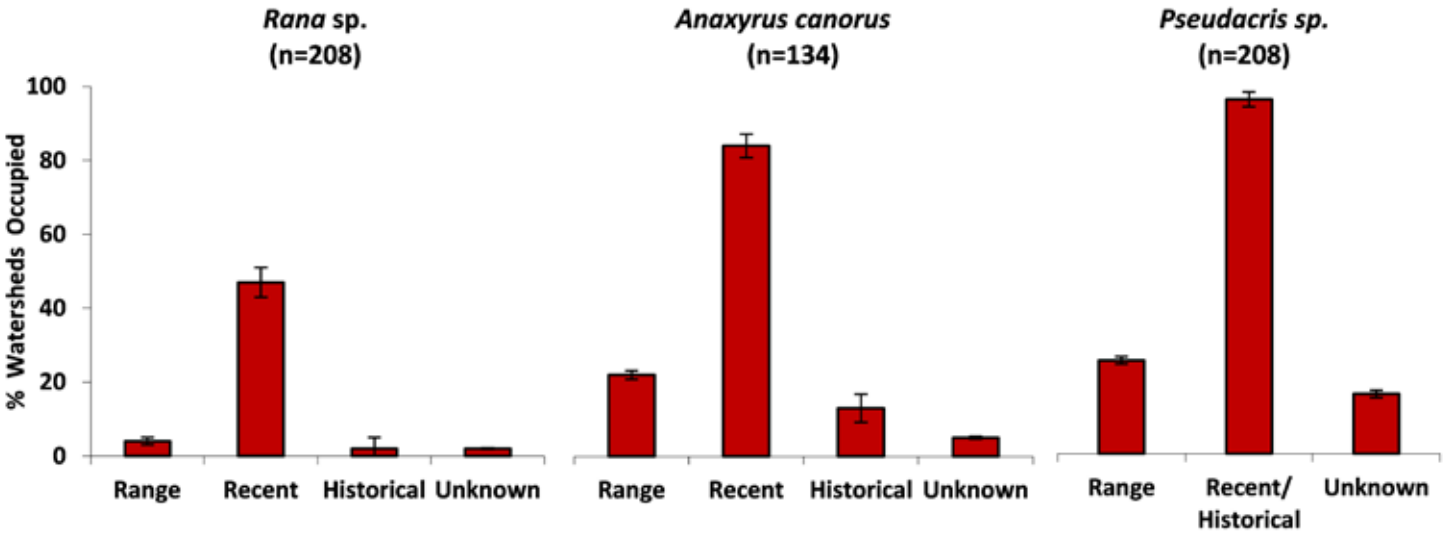

Fig. 4: Estimated proportion of watersheds with evidence of reproduction by *Rana sp.*, *A. canorus*, and *Pseudacris sp.* rangewide and for Recent, Historical, and Unknown watersheds from 2002-2009 in the Sierra Nevada, CA, U.S. Evidence of reproduction includes eggs, tadpoles or metamorphs. n is the number of watersheds sampled. Standard errors bars are shown.

Topsin-M®: How One Pesticide is Changing How We Look at Amphibian Toxicology

By Shane M. Hanlon

Every year, at what has been dubbed the "Amphibian Decline Meeting" in Tempe, AZ, a group of researchers gather to present and discuss their research on the causes of amphibian declines. The meeting is decidedly heavy on studies pertaining to chytridiomycosis, an infectious disease of amphibians caused by the pathogenic fungus *Batrachochytrium dendrobatidis* (*Bd*); however, other factors such as exotic trade, habitat destruction, and global climate change are discussed. One area, environmental contamination, receives little attention, not because it is not important, but because for all the research that has been conducted, there is still not a clear link between contamination and amphibian declines (*1*). Anthropogenic changes (e.g., pesticides) present a wide variety of environmental perturbations also associated with amphibian declines, and yet little is known about how these affect susceptibility to *Bd* or the resultant impact of infection.

Adult Southern leopard frog *Lithobates sphenocephalus*. Photo: Shane M. Hanlon.

The primary goal of my research is to determine the effects of pesticides on host-pathogen interactions in amphibians. While much research has been conducted on the separate effects of disease and pesticides, surprisingly few studies have focused on the combined effects of these stressors on amphibians. Common logic would predict that the combination of two detrimental perturbations would result in further negative effects to affected organisms. However, the possibility also exists that a pesticide would directly effect the pathology of *Bd* to amphibian hosts. With this in mind, I set out to test the hypothesis that pesticides would alter the affect of *Bd* on amphibian hosts.

At the time my research on this topic began three years ago, there was one published study that examined the interactive effects of pesticides and *Bd* on amphibians (2). Upon writing this piece, there are still less than ten. Because of this dearth of data, determining where to start proved to be a difficult decision. With the help of my advisor, Dr. Matt Parris at the University of Memphis, we decided to begin with a local *Bd* isolate and three pesticides that spanned a range of different target pests: Roundup®- an herbicide, Sevin®- an insecticide and Topsin-M®- a fungicide. While I conducted tests with all three pesticides, what I found with Topsin-M was the most surprising.

Topsin-M is the commercial formulation of thiophanate-methyl, a fungicide used to suppress mycorrhizal fungi. It is used on a variety of crops and is especially popular in the Mississippi River Basin, the area where our research is conducted. Thus, it was a good candidate to fit the "fungicide" classification. To determine the direct effects of Topsin-M on *Bd*, I first examined the effects of pesticides on *Bd* outside of hosts. Because *Bd* is grown on tryptone agar plates in the lab, I grew *Bd* in culture, then applied Topsin-M (and other pesticides) directly to the plates. What I found (not surprisingly) was that pesticides reduced *Bd* growth (3). From here, I wanted to determine how the fungicide would alter the effects of *Bd* on hosts.

In lab settings, I infected tadpoles of Southern leopard frogs (*Lithobates sphenocephalus*) with *Bd*. Once infected, tadpoles were repeatedly exposed to Topsin-M throughout the larval period. Upon metamorphosis, the frogs were tested for *Bd* infections and life history measures of size and developmental rates were assessed. Not only did Topsin-M clear infections 100% in infected frogs, but also increased size and developmental rates. Moreover, tadpoles exposed to both *Bd* and TM grew larger and faster than those in any other treatment (4). While the exact mechanism(s) for these observations is currently unknown, it is clear that the fungicide is superficially "beneficial" to *L. sphenocephalus* tadpoles.

While the preceding results were promising (and unexpected), I wanted to determine if these results were applicable in natural settings. Accordingly, I repeated the experiment in experimental mesocosms at the Edward J. Meeman Biological Research Station in Memphis, TN. Tanks were first filled with water, dosed with natu-

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Experimental array of aquatic mesocosms at Edward J. Meeman Biological Field Station. Photo: Shane M. Hanlon

Experimental mesocosm prior to Topsin-M introduction (top) and after eight weeks of Topsin-M exposure (bottom). Photo: Shane M. Hanlon

ral pond aliquots, leaf litter and substrate was added and the tanks were allowed to sit for ~ two weeks to facilitate nutrient turnover. Next, tadpoles (either exposed to *Bd* or water control in the lab) were added to the tanks. Throughout the experiment, Topsin-M was added weekly at lower concentrations than were used in the lab study. The project was to proceed until all tadpoles completed metamorphosis; however, not a single tadpole survived, independent of *Bd* treatment, in any tank that was dosed with Topsin-M. Similar to Relyea and Diecks (*5*), Tospin-M caused algal blooms and initiated a trophic cascade that ultimately resulted in the complete mortality of all tadpoles. From these results, Dr. Parris and I concluded that the facilitative properties of Topsin-M in the lab may not be applicable in field settings.

The following summer, I repeated the field experiment with the added treatment of three different Topsin-M concentrations. The highest concentration was the same as the one used the previous summer and from that two serial dilutions were produced. The goal was to determine if there was a "perfect" concentration at which the facilitations from the lab would be observed without the trophic alterations. What I found was surprising. Unlike the previous year, complete mortality was not observed in any treatments; however, the greatest mortality did occur in the highest Topsin-M treatment. Surprisingly though, tadpoles infected with *Bd* and exposed to the highest Topsin-M concentration experienced higher survival rates than those exposed to Topsin-M in the absence of infection. So, while the facilitations from the lab experiment were not observed, it appeared that the Topsin-M by *Bd* interaction "protected" tadpoles from the increased mortality observed from Topsin-M alone.

I must reiterate that I do not promote the use of pesticides to treat *Bd* in natural systems. However, although we do not know the mechanism(s) for the effects of Topsin-M on tadpoles, our results clearly show that this pesticide (and perhaps others) may have counter-intuitive effects on *Bd* infections in amphibians. While studies have focused on the effects of external perturbations on host-pathogen dynamics, such situations are not mutually exclusive and we must examine how external pressures alter host-pathogen interactions.

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Monitoring Environmental Pollution and Sublethal Effects in Giant Toad from Coatzacoalcos, Mexico

By [1]César Ilizaliturri-Hernández, [1]Guillermo Espinosa-Reyes, [1]Donaji Gonzalez-Mille & [2]Alma Nava-Montes

Fig 1: Industrial Wastewater Discharge. Photo: C. Ilizaliturri-Hernández and G. Espinosa-Reyes.

Wildlife can be used to detect chemical pollution as well as to evaluate the ecosystems health through using the species as systematic models in the risks evaluation associated to exposure routes and hazard assessment of chemical compounds. Wildlife species residing in polluted sites are exposed to complex mixtures of pollutants by multiple pathways which could hardly be evaluated in lab studies (*1*). From the ecotoxicological point of view, exposure to pollutants does not always result in lethal effects in the biota, but on the other hand, subtle sublethal effects can be produced at different levels of biological organization which eventually can influence the survival of the populations in the long-term. Amphibians have been recently proposed as biomonitors of environmental conditions due to the characteristics associated with their metabolism, life cycle and ecology (*2*). Persistent Organic Pollutants (POPs) and heavy metals are contaminants of national concern; however there are few studies demonstrating their presence and bioaccumulation, as well as their effects on amphibian populations in Mexico. This study provides baseline data on the status of POPs and lead pollution in the region of Coatzacoalcos, Veracruz.

Coatzacoalcos river basin has been one of the most diverse biological areas in Mexico; unfortunately since of 70´s, Coatzacoalcos estuary has experienced a fast industrial and urban growth which combined with other productive activities such as agriculture and cattle farming have triggered a severe impact in the region's ecosystems (Fig. 1). Actually, the Coatzacoalcos estuary houses one of the biggest and most important petrochemical industrial complexes of Mexico and Latin America (total production of petrochemical products is approximately 1.6 thousand million tons/year) and supports a population of 607,919 habitants. At present, the Coatzacoalcos River's downstream is considered one of the most polluted sites in Mexico. Environmental research in the area has shown the presence of hydrocarbon, volatile organic compounds, persistent organic pollutants and heavy metals (*3-5*).

The Giant toad (*Rhinella marina*) is a native and geographically widespread species in Mexico and Central America (*6*). It is an omnivorous and opportunistic species, which indicates that toads would reflect different exposure routes due to the ingestion of a wide variety of food items and amphibious living habits. The giant toad is one of the largest amphibians in Mexico (adult body length ranges from 10 to 17 cm), with a life expectancy of 10 to 15 years in the wild. The high lipid-somatic index (2 to 10 % compared to less than 0.1 % in most anuran species after the spawning period) and the elevated hepatosomatic index along with its breeding biology make this species prone to bioaccumulation of organic and inorganic pollutants and their toxicological effects. Recently, the giant toad has been used as an aquatic ecosystem biomonitor in the evaluation of air pollution, infectious diseases, organochlorine pesticides and endocrine disruptors.

In 2006 and 2008 we studied sexually mature toads collected from two adjacent sites to the Coatzacoalcos River (Fig. 2). The sites characteristics are: A) **Urban zone**. It is located by the banks of the Calzadas River, seven kilometers east of the Pajaritos petrochemical complex; this site was considered a reference area (minor exposure of pollutants); and, B) **Industrial zone**. It is located in favor of dominant winds at two kilometers south of the Pajaritos-Cangrejera petrochemical complexes. At this area, important pollutants such as dioxins, hexachlorobenzene and volatile compounds have been found on environmental matrices. Sexually mature toads were collected from each site using nets in nocturnal transects within an area of 10,000 m^2 and transported to the laboratory. In the laboratory, blood samples were collected. All toads' were collected with a Scientific Collector`s Permit (Colector Científico de Flora y Fauna Silvestre) issued by SEMARNAT (Secretaría de medio ambiente y recursos naturales, No. FAUT-0133) and handled in the lab in accordance with the mexican normative (NOM-062-ZOO-1999). Additionally, Surface soil samples (1-5 cm) were obtained on approximately 1 m^2 surface area and stored for further analysis. Sixteen insecticides, fourteen polychlorinated biphenyls (PCBs) and lead were evaluated in soil and blood samples. Inhibition of the activity of the enzyme delta aminolevulinic acid dehydratase (δ-ALAD) and comet assay were measured as integrative indicators of stress in order to obtain an indication of adverse health effects. The inhibition of the δ-ALAD activity is one of the most important biomarkers and a well-known indicator of lead toxicity and the comet assay is widely used to detect damage in vitro or in vivo caused to DNA by a broad spectrum to genotoxic agents in a wide range of ecological receptors. Exposure (*7,8*) and effects (*9,10*) analysis was performed according to the methods published by our group and are summarized in Table 1.

We found that concentration of lead and POPs were higher in soil samples from industrial sites. Of the 31 compounds and one element evaluated, only 28 were detected in the soil samples -Lead (Pb), Hexachlorobenzene (HCB), α,β, γ – Hexachlorocyclohexane

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Parameter		Urban zone	Industrial zone
Soil	Lead (mg/Kg)	6.6 ± 1.0	8.2± 1.2
	∑POPs (ng/g d.w)	1108.4 ± 377.8	1488.7 ± 612.5
Blood	Lead (µg/mL)	5.5 ± 0.9	10.9 ± 2.4
	∑POPs (ng/g l.w)	2547.8 ± 740.3	4509.4 ± 1739.3
Sublethal Effects	δ-ALAD activity (µmol/min*L RBC)	48.8 ± 6.5	5.5 ± 0.9
	DNA Damage (µm)*	9.7 ± 0.5	14.1 ± 0.8

Values represent the mean and standard error; ∑POPs: Total of Persistent Organic Pollutants; δ ALAD: Delta aminolevulinic acid dehydratase; *DNA damage as comet tail length; d.w: Based dry weight; l.w: Based lipid weight.

Table 1: Comparisons of environmental pollution, body burden and sublethal effects parameters.

Fig 2: *Rhinella marina*. **Photo: C. Ilizaliturri-Hernández and G. Espinosa-Reyes.**

(HCH), Dichlorodiphenyltrichloroethane (DDT), Dichlorodiphenyldichloroethylene (DDE), Mirex and 20 PCBs congeners- collected from the study area. PCBs congeners detected were 52, 70, 74, 82, 87, 90, 101, 105, 110, 118, 128, 132, 138, 149, 151, 153, 156, 158, 170 and 180. The presence of these compounds may represent a high potential risk to biota in the region. The levels of POPs and lead in toads' blood which were from industrial site were higher than the levels of toads from urban site. Our data shows a possible relation between POPs and lead concentrations in soil and the Body burden. Various studies on food preferences of the giant toad have identified a great variety of stomach contents, as well as rock fragments and soil particles; this also suggests that accidental ingestion of soil can be considered an important pathway to exposure. Also, we observed a significant decrease in δ-ALAD activity in toads from industrial sites, in comparison with toads from urban zones. Decrease pattern of δ-ALAD concentration by site can be affecting the synthesis of protoporphyrin IX and hemoglobin (secondary anemia) in toads with high lead levels in blood, especially in sites with industrial influence. The damage to DNA was higher in the toads from industrial sites. DNA damage recorded in the toads of the lower basin of the Coatzacoalcos River is evidence of genotoxic pollutant exposure. This may reflect the degree of environmental stress of aquatic and terrestrial organisms in the region.

Because of ethical and preservation reasons, the development and use of non-destructive biomarkers in wildlife organisms have been promoted. Blood parameters evaluated in this study may be very useful in field conditions because a greater number of samples can be obtained with the minimum of stress for the animal and its population (it is not necessary to kill the animals); another advantage is that it is possible to follow the organisms in time and space, providing priceless information over the efficiency of remedy program applications as well as other intervention and biological conservation measures in polluted sites. We consider that more studies are required in the amphibian populations in Coatzacoalcos with the purpose to identify and quantify the associated risks in the populations of amphibians in the region, as well as their implications in human health and ecosystems.

Acknowledgments

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Implementing Recovery Actions for the Threatened Coquí Guajón (*Eleutherodactylus cooki*): Where to Start?

By [1]Alberto L. López-Torres & [2]Ana V. Longo

In Puerto Rico, more than 70% of all endemic amphibians are classified as threatened or extinct due to habitat loss, climate change or disease (*1*). Therefore, the implementation of species recovery plans is critical to guarantee the long-term viability and stability of existing populations in their protected habitats. However, only three species currently have a Recovery Plan: the Puerto Rican crested toad (*Peltophryne lemur*), the Golden coqui (*Eleutherodactylus jasperi*) and Coquí Guajón (*E. cooki*). Their conservation history provides valuable lessons of failures and successes regarding the implementation of recovery actions, which will improve our strategies for other species in the near future.

Male frog attending an egg clutch laid in the surface of rocks. Photo by: Alberto L. López-Torres.

Learning from previous mistakes

In 1973 the distribution of Golden coqui (*E. jasperi*), one of the few ovoviviparous species of amphibians in the world, was restricted to only 26 ha in privately owned lands (*2*). Local and federal agencies did not respond fast enough to protect and manage these small fragments threatened by habitat destruction and possibly disease, thus contributing to the extinction of the Golden coqui by 1981 (*3*). Three years later, U.S. Fish and Wildlife Service (USFWS) released a recovery plan, but its implementation was too late.

In contrast, the Puerto Rican crested toad (*Peltophryne lemur*) benefited from having part of its distribution occurring within locally protected lands. Although the crested toad was listed as threatened in 1987, conservation efforts started a few years before its recovery plan was officially released in 1992. Recovery actions included the management and construction of breeding pools, intensive population monitoring, captive breeding and public outreach. The protected population became essential for breeding programs, because it supplied 90% of the founder toads used to establish the first colonies (*4*). Supported by local and federal agencies, non-profit organizations (NGOs), zoos and universities, crested toad populations are now gradually recovering.

Coquí Guajón: déjà vu all over again?

Coquí Guajón (*Eleutherodactylus cooki*) was listed as a threatened species in 1997, and its recovery plan was released in 2004. Just like the Golden coqui, its distribution is limited to privately owned lands, and it is facing major threats mainly due to habitat destruction and fragmentation. We have learned from our previous experiences and urged the USFWS to declare critical habitats, which was successfully accomplished in 2007. However, again local and federal agencies did not set aside any lands for the Guajón. Fortunately, the Puerto Rican Conservation Trust, a local NGO, acquired 70 ha securing some of the critical habitats for perpetuity.

Establishing Cooperative Agreements

Proyecto Coqui partnered with USFWS to evaluate some of the factors contributing to the high level of threat of Coquí Guajón. This direct-developing frog is a habitat specialist that depends on caves formed by large granite boulders along streams, and only occurs in the southeastern region of Puerto Rico. Because all of *E. cooki*'s critical habitats are fragmented and confined to private lands, we need to understand how this species responds to anthropogenic threats caused by agricultural and urban development. Using populations from the critical habitats as focal points, we are: 1) monitoring the population status by performing demographic studies, 2) determining the impacts of the pathogen *Batrachochytrium dendrobatidis* (*Bd*) and parasitic tick *Ornithodoros talaje*, 3) assessing the genetic diversity and patterns of gene flow (connectivity) between localities, and 4) developing an outreach program to promote species awareness and to improve habitat quality. Our results will help achieve most of the conservation actions from *E. cooki*'s Recovery Plan designed to protect and stabilize its populations.

Current progress

With the help of students, volunteers and USFWS staff (J.P. Zegarra), we visited eight of the 17 critical habitats, monitored populations, marked individuals and took tissue and skin swab samples. Our surveys allowed us to discover new aspects about its reproductive biology, estimate baseline population sizes and evaluate *Bd* and tick prevalence. We found individuals at all life stages, frogs in amplexus, clutches in non-traditional nesting sites and males

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Coqui Guajón's habitat is formed by large granodiorite boulders with flowing water in southeastern Puerto Rico. Photos by: Alberto L. López-Torres and Louis Santiago.

guarding eggs, indicating that populations are reproductively active. However, we were unable to find any Guajones in one of the critical habitats (Jacaboa Unit, Patillas, PR). This finding concerned us because we recently confirmed the presence of the chytrid fungus (*Bd*) in several of these populations. In addition, we observed multiple skin lesions and severe swelling caused by ticks in heavily parasitized individuals. Our preliminary genetic studies showed little genetic differentiation among the eight populations sampled, based on the mitochondrial marker cytochrome *b*. We are in the process of adding more nuclear to further characterize the population structure of Coquí Guajón.

Public outreach is essential

Accessing the sites required talking with landowners to explain the importance of our visit for the species recovery. Although the Guajón is present in streams in their properties, some of them had never seen or heard it before. For this reason, we developed several educational materials under the slogan "¿Conoces al Coquí Guajón?" ("Do you know the Coquí Guajón?"). This campaign includes informative posters, a website (www.coquiguajon.org), and promotional products, that will help us increase awareness among landowners.

More than just a start

The conservation lessons from the crested toad and the Golden coqui gave us a head start towards implementing some of the recovery actions for the Guajón. Certainly, knowing about the natural history and evolutionary biology will inform future management decisions. However, we still have to promote stronger conservation actions including the acquisition and development of land stewardships, as well as habitat restoration programs.

We removed ticks that were causing lesions and swelling. During the wet season, land slides and collapsed roads may increase the flow of sediments into *E. cooki*'s habitats. Photos by: Alberto L. López-Torres.

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We thank J.P. Zegarra, H. J. Claudio-Hernández, C.A. Rodríguez, O. Monzón, and Dr. P. Torres Morales for assisting us in the field, and L. Santiago provided a photograph of *E. cooki*'s cave habitat. We also thank all the landowners who kindly provided access to their properties.

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Our project for training youth in sustainability based IT can reduce deforestation. Photo: Robert Browne.

A Unique Collaboration

By Robert Browne, Craig Hassapakis & Howard Clark

Amphibian and Reptile Conservation journal is establishing a unique collaboration to develop Internet based biodiversity conservation training for youth in a developing country. Since collaborating with Sustainability for Sarteneja as an Internet project base, Amphibian and Reptile Conservation has achieved a new stature and continues to grow in publications, web visits, and social networking sites. It now has a Facebook reach of over 30,000.

