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froglog

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Conservation news for the herpetological community

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Regional Focus

North and Central America, and the Caribbean

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Robin Moore / iLCP

Editorial

FrogLog has undergone something of a metamorphosis in the past year or so with thanks to the vision of James Lewis and input of the ASG community. A year ago, almost to the day, we launched the first "Regional Focus" edition to shine the spotlight on the impressive array of work being done in different regions, both in understanding and combating amphibian declines. The response to this new format was inspiring, as are the stories of success from around the world.

We are bombarded on a daily basis by negative stories from the global media, and news about the state of our environment is rarely positive. But every now and then we are offered glimmers of hope that can serve to empower and inspire. Earlier this month Carlos Vasquez Almazan traveled to London to receive a prestigious Whitley Award for his incredible contribution to amphibian conservation in Guatemala. Carlos has been a key player in identifying and securing key habitat for amphibians including the Sierra Caral, home to a dozen globally threatened amphibians, five of which are found nowhere else in the world. Carlos is an inspiring example of an individual working tirelessly to bridge the gap between science and conservation, and has been a valuable ASG partner. The ASG is also proud to have supported the recent creation of the *Rana Terribilis* Amphibian Reserve in Colombia, another conservation victory thanks to an collaboration among international and local partners. And then of course there is the Search for Lost Frogs, a simple idea that has caught the attention of the masses from scientist to school children.

While we should not mislead people to believing that there is not a problem, we must give them something to believe in if we want them to support the cause to protect amphibians. This sentiment was echoed in a recent comment on Facebook in response to a post about the conservation efforts for the Mountain chicken frog (*Leptodactylus fallax*): "Successful programs like this need to become more widely known. I think people are willing to support them if they are aware that they pay off." So let's keep raising awareness about the plight of amphibians, let's document and learn from our failures, but let's also be sure to celebrate and spread the word about our successes, no matter how small or trivial they may seem, because these success stories may just help foster support for the cause.

Robin D. Moore
ASG Program Officer

Froglog

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Amphibian Red List Internship Available

The Amphibian Red List Authority seeks a volunteer intern to assist with maintenance and update of the amphibian conservation assessment database in the IUCN Red List of Threatened Species. The intern will receive formal training in the IUCN Red List Categories and Criteria, IUCN's Species Information System (SIS) database and IUCN's mapping standards. The internship will involve helping update the amphibian database and liaising with experts as needed to complete draft assessments. Potential interns should have a good understanding of amphibian taxonomy, declines and conservation, strong internet-search and data-mining skills and experience with GIS. Familiarity with operating and managing web systems as well as knowledge of the IUCN Red List Categories and Criteria will be an asset. All interns must have an advance level of English, both spoken and written. While this is an unpaid internship, the intern will receive a free copy of Threatened Amphibians of the World at the end of the internship. For more information please contact Ariadne Angulo - ariadne.angulo@iucn.org.

Lost Frogs update – Research Assistant Needed

Thank you to all those who provided updates to the current lost frogs list published in the last edition of FrogLog. There have been a number of updates to the list, discussed below. The continued response we are receiving to in relation to this initiative means that we are looking for some additional support to help manage and develop the global initiative. We are looking for someone ideally working on an MSc or Phd who would be able to utilize the data generated by the searches for lost species.

The position would initially be on a voluntary basis however once in place there would be the opportunity to develop funding proposals in conjunction with the ASG and our partners. The focus of the position would be to:

- Develop a system for managing data related to lost species
- Collaborate with the Amphibian Red List Authority to update assessments relating to Lost species.
- Develop and refine our understanding and definition of what constitutes a lost and a rediscovered species
- Assist with raising funds to support ongoing searches and post rediscovery conservation and research efforts

Candidates will require a sound knowledge of database and web site development, and an interest in amphibian conservation. Experience with fund raising, media relations, GIS and the Red Listing process would all be beneficial.

If you are interested in taking on this fantastic opportunity to be at the forefront of the Search for Lost Frogs please contact Robin Moore rdmoore@amphibians.org .

The Second Call for Proposals for SOS - Save Our Species is now open

SOS – Save Our Species is a joint initiative of IUCN, the Global Environment Facility and the World Bank supported by Nokia and the Fonds français pour l'environnement mondial (FFEM). Its objective is to ensure the long-term survival of threatened species and their habitats. Since its launch in October 2010 in Nagoya, SOS has already funded 28 projects targeting threatened mammals, birds and amphibians in more than 27 countries.

Applications for Threatened Species Grants will be accepted for the following Strategic Directions:

- Threatened tropical Asian vertebrates
- Threatened small marine mammals
- Threatened cycads
- Threatened freshwater African animals

Detailed information for applicants, including eligibility criteria and application templates are on the SOS website. www.sospecies.org/sos_projects/apply_for_a_grant/

EPA and the Ecological Risks from the Use of Atrazine

FIFRA Scientific Advisory Panel; Notice of Public Meeting

There will be a 4-day meeting of the Federal Insecticide, Fungicide, and Rodenticide Act Scientific Advisory Panel (FIFRA SAP) to consider and review Problem Formulation for the Reassessment of Ecological Risks from the Use of Atrazine. The meeting will be held on June 12-14, 2012, from 9 a.m. to approximately 5:30 p.m. and on June 15, 2012, from 9 a.m. to approximately 12:30 p.m.

The Agency encourages that written comments be submitted by May 29, 2012, and requests for oral comments be submitted by June 5, 2012. However, written comments and requests to make oral comments may be submitted until the date of the meeting, but anyone submitting written comments after May 29, 2012, should contact the Designated Federal Official (DFO. Further information can be found at https://www.federalregister.gov/articles/2012/04/04/2012-8085/fifra-scientific-advisory-panel-notice-of-public-meeting

Updates to the Lost Frogs list

Species reported as Found: *Isthmohyla tica, Odontophrynus moratoi, Incilius holdridgei, Craugastor fleischmanni, Centrolene heloderma* and *Litoria castanea*.

Species added to the Lost list: *Atelopus senex* and *Craugastor andi*

See the full list on our web site: www.amphibians.org

FrogLog Feature Editors Wanted

A year on from our first "Regional Focus" edition FrogLog continues to grow in readership and diversity of content. Not wanting to stand still however, and in response to readers requests, we are interested in producing FrogLog editions focused around thematic areas. We are looking for volunteer guest editors willing to leading the production of an edition focused on any thematic area of choice related to amphibian conservation.

As the ASG was formed largely in response to the creation of the Amphibian Conservation Action Plan, we would be particularly interested in focal editions around any of these topics:

- Designing a Network of Conservation Sites for Amphibians
- Freshwater Resources and Associated Terrestrial Landscapes
- Climate Change, Biodiversity Loss, and Amphibian Declines
- Infectious Diseases
- Over-harvesting
- Evaluating the Role of Environmental Contamination in Amphibian Population Declines
- Captive Programs
- Genome Resource Banking
- Reintroductions
- The Continuing Need for Assessments: Making the Global Amphibian Assessment an Ongoing Process
- Systematics and Conservation
- Bioresource Banking Efforts in Support of Amphibian Conservation

There are of course many other topics we would be interested in covering, such as the role of citizen science and capacity building within the amphibian conservation community, and so we are also open to suggestions. We hope that this will provide an opportunity to bring together different groups to showcase some of the current thinking and provoke discuss around the topics while identifying new challenges and opportunities for the ASG.

The Regional format of FrogLog will continue with thematically-focused material being included when ready for publication. If sufficient material is available there will be the opportunity to publish a special edition, supplementary to the regular schedule.

If you are interested in being a guest editor for FrogLog and developing a thematically-focused edition please contact James Lewis – jplewis@amphibians.org.

Wanted: Partnership in Husbandry Research on the Frogs of Andasibe, Madagascar

The community-run conservation organization Mitsinjo operates a 185 m^2 biosecure amphibian captive breeding and husbandry research facility, one of the goals of which is to conduct husbandry studies on frog species which have been assessed as being at high-risk from amphibian chytrid, or which can serve as surrogate species for other already threatened frog species from elsewhere in Madagascar. Of the more than 286 species of frogs endemic to the island, only a couple dozen have ever been managed in captivity. To facility *ex-situ* conservation actions as well as potential rescue operations due to the threat of infectious diseases, it is necessary to understand the captive husbandry of the species not yet kept or bred in captivity.

We currently are seeking a partnership with 1 to 2 academically-minded people to help design hypothesis driven husbandry studies on frog species from Andasibe, and to assist with analyzing the resulting data. The ideal person(s) would have a broad understand of amphibian biology and captive husbandry, as well as a sound scientific background. Masters and PhD candidates that could adapt this work into their current studies would be perfect, as would others with a strong academic background, as well as skilled zoo keepers or private breeders. Mitsinjo technicians would conduct and carryout the studies at the breeding facility, and then work with the partner to interpret the results.

Communication and discussion about the research projects will be discussed through email or Skype and it is not required to travel to Madagascar, however, if desired the project partner would be welcome in Andasibe with free housing provided by Mitsinjo. If interested, please email a current CV and Cover Letter addressed to Devin Edmonds devin@amphibiancare.com. Additional information about Mitsinjo and our amphibian conservation activities can be found at www.mitsi njo.org and www.sahonagasy.org

NHBS Gratis Books Scheme

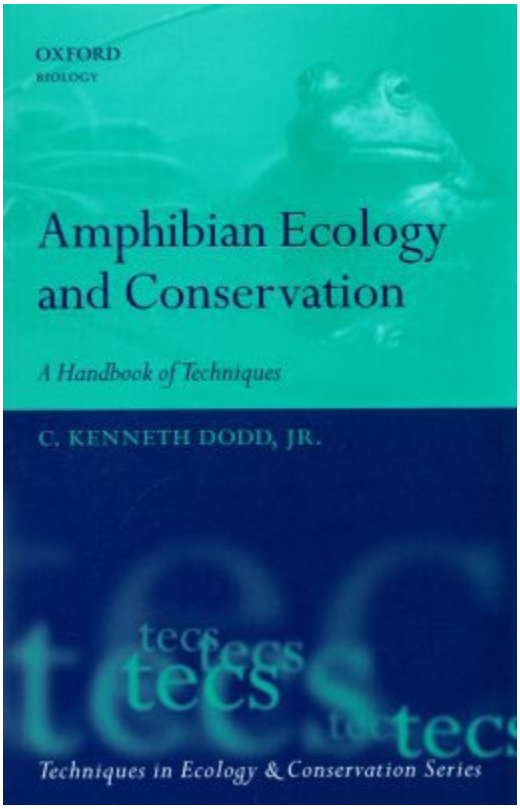
The aim of this scheme is to provide ecology and conservation books to those from outside Western Europe, North America, Japan, Australia and New Zealand who would otherwise be unable to obtain them. The simple purpose of this scheme is to spread ecological knowledge as widely as possible.

This scheme is a collaboration between the British Ecological Society (who pay for the postage), the NHBS — Everything for wildlife, science & environment (who co—ordinate and organise the distribution) and the publishers and authors of the books (who provide the books for free). We plan to provide a number of books per year. They will usually be distributed three months after the book has been published. Individuals may request the book for themselves or suggest the book for others. It is likely that some of these schemes will be oversubscribed: all applications will be considered together and the available copies will then be awarded to those considered most able to benefit from them.

The first title to be distributed under the Gratis Books Scheme was William J Sutherland's Conservation Handbook. Over 3000 copies have been donated.

We regret that gratis books are not available to students currently doing degrees in Western Europe, North America, Japan, Australia and New Zealand. They are also not available to those from Western Europe, North America, Japan, Australia and New Zealand but currently working outside those areas.

Amphibian Ecology and Conservation A handbook of techniques by Kenneth Dodd is currently available through the NHBS Gratis Books Scheme, please visit http://www.nhbs.com/Conservation/gratis-books.php for further information.



Amphibian Ecology and Conservation A handbook of techniques. Kenneth Dodd (Oxford University Press, 2009)

This practical manual of amphibian ecology and conservation brings together a distinguished, international group of amphibian researchers to provide a state—of—the—art review of the many new and exciting techniques used to study amphibians and to track their conservation status and population trends. The integration of ecology and conservation is a natural outcome of the types of questions posed by these disciplines: how amphibians can and should be sampled, marked, and followed through time; how abundance and population trends are measured; what are the robust statistical methods that can be used in ecology and conservation; what roles do amphibians play in community structure and function; how do animals function in their environment; and what affects the long-term persistence of species assemblages?

Although emphasizing field ecology, sections on physiological ecology, genetics, landscape ecology, and disease analysis are also included. The book describes the latest statistical approaches in amphibian field ecology and conservation, as well as the use of models in interpreting field research. Much of this information is scattered in the scientific literature or not readily available, and the intention is to provide an affordable, comprehensive synthesis for use by graduate students, researchers, and practising conservationists worldwide.

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Robin Moore / iLCP

WANTED ALIVE

One rainy evening in May 1989, a lone Golden toad appeared at a pool high in a Costa Rican cloud forest. He was the last Golden toad ever seen. Join the Amphibian Specialist Group in the search for this and other lost amphibians. Countries and dates indicate where and when these "top ten" were last seen alive. **Reward:** Pest control, nutrient cycling and other services provided by amphibians for people worldwide.

Golden toad

Coasta Rica 1989

Gastric brooding frog

Australia 1985

Mesopotamia beaked toad

Colombia 1914

Jackson's climbing salamander

Guatemala 1977

African painted frog

Rwanda 1950

Chile's Darwin Frog

Chile 1980

Turkestanian salamander

Turkmenistan 1909

Scarlet harlequin frog

Venezuela 1990

Dutoit's torrent frog

Kenya 1962

Yunnan Lake Newt

China 1979

Regional Updates

North and Central America and the Caribbean

Each edition of FrogLog focuses on one of the six geographical areas as outlined in FrogLog 96 (pg 6-7). This format provides regional ASG groups with an opportunity to showcase their conservation efforts and publicize issues of concern.

Caribbean

Meeting the Challenge: Getting people to value amphibians as a means to conservation action in Puerto Rico

As leaders of a conservation group, Proyecto Coquí, and Co-Chairs of the local Amphibian Specialist Group (ASG), our goal has been to try to find a balance between the time and effort devoted to both research and outreach activities. We have learned that when the general public is educated on the risks that threaten our amphibian diversity, and exposed to the uniqueness of the species that live in their backyards, and nearby habitats, they are keener to want to protect them. Children are eager to learn more about animals and their interactions with nature, and by offering family-oriented activities we have been able to get the adults motivated as well. When the population gains knowledge of nature they will value it, they will get involved in conservation initiatives, they will donate money to nature causes, and they will support petitions to legislation. In this issue of FrogLog we would like to share a few of our efforts in this direction.

Over the years we have taken advantage of diverse outreach opportunities, and encouraged our graduate students to participate. In January, the teacher of a local elementary school asked us if we could give a talk to third-graders on the importance of protecting the environment. So off we went to the grade school with our well-illustrated presentation showing the consequences of deforestation, contamination, and human-influenced global warming, on natural habitats and biodiversity. We spoke about how to help: what, how and where to recycle, ways to save energy at home, car-pooling, etc. Examples of course, were mostly of amphibians, and their global decline was discussed. At the end, every child had to draw a picture illustrating his/her compromise to make a difference.

Students visiting Puerto Rican frog photographic exhibit at our "Leaping ahead of extinction" activity.

Our graduate student, Patricia Caligari, has given talks about her research on amphibians affected by *Batrachochytrium dendrobatidis (Bd)* in Puerto Rico, and aided by the undergraduate students in our lab, offered guided nocturnal walks to see the amphibians. As a consequence, motivated youngsters have volunteered to get involved and help in her field work.

Participants of SEPARC -Caribbean Herpetofauna Conservation Symposium, from left to right (standing) Fedric Burton, Kent A. Vliet, Carlos Martinez, and Chris Jenkins; (sitting) Rafael Joglar, Patricia Burrowes and Jenifer Stabile.

Last February 29, Proyecto Coqui joined Amphibian Ark's worldwide initiative, and celebrated extant Puerto Rican frogs with a special "Leaping ahead of extinction" activity at the University of Puerto Rico in San Juan. The University community and the general public were invited to a half day venture that started at noon with a photographic exhibition of the local species by R. L. Joglar, and live frog exhibits entertained by graduate and undergraduate students at the Joglar and Burrowes herpetology lab. At 6:00 pm P. A. Burrowes offered a talk on "The coquis of Puerto Rico: marvels, curiosities and why we should protect them" that was attended by over 100 people many of which were kids with their parents. After the talk, those that were interested stayed along for guided tours around the university campus to look for coquis. We had three groups of approximately 20 people

each, that had the opportunity to learn to distinguish visually and acoustically four of our endemic species. The grand prize of the night was the encounter of a male *Eleutherodactylus coqui* guarding an egg clutch! One of the keys to the success of this event was that we had excellent media coverage through radio and TV, not only to invite the public to attend, but to help us disseminate the message of the importance of conserving the amphibians that have "leaped ahead of extinction" in spite of all the many threats they face.

Another important part of outreach is getting involved, as experts, in local projects that will have detrimental consequences to the environment. In this respect, we have devoted many hours attending governmental forums and public hearings to inform the public about how building the proposed natural gas pipeline across 92 miles of Puerto Rican landscape, will be harsh on amphibians and other biodiversity.

Breaching to international efforts is also valuable. Last February, we participated in the South-Eastern Partners in Amphibian and Reptile Conservation (SEPARC) meeting in Tennessee where we gave two talks about the risks threatening Puerto Rican amphibians, with emphasis in disease *(Bd)* and the expansion of exotic herps. SEPARC is interested in reaching out to the Caribbean in their efforts to protect amphibians and reptiles and thus, sponsored a Caribbean symposium that culminated in a discussion between participants, to consider ways in which this could be done more effectively.

We acknowledge that juggling between duties in our academic and/or administrative posts and trying to do these types of activities can be difficult. However, it is our responsibility as an educated community to share this knowledge with others, in order to give them the opportunity to make educated decisions and truly get involved in the ever most challenge of protecting this group of animals that is confronting a growing diversity of threats in this century.

Patricia A. Burrowes and Rafael L. Joglar (Co-Chairs) Caribbean Amphibian Specialist Group.

USA

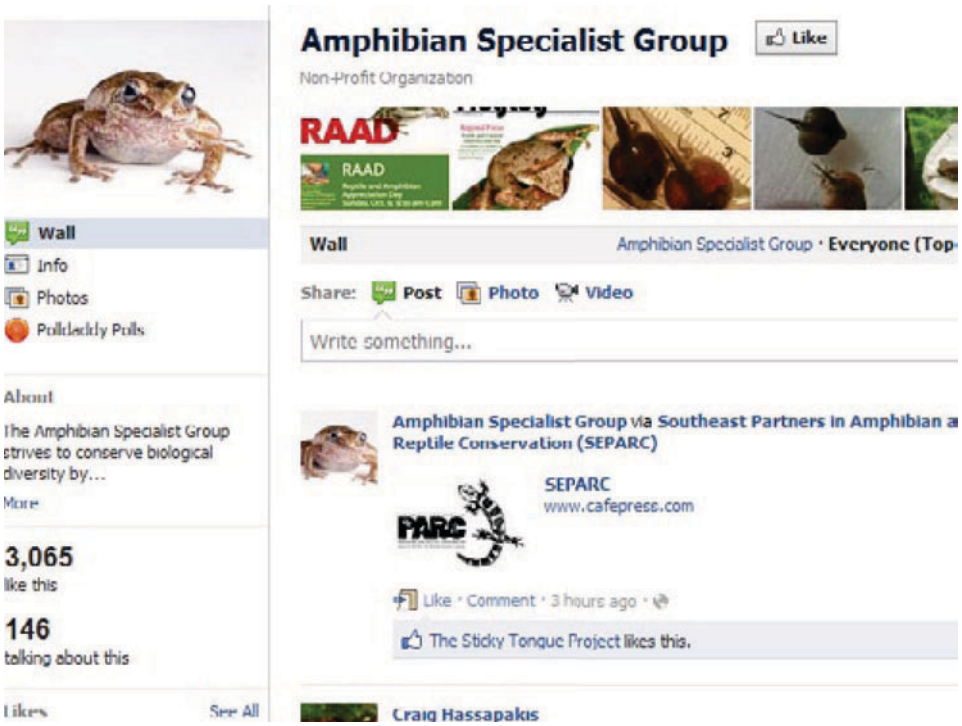
2012 North American Amphibian Ark Assessment

From March 5th through March 8th, 2012, the North American Amphibian Ark Assessment Team, led by Richard Gibson assisted by Diane Barber and Paul Crump of the AZA Amphibian Taxon Advisory Group, met at the Fort Worth Zoo. The team considered each of the 285 currently recognized North American Amphibian species with respect to factors such as biological distinctiveness, extinction risk, available protected habitat, threat mitigation, scientific importance, cultural/economic importance, education potential, and conservation mandate. The team identified the following species as candidates for in-situ research programs to aid in answering conservation-related issues:

Ambystoma californiense
Ambystoma cingulatum
Amphiuma means
Amphiuma pholeter
Amphiuma tridactylum
Anaxyrus baxteri
Anaxyrus houstonensis
Batrachoseps major
Cryptobranchus alleganiensis
Desmognathus abditus
Eurycea chisholmensis
Eurycea neotenes
Eurycea sosorum
Eurycea tonkawae
Eurycea wallacei
Gyrinophilus porphyriticus
Hydromates platycephalus
Lithobates chiricahuensis
Lithobates hecksheri
Lithobates sevosus
Phaeognathus hubrichti
Plethodon ainsworthi
Plethodon amplus
Plethodon cheoah
Plethodon jordani
Plethodon petraeus
Plethodon shenandoah
Rana draytonii
Rana muscosa
Ranna sierrae
Rana pretiosa
Urspelerpes brucei

In addition, we recommended changes to the IUCN Red List status of 21 species.