Sustainability for Sarteneja is located in Belize, Central America, and provides an ideal partner for testing innovations in biodiversity conservation. Belize is a safe and stable country in a biodiverse tropical region with a high percentage of forest cover and a high population growth.

Belize is a small country and has a low population density with a population growth among the highest in the in the western hemisphere of 3%; 36% of the population is aged 14 years or younger and 56% aged 24 years or younger. However, there is limited environmental education and very little education concerning sustainability. Belize has the greatest percentage 65% of forest cover of any country in Central America. Unfortunately, the rate of deforestation is increasing in Belize. Our project for training youth in sustainability based IT can reduce the rate of deforestation.

Although a strong environmental consciousness is encouraged in Belize there is not a supportive Internet based information system, formal education, or training in sustainability. Photo: Robert Browne.

In developing countries like Belize the youth are devoted to the use of electronic communication including IT. Many of these are top students and are keen to excel, have family support, and a fundamental respect for the environment; however, they lack training and facilities.

Sustainability IT training will use our established environmental websites for practical experience. We will publish our theoretical

and technical training programs as free Internet Open Access. The use of environmental websites for IT training will immerse the trainees in the presentation of sustainability information. Trainees will learn search engine optimization techniques and the use of IT based information and marketing tools.

Sustainability based IT training offers an opportunity for these talented youth to make a substantial contribution to biodiversity conservation and environmental education while developing and using their Internet skills to further their careers. Trainee's skills will also contribute to the Internet capacity of both the Amphibian and Reptile Conservation journal and Sustainability for Sarteneja. Our recommendations from both private industry and NGOs will provide trainees with a powerful tool to increase employment opportunities. Overall, our global goal is to provide increased knowledge and experience for similar IT projects in the developing world, and through reverse innovation in the developed world.

Sustainability of amphibian conservation is becoming an emerging theme in Central America and the Caribbean Region (*1*). An important consideration in the sustainable management of amphibians is that 30% of tropical biodiversity cannot be preserved through national parks. The preservation of this biodiversity depends on the maintenance of sustainable landscapes in developing areas.

Sustainable management is also becoming the central theme for many biological fields including biodiversity conservation (2). A major research theme of Sustainability for Sarteneja is the comparison of amphibian and reptile biodiversity between blocks of rainforest at various stages of succession before and after slash burn. Sarteneja is ideal for this project as B'alam Ja Way is centered in an area with a mosaic of blocks at various stages of slash burn and forest succession. This field project will be central in our biodiversity focused educational and student activities.

There is less potential for biodiversity conservation when not linked to sustainability. The home of Sustainability for Sarteneja, B'alam Ja Way, provides an ideal base for international visitors. Direct interaction between students and international visitors will provide new knowledge, innovation and incentives. Sustainability for Sarteneja offers students onsite projects that they can document and place on websites. B'alam Ja Way, is landscaped for safely presenting ecological models of all stages of forest regrowth and has a wetland. B'alam Ja Way also has a Maya large ceremonial mound and caves, and demonstrates sustainable building technologies and agriculture. Landscaping has been especially designed to provide habitat for amphibians, however, large mammals such as Baird's tapir and jaguars are found and a plethora of reptiles, mammals, birds, butterflies and other arthropods.

Youth in the developing world face a challenging employment environment unless provided with knowledge based skills. Photo: Robert Browne.

Sarteneja is reliant on traditional fishing for income but catches are declining and alternative industries are needed especially to provide youth employment. Photo: Robert Browne.

This collaboration between two diverse entities, an Internet based amphibian and reptile conservation journal and a sustainability project, has resulted in the testing of a new approach to conservation in the developing world.

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Preventing Amphibian E
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Extinction in Honduras: uras Amphibian Rescue and ion Center

Plectrohyla exquisita. Photo by: Jonathan Kolby.

Duellmanohyla soralia . Photo by Jonathan Kolby.

By Jonathan Kolby

The long-term survival of amphibian species worldwide is becoming increasingly jeopardized by habitat destruction and emerging infectious disease. These threats have been growing over the past few decades and together, are now driving an amphibian extinction crisis. This loss of biodiversity has been especially apparent in Central America, where a growing number of species now require *ex situ* management efforts to ensure long-term survival. Although successful captive management programs have been established in nearby Costa Rica and Panama, no such operations exist in Honduras, where an amazing diversity of Critically Endangered (32 spp.) and Endangered (24 spp.) amphibian species are found (*1*). In response to the growing need for applied conservation efforts in the country, the Honduras Amphibian Rescue and Conservation Center (HARCC) is now being established to protect Honduran amphibian biodiversity.

Jonathan Kolby with an adult *Plectrohyla exquisita* in situ. Photo by Sara Ramirez.

This new long-term program is designed to ensure the survival of Critically Endangered amphibians found in El Parque Nacional Cusuco (PNC), a small fragment of montane rainforest located in the Sierra de Omoa range of the Merendón mountains in northwestern Honduras. PNC is surrounded by deforested land modified for agriculture, and provides a small "island" (approx. 200 km^2) of critical cloud forest habitat for 16 Endangered and Critically Endangered amphibian species. Since 2004, the UK-based conservation group Operation Wallacea has been monitoring the biodiversity of PNC and surveys have indicated an overall decline in the presence of stream-associated amphibians. Concurrent disease surveys were initiated in 2007 and many species demonstrated a high prevalence of infection with amphibian chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) (2). This fungus is responsible for amphibian chytridiomycosis, an emerging infectious disease associated with global amphibian declines and extinctions, even in remote locations where habitats remain intact. Enigmatic declines of amphibians in north-

Montane cloud forest habitat within PNC. Photo by: Jonathan Kolby.

ern Central America appear to have begun in the 1980's to early 1990's, and a retrospective study indicated the presence of *Bd* in PNC by 1996 (*3*).

Three Critically Endangered species in PNC have been selected as priority candidates for conservation management by combining information in IUCN Red List assessments, results from a five-year *Bd* infection survey, and field observations of habitat degradation. The species to receive attention include: *Duellmanohyla soralia*, *Plectrohyla dasypus* and *Plectrohyla exquisita*. PNC is recognized by the Alliance for Zero Extinction as the only site of existence for *P. dasypus* and *P. exquisita*, and also represents the largest stronghold of several fragmented localities for *D. soralia*. A recent catastrophic rise in illegal deforestation combined with a severe loss of juvenile amphibian recruitment caused by chytridiomycosis are together impacting each of these species and pushing them closer towards extinction. The situation is especially grave for *P. dasypus*, which is reported to have already experienced, "... a drastic population decline, estimated to be more than 80% over the last ten years, inferred from the apparent disappearance of most of the population..." (*4*).

The HARCC will address this conservation challenge and work to rebuild declining amphibian populations through a combination of *ex situ* and *in situ* management platforms while placing strong emphasis on local capacity-building. Captive management activities will occur at Lancetilla Botanical Garden and Research Institute, in Tela, Honduras where biosecure isolated amphibian rooms will house up to 600 amphibians in a *Bd*-free environment. These amphibian rooms are being modeled after those located at the Henry Doorly Zoo in Omaha Nebraska, employing a high standard of biosecurity to control disease. The core activities of this rescue effort will take two forms: 1) Annual population supplementation via head-starting to increase the number of wild animals that may survive to adulthood, and 2) Establishment and maintenance of long-term captive assurance populations to buffer against the risk of extinction in the wild.

Head-starting activities will entail the collection of tadpoles and recently metamorphosed frogs from PNC, treatment for *Bd* infection and maintenance in a biosecure facility, and subsequent reintroduction of healthy adults back to original collection sites. Long-term data previously collected in PNC suggests that the adults of these species possess increased resistance to chytridiomycosis and are able to survive *Bd* exposure, but that most larvae are highly susceptible and few survive far beyond metamorphosis. This observation is not unexpected, as larval and adult frogs have also been found to exhibit a similar pattern of susceptibility in other *Bd* investigations (*5,6*). Therefore, annual supplementation of healthy adults back into remnant wild populations is likely to be a highly effective technique to increase the number of breeding adults surviving in the wild, despite exposure to a *Bd*-infected habitat in PNC. This supplementation is similarly expected to promote an increase in the volume of offspring produced by these species, in turn increasing the number of animals that survive past metamorphosis. In this fashion, reintroduction can both directly and indirectly re-establish declining adult populations, and eventually may reach self-sustaining levels in the wild that can compensate for the disease-driven additive mortality. All reintroduced frogs will be marked with either alpha-numeric visual implant elastomers or PIT tags, to allow every individual to be recognized in the field. For the

Clockwise from upper left: *Bothriechis marchi*, *Plectrohyla dasypus* (recent metamorph), *Plectrohyla exquisita* (brown phase), *Plectrohyla exquisita* (green phase), *Duellmanohyla soralia* (recent metamorph). Photos by: Jonathan Kolby.

One of three focal study sites in PNC where HARCC efforts will be concentrated. All three priority species (*D. soralia, P. dasypus*, and *P. exquisita*) are found in this river system, where *Bd* is highly prevalent. Photo by: Jonathan Kolby.

duration of this program, annual mark-recapture surveys will be performed in PNC to continually monitor adult frog populations, measure the survival of reintroduced frogs, and evaluate the overall impact of management efforts. In addition, some adults will be retained at HARCC to develop captive assurance breeding populations and ensure the long-term survival of each species in the event of sudden population crashes, which remain likely.

Jonathan Kolby swabbing *Plectrohyla exquisita* to detect *Bd* infection. Photo by Sara Ramirez.

This program will provide one of few examples whereby amphibian populations threatened by *Bd* are protected from extinction in the wild, and not solely in captivity. Similar head-start and reintroduction efforts have been performed by the Taronga Zoo (Sydney, Australia) and appear to be successfully recovering wild populations of the Southern corroboree frog (*Pseudophryne corroboree*) decimated by *Bd* in Kosciuszko National Park, Australia. Recent efforts to reintroduce the Kihansi spray toad (*Nectophrynoides asperginis*) to Kihansi Gorge, Tanzania also appear successful thus far, again despite being returned to a *Bd*-infected habitat. Although *ex situ* captive assurance programs can prevent species extinction, limited space and financial resources dictate this to be a feasible long-term solution only for a small fraction of species that may require assistance.

Reintroduction efforts are also needed in order to maintain healthy ecosystems in locations where amphibians have suffered dramatic declines or extirpation. Amphibians provide a significant food source for other wildlife species in tropical ecosystems, such as the Palm viper (*Bothriechis marchi*) in PNC, which has a specialized diet of frogs and is now believed to be in decline in response to the lower abundance of amphibian prey (*7*). The activity and feeding behavior of larval amphibians has also been shown to affect aquatic ecosystem structure; their disappearance is likely to promote algal blooms, reduce freshwater aquatic invertebrate diversity and reduce water quality. Therefore, *ex situ* management programs that explore possibilities for active reintroduction alongside captive assurance propagation will provide the means to both restore wild populations and ecosystem services amphibians provide. It is unknown whether population recovery via the methods described herein would be effective for a broad range of species affected by chytridiomycosis, but the strong promise for long-term recovery via relatively basic techniques warrants serious consideration. Therefore, the HARCC aims to provide one of the first such examples of integrated *ex situ* and *in situ* species management in Central America.

The distribution and prevalence of *Bd* throughout Honduras is largely unknown aside from PNC (*2*) and Parque Nacional Pico Bonito (*8*), the only documented localities, but it is likely that amphibians in many of the country's national parks are similarly being affected. Additional observations of amphibian declines throughout the region have been reported (*9,10*), and despite widespread habitat destruction, the influence of disease cannot be ruled out as a significant driver of these declines. There is a great need for additional research to map the distribution of *Bd* in all of the country's national parks to develop information that can be used to prioritize future candidate species in need of *ex situ* conservation. HARCC will serve as a model for effective species management of the Critically Endangered species of PNC, but it is expected that additional facilities within Honduras will become necessary to protect species of other regions once the true extent of *Bd*-associated declines in Honduras becomes recognized.

Construction of the HARCC will take place in August 2013 and the first collection of larval amphibians is targeted for August 2014 (pending funding to support the long-term care and maintenance). For more information about this program or for future opportunities to become involved in this project, please contact Jonathan Kolby (Jonathan.Kolby@my.jcu.edu.au).

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Captive Breeding of the Frog *Mannophryne herminae* (Anura: Aromobatidae) and Releases to the Wild in Venezuela

By [1]César Molina, [2]Claudia Camacho & [3]Juan Vicente Hernández

The frog *Mannophryne herminae* (Anura: Aromobatidae) (Fig. 1) has been categorized as Near Threatened by the IUCN and Red Book of Venezuelan Fauna (*1*, *2*), due to human intervention in most of its range distribution. This has attracted interest as an object of study. This species endemic to Venezuela is small, diurnal, territorial and presents sexual dimorphism in size and color. Shows preference for rocky streams with small waterfalls, in areas covered by woody vegetation in mountainous areas. Their geographical distribution covers the entire Central Coast Cordillera, in northern Venezuela, between 351 and 1,610 m.a.s.l (*3*, *4*).

Studies of *M. herminae* in the field have encompassed population, reproductive, trophic and ecological aspects (*3*, *5*). However, there are no studies on ex situ breeding and captive management at different stages of the life cycle. These aspects are essential for addressing conservation actions involving the introduction of individuals to the wild, either to strengthen existing populations or to restock sites previously inhabited by this focal species (*6-8*).

Here we report results from a study to investigate the basic requirements for breeding and maintenance of larval forms of this species in ex situ conditions. We developed a set of experimental protocols with tadpoles of *M. herminae* at stages 25 and 26 (Fig. 2), taken from the Sabaneta creek at Fundo Ecológico Santa María (10°22'53.82''N; 66°47'21.97''W, 999 m.a.s.l). We evaluated the effect of food type, population density, temperature and chemicals on responses variables of growth, size, weight, survival and time to reach metamorphosis (*9*).

We evaluated five foods whose protein composition varied between 33 and 35% and fat composition 3.4 to 5.0%. Tadpoles had increased survival, higher rates of growth and development with the commercial fish food brand Kantal™. Moreover, higher survival, rates of growth and development were achieved at optimal densities of 3 ind/l and at an average temperature of 25 °C.

The results and learning best practices from this experience could be used as preliminary captive breeding protocols for purposes of reintroducing *M. herminae* in some locations within the historic range (Coastal Cordillera Range) where this species has disappeared in recent times due to habitat destruction and urbanization pressures, such as some of the streams that flow from the National Park Guaraira Repano leading into the metropolitan area of Caracas. Furthermore, this protocol can be replicated for other species with some degree of increased threat, whether they belong to the same genus or species of other genera that share similar aspects of its biology and ecology.

However, ex situ conservation can be an optimal strategy when it complements in situ conservation efforts (*10*, *11*), such as reintroduction and reinforcement of populations with individuals bred in captivity (*12*). That is why we decided, in a second phase of this study, to experimentally reinforce preexisting population of that species. We raised tadpoles using optimal breeding protocols previously evaluated. Again, the tadpoles were from the Sabaneta creek, which were raised at the Caracas Terrarium facilities located in the Park G. Francisco de Miranda. Individuals were raised to metamorphosis (Fig. 3). Small frogs were marked as two different batches (225 and 175 individuals, respectively) and were fed with ants and crickets for at least two weeks (Fig. 4), before release to the same creek (Fig. 5).

The study has just started to track movements of marked indi-

Fig. 1: *Mannophryne herminae* male with tadpoles. Photo: C. Molina.

Fig. 2: Tadpoles of *M. herminae* in a body of water with sandy bottom. Photo: C. Molina.

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Fig. 3: Facilities captive breeding: A) Containers with individuals, B) Concealment with pieces of cardboard to avoid stress of individuals. Photos J. V. Hernandez.

Fig. 4: A) Individuals with tail fully reduced. B) Individuals feeding on ants. It details the piece of paper with honey used as a trap to catch ants. Photos J. V. Hernandez.

Fig. 5: Liberation of individuals of *M. herminae* and microhabitat data collection. Photos A. Grajal.

viduals, which involves an survey every week for the first month after release and then a monthly evaluation. In each of these monitoring events each animal recaptured is being measured in its body length and weighted in order to assess their condition through an index. Distance from the release point will indicate the patterns of spatial dispersion of released individuals and microhabitat use. These data will be compared with those obtained from unmarked individuals recorded during the monitoring (*13*).

Finally we should note that this study is embedded in a project to develop and implement protocols for breeding and release of endangered amphibians in an ex situ conservation program for Venezuela. This study will allow, in the future when conditions are ripe, to work directly with species categorized at a higher level of threat, such as *Atelopus cruciger* (Anura: Bufonidae).

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Amphibians of the Tama National Park: Hidden Biodiversity and New Salamander Species from Colombia

By Aldemar A. Acevedo, Rosmery Franco & Karen Silva

Colombia has 720 species of amphibians, occupying the second place in amphibians diversity in the world, yet 213 of these species are listed as Threatened species under the IUCN Red List categories (as either Extinct, Critically Endangered, Endangered or Vulnerable). In addition, many regions in Colombia have a serious lack of information, leading to 129 species of amphibians listed as Data Deficient (*1*). The lack of studies particularly on natural history, distribution, ecology and diversity, have limited the accurate assessment of levels of threat, and therefore delayed the design of management plans and conservation strategies (*2*). This is the case of Tama National Natural Park, a legally protected area of 48,000 hectares located in the northeast of Colombia, in the State Norte de Santander (Fig. 1). The knowledge of the diversity of amphibians in this region is particularly poor; only five species of amphibians were known to science prior to the "Tama Amphibians Project". There are multiple reasons that explain this lack of information, on the one hand the political instability and rules and regulations at the national level, that make it difficult to perform scientific research, although this has improved considerably in recent times, and on the other hand the absence of specific research groups and the consequent deficiency of economic resources and/or human capital seeking such resources. With this in mind, since 2010 our team has been working in various localities of the Tama National Natural Park, with the objectives of identify and prioritize species of amphibians that are at risk of becoming extinct and evaluating threat factors such as climate change, habitat degradation and, particularly, identifying locations and species infected by the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*), considered the main cause of the global decline of amphibians (*3*,*4*). After two years of field work, we have identified 34 species of amphibians within the park, including 10 new records for the region, three new records of *Pristimantis* species for Colombia and two species of salamanders new to science, which is a clear but still small example of the hidden diversity of this region.

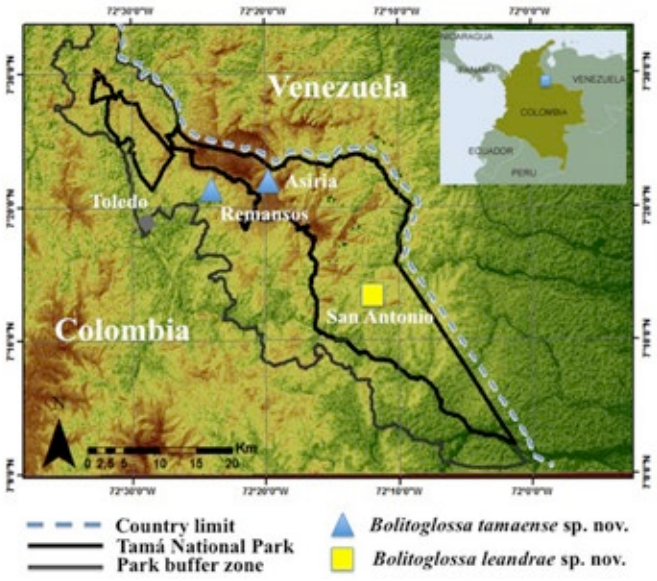

Fig. 1: Localities of new species of *Bolitoglossa*. National Natural Park Tama, State Norte de Santander, Colombia.

New species of salamanders

Discovering a new species is something that many of us working with animal or plant species desire silently. In our work inventorying, monitoring or working in conservation assessments of species, we spent long hours exploring remote places and living with local communities. These spaces become our home with lush forests and rivers nestled between high mountains, which represent a complex mosaic of nature and human activities that move in the dynamics of natural resource protection and the use and knowledge of them.

The most important discovery in our study occurred in 2010 when we found two species of salamanders, which immediately called our attention, because we knew beforehand that species of this group had not been reported for this region. However, the surprise could not be bigger when noting they exhibited very apparent differences when compared to species of salamander known in Colombia and Venezuela, reason for us to catalogue them as undescribed species of the genus *Bolitoglossa*. Later these species became the recently described *Bolitoglossa tamaense* and *B. leandrae* (*5*) (Fig. 2). Twenty-two species of salamanders have been described in Colombia so far, more than any other South American country (*1*, *6*). However, very little is known about their natural history, ecology and distribution, as they are often known from just the type locality, and even the type specimen or series.

Fig. 2: A. *Bolitoglossa tamaense*. B. *Bolitoglossa leandrae*. Photo: Aldemar A. Acevedo.

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Characteristics of the new species

Bolitoglossa tamaense is a species of medium size, with an average SVL of 47 mm. It has a wide range of variations in coloration with bodies varying from yellow to orange in color, and some individuals having the tail, head and legs slightly reddish and others with a cream colored back with dark gray color. *Bolitoglossa leandrae*, on the other hand, is one of the smallest species of salamanders with an average SVL of 30.3 mm and a less variable coloration than *B. tamaense*. *Bolitoglossa leandrae* has tail and dorsal torso dark brown, with thin yellow stripes along the length of the body; the ventral area is predominantly gray with small brown spots. The dorsal surface of the female is brown and reddish copper.

Distribution and natural history

The two species of salamanders have similar behavior with slow and cautious movements going unnoticed in the forests. Individuals are usually refuged under rocks, litter or protected in the roots of plants, and will only begin to leave their refuges when the sun goes down and there is no heavy rain. The will claim plants, to finally settle on the ferns, bromeliads and low vegetation, where they stay most of the night, either in search of prey or a reproductive mate.

Bolitoglossa Tamaense is only known from high Andean forests, on the eastern flank of the Cordillera Oriental, in the town of Toledo, State Norte de Santander, Colombia (Fig. 1). It has only been recorded from two localities located in small patches of forest associated with streams and riparian vegetation, one population has been found at 2,000 m of elevation, and the other population at 2,700 m. *Bolitoglossa leandrae* is only known from the lowland of the Tama Park. Only one population has been recorded in small patches of secondary rainforest at 600 m. Expeditions to altitudes ranging from 600 to 3,300 m within the park revealed no additional localities for both species of *Bolitoglossa*.