Michael J. Lannoo (Co-Chair) USA Amphibian Specialist Group.



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Conservation Status and Ecological Notes of the Previously Extinct Toad *Incilius holdridgei* (Taylor, 1952), Costa Rica

By Juan G. Abarca

Holdridge's toad *Incilius holdridgei* (*1*) was described based on a single individual collected on the eastern slope of Volcán Barva, Costa Rica by the eminent naturalist Leslie Holdridge, to which the species was dedicated (*2*). Adults are dark brown with intense orange colors on the back, limbs, and head during the breeding season; the young are brown or black (Fig. 1). In the 1970's herpetologist Douglas Robinson made a series of reproductive and ecological observations on this species (*3*). Unfortunately, the population declined and the last living specimen was observed in 1985, and in 2007 it was declared extinct (*4*). *I. holdridgei* remained unseen until Abarca *et al.*, (2010) reported on their appearance after a 25 year absence. Since then, very few individuals have been observed. In the following article I show the locations, condition, and ecology notes of this rare species.

Fig. 1: Subadult of *Incilius holdridgei*. Photo: J. Abarca.

Methods

Field observations were made in Alto del Roble, San Rafael, Heredia Province, Costa Rica, between May 1, 2008 and July 29, 2011 for a total of 40 visits in four years.

One and two day visits were made, which included both day and night sampling. The toads were found using Visual Encounter Survey (VES) within suitable habitat. Measurements, weight, activity, time of day, location, description of habitat, weather conditions, and proximity to other individuals were documented in situ. The maturity of the individual was based on the data provided by Novak and Robinson (1975): juveniles from 7 mm to 30 mm, subadults between 30 mm and 40 mm, and older adults over 40 mm. A group of tadpoles were collected from several temporary ponds to verify the identity of the species, which were later confirmed to belong to *Incilius holdridgei* and *Isthmohyla pseudopuma*.

Natural history

Incilius holdridgei is only known from the Central Volcanic Cordillera of Costa Rica. In the Alto del Roble, above 2000 m on the southern slope of Volcán Barva, three localities are known for this species. These areas are palustrine clearings where the soil retains much water. Characteristic vegetation is rush (*Juncacea*), ferns (*Alsophila*), bamboo (*Chusquea*), club mosses (*Sphagnum*), and associated wetland vegetation. Some of these gaps are lower strata forest where cover and shade are sparse, with species such as Copey (*Clusia sp.*) and Aguacatillo (*Ocotea sp.*) providing plenty of leaf litter around these openings. Open areas appear to benefit this species, especially for adults during the reproduction period, while juveniles need leaf litter and forest areas for food and protection (Fig. 2).

Fig. 2: Habitat of *Incilius holdridgei*. Photo: J. Abarca.

Adults are fossorial, surfacing only to breed, subadults are semifosso-

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Fig. 3: Eggs of *Incilius holdridgei.* Photo: J. Abarca.

Fig. 5: Breeding ponds of *Incilius holdridgei.* Photo: J. Abarca.

Fig. 6: Reproductive site of *Incilius holdridgei.* Photo: J. Abarca.

Fig. 4: *Incilius holdridgei* with eggs. Photo: J. Abarca.

rial, and juveniles move more actively on leaf litter and low vegetation. While mainly terrestrial, *I. holdridgei has* a great facility for climbing and many were seen moving on vertically inclined moss on tree trunks, roots, branches, and low vegetation over 30 cm from the soil. The toads' activity is both diurnal and crepuscular. Their movements are slow, and like many other highland toads, *I. holdridgei* prefer walking instead of jumping. This lethargic form of locomotion along with their ability to blend in with the environment makes the juveniles almost imperceptible under the tangle of grass and tall reeds.

Eggs are laid in water in long free floating rows (Fig. 3); a clutch may have 40 to 150 eggs. Although Robinson described the eggs in his 1975 account, no photographs were available before the present article. A description of the eggs is as follows: The relatively large eggs (2.76 mm in diameter) are black (animal pole) and cream (vegetal pole), and surrounded by a gelatinous mass with a cylindrical diameter of 5 mm; this mass effectively protects the eggs against desiccation. Although reports from 1975 described aggregations of hundreds of individuals, at present only two reproductive events have been observed: the presence of tadpoles in June 2010 and a mass of eggs and six adults at the same location in May 2011 (Fig. 4). Amplexus was not observed during either event. The oviposition site was a cavity 15 cm deep and 50 cm in diameter, located in a clear area under ferns and grasses at one of the sites (Fig. 5 & 6). The oviposition site was surrounded by small pools of water less than 5 cm deep, where eggs and tadpoles from *Isthmohyla pseudopuma* were collected. The average water temperature was 15 ° C.

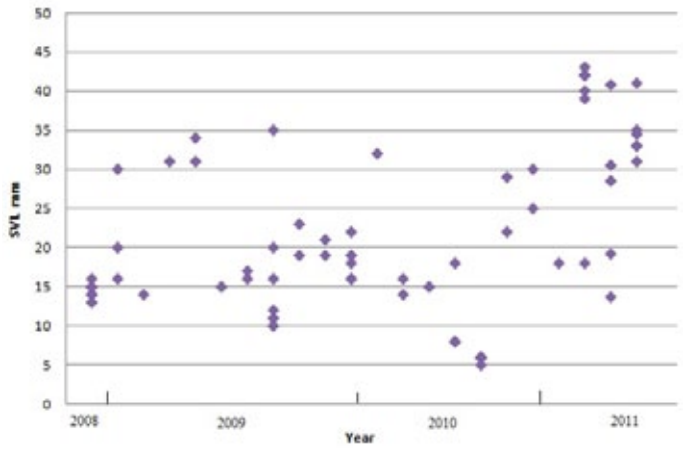

Fig. 7: Observation between 2008 and 2011.

Fig. 8: Breeding coloration of *Incilius holdridgei*. Photo: J. Abarca.

Conservation Status

Of the 40 visits, individuals were observed on 22 occasions, accounting for a total of 67 individual observations over four years of sampling (Fig. 7). There have been 45 juveniles weighing between 0.3 g and 2 g. Juveniles were observed 19 times under clear and sunny conditions and 27 times under cloud cover and rain. Fourteen subadults have been observed with an average weight of 4.5 g and under primarily cloudy, and rainy conditions. Eight adults in aggregation were observed in the same pond, under overcast conditions after a week of heavy rain (Fig. 8). Most of the adults and juveniles have been observed during the months of May through August, with the majority in May; this is consistent with the data presented by Novak and Robinson (1975).

Incilius holdridgei is still listed by the IUCN Red List as Extinct (*6*), however is currently being reassessed. I have calculated the potential extension of their presently known range, based on suitable habitat, by only 2100 km^2. The finding of eggs, tadpoles, and individuals of all age classes indicate constant reproductive activity. Although one might think that the population of *I. holdridgei* is recovering, information on the only known populations is still very limited, showing an imperative that further studies be performed to determine the population dynamics, environmental factors of the breeding sites, population health and genetics. While these studies are currently being conducted by several researchers, the acquisition of additional funding is crucial for the continued conservation of this vulnerable species.

An adult Holdridge's toad *Incilius holdridgei*. Photo: J. Abarca.

Acknowledgments

Gilbert Alvarado, Víctor Acosta, Heinz Hoffman and Don Filipiak.

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Assessment of Risk of Local Extinction, a Fast - Acting Method in Mexico

By Edgar Toribio-Hernández

During the 1960s it was globally acknowledgment that many amphibian populations were in decline. The following decades led to numerous long - term studies in diverse parts of the world producing supporting evidence of these population declines, and in some cases, species disappearance altogether. The first World Congress of Herpetology, held in 1989, brought together much of this knowledge to confirm the phenomenon on a global scale. The 1990s saw several international institutions focus their efforts on studying the causes of these declines with a focus towards Europe, the United States, and equatorial Neotropics (*1-4*).

In Mexico, the study of this phenomenon is still in its infancy. There are only a limited number of long - term monitoring programs in place and still many gaps in the understanding of the basic ecology of some species, consequently few studies have demonstrate regional declines directly (*5*). The amphibian population declines that have been reported in countries to the north and south of Mexico reflect the urgent need to develop short and efficient population trend evaluations techniques. A strategy currently being proposed for use in Mexico, utilizes a method to assess the risk of local extinction using previous knowledge of the historical populations (*6-9*).

Based on studies conducted between 2000 – 2011 there is evidence of amphibian population declines in nine of the 32 states in Mexico (Fig. 1). An example of one such study is presented here. Between 2006 - 2007, historically known populations of amphibians were monitored and results compared with records collected between 1900 and 1995 in one of the central states of Mexico. The study area has undergone significant anthropogenic change over the years and

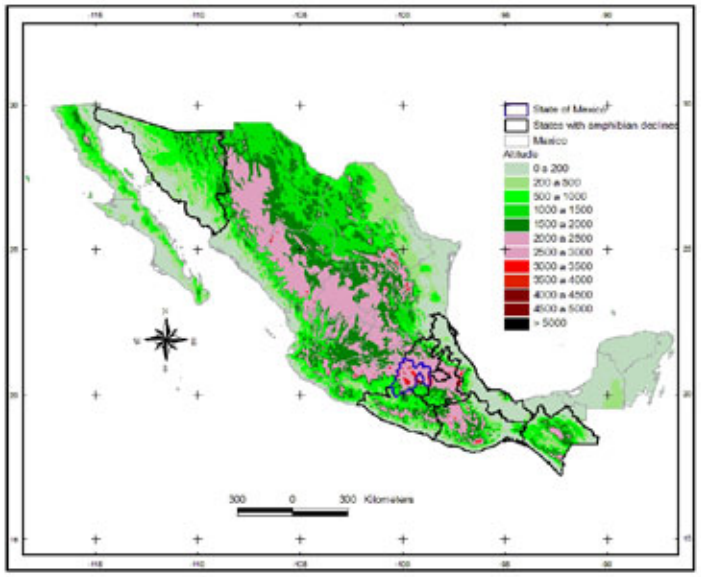

Fig. 1: The States of Mexico where the declination of amphibians has been demonstrated.

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Fig. 2: A pond of the studied area surrounded with cultures.

Fig. 3: Part of the studied area with wide skylights inside the vegetation.

Fig. 4: Cleared vegetation in the studied area.

as a consequence has been subject to soil and water contamination and significant levels of forestry and logging (Fig. 2-4). The study aimed to establish the population status of all detected amphibian species, assess the level pollution in locations with historical records and identify any correlation between these two factors. Furthermore information was collected on local precipitation and temperature and, the regional conservation status of each species. The regional conservation status provides an indication of the risk of extinction of wild species in Mexico (MER, for its acronym in Spanish) (*10*) using a method based on the IUCN Red List Categories and Criteria.

The results of the study identified nine species with highly fluctuating populations during the 2006 (year with abnormal drought) and 2007 (with abundant rains) study periods. According to local meteorological data recorded between 1970 - 2007, precipitation levels during 2006 and 2007 were extremes. These data allows deducing the reliable limits of the population tendencies of each species. During both years there was a negative correlation between pollution levels and species abundance. The regional conservation status of five species were: Endangered (*Hyla smithii, Hyla plicata*, *Syrrhophus nitidus*, *Ambytoma bombypellum*), Critically Endangered (*Pseudoeurycea leprosa*; Fig. 5-6), and four as Extinct in the wild. Further information on the regional conservation status assessment process can be supplied by the authors at written request.

This rapid method, to evaluate the risk of local extinction in order to prepare immediate plans for conservation action, has been widely used in first world countries. The economic challenges faced in Mexico, and many other parts of the world, mean undertaking long – term national monitoring program is currently unlikely. The rapid assessment method discussed here, which relies on a combination of historic data, knowledge of species biology and ecology (*9*, *10*), and current population data, is likely to prove a far more viable and efficient assessment strategy. Following a precautionary approach to conservation action, this method is already contributing to avoiding further populations declined without the need for long - term monitoring (*11*). At present, local and national governments in Mexico are using this method to develop conservation action plans.

Fig. 5: Adult of *Pseudoeurycea leprosa*.

Fig. 6: Adult (up) and juvenile (down) of *Pseudoeurycea leprosa*.

Acknowledgments

Autonomous University of the State of Mexico, IUCN, Idea Wild, RANA, and James Lewis for his comments.

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PANAMA


E.V.A.C.C.
EL VALLE AMPHIBIAN CONSERVATION CENTER
AMPHIBIAN RESCUE & CONSERVATION PROJECT
PANAMA

Amphibian Rescue and Conservation Project - Panama

By Brian Gratwicke, Paul Crump, Eric Baitchman, Matt Evans, Della Garelle, Cindy Hoffmann. Roberto Ibáñez, Lindsay Renick Mayer & Heidi Ross

Who we are

The mission of the Panama Amphibian Rescue and Conservation project is to rescue and establish assurance colonies of amphibian species that are in extreme danger of extinction throughout Panama. We also focus our efforts and expertise on developing methods to reduce the impact of the invasive fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*) so that captive amphibians may eventually be re-introduced to the wild.

History

In 1999, Project Golden Frog (PGF- www.ranadorada.org) was formed to address the inevitable decline and possible extinction of the Panamanian golden frog (*Atelopus zeteki*) in western-central Panama. Through research (*1*) and *ex situ* conservation efforts (*2*), a thriving cooperatively managed population of golden frogs was established in institutions of the Association of Zoos and Aquariums (AZA). One of the major goals of PGF was to establish an in-country conservation center for keeping and breeding golden frogs, amongst other species, to offset the illegal collection for the pet trade. In 2004, building on the infrastructure and contacts of PGF, and spurred to action by the declines in western Panama (*3*) the Amphibian Recovery and Conservation Coalition (ARCC) was launched. ARCC attempted an assemblage-level project focusing on the feasibility of reactive and proactive rescue approaches. ARCC noted again that it was necessary to export amphibians to the USA for conservation purposes, as no facilities existed in Panama for *ex-situ* amphibian conservation (*4*). Thus in 2005, the Houston Zoo and partners established the El Valle Amphibian Conservation Center (EVACC) to house endangered amphibians that were declining rapidly as a result of *Bd* in western Panama (*3, 4*). By 2008, which was Year of the Frog, *Bd* had spread east of the Panama Canal (*5*) and conservationists agreed that additional capacity was needed to establish assurance populations of amphibians at risk in eastern Panama. Amphibian Ark, along with the American AZA, helped us persuade new partners to pledge funding, and expertise to build a second Panamanian facility, in order to establish *ex-situ* colonies of amphibians from eastern Panama. The Panama Amphibian Rescue and Conservation Project was officially launched in 2009, and now both in-country facilities fall under the umbrella of this project, Roberto Ibáñez is the in-country project director and Brian Gratwicke is the international coordinator. The project's partners include Africam Safari, Autoridad Nacional del Ambiente (ANAM), Cheyenne Mountain Zoo, Defenders of Wildlife, Houston Zoo, Parque Municipal Summit, Smithsonian Tropical Research Institute (STRI), Smithsonian Conservation Biology Institute and Zoo New England.

Capacity Building

Before the official launch of the Panama Amphibian Rescue and Conservation project in 2008, a workshop was hosted at STRI to look at all 200 known species from Panama and determine the 20 priority species for rescue based on methods developed by the Amphibian Ark (www.amphibianark.org). *Atelopus* species as a group stood out as the most highly ranked taxa in need of *ex-situ* assurance populations, as they have declined across much of their range due to chytridiomycosis (6). Amphibian Ark also conducted an important workshop on husbandry at the beginning of the project, and has continued providing advice and technical assistance as part of their wider global capacity-building strategy. Other key capacity-building activities included training of molecular technicians leading to the establishment of the first qPCR *Bd* diagnostics lab in Panama, and a workshop on isolation, culture and cryopreservation of *Bd* that established the first live *Bd* cultures in Panama. These living cultures and diagnostic capabilities will be a vital resource for future *Bd* research in Panama. We have also established colonies of domestic crickets, fruit flies, springtails, mealworms, superworms, earthworms and cockroaches. This is critical because keeping healthy frogs is only possible if they can be fed. We now have a full-time technician at our facility in central Panama focused on rearing a diverse array of insect food items to feed the frogs, and we intend to hire a full-time equivalent at EVACC. Some food items, such as katydids, have proven difficult to rear and children in El Valle still earn pocket money collecting them from the wild. Many of the priority amphibian species have never been maintained successfully in captivity before, and understanding nutritional needs at different life stages is an obvious aspect that would require attention and some research in the future.

Fifty species are housed in this building and have four people managing them on a day to day basis. Volunteers often assist, preference is for spanish speakers who can devote substantial time to assisting as it often takes a long time just to train volunteers. Photo: Panama Amphibian Rescue and Conservation Project.

Dr. Eric Baitchman from Zoo New England oversees the veterinary program with support from Dr. Della Garelle from Cheyenne Mountain Zoo. They oversee protocols and provide ongoing training to our staff so that they can monitor signs of animal health, screen fecal samples for parasites and administer routine treatments for common conditions. Eric and Della are available to staff by phone and e-mail for consultation and they have developed close working relationships with all project staff. All animal morbidities and mortalities are recorded in an animal management database. Preserved carcasses are prepared for shipment to Allan Pessier from San Diego Zoo, who has generously performed histopathology on priority cases and is providing feedback.

EVACC

El Valle Amphibian Conservation Center (EVACC) is located in El Valle, a town inside a dormant volcanic crater in the Central Province of Coclé, on the grounds of the Níspero Zoo. The amphibian rescues at EVACC began in early 2006, in *Bd* infected streams and forests of the surrounding area. The initial rescues occurred in the surrounding areas of the crater, and as the disease continued to wipe out populations, the rescue was extended to the eastern part of the country to ensure viable numbers for population management in captivity. El Valle is a tourist destination in Panama, and to share Panama's rich diversity with the many national and international visitors, EVACC has a public exhibition area inside the zoo. EVACC receives no money from gate fees, and does not charge any additional fees to enter the frog hall. Fifteen priority species were rescued, including the Panamanian Golden Frog, *Atelopus zeteki*. Of those priority species ten have been reproduced in captivity with varying success rates (e.g., see Gagliardo *et al.*, 2010).

Gamboa

Due to the success of EVACC, and the lack of space in the facility, the Panama Amphibian Rescue and Conservation Project was formed and a second facility established in central Panamá at the Summit Municipal Park in 2009. This consists of three outfitted 400sq ft

Atelopus certus pair in amplexus - we hope they'll be laying eggs soon at the Summit Municipal Park. Photo: Panama Amphibian Rescue and Conservation Project.