A brief history of *Bolitoglossa leandrae*

The journey is long, through dirt and inaccessible roads that lead to a small village of no more than ten houses, where the field guide awaits us. Our guide is a young farmer who will help us in our work, along with his two horses, on whose backs we transport our field equipment and supplies. A short break and we embark on a new path to the study site, which is situated some six hours walking, passing through different agricultural landscapes, forests and rivers. Step by step it is revealed the majesty of nature, green in a variety of shades that blend with the pleasant sounds of a large number of creeks that merge into the distance with echoes of birdsongs. Night falls and we have reached a house immersed in the mountains, enlightened by a few candles already depleted and some frogs croaking in the distance. The pleasure of being in that place is absolute, smelling great tranquility and a welcoming hug.

We are greeted with a cup of "*Guarapo*" a fermented beverage made from sugar cane cultivated in these rural areas, which retrieves us almost instantly from the grueling journey. Several children live in the house and among the dim candlelight are approaching with curiosity. The oldest, an 11 year old girl named Leandra (Fig. 3) remains in the distance, but her eyes are brimming with the desire to ask and ask. We explained to the inhabitants of the house our purpose and why we are in this place "we come to find frogs and toads" we tell them, they smile, and we patiently tell them with laughter and anecdotes our purpose and the importance of our work.

The following days are of hard work, waking up early and walking around the area. It's been five days and in the company of Leandra and other children we already have found incredible amphibian species, many of whom we do not know. The six night as late, we reached a small patch of forest and in the middle of the walk we watched with astonishment a small animal perched on a leaf, the excitement was indescribable, we found a salamander, it was the first time we saw one live. The energies were recharged and we did not stop looking through most of the night around that area, finding many individuals. Months after, those creatures that amazed us that night were confirmed to be a new species, naming *Bolitoglossa leandrae*, in honor of the curious child who knew the hidden beauty that haunted her house in the farthest mountains of the Tama Park in Colombia.

Fig. 3: Leandra Mojica, a girl living in the rural area of San Antonio, the type locality of *B. leandrae*. She represents the children of the rural area in the Tama National Park, who show so much enthusiasm for learning about amphibians. Photo: Rosmery Franco.

The conservation threats

The Tama Park has a large hidden diversity of amphibian species; our project has reported several species that were not known to this area, two new species to science and still several remain to be described. Moreover, we have been able to identify some factors that threat the existence of amphibian species, which merit urgent attention. However, information on the conservation status is still incipient. Out of the 34 species of amphibians recorded 23 species that comprise seven families of amphibians are infected with *Bd* (Fig. 4), distributed from tropical forests at 600 m, up to the paramo at 3,200 m of elevation. Additional factors such as the clearing of forests, is probably the main threat facing amphibian species in the region. New species of salamanders are found in small isolated patches of forest and surrounded by a mosaic of agriculture and livestock. Furthermore, many salamander specimens were diag-

nosed positive for *Bd*, which increases their risk of becoming extinct in the short term.

Conservation actions

Social programs have focused on working with local residents through the organization of seminars and workshops, in order to facilitate their integration in the conservation strategies and helping them to make direct contact and better understand current conservation issues. Today we have an overview of the biodiversity and the threats affecting amphibians of the park. Educational activities and active participation of local communities have created a natural resource ownership, generating an exchange of knowledge. Working with local communities, environmental agencies and the education sector open the possibilities to develop a management plan for threatened habitats of the Tama Park, with the aim of increasing the programs for long-term restoration of forests to ensure the survival of amphibian species that live in these habitats.

Acknowledgments:

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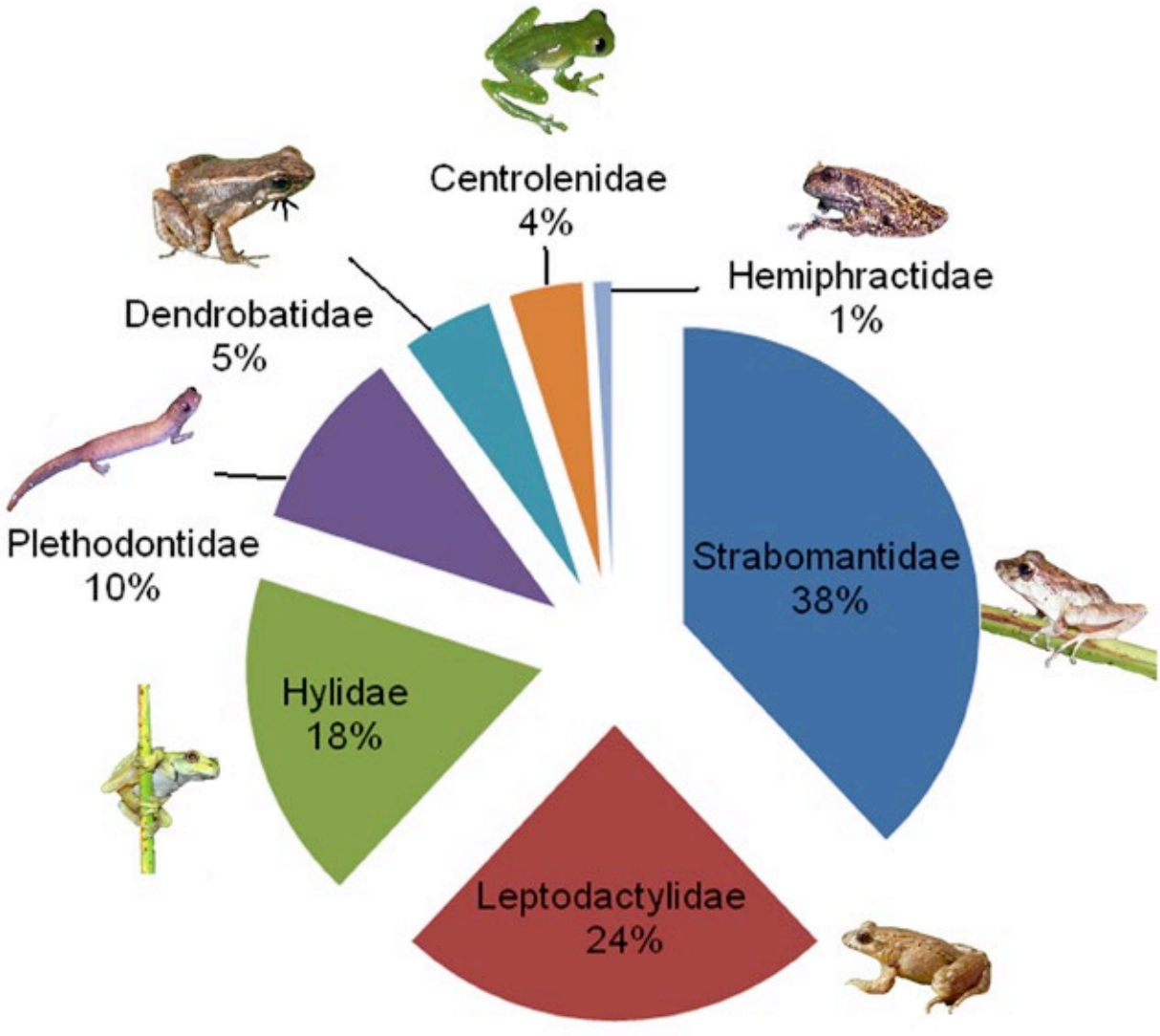

Fig. 4: Percentage of amphibian families infected with *Bd* in the Tama Park

Lethal and Lovely: An Interview with the Founders of the Golden Poison Frog Reserve

The Golden poison frog. Photo: Fundación ProAves.

By Manuel Beterams

" I would like to dedicate this article to my father, René Beterams, who has passed away too young. He has taught me the value of pursuing knowledge and he has taught me the love for nature; for this I am eternally grateful to him. And I will always remember him as a loving and supporting man, may he rest in peace."

If you ever would be so happy to stumble upon a Golden poison frog in the wild, chances are you are more enticed by this creature than frightened of it. With a Latin name like "terribilis", referring to how terrifying this creature is, you might expect to see a more ferocious animal. However it definitely has earned its Latin name considering it is the most poisonous animal on earth. Therefore, it would be very wise not to touch it; the skin of one frog contains enough poison to kill 20 grown men. Its toxicity has proved to be a very useful feature for indigenous people living in the western part of Colombia: these people load their blowguns with darts that are made toxic by simply rubbing them against the backs of living Golden poison frogs. This custom is very neatly described by Myers *et al.* (*1*) in a lengthy article announcing the discovery of the species in 1978. Why it is so toxic and where it derives its toxicity from is still up for debate: long it was believed that ants in their diet were the primary source of the alkaloids that make up the poison, however the source now most likely is a small beetle that is part of the species' diet. I will leave the debate with this and will focus now on conservation efforts, because unfortunately, even being the most poisonous animal on earth has not safeguarded this species from humanity's march up on the Olympus. For it is our species that is continuously extending its sphere of influence over the earth and thereby leaving less and less habitat for other species to occupy. With its global population declining the Golden poison frog, like many other amphibians, seems to have a grim future ahead.

Luckily there are people who try their best to conserve this species; in May of last year Fundacion ProAves founded a reserve, with the help of World Land Trust, American Bird Conservancy, Global Wildlife Conservation, Conservation International and the IUCN SSC Amphibian Specialist Group, in the Choco rainforest of Colombia, named the *Rana Terribilis Amphibian Reserve,* that is comprised of about 50 hectares of primary rainforest. The reserve is situated near the bank of the Timbiqui River, a short distance from the municipality of Timbiqui within the Cauca Department. This is the first effort to save this species, because until then the Golden poison frog was completely unprotected. Because I was interested to know more about this species and the reserve I contacted Fundacio ProAves and had the pleasure of asking some questions to Luis Gabriel Mosquera, subdirector of the natural reserves in this area and in charge of the *Rana Terribilis Amphibian Reserve* . He provided me with some answers about the reserve and the state of the species.

How many individuals are estimated to live within the Reserve?

"At this moment it is very hard to estimate the number of individuals of this species within the reserve, because we haven't been able to carry out a proper investigation that would give us the necessary data. Within our team we have people at the moment that are specialized in collecting these data. Therefore, we hope to obtain better estimates in the near future."

Is the Reserve large enough to sustain a healthy population?

"No, with only 47 hectares, the reserve is probably too small to ensure a healthy population and save the species, but fortunately we are planning to acquire a larger area. And as well, we count on the people of the surrounding communities to become more involved in conservation efforts and obtain a better knowledge of the species, thereby promoting the conservation of the frog."

How do you manage the Reserve? Any specific management you use to promote the survival of the frogs?

"At this moment we are developing research activities that are aimed at increasing knowledge on the life history of the frog. Also, we have other activities planned that should help us with conservation of the species, some examples are:

- Developing educational activities including trips into the field with youngsters from the region.
- Getting the word out to the surrounding communities about the location of the reserve and the activities that are being implemented there, to make sure that the reserve is respected as a protected area within the surrounding area. In this way we hope to bind people to our conservation efforts for the Golden poison frog and other species in the area.
- We are trying to connect with other institutions that could be helpful for the project, like the mayor of Timbiqui (to create forested zones), community leaders (to create conservation zones within their areas), the Colomian institute for rural development (INCODER), etc..."

What are the greatest threats to this species?

"Among the greatest threats that are imposed on this species are the human pressure on its environment and activities like deforestation that are reducing the area of habitat for this species. That's why it might become hard to find this species in its natural habitat in the future." (in addition to this, the press release of the World Land Trust mentions that "improved security in the region has increased deforestation, illegal gold-mining, illicit coca cultivation and logging") (2).

What is for you the most fascinating thing about this species?

"What I find fascinating is that evolution has enabled such a small and apparently harmless creature to become one of the most poisonous vertebrates in the world. "

The Golden poison frog. Photo: Fundación ProAves.

View of the Rio Timbiqui and the *Rana terribilis* reserve. Photo: Fundación ProAves.

Why is it so important to preserve this species in the reserve?

"This species is known as the most poisonous vertebrate on earth. Moreover, this species is endemic to the coast of Colombia and Panama, which indicates that these frogs cannot be found in any other part of the world. Unfortunately the Golden poison frog is an Endangered species and therefore conservation is of the highest priority. "

As you have been able to read above the people managing the *Phyllobates terribilis* reserve are trying to spread the conservation message throughout the community and trying to involve as many institutions as they can. I think it is very wise of them to look for that support in the surrounding communities and the government. Because as well as it is us people that are putting an increasing pressure on the habitat of the Golden poison frog, it is us as well that can make a valuable contribution to the survival of the species. This is my small contribution towards that goal and I want to ask you to make your own contribution by spreading the word and maybe making a financial contribution. To do the latter, go to the website of Fundacion Proaves or the World Land Trust.

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Ecosystem Services Provided by Neotropical Amphibians and Reptiles: A General Overview

By [1]Anyelet Valencia-Aguilar, [2]Angela M. Cortés-Gómez & [3]César Augusto Ruiz-Agudelo

The strategy of "Colombia's Natural Capital" (Capital Natural de Colombia), https://sites.google.com/site/capital-naturalcolombia/iniciativa-capital-natural-colombia, was initiated in 2010 by Conservation International Colombia and the Colombian government. Within the development framework of this strategy, a series of studies have been initiated to gain knowledge of this natural capital. In 2012, part of this exercise focused on understanding the role of certain elements of biodiversity, such as amphibians and reptiles, and their direct benefits to human wellbeing. The goal of this review was to identify and describe the role of amphibians and reptiles in neotropical ecosystems, in addition to providing a general overview on their importance, not only for the functioning of ecosystems, but also for human wellbeing.

Ecosystems provide societies with a significant amount of goods and services through regulation processes (biological control, pollination, seed dispersal, climate regulation, soil stabilisation), supply of products and services (food, fibres, medicines), support systems (nutrient cycling, soil formation, primary production) and cultural benefits (aesthetic, educational, spiritual, recreational) that improve human wellbeing (*1*,*2*). In this sense, amphibians and reptiles play a key role in the ecosystem and in the human wellbeing. They can be herbivores or carnivores regulating the dynamics of aquatic ecosystems by reducing natural eutrophication or certain insect populations in the terrestrial habitats. Certainty some of these insects are hosts for human pathogens; others may affect crops of economic importance (*3*). Similarly, amphibians and reptiles can potentially play a role in energy flow and nutrient cycling in both terrestrial and aquatic ecosystems (*4*), seed dispersal (*5*,*6*) and pollination of certain plant species (*7*).

***Chelonoidis carbonaria*, dispersal agent of numerous seeds in different terrestrial neotropical ecosystem. Photo: A. M. Cortés-Gómez.**

Roles played by different species within ecosystems can directly or indirectly influence the function of ecosystems and a portion of these functions translate into services that are used or enjoyed by society (*8*). These ecosystem services or values are used by people according to their preferences and needs. In this instance, the ecosystems and their services have a value for human societies because a person can obtain direct or indirect benefits from them (*1*).

Provision Services: Food and Commerce

Humans obtain safe and nutritious food from ecosystems and ecosystem components to fulfil their nutritional needs (*9*). In this way, wildlife has been a natural resource for societies for a long time, and even today, numerous species of reptiles are considered important food sources, particularly as sources of protein (*10*,*11*). The life histories of amphibians and reptiles make them adequate food sources in lowland tropical areas (*12*). In addition, leather from several species of reptiles are sold in the international market to make clothes and accessories, such as shoes, shawls and purses, as well as decorations and furniture, such as rugs, amulets and trophies(1). For example, the countries in the neotropical region have a great diversity of amphibians and reptiles, and such species as frogs, salamanders, lizards, snakes, turtles and crocodiles are sold for their skin and meat, and others are sold live as a pet and for research purposes (*13*).

The meat and eggs of the turtles, such as Chelonoidis carbonaria, Kinosternon scorpioides, Podocnemis expansa, Rhinoclemmys melanosterna and crocodiles such as Caiman crocodilus, Melanosuchus niger, Paleosuchus trigonatus, etc., have been hunted by rural communities for centuries. The kills are used for human consumption (meat), and the skins and other parts are used for different purposes. For example, in countries like Argentina, Bolivia, Brazil, Colombia, Mexico, Paraguay and Venezuela, the consumption of "wildlife meat" is a common practice in rural areas, comprising up to 70% of the protein consumed by a family (*14*). Also, the hunt of lizards (Tupinambis), to sell the skins is a practice that not only provides income to these people but is also considered an important activity for the economy of these countries, being valued in millions of dollars per year in exports to the United States, Canada, Mexico, Hong Kong, Japan and certain European countries (14,15).

Regulation Services: Pollination, Seed Dispersal and Biological Control

In the last decades, numerous researchers have evaluated the role of amphibians and reptiles in process like pollination and seed dispersal in terrestrial neotropical ecosystems, identifying approximately 14 species of this groups (frog, lizards and turtles) responsible for the dispersal of seeds of at least 56 species of plants (*5*,*6*).

In recent decades, there has been an interest in implementing handling methods less aggressive like biological control, to help to reduce the economic and environmental costs of pest species (16). In this sense, the role of generalist predators, such as the amphibians, for biological control has become important in the past several years, because it has been shown that species in this group consume

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Caiman crocodiles (left) and *Iguana iguana* (right), species used as a protein source by many rural communities in Neotropics. Photo: A. Valencia & A.M. Cortés-Gómez.

numerous arthropods including vectors for human diseases and harmful for the crops (*3*).

The roles of the amphibians and reptiles in processes such as nutrient cycling and energy flow could be helping to maintain the structure and function of the ecosystems that they inhabit and may also influence the stability of different ecosystems and thereby benefit human societies. This review is part of a series of publications in which we analyse a large amount of literature and information supplied by national and international research in the Neotropical region (http://www.conservation.org.co/?page_id=5415).

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The Importance of Floating Meadows for Amphibians in a Flooded Forest

By Katy Upton

Peru has very high levels of biodiversity, including over 520 species of amphibians (*1*). This diversity is due to Peru's wide range of habitats from arid deserts, high Andean plateaus to tropical rainforests. Amphibian diversity is being assessed as part of long-term biodiversity assessments in the Pacaya Samiria National Nature Reserve (PSNNR) within the Loreto region of Peru in the north-east of the country. PSNNR consists of low lying varzea forests which are seasonally inundated with floodwaters. This dynamic environment creates a wide range of niches which supports many species. Unlike many tropical forests this area of Peru does not have defined wet and dry seasons. Instead, seasonal inundation of floodwaters is the norm. The PSNNR is one of the largest protected areas in Peru and can endure several months in which 95% of the land is under water.

This reserve contains a variety of habitats, which in turn support many different species. Some of these habitats can be seasonal or temporary and therefore can be easily overlooked, or even ignored, as they may be deemed unimportant. However, some can be very important; the floating meadow is one such habitat (Fig. 1). Found in great abundance within PSNNR, floating meadows are vital for many different species from birds to fish (*2*). Floating meadows are made of herbaceous water plants and grasses which have free floating root systems, enabling movement with the rising water levels. They often grow in shallow, nutrient rich water along the banks of rivers or around lake edges (*3*). Some of these meadows are in direct sunlight, and therefore are able to form vast mats of vegetation with a deep root system. This creates a habitat with very high invertebrate diversity. Consequently, many vertebrate species can be found here taking advantage of the food available (*2*).

After heavy rainfall the floating meadows can break down and wash out of lakes and channels and into the main river channel where they can accumulate. Sections of the floating meadow which break away create rafts which are transported down river (Fig. 2). Amphibians can become trapped on these rafts and dispersed (*4, 5*). Very little research has been undertaken on the role of floating meadows as amphibian habitats or as a mechanism for dispersal.

Fig. 1: The floating meadow habitat along a lake edge within the PSNNR. Photo by: Katy Upton.

Durrell Institute of Conservation and Ecology (DICE), University of Kent

Fig. 2: The author surveying a floating raft found on a Lake in PSNNR, a large potentially gravid female *Dendropsophus triangulum* was found on the raft. Photo by: Isabel Rogers.

Some of the original work was completed by Junk (1973), who surveyed a wide range of species found on floating meadows as well as surveying the plant diversity. This work, however, only mentions amphibians once, stating that they were rarely encountered on floating meadows (*2*). Working on the Solimoes River near Manaus, Brazil, Hodl (1977) found 15 amphibian species on floating meadows and investigated breeding activities (*6*). Schiesari et al. (2003) surveyed floating rafts rather than whole meadows on the Solimoes River and observed nine species.

Amphibian surveys

Research on amphibians with the PSNNR reserve has been undertaken by DICE, Operation wallacea students and Earthwatch volunteers. This research has observed 65 frog species, one salamander and one caecilian representing twelve families of amphibians.

These species are seasonal in their activities. In 2012 surveys were completed during the high flooding (April), transitional period (May-June) and during the low water (September). Surveys were conducted on the same transects across all seasons, and the amphibian assemblages changed dramatically. Of the 39 species caught in the forests, 19 were found in only one flooding season, nine in the high water, nine in the transition period and only one in the low water. Thirteen species were found in two seasons while only eight species were found across all three seasons. Many of the more common species are associated with the leaf litter of the forest floor and therefore were unable to escape the flooding. Instead they were observed on debris on the water surface and leaves just above the flood water (Fig. 3). However, most of the species only found in a single season (five during high water and six during the transition period) are hylids which have the ability to move up and down in the canopy in relation to flooding, potentially explaining why 19 species were only observed in one season.

In 2012, more intensive surveys were carried out on floating meadows. Seventeen species were found on the floating meadow habitat between March-September 2012. Of these, six species have not yet been recorded on this habitat (*5, 6*).

Fig. 3: A very unusual observation of *Rhinella margaritifera* above the forest floor, it may have been displaced due to the extremely high flooding observed in 2012. Photo by:

Fig. 4: An unusual encounter of *Pipa pipa* as it is a fully aquatic species which is rarely captured. This individual was seen swimming very close to the surface of the water in a channel while undertaking a floating meadow survey. Photo by: Katy Upton.

Thirteen out of the 17 species found on floating meadows were hylids. These frogs are thought to be arboreal often only venturing down to lower levels and the forest floor to breed (7). Only three other families were represented in this habitat, two by only one species and the third by only two species. Bufonidae was represented by *Rhinella marina*, this species and the two leptodatylid species; *Leptodactylus leptodactyloides* and *Leptodactylus petersii* are probably only using this habitat as a refuge during the high waters. This is supported by the fact that neither species was found showing mating behaviour, also no tadpoles or frog spawn where found on this habitat. One other species *Pipa pipa* (Fig. 4) was observed swimming in the river alongside the floating meadow.