This paper was written by the implementation team of the Panama Amphibian Rescue and Conservation Project www.amphibianrescue.org. Corresponding author Brian Gratwicke, Smithsonian Conservation Biology Institute, gratwickeb@si.edu

Angie Estrada (left) - is Volunteer Coordinator and helps to run and manage the ex-situ collection of the Panama Amphibian Rescue and Conservation Project. Jorge Guerrell (right) - conservation technician performing parasite load analysis on fecal samples. Photo: Panama Amphibian Rescue and Conservation Project.

shipping containers and is staffed by four amphibian conservation technicians. Unfortunately, unreliable electric supply have plagued us at this location and we are in the process moving the facility to a new site based in Gamboa that will have more reliable electricity, a back-up power generator and wastewater treatment facilities. The new location is in close proximity to scientific infrastructure at the STRI and this should facilitate further scientific collaborations. We have secured the donation of three additional shipping containers from Maersk Line and will outfit them to house amphibians and insectariums to produce food for the living collection. We are actively fundraising to construct an adjacent building that will provide a laboratory, quarantine, storage and office space and maximize the scientific opportunities of having a valuable living collection of animals. This facility currently houses founding populations of *Atelopus limosus, Atelopus certus* and *Atelopus glyphus* which have all been successfully bred in captivity and we feel confident that these species will be secure in *ex-situ* populations for the immediate future. We also house two undescribed species at risk of extinction and *Hyloscirtus colymba,* but to date we have not been successful at raising offspring from these species to adulthood.

The first-ever captive-bred *Atelopus limosus,* lowland color form, juvenile. Photo: Panama Amphibian Rescue and Conservation Project.

Population management and assisted reproduction

With the commitment to take on an *ex-situ* population of a species comes responsibility to manage the population in a way that will preserve the genetic integrity of the species over the longer term. We recently hosted the first of what we hope will be a series of workshops to train conservation technicians on amphibian population management guidelines (*8*). We now maintain animal records in a studbook and analyze population data and optimal pairings using SPARKS and PopLink management software. In addition, we are developing assisted reproduction methods for *Atelopus*. These tools may one day allow us to cryobank gametes of valuable founding animals and provide further insurance to help maintain the genetic diversity of these valuable animals well into the future.

Exploring methods to mitigate chytridiomycosis

One of the main scientific goals of this project is to develop tools mitigate the threat of chytridiomycosis, aiming to reestablish wild populations of amphibians from *ex-situ* colonies in *Bd* -positive areas. We currently have few viable tools to manage chytridiomycosis in a wild situation (*9*), but recent experiments successfully demonstrated that some skin bacteria produce anti-*Bd* metabolites and offer anti-fungal protection to their amphibian hosts (*10,11*). We conducted trials using *Janthinobacterium lividium* as a potential probiotic agent on Panamanian golden frogs provided through the golden frog species Survival Plan. Unfortunately, *J. lividium* did not grow well on golden frog skin and it did not protect the frogs from chytridiomycosis (*12*). We are now conducting laboratory trials using four anti-*Bd* bacteria candidates isolated from Panamanian toad species to determine if any of them are effective at protecting golden frogs from *Bd*. Furthermore, our collaborators, led by Lisa Belden from Virgina Tech, have secured an NSF "dimensions of diversity" grant to investigate how skin microbe diversity may affect disease outcomes in Panamanian amphibians over the next four years. The recent discovery of genes linked to *Bd* resistance in *Lithobates* species (*13*) is another promising direction that we intend to explore in coming years.

Education and outreach

Our primary education and outreach tool has been our website http://amphibianrescue.org. We have a Spanish and English version and receive about 50,000 unique visitors per year. Our communications committee has developed an award-winning cam-

Kids learning about Panamanian golden frog conservation at the Smithsonian National Zoo's Reptile Discovery Center Exhibit. Photo: Panama Amphibian Rescue and Conservation Project.

paign that has resulted in about 50 unique news stories about our project each year, a blog that is updated twice weekly and a feature-length documentary produced by the Smithsonian Channel entitled "Mission Critical: Amphibian Rescue." Our social media strategy has attracted a highly engaged audience of about 5,000 Facebook fans and 1,500 Twitter (@amphibianrescue) followers and this network of supporters has been a valuable resource for fundraising and recruiting volunteers. The exhibit at EVACC has made the El Níspero Zoo a real tourist destination, drawing more than 100,000 Panamanian and international visitors each year. In addition, we have two major amphibian conservation exhibits at the Smithsonian's National Zoo highlighting this project and the importance of zoos in conservation to 2 million annual visitors.

Perhaps one of the most exciting developments in Panama has been legislation passed in 2010 declaring August 14th National Golden Frog Day. Our first official celebrations last year included a parade in El Valle, a special activity day at the Summit Zoo and art competitions, plays and lectures. This was well received and has huge potential to engage Panamanians and get them excited about their incredible amphibian biodiversity heritage. In addition to this, in 2009, Panama's environmental authority -ANAM- issued the legal resolution No. AG 0467-2009 urging the drafting and implementation of a national action plan for the conservation of the amphibians of Panama. ANAM recently launched a country-wide amphibian conservation action plan that identifies various needs for *in-situ* and *ex-situ* conservation that will help us to mobilize further resources to build capacity to conserve Panama's amphibians as part of a larger, coordinated national effort (*14*).

Acknowledgements

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Water fall at Sierra Llorona, a *Bd* monitoring site in central Panama. Photo: Panama Amphibian Rescue and Conservation Project.

Protecting Rare Amphibians Under the U.S. Endangered Species Act

By Collette L. Adkins Giese

Frogs, salamanders and other amphibians are some of the most rapidly disappearing species on Earth. Every day, species here in the United States are beset by habitat destruction, pollution, toxins, climate change and other factors that drive extinction.

Surprisingly, though, just 23 of the nearly 1,400 species protected under the U.S. Endangered Species Act are amphibians. That's partly because they've been woefully underrepresented when it comes to wildlife protection efforts by environmental organizations in the United States. The Center for Biological Diversity aims to change that.

Almost since its inception two decades ago, the Center has worked to protect endangered amphibians. By petitioning the U.S. Fish and Wildlife Service to provide Endangered Species Act protection for imperiled amphibians – and following up with lawsuits when necessary – the Center is working to obtain federal safeguards and protected habitat for frogs, toads and salamanders across the country. And after hiring the nation's first full-time attorney dedicated to conserving amphibians and reptiles, the Center is expanding its work on behalf of rare amphibians. But there isn't much time to lose.

A Powerful Tool

In the United States, 56 species of amphibians (more than 20 percent of those evaluated) are endangered or vulnerable to extinction, according to the 2011 Red List of the International Union for Conservation of Nature (IUCN). Moreover, scientists lack sufficient information to assess the status of an additional five percent of the nation's amphibians (*1*).

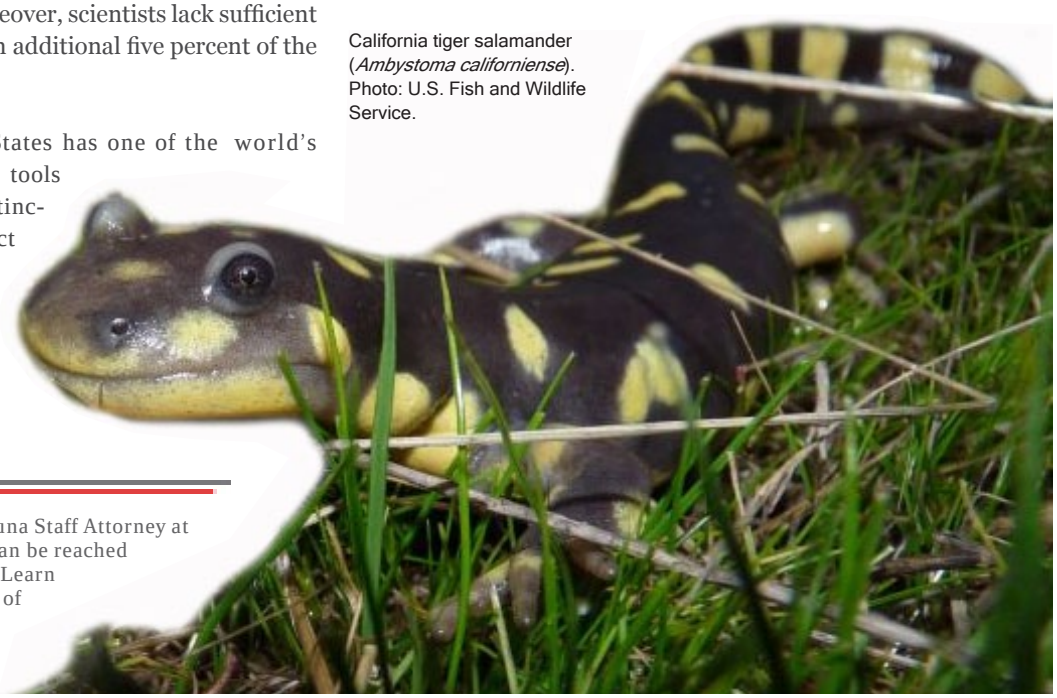
The good news is that the United States has one of the world's most powerful and successful legal tools for protecting species at risk of extinction: the Endangered Species Act (ESA). Passed by Congress in 1973, the ESA is the best and possibly the last chance Americans have to secure a future for diverse native wildlife and the natural environments they depend on.

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

The ESA is, by any measure, a success: 99.9 percent of species protected by the Act have been kept from extinction and, where measured, 93 percent of protected species are stable or moving toward recovery (*2, 3*). The longer a species is listed under the Act, the more likely it is to be recovering (*4*).

More Protections Needed

Currently, just six frogs, four toads and 13 salamanders are protected in the United States under the ESA. Yet dozens more urgently need federal protection but do not currently receive it, such as the Foothill yellow-legged frog (*Rana boylii*), Black toad (*Anaxyrus exsul*), and the Inyo mountain salamander (*Batrachoseps campi*). In fact, 41 species lack protection under the ESA even though they are considered endangered or vulnerable to extinction by the IUCN. The Center (which has helped secure ESA protections for more than 500 plants and animals over the last 20 years) is working to secure ESA protection for these amphibians through petitions, lawsuits and negotiations.



California tiger salamander (*Ambystoma californiense*). Photo: U.S. Fish and Wildlife Service.

Collette L. Adkins Giese is the Herpetofauna Staff Attorney at the Center for Biological Diversity. She can be reached at cadkinsgiese@biologicaldiversity.org. Learn more about the Center's efforts on behalf of imperiled amphibians at http://www.biologicaldiversity.org/campaigns/amphibian_conservation/index.html.

Last year, the Center struck a historic legal settlement with the U.S. Fish and Wildlife Service requiring the agency to decide whether to add 757 imperiled plants and animals – including more than 30 amphibian species — to the endangered species list by 2017. The settlement caps a decade-long effort by the Center's scientists, attorneys and activists to secure federal help for some of the country's least protected, but most imperiled, species.

The agreement has already yielded important results. The Service recently found that protection of 374 freshwater species in 12 southeastern states — including 13 species of amphibians — may be warranted under the ESA (The Center and other groups petitioned to protect those species in 2010). As a result, the Neuse river waterdog (*Necturus lewisi*), Gulf hammock dwarf siren (*Pseudobranchus striatus lustricolus*), One-toed amphiuma (*Amphiuma pholeter*) and 10 other salamander species will move closer to ESA protection. The Center's petition to list a population of the Boreal toad (*Anaxyrus boreas boreas*) — whose numbers plummeted from disease and habitat destruction — received a positive initial finding from the Service in April with a final listing decision expected next year.

Juvenile Boreal toad (*Anaxyrus boreas boreas*). Photo: J.N. Stuart.

Looking forward, the Center plans to file a large-scale petition this year seeking ESA protection for nearly 50 species of amphibians and reptiles across the United States. The status of these species was initially evaluated using information from NatureServe Explorer, the IUCN Red List, AmphibiaWeb and scientific journals. The Center is now seeking additional guidance from the scientific community on species that should be included in this listing petition.

Saving Habitat

Beyond including additional amphibians on the list of endangered and threatened species, the Center also seeks to reduce threats to those species already on the list. Habitat destruction is a primary threat to endangered amphibians, and as such, protection of critical habitat is literally critical. In fact, a study by the Center found that species with this federally protected habitat are more than twice as likely to be moving toward recovery than species without it (*4*). Strictly defined, critical habitat includes specific areas within a species' current range that have "physical or biological features essential to the conservation of the species," as well as areas outside the species' current range upon a determination "that such areas are essential for the conservation of the species." 16 U.S.C. § 1532(5)(A).

Critical habitat provides key protections for listed species by prohibiting federal agencies from permitting, funding or carrying out actions that "adversely modify" designated areas. Designating critical habitat also provides vital information to local governments and citizens about where important habitat for endangered species is located — and why they should help conserve it.

The Center works to ensure that the U.S. Fish and Wildlife Service designates critical habitat for listed species. For example, after the agency refused to designate any critical habitat for the Sonoma County population of the California tiger salamander (*Ambystoma californiense*), the Center brought a lawsuit arguing that this decision was based on undue political influence rather than the best available science, as the ESA requires. These efforts paid off last year when the agency finally designated more than 47,000 acres of critical habitat for the Sonoma County salamanders.

The Center also works to prevent destruction of essential amphibian habitats, such as the last viable breeding pond for the highly endangered Mississippi gopher frog (*Rana capito sevosa*). A private developer in Mississippi is building a town called "Tradition" with as many as 35,000 people on land near the pond. In response to the threat of litigation under the ESA, the developer began discussing with the Center how the development project could proceed while still protecting the frog. Since then, the Center and its local partner negotiated a memorandum of understanding with the developer that outlines steps the parties will take to facilitate a proposed land exchange between the developer and the U.S. Forest Service that would keep development away from the essential habitat.

The Center's campaign against stocking of nonnative fish provides another example of its efforts to protect amphibian habitats. Nonnative trout stocking is causing amphibian declines in California and throughout the western United States, with introduced fish preying upon amphibians like the mountain Yellow-legged frog (*Rana muscosa*), Yosemite toad (*Anaxyrus canorus*), and Arroyo toad

(*Bufo californicus*). Litigation brought by the Center against California Department of Fish and Game forced the agency to consider the impacts of trout stocking on amphibians, which resulted in many lakes being made off limits to nonnative fish stocking.

Mississippi gopher frog (*Rana capito sevosa*). Photo: U.S. Fish and Wildlife Service.

Protection from Pesticides

Pesticides pose another significant threat to endangered amphibians, which are particularly sensitive to pesticides and other toxins because of their permeable skin. To address this threat to amphibians and other wildlife, the Center has brought a series of lawsuits against the U.S. Environmental Protection Agency (EPA), which is tasked with ensuring that pesticide registrations do not pose unreasonable adverse effects on the environment. Unfortunately, the EPA registers most pesticides without fully analyzing the impacts on endangered and threatened species.

In 2011, the Center filed a legal complaint challenging the EPA's failure to consult with the U.S. Fish and Wildlife Service on pesticides' harmful effects on endangered and threatened species, including 16 amphibian species. This is a landmark case — the largest of its kind — and it seeks to protect more than 200 species from the harmful effects of approximately 400 pesticides. Last year the Center also filed a complaint against the U.S. Fish and Wildlife Service and the EPA for failing to study and act on threats posed by pesticides to the threatened California red-legged frog (*Rana draytonii*).

Atrazine is a pesticide with particularly harmful effects on amphibians. Atrazine is an endocrine disruptor that chemically castrates and feminizes male frogs – even when it's used at levels lower than those currently allowed in drinking water by the EPA (5). Its danger to humans and wildlife is so serious that it was banned in the European Union in 2004. Yet it's the most commonly used herbicide in the entire United States. The Center and its allies have been pushing the EPA to ban atrazine in the United States. The Center also supports legislative efforts to ban atrazine; a bill was introduced earlier this year.

More Work Ahead

Efforts by the Center for Biological Diversity and other organizations are urgently needed because the United States is experiencing amphibian declines symptomatic of the global amphibian extinction crisis. Ubiquitous toxins, global warming, nonnative predators, over collection, habitat destruction and disease are key factors leading to the demise of amphibians in the United States and across the globe.

The Center uses biological data, legal expertise and the citizen petition provision of the powerful Endangered Species Act to obtain legally binding protections for rare amphibians and other wildlife across the country. But stemming the amphibian extinction crisis means attacking it on every front. The nation needs conservation efforts as diverse as the animals we're working to protect.

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Conservation of the Florida Bog Frog, One of North America's Rarest Amphibians

By Thomas A. Gorman[1], James A. Austin[2] & Carola A. Haas[1]

The Florida bog frog (*Lithobates okaloosae*) is a narrow endemic restricted to an estimated 25 streams that span three counties in northwestern Florida. The species occurs in a unique riparian habitat that is associated with acidic streams and that originates from seepage and steephead habitats. The restricted geographic range is a concern for the long-term conservation of the Florida bog frog (*1*). Thus, Eglin Air Force Base has been a critical partner in this conservation effort, as >90% of all known breeding sites occur within its borders.

Throughout its range, Florida bog frog occur syntopically with a more common congener, Bronze frog (*L. clamitans*; see Figure 1). In contrast to the bog frog, the Bronze frog is widely distributed and occurs throughout most of eastern North America. The Bronze frog is closely related to the Florida bog frog (*2*) and has been hypothesized to be a potential interspecific competitor (*3, 4*). In addition to possible competition, putative hybrids had been identified anecdotally based on phenotypic characteristics (*5*).

Over the past 8 years, our research has focused on elucidating the interspecific interactions of these two species. We have examined microhabitat use and space partitioning, tadpole behavior, and patterns of potential hybridization between these two species. We have cooperated with Eglin AFB land managers who can use information from our work to actively inform their management decisions.

To understand how male Florida bog frog and Bronze frog use space within a breeding area, we monitored movements and spatial interactions to quantify intra- and inter-specific interactions. Our results suggested that both species were distributing themselves in the same areas of the wetland (in a clumped distribution) and that they did not appear to be influenced by the presence of other conspecifics or heterospecifics (i.e., positive interactions outweighed negative interactions; (*6*)).

This work led us to want to more fully understand the microhabitats these two species were using within breeding sites (see Figure 2). Therefore, we examined microhabitat characteristics of both the Florida bog frog and Bronze frog to understand if there was overlap in microhabitat use of male calling sites. We hypothesized that the bog frogs would select a narrower range of characteristics compared to the Bronze frog and we predicted that factors influencing site selection would include (i) cover for protection to calling males from predators, (ii) suitable conditions for oviposition, or (iii) variables that may encompass both increased cover and oviposition potential. The results of our research supported the notion that bog frog select a narrow range of characteristics at male calling sites, and that in these same sites, Bronze frog selected a more general suite of habitat conditions (*7*). Specifically, bog frog calling sites were best described by habitat features related to mi-

Fig. 1: Florida bog frog (*Lithobates okaloosae*) breeding habitat on Eglin Air Force Base, Florida, USA. Photo: Thomas Gorman.

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crohabitat cover (i.e., submergent vegetation, emergent vegetation, woody debris, frog-level canopy cover, distance to cover), whereas the Bronze frog selected sites based on features that may be favorable for female oviposition or egg survival (i.e., depth, water movement, depth X water movement interaction). At the microhabitat scale, it is interesting to note that these two species appear to select microhabitat differently, with some overlap in selected variables like percent emergent vegetation and distance to cover (*7*).

Understanding the nature of ongoing hybridization is an important aspect of any long-term management plan. We took a molecular genetic approach to quantify the occurrence and degree of contemporary hybridization between these two species. Our results indicated that approximately 5-10% of the 350 frogs examined were hybrids. This estimate included approximately half that were considered putative hybrids when captured, and half that were not identified as such, suggesting that visually identifying hybrids in nature may be difficult (*8*). While this level of hybridization may not impact the genetic integrity of bog frogs, the rarity of this species does increase the risk that hybridization may influence its population demographics. For example, it is possible that introgressive hybridization between these species could facilitate extinction through genetic assimilation. However, without additional research to determine the ecological mechanisms that are driving this hybridization and the extent of backcrossing that is occurring between hybrids and putative bog frogs, impacts will be difficult to determine.

Combined, our recent work on the ecology and genetics has allowed us to increase our knowledge about the Florida bog frog and its interactions with the Bronze frog. While this work is a step in the right direction, many interesting questions remain. Our genetic work has identified genetic differences between the bog frog and Bronze frog that suggested management and conservation efforts will need to be specifically directed to the bog frog (*9*). To this end, the Florida bog frog may benefit from targeted habitat management, including prescribed fires that penetrate the riparian habitat where they breed. This type of habitat management may increase habitat suitability and perhaps limit opportunities for hybridization to occur, however this link needs further exploration. We are currently investigating how fire structures vegetation in riparian habitat by comparing pre-treatment data in riparian areas with post-treatment data of sites that were burned by both wildfire and prescribed fire. Lastly, while hybridization appears to be low, future efforts should focus on genetic monitoring at a suite of breeding sites to ensure demographic changes are not taking place (*8*).