Of the hylids found on floating meadows those found in the highest abundance were *Dendropsophus triangulum* and *Hypsiboas punctatus* (Fig. 5). All other hylids were found in much lower abundance. As only four of the hylids were also found on terrestrial transects, they appear to be floating meadow specialists. Rather than descending from trees to use temporary pools on the forest floor, hylids may be using floating meadows for breeding. In the flooded forests of PSNNR these temporary pool habitats are less available. Therefore if hylid frogs climb down to breed when the forest is flooded there is likely to be high predation of their tadpoles. Fish migrate into the forests to spawn during the high water and the juvenile fish remain here to develop as there are fewer large predators within the flooded forest and more leaf litter/debris to

Fig. 5: The two most common species found on the floating meadow habitat *Hypsiboas punctatus* (above) and *Dendropsophus triangulum* (below). Photos by: Katy Upton..

provide cover. The lakes and channel will at this time contain fewer predators for tadpoles and the large root systems of the floating vegetation provide protection and food. Evidence of breeding was observed in many of the hylid species found in this habitat including frogspawn, individuals in amplexus and calling males (Fig. 6).

Rafts are made of sections of vegetation which break away from the floating meadows during heavy rainfall. Seven species were re-

Fig. 6: Breeding evidence was found for many of the floating meadow species such as *Dendropsophus triangulum*, these photos show a calling male, a mass of frog spawn and the tadpoles (all *D.triangulum*). Photos by: Katy Upton.

corded on rafts; the species found were represented by both adults and juveniles. These rafts potentially can transport individuals down river, aiding gene flow and dispersal.

Conservation Importance

The floating meadow habitat offers a unique opportunity to study hylid species which may not be encountered in other habitats. If this habitat was unavailable or not surveyed it is possible that several species would be missed from amphibian inventories. In recent years the flooding in the Loreto region of Peru has experienced some extreme highs and lows. The flood waters result from rainfall in the Andes and 2012 saw the highest recorded water level since the previous highest flood event in 1986. We do not know the impact that this extreme event may have on the floating meadows, or the subsequent impacts on the species which may rely on it for breeding.

Longer term research will always be more informative than short term or rapid inventories. This work highlights the importance of surveying all habitats, including those that are only temporary. It also highlights the importance of surveying across different seasons in order to gain a full picture of amphibian diversity.

Acknowledgements

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Monitoring an Endemic Amphibian Along a Natural Gas Pipeline in the Peruvian Andes

By [1]Jessica Deichmann, [1]Catherine Sahley, [2]Victor Vargas, [2]Oscar Chipana, [3]Edwin Smith, [2]Wilson Velazco & [4]Alessandro Catenazzi

Industrial operations and the development of linear infrastructure are an increasingly common threat in South American ecosystems (*1*). Unfortunately the impacts of such development on biodiversity and habitats have rarely been evaluated scientifically, particularly in high elevation tropical habitats. In 2008, the Smithsonian Conservation Biology Institute and the company PERU LNG established a Biodiversity Monitoring and Assessment Program (BMAP) to integrate biodiversity research and conservation into operations of a natural gas pipeline in Peru. This underground pipeline stretches 408 km from the eastern slopes of the Andes mountains, across the Andean *cordillera*, ending at the Pacific coast of Peru. Habitats traversed by this pipeline are extremely diverse and include montane tropical forest, high Andean grasslands and arid coastal areas. Knowledge on the diversity, abundance and conservation status for most species in the area of the pipeline is scant. The objectives of the BMAP, therefore, are to 1) assess the status, distribution and abundance of species and habitats of conservation concern; 2) evaluate the potential impacts of the project on these species and habitats and 3) provide management and conservation recommendations for effective mitigation of project impacts on studied systems.

The Andes are home to a variety of freshwater habitats that are poorly studied, but heavily utilized by local human populations as well as many species of birds, fish, and, of course, amphibians. Natural springs, bofedales (Andean wetlands), rivers and streams are all highly vulnerable to changes in land use. In particular, the construction of drainage canals and conversion of land to agriculture or pasture can have significant effects on watersheds through increased sedimentation as a result of erosion, changes in hydrologic cycle and flow patterns, and increased runoff of pesticides and other pollutants. The construction and operation of a pipeline might have similar impacts if care is not taken to avoid these.

Because of the importance of aquatic habitats along the pipeline, it was clear that species strongly tied to water would be important indicator taxa. The high Andean frog *Telmatobius jelksii* was one of the six amphibian species identified in the area of the pipeline during the Environmental and Social Impact Assessment (*2*). *Telmatobius jelksii* (Fig. 1) belongs to a genus of aquatic anurans endemic to the Andes. More than half of the species in this genus are listed as Endangered or Critically Endangered by the IUCN. In fact, none of Ecuador's three endemic *Telmatobius* species have been seen in years (*3*) and declines in *Telmatobius* populations have been documented in the Peruvian Andes as well (*4, 5*). Although *T. jelskii* is not considered threatened by the IUCN (*6*), this species faces threats similar to those faced by its congeners: habitat loss, disease, pollution and overharvesting. This made *T. jelskii* stand out as an important candidate for assessment and continued monitoring under the BMAP plan.

Fig. 1: Adult male (A), female (B) and tadpole (C) of *Telmatobius jelskii* encountered during the third sampling season. Photos: Jessica Deichmann (A&B), Victor Vargas (C).

The assesment and monitoring program

We began the assessment of *T. jelskii* populations in the wet season (March-April) of 2010, shortly after construction of the pipeline was completed. We have since repeated sampling in the dry season (July) in 2010, the rainy season (December) in 2011 and the dry season (October) in 2012. Monitoring is conducted along the pipeline across a spectacular array of seven different habitats in the Departments of Ayacucho and Huancavelica including: montane forest of the Apurimac River Valley, the dry forests of the Torobamba River Valley, the divide of the Huamanga and Vischongo River basins,

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Fig. 2: *Telmatobius jelskii* **in a rocky-bottomed stream in the Andean highlands. Photo: Alessandro Catenazzi.**

the Vinchos River Valley, the Apacheta highlands, the Pampas-Palmitos River basin and the high plains and peaks of Huaytará (*7*). *Telmatobius jelskii* is known to inhabit rocky-bottomed streams (Fig. 2) as well as soft-bottomed springs (*8*). Based on this knowledge, transects are established in springs, streams and rivers that are intersected by the pipeline. At each sampling location, a "control" transect is established upstream of where the pipeline crosses the water body (Fig. 3), and an "impact" transect downstream of the crossing point. Teams search all pools along each transect, reaching down to the bottom, in mud, under rocks and in crevices where adults, juveniles and tadpoles often hide. All larval and adult amphibians are identified, measured, weighed, sexed or larval state (Gosner stage) identified and a swab of skin (adults) or mouthparts (tadpoles) is taken for later analysis to determine the prevalence of *Batrachochytrium dendrobatidis* (*Bd*) in the population. Physical characteristics of the water bodies in which individuals are found are also measured to determine microhabitat preference for the species (Fig. 4).

Fig. 3: A "control" sampling transect along a stream inhabited by *T. jelskii*. **Photo: Jessica Deichmann.**

After four sampling seasons, this program has trained three biologists from Ayacucho, Peru in field work techniques for population monitoring and disease detection in amphibians. This assessment program has allowed us to determine the current geographic and elevational range of *T. jelskii* along the pipeline, collect baseline data on relative abundances of tadpoles and post-metamorphic life stages, characterize habitats used by tadpoles along the pipeline and conduct within-stream comparisons of frog populations downstream and upstream of pipeline crossings. As a result of this program, we have determined prevalence and levels of *Bd* infection in frog populations in our study area. We have also documented occurrence of other amphibians: we have found juveniles, adults and tadpoles of *Pleurodema marmoratum*, *Rhinella spinulosa* and *Gastrotheca marsupiata*, indicating persisting sympatric populations of these anurans in the area impacted by the pipeline. Results from the program thus far have been disseminated in a book chapter (*9*),

Fig. 4: Research team taking physical measurements of a pool along a transect. Photo: Alessandro Catenazzi.

a peer-reviewed journal publication (*10*) and in a public presentation in Ayacucho.

Population characteristics and persistence

Telmatobius jelskii inhabits small streams between 2,500 and 4,500 masl across a variety of habitats over a 180 km stretch of pipeline (Fig. 5), yet this frog is absent from some sites in our study area with seemingly appropriate habitat. Conversations with local people indicate that *T. jelskii* was once abundant in these areas, but has disappeared within the last 10-15 years (VV, WV, JD, pers. comm.). The reasons for this are unknown, but the area boasts a significant human presence with heavy agriculture, and some *Telmatobius* species have been overharvested in many parts of the Andes (*11*). *Batrachochytrium dentdrobatidis* has been detected in each of the populations surveyed along the pipeline (*10*) and this also provides a plausible explanation for the disappearance of *T. jelskii* in some areas. As for the impact of the gas pipeline, our data thus far reveal no detectable influence of pipeline operations on presence/absence or abundance of *T. jelskii*. It is important to note that we were unable to assess the impact of the pipeline's construction because the first assessment did not take place until after construction was completed.

Analyses of swabs of frog skin and tadpole mouthparts were conducted through collaboration with the Vredenburg lab at San Francisco State University. The prevalence of *Bd* among tadpoles averaged 53%, and 8 out of 13 streams inhabited by *T. jelskii* had a prevalence greater than 50% (10). Our team also found that prevalence of *Bd* was higher during the dry season and increased with the age of the tadpoles. Tadpoles do not typically suffer high mortality from *Bd* (only keratinized mouthparts are infected), and are thought to be important reservoirs (*12*) that can enhance the persistence of *Bd* (*13*). Previous research suggests that, given appropriate habitat and favorable conditions, *T. jelskii* can reproduce throughout the year (*8, 14*). Our data support this finding. In areas where multiple tadpoles were found, there was evidence of multiple cohorts in a site at one sampling time (*10*). Multiple cohorts combined with a prolonged larval period (*8*) ensure a constant source of infection for juveniles and adults, and could play an important role in triggering disease outbreaks. Although we lack data on the frog's susceptibility to *Bd* infection, we have found dead and dying juvenile frogs at some sites. The transition from tadpole to adult is a vulnerable stage for amphibians with chytridiomycosis because as their skin becomes more keratinized, they are more likely to succumb to a fatal infection (*15*). Thus, based on what we know about susceptibility to *Bd* in other *Telmatobius* species (*16*), there is reason to be concerned about the continued persistence of these frogs in the central Andes where we work.

The good news is that the numbers of tadpoles and adults of *T. jelskii* have not decreased since our first survey in March 2010. However, continued monitoring of these populations is extremely important. When *Bd* first arrives to a site, the amphibian community typically experiences a widespread outbreak of chytridiomycosis, but the intensity of infection in an area depends on the density of individuals in the population and the local environmental conditions. If some individuals survive the initial outbreak, population

persistence is possible, and *Bd* is likely to persist in the population in an endemic, rather than an epidemic, state (*13*). Because *Bd* is already present at our monitoring sites, we need to understand if these *T. jelskii* populations have experienced a recent outbreak that might have caused regional declines and if the state of *Bd* is now endemic in the environment.

Future work with *T. jelskii*

Four sampling seasons have resulted in the collection of valuable data on *T. jelskii* range and distribution, habitat use, reproduction, population viability and other natural history information, as well as prevalence and intensity of infection with *Bd*. Since the initiation of surveys in 2010, there have been no major changes in population abundances along the pipeline, either upstream or downstream of the pipeline crossing; however continued monitoring of these populations is essential to understand the potential impacts of infrastructure development and evaluate restoration measures post-impact. We will also continue surveying levels of *Bd* at all sites so that we can determine the infection dynamic in this system and if possible, ameliorate disease outbreaks and potential population crashes. Future efforts will focus on investigating possible symbiotic *Bd*-resistant bacteria and evaluating the potential of probiotics as a way to protect frogs from chytridiomycosis. Finally, it is important to consider that the effects of global climate change, including increased loss of glaciers in the central Andes, reduced rainfall, and extreme climatic fluctuations will affect the availability of aquatic habitats used by *T. jelskii* and will likely have great repercussions for the persistence of high Andean frogs (*17*). We plan to continue evaluating existing populations and to search for additional populations of *T.jelskii* in areas with appropriate habitat, but where we have not yet found any individuals, in order to add to the body of knowledge surrounding this species with the hope of contributing to its continued persistence.

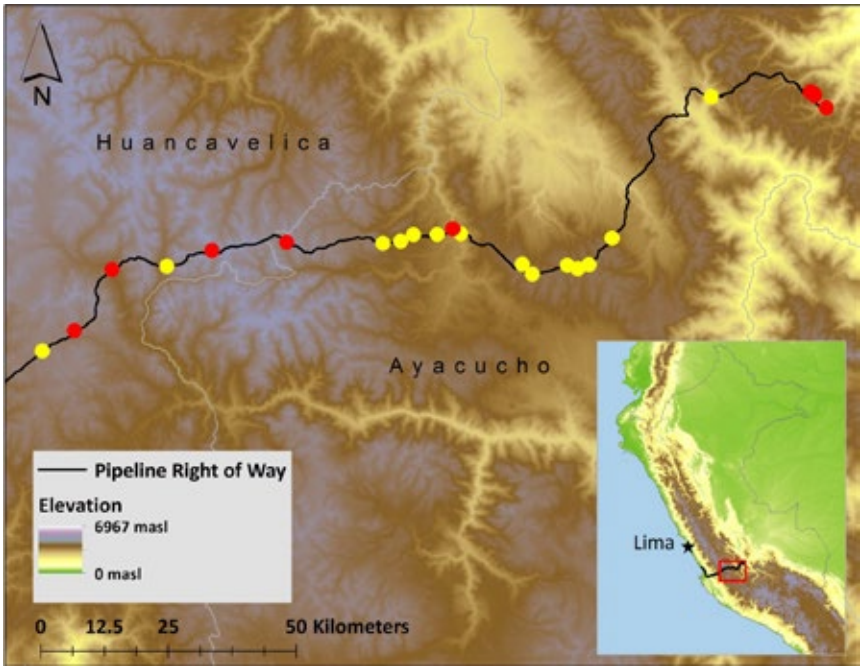

Fig. 5: Map depicting *T. jelskii* assessment and monitoring points along the pipeline. Yellow dots indicate sites where *T. jelskii* is present; red dots indicate sites which have been searched but the species has not been found.

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The 2012 Field Course on Amphibian Conservation in Peru

By Alessandro Catenazzi

Last June a group of 14 students and conservation practitioners from Peru, the US and the UK gathered at Wayqecha Biological Station, near Manu National Park, for a week-long field course on disease ecology and amphibian conservation. The course was organized by San Francisco State University (SFSU) and supported by funds from the National Science Foundation (Grants 1120283 and DBI-1103087), the Rufford Small Grants Foundation, the Asociación para la Conservación de la Cuenca Amazónica and Rainforest Expeditions. The bilingual course combined lectures with field and lab work at the station and its surrounding cloud forest and grassland. Lectures were given in Spanish or English, with simultaneous translation provided; participants also received copies of all presentations, PDFs of relevant literature and other bibliographic resources, as well as a photographic guide to the local amphibian fauna. The instructors were Drs. Vance Vredenburg and Alessandro Catenazzi from SFSU, Dr. Andrea Swei from the University of California San Francisco, Dr. Rudolf von May and M.Sc. Jenny Jacobs from the University of California Berkeley and M.Sc. Vicky Flechas from the Universidad de los Andes in Bogotá. Lectures ranged from principles of disease ecology to amphibian diversity, with a focus on the local frog fauna and the study of the frog-killing fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*). Field activities and training at the spectacular setting of Wayqecha with views over the cloud forest and Amazon lowlands, included leaf-litter plots, visual encounter surveys, call recordings and the use of agar frog models and temperature sensors to collect microclimatic data. These activities were followed in the lab with protocols for frog physiological measurements, swabbing for detection of *Bd*, treatment of infected frogs, animal husbandry, infection susceptibility trials and microbiological studies of amphibian skin microbiota. The diverse group of participants included undergraduate and graduate students, professional biologists, ecotourism guides and a lawyer. The course ended with a pachamanca of potatoes, beans and lamb and a dancing party in the midst of a marsupial frog chorus.

Instructors Vance Vredenburg (center), Rudolf von May (right) and Vicky Flechas (center-left, in white shirt) teach students how to survey grassland amphibians. Photo: A. Catenazzi.

Course participants. Photo: Celeste Dodge.

Lecture at Wayqecha Biological Station. Photo: A. Catenazzi.

Participants looking for frogs in a 10 x 10 m plot in the grassland of Wayqecha Biological Station. Photo: A. Catenazzi.

Batrachochytrium dendrobatidis in Peru

By Tiffany A. Kosch

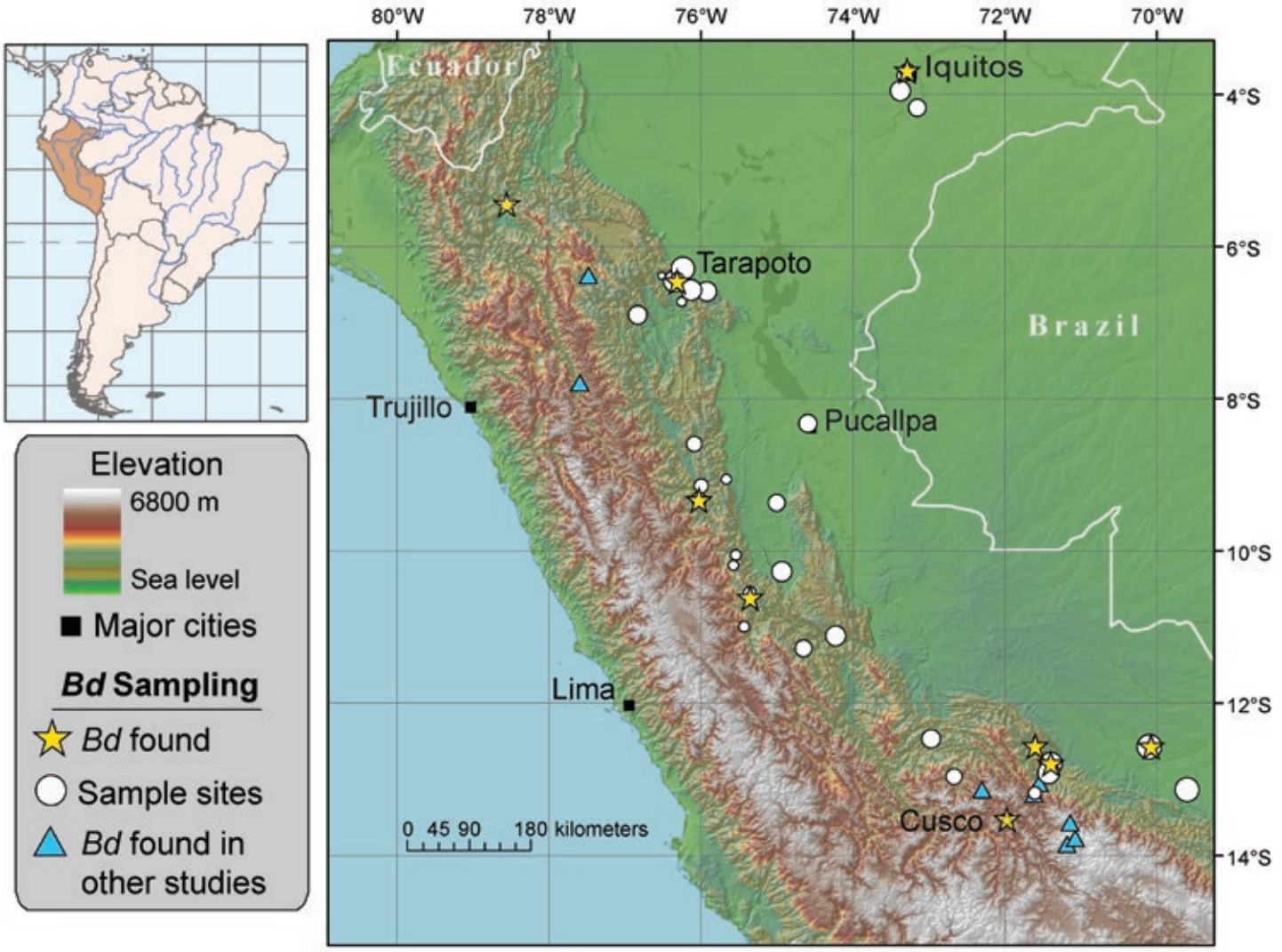

Fig. 1: Study sites in Peru sampled for *Batrachochytrium dendrobatidis* (*Bd*). Circles = sample sites with circle diameter proportional to number of samples; stars= sites where *Bd* was detected, and triangles= *Bd*-positive sites reported by others (see Table 3, *2*). Source: Kosch *et al.* (*2*).

The amphibian chytrid fungus, *Batrachochytrium dendrobatidis* (*Bd*), has been well-studied in Australia, North and Central America and Europe, but relatively little information is available concerning the status of this pathogen in South America. This is especially disturbing given that South America contains the highest diversity of amphibian species in the world (*1*). In Peru, sampling for *Bd* has been especially sparse, with only six studies in localized regions conducted to date (*2*). The aim of our investigation was to conduct a systematic survey of the current distribution of *Bd* in amphibian populations throughout Peru. Our main goal was to obtain a "snapshot" of the current distribution of the infection to aid in planning future research and management of the disease.

Field surveys of *Bd* prevalence were conducted during June–August of 2007 and May–July of 2008, coinciding with the dry season. We surveyed 39 sites along the eastern slopes of the Andes, providing a collection of samples along both latitudinal (3.68572°S, 73.28350°W to 13.17956°S, 71.60561°W) and altitudinal gradients (90–3240 m). Amphibians were sampled for *Bd* using the methods outlined in Kosch *et al.* (*2*). A total of 983 amphibian skin swabs were collected from 39 sites throughout Peru. The results of our PCR assay showed that 11 of 983 individuals sampled were positive for *Bd* (overall prevalence = 1.0%). *Bd* was detected in amphibians at 9 of 39 sites across a broad range of altitudes (96–3240 m, Fig. 1). Among-site prevalence ranged from 0 to 25%. The majority of *Bd*-positive individuals had reproductive modes associated with permanent bodies of water (6/11; *Allobates marchesiansus*, *Hypsiboas melanopleura*, *Hyloscirtus cf. phyllognatus*, *Hyloxalus shuar*, *Osteocephalus buckleyi*, *Telmatobius cf. marmoratus*) and/or had an aquatic tadpole stage (9/11; *A. marchesiansus*, *Engystomops petersi*, *Hypsiboas melanopleura*, *Hyloscirtus cf. phyllognatus*, *Hyloxalus shuar*, *Leptodactylus petersii*, *O. buckleyi*, *Scinax garbei*, *T. cf. marmoratus*). Five of the 11 *Bd*-positive individuals are known to utilize streams for reproduction (*Allobates marchesiansus*, *Hyloscirtus cf. phyllognatus*, *Hyloxalus*

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Fig. 2: Three species that tested positive for *Batrachochytrium dendrobatidis* (*Bd*) in this study: A) *Hyloscirtus c.f. phyllognatus*, B) *Telmatobius c.f. marmoratus*, and C) *Gastrotheca excubitor*. Source: Kosch *et al.* (*2*).

shuar, *O. buckleyi*, *T. cf. marmoratus*). The family with the highest number of infected individuals was Hylidae with four infected individuals. *Bd* was not detected in any of the *Atelopus* that we sampled (3 *A. pulcher*, 20 *A. cf. andinus*). One of the *Bd*-positive samples came from two *Telmatobius marmoratus* purchased at San Pedro Market in central Cusco (of unknown origin) where they were being sold for human consumption. Obvious clinical abnormalities consistent with possible signs of the disease chytridiomycosis were only detected in three of 983 individuals (all *Gastrotheca excubitor*), which had retained shed skin on their toe pads. One of these three individuals tested positive for *Bd* with the PCR assay.