Fig. 2: Florida bog frog (*Lithobates okaloosae*; left) and bronze frog (*L. clamitans*; right). Photo: Thomas Gorman.

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Incorporating Search Method and Age in Mark - Recapture Studies of an Indicator Species, the Red -Backed Salamander

By Eric B. Liebgold[1] & Frances E. Buderman[2]

Abundance estimates are critical for understanding population trends in amphibians and identifying populations of concern. Determining whether the search methods used by researchers affect the likelihood of catching amphibians and the accuracy of estimates of population size is especially important in species of conservation concern and species that are indicators of ecosystem health, such as woodland salamanders in the family Plethodontidae (*1*).

Fig. 1: A clutch of red-backed salamanders (*Plethodon cinereus*) hatching. Fig. 2 Undergraduates searching for salamanders at night. Fig. 3 12 x 12 m plot where over 700 salamanders were captured and marked in the three years. Photo: Eric B. Liebgold.

The red-backed salamander, *Plethodon cinereus*, is a fully terrestrial salamander that has no aquatic larval stage, but hatches into a miniature adult (Fig. 1). Terrestrial salamanders are often counted by searching under moist rocks and logs during the day or by monitoring under wooden boards set out by researchers (*1*). A less commonly used method for monitoring salamanders is night searches on wet nights for salamanders that have emerged from underground retreats like earthworm burrows (*2*) to forage on the ground surface.

Data from animal surveys are typically collected as count data, the numbers of animals observed during each search, and this data is commonly used to infer population trends. However widespread, count data may not be an accurate representation of population estimates. This is because the number of animals counted during a survey varies widely based on the likelihood that an animal is encountered. As a result, factors like search methodology (e.g., time of day), and variation in recent environmental conditions, such as precipitation between surveys, which affect amphibian activity and choice of microhabitat, make it difficult to interpret count data for salamanders.

Marking and recapturing animals avoids this difficulty because it enables researchers to estimate changes in survival and encounter probabilities between surveys (*3*). For salamanders, simple mark-recapture modeling still includes unexplained variation due to the large proportion of time salamanders spend underground, which complicates estimation of encounter probability and obscures abundance estimates (*4*). This may be alleviated in part by increasing search effort and improving the likelihood of capturing and recapturing individuals by using multiple types of searches.

We tested the hypotheses that mark - recapture models that included search method and age of salamanders would be better fits to mark - recapture data and improve estimation of abundances. We used day searches looking under natural rocks and logs and night searches of the ground surface on wet nights (Fig. 2) to mark over 700 salamanders in 2005, 2006, and 2009 on a single 12 x 12 m plot (Fig. 3) near Mountain Lake Biological Station in Virginia in the southern Appalachian mountains, which have the greatest species diversity of salamanders in the world and the highest densities of Red-backed salamanders (2.8 per m^2: (5)). Captured salamanders were marked by injection of combinations of UV fluorescent colored elastomers under the translucent skin of their bellies in five locations so that each salamander had a unique color code (Fig. 4). We also estimated age of salamanders, determining whether they were sexually immature juveniles (1 - 2 years old; typically 3 - 5 cm long) or adults (>8 cm long). After marking and measuring salamanders, they were released. Red-backed salamanders in this area have home ranges of 1.37 m^2 (*6*), and can live at least 12 years (unpublished data). As a result, we recaptured salamanders nearly 1000 times on our plot and some salamanders were found under the same rock or log it was originally captured under consistently over the five - year period.

We designed our survey schedule using Pollock's Robust Design (*7*). This design for mark-recapture data collection splits survey dates up in two ways in order to disentangle and estimate encounter rates and survival rates. Primary time periods are separated by

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Fig. 4: Female Red-backed salamander #211 with unique fluorescent elastomer five-color code. Photo: Eric B. Liebgold.

long lengths of time that include deaths, immigrations, and emigrations. Each of our primary time periods (five per summer) included 3-5 secondary searches where the only variation between searches should be encounter probabilities (i.e., not survival or emigration), allowing estimation of initial and re-encounter probabilities as well as population size. We calculated these probabilities in Program MARK and used a model selective framework to determine what factors were most important for inclusion in mark-recapture models (*8*).

We found that night searches were consistently better than day searches for initially capturing salamanders (Fig. 5). In 2005 and 2006, night surveys were 66% better at detecting salamanders, regardless of salamander age. This makes sense because red-backed salamanders are territorial, with large adults often excluding other salamanders from high-quality territories under rocks and logs (*5*). This territoriality may have also led us to be twice as likely to recapture marked salamanders during day searches compared to night searches (Fig. 6). As a result, all of the top mark-recapture models included search method in their calculations. In other words, not only did utilizing both day and night searches increase our ability to both detect and recapture salamanders, but including information on how we found individual salamanders in models gave better estimates of encounter rates and abundance.

In contrast, there were no differences between the likelihoods of capturing adults and juvenile salamanders using either search method (Figs. 5-6), nor were abundance estimates affected by inclusion of age in our mark-recapture models. However, there was huge yearly variation in salamander abundance estimates (Fig. 7), from over 6 salamanders/m^2 in 2006 to less than 1/m^2 in 2009, mostly due to yearly differences in the abundance of juveniles.

Our general conclusion is that wildlife managers and researchers should consider using multiple search methods when estimating abundance and population trends because different methods are likely to have different capture and recapture probabilities. Importantly, if multiple methods are used, inclusion of methodology when modeling mark-recapture data helps refine these estimates. For studies monitoring terrestrial amphibian populations, we specifically recommend a) including night searches on wet nights in order to more accurately assess population sizes and trends and b) conducting surveys over multiple years in order to account for potentially extreme variation in juvenile abundances.

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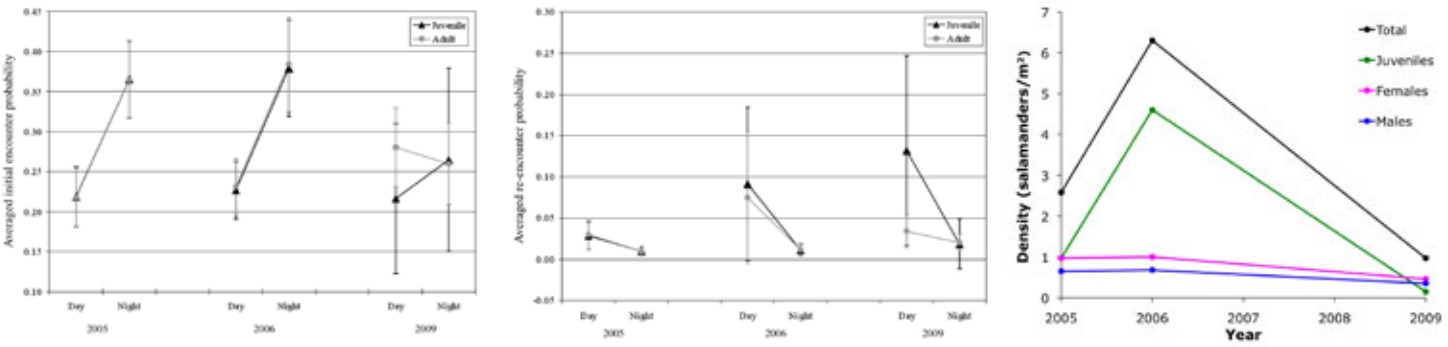

Fig. 5: Initial encounter probabilities (averaged across all mark-recapture models) were greater during night surveys than day surveys except for juveniles in 2009. Fig. 6 Recapture probabilities (averaged across all mark-recapture models) were greater during day surveys than night surveys. Fig. 7 Density estimates for red-backed salamanders at our site at Mountain Lake Biological Station. Note the extreme variation in juvenile (1-2 years old) density between years.

Concern Over Destroying Frog Habitat on the Occasion of Save The Frogs Day in Bangladesh

M. Nurul Islam[1,2], A. H Shaikat[2], S. A Khan[2], M. M Hassan[2,3] & Kerry M. Kriger[1]

Frog lovers and environmentalists of all ages celebrated the 4th Annual Save The Frogs Day on April 28, 2012. More than 200 events took place in 38 countries including the USA, Bangladesh, Australia, Ghana, Brazil, India and Pakistan. In Bangladesh, 18 Save The Frogs Day events took place around the country, through the assistance of SAVE THE FROGS! USA, One Health Young Voice and Chittagong Veterinary & Animal Sciences University (CVASU).

The central events in Bangladesh on the big day took place at CVASU. Save The Frogs! Bangladesh volunteers arranged a frog painting competition and an art exhibition. A colorful parade also brought attention in the Chittagong Metropolitan area and later a central seminar was held in the CVASU conference room. The theme of the seminar was "Our Role in Frog Conservation in Bangladesh." In the seminar the speakers expressed their deep concern over habitat destruction by filling the wetland and use of harmful pesticides on crops. An effective discussion was carried out to determine the best method to track the illegal hunting of amphibians for trading, and overharvesting of wild frogs by the tribal people for eating purposes.

Another scientific conference took place in Faculty of Fisheries in Bangladesh Agricultural University (BAU). Professor Dr. Muklesur Raman, Bangladesh's famous amphibian biologist, presented the keynote paper on the seminar. He gave emphasis on the conservation issue of and possible causes of amphibian declines in Bangladesh. He also requested a cessation of inhumane dissection of wild caught frogs for educational and research purposes in Bangladesh and requested the authorities introduce digital dissecting software in Bangladesh. Another 16 events took place around the country including Shahjalal University of Science and Technology by Green Explorer Society, Sylhet Agricultural University, Sirajgong, Chapai Nawabgong, Noagoan, Chandpur, Coxsbazar and Safari Park. All the events were focused on the role of frogs in nature and the causes of their declines in Bangladesh and across the world. The events also emphasized how we can help to protect amphibians from unprecedented rate of decline. More than 1,500 participants on those events including school children, Veterinarians, Zoologists, Teachers, Journalists, Politicians, and Farmers got educated regarding the importance of frogs, causes of decline and ways to help the frog populations in Bangladesh.

Unfortunately the amphibian conservation issue is a neglected matter in Bangladesh. No major studies have been conducted yet to identify and quantify the threats to amphibian populations in Bangladesh. It's an urgent need to identify the diversity and hotspots of frog populations in Bangladesh and to develop proper management strategies to protect the amphibians in Bangladesh. It is hoped that the activity on Save The Frogs Day will generate media attention and community engagement to protect the frogs in Bangladesh.

We aim to establish an international branch of SAVE THE FROGS! to conduct educational programs and research regarding amphibian conservation in Bangladesh. We appreciate your kind help and suggestions on acquiring funding for SAVE THE FROGS! Bangladesh. You can find us at www.savethefrogs.com/countries/bangladesh and on Facebook at www.facebook.com/savethefrogsbangladesh and don't hesitate to contact me at nurul@savethefrogs.com for any details.

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Eastern Afromontane Amphibian Assessment

Trento Italy, 2nd June – 4th June 2012

Summary

The 2004 Global Amphibian Assessment (GAA) was a landmark initiative that served as the foundation for many conservation actions worldwide. The assessment provided evidence of alarming declines in amphibians, with almost one third of the then >5,700 species assessed to fall into one of IUCN's Threatened categories. However, it is now eight years since this first comprehensive assessment including African amphibians was published. Since 2005, nearly 1,300 amphibian species have been described or revalidated. In addition, a wealth of new field data can potentially inform the conservation status of species. The identification of priority species and areas is achieved in great part due to conservation assessments, and tracking changes in species and areas is thus fundamental for informing conservation action both locally as well as on a regional and global scale. This however is only possible if assessments are maintained to make them current and informative.

Sub-Saharan Africa has a rich and unique amphibian fauna, with mainly endemic families and genera. The region contains 1011 species. Of these, 263 are currently assessed as Threatened, with many having highly restricted distributions. One region of high amphibian species richness is the Eastern Afromontane region, a recognized global biodiversity hotspot with 181 amphibian species, 167 of which have been assessed. Of these, 40% are Threatened — a higher proportion than the global average. A remarkable 39 species of the 799 EDGE (Evolutionary Distinct – Greatly Endangered) amphibian species are also located in this region (5%). Habitat loss, detrimental environmental change and/or spread of emerging infectious diseases are all likely having a big impact on amphibians in this region. However, currently our understanding of the amphibian fauna of this region is patchy in coverage and quality and requires disparate pieces of information to be linked. Focus on this region is urgently required.

In conjunction with the African Amphibian Working Group Meeting in May 2012, we aim to reassess all Eastern Afromontane African amphibians. We expect the assessment to provide a vital update to the status of the highly Threatened amphibian fauna of Africa, and thereby contribute to its long-term conservation and preservation.

Timetable

Following the AAWG 2012 meeting 28th May-31st May (Monday-Thursday), we will conduct a three day workshop from 2nd June-4th June (Saturday-Monday).

Pre-Workshop Objectives

In order to successfully meet the goals of the workshop we need experts interested in the Eastern Afromontane amphibian fauna to update information on taxonomy, spatial distribution of species and their conservation. We have distributed species lists to experts to form a basis for informing us on what species you can update.

We will prioritize the reassessment of Threatened species. First we plan to assess all Threatened amphibians, including EDGE species. In addition, we plan to re-assess Data Deficient species for which there is recent information relevant to their conservation status. Lastly, we will re-assess range-restricted species that are considered as Least Concern following IUCN categories. Overall, we expect to collate new data on taxonomy, distribution, and conservation for ca. 100 Threatened and Data Deficient species.

Workshop Objectives

Preliminary draft assessments of species will be created and new information will be compiled prior to the workshop. Workshop assessments will be completed with IUCN facilitators. During the meeting we therefore aim to: (1) Re-assess all Threatened Eastern Afromontane African amphibians, (2) Discuss action plans for all Critically Endangered Eastern Afromontane amphibian species, (3) Utilize the data to update web-based databases (e.g., IUCN Red List of Threatened Species™), (4) Compile a top 10 list for the most threatened Eastern Afromontane African amphibians and use it to lobby for their conservation.

Contributors

Please can you inform us of your willingness to participate by either i) Filling out the registration form for the AAWG 2012 meeting asap if your attending, or ii) directly email Fabio Pupin (fabiopupin@inventati.org) stating your interest in contributing to the assessment, either remotely or just by attending the workshop (2nd-5th June). We will then contact you directly with an excel database for you to fill out. We need to have an estimation of participants so that we can calculate costs and make bookings for accommodation.

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Recent Publications

Conservation and Ecology

Linking extinction-colonization dynamics to genetic structure in a salamander metapopulation

By Bradley J. Cosentino, Christopher A. Phillips, Robert L. Schooley, Winsor H. Lowe & Marlis. R. Douglas

In metapopulations with demographic turnover, theory predicts that founder effects have a primary role in determining spatial genetic structure. However, among-patch heterogeneity in ecological factors that affect extinction and colonization probabilities may also create spatial variation in the strength of genetic drift and migration, obscuring genetic founder effects. We used microsatellite markers to test the hypothesis that ecological factors underlying extinction-colonization dynamics influenced the genetic structure of a Tiger salamander (*Ambystoma tigrinum*) metapopulation in Illinois, USA. We used empirical data on metapopulation dynamics to make *a priori* predictions about the effects of population age and ecological factors on genetic diversity and divergence among 41 populations. Previous research demonstrated that metapopulation dynamics of *A. tigrinum* were influenced by wetland area, spatial connectivity, and presence of predatory fish. We found that newly colonized populations were more genetically differentiated than established populations, suggesting founder effects influenced genetic structure. However, ecological drivers of metapopulation dynamics were more important than age in predicting genetic structure. Consistent with demographic predictions from metapopulation theory, genetic diversity and divergence depended on wetland area and connectivity. Divergence was greatest in small, isolated wetlands where genetic diversity was low. Our results show that ecological factors underlying metapopulation dynamics can be key determinants of spatial genetic structure, and that habitat area and isolation may mediate the contributions of drift and migration to divergence and evolution in local populations.

Ambystoma tigrinum (Tiger salamander) is a pond-breeding amphibian in North America. The genetic structure of an *A. tigrinum* metapopulation in Illinois, USA was strongly dependent on the size and spatial isolation of breeding ponds. Photo: Brad Cosentino.

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A comparison of two non-lethal methods for dietary studies in terrestrial salamanders

By Federico Crovetto, Antonio Romano & Sebastiano Salvidio

Non-lethal methods should always be preferred to investigate food habits in amphibians, but there are no studies that compared the dietary data obtained by different non-lethal methods from the same individuals. Therefore, we analysed the dietary habits obtained by stomach flushing and faecal analysis from the same 31 *Speleomantes strinatii*, a terrestrial salamander living in Italy and belonging to the family Plethodontidae. Salamanders were stomach flushed in the field and individually caged in the laboratory to obtain also faecal samples. In addition, a reference collection of prey items was collected in the same habitat and at the same time as the salamander sample. The number of prey taxa was similar in the two dietary samples and there were no differences in the number of indeterminate prey items. However, niche overlap between the two samples was relatively low (Pianka's index O_{jk}= 0.58) and prey diversity was significantly higher in the stomach contents (Simpson's diversity index 0.86 ± 0.02 s.d.) that than in the faeces (0.77 ± 0.04 s.d.; permutation test P = 0.001). The number of fly larvae and springtails was significantly higher in the stomachs, while the reverse was true for ants that were about six times more abundant in the faecal samples. Our study demonstrates that different non-lethal dietary methods, even if applied on the same individual salamanders, produced different data-sets and allowed different interpretations about the trophic niche of the study population. Stomach flushing was more informative and gave more accurate insights on the diversity and the trophic strategy of the study population. This because heavy armoured prey items, such as ants, tend to accumulate along the salamander digestive tract becoming the dominant prey in the faecal samples, while slightly *chitinised* soil invertebrates, such as fly larvae and springtails, are almost completely degradated during the digestive process. These results have relevant ecological implications, in particular when the study of the ecological role of salamanders, that consume and regulate the populations of soil-dwelling invertebrates in temperate forest ecosystems, is the aim of the study.

Flushed stomach content (left) and faecal sample (right) from the same individual salamander. Photo: Sebastiano Salvidio.

F. Crovetto, A. Romano, S. Salvidio, *Wildlife Res.* **39**, 266 (2012).

Terrestrial herpetofaunal assemblages in secondary forests and exotic *Lophostemon confertus* plantations in South China

By Yik-Hei Sung, Nancy E. Karraker & Billy C.. Hau

Deforestation is one of the major causes of biodiversity loss. Globally, large areas of primary forest have been replaced by secondary forests and plantations, and changes in tree species composition have often led to alteration of plant and animal communities that were associated with those primary forests. Hong Kong has experienced a long history of deforestation and may be one of the pioneers in using exotic tree plantations for forest restoration in Asia. In this study, we sampled herpetofaunal assemblages in native, secondary forests and exotic *Lophostemon confertus* plantations by transect surveys, pitfall traps, and coverboards. Amphibians were more abundant in secondary forests than in plantations, while the abundance of reptiles and species compositions of both amphibians and reptiles were similar in both forest types. Body condition of the most common reptile, *Sphenomorphus indicus* (Indian forest skink), was similar

An adult of Indian forest skink (*Sphenomorphus indicus*). Photo: Yik-Hei Sung.

between secondary forests and plantations. However, higher proportions of S. indicus exhibited autotomized or missing tails in secondary forests than in plantations, which may be related to higher percentages of rock and wood cover on the forest floor potentially increasing the chance of escape from predators in secondary forests. Secondary forests may provide a better habitat for herpetofauna, we suggest that planting of a mixture of native tree species and thinning of exotic trees may be favorable management efforts to enhance biodiversity in future plantation in South China.

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A quantitative comparison of two common amphibian sampling techniques for wetlands

By Robert D. Denton & Stephen C. Richter

Determining the most appropriate sampling protocol for aquatic amphibians is critical for obtaining reliable survey data. The most widely used active sampling technique for aquatic amphibians is dipnetting, often by controlling for time or effort. Aquatic minnow nets are a common passive technique for surveying lentic habitats. We compared these two methods over a four—month period in 19 wetlands on the Daniel Boone National Forest, Kentucky in order to determine which method is most appropriate for individual species. Each pond was surveyed for a three-day period per month (May–August 2010) using standardized dipnetting and aquatic minnow traps with aquatic drift fencing. Out of a total of 13 detected species, 12 were captured by both methods. Four-toed salamanders (*Hemidactylium scutatum*) were only detected as larvae and only by dipnetting. For the remaining species and associated life stages, we compared abundances from dipnetting and minnow trapping using Wilcoxon sign-rank tests, and seven of these comparisons were significant. Larval Spring peepers (*Pseudacris crucifer*), Four-toed salamanders, Spotted salamanders (*Ambystoma maculatum*), Jefferson salamanders (*A. jeffersonianum*), and Red-spotted newts (*Notopthalmus viridescens*) were captured in significantly higher abundances using dipnetting. Adult Green frogs (*Lithobates clamitans*) and Red-spotted newts were captured in significantly higher abundances using minnow trapping. Overall trends indicated that dipnetting resulted in higher abundances for fast-moving and small larvae such as ambystomatids, four-toed larvae, and toads (*Anaxyrus sp.*). Minnow trapping was more effective for species that had the highest densities in the ponds during their breeding period, including Gray tree frog (*Hyla chrysoscelis*) larvae, Wood frog (*L. sylvaticus*) larvae, and Red-spotted newt adults. Because species-specific biases were apparent when using a single sampling method, we suggest a combination of survey techniques and calculations of detection probabilities in order to obtain the most accurate amphibian survey data.

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Captive management and reproductive biology of Orlov's treefrog, *Rhacophorus orlovi* Ziegler & Köhler, 2001 (Amphibia: Anura: Rhacophoridae), including larval description, colour pattern variation and advertisement call

By Marlen J. Wildenhues, Mikhail F. Bagaturov, Andreas Schmitz, Tran Thi Anh Dao, Ralf Hendrix & Thomas Ziegler

In times of the global amphibian crisis *ex-situ* captive-breeding programs have proven to be valuable tools both in species preservation and natural history as well as larval development research. The importance of zoos and aquariums for amphibian conservation breeding is gaining more attention recently. Breeding of endangered species can help to build up reserve populations or to provide specimens for reintroduction into nature. By keeping and breeding endangered or barely known species also important insights into the reproductive biology can be achieved and the morphological descriptions of the larval stages help to identify early stages

Couple of *Rhacophorus orlovi* in amplexus building a foam nest in Leningrad Zoo St. Petersburg. Photo: M. F. Bagaturov.

FrogLog Schedule

January - South America
March - Europe, North Africa and West Asia
May - North and Central America and the Caribbean
July - Sub Saharan Africa
September - Mainland Asia
November - Maritime Southeast Asia and Oceania

Robin Moore / iLCP

of amphibians in the field as prerequisite for subsequent ecological research and adequate conservation measures. The successful keeping and breeding of *Rhacophorus orlovi*, Ziegler and Köhler, 2001, from Vietnam in the Leningrad Zoo, St. Petersburg is described for the first time. In addition, we provide the morphological description of the larva of *R. orlovi*, based on specimens identified by DNA barcoding and we describe so far unknown subadult and adult colour patterns as well as the advertisement call of this species from Vietnam. Breeding took place based on two couples from northern Vietnam at low temperatures of 19°-24° C and an ambient humidity of around 90% with daily misting. The species turned out to have a noticeably rapid development. Slightly more than two weeks after foam nest building, larvae developed hindlimbs, and after about four weeks, all larvae were metamorphosed, with a snout-vent length of 10.5 mm. Tadpoles of *R. orlovi* are of rather generalized morphology with a labial tooth row formula of 4(2-4)/3(1). Compared to other rhacophorid larvae the larva of *R. orlovi* is quiet small and has the low number of four keratodont rows at the upper labium. In adult, wild and captive held, individuals of *R. orlovi* we could observe a yellowish-green back pattern. Facing the advertisement call of the investigated species there is a similarity of the call structure to the species *Polypedates leucomystax*. The implementation of the breeding of a species and the setting up of captive breeding programs might be one step towards the important role of zoos and aquariums as institutions being involved in species conservation action. Hopefully, the pending paper motivates others to publish similar articles of different frog species.

M. J. Wildenhues *et al.*, *Zool. Garten N.F.* **80**, 287 (2011).

Husbandry, captive breeding, larval development and stages of the Malayan horned frog *Megophrys nasuta* (Schlegel, 1858) (Amphibia: Anura: Megophryidae)

By M. Wildenhues, A. Rauhaus, R. Bach, D. Karbe, K. van der Straeten, S. T. Hertwig & T. Ziegler

We report our long-term experience with the successful keeping and breeding of *Megophrys nasuta* at the Cologne Zoo's amphibian breeding unit and compare our data with other breeding reports. We also document the development and morphology of different larval stages of *M. nasuta*. Diagnostic morphological characters are provided for Gosner (1960) larval stages 18–22 and

Megophrys nasuta larvae in stages 41 to 46. Photo: M. Wildenhues.

25–46. Ovipositions were not seasonal and took place after a drier phase in the terrarium followed by intensive spraying to simulate the natural rain period. The larvae hatched about one week after egg deposition. The characteristic funnel-shaped oral disc became discernible about two weeks after egg deposition at Gosner stage 21 and degenerated at Gosner stage 42. The mean total developmental time observed by us for *M. nasuta* was 2.5–3.5 months. Larvae developed faster at higher temperatures and lower densities. The triangular projections at the upper eyelids, which are characteristic for the advanced terrestrial stages, began to develop after about two or three weeks after completion of metamorphosis.

M. Wildenhues *et al.*, *Amphib. Rept. Conserv.* 5, 15 (2012).

The role of forest harvesting and subsequent vegetative regrowth in determining patterns of amphibian habitat use

By Viorel D. Popescu, David A. Patrick, Malcolm L. Hunter Jr. & Aram J. K. Calhoun

Despite increased efforts towards integrating forest management and biodiversity conservation, little is known about the short-term vegetative regrowth post-harvesting on forest amphibians. We studied the response of eight species (four forest specialists: Wood frog - *Lithobates sylvaticus*, Spotted salamander - *Ambystoma maculatum*, Red-backed salamander - *Plethodon cinereus*, Red-spotted newt - *Notophthalmus viridescens*, and four habitat generalists: Green frog - *L. clamitans*, Bullfrog - *L. catesbeianus*, Northern leopard frog - *L. pipiens*, Pickerel frog - *L. palustris*) to four forestry treatments (partial harvest, clearcut with coarse woody debris [CWD] removed, clearcut with CWD retained, and uncut control, all centered on a breeding pool) over a six year period in Maine, USA. Forest amphibians showed a strong negative response to clearcutting through the duration of the study, regardless of the presence of CWD, but only during the post-breeding season (i.e., summer). The spring breeding migrations of *L. sylvaticus* and *A. maculatum* to experimental pools were not affected by the forestry treatments. The use of partial cut treatments by forest amphibians differed between animals emerging from experimental pools (i.e., juvenile *L. sylvaticus* and *A. maculatum*), and animals originating from outside the experimental arrays (i.e., adults of all forest species, juvenile *L. sylvaticus* and *A. maculatum*). Animals emerging from our experimental pools showed no difference in the use of control and partial cut treatments, while all the other animals preferred control plots. In addition, we found a modest increase in the use of clearcuts over the six years following harvesting by juvenile *L. sylvaticus* leaving experimental pools (from

Aerial image of one of the four experimental sites 2 years post-harvesting (project LEAP - Land Use Effects on Amphibian Populations); clearcuts are located opposite of each other and the breeding pool is located in the center. Photo: M.L. Hunter.

an eight fold difference between forest and clearcut treatments in the first year post-clearcutting to a three fold difference during years 3–5). Forest specialists declined in abundance in all treatments beginning 2–3 years post-disturbance, and there was a shift in relative abundance towards habitat generalists, most notably *L. clamitans*. Habitat generalists were not affected by clearcutting or vegetative regrowth. These general patterns of habitat use were overridden at the local scale by site-specific variation in the use of forestry treatments, most evident in emigrating juvenile *L. sylvaticus*. From a management standpoint, implementing broad silvicultural prescriptions could be a viable strategy in extensively forested landscapes, but local variation in habitat use has to be acknowledged when managers focus on a limited area.

V. D. Popescu, D. A. Patrick, M. L. Hunter Jr., A. J. K. Calhoun, *For. Ecol. Manage.* **270**, 163 (2012).

Estimates of sex ratio require the incorporation of unequal catchability between sexes

By Evan J. Pickett, Michelle P. Stockwell, Carla J. Pollard, James I. Garnham, John Clulow & Michael J. Mahony

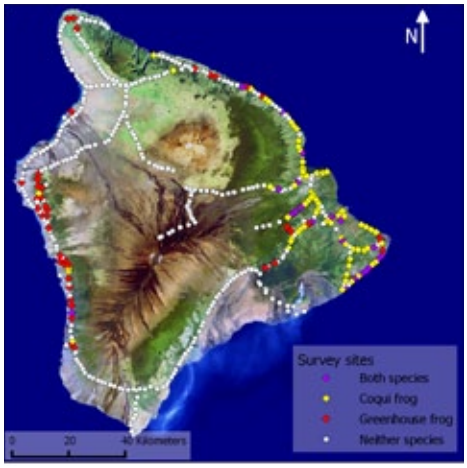
Estimates of the sex ratio of a population are a common summary statistic used for ecological studies and conservation planning. However, methods to determine the sex ratio often ignore capture probability which can lead to a perceived bias in the sex ratio when the sexes are detected at different rates. Our aim was to illustrate the bias from conventional count-based analysis methods for determining sex ratio by comparison with analytical methods which include capture probability. Closed population mark-recapture analysis was used to determine the population size of each sex within a population of Green and golden bell frogs (*Litoria aurea*). This was then compared to the traditional count-based methods of estimating sex ratio to determine the effect of incorporating capture probability on the sex ratio estimate. More males were detected during surveys than females, producing a male-biased sex ratio when there was no incorporation of capture probability. Mark-recapture results indicated a similar population size between the two sexes, suggesting the sex ratio is closer to even. Methods to estimate sex ratios that incorporate capture probability can significantly reduce the bias obtained from count data. We suggest that population studies must incorporate capture probability to determine the sex ratio of a population.

E. J. Pickett *et. al.*, *Wildl. Res.* (2012) DOI: 10.1071/WR11193

Detection probabilities of two introduced frogs in Hawaii: implications for assessing non-native species distributions

By Christina A. Olson, Karen H. Beard, David N. Koons & William C. Pitt

Understanding the distribution of non-natives, especially cryptic non-natives, is critical in determining their degree of invasiveness and managing their spread. Two non-native Caribbean frogs, the Puerto Rican Coqui and the Cuban Greenhouse frog, recently invaded Hawaii. Because of its louder breeding call, management efforts have focused on the Coqui, while little has been done to address the quieter Greenhouse frog, even though it may be as widespread and have similar ecological impacts. We determined the distribution and detection probability of both species on the island of Hawaii



Coqui and Greenhouse frog presence/absence points on the island of Hawaii, USA. If a frog was detected during any survey, it was included as present. (Source: Landsat –http://hawaii.gov/dbedt/gis/)

by conducting a breeding call presence/absence survey at 446 sites every 2 km along major road networks. We re-surveyed 125 sites twice to determine detection and occupancy probabilities. Greenhouse frog detection probabilities were lower than Coqui detection probabilities and increased with visits while those of the Coqui did not. Greenhouse frog detection probabilities were lower in the presence of Coquis for the first two surveys than in sites with Greenhouse frogs alone, while Greenhouse frogs had no effect on the detection of Coquis. Overall site occupancy estimates for both species were similar, suggesting they are equally widespread. Results suggest multiple visits to sites are required to detect the Greenhouse frog, and that accounting for detectability is essential when determining the distribution of cryptic species.

C.A.Olson, K. H. Beard, D. N. Koons, W. C. Pitt, *Biol. Inv.* **14**, 889 (2012).

Maintenance of biodiversity in vineyard-dominated landscapes: a case study on larval salamanders

By Matteo Tanadini, Benedikt R. Schmidt, Pierre Meier, Jérôme Pellet & Nicolas Perrin

Large pristine areas are rare in Europe. The vast majority of the land is covered by settlements and agricultural areas. The latter cover about two thirds of the European surface, and thus play an extremely important role in the conservation of species. Amphibians are often encountered in seminatural areas such as agricultural fields and adjacent set aside areas.

The paper looks at the influence of aquatic and terrestrial habitats on the abundance of Fire salamander larvae (*Salamandra salamandra*) in a vineyard-dominated landscape close to Lake Geneva in Switzerland.

Results confirmed previous studies by highlighting the importance of landscape complementation for amphibians. Models including both stream and landscape variables best explained larval abundances.

Our study indicates simple practices for farmers to favour Fire salamanders and most likely wildlife in general in vineyard-dominated landscapes, e.g. allowing riparian vegetation to grow along streams and reduce grass removal within vineyards.

Larval densities were higher in the vineyard landscape than in natural ecosystems. This suggests that salamanders can persist in a high-intensity agricultural landscape such as vineyards. Moreover the highest local densities were encountered very often in stream sections with man-made stream elements (weirs). Hence habitat modifications do not always result in negative impacts for wildlife. Thus conservationists should not underestimate the conservation value of man-made habitats, especially in human-dominated regions.

Adult Fire salamander (*Salamandra salamandra*). Photo: Jérôme Pellet.

M. Tanadini, B. R. Schmidt, P. Meier, J. Pellet & N. Perrin, *Anim. Conserv.* **15**, 136 (2012).

Diet of the introduced Greenhouse frog in Hawaii

By Christina A. Olson & Karen H. Beard

The Greenhouse frog (*Eleutherodactylus planirostris*) is a nocturnal, terrestrial species native to Cuba and the Bahamas that has invaded areas of the southeastern United States, Jamaica, Guam, and five Hawaiian Islands. First recorded in Hawaii in 1994, the invasion has not been well-studied. Because it is an insectivore, the greatest threat is to Hawaii's native invertebrates. We conducted a stomach content analysis of frogs from the island of

Photograph of adult female Greenhouse frog (*Eleutherodactylus planirostris*) taken in Hawaii. Photo: Christina A. Olson.

Hawaii and determined that the dominant prey items consisted of ants, mites, and springtails. Invertebrate orders that contain native species in Hawaii found most often in the stomachs included mites, springtails, spiders, and booklice, although 43% of their diet was non-native invertebrates, including ants, isopods, and amphipods. At each study site, we collected invertebrates in the environment to determine if diets were representative of the available resources, and found that greenhouse frogs predominantly consumed leaf litter invertebrates and selected proportionately more ants than were available in the environment. We estimated population density at one study site to be 12,500 frogs ha^{-1}. Based on this density estimate and the mean number of prey consumed, we estimated that Greenhouse frogs consume 129,000 invertebrates per night at some sites. This research highlights the need to understand the direct and indirect effects of predation by Greenhouse frogs on invertebrates in Hawaii.

C. A. Olson, K. H. Beard, *Copeia* **2012**, 121 (2012).

Use of a native predator for the control of an invasive amphibian

By Gerald Louette

The control of alien invasive species is essential for securing native biodiversity. However, control tools that combine cost-effectiveness and insure a sustainable outcome are not that readily available. As for invasive American bullfrog *Lithobates catesbeianus*, known to cause ecological damage around the globe, comprehensive management techniques are currently absent. To fill this gap, opportunities arising from habitat restoration were explored. A multiannual biomanipulation experiment was performed in small shallow fish ponds, and effects of complete drawdown (with amphibian and fish removal) and predation (introduction of originally occurring native Northern pike *Esox lucius*) on bullfrogs were investigated. Furthermore, effects of both performed measures were assessed on the whole amphibian and fish community, as well as how the overall ecological quality of the water bodies responded. The presence of pike lead to a strong decline in bullfrog tadpole numbers, while no effect of drawdown was observed. Also, communities receiving pike harbored substantially less small and mostly planktivorous fish species (e.g. Pumpkinseed *Lepomis gibbosus* and Topmouth gudgeon *Pseudorasbora parva*). Moreover, these pike-inhabited ponds tended to have a higher overall ecological quality. The reduction in bullfrog tadpoles may be assigned to both direct and indirect effects induced by pike. First, direct pike predation on tadpoles was observed. Second, as the occurrence of macroinvertebrate-feeding Pumpkinseed was low in the presence of pike, the indirect effect of predation by macroinvertebrates on tadpoles may significantly increase, leading to tadpole decline. Habitat restoration of fish ponds, leading to a change in food web interactions, can thus be regarded as an effective tool for the

Control of invasive American bullfrog may be facilitated through biomanipulation (e.g. adding native pike) of small shallow fish ponds. Photo: Sander Devisscher.

control of invasive bullfrog. Piscivorous fish introduction may be applied because of its inexpensive character. This technique, however, requires careful consideration of the indigenous status of the introduced species, angling purposes, or specific nature values. On the long run, however, repeated stocking may be necessary to maintain effects on both bullfrog populations and the overall ecological quality of aquatic habitats.

G. Louette, *Wildl. Res.* **39,** 271 (2012) http://dx.doi.org/10.1071/WR11125.

Genetic structure of the marsh frog (*Pelophylax ridibundus*) populations in urban landscape

By Peter Mikulicek & Peter Pisut

Urbanization is a pervasive process causing habitat fragmentation, spatial isolation of populations and reduction of biological diversity. In this study, we applied eleven microsatellite loci and Bayesian analyses to investigate genetic diversity and population structure in Marsh frogs (*Pelophylax ridibundus*) living in two types of environment – highly fragmented urban landscapes, and landscapes characterized by the presence of a river and artificial canals. Our results show reduced genetic diversity, lower effective population sizes and higher genetic differentiation for spatially isolated urban populations in comparison with populations outside intensely urbanized areas. Reduction of allelic diversity in urban localities isolated for 13 – 37 generations is more conspicuous than reduction of expected heterozygosity. Populations living close to the River Danube, its branches and artificial canals are genetically more homogenous. Our results also suggest that the Danube in Bratislava is not a natural barrier to gene flow. In contrast, it acts as a natural corridor for water frog dispersal. Population structure of *P. ridibundus* also shows higher genetic connectivity within water paths than between them, suggesting limited overland dispersal, and reflects the historical landscape structure associated with the distribution of the lost river branches.

P. Mikulicek, P. Pisut, *Eur. J. Wildl. Res.* DOI 10.1007/s10344-012-0631-5 (2012).

In search of Critically Endangered species: the current situation of two tiny salamander species in the Neotropical mountains of Mexico.

By Adriana Sandoval-Comte, Eduardo Pineda & José L. Aguilar-López

One in three of the amphibian species on the world is endangered and salamanders have the highest portion of their species in one of the risk categories, even higher than the frogs. *Parvimolge townsendi* and *Thorius pennatulus* are two tiny salamanders that are found at intermediate elevations in the mountains of the northern Neotropics. Historically both species were considered abundant, but a few years ago a warning was sounded about the dramatic declines in their populations with the main cause cited as the destruction and modification of their natural habitat. Both species are considered to be Critically Endangered by the IUCN; in fact, *P. townsendi* has been proposed as Possibly Extinct. We evaluated the current situation of *P. townsendi* and *T. pennatulus* by carrying out exhaustive field surveys, between June and November 2010, in both historical and potentially suitable localities (those with bioclimate attributes and vegetation types similar to those of the historical

Individuals of *Parvimolge townsendi* (A) and *Thorius pennatulus* (B) found during field survey. The black bar represents one centimeter. Photos: J.L. Aguilar-López.

localities). We evaluated the abundance of both species and the characteristics of their habitats, and we estimated their potential geographic distribution. We repeatedly visited 22 localities, investing 672 person-hours of sampling effort in the surveys, and found 201 *P. townsendi* salamanders in 11 localities and only 13 *T. pennatulus* salamanders in five localities. Both species were preferentially found in cloud forest remnants that were well conserved or only moderately transformed, and some of the salamanders were found in shade coffee plantations. The potential distribution area of both species is markedly fragmented and we estimate that it has decreased by more than 48%. Although *P. townsendi* and *T. pennatulus* are in the same category of Critically Endangered according to the IUCN, the results of our study suggest that these two species may be facing different levels of extinction risk. That of *P. townsendi* appears to be less worrisome than that of *T. pennatulus*, because the former was found in a greater number of sites, its total abundance was notably higher and it is capable of inhabiting environments with a certain degree of disturbance or management. The approach we used to identify the current status of the two endangered salamander species may be also useful for studying other endangered species.