The results of this and previous studies indicate that *Bd* is widespread throughout Peru, and from our limited data it appears that *Bd* presence may not be as tightly linked with altitude as suggested by others (Fig. 1, *3*). This adds to the growing body evidence (*4*) that altitude may not have as strong an influence on disease prevalence as originally believed. In this study, the majority of *Bd*-positive individuals had reproductive modes associated with permanent bodies of water and/or an aquatic tadpole stage, characteristics that are have been shown to be associated with high transmission and prevalence of chytridiomycosis (*5*). Of these species, five are known to utilize streams for reproduction, a characteristic associated with susceptibility and declines in other regions (*5, 6*). There was a greater frequency of *Bd*-positive individuals in the family Hylidae. Other researchers have reported a higher frequency of chytridiomycosis in this family (*7*), which may be linked to reproductive mode (*8*) and/or evolutionary history (*9*). Contrary to our expectations, we did not detect *Bd* in any of the *Atelopus* that we sampled. Although this is the first time that a population of *A. cf. andinus* has been sampled for *Bd*, this disease has been previously detected in other Peruvian *Atelopus* (*A. pulcher*, *10*, *A. patazensis*, *11*). Our data showing *Bd* in one of two *Telmatobius marmoratus* tested from the San Pedro market further supports the idea suggested by Catenazzi *et al.* (*12*), that the trade of amphibians in Peru is contributing to the dispersal of *Bd* throughout the country.

Although the results of this and previous studies demonstrate that *Bd* is widespread in South America (e.g., *13, 14*), the consequences of this disease are still relatively unknown due to the near complete absence of general population monitoring and *Bd* studies in these countries. In Peru, significant declines have been reported in multiple high-elevation species in southern Peru (*6*), and several *Atelopus* and *Telmatobius* species are believed to already be extinct (*11, 15*). Peru contains some of the highest amphibian diversity on record and the loss of this diversity could have severe consequences. This makes the lack of knowledge on *Bd* and population status of Peru's amphibians especially alarming.

Acknowledgements:

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Agricultural Frontier Expands on par with Constraints to Protect Amphibians and Reptiles

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The transformation of natural ecosystems to croplands represents one of the greatest threats to global biodiversity. This process, and its expansion, generate clear problems for ecosystems and biodiversity, as well as hinder conservation efforts. Given these problems, the primary response from conservation biologists has been to develop approaches for the establishment of protected areas that adequately protect biodiversity, through a protocol known as Systematic Conservation Planning (SCP). The impossibility of using certain places (crops) for conservation, however, creates more problems, constraints and impossibilities for SCP. Therefore, certain conservation measures should be taken urgently, before it is too late.

Figure Legend: A: *Rhinella achalensis*; B: *Odontophrynus achalensis*; C: *Chacophrys pierottii*; D: *Phyllomedusa sauvagii*; E: *Lepidobatrachus asper*; F: *Pleurodema kriegi*.

Our research focuses on how the spatial configuration of Priority Conservation Areas for amphibians and reptiles change with the progressive conversion of ecosystems to intensive anthropic land-uses. In our study, we used a standardized methodology to calculate Priority Conservation Areas under different human land-use scenarios (one hypothetical in which intensive human uses are not considered and two real scenarios [or times] of land-use), while maintaining the same conservation objectives. Our study area was Cordoba, a highly deforested province in Argentina, where the deforestation rate is more than 12 times the global average, with more than 80% of its closed forest being lost in the last 20 years. Moreover, in Córdoba province inhabit several endemic, threatened and range restricted species of amphibians and reptiles, many of them listed on different categories of threat in the IUCN Red List (e.g. *Rhinella achalensis*; *Pleurodema kriegi*, *Odontophrynus achalensis*; see Figure).

Our results clearly show that, as time passes, the constraints to protect amphibians and reptiles expand on par of the agricultural frontier. Not only will more land be necessary to meet a given conservation objective, but also, Priority Conservation Areas become increasingly less efficient, meaning that a given area will have to have a much greater perimeter, a larger number of fragments and a less appropriate shape to obtain the same conservation objective.

With respect to the Cordoba province, we found that the vast majority of protected surfaces are located in unproductive places such as salt flats or occupied by water, with only around 1% having the potential for intensive agriculture. As noted in many places around the globe, this leads us to consider that land productivity (or lack thereof) is the most important criteria in the selection of protected areas in the region. The case of Cordoba province is even more alarming because despite the environmental damage suffered by the province, political decisions in the territory continues to promote agricultural expansion. Under the framework of a national law for the management of the national forests, several researches together with NGOs presented in Córdoba a project aimed at protecting the scarce forest remnants; unfortunately this project fell on deaf ears, instead the "Ley 9814 de la Legislatura de la Provincia de Córdoba" was approved, with a clearly different goal leaning towards more production than conservation.

Regrettably, unless radical political changes occur, the forecast for many species of amphibians and reptiles of our province is highly uncertain. More troubling still is the fact that the pattern shown here is probably applicable to others places with high rates and degrees of deforestation in which reserves have also been selected in an *ad hoc* manner. Therefore, it is imperative that decisions be made with the aim of protecting the remaining natural areas in regions dominated by agriculture in the best way possible, despite the fact that it may involve high costs. This decision, as stated before, should be taken urgently, before it is too late.

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Some Updates in the Conservation of Bolivian Amphibians

By Muñoz Arturo

Telmatobius sp. One of the species we are breeding. Photo: Arturo Muñoz.

After four years of the Bolivian amphibian initiative, information about amphibians and their habitat is being gathered. We are continuing to learn more about the patterns of distribution, abundance, threats and also gaining important insights into the populations of the regional amphibians. These results are helping us to prioritizing our conservation efforts. Our efforts are helping identify those population at most risk and helping us understand which populations might have already been lost. For this reason we want to focus our work on the aquatic frogs of the genus *Telmatobius* and one of these species is the Critically Endangered Titicaca water frog *Telmatobius culeus*. Working with this species we are locating the highest priority sites for conservation action on the Bolivian side of Lake Titicaca and collecting data that will allows us to understand the status and population trends of this species in the lake.

We found populations that are in very bad conditions, where almost 50% of the frogs were dead. In some areas it seems that pollution is affecting frog populations. A very worrying finding is the trade of this species for use in local fairs and restaurants where they use the species as food and also as medicine or a tonic for almost anything. So there is an urgent need to coordinate a conservation strategy for this species together with local stakeholders, governmental institutions and conservation organizations.

Finding dead frogs in Titicaca Lake. Photo: Arturo Muñoz.

We also found that some species of *Telmatobius* from mountain cloud forests are not present in areas where they were previously common. This is not the case for species from the dryer areas in the Andes where it seems that those amphibian populations are still there or are less affected. We also found areas where Chytridiomycosis has recently arrived and during fieldwork we are encountering an increasing number of dying frogs. We don't yet know the exact reason but we are working to understand what is really happening there.

Museo de Historia Natural Alcide d'Orbigny. For more information please visit www.bolivianamphibianinitiative.org, www.bolivianamphibianinitiative.blogspot.com or contact Arturo Muñoz hyla_art@yahoo.com

A couple of *Telmatobius culeus* in amplexus. Photo: Arturo Muñoz.

A tadpole of *Telmatobius culeus* from our first captive offspring. Photo: Sara Dykman.

Due to the bad situation of the frogs in Bolivia we see an urgent need to do something and we are taking some individuals from the field to try and learn how to keep and breed them in captivity.

Working with the captive breeding program of the aquatic frogs of the genus *Telmatobius*, we have been able to successfully breed four species; *Telmatobius hintoni, Telmatobius culeus* and two other species of *Telmatobius* whose identity is not clear yet, and possibly they are new species to science. These experiences allowed us to learn about their natural history and their breeding requirements. We are already holding six species of Bolivian *Telmatobius* and we hope we will be able to learn how to breed all of them soon.

We are also taking advantage of captive bred frogs to test new methods to be used in the future on wild amphibians. This is also a great opportunity to develop education programs using some of the captive individuals to create museum exhibits.

We are focusing our efforts in the genus *Telmatobius* because that group is at high risk of extinction, from the 14 Bolivian species, 13 are endemic and all of them are listed in the IUCN Red List and most of them have a very restricted distribution, facts that make this group of amphibians very susceptible to different threats that are affecting Bolivian amphibians.

We are also working with local communities and people in general to raise awareness about the amphibian crisis, both in rural and urban areas with the goal to change negative attitudes that can affect amphibians. We are also training young biologists and conservationists during our field campaigns and through the different activities of the project and once a year we organize a training course for students and young conservationists from all over Bolivia. In this way we can work together with a bigger number of people for the conservation of Bolivian amphibians.

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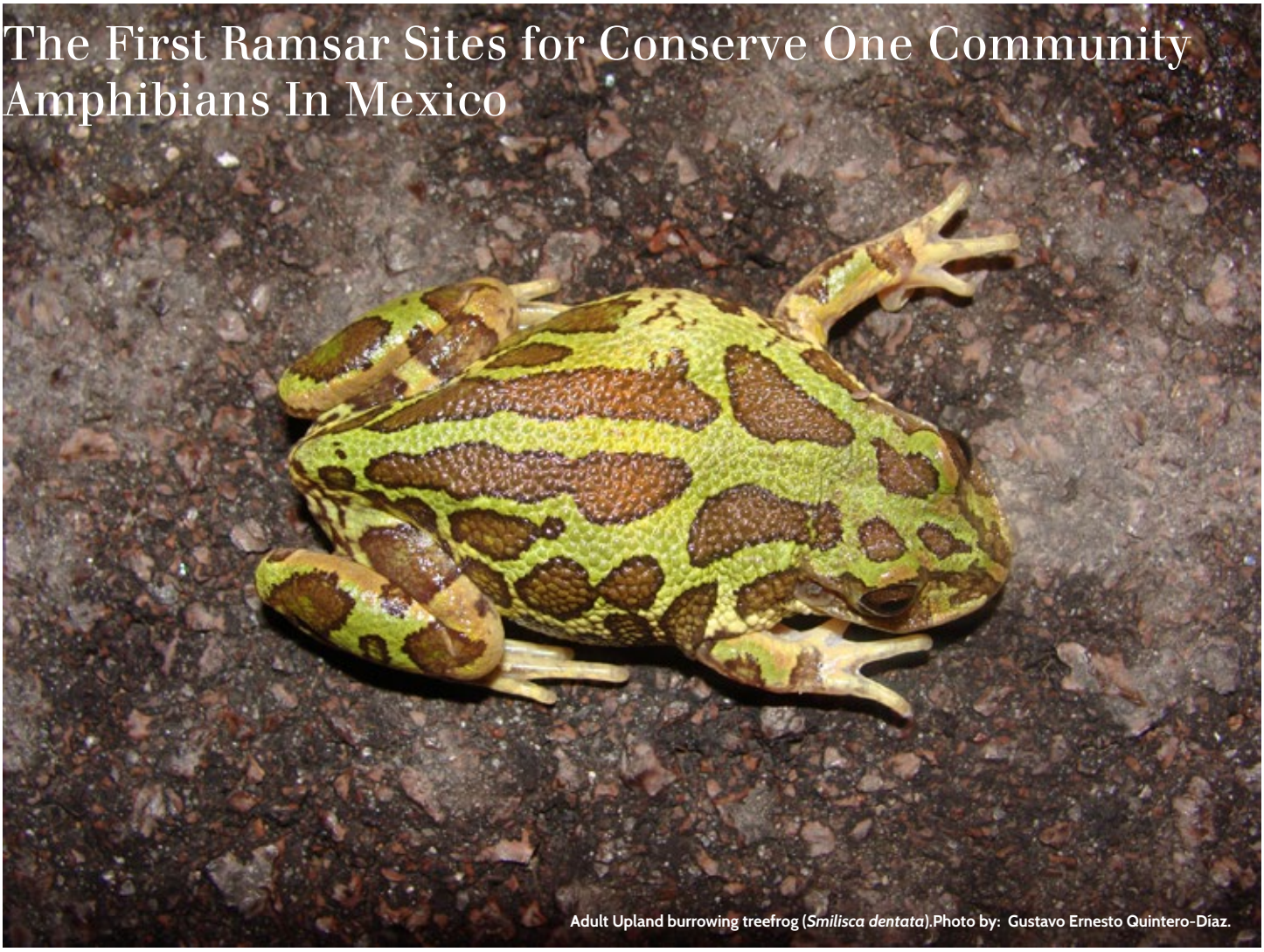
This work of the Bolivian amphibian initiative is thanks to the support of several institutions and people such as Amphibian Ark, IUCN SSC Amphibian Specialist Group, Rufford Small Grants, Durrell Wildlife Conservation Trust, European Association of Zoos and Aquaria, Denver Zoo, Fresno Chaffee Zoo, Museo de Historia Natural Alcide d'Orbigny and Idea Wild among others.

The First Ramsar Sites for Conserve One Community Amphibians In Mexico



Adult Upland burrowing treefrog (*Smilisca dentata*).Photo by: Gustavo Ernesto Quintero-Díaz.

By Gustavo Ernesto Quintero-Díaz.

Mexico has the privilege of being one of the six most biologically diverse countries in the world, along with Colombia, Brazil, Zaire, Madagascar and Indonesia (*1*). There are 7,093 amphibian species known in the world (2) of which 373 are present in Mexico (*3, 4*), one of the highest levels of amphibian species richness in the world only after Brazil, Colombia, Ecuador and Peru (*5*). These numbers help to illustrate the importance of the Mexico for amphibian fauna.

Unfortunately, a third of the amphibian species worldwide are facing an array of challenges just to survive and many others have disappeared in the last thirty years. The IUCN Red List of Threatened Species™ has identified amphibians as being the most threatened vertebrate group assessed so far, with around 41% at risk of extinction. Although there is no documentation of recent Mexican amphibian extinctions, with the possible exception of the Tlaloc's leopard frog *Lithobates tlaloci* (*6*), it has been indicated that the Upland burrowing treefrog (*Smilisca dentata*) that inhabits Central Mexico could be at risk of extinction (*7-10*).

Among the 414 vertebrate species known for the state of Aguascalientes, the Upland burrowing treefrog is the only near endemic to the state, it is distributed in the southern part of Aguascalientes and northern Jalisco, in Central Mexico and has been the subject of numerous natural history and ecology studies by the authors.

The Upland burrowing treefrog (*S. dentata*) is a member of the treefrog family (Hylidae). These frogs are characterized for having broad tips of the digits that secrete a substance (polysaccharide) that allows them to adhere to slippery surfaces and climb easily (*11*). Although a member of the Hylid family, this species has lost the broad fingertips almost entirely as a consequence of its fossorial habits. As many frogs, *S. dentata* possess a beautiful green color skin with tone variations in the dorsum, several blotches in dark brown forming ovals or bars, or a combination of the two; these blotches are always surrounded by thin black lines. The ventral part in most of them is whitish and only a handful of specimens have a creamy coloration in the belly; both the skin in the dorsum and the belly have a warty texture.

The adult males have a pair of vocal sacs, one in each side of the throat, that are used to call during the rainy nights. The Upland burrowing treefrog has a robust body; the adult size between males and females differs little.

One of the major threats to this frog and anuran community in Buenavista de Peñuelas, Aguascalientes, Mexico is the proximity of the frog population to the main road leading to the airport. Al-

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though the road plays an important role in delivering both social and economic benefits, opening areas in this way, just like many other human endeavors, can cause negative impacts on the environment. Understanding those negative impacts is important for developing and implementing conservation strategies that avoid and/or mitigate those impacts (*12*).

Installing barrier. Photo by: Gustavo Ernesto Quintero-Díaz.

From 2005 to 2012, a comprehensive study on the distribution, population size and ecology of *S. dentata* took place in Aguascalientes, Mexico. Our research, the activities from the civil organization "Conservación de la Biodiversidad del Centro de México" (BIODIVERSIDAD A. C.), and the Social Service campaign that the students from the Preparatoria (high school) from the Universidad Autónoma de Aguascalientes, have all resonated with the people of Aguascalientes and the scientific community at national and international levels.

The impact has been so important that the students from the preparatoria were awarded the prize "Premio Estatal al Mérito Ecológico Ambiental 2007" and instituted The Day of the Upland burrowing treefrog in Aguascalientes the last Friday in November. In 2012, the day of the Upland burrowing treefrog, was celebrated for the seventh year in a row. An explanatory article in the magazine ESPECIES was published, in which the urgency to establish a conservation strategies for this Mexican endemic frog is highlighted (*13*). As early as January 2008, members of the Committee of the Environment from the legislative "Cámara de Diputados" from Distrito Federal established direct communication with us. As a result of this, we sent a protection proposal to the Committee of the Environment, that showed a remarkable interest in the conservation of the area where the Upland burrowing treefrog lives. We were invited by the House of Representatives (Cámara de Diputados) to the Federal Legislative Palace (Palacio Legislativo) in San Lázaro to give a talk about the problems faced by the Upland burrowing treefrog. On January 23rd, 2008 a federal motion was approved to start a technical study to justify the creation of a natural protected area in the category of Sanctuary. This motion asked that the National Commission of Protected Natural Areas (CONANP) and the Environment and Natural Resources Secretariat (SEMARNAT), enforce the General Law for Ecological Equilibrium and Environmental Protection regarding protected natural areas (Gaceta parlamentaria, 2008). On February 28th, 2008, Decree 49 was unanimously approved at state level, requiring that the competing instances, such as the Aguascalientes office of SEMARNAT and the Ministry of Environment of Aguascalientes state (SMAE) to also enforce the General Law for Ecological Equilibrium and Environmental Protection regarding protected natural areas by promoting the required technical work that would allow the creation of the protected natural area Sanctuary of *S. dentata*, in the Ejido Buenavista de Peñuelas, Aguascalientes.

Passage tunnels wildlife. Photo by: Carolina Chávez- Floriano.

This protected natural area will allow the only vertebrate species endemic to the state of Aguascalientes to persist, and help support the entire amphibian community that share the habitat in Buenavista de Peñuelas, along with other endemic invertebrates, like the fresh water crustaceans *Karualona penuelasi* (*14*) and *Macrothrix agsensis* (15), *Macrothrix smirnovi* (*16*), a couple of new species for México of fresh water "cacerolitas" (*Triops* sp.), two rotifers *Keratella mexicana* (*17, 18, 19*) and *Brachionus josefinae* (*20*). Three species of anurans that inhabit the area are listed in the NOM-059-SEMARNAT-2010 (the document that contains the species of animals and plants that are federally protected in México): Montezuma's frog (*Lithobates montezumae*) is in the category "sujetas a protección especial" or species subject to special protection, and *Lithobates neovolcanicus* and *Smilisca dentata* are in the category "amenazada" or threatened.

Several TV stations at state and national level were interested in the problems faced by the species, and two programs were recorded for Aguascalientes TV and an informative spot for the nightly news program on TV Azteca. Those TV programs help draw attention to the conservation effort surrounding the Upland burrowing treefrog in the state of Aguascalientes.

By March 2009, the state government in Aguascalientes had a considerably better understanding of the conservation issues surrounding the species and the mitigation strategies proposed to deal with those issues. With the involvement of the "Secretaría de Obras Públicas", a concrete wall, 180 m long by 1.2 m high, was constructed to prevent frogs being run over while moving between their two most important breeding sites, two temporary pools that are part of the site proposed as a RAMSAR site. Seven bridges were also built to allow the fauna to pass as a strategy to mitigate the habitat fragmentation due to the recent construction.

Furthermore 1,000 m of fencing was installed in the main area along with about 6,000 m of additional barrier along the road that goes to the community of Tanque de los Jiménez. To complement the above strategies the lighting system along the airport road was replaced by technology based on "LEDs" that are less harmful to

insects and more energy efficient. This action by itself will save 75% in the money paid to the energy company, without mentioning that the LED lights have a useful life of about 15 years instead of one or two years. In 2009 we evaluated the effect that this new lighting system, the barriers and the faunal bridges had on the amphibian fauna. There is still a need for a better traffic signaling system so the drivers lower their speed in the rainy season along the six kilometers that have been identified as key points for amphibians crossing the road. There is also evidence to support the need for better environmental impact studies before the construction of more roads.

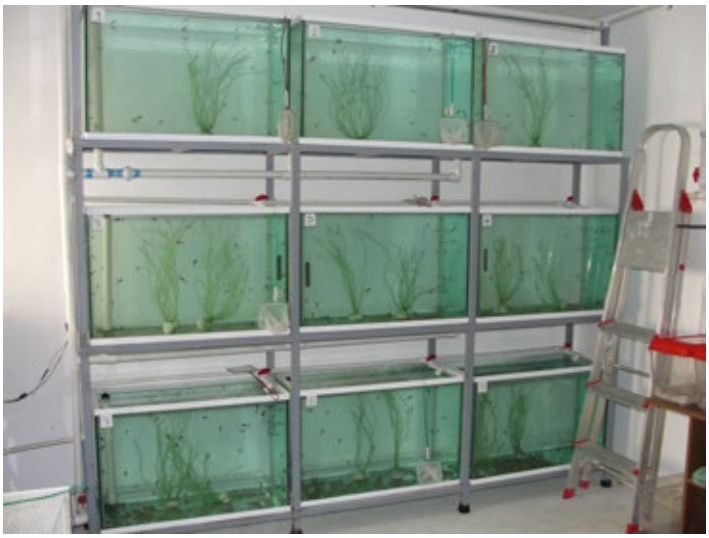
In an effort to sensitize the local communities in the state, the Aguascalientes municipality and TETRAPAK created a theater act, which was presented successfully during the "Feria Nacional de San Marcos 2009 - 2012", and during the month of June in the municipalities of Aguascalientes state and in the "Centro de Educación Ambiental y Recreativo Rodolfo Landeros Gallegos". The show narrates, in a very accessible way, the conservation issues that are faced by the Upland burrowing treefrog in Aguascalientes. Members of BIODIVERSIDAD A. C., during the campaign titled "Cleans México", launched by TV Azteca Aguascalientes, coordinated the cleaning of the area on May 30th, 2009. Such cleaning effort were very successful and attracted more than 90 people that worked hard all day to clean the area where the Upland burrowing treefrog lives.