A. Sandoval-Comte, E. Pineda, J. L. Aguilar-López, *PLoS ONE* **7,** e34023. (2012). doi:10.1371/journal.pone.0034023

Genetic structure among Coastal tailed frog populations at Mount St. Helens is moderated by post-disturbance management

By Stephen F. Spear, Charles M. Crisafulli & Andrew Storfer

Large landscape disturbances often serve as natural experiments that allow greater understanding of ecological response to landscape change. The 1980 eruption of the Mount St. Helens volcano in Washington, USA provided an opportunity to test hypotheses relating to both the effects of natural disturbance as well as post-disturbance management. Despite severe alteration of nearly 600 $km^2$of habitat, Coastal tailed frogs (*Ascaphus truei*) were documented within a portion of the blast area five years post-eruption. We investigated the genetic source of re-colonization within the blast area and tested whether post-eruption management, such as salvage logging and subsequent tree planting, influenced tailed frog movement patterns. Our results support widespread re-colonization across the blast area from outside the blast area, as all sites are grouped into one genetic cluster regardless of disturbance history. Landscape genetic regression analyses suggest gene flow through the unmanaged portion of the blast area is influenced only by distance between sites and frost-free period (r^2 = 0.74). In contrast, gene flow within the blast area where salvage logging and replanting occurred post-eruption is strongly limited (r^2 = 0.83) by the physiologically important variables of heat load and precipitation. These data suggest the lack of understory and coarse wood (downed and standing dead tree boles) refugia in salvaged areas may create a less hospitable matrix for frog movement due to increased susceptibility to desiccation and mortality than frogs moving through the naturally regenerated area. Simulated populations based on the landscape genetic models show an increase in the inbreeding coefficient in the managed area relative to the unmanaged blast area, suggesting future population-level consequences. In summary, we demonstrate resilience of an amphibian species to an extreme disturbance, and we suggest that, at least for this species, naturally regenerating forest habitat may better maintain long-term genetic diversity of populations than actively managed habitat.

Comparison of unmanaged blast area (top) and managed blast area (bottom). Note the high density of coarse woody debris in the unmanaged blast area. Photo: Stephen Spear.

S. F. Spear, C. M. Crisafulli, A. Storfer, *Ecol. App.* **22**, 856 (2012).

Using sighting records to infer extinction in three endemic Argentinean marsupial frogs

By Mauricio S. Akmentins, Laura C. Pereyra & Marcos Vaira

Three species of marsupial frogs of the genus *Gastrotheca* are at the forefront of amphibian conservation concern in Argentina. One of the main reasons is the sudden lack of records of the three species in the last two decades despite intensive searches. Fortunately in early 2011 tadpoles and metamorphs of *Gastrotheca gracilis* were recorded in two repeatedly surveyed historic localities.

Most of the data available on these amphibian species in Argentina are in the form of museum collections. This historical data provides important information on the distribution of taxa through time and space and represents the primary, verifiable observations. A number of methods have been developed in the last few years, which provide a probabilistic basis for testing extinction based on sighting records. These methods consider that extinction should be

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Juvenile of *Gastrotheca gracilis* from one of the rediscovered populations in Argentina after 20 years of the last sight. Photo: M. S. Akmentins.

based on the time since the species was last observed and the frequency with which it was seen before the last observation.

The rediscovery after 20 years of *Gastrotheca gracilis*, gives us an opportunity to test the applicability of these probabilistic methods to infer extinction in such anuran species with cryptic life habits, and could help us to infer the conservation status of the other two Argentinean marsupial frog species, *Gastrotheca christiani* and *Gastrotheca chrysosticta* .

We used seven probabilistic statistical methods and a trend analysis. The first seven approaches test the null hypothesis that the species persist against the alternative hypothesis that it is Extinct. The trend analysis is not probabilistic, but it is designed to determine population trends.

Taking into account the results of quantitative probabilistic methods and the trend analysis performed, we can conclude that these three species are still extant but had suffered some degree of population decline.

The results of this study highlight the usefulness of quantitative methods to make use of available data in museum collections to allow prioritization of further conservation actions for amphibians. These probabilistic methods, in combination with complementary tests and information, should enhance the overall ability to detect species declines.

M. S. Akmentins, L. C. Pereyra, M. Vaira, *An. Cons.* **15**, 142 (2012).

Batrachochytrium dendrobatidis infection of amphibians in the Doñana National Park, Spain

By Judit Hidalgo-Vila, Carmen Díaz-Paniagua, Marc Antoine Marchand and Andrew A. Cunningham

Amphibian chytridiomycosis, caused by the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*), is a highly virulent, recently emerged infectious disease that has caused amphibian population declines and extinctions globally. *Bd* infection has been reported from across much of western Europe. The Doñana National Park (DNP) is an important protected area located in southwestern Spain, a European country with widespread *Bd* infection. This area has a great diversity of aquatic habitats that constitute important breeding habitats for 11 native amphibian species (*Bufo bufo*, *Bufo calamita*, *Pelobates cultripes*, *Rana perezi*, *Hyla meridionalis*, *Pleurodeles waltl*, *Pelodytes ibericus*, *Discoglossus galganoi*, *Triturus pygmaeus*, *Lissotriton boscai* and *Alytes cisternasii*), the last five species, endemic to the Iberian Peninsula. During the first survey for *Bd* infection in DNP, we sampled 625 amphibians of nine species in two periods corresponding with the early and intermediate breeding seasons for amphibians. There was a seasonal effect, and no infection was detected in the amphibians sampled in the earlier period. However, there was an overall 34% prevalence of infection during the intermediate breeding season, with seven of the species infected by *Bd*. Most of the amphibians infected were captured in human-made permanent ponds in which these individuals could act as reservoirs for the fungus when temporary ponds dry out during the summer season. Although some amphibians tested had higher intensities of infection than others, all animals sampled were apparently healthy and, so far, there has been no evidence of either unusually high rate of mortality or amphibian population declines in the DNP. Anyway, we recommend continued monitoring of both amphibians populations and their *Bd* status in the DNP in order to further investigate this host-parasite relationship and determine if any ecological factors, such as prolonged wet periods or periods of drought, may alter the current situation.

J. Hidalgo-Vila, C. Díaz-Paniagua, M. A. Marchand, A. A. Cunningham, *Dis. Aquat. Org.* **98**, 113 (2012).

Current state of *Bd* occurrence in the Czech Republic

By Petr Civiš, Jiří Vojar, Ivan Literák & Vojtech Baláž

A relatively new amphibian disease chytridiomycosis, caused by the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) is recognized as a key threat to global amphibian biodiversity. Central Europe is home to many amphibian species potentially susceptible to the disease, nevertheless no adverse impact on populations or *Bd*-mediated deaths have been observed so far. Only negative results from limited sample sets describing *Bd*'s

European fire-bellied toad (*Bombina bombina*) - a species that carried most of *Bd* zoospores in our survey

status in Europe were available until 2007. This article presents recent results of *Bd* sampling done in the Czech Republic. We detected this pathogen in 41 of 466 animals sampled overall, in 4 of 10 species sampled, and at 8 of 11 sites sampled during the 2010 season. The genus *Bombina* carried most of the infection in populations tested, however our results show very low zoospore loads in almost all cases.

P. Civiš, J. Vojar, I. Literák, V. Baláž. *Herp. Rev.* **43** (1), 75 (2012).

Acute toxicity of three strobilurin fungicide formulations and their active ingredients to tadpoles

By Emily A. Hooser, Jason B. Belden, Loren M. Smith & Scott T. McMurry

Strobilurin fungicide applications in the United States have increased tenfold in the last five years, which is of concern since fungicide formulations have been demonstrated to cause acute mortality to amphibian life stages at environmentally relevant concentrations. The non-selective mode of action for the strobilurin fungicide active ingredients (AIs) is mitochondrial respiratory inhibition creating potential to affect non-target organisms. Further, some formulated fungicides contain adjuvant chemicals such as naphthalene and petroleum distillates that could contribute

to the toxicity of the formulations. Depressional wetlands are found across the intensely cultivated Great Plains and have the potential to receive fungicide overspray or runoff. Therefore, there is a likelihood of exposure for larval Great Plains toads (*Bufo cognatus*) as they are found throughout the Great Plains in these wetlands. We designed a study to compare the differences in acute toxicity to larval *B. cognatus* after exposure to Headline®, Stratego®, and Quilt® (commonly used strobilurin fungicides) and their AIs. Headline® was the most toxic fungicide tested eliciting 79% mortality at 3% of an environmentally relevant concentration. Quilt® was least toxic, resulting in mortality at 27x the environmentally relevant concentration. Stratego® had a steep increase in mortality making it a riskier fungicide as 100% mortality occurred at 2x environmentally relevant concentration. No differences were found between formulations and AIs for Stratego® or Quilt®. However, differences in toxicity were found in Headline®, where the formulation caused 79% mortality at 5 ppb, and the AI resulted in 0% mortality at 5 ppb. Overall, our study showed that the AIs are toxic to tadpoles; however, in the case of Headline®, adjuvant chemicals may be contributing to the toxicity to *B. cognatus* larvae.

E.A. Hooser, J.B. Belden, L.M. Smith, S.T. McMurry, *Ecotoxicology* (2012) DOI 10.1007/s10646-012-0899-y

Batrachochytrium dendrobatidis shows high genetic diversity and ecological niche specificity among haplotypes in the Maya Mountains of Belize

By Kristine Kaiser & John Pollinger

The pathogen *Batrachochytrium dendrobatidis* (*Bd*) has been implicated in amphibian declines around the globe. We collected 524 *Bd* swabs from amphibians in the Maya Mountains of Belize, Central America from June – August 2006-2008. We sequenced the 5.8s and ITS1-3 regions of 72 samples that had tested positive for *Bd* at least once by qPCR; 30 of these were verified as *Bd*. We found high diversity in these samples, with eight unique *Bd* haplotypes, five of which were previously undescribed. Using Maxent modeling, we identified distinct ecological niches for the two most common haplotypes (KK5 and KK15), and found that the environmental factors driving their distributions were dissimilar: although vegetation cover was an important factor in the distribution of both haplotypes, KK5 was driven primarily by temperature and KK15 by precipitation variables. Other studies have shown that different strains vary in virulence. Combined with this information, the diversity shown in the 5.8s-ITS region found here suggests that there may be substantial differences among populations or haplotypes. Future work should focus on whether specific haplotypes for other genomic regions and possibly pathogenicity can be associated with haplotypes at this locus, as well as the integration of molecular tools with other, ecological tools to elucidate the ecology and pathogenicity of *Bd*.

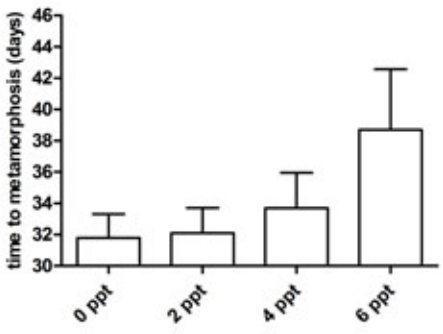
Critically Endangered *Agalychnis moreletii* carrying *Bd*. Photo: Kristine Kaiser.

K. Kaiser, J. Pollinger, *PLoS ONE* **7**, e32113 (2012). Open Access: doi:10.1371/journal.pone.0032113

Effects of salinity on early life stages of the Gulf Coast toad, *Incilius nebulifer* (Anura: Bufonidae)

By Laura G. Alexander, Simon P. Lailvaux, Joseph H. K. Pechmann & Philip J. DeVries

Anuran amphibian populations worldwide are in decline due to a variety of factors including habitat destruction, climate change, disease, introduction of non-native species, and environmental contamination. We conducted a laboratory trial with *Incilius nebulifer* (synonym: *Bufo nebulifer*) to determine at what level salinity negatively affects hatching and metamorphosis, and how exposure to salinity during development affects metamorp characteristics that influence adult fitness. Embryos exhibited 95.5–99.5% hatching success at salinities of 0, 2, and 4 parts per thousand (ppt); 74.4% success at 6 ppt; and no hatching at 8 or 10 ppt. Salinity affected hatching success and larval survival, and we found linear trends between increasing salinity and decreasing fractions of hatched embryos and living larvae. The odds of hatching were about the same for 0, 2, and 4 ppt, significantly lower for 6 ppt, and zero for 8 and 10 ppt. The odds of survival to metamorphosis were significantly lower in 6 ppt relative to 0, 2, and 4 ppt combined. Time to metamorphosis, mass, and hind limb length of recent metamorphs showed significant differences among treatment groups, with salinity having large effects on these variables. Development time was longer, mass was lower, and hind limb length was shorter in the 4 and 6 ppt treatments compared to 0 or 2 ppt. We showed that salinity affected the survival of early life stages of *Incilius nebulifer* and characteristics that have been linked to adult fitness. Our study suggests that low levels of salinity may affect the survival and fitness of other anurans.



Phase II mean (+ SD) results for time to metamorphosis.

Full article: L. G. Alexander, S. P. Lailvaux, J. H. K. Pechmann, P. J. DeVries, *Copeia* **1**, 106 (2012).

Chytrid fungus, *Batrachochytrium dendrobatidis* (*Bd*) detected at lower elevations in Puerto Rico: Implications for conservation of the Puerto Rican crested toad (*Peltophryne lemur*)

By Diane M. Barber

Recovery efforts for the Critically Endangered Puerto Rican crested toad, *Peltophryne lemur*, include a reintroduction program directed by the United States Fish and Wildlife Service, The Puerto Rican Department of Natural and Environmental Resources, and The Association of Zoos and Aquariums Puerto Rican Crested Toad Species Survival Plan. Intensive reintroduction of tadpoles began in 1992 to establish additional populations of *P. lemur* on the island and monitoring efforts include random sampling for the chytrid fungus, *Batrachochytrium dendrobatidis* (*Bd*), in order to assess the overall health of wild *P. lemur* populations and mitigate threats that may be introduced via disease from other amphibian species.

The earliest record of chytridiomycosis detected in Puerto Rico was from preserved *Eleutherodactylus* specimens collected in 1976 and has only been reported at elevations above 600 m. *Bd* has now been detected at much lower elevations from two coqui species residing within *P. lemur*

An adult male Puerto Rican crested toad, *Peltophryne lemur*. Photo: Dustin Smith.

habitat. The susceptibility of *P. lemur* to an outbreak of *Bd* remains unknown and these findings have bearing on future reintroduction efforts and management decisions for wild populations of *P. lemur* within Puerto Rico.

D. M. Barber, *Herp. Rev.* **43**, 73 (2012).

Changes in urinary testosterone and corticosterone metabolites during short-term confinement with repeated handling in wild male Cane toads (*Rhinella marina*)

By Edward J. Narayan, Frank C. Molinia, John F. Cockrem & Jean-Marc Hero

Stressors generally decrease testosterone secretion and inhibit reproduction in animals. Urinary testosterone and corticosterone metabolite concentrations were measured in adult male Cane toads (*Rhinella marina*) at the time of capture from the wild and during 24 hours of confinement with repeated handling. Mean urinary testosterone concentrations increased two hours after capture, were significantly elevated above initial concentrations at five hours, and then declined. Mean testosterone concentrations remained elevated 24 hours after capture. Mean urinary corticosterone concentrations

Herpetologists (Dr. Edward Narayan and A. Prof Jean Marc Hero) sampling Cane toads on the field at the Parkwood International Golf Course, Queensland, Australia. Photo: Edward Narayan.

increased after capture, were significantly elevated above initial concentrations at two hours, and remained elevated thereafter. This is the first report in amphibians of an increase in testosterone excretion after capture from the wild, with previous studies showing either no change or decline in testosterone. This finding may be associated with the mating strategy and maintenance of reproductive effort in the Cane toad, a species that shows explosive breeding and agonistic male–male interactions during breeding. The finding that testosterone excretion increases rather than decreases after capture in male Cane toads shows that it should not be generally assumed that reproductive hormone secretion will decrease after capture in amphibians.

E. Narayan, F. Molinia, J. Cockrem, J. M. Hero, *Aust. J. Zool.* **59**, 264 (2011).

Species diversity reduces parasite infection through cross-generational effects on host abundance

By Pieter T. J. Johnson, Daniel L. Preston, Jason Hoverman, Jeremy S. Henderson, Sara H. Paull, Katherine L. D. Richgels & Miranda D. Redmond

With growing interest in the effects of biodiversity on disease, there is a need for studies that identify the empirical mechanisms underlying the diversity–disease relationship. Here, we combined wetland surveys with mechanistic experiments to evaluate competing explanations for the dilution effect. We focused our study on the multi-host parasite, *Ribeiroia ondatrae*, which is known to cause severe limb malformations in amphibians. Sampling of 320 California wetlands indicated that snail host communities were strongly nested, with competent hosts for the *R. ondatrae* predominating in low-richness assemblages and unsuitable hosts increasingly present in more diverse communities. Moreover, competent host density was negatively associated with increases in snail species richness. These patterns in host community assembly support a key prerequisite underlying the dilution effect. Results of multi-generational mesocosm experiments designed to mimic field-observed community assemblages allowed us to evaluate the relative importance of host density and diversity in influencing parasite infection success. Increases in snail species richness (from 1 to 4 species) had sharply negative effects on the density of infected hosts. However, this effect was indirect; competition with non-host species led to a 95% reduction in host density, owing to reduction in host reproduction. There were no differences in infection prevalence among susceptible hosts as a function of community structure, indicating a lack of support for a direct effect of diversity on infection. In monospecific conditions, higher initial host densities increased infection among adult hosts; however, compensatory reproduction in the low-density treatments equalized the final number of infected hosts by the next generation, underscoring the relevance of multigenerational studies in understanding the dilution effect.

P. T. J. Johnson *et al.*, *Ecology* **93**, 1 (2012).

Populations of a susceptible amphibian species can grow despite the presence of a pathogenic chytrid fungus

By Ursina Tobler, Adrian Borgula & Benedikt R. Schmidt

It is well known that amphibian populations can decline rapidly after an area was invaded by the amphibian chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*). Much less is known how *Bd* affects amphibian populations years, or even decades, after the initial invasion. We studied the effects of *Bd* on the dynamics of 26 populations of the midwife toad *Alytes obstetricans* in Switzerland. *Alytes obstetricans* is known to be susceptible to *Bd*. An analysis of museum specimens showed that *Bd* has been present in Switzerland since at least the 1980ties. Times series data was available for the years 2002-2008 and *Bd* survey data for 2007, 2008 or 2009 (every populations was surveyed in only one year). *Bd* was present in 16 populations. When *Bd* was present, average prevalence was 57%. We also found some dead metamorphs that tested positive for *Bd*. We used a state-space model that accounts for observation error to analyze the short time series of counts of calling males. Unexpectedly, we found no effect of presence or absence of *Bd* on population trends. We offer three alternative explanations for this result. First, Alytes

Several *Alytes obstetricans* were hiding under a stone. *Alytes obstetricans* is a species with male parental care: Two males are carrying egg strings. Photo: Corina Geiger.

obstetricians may be susceptible to *Bd* in the laboratory but not in the field where environmental conditions may not favor *Bd*. Second, populations may be able to compensate for *Bd*-induced mortality (e.g. through increased adult survival or increased recruitment). Third, *Bd* may have caused population declines in the past and populations may have stabilized at low abundance such that an effect of *Bd* is no longer detectable (such a pattern was predicted by the model published by Briggs et al. 2005). In conclusion, the results show that amphibian populations can persist with enzootic *Bd*.

U. Tobler, A. Borgula, B. R. Schmidt, *PLoS ONE* 7, (2012) e34667. doi:10.1371/journal.pone.0034667

Chronic toxicity of copper on embryo development in Chinese toad, *Bufo gargarizans*

By Kun Xia & Hongyuan Wang

Copper is a common trace element in the environment. In recent years, concentrations of copper in the environment have increased dramatically due to widespread use. Excessive copper could lead to water contamination and poor water quality. Therefore, copper has received much attention due to its toxicity, especially to aquatic organisms. This study examined the effects of copper exposure on embryonic development of Chinese toad, *Bufo gargarizans*. The acute toxicity assays were carried out in static fashion for 96 h. And, the LC_{50} values from 24 to 96 h of exposure were 3.61×10^{-6} M, by means of a 4 d toxicity test with *B. gargarizans* embryos.

Chinese toad (*Bufo gargarizans*) in our laboratory spawning.

In long-term exposure experiment, Chinese toad embryos were exposed to 10^{-9}-10^{-6} M copper from mid gastrula stage to operculum completion stage. Measurements included mortality, tadpole weight, tadpole total length, growth retardation, duration of different embryo stages and malformation. Results indicated that embryonic survival was not affected by copper. Relative to control tadpoles, significantly decreased weight and total length were found at 10^{-9}-10^{-6} M reduced percentage of the embryos in right operculum stage after 10 d exposure to copper and reduced percentage of embryos in operculum completion stage after 12 d exposure to copper were also observed. Moreover, the duration of embryonic development was increased at neural, circulation and operculum development stage in copper-treated groups. For the scanning microscope and histological observation, the abnormalities were malformation of wavy dorsal fin, flexural tail, curvature body axis, yolk sac oedema and reduced pigmentation in the yolk sac. Histopathological changes in olfactory, retinal epithelium and skin were observed. The histological morphology of gill and adhesive organ of copper exposed embryos had no difference from controls. DNA strand breaks exposed to the copper were measured by DNA ladder. DNA fragmentation was seen increasing steadily over 1×10^{-6} M, while there was no fragmentation in control group. Higher dose copper exposure is supposed to have more serious effect on DNA fragmentation than low-dose. In conclusion, copper induced toxic effects on *B. gargarizans* embryos. The present study indicated chronic toxicity tests may provide more accurate way in formulating the "safe levels" of heavy metals to amphibian.

K. Xia, H. Zhao, M. Wu, H. Wang, *Chemosphere* **87**, 1395 (2012).

Daphnia in tadpole mesocosms: trophic links and interactions with *Batrachochytrium dendrobatidis*

By Phineas T. Hamilton, Jean M.L. Richardson & Bradley R. Anholt

The involvement of *Batrachochytrium dendrobatis* (*Bd*; the fungus causing amphibian chytridiomycosis) in ongoing amphibian declines has spurred research into the ecological factors that can influence the transmission and effects of the pathogen. *Bd* reproduces through a motile zoospore stage, and thus biotic and abiotic factors that affect zoospores may influence the transmission of disease. In this study, we investigated the effects of predation on zoospores in the water column by grazing *Daphnia* zooplankton. First, we assessed the ability of *Daphnia* to reduce zoospore density in laboratory microcosms, and found that *Daphnia* effectively removed zoospores from the water column. Second, we used outdoor mesocosms to test the effects of *Daphnia* in Red-legged frog (*Rana aurora*) tadpole communities exposed to *Bd*. Interestingly, the presence of *Daphnia* had dramatic beneficial effects for tadpoles, with tadpoles growing nearly twice as massive in the presence of *Daphnia*. Tadpoles also consumed large numbers of *Daphnia* – this was surprising given the prevailing view that anuran tadpoles are herbivorous. In mesocosms, between-tadpole transmission of *Bd* was very low and we were unable to detect an effect of *Daphnia* on *Bd* transmission, although *Bd* and *Daphnia* presence combined to affect tadpole survival. Links between *Daphnia* and *Bd* have yet to be fully clarified, but this study suggests that the presence of *Daphnia* in experimental systems and imperiled wild ecosystems will have major consequences for some amphibians.

P.T. Hamilton, J.M.L. Richardson, B.R. Anholt, *Freshwat. Biol.* **57**, 4 (2012).

Myxozoan parasite in brain of Critically Endangered frog

By Ashlie Hartigan, Cheryl Sangster, Karrie Rose, David N. Phalen & Jan Šlapeta

Frogs worldwide are threatened by habitat destructions and infectious diseases, with many species extinct or close to extinction. One of such extinct frog examples was the Yellow-spotted bell frog (*Litoria castanea*), last seen in 1970th. Fortunately, the frog was recently rediscovered, but animals relocated to captivity have died suddenly. Histopathology revealed acute severe encephalomalacia with directly associated small parasitic stages demonstrated to be myxozoan parasites. Myxozoan parasites are well known in aquaculture and fish hosts where they cause serious disease. It is alarming that virtually nothing is known about these parasites in amphibians, birds or mammals. Results of frog necropsies at the Australian Registry of Wildlife Health revealed remarkable

A new myxozoan parasite *Cystodiscus axonis* inside the axons of the caudal brainstem of a captive Yellow spotted bell frog. The histological section is stained with the Luxol fast blue myelin stain and the Holmes silver nitrate stain for nerve fibers. Photo: Jan Šlapeta.

pathological findings in CNS associated with a newly discovered myxozoan parasite *Cystodiscus axonis* confirmed by DNA amplification and sequencing. Moreover, a retrospective examined of stored material of an Endangered Booroolong frog (*Litoria booroolongensis*) demonstrated presence of similar brain lesions and intralesional myxozoan parasites (*Cystodiscus axonis*). This parasite is characterized by its developmental stages found predominately within axons (Figure). The intraxonal location of the parasite is intriguing, because disruption of the neurological function could be disastrous for a tadpole or frog trying to evade predators. As of yet, we are unaware of any other infectious disease in frogs causing such marked brain pathology. Based on the remarkable findings we encourage other investigators to consider the potential role that myxozoan parasites play in wild and captive populations of declining frogs worldwide.

A. Hartigan, C. Sangster, K. Rose K, DN Phalen, J. Šlapeta, *Emerg. Infect. Dis.* **18,** 693 (2012). http://dx.doi.org/10.3201/eid1804.111606

Effects of herbicides and the chytrid fungus *Batrachochytrium dendrobatidis* on the health of post-metamorphic Northern leopard frogs (*Lithobates pipiens*).

By Linda J. Paetow, J. Daniel McLaughlin, Roger I. Cue, Bruce D. Pauli & David J. Marcogliese

Environmental contaminants and pathogens have been listed as two contributors to the global crisis of declining amphibian populations. It is likely that amphibian populations are exposed to these factors simultaneously, so it is important to assess their combined effects on amphibian health. Pesticides are common aquatic pollutants that can have significant immunomodulatory effects on amphibians. We therefore conducted an experiment to test the hypothesis that exposure to agricultural herbicides (atrazine or glyphosate) would increase the susceptibility of post-metamorphic Northern leopard frogs (*Lithobates pipiens*) to an important amphibian fungal pathogen, *Batrachochytrium dendrobatidis* (*Bd*). Initial exposure to the pesticides in a static renewal system for 21 days was followed by pulse exposure to *Bd*, and the independent and interactive effects of these exposures on the health and survival of the frogs were evaluated. Compared to controls, the glyphosate formulation reduced the snout-vent length of the frogs during the pesticide exposure (at day 21), and the atrazine formulation reduced gain in mass over the longer term (at day 94, when the experiment was terminated). However, there did not appear to be an influence of the pesticide exposure on the incidence of infection or the virulence of *Bd*, as no treatment affected survival or various other biomarkers of animal health or immune function. The observed effect of herbicide exposure on growth of the metamorphs is a concern, as similar effects of atrazine and glyphosate on amphibian growth have been observed previously, and they have been reported to affect population stability.

L. J. Paetow, J. D. McLaughlin, R. I. Cue, B. D. Pauli, D. J. Marcogliese, *Ecotoxicol. Environ. Saf.* **80**, 372 (2012). DOI: http://dx.doi.org/10.1016/j.ecoenv.2012.04.006 (linda.paetow@gmail.com)

Emerging fungal threats to animal, plant and ecosystem health

By Matthew C. Fisher, Daniel. A. Henk, Cheryl Briggs, John S. Brownstein, Lawrence C. Madoff, Sarah L. McCraw & Sarah J. Gurr

The last two decades have seen an increasing number of virulent infectious diseases in natural populations (including amphibians widely) and managed landscapes. An unprecedented number of fungal and fungal-like diseases, in both animals and plants, have been causing some of the most severe die-offs and extinctions ever witnessed in wild species and are further jeopardising food security. Human activity is intensifying fungal disease dispersal by modifying natural environments and so creating new opportunities for evolution. This article argues that nascent fungal infections will cause increasing attrition of biodiversity, with wider implications for human and ecosystem health, unless steps are taken to tighten biosecurity worldwide.

One of few surviving Southern mountain yellow-legged frogs (*Rana muscosa*) in Kings Canyon National Park, California, where chytrid fungus has all but wiped them out. Photo: Joel Sartore/joelsartore.com

M. C. Fisher, D. A. Henk, C. Briggs, J. S. Brownstein, L. Madoff, S. L. McCraw, S. Gurr, *Nature* **484**, 186 (2012).

Absence of infection with the amphibian chytrid fungus in the terrestrial Alpine salamander, *Salamandra atra*

By S. Lötters, J. Kielgast, M. Sztatecsny, N. Wagner, U. Schulte, P. Werner, D. Rödder, J. Dambach, T. Reissner, A. Hochkirch & B. R. Schmidt

A team of Austrian, German and Swiss herpetologists tested whether the Alpine salamander *Salamandra atra* is infected with the amphibian chytrid fungus (Bd). Swabs and qPCR was used to test for the presence of Bd on the salamander skin. 310 salamanders from nine localities across the species distribution range in Switzerland, Germany and Austria were tested for Bd. None was positive. Sample sizes were large enough to reliably infer the absence of Bd in this species. The appendix to the paper contains WinBUGS code that can be used to computer Bayesian 95% credible intervals.

S. Lötters *et al.*, *Salamandra* **48**, 58 (2012).

The ebb and flow of antimicrobial skin peptides defends Northern leopard frogs (*Rana pipiens*) against chytridiomycosis.

By James D. Pask, Douglas C. Woodhams, and Louise A. Rollins-Smith

Many amphibian populations and species are currently threatened by the emerging infectious disease, chytridiomycosis, caused by the chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*). Antimicrobial peptides (AMPs) secreted into the mucus of some amphibians are thought to be an important defense against chytridiomycosis. However, little is known about the quantities of AMPs secreted under natural conditions, whether they are sufficient to protect against this pathogen, and how they interact with commensal microbes. To understand how defensive peptides and skin microbes may interact, it is essential to know the precise quantities of AMPs present under natural conditions. Using matrix-assisted laser desorption time-of-flight (MALDI-TOF) mass spectrometry and growth inhibition

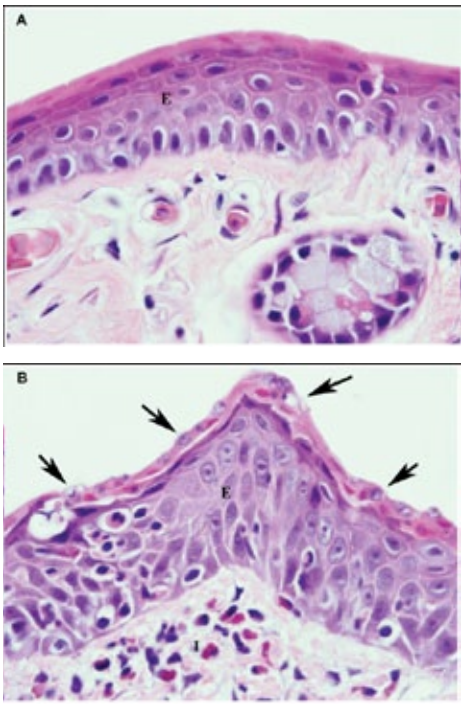
assays, we recently showed that resting Northern leopard frogs (*Rana pipiens*) constitutively release low amounts of AMPs that inhibit *Bd* in vitro, and AMP defenses are elevated following a simulated predator attack. Using a synthetic peptide analog of brevinin-1Pb as an external control, we quantified the amounts of four abundant AMPs (brevinin 1Pa, brevinin-1Pb, brevinin-1Pd, and ranatuerin-2P) at several time points after secretion. Once secreted onto the skin, the peptides were most active for fifteen minutes, and small quantities persisted for at least two hours. These data suggest that small amounts of AMPs are rapidly available and quite stable on the skin of *R. pipiens*. They are effective inhibitors of *Bd* at these low constitutive concentrations but degrade within two hours, protecting the integrity of the skin and commensal bacteria.

J. D. Pask, D. C. Woodhams, L. A. Rollins-Smith, *Global Change Biol.* **18**, 1231 (2012).

A reservoir species for the emerging amphibian pathogen *Batrachochytrium dendrobatidis* thrives in a landscape decimated by disease

By Natalie M.M. Reeder, Allan P. Pessier & Vance T. Vredenburg

Chytridiomycosis, a disease caused by the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*), is driving amphibian declines and extinctions in protected areas globally. The introduction of invasive reservoir species has been implicated in the spread of *Bd* but does not explain the appearance of the pathogen in remote protected areas. In the high elevation (>1500 m) Sierra Nevada of California, the native Pacific chorus frog, *Pseudacris regilla*, appears unaffected by chytridiomycosis while sympatric species experience catastrophic declines. We investigated whether *P. regilla* is a reservoir of *Bd* by comparing habitat occupancy before and after a major *Bd* outbreak and measuring infection in *P. regilla* in the field, monitoring susceptibility of *P. regilla* to *Bd* in the laboratory, examining tissues with histology to determine patterns of infection, and using a soak technique to determine individual output of *Bd* zoospores in water. *P. regilla* persists at 100% of sites where a sympatric species has been extirpated from 72% in synchrony with a wave of *Bd*. In the laboratory, *P. regilla* carried loads of *Bd* as much as an order of magnitude higher than loads found lethal to sympatric species. Histology shows heavy *Bd* infection in patchy areas next to normal skin, a possible mechanism for tolerance. The soak technique was 77.8% effective at detecting *Bd* in water and showed an average output of 68 zoospores per minute per individual. The results of this study suggest *P. regilla* should act as a *Bd* reservoir and provide evidence of a tolerance mechanism in a reservoir species.



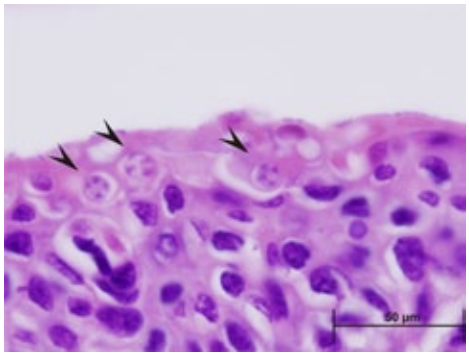
Histological views of the skin from a single infected but asymptomatic individual showing patchy distribution of *Bd* zoosporangia. Alan P. Pessier.

N. M.M. Reeder, A. P. Pessier, V. T. Vredenburg, *PLoSONE*, **7**, e33567. (2012) doi:10.1371/journal.pone.0033567.

First record of *Batrachochytrium dendrobatidis* in *Physalaemus fernandezae* (Anura: Leiuperidae) for Buenos Aires Province, Argentina

By Cristian S. Delgado, Guillermo S. Natale, Raúl A. Herrera & Diego A. Barrasso

Batrachochytrium dendrobatidis (*Bd*) was first reported for Argentina in 2002, after finding dead adult specimens of *Leptodactylus latrans* in locations from Buenos Aires Province. It was later registered in other provinces e.g. Córdoba, Misiones, Neuquén, San Luis, Salta and Tucumán. This study provides the first *Bd* infection record in a population of *Physalaemus fernandezae* from Punta Lara Natural Reserve (34.8033˚S, 58.0099˚W), Buenos Aires Province. Seven adult specimens were collected on September 2007; ventral skin samples were taken from each specimen, and the presence/absence of *Bd* was determined from histological studies. Cases of chytridiomycosis were found in 3 out of 7 samples. It is noteworthy that the individuals used in this study were sampled during reproductive activity. Moreover, despite numerous field studies carried out locally in the Reserve, no mortality episodes have been recorded for this species.



Histologic section of ventral epidermis of an adult specimen of *Physalaemus fernandezae* from Punta Lara Natural Reserve, Buenos Aires Province, Argentina. Arrows indicate zoosporangia of *Batrachochytrium dendrobatidis* with zoospores.

C. S. Delgado, G.S. Natale, R.A Herrera, D. A. Barrasso, *Herpetol. Rev.* **43**, 84 (2012).

Toxicity of the fungicide trifloxystrobin on tadpoles and its effect on fish-tadpole interaction

By Celina M. Junges, Paola M. Peltzer, Rafael C. Lajmanovich & Andrés M. Attademo

The use of pesticides in agriculture is increasing worldwide, and as a result, non-target aquatic animals in nearby ponds are increasingly exposed to pesticides through processes like runoff or through spray drift, direct overspray, atmospheric transport, and movement of animals through fields during application. Indeed, several studies have demonstrated their influence on predator-prey relationship and community structure. The purpose of our study was determined the toxicity of the fungicide trifloxystrobin (TFS) on four anuran tadpole species (*Rhinella arenarum*, *Physalaemus santafecinus*, *Leptodactylus latrans*, and *Elachistocleis bicolor*), and examine the effects of sublethal exposure to TFS on predation rates of tadpoles using eels (*Synbranchus marmoratus*) as fish predator under laboratory conditions. We also investigated whether eels preyed differentially on tadpoles exposed or not to TFS, and whether predation differed among anuran species. The results of our laboratory experiments suggest that the TFS was not equally toxic to the four tadpole species, *E. bicolor* being the most sensitive species, followed by *P. santafecinus*, *R. arenarum*, and *L. latrans*. Indeed, LC50 values of the most sensitive species were at least twice as high as those of the least sensitive species

(*E. bicolor* = 0.1 mg/L and *L. latrans* = 0.26 mg/L), indicating that larval species had differential sensitivity to TFS. On the other hand, predation rates were evaluated using different treatments that combined predator and prey exposed or not to this fungicide. TFS would alter the outcome of eel-tadpole interaction by reducing prey movements; thus, prey detection would decrease and therefore tadpole survival would increase. In addition, eels preyed selectively upon non-exposed tadpoles avoiding the exposed ones almost all throughout the period evaluated. Predation rate differed among prey species; such differences were not due to TFS exposure, but to interspecific differences in behaviour. The mechanism that would explain TFS-induced reduction in predation rates remains unclear; however, what is clear is that sublethal TFS concentrations have the potential to alter prey behaviour, thereby indirectly altering predator-prey interactions. In addition, we consider that predator–prey relationships are measurable responses of toxicant exposure and provide ecological insight into how contaminants modify predator–prey interactions.

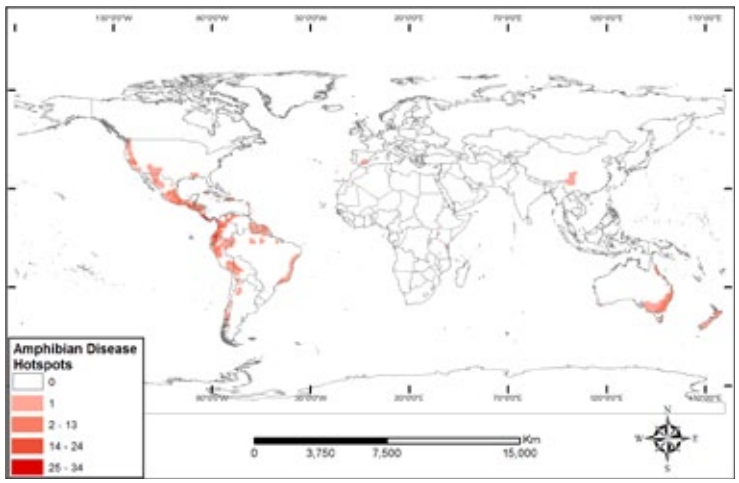
C. M. Junges, P.M. Peltzer, R.C. Lajmanovich, A.M. Attademo, M.C. Cabagna-Zenklusen, A. Basso, *Chemosphere.* **87,** 1348 (2012). http://anfibios-ecotox-conser.blogspot.es/

Ill nature: disease hotspots as threats to biodiversity

By Daniel Brito, Danielle O. Moreira, Bruno R. Coutinho & Monik Oprea

The disruption of natural disease dynamics is a serious side-effect of human actions that threatens biodiversity worldwide. We compiled range maps for species listed as threatened by disease for all the three globally assessed vertebrate taxa (amphibians, birds and mammals). Then we overlaid such maps to identify global disease hotspots. Amphibians are the group most affected by this threat, with 548 species subject to the negative effects of disrupted disease dynamics, mostly due to the invasive chytridiomicosis. We identified as disease hotspots for amphibians the regions of Mesoamerica, the Andes, the western coast of the USA, Mexico, the Guyana Shield, the Atlantic Forest, southeastern Australia and New Zealand. Amphibian species, not currently registered to suffer from diseases, but which geographic range overlaps with the disease hotspots, are expected to be more vulnerable to the onset of infections and/or epidemic events. This may drive threatened species closer to extinction and/or cause serious population declines in currently non-threatened species, increasing their chances to be listed in the future.