The Upland burrowing treefrog is a clear example of the environmental crisis that we are living in today. This frog is disappearing in front of us, but it is not the only species that faces the same future in our country, and we want to take a brief moment to mention some statistics that attest to the gravity of the situation.

One hundred and ninety four (52%) of Mexican amphibians are listed in one of the four country based risk categories (Norma Oficial Mexicana NOM-059-SEMARNAT-2010). From those 194 species, 105 species (28.15%) belong to the Order Anura (frogs and toads), 87 species (23.32%) belong to the Order Caudata (salamanders) and two species (0.53%) to the Order Gymnophiona (caecilias). It is worth noting that no species of amphibian in our country is considered in the most serious of the four categories: Endangered or "En peligro de extinción". The Upland burrowing treefrog is considered to be Threatened species ("amenazada"). In addition the IUCN Red List of Threatened Species™ (IUCN) categorizes the Upland burrowing treefrog as "Endangered" meaning the species faces a very high risk of extinction in the wild.

Would it be possible that a charismatic frog species of restricted distribution, endemic to Central Mexico and that possibly only survives in a few localities in Aguascalientes and Jalisco states will disappear in front of us just as it happened with the Golden toad?

Theater act. Photo by: Gustavo Ernesto Quintero-Díaz.



Captive breeding. Photo by: Gustavo Ernesto Quintero-Díaz.

The biggest threat to the Upland burrowing treefrog, like for most species of flora and fauna worldwide, is habitat destruction in favor of activities that benefit the humans. In the long term, what will save this species will be our understanding and our ability to harmonize our necessities as a species with the other living beings on the planet.

Fortunately, as of 2nd February 2011 the primary habitat of the Upland burrowing treefrog was declared a Ramsar site. In 2012 we release to 121 individuals in the Ramsar Site, these individuals were the product of an ex-situ breeding initiative. To learn more about this species and the work underway to save it please read the book "Natural History of a very Mexican frog" by Gustavo Ernesto Quintero Díaz y Joel Vázquez Díaz.

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Team members of BIODIVERSIDAD A. C. Photo by: Gustavo Ernesto Quintero-Díaz

The Catalogue of Colombian Amphibians and Reptiles

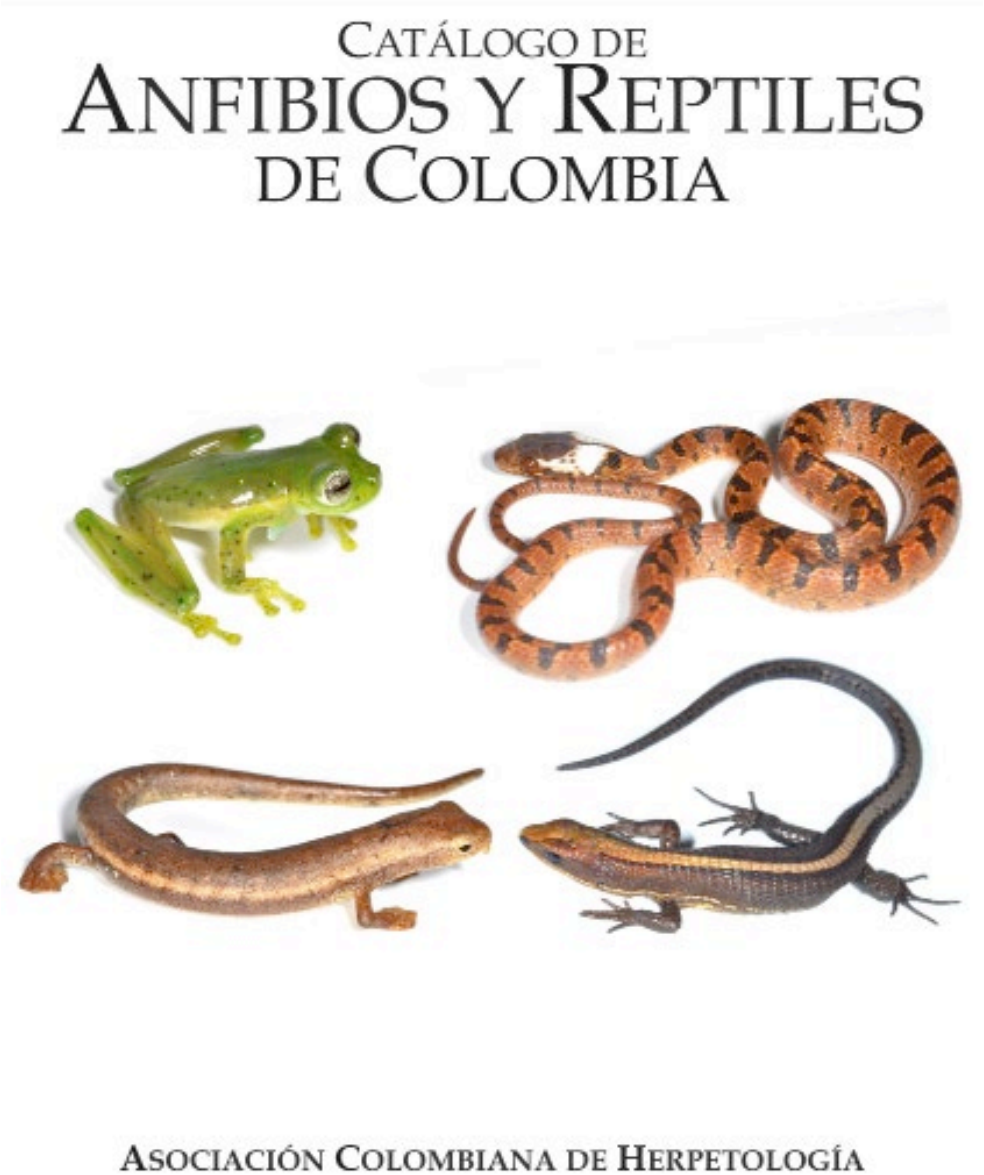
By: Mauricio Rivera-Correa

The Catalogue of Colombian Amphibians and Reptiles (Catálogo de Anfibios y Reptiles de Colombia in Spanish) is an initiative of the Colombian Herpetology Association. It is conceived as a scientific contribution that summarizes the current knowledge of the species of amphibians and reptiles distributed in Colombia.

In addition to contributing to a thorough literature review, it is a mechanism to present information that has not been published by a peer review process but that deserves to be released to help better understand the attributes of species, natural history and the conservation status. This unpublished information includes photographs, maps, museum records and undergraduate theses.

The Catalogue is an initiative built around the ACH members and led by a team of professors and researchers who will assist in the species account review process of more than 1400 species of amphibians and reptiles. The Catalogue will be published online including about ten species accounts per issue and also individual species accounts in pdf format.

We are convinced that the Catalogue of Amphibians and Reptiles of Colombia will become an excellent source of reference for young herpetologists and professional biologists interested in the knowledge and conservation of this highly diverse group. Please visit the website www.acherpetologia.org for more details about this initiative.



Conservation and Ecology

Lost and found: One of the world's most elusive amphibians, Pseudophilautus stellatus (Kelaart 1853) rediscovered

By L. J. M. Wickramasinghe, D. R. Vidanapathirana, S. Airyarathne, G. Rajeev, A. Chanaka, J. Pastorini, G. Chathuranga & N. Wickramasinghe

Pseudophilautus stellatus (Kelaart 1853) has been rediscovered from the Peak Wilderness, Central Hills of Sri Lanka. The species, till now Fejervarya known only from its lost holotype, was the first shrub frog described from Sri Lanka, and had not been reported since then. It was thought to

have become extinct for nearly 157 years, being the amphibian species "lost" for the longest amount of time. Here we designate a neotype from the material collected at what we consider its type locality, having considered characters of the lost holotype and provide a complete description. We have conducted a molecular phylogenetic analysis, on which basis the species is well differentiated from all other *Pseudophilautus* sequenced so far, and placed in a clade together with *P. femoralis, P. frankenbergi, P. mooreorum* and *P. poppiae.*

L. J. M. Wickramasinghe *et al., Zootaxa* **3620**, 112 (2013). http://dx.doi.org/10.11646/zootaxa.3620.1.5

Uncovering salamander ecology: A review of coverboard design

By Kyle Miller Hesed

Coverboards have been used for decades in research on amphibians and reptiles, but their characteristics have varied widely. This diversity in design may both complicate comparisons among studies and preclude assessment of how coverboards could be deliberately tailored to specific study objectives. Although numerous studies have evaluated the effectiveness of various aspects of coverboards, a general synthesis of these results as they relate to salamanders is lacking. Here, I summarize and evaluate information relating to coverboard design and potential concerns for using coverboards in studies of salamanders. Although many salamander species have been encountered under coverboards, coverboard design may have been optimized for *Plethodon cinereus*, a terrestrial species found in eastern North America. Altered designs (e.g., material, dimensions, placement, location or spacing) may prove superior for other species. With reported declines in salamander populations at both of their global centers of diversity, now may be a crucial time to expand the use of coverboards for studies of a wider variety of species. Further work also should evaluate the ability of a given design to address specific hypotheses and study objectives. In future studies, it should be possible to better tailor coverboard designs to the species, site and study questions at hand.

K. Miller Hesed, *J. Herpetol.* **46**, 442 (2012).

Cues from introduced fish alter shelter use and feeding behavior in adult Alpine newts

By Laurane Winandy & Mathieu Denoël

Amphibians are particularly affected by alien fish introductions and are declining worldwide. However, the behavioral mechanisms behind the observed cases of coexistence and exclusion patterns between adult amphibians and fish are poorly understood. In the present study, we aimed at testing the hypothesis that adult newts display different feeding and space use behavior in the presence of fish cues (i.e. access less food resources and use more shelters than when fish cues are absent). To achieve this we measured behavioral patterns in 100 adult Alpine newts (*Mesotriton alpestris*) in a replicated laboratory design (20 tanks × 7 replicates across time). Half of trials involved individuals in indirect (visual and olfactory) contact with goldfish (*Carassius auratus*), a non-predatory species for adult newts. In the presence of fish, significantly more newts hid under shelters than in their absence, but this difference decreased over time. A lower number of newts fed in comparison with controls. These results show that newts responded to fish presence even in the absence of direct contact, but the differences were small. Newts decreased vital activities such as exploration of open areas and feeding. They also adjusted shelter use over time, suggesting a process of habituation or a risk assessment in the absence of direct risk. These results reveal that exploring behavioral patterns can aid in understanding the causes of exclusion and coexistence patterns between fish and amphibians.

L. Winandy, M. Denoël, *Ethology* **119**, 121 (2013). http://hdl.handle.net/2268/135870

Illegal trade on non-native amphibians and reptiles in southeast Brazil: The status of e-commerce

By André L. B. Magalhães & Vinícius A. São-Pedro

In Brazil, the pet trade as a vector for amphibian and reptile introduction has received little attention. The introduction of non-native herpetofauna through the release of pets has occurred in many regions of Brazil, resulting in the establishment of feral populations. Although the Brazilian government has declared the importation of non-native herpetofauna illegal (Ordinance No. 93 of 1998), the efficacy of this regulatory policy has not been determined, and data on trade are incomplete and likely inaccurate, especially regarding trade carried out through the Internet. Species were surveyed in the social network Orkut from mid-2006 to mid-2012. We calculated the absolute and relative frequencies of dealers selling amphibian and reptile species in the three main cities of pet trade in southeastern Brazil. A total of 49 non-native species (three frogs, two salamanders, 16 lizards, 26 snakes, two turtles) were detected during the seven year survey period. Corn snakes (*Pantherophis guttatus*), Milk snakes (*Lampropeltis triangulum*), Central bearded dragons (*Pogona vitticeps*), Ball pythons (*Python regius*), and African clawed frogs (*Xenopus laevis*) comprised the largest percentage of e-commerce trade (43.37%, 9.40%, 8.67%, 6.99% and 4.82%, respectively). Seventeen species identified in our survey such as the Axolotl (*Ambystoma mexicanum*), Jackson's chameleon (*Trioceros jacksonii*) and the Royal python (*Python*

regius) are considered Endangered by the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). São Paulo was the city with the greatest demand (N=169 Internet dealers) and anonymous dealers were the ones that most negotiated animals (N=209). The reasons for a lack of compliance with existing Brazilian law are probably related to: (1) a lack of enforcement by Brazilian Institute of Environment and Renewable Natural Resources (IBAMA) on wildlife e-commerce; (2) ignorance of the law by most people involved in the pet trade, especially pet owners. We strongly suggest two measures: (1) IBAMA should work with web administrators, such as Google and Yahoo, to provide warnings to dealers and enthusiasts about Brazilian law regarding non-native wildlife trade, as well as the dangers of releasing non-native amphibians and reptiles into the habitats; (2) Orkut and other social media must intensify their educational campaigns concerning the illegal pet trade and popularize the goals of CITES to ensure that the international trade does not threaten species survival. Our study examined a small portion of the extent of the Brazilian Internet pet trade. Indeed, the problem seems to be much larger than we can imagine.

A. L. B. Magalhães, V. A. São-Pedro, *Phyllomedusa* **11**, 155 (2012). http://www.phyllomedusa.esalq.usp.br/table.php?V=11&N=2

Spatial analysis of amphibian road mortality levels in northern Portugal country roads

By Cátia Matos, Neftalí Sillero & Elena Argaña

By displaying higher biological susceptibility to the impact of roads, amphibians are the most affected group and road fatalities have a significant impact on population dynamics and viability. Iberian studies have shown the importance of country roads on amphibian road mortality, but still, little is known about the situation in northern Portugal. In contrast to highways, which act as barriers, country roads are more permeable to amphibian passage and represent a greater source of mortality. To plan and apply successfully mitigation measures in the extensive country road network, the identification of precise locations (*hotspots*) and variables related to animal-vehicle collision is needed. The aim of the study was to analyze the spatial occurrence and related factors linked to amphibian mortality on a number of country roads in northern Portugal, using spatial statistics implemented in GIS and applying a binary logistical regression. We surveyed 631 km

Male B. Bufo rescued from being road-killed. Photo: Cátia Matos.

of road corresponding to seven transects that represented country roads found in the study area (mainly urban landscape). We observed 404 individual amphibians: 74 (18.3%) alive and 330 (81.7%) road-killed. *Bufo bufo* represented 80% of the mortality records. Three transects showed clustered distribution of road-kills, and broadleaved forests and road ditches were the most important factors associated with hotspots of road-kill. Logistic regression models showed that habitat quality, *Bufo bufo*'s habitat preferences, and road ditches are positively associated with amphibians' road mortality in northern Portugal, whereas average altitude and length of walls were negatively associated. This study is a useful tool to understand spatial occurrence of amphibian road-kills in the face of applying mitigation measures on country roads. Also, it is considered the necessity of assessing the condition of amphibian local populations to understand road-kills spatial patterns. Future populations are at risk and the future may be reflected in the low kill/live rates detected, consequently there is an urgency to apply mitigation measures on country roads.

C. Matos, N. Sillero, E. Argaña, *Amphibia-Reptilia* **33,** 469 (2012).

River islands, refugia and genetic structuring in the endemic brown frog *Rana kukunoris* (Anura, Ranidae) of the Qinghai-Tibetan Plateau

By Weiwei Zhou, Fang Yan, Jinzhong Fu, Shifang Wu, Robert W. Murphy, Jing Che & Yaping Zhang

Frequently, Pleistocene climatic cycling has been found to be the diver of genetic structuring in populations, even in areas that did not have continental ice sheets, such as on the Qinghai-Tibetan Plateau (QTP). Typically, species distributed on the plateau have been hypothesized to retreat to south-eastern refugia, especially during the Last Glacial Maximum (LGM). We evaluated sequence variation in the mitochondrial DNA gene Cytb and the nuclear DNA gene RAG-1 in *Rana kukunoris*, a species endemic to the QTP. Two major lineages, N and S, were identified and lineage N was further subdivided into N1 and N2. The geographical distribution and genealogical divergences supported the hypothesis of multiple refugia. However, major lineages and sublineages diverged prior to the LGM. Demographical expansion was detected only in lineage S and sublineage N2. Sublineage N1 might have survived several glacial cycles in situ and did not expand after the LGM because of the absence of suitable habitat; it survived in river islands. Genetic analysis and environment modeling suggested that the north-eastern edge of QTP contained a major refugium for *R. kukunoris*. From here, lineage S dispersed southwards after the LGM. Two microrefugia in northern Qilian Mountains greatly contributed to current level of intraspecific genetic diversity. These results were found to have important implications for the habitat conservation in Northwest China.

W. W. Zhou *et al.*, *Mol. Ecol.* **22**, 130 (2013).

Spatial ecology of *Scaphiophryne gottlebei* in the canyons of the Isalo Massif, Madagascar

By Franco Andreone, Paolo Eusebio Bergò, Vincenzo Mercurio & Gonçalo M. Rosa

The Rainbow Frog *Scaphiophryne gottlebei* lives within the humid canyons that cross the Isalo Massif, central-southern Madagascar. Knowing that a single haplotype dominates the largest part of its distribution range raised questions about the dispersal ability of the species. We affixed external radio transmitters to 36 individuals of S. gottlebei to understand whether the adults of this species actively displaced from the canyon they inhabit. We studied 13 males and seven females in 2009 and seven males and nine females in 2011 over two periods (November–December 2009 and January–February 2011). Study sessions were chosen due to the different meteorological conditions: the first is the beginning of the rainy season, which corresponds to the start of reproduction; and the second being the end of the rainy season, which corresponds to

a wetter period during which individuals are more likely to be feeding in order to increase body weight. Our results revealed that there is no significant difference in the activity patterns between sexes and that rain and temperature stimulate the dispersal rate. The distance covered by the individuals did not differ between males and females; the range varied from a few centimeters to approximately 50 m, although two individuals displaced more than 100 m in a single day. These data suggest that individuals of this species are quite phylopatric to the canyons they inhabit. The generalized haplotype sharing observed might then be explained by passive dispersal of larvae and metamorphosed individuals during the intense cyclonic floods.

F. Andreone, P. E. Bergò, V. Mercurio, G. M. Rosa *Herpetologica* **69**, 11 (2013).

Bullfrog *Lithobates catesbeianus* (Amphibia: Ranidae) tadpole diet: Description and analysis for three invasive populations in Uruguay

By Mariana Ruibal & Gabriel Laufer

The North American bullfrog, *Lithobates catesbeianus* is an invasive species that has been introduced for aquaculture and escaped conducting to many invasions worldwide. This species is considered as one of the worst invasive species by the IUCN. Even though the majority of studies that evaluate bullfrog ecological effects are focused in adults or post-metamorphic stages it is noticeable that due to experimental evidence and its ecological attributes, the *L. catesbeianus* tadpole could also have significant impact on invaded communities. Bullfrog tadpoles have big body sizes and usually appear at a high density in invaded systems.

The aim of this paper was to perform a detailed qualitative and quantitative exploration of the *L. catesbeianus* larval diet. We studied and compared the gut content for three bullfrog populations in Uruguay (Departments of Canelones, Soriano and Cerro Largo). Due to its digestive length, we used a set of subsamples for each individual for the identification of consumed preys (based on regional identification keys). The most frequent preys were a broad range of microscopic algae, but the diet also included small invertebrates (especially rotifers) and eggs. Despite several differences in diet composition and number of items were found among the three study sites, one feature was common to all: the ingestion of prey with high protein levels (filamentous algae and animal prey). The trophic profile

Image of the larvae cached by a trawl net in a pond invaded by bullfrogs in the locality of Aceguá. Note the high density and biomass of bullfrog larvae in these systems. Photo: Noelia Gobel.

presented in this study suggest that bullfrog larva is an omnivorous and suggest their preference for consuming prey from lower trophic levels. Nonetheless this also could indicate a phenotipic plasticity with a consequent adaptation to a great variety of preys. Both of these hypotheses could explain the substantial growth rate of these larvae and their rapid adaptation to novel places. Native tadpoles appear to have a much poorer diet than bullfrogs. Therefore we can assume that the *L. catesbeianus* larvae will be a major competitor and a potential predator for native communities. We propose that *L. catesbeianus* tadpoles may have significant effects on invaded communities, and should be considered in future research and managements plans.

This research was partially supported by the IUCN SSC Amphibian Specialist Group, Chester Zoo Seed Grant to GL.

M. Ruibal, G. Laufer, *Amphibia-Reptilia* **33**, 3-4 (2012).

Sympatry and syntopy of the cricket frogs *Acris crepitans* and *Acris gryllus* in southeastern Virginia, USA and decline of *A. gryllus* at the northern edge of its range

By Jonathan P. Micancin, Anikó Tóth, Rachel Anderson & Jeff T. Mette

The three species of *Acris* (cricket frogs) have experienced widespread declines in the northern portions of their ranges in the eastern United States since the middle of the 20th Century. In *A. blanchardi* and *A. crepitans*, these declines have been observed for decades but remain unexplained. The recently-discovered decline of *A. gryllus* in North Carolina was obscured by sympatry and syntopy with its cryptic sibling species, *A. crepitans*, which is stable or expanding. The region of decline of *A. gryllus* is adjacent to its global northern limit, where sympatry with *A. crepitans* conceals the range limits and conservation status of both species. We investigated the historic and current ranges of sympatric *Acris* in southeastern Virginia. We established the 20th Century ranges of *Acris* using 1,769 museum catalog records from 282 collection sites and morphometric analysis of 205 specimens from 42 sites. We acoustically identified *Acris* at 140 choruses in southeastern Virginia in 2010 and 2011. Before 1990, *A. gryllus* might have ranged further north and west than expected from museum records or published maps. On the Virginia Peninsula at the putative global northern limit of the range of *A. gryllus*, the species has declined and now occurs only in syntopy with *A. crepitans*. In contrast, *A. gryllus* persists in sympatry with *A. crepitans* in the Chowan Basin and remains in allopatry in the Great Dismal Swamp. We propose adapting our approach to sympatric *A. gryllus* and *A. crepitans* to investigate other potential declines among sibling amphibians in the Atlantic Coastal Plain.

J. P. Micancin, A. Tóth, R. Anderson, J. T. Mette, *Herp. Con. Bio.* **7**, 276 (2012).

Eight new species of Pseudophilautus (Amphibia: Anura: Rhacophoridae) from Sripada World Heritage Site (Peak Wilderness), a local amphibian hotspot in Sri Lanka

By L. J. Mendis Wickramasinghe, Dulan Ranga Vidanapathirana, M. D. Gehan Rajeev, S. Chathuranga Ariyarathne, A. W. Amila Chanaka, L. L. Dharshana Priyantha, Imesh Nuwan Bandara & Nethu Wickramasinghe

Eight new species of Pseudophilautus (*Pseudophilautus bambaradeniyai, P. dayawansai, P. jagathgunawardanai, P. karunarathnai, P. newtonjayawardanei, P. puranappu, P. samarakoon,* and *P. sirilwijesundarai*) were discovered as a result of a survey carried out to study the herpetofaunal diversity with the changes in elevation in the Sripada World Heritage Site (Peak Wilderness), Central Hills of Sri Lanka. Detailed descriptions of new species along with color photographs and line drawings for each species are provided herein. The new species possess unique morphological characters and are well distinguishable from one another that could be easily identified in the field. The conservation status of all species described here, have been considered Critically Endangered, except for *P. newtonjayawardanei*, as all the new species are recorded from single locations, and their habitats are under severe threat.

L. J. M. Wickramasinghe *et al., J Threat. Taxa,* **5**, 3789 (2013).

West Africa—A Safe Haven for Frogs? A Sub-Continental Assessment of the Chytrid Fungus (*Batrachochytrium dendrobatidis*)

By Johannes Penner, Gilbert B. Adum, Matthew T. McElroy, Thomas Doherty-Bone, Mareike Hirschfeld, Laura Sandberger, Ché Weldon, Andrew A. Cunningham, Torsten Ohst, Emma Wombwell, Daniel M. Portik, Duncan Reid, Annika Hillers, Caleb Ofori-Boateng, William Oduro, Jörg Plötner, Annemarie Ohler, Adam D. Leaché & Mark-Oliver Rödel

A putative driver of global amphibian decline is the panzootic chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*). *Bd* has been documented widely across continental Africa with high number of occurrences in South and East Africa and a few in Central and North Africa which most likely rather reflects sampling intensity than real abundances. Its distribution in West Africa (west of Nigeria) remains ambiguous.

We tested 793 West African amphibians (belonging to one caecilian and 61 anuran species) for the presence of *Bd*. The samples originated from seven West African countries—Bénin, Burkina Faso, Côte d'Ivoire, Ghana, Guinea, Liberia, Sierra Leone—and were collected from a variety of habitats, ranging from lowland rainforests to montane forests, montane grasslands to humid and dry lowland savannahs. The species investigated comprised various life-history strategies, but we focused particularly on aquatic and riparian species. We used diagnostic PCR to screen swabs from 656 and histology to analyze toe clips from 137 specimens.

All samples were tested negative for *Bd*, including a widespread habitat generalist *Hoplobatrachus occipitalis* which is intensively traded, even across countries, on the West African food market and thus could be a potential dispersal agent for *Bd*. Continental fine-grained (30 arc seconds) environmental niche models suggest that *Bd* should have a broad distribution across West Africa that includes most of the regions and habitats that we surveyed.

Photographs of the four species mentioned in the abstract and of special concern with respect to Bd and conservation in West Africa: *Hoplobatrachus occipitalis* (top left), *Conraua alleni* (top right), *Petropedetes natator* (bottom left) and *Nimbaphrynoides occidentalis* (bottom right) (Credits: Mark-Oliver Rödel (top left), Johannes Penner (top right & bottom left), Laura Sandberger (bottom right))

The surprising apparent absence of *Bd* in West Africa indicates that the Dahomey Gap may have acted as a natural barrier. This is a natural gap in the otherwise continuous rain forest biome along the West African coast, consisting of savannah. In the published paper we highlight the importance of this *Bd*-free region of the African continent—especially for the long-term conservation of several threatened species depending on fast flowing forest streams (*Conraua alleni* ("Vulnerable") and *Petropedetes natator* ("Near Threatened")) as well as the "Critically Endangered" viviparous toad endemic to the montane grasslands of Mount Nimba (*Nimbaphrynoides occidentalis*) which might be impacted heavily by *Bd* if it would occur in West Africa.

J. Penner *et al.*, ***PLoS ONE*** 8, e56236 (2013).

Comparative acute and chronic sensitivity of fish and amphibians: A critical review of data

By Lennart Weltje, Peter Simpson, Melanie Gross, Mark Crane & James R. Wheeler

The relative sensitivity of amphibians to chemicals in the environment, including plant protection product active substances, is the subject of ongoing scientific debate. The objective of this study was to systematically compare the relative sensitivity of amphibians and fish to chemical exposure. Acute and chronic ecotoxicity data were obtained from the U.S. Environmental Protection Agency (U.S. EPA) ECOTOX database supplemented with additional data from the scientific and regulatory literature. Fish and amphibian toxicity data are highly correlated. Fish are generally more sensitive to chemical exposure than amphibians (after both acute and chronic exposure). In terms of acute sensitivity, amphibians were between 10- and 100-times more sensitive than fish in only four out of 55 comparisons and more than 100-times more sensitive in only two comparisons. A detailed inspection of the instances of greater amphibian sensitivity revealed a similar acute sensitivity of fish and amphibians once other data sources were also considered. Comparisons of chronic fish and amphibian ecotoxicity were possible for 52 chemicals. Amphibians were between 10- and 100-times more sensitive than fish on only two occasions (carbaryl and dexamethasone) and greater than 100-times more sensitive than fish on only a single occasion (sodium perchlorate). The comparison for carbaryl was subsequently determined to be unreliable and that for sodium perchlorate is considered to be a potential artefact of the exposure medium used in the study. Only a substance such as dexamethasone, which interferes with a specific aspect of amphibian metamorphosis, has the potential not be detected using fish tests. However, several other compounds known to influence amphibian metamorphosis were included in the analysis, and these did not affect amphibians disproportionately. These analyses suggest that fish are an appropriate surrogate for amphibians during chemical risk assessment and that additional routine testing with amphibians will not be necessary to ensure environmental protection.

L. Weltje, P. Simpson, M. Gross, M. Crane, J. R. Wheeler, *Environ. Toxicol. Chem.* accepted (2013). DOI: 10.1002/etc.2149

Iezilor Lake, Retezat National Park, Romania. Photo: Dan Cogălniceanu.

Aquatic habitat use by amphibians with specific reference to *Rana temporaria* at high elevations (Retezat Mountains National Park, Romania)

By Dan Cogălniceanu, Raluca Băncilă, Rodica Plăiaşu, Ciprian Samoilă & Tibor Hartel

Alpine areas are extreme habitats that require special adaptations and involve major trade-offs in terms of life history. Amphibians have the ability to adapt both their life history and developmental traits to alpine environments. Temperate amphibians depend on the quality and availability of aquatic habitats for reproduction. We explored the aquatic

habitat used by amphibians in the alpine area of Retezat Mountains, Southern Carpathians, Romania. We surveyed 40 aquatic habitats in a 380 ha area delimited by mountain crests and drained by a steep valley. Each aquatic habitat was characterized using 10 environmental variables. Only three amphibian species occur at elevations above 1,900 m, the most widespread being the Common frog *Rana temporaria*. The Common frog showed preference for breeding aquatic habitats, the variables of importance being altitude, solar radiation, water chemistry and grazing. Higher elevation and lower solar radiation decreased frog occurrence, while the impact of grazing favored the use of water bodies. Acidification is eminent in the area with pH dropping below 5 in 20% of the water bodies. Overall, amphibian occurrence in alpine area can be partly explained by the characteristics of aquatic habitats.

D. Cogălniceanu, R. Băncilă, R. Plăiaşu, C. Samoilă, T. Hartel, *Ann. Limnol. Int. J. Lim.* **48,** 355 (2012).

Diseases and Toxicology

Host identity matters in the amphibian-*Batrachochytrium dendrobatidis* system: Fine-scale patterns of variation in responses to a multi-host pathogen

By Stephanie S. Gervasi, Carmen Gondhalekar, Deanna H. Olsen & Andrew R. Blaustein

Species composition within ecological assemblages can drive disease dynamics including pathogen invasion, spread and persistence. In multi-host pathogen systems, interspecific variation in responses to infection creates important context dependency when predicting the outcome of disease. Here, we examine the responses of three sympatric host species to a single fungal pathogen, *Batrachochytrium dendrobatidis*, which is associated with worldwide amphibian population declines and extinctions. Using an experimental approach, we show that amphibian species from three different genera display significant differences in patterns of pathgen-induced mortality as well as the magnitude and temporal dynamics of infection load. We exposed amphibians to one of four inoculation dose treatments at both larval and post-metamorphic stages and quantified infection load on day eight and day 15 post-inoculation. Of the three species examined, only one (the Pacific treefrog; *Pseudacris regilla*) displayed "dose-dependent" responses; survival was reduced and infection load was elevated as inoculation dose was increased. We observed a reduction in survival but no differences in infection load across pathogen treatments in Cascades frogs (*Rana cascadae)*. Western toads (*Anaxyrus boreas*) displayed differences in infection load but no differences in survival across pathogen treatments. Within species, responses to the pathogen varied with life history stage and the most heavily infected species at the larval stage was different from the most heavily infected species at the post-metamorphic stage. Temporal changes in infection load were species and life history stage-specific. We show that variation in susceptibility to this multi-host pathogen is complex when viewed at a fine-scale and may be mediated through intrinsic host traits.

S. Gervasi, C. Gondhalekar, D. H. Olson, A. R. Blaustein, *PLoS ONE* **8**, e54490 (2013). DOI:10.1371/journal.pone.0054490

A non-invasive stress assay shows that tadpole populations infected with *Batrachochytrium dendrobatidis* have elevated corticosterone levels

By Caitlin R. Gabor, Matthew C. Fisher & Jaime Bosch

Batrachochytrium dendrobatidis (*Bd*) is a fungus that causes the disease chytridiomycosis and is associated with widespread amphibian declines. Populations vary in their susceptibility to *Bd* infections, and the virulence of the infecting lineage can also vary. Both of these factors may manifest as a differential physiological stress response. In addition, variation in disease susceptibility across amphibian populations may be influenced by immunosuppression caused by chronic stress imposed by environmental factors. Here, we use a non-invasive water-borne hormone technique to assess stress levels (corticosterone) of free-living tadpole populations that are infected by *Bd*. We found that corticosterone release rates were higher in infected populations of two species of tadpoles (*Alytes obstetricans* and *A. muletensis*) than in an uninfected population for both species. The relationship between corticosterone and the intensity of infection differed between species, with only the infected *A. obstetricans* population showing a significant positive correlation. The higher corticosterone release rates found in *A. obstetricans* may be an outcome of infection by a highly virulent lineage of *Bd* (*Bd*GPL), whereas *A. muletensis* is infected with a less virulent lineage (*Bd*CAPE). These results suggest that different lineages of *Bd* impose different levels of stress on the infected animals, and that this may influence survival. The next step is to determine whether higher corticosterone levels make individuals more susceptible to *Bd* or if *Bd* infections drive the higher corticosterone levels.

Alytes muletensis **tadpole during 1hr water-borne hormone collection period. Photo: Caitlin Gabor**

C. R. Gabor, M. Fisher, J. Bosch. *PLOSone* 8, e56054 (2013). http://www.plosone.org/article/info%3Adoi%2F10.1371%2Fjournal.pone.0056054

Terrestrial pesticide exposure of amphibians: An underestimated cause of global decline?

By Carsten A. Brühl, Thomas Schmidt, Silvia Pieper & Annika Alscher

Amphibians, a class of animals in global decline, are present in agricultural landscapes characterized by agrochemical inputs. Effects of pesticides on terrestrial life stages of amphibians such as juvenile and adult frogs, toads and newts are little understood and a specific risk assessment for pesticide exposure, mandatory for other vertebrate groups, is currently not conducted. The permeable skin of amphibians increases uptake processes of pesticides. The effects of seven pesticide products on juvenile European common frogs *(Rana temporaria)* were tested in an agricultural overspray scenario in a research project for the German federal Environment Agency under the lead of Carsten Brühl, University Koblenz-Landau, Germany. Acute mortality ranged from 100% after one hour in a fungicide to 40% after seven days at the recommended label rate of currently registered products. Three products showed a mortality of 40% after seven days at the lowest rate tested (10% of the label rate). Effects were not restricted to a specific class of pesticides

and seem to be influenced not only by the active substance but also the formulation additives. The demonstrated toxicity is alarming and a large-scale negative effect of terrestrial pesticide exposure on amphibian populations seems likely. Terrestrial pesticide exposure might be underestimated as a driver of their decline calling for more attention in conservation efforts and the risk assessment procedures in place do not protect this vanishing animal group.

C. A. Brühl, T. Schmidt, S. Pieper, A. Alscher, Sci. Rep. 3, 1135 (2013). DOI: 10.1038/srep0113 Open access: http://www.nature.com/srep/2013/130124/srep01135/full/srep01135.html

Blood lead levels, δ-ALAD inhibition, and hemoglobin content in blood of Giant toad (*Rhinella marina*) to assess lead exposure in three areas surrounding an industrial complex in Coatzacoalcos, Veracruz, Mexico

By César A. Ilizaliturri-Hernández, Donaji J. González-Mille, Jesús Mejía-Saavedra, Guillermo Espinosa-Reyes, Arturo Torres-Dosal & Iván Pérez-Maldonado.

Coatzacoalcos river basin has been one of the most diverse biological areas in Mexico. Actually, the Coatzacoalcos estuary houses one of the biggest and most important petrochemical industrial complexes of Mexico and Latin America. Lead is an ubiquitous environmental pollutant which represents a great risk to human health and ecosystems; it is not an essential element and it can be absorbed by many means; all of its forms are toxic. Nowadays, the magnitude of lead exposure in Mexico and other countries from Latin America is critical due to the industrial growth in the region. Amphibian populations have been recognized as biomonitors of changes in environmental conditions. The Giant toad (*Rhinella marina*) is a native and geographically widespread species in Mexico and Central America. The purpose of this research is to measure exposure to lead and evaluate hematological and biochemical effects in specimens of Giant toads taken from three areas surrounding an industrial complex in the Coatzacoalcos River downstream. Lead levels in toads' blood are between 10.8 and 70.6 μg/dL and are significant higher in industrial sites. We have found a significant decrease in the δ-ALAD activity in blood from 35.3 to 78% for the urban-industrial and industrial sites, respectively. In addition, we have identified a strong inverse relationship between the δ-ALAD activity and the blood lead levels (r=-0.84, p<0.001). Hemoglobin and mean

Giant toad (*Rhinella marina*). Photo: C. A.Ilizaliturri.

corpuscular hemoglobin levels, as well as the condition factor is found to be lower at industrial sites compared with the reference sites. Decrease pattern of hemoglobin concentration and media corpuscular hemoglobin by site can be interpreted as a secondary anemia in toads with high lead levels in blood, especially in sites with industrial influence. Our results suggest that the *Rhinella marina* can be considered a good biomonitor of the δ-ALAD activity inhibition and hematological alterations at low lead concentrations. We consider that more studies are required in the amphibian populations in particular, about the evaluation of various effects of ecological relevance, with the purpose to identify and quantify the associated risks in the populations of amphibians in the region, as well as their implications in human health and ecosystems.

C. A. Ilizaliturri-Hernández *et al*., *Environ Monit Assess*. **185**, 2 (2013).

Effects of the herbicide imazapyr on juvenile Oregon spotted frogs

By Amy E. Yahnke, Christian E. Grue, Marc P. Hayes & Alexandra T. Troiano

Conflict between native amphibians and aquatic weed management in the Pacific Northwest is rarely recognized because most native stillwater-breeding amphibian species move upland during summer, when herbicide application to control weeds in aquatic habitats typically occurs. However, aquatic weed management may pose a risk for aquatic species present in wetlands through the summer, such as the Oregon spotted frog (OSF, *Rana pretiosa*), a state endangered species in Washington. Acute toxicity of herbicides used to control aquatic weeds tends to be low, but the direct effects of herbicide tank mixes on OSFs have remained unexamined. We exposed juvenile OSFs to tank mixes of the herbicide imazapyr, a surfactant, and a marker dye in a 96-h static-renewal test. The tank mix was chosen because of its low toxicity to fish and its effectiveness in aquatic weed control. Concentrations were those associated with low-volume (3.5L/ha) and high-volume (7.0L/ha) applications of imazapyr and a clean-water control. Following exposure, frogs were reared for two months in clean water to identify potential latent effects on growth. Endpoints evaluated included feeding behavior, growth, and body and liver condition indices. We recorded no mortalities and found no significant differences for any end point between the herbicide-exposed and clean-water control frogs. The results suggest that imazapyr use in wetland restoration poses a low risk of direct toxic effects on juvenile OSFs.

A. Yahnke, C. Grue, M. Hayes, A. Troiano *Environ. Toxicol. Chem*. **32**, 228 (2013). http://onlinelibrary.wiley.com/doi/10.1002/etc.2048/abstract

Effects of a sublethal pesticide exposure on locomotor behavior: A video-tracking analysis in larval amphibians

By Mathieu Denoël, Sylvie Libon, Patrick Kestemont, Catherine Brasseur, Jean-François Focant & Edwin De Pauw

Organochlorine pesticides such as endosulfan have been shown to have both lethal and sublethal effects on amphibians. In this context, behavioral endpoints have proved their usefulness in evidencing impacts of such chemicals at environmental concentrations that do not necessarily cause mortality. The recent development of video-tracking technologies now offers the possibility of accurately quantifying locomotor behaviors. However, these techniques have not yet been applied to evaluating the toxicity of pesticides in amphibians. We therefore aimed at determining the potential toxicity of endosulfan on endpoints associated with locomotion after short-term environmental endosulfan exposure in *Rana temporaria* tadpoles and at using these data as warning systems for survival alterations after a longer exposure. To this end, we analyzed video-tracks of 64 tadpoles (two pesticide treatments: 5 and 50 μg L−1, one control and one solvent-control) with Ethovision XT 7 software. The highest endosulfan concentration had a significant effect on all four behavioral endpoints. Contaminated tadpoles traveled shorter distances, swam less often, at a lower mean speed and occupied a less peripherical position than control tadpoles. The lowest endosulfan concentration had similar but lower effects, and did not affect mean speed during swimming. Survival was reduced only after a long-term exposure to endosulfan and was associated with short-term behavioral dysfunctions. These results show that endosulfan strongly affects the behavioral repertory of amphibian tadpoles, but in different ways depending on concentration, thus suggesting that the

pesticide has complex modes of action. Given the importance of locomotion and space use in tadpole success in their aquatic environment, these results confirm the toxic action of endosulfan. By highlighting effects before mortality markers, video-tracking systems also show their potential as sentinels of sublethal effects of pesticides.

M. Denoël *et al.*, *Chemosphere* **90**, 945 (2013). http://hdl.handle.net/2268/127925

Amphibian chytrid prevalence in an amphibian community in arid Australia

By Joanne F. Ocock, Jodi J. L. Rowley, Trent D. Penman, Thomas S. Rayner & Richard T. Kingsford

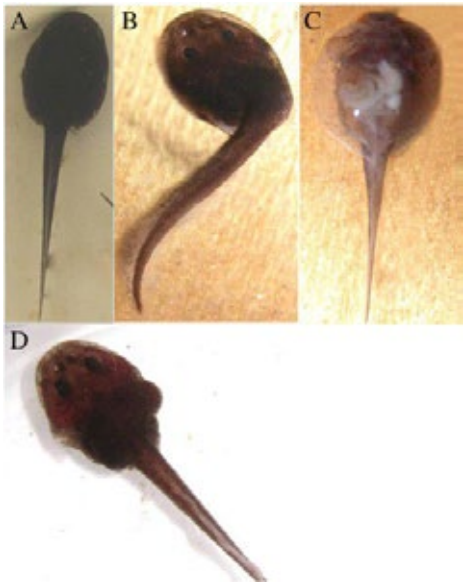
Chytridiomycosis, caused by the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*), has been implicated in amphibian population declines on most continents around the world. Surveys for *Bd* have been biased towards mesic regions, potentially affecting current understanding of the impact of *Bd*, particularly in arid regions. We systematically surveyed for the prevalence and intensity of *Bd* at a site in arid Australia predicted to be unsuitable for *Bd* on the basis of climatic suitability models. A total of 271 individuals were sampled, and 17 individuals from seven species tested positive for *Bd*. Overall infection prevalence was 6.36% (95% CI 3.40 – 9.85%), relatively low compared with other regional surveys. Infection intensities were low (<10 Z swab) with no evidence of amphibian morbidity, mortality or population declines at our site. The presence of *Bd* was not significantly associated with habitats or species. Combined with two recent *Bd* records, our study shows that contrary to predictions, *Bd* is present and may pose a threat to arid amphibian populations in Australia. Large scale *Bd* modelling is limited by true absences of *Bd* occurrence and therefore unable to accurately predict *Bd* occurrence. Further research is needed to increase our understanding of the prevalence and potential impact of *Bd* on arid amphibians.

J. F. Ocock, J. J. L. Rowley, T. D. Penman, T. S. Rayner, R.T. Kingsford, *Ecohealth* DOI 10.1007/s10393-013-0824-8 (2013).

Malformations and mortality in the Asian common toad induced by exposure to *pleurolophocercous cercariae* (Trematoda: Cryptogonimidae)

By Uthpala A. Jayawardena, Vasyl V. Tkach, Ayanthi N. Navaratne, Priyanie H. Amerasinghe & Rupika S. Rajakaruna

Malformations and increased mortality due to infection by the digenetic trematode, *Riberioa ondatrae* have been reported for many species of amphibians. Severe malformations have also been reported in the Common hourglass tree frog, *Polypedates cruciger* induced by pleurolophocercous cercariae in Sri Lanka in addition to the changes in the behavior, development and survival of the host. We exposed pre-limb bud stage tadpoles (Gosner stages 25–26) of the Asian common toad, *Duttaphrynus melanostictus* to the same pleurolophocercous type cercariae under laboratory conditions. Molecular and morphological identification showed that these cercariae belonged *Acanthostomum burminis* infecting freshwater snakes as definitive hosts. These cercariae induced malformations (27.8%) and reduced survival to metamorphosis (53.8%). The magnitude of the effects increased with the dose of cercariae. Types of malformations were mainly axial, such as scoliosis and kyphosis. Severe limb malformations such as extra or missing limbs as reported for amphibians exposed to *R. ondatrae* were not observed in the *D. melanostictus*. Same authors reported a higher percentage of malformations previously when *P. cruciger* was exposed to the cercariae *A. burminis* compared to *D. melanostictus*. However, tadpoles of *D. melanostictus*, which are smaller compared to those of *P. cruciger*, experienced higher mortality than *P. cruciger* tadpoles. Trematode induced malformations and mortality in amphibians are highly variable and depend on multiple factors such as host species differences such as resistance to infection and tolerance, life-history characteristics such as size at metamorphosis and length of the metamorphosis period, and other factors such as size of the amphibian at the time of trematode exposure.



Tadpoles of *Duttaphrynus melanostictus*, (A) Uninfected (B) infected tadpoles with signs of early kyphosis, (C) infected tadpole with an edema on the left side of the body (D) infected tadpole with a lump on the right side of the body.

U. A. Jayawardena, V. V. Tkach, A. N. Navaratne, P. H. Amerasinghe, R. S. Rajakaruna, Parasitol. int. 62 (3). 246-252.

Techniques for minimizing the effects of PCR inhibitors in the chytridiomycosis assay

By Tiffany A. Kosch & Kyle Summers

Chytridiomycosis is an amphibian disease of global conservation concern that is caused by the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*). Since the discovery of *Bd* in 1998, several methods have been used for detection of *Bd*; among these polymerase chain reaction (PCR) from skin swabs is accepted as the best method due to its noninvasiveness, high sensitivity and ease of use. However, PCR is not without problems—to be successful, this technique is dependent upon the presence of nondegraded DNA template and reaction contents that are free from inhibitors. Here, we report on an investigation of several techniques aimed at improving the reliability of the *Bd* PCR assay by minimizing the effects of humic acid (HA), a potent PCR inhibitor. We compared the effectiveness of four DNA extraction kits (DNeasy, QIAamp DNA Stool, PowerLyzer Power Soil and PrepMan Ultra) and four PCR methods (Amplitaq Gold, bovine serum albumin, PowerClean DNA Clean-up and inhibitor resistant Taq Polymerase). The results of this and previous studies indicate that chytridiomycosis studies that use PCR methods for disease detection may be significantly underestimating the occurrence of *Bd*. Our results suggest that to minimize the inhibitory effects of HA, DNeasy should be used for sample DNA extraction and Amplitaq Gold with bovine serum albumin should be used for the *Bd* PCR assay. We also outline protocols tested, show the results of our methods comparisons and discuss the pros and cons of each method.

T. Kosch and K. Summers, *Mol. Ecol. Res.* **13**, 230-236 (2013).

Parallels in amphibian and bat declines from pathogenic fungi

By Evan A. Eskew & Brian D. Todd

Pathogenic fungi have substantial effects on global biodiversity, and two emerging pathogenic species—the chytridiomycete *Batrachochytrium dendrobatidis* (*Bd*), which causes chytridiomycosis in amphibians, and the ascomycete *Geomyces destructans*

(*Gd*), which causes white-nose syndrome in hibernating bats—are implicated in the widespread decline of their vertebrate hosts. We synthesized current knowledge for chytridiomycosis and white-nose syndrome regarding disease emergence, environmental reservoirs, life history characteristics of the host and host–pathogen interactions. We report striking similarities between these aspects of chytridiomycosis and white-nose syndrome. Genetic evidence suggests that both diseases are largely driven by novel pathogens introduced to new geographic regions, likely due to anthropogenic activities. *Bd* and *Gd* are both host-generalist pathogens with abiotic reservoirs and may therefore persist in the environment even when the density of host species is low. Furthermore, the life history characteristics of many amphibian and bat species result in high host densities, high intraspecific contact rates, and reduced immune function during specific life stages or seasons, all of which contribute to pathogen persistence and transmission within a host community. Finally, both *Bd* and *Gd* are dermatophyte fungi that appear to successfully overcome vertebrate host defenses in some of their primary host species through mechanisms that are not yet well understood. We reiterate previous calls for improved biosecurity and additionally suggest targeted monitoring of species that are most likely to suffer from emerging fungal diseases as management actions that should help mitigate the negative impacts of pathogenic fungi.

E. A. Eskew, B. D. Todd, *Emerg. Infect. Dis.* **19**, 379 (2013).

Promoting amphibian conservation through the college classroom: Detection of *Batrachochytrium dendrobatidis* among local amphibians

By Julie L. Wunder, Noel M. Lampazzi, Kelsey D. Acre, Nicholas J. Bent, Sadie A. Canter, Alexandra M. Chapman, Margaret A. Davies, David Kashan, Jonathan W. Keiley, Rachel I. MacIntyre, Tamara F. Milton, Kara L. Weichler, Matt J. Wilson & Mizuki K. Takahashi

Many global amphibian declines have been linked to the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*). The knowledge on *Bd* distribution provides a fundamental basis for amphibian conservation planning. Yet, such *Bd* distribution information is currently insufficient, in particular at a regional scale. The college classroom provides an excellent opportunity to expand the knowledge of *Bd* distribution. Here we provide an example of such research projects to detect *Bd* prevalence among local amphibians in a college course setting and present the results of work conducted in central Pennsylvania, USA. We collected toe clips and conducted PCR assays of six species, *Plethodon cinereus, Desmognathus fuscus, Notophthalmus viridescens, Lithobates catesbeianus, L. clamitans* and *L. sylvaticus* (59 individuals). Four groups of students independently conducted entire projects, orally presented their findings and submitted manuscripts to the professor at the end of the semester. This example demonstrates that it is feasible for an undergraduate class to complete a *Bd*-detection project within a single semester. Such a project not only contributes to *Bd* research but also promotes conservation

Field sample collection by students in an Amphibian Biology course at Bucknell University. Photo: Mizuki K. Takahashi.

education among students through hands-on research experiences. We found *Bd* infection in only one sample of *N. viridescens*, but no sign of infection in the rest of the samples. As a relatively high prevalence of *Bd* has been reported in surrounding areas, our results suggest spatial heterogeneity in *Bd* occurrence at a regional scale and thus, the need for continued efforts to monitor *Bd* prevalence.

Wunder *et al., Herpetol. Conserv. Biol.* **7**, 462 (2012).

Call for recent publication abstracts

If you would like to include an abstract from a recent publication in this section of *FrogLog* please email: froglog@amphibians.org. We also encourage all authors of recent publications to inform Professor Tim Halliday (formerly DAPTF International Director) (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

Upcoming Meetings & Workshops

April

19-21, Joint meeting of North Carolina PARC and the North Carolina Herpetological Society-NC Zoo, Asheboro, NC

May 2013

13–24, Graduate and Professional Course - Species Monitoring and Conservation: Reptiles - Smithsonian Conservation Biology Institute, Front Royal, VA, USA

13–24, Coastal Herpetology Course - Univeristy of Southern Mississippi, Gulf Coast Research Laboratory, Ocean Springs, MS

June 2013

12–15, 8th Annual Great Basin Kingsnake Survey – Great Basin National Park, Baker, NV.

July 2013

8–12, 2013 Spotted Frog Survey – Indian Valley, NV.

8–10, Southwest PARC Annual Meeting – University of New Mexico, Albuquerque, NM.

10–15, Joint Meeting of Herpetologists and Ichthyologists - Hosted by University of New Mexico Museum of Southwestern Biology – Albuquerque Convention Center, Albuquerque, NM

16, World Snake Day!

Internships & Employment

The following information can be found at: http://www.parcplace.org/resources/job-listings.html. Herp jobs are posted as a service to the herpetological community. If you would like to list a job opening for your organization, please send the announcement to: herpjob@parcplace.org

Priority Amphibian and Reptile Conservation Areas (PARCAs) Field Technician - The Orianne Society

North Carolina, South Carolina, Georgia, USA (Posted 03/21/2013; Closing March 31, 2013)

Timber Rattlesnake Spatial Ecology Technician - The Orianne Society (TOS)

Nantahala Mountains of Georgia and North Carolina (Posted 03/21/2013; Closing March 28, 2013)

Flat-tailed Horned Lizard Occupancy study, Arizona Game and Fish Department

Yuma Desert of Arizona (Posted 03/21/2013; Closing 03/22/2013)

Northeast Regional Blanding's Turtle Field Technician, Massachusetts Cooperative Fish and Wildlife Research Unit

Amherst, MA. (Posted 03/21/2013; Closing 4/3/13)

Wyoming Toad Field Technician Position - Mortenson Lake National Wildlife Refuge

Laramie, WY (Posted 03/14/13; Closing March 25, 2013)

Amphibian Research Internship - USGS Northeast Amphibian Research and Monitoring Initiative

Patuxent Wildlife Research Refuge, Laurel, MD (Posted 03/11/13; Open till filled)

Desert Tortoise Telemetry Research Associate

Las Vegas, NV (Posted 03/01/13; Closing March 14, 2013)

Research Assistant for Bog Turtle and Bat Work (Seasonal) - Delaware Division of Fish and Wildlife

Smyrna, DE (Posted 02/28/13; Closing March 22, 2013)

Desert Tortoise Research Assitant - Penn State University

Fort Irwin National Training Center near Barstow, CA and Henderson, NV (Posted 02/28/13; Closing Mid - Late March)

Wildlife Inventory Intern Vacancy - HRM

Ann Arbor and Lansing areas, MI (Posted 02/28/13; Closing March 22, 2013)

GIS Intern Vacancy - HRM

Ann Arbor and Lansing areas, MI (Posted 02/28/13; Closing March 22, 2013)

Fisheries / Wildlife Technicians (7) - USGS Forest and Rangeland Ecosystem Science Center

Corvallis, OR (Posted 02//28/13; Closing 20 March, 2013)

Wood Turtle Surveyor - Massachusetts Natural Heritage and Endangered Species Program

West Boylston, MA (Posted 02/21/13; Closing March 6, 2013)

Blanding's Turtle Surveyor - Massachusetts Natural Heritage and Endangered Species Program

West Boylston, MA (Posted 02/21/13; Closing March 6, 2013)

Graduate Student Position in Stream Salamander Ecology and Conservation - University of Arkansas

Fayetteville, AR (Posted 02/15/2013; Closing 03/15/13)

Wood Turtle Monitors

Northeast US (Posted 02/15/2013)

Venomous Snake Construction Monitor

Northeast US (Posted 02/15/2013)

Field Technician, Texas AM University

Maljamar, New Mexico, U.S.A. (Posted 02/09/2013; Closing 03/01/13)

Biologist Supervisor, State of Louisiana

Lafayette, LA (Posted 01/29/2013; Closing 02/11/13)

Biological Science Technician, Humboldt-Toiyabe National Forest

Austin, NV (Posted 01/29/2013; Closing 03/01/13)

Volunteer Herpetological Field Technician, Southern Illinois University

Nachusa Grasslands, Franklin Grove, IL (Posted 01/29/2013; Closing 04/30/13)

Unpaid Internship/Volunteer Position - Northern Pacific Rattlesnake/California Ground Squirrel Research - Ohio State University

Central and Northern California, USA (Posted 1/24/13; open till filled)

Graduate Research Position - Applied Population Ecology - Penn State University

University Park, PA (Posted 1/23/13; Closing 03/01/2013)

Summer Research Internship at The Wetlands Institute through CNAH

The Wetlands Institute, Cape May Peninsula of southernmost New Jersey (Posted 1/14/13; Closing 03/01/2013)

Field technicians (3 Positions Available), Columbia Spotted Frogs in the Bighorn Mountains of Wyoming

The University of Wyoming, Bighorn Mountains (north central Wyoming) (Posted 1/11/13; Closing 03/08/2013)

Sea Turtle Research Jobs in Australia

Kensington (Perth), Western Australia (Posted 1/8/13; Closing 1/24/13)

Field Technicians - Anuran and Turtle Research in Western Iowa

Western Iowa (Posted 1/2/13; Closing 2/4/13)

Biological Science Positions (2 openings), Yosemite National Park

Yosemite, CA (Posted 12/22/12; Closing Jan. 10, 2013)

Graduate Opportunities to Study Salamanders, Department of Biology at San Francisco State University

San Fransisco, CA (Posted 12/19/12; No Closing Date Provided)

Full-Time Laboratory Technician in Reptile Facility, Stowers Institute for Medical Research

Kansas City, MO (Posted 12/19/12; No Closing Date Provided)

Herpetological Researcher/Educator Internship - Research 4 Reptiles LLC

Wilmington, IL (Application Deadline April 1, 2013)

Funding Opportunities

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

April 2013

Critical Ecosystem Partnership Fund (CEPF)—Biodiversity Conservation in the Eastern Afromontane Region. CEPF announces a second round of grants 2012-2017 for biodiversity conservation in the Eastern Afromontane countries: Burundi, Dem Rep Congo, Eritrea, Ethiopia, Kenya, Malawi, Mozambique, Rwanda, South Sudan, Tanzania, Uganda, Yemen, Zambia, and Zimbabwe. Eligible applicants are NGOs, community groups, private enterprises, universities, and other civil society organizations. Government institutions are eligible in certain circumstances. The deadlines for letters of inquiry (LOIs) are 01 April 2013 for large grants (i.e., above US$20 thousand), and 19 April 2013 for small grants. Link

European Commission (EC)—Forest Law Enforcement, Governance, and Trade in Guyana. The EC will support the participation of national civil society organizations and national private sector federations and entities in the EC's program for Forest Law Enforcement, Governance, and Trade (FLEGT). Grants ranging from €100 thousand to €150 thousand are available to non-state organizations legally established in Guyana, possibly working with international partners. The deadline for concept notes is 01 April 2013. Link

European Commission (EC)—Support for FLEGT in the Ivory Coast. The EC announces funding to help increase the participation of civil society organizations and the private sector in processes related to Forest Law Enforcement, Governance, and Trade (FLEGT) in Cote d›Ivoire. The program is open to NGOs and government entities in EU member states and Ivory Coast, and to international organizations. Grants range from €250 thousand to €400 thousand, depending on cost shares. Reference EuropeAid/133775/L/ACT/CI. The application deadline is 30 April 2013. Link

International Association for the Study of the Commons—Elinor Ostrom Award. With financial support from the Ford Foundation, the International Association for the Study of the Commons (IASC) requests nominations for the first Elinor Ostrom Award on Collective Governance of the Commons, in honor of the pioneering work in this area by Professor Ostrom. The award has three categories: young scholars; senior scholars; and practitioners. Three winners will be selected in each category. First-place winners will receive US$10 thousand; second-place winners will receive US$5 thousand. All winners will be awarded travel grants for IASC's 2013 conference in Japan. The closing date for nominations is 08 April 2013. Link

International Development Research Center—Climate Change Adaptation in Africa and Asia. Canada's IDRC, jointly with the UK's Department for International Development (DFID), will fund three research consortia under the Collaborative Adaptation Research Initiative in Africa and Asia (CARIAA). The goal is to increase the resilience of the poorest and most vulnerable populations in the climate "hot spots" of CARIAA's eligible countries, drawing on regional and inter-regional collaboration. The program is open to research teams from academic institutions; private sector organizations that have a research mandate; NGOs and other civil society organizations; and research-based organizations. Grants are up to CAN$15 million per consortium. The deadline for concept notes is 04 April 2013. Link

International Union for the Conservation of Nature (IUCN)—Mangrove Management in India. IUCN-India invites applications for grants in support of Mangroves for the Future (MFF). The grants will fund small-scale coastal restoration and management by local communities, small-scale businesses, or research organizations in priority geographical locations of India. Grants are from US$10 thousand to US$20 thousand for projects of 12-18 months. The deadline for proposals is 05 April 2013. Link

Korea Green Foundation—Green Asia Grants 2013. The Korea Green Foundation makes grants to civil society organizations in the Asia-Pacific region that work on projects related to climate change, renewable energy, and other environmental issues in local areas. Grants of US$5 thousand support education, campaigns, and research. The application period closes 05 April 2013. Link

Seeding Labs—Equipment Transfer Program 2013. Seeding Labs aims to help talented scientists in the developing world by transferring high-quality equipment for their teaching and research in areas related to the natural sciences. Applicants for the equipment are public universities in low and middle-income countries. The deadline for preliminary submissions is 01 April 2013. Link

May 2013

Council for the Development of Social Science Research in Africa (CODESRIA)—Comparative Research Networks 2013. CODESRIA supports researchers in African universities and research centers through funding for Comparative Research Networks. The networks address themes within CODESRIA's strategic plan, including (among others) research on climate change, natural resources, and development. Most grants range from US$10 thousand to US$30 thousand. The application deadline is 31 May 2013. Link

European Commission (EC)—Modernization of the Forestry Sector in Honduras. The EC will support the government of Honduras to improve the governance of forests, protected areas, and wildlife in Honduras. Grants will focus on building the capacity of local governments and communities in forest protection and management. The program is open to nonprofit organizations in the EU, Honduras, countries of the EU›s Development Cooperation Instrument, and international organizations. Grants are €100 thousand to €500 thousand, subject to co-financing shares. Reference EuropeAid/134023/M/ACT/HN. The deadline for proposals is 27 May 2013. Link

Kurita Water and Environment Foundation – Research Grants 2013. The KWEF makes grants in China and Southeast Asia 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 500 thousand yen per project per year. Applicants are preferably under age 40. Each applicant should have a Japanese advisor in Japan or in the researcher's home country. The application period is 01 April 2013 through 16 May 2013. Link

University of Cape Town—CSAG Winter School 2013. The Climate Systems Analysis Group (CSAG) at the University of Cape Town organizes a "winter school" intensive course in July. The course is presented by facilitators from CSAG and other institutions to take participants through the full spectrum of topics related to climate and climate change, with an emphasis on issues in developing countries. CSAG will offer partial sponsorships to some participants. The application deadline is 03 May 2013. Link

U.S. Fish and Wildlife Service—Conservation of Amphibians in Decline 2013. Amphibians in Decline supports activities that address threats to frogs, toads, salamanders, newts, and caecilians that face an unprecedented threat of extinction. Eligibility for grants extends to individuals, multi-national secretariats, government units (all levels), nonprofit NGOs, and institutions of higher education. Preference is for proposals that request less than US$25 thousand, although larger proposals will be considered. Proposals should demonstrate in-kind or financial matching. The closing date for applications is 01 May 2013. Link

U.S. Fish and Wildlife Service—Critically Endangered Animals Conservation Fund 2013. The Critically Endangered Animals Conservation Fund makes grants for projects that conserve the world's most endangered species. Species eligible for funding are those that face a very high risk of extinction in the immediate future; that are not located in North America or high-income countries of Europe; and that are not included in other USFWS programs. Eligibility for grants is unrestricted. The USFWS favors proposals that request under US$25 thousand, and that contribute complementary resources. Funding Opportunity F13AS00106. The deadline for applications is 01 May 2013. Link1 Link2

U.S. National Science Foundation—Dimensions of Biodiversity 2013. The goal of Dimensions of Biodiversity is to promote novel and integrated approaches to understand the evolutionary and ecological significance of biodiversity. The program currently focuses on the integration of genetic, taxonomic/phylogenetic, and functional dimensions of biodiversity. It includes international partnerships when relevant, such as with research organizations in China and Brazil. The deadline for full proposals is 06 May 2013. Link

July 2013

Blue Earth Alliance—Support for Photography Projects. Blue Earth sponsors photography projects that educate the public about threatened cultures, endangered environments, and other social concerns. Blue Earth provides assistance with fund raising, publishing, and publicity. However, it does not make direct grants to sponsored projects. The deadlines for submissions are 20 January and 20 July of each year. Link

Club300 Foundation for Bird Protection—Grants 2013. The Club300 Foundation, based in Sweden, makes grants of up to US$5 thousand for bird protection on a worldwide basis. The objective is research and conservation of the world's highly threatened and poorly known bird species (IUCN's Red List). The deadline for applications (English or Swedish) is 31 July 2013. Link

Ford Motor Company in the Middle East—Grants for Conservation and Environment 2013. Ford Middle East makes grants for non-profit conservation initiatives in the Middle East. Categories are conservation engineering; protection of the natural environment; and environmental education. Ford will allocate US$100 thousand for projects in Bahrain, Iraq, Jordan, Kuwait, Lebanon, Oman, Qatar, Saudi Arabia, and the United Arab Emirates. The application deadline is 01 July 2013. Link

August 2013

Christensen Fund—Biodiversity and Cultural Diversity 2013. The Christensen Fund makes grants to indigenous-led and community-based organizations for projects that combine biodiversity with cultural diversity. Most grants are in the range of US$50 thousand to US$100 thousand for one or two years. Application dates are 01 August 2013 through 31 August 2013 for project pre-proposals in the following bio-cultural regions: African Rift Valley; Central Asia; and Greater Southwest (i.e., southwestern USA and northern Mexico). The application period is 01 September 2013 through 30 September 2013 for projects in Melanesia. Link

Fulbright Scholar Program—Core Program for U.S. Faculty and Professionals 2014-2015. 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 01 August 2013. Link

U.S. National Science Foundation—International Research Experiences for University Students 2013. Through the program IRES, the National Science Foundation supports U.S. university students (graduate and undergraduate) to engage in international research. Past fellowships include several in developing countries in subject areas such as ecology, renewable energy, climate change, and others. Proposals are due 20 August 2013. Link

Selected Advance Postings

Christensen Fund—Biodiversity and Cultural Diversity 2013. The Christensen Fund makes grants to indigenous-led and community-based organizations for projects that combine biodiversity with cultural diversity. Most grants are in the range of US$50 thousand to US$100 thousand for one or two years. Application dates are 01 August 2013 through 31 August 2013 for pre-proposals representing the following bio-cultural regions: African Rift Valley; Central Asia; and Greater Southwest (i.e., southwestern USA and northern Mexico). The application period is 01 September 2013 through 30 September 2013 for projects in Melanesia. Link

International Land Coalition—Protecting Land Rights for the Rural Poor. The International Land Coalition announces the «Facility in Support of Innovative and High-Impact Targeted Interventions» (FTI) as a new program to help the rural poor gain access to and control over land and other natural resources. The FTI is primarily intended to support national and local civil society organizations in developing countries worldwide. Most grants will be under US$30 thousand for interventions of up to one year. FTI requires a minimum 20 percent co-funding. Proposals are invited in English, Spanish, and French on a continuous basis through October 2015. Link

U.S. Agency for International Development—Global Development Alliance 2013. The Global Development Alliance seeks to leverage resources from partner organizations to complement USAID›s program funding. USAID welcomes applications from private businesses, business and trade associations, foundations, NGOs, faith-based organizations, international organizations, colleges and universities, civic groups, regional organizations, and others—in the USA and internationally. Proposals must offer resource contributions of at least 1:1 with USAID. Proposals should be directly related to USAID's programs at the country level (i.e. the USAID missions), or to programs based in USAID-Washington. Funding Opportunity APS-OAA-13-000003. The closing date for applications is 31 January 2014. 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.

Production Editor

Candace M Hansen: cmhansen@amphibians.org

Editorial Committee

Claude Gascon (ASG Co-Chair)
Phillip J. Bishop (ASG Co-Chair)
James P. Lewis (ASG Program Officer)
Robin D. Moore (ASG Program Officer)
Candace M. Hansen (ASG Program Officer)
Craig Hassapakis (Co-editor, Amphibian and Reptile Conservation)

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 English.

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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Hypsiboas punctatus. Photo by: Katy Upton.