D. Brito, D. O. Moreira, B. R. Coutinho, M. Oprea, *J. Nat. Conserv.* **20**, 72 (2012).

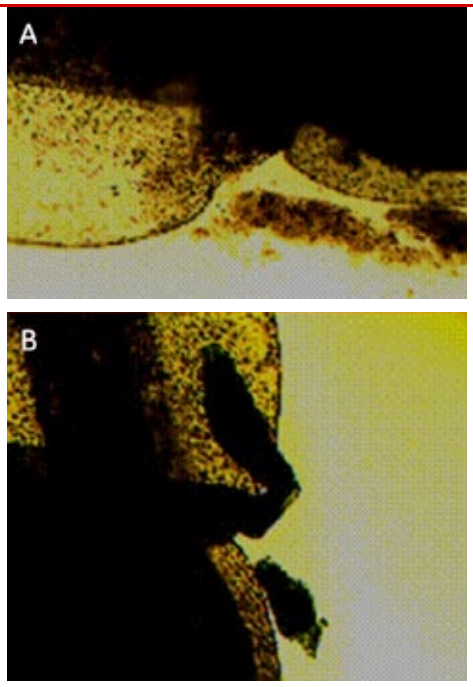


Global amphibian disease hotspots

Polyaniline nanofibers: Acute toxicity and teratogenic effect on *Rhinella arenarum* embryos.

By Edith I. Yslas, Luis E. Ibarra, Damián O. Peralta, César A. Barbero, Viviana A. Rivarola & Mabel L. Bertuzzi

Nanotechnology is a novel field that employs chemical techniques for the synthesis of extremely small molecules for their use in medicine, biomedical research, radiology, cosmetics, optics, fabrics and electronics. Due that these nanomaterials are also frequently used in high volumes in various industries and as such may enter in the environment is very important determine ecotoxicological effects. Given the uncertainty about their potential hazards, there is an immediate need for toxicity data and field studies to assess exposure to nanomaterials. The purpose of this study is to evaluate the toxicological potential of PANI nanofibers in the larvae *Rhinella arenarum* by means of AMPHITOX test. Acute toxicity of PANI nanofibers was evaluated in the premetamorphosis (stage 25) larvae. The exposure of *R. arenarum* larvae at dose of 150-400 mg L-1 resulted in 100% viability within 96 h exposure. Also, the teratogenic effects, was studies by means of an attractive alternative testing method for embryotoxicity/teratogenicity. In this assay, the embryos at 2-4 blastomers stage (early life stage teratogenic test) revealed that embryos were not killed and no teratogenic effects were observed when embryos were incubated with PANI nanofibers in concentrations of 150 and 250 mg L-1, while only a growth retardation of embryos was induced at levels of 250 mg PANI nanofibers L-1. While on the contrary at concentration of 400 mg L-1, a reduction in the body length of larvae and tail malformation was observed.



Optical micrographs of *Rhinella arenarum* larvae grown A with food fish and B with PANI nanofibers as the only carbon source.

These results suggest that a concentration-dependent teratogenic effect is operative, typified by phenotypes that had abnormal body axes and growth retardation of embryos. Also was observed the presence of PANI nanofibers in gut contents and its excretion by larval stages of *R. arenarum*. The results demonstrate the necessity to use a large variety of different tests for a comprehensive risk assessment. Therefore, these studies in embryonic and larval stage of *R. arenarum* could be included in a test system for nanomaterials toxicity determination. In the best of our knowledge, this is the first use of larvae in nanotoxicology.

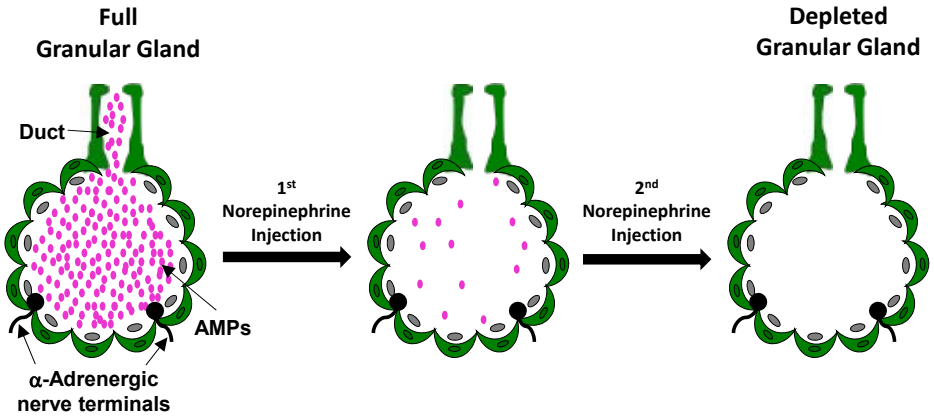
E. I. Yslas *et al.*, *Chemosphere* **87**, 1374 (2012).

Norepinephrine depletion of antimicrobial peptides from the skin glands of *Xenopus laevis*

By Whitney M. Gammill, J. Scott Fites & Louise A. Rollins-Smith

Our previous studies have suggested that the antimicrobial peptides (AMPs) produced in the dermal granular glands of many amphibian species play an important role in protection of these species from pathogens that infect amphibian skin. To further understand the role of AMPs in protection against *Batrachochytrium dendrobatidis*, the fungal pathogen that causes the potentially lethal skin disease chytridiomycosis, we developed a method to deplete skin peptides by norepinephrine induction of peptide secretion. Our model species, the South African clawed frog, *Xenopus laevis*, contains AMPs that are normally secreted following local nerve stimulation. Granular gland secretion can be stimulated in the laboratory by norepinephrine injection. Our studies showed that two injections of 80 nmol/g norepinephrine were necessary to fully deplete the AMP stores. One injection resulted in the secretion of most of the stored peptides. A second injection, two days later, released a small amount of additional AMPs that were not different in their composition from those released at the first injection. A third injection, four days after the first, resulted in no further AMP release. Hematoxylin and eosin staining confirmed that glands were depleted after two injections. Periodic acid-Schiff staining indicated that mucus gland secretion was also affected by norepinephrine.

W. M. Gammill, J. S. Fites, L. A. Rollins-Smith, *Dev. Comp. Immunol.* **37**, 19 (2012).



Graphical representation of amphibian granular gland with full complement of peptides and after one or two injections of norepinephrine to deplete the gland contents. [Original artwork of W. M. Gammill as published in Gammil *et al.,* 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.

Environmental persistence of amphibian and reptilian ranaviruses

By Jawad Nazir, Marisa Spengler & Rachel E. Marschang

Ranaviruses infect fish, amphibians, and reptiles. The present study was conducted to compare the persistence of amphibian and reptilian ranaviruses in the pond habitat. The four viruses used in this study included two amphibian ranaviruses: FV 3 (the type species of the genus *Ranavirus*) and an isolate from a frog and two ranaviruses of reptile origin (from a tortoise and from a gecko). A sandwich germ carrier technique was used to study the persistence of these viruses in sterile and unsterile pond water (PW) and soil obtained from the bank of a pond. For each virus, virus loaded carriers were placed in each of the three substrates and incubated at 4 and 20 °C and titrated at regular intervals. Serial data were analyzed by linear regression model to calculate T-90 values. Resistance of the viruses to drying was also studied.

All four viruses were resistant to drying. At 20 °C, T-90 values of the viruses were 22-31 days in sterile PW and 22-34 days in unsterile PW. Inactivation of all four viruses in soil at this temperature appeared to be non-linear. T-90 values at 4 °C were 102-182 days in sterile PW, 58-72 days in unsterile PW, and 30-48 days in soil. Viral persistence was highest in the sterile PW followed by the unsterile PW and the lowest in soil. There were no significant differences in the survival times between the amphibian and reptilian viruses. The results of the present study suggest that ranaviruses can survive for long periods of time in pond habitats at low temperatures.

J. Nazir, M. Spengler, R. E. Marschang, *Dis. Aquat. Org.* **98**, 177 (2012), doi: 10.3354/dao02443.

FrogLog Schedule

January - South America
March - Europe, North Africa and West Asia
May - North and Central America and the Caribbean
July - Sub Saharan Africa
September - Mainland Asia
November - Maritime Southeast Asia and Oceania

Robin Moore / iLCP

AmphibiaWeb Recent Publication List

This reference list is compiled by Professor Tim Halliday (formerly DAPTF International Director; tim.r.halliday@gmail.com). It lists papers on amphibian declines and their causes and amphibian conservation, with an emphasis on those that describe methods for monitoring and conserving amphibian populations. Tim is always delighted to receive details of forthcoming papers from their authors.

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Wildenhues, M. *et al.* (2011) Husbandry, captive breeding, larval development and stages of the Malayan horned frog *Megophrys nasuta* (Schlegel, 1858) (Amphibia: Anura: Megophryidae). *Amphibian & Reptile Conservation* **5:** 15-28. (marlen.wildenhues@gmx.de)

Wu, C.-S. *et al.* (2012) Irreversibility of a bad start: Early exposure to osmotic stress limits growth and adaptive developmental plasticity. *Oecologia* **169:** 15-22. (biyckam@thu.edu.tw)

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FrogLog Schedule

January - South America
March - Europe, North Africa and West Asia
May - North and Central America and the Caribbean
July - Sub Saharan Africa
September - Mainland Asia
November - Maritime Southeast Asia and Oceania

General Announcements

Upcoming Meetings & Workshops

May

28 — 31. African Amphibian Working Group, Trento, ITALY.

June

2 — 4. Fifth Asian Herpetological Conference, Chengdu, Sichuan Province in The Peoples Republic of China.

July

24 — 26. Northeast PARC Annual Meeting, Highlands Center, Scenic White Mountains Region, Crawford Notch, NH.

August

8 — 14. 7th World Congress of Herpetology (WCH), Vancouver.

16 — 19. 10th Annual Symposium on the Conservation and Biology of Tortoises and Freshwater Turtles, Tucson, Arizona.

September

6 — 15. IUCN World Conservation Congress, Jeju, Korea.

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

MS Assistantship - Clemson University - Wetland Herpetofauna Research. Clemson, SC (5/8/12).

Desert Tortoise Telemetry Technician. Las Vegas, NV (5/3/12).

Gopher Tortoise Seasonal Field Technician. Southern GA (4/25/12).

Copperhead Research Assistant - Ball State University. South-Central Indiana (4/22/12).

Herpetofaunal Research Intern - Alabama A&M University. Bankhead National Forest, northern AL (4/21/12).

Research Associate - Florida Fish and Wildlife Conservation Commission. Gainesville, FL (4/18/12).

Research Manager - Brown Treesnake Research. Guam (4/11/12).

Volunteer Field Assistants - Ornate Tree Lizard Research. Southeast, AZ (4/3/12).

MS or PhD Graduate Research Assistantship - Oregon State University. Corvallus, OR (4/3/12).

Biological Technician - Reptile and Amphibian Research. Snake River Field Station, Biose, ID (4/1/12).

Zoo Keeper - Herpetology - Houston Zoo. Houston, TX (3/30/12).

Further volunteer positions can be seen on page XX

Keep In Touch

If you would like to be added to the ASG mailing list, please send an email to froglog@amphibians.org with the subject heading “add me to mailing list.”
Also, follow us on Facebook for regular updates on the herpetological conservation community and the latest news from the ASG.
http://www.facebook.com/AmphibiansDotOrg

Funding Opportunities

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

May 2012

African Network of Scientific and Technical Institutions (ANSTI) - ANSTI/DAAD Post-Graduate Fellowships. The German Academic Exchange Service (DAAD) cooperates with ANSTI by offering financial support for Masters and Ph.D degrees at institutions in Sub-Saharan Africa. The fellowships are awarded to nationals in Sub-Saharan Africa for studies outside the applicants' home countries. Participants must be from ANSTI member institutions, and be less than 36 years old at the time of application. The application deadline is 31 May 2012. **Link**

French Foundation for Research on Biodiversity - CESAB Call for Research Proposals 2012. CESAB (CEnter for Synthesis and Analysis on Biodiversity) makes grants to working groups that develop syntheses of ideas and concepts, and/or data analyses, to improve understanding of biodiversity. Working groups should address a limited number of specific questions; be international in their composition; and be led by a well-recognized scientist belonging to a French scientific organization. Proposals from natural and social sciences are welcome. Interdisciplinary groups and mixed groups linked to stakeholders are particularly welcome. Working groups will be funded for a maximum of €190 thousand for three years. The deadline for pre-registration is 25 May 2012. Link

Global Development Network (GDN) - Global Research Competition 2012. The GDN invites researchers from developing and transition countries to submit research proposals for the 2012 Global Research Competition. The themes in 2012 - which is the pilot year of this program - include food security (among others). The research competition is inter-regional and multi-disciplinary. Proposals can be submitted individually, or in teams. An online platform will help individuals and incomplete teams to find collaborators. Each winning team will be paired with world-class mentors. Grants are up to US$32 thousand for projects of up to 18 months. The submission deadline is 21 May 2012. **Link**

Zoological Society of London – Call for EDGE Fellows 2012. ZSL's EDGE of Existence Program provides fellowships for two-year projects in research and conservation of EDGE (Evolutionarily Distinct and Globally Endangered) mammals, amphibians, and corals. Applicants must be early-career conservation biologists (less than 5 years experience) who are nationals or residents of the country in which the EDGE species occur. The Fellowship includes a grant of up to £5 thousand per year, plus training and technical support. Applicants are strongly advised to discuss their projects with the EDGE team before applying. The application deadline is 31 May 2012. **Link**

June 2012

African Enterprise Challenge Fund - Support in Post-Conflict Countries. The AECF offers grants and loans to support businesses in Africa working in agriculture, financial services, renewable energy, and technologies for adapting to climate change. AECF's Post Conflict Window is seeking proposals from Dem Rep Congo, Liberia, Sierra Leone, and Somalia/Somalialand. The deadline for applications is 15 June 2012. **Link**

l'Agence inter-établissements de recherche pour le développement (AIRD) - AIRD-CIRAD Doctoral Program. France's AIRD, in collaboration with CIRAD, will fund up to 15 doctoral degrees for agricultural researchers from developing countries. The support is for agriculture in the broad sense, i.e., including agriculture, environment, natural resources, etc. The research should be carried out in partnership with CIRAD. Applicants should be from inter-tropical and Mediterranean countries, and under 40 years of age. Applications (French, English) are due before 18 June 2012. **Link**

Both ENDS - JWH Initiative to Promote Leadership of Young Environmentalists. The Joke Waller-Hunter Initiative offers grants to advance the leadership and learning of junior persons working for - or affiliated with - environmental civil society organizations in developing and emerging countries. JWH especially encourages the nomination of young women and local community leaders. Grants can be applied for a wide range of activities, but candidates should have a clear idea of how they intend to use the grant. The grants range from €2,500 to €10,000 each, and 15 grants are offered twice a year. The nomination deadlines are 01 June and 01 December. **Link**

Crawford Fund - Australian Support for Asia-Pacific Research in Agriculture, Fisheries, Forestry. The Crawford Fund Fellowship supports a scientist in agriculture, fisheries, or forestry from the Asia-Pacific countries for additional training in Australia. The Fellowship is offered annually to an agricultural scientist below the age of 35 years who is a citizen of (or who is working in) one the following countries: Bangladesh, Bhutan, Burma, Cambodia, East Timor, Fiji, Indonesia, Laos, Nepal, Niue, Papua New Guinea, Samoa, Solomon Islands, Tonga, Tuvalu and Kirabati, Vanuatu, and Vietnam. The closing date for applications is 29 June 2012. **Link**

CRDF Global - Grants for Environmental Technologies in Azerbaijan. CRDF Global, in partnership with Azercell Telecom and the Barama Innovation Center, is sponsoring a business plan competition in Azerbaijan to encourage new technology-based businesses in ICT and green (environmental) technologies. The program is open to students, researchers, and technology entrepreneurs who are legal residents of Azerbaijan. With funding from the U.S. Department of State, CRDF will make four grants ranging from US$10 thousand to US$20 thousand for the best submissions. The application deadline is 24 June 2012. **Link**

European Union - Central European Initiative (CEI), Cooperation Activities 2012. The CEI makes grants of up to €20 thousand to support its action plan. Cooperation projects are mainly for seminars, workshops, training courses, and other kinds of meetings. Priority themes include climate, environment, and sustainable energy; and sustainable agriculture. Projects can be submitted by all public and private bodies in CEI's member states, as well as by international and regional organizations. Preference is for proposals submitted from CEI's non-EU member states (Albania, Belarus, Bosnia and Herzegovina, Croatia, Macedonia, Moldova, Montenegro, Serbia, and Ukraine). The application deadline is 30 June 2012. **Link**

European Union - Central European Initiative (CEI), Know-How Exchange 2012. The CEI's Know-How Exchange Program (KEP) connects knowledge providers in some EU countries with recipient partners in Albania, Belarus, Bosnia and Herzegovina, Macedonia, Moldova, Montenegro, Serbia, and Ukraine. Applicants are organizations in the CEI which are also members of the EU. Priority themes in

the exchange program include agriculture, energy, and environment (among others). Grants are up to €40 thousand. The application deadline is 30 June 2012. **Link**

French Ministry of Foreign and European Affairs - Research Cooperation France-Indonesia 2013. The French Ministry of Foreign Affairs, in partnership with the French Ministry of Higher Education and Research, announce "Nusantara 2013" to promote research cooperation between France and Indonesia. Focus areas include environmental protection, marine science, prevention of natural disasters, renewable energy, and several others. Contact information (in each of France and Indonesia) is provided in the announcement. The deadline for proposals is 15 June 2012. **Link**

French Ministry of Foreign and European Affairs - Biodiversity and Natural Products in Asia. The French government announces its third call in the program Bio-Asie to foster French-Asian collaboration in research and development of natural products that may have applications in medicine, cosmetics, agriculture, renewable energy, etc. Each funded project should include at least one French team combined with partners in two or more eligible Asian countries: Brunei, Cambodia, China, India, Indonesia, Laos, Japan, Malaysia, Myanmar, Pakistan, Philippines, Singapore, South Korea, Taiwan, Thailand, Vietnam. Grants will range from €23 thousand to €40 thousand for projects of two years. The application deadline is 25 June 2012. **Link**

French National Research Agency (ANR) - Adaptation to Environmental Changes in the Mediterranean Sea. France's ANR will fund planning to bring together potential research partners to address environmental issues of the Mediterranean Sea in relation to fisheries, ecosystem risks, impacts of marine transport, and others. The ANR prefers a consortium approach and North-South inclusion. A planning grant (ARP) can range from €50 thousand to €200 thousand for 18 months. The deadline for proposals is 29 June 2012. **Link**

Higher Education for Development (HED) - Conservation in the Andean Amazon. HED, in cooperation with the U.S. Agency for International Development, calls for applications for the second phase of the "Initiative for Conservation in the Andean Amazon" (ICAA II) in Bolivia, Colombia, Ecuador, and Peru. HED anticipates making one award per country for initiatives in higher education that promote the conservation of Andean-Amazon biodiversity. Each award is up to US$750 thousand over three years. The program is open to U.S. colleges and universities that apply individually or in partnership with other institutions. Partnerships are encouraged. The closing date for applications is 28 June 2012. **Link**

Humane Earth Foundation - Environment, Biodiversity, and Sustainable Agriculture in Europe. The Foundation calls for project proposals in environment, biodiversity, and sustainable agriculture. Priority is for grants to small organizations in Western Europe, but also including some grant making in the Balkan countries and Eastern Europe. The next deadline is 04 June 2012. **Link**

Japan International Research Center for Agricultural Sciences - International Awards for Young Agricultural Researchers 2012. JIRCAS makes awards of US$5 thousand each for up to three young agricultural researchers in developing countries who contribute to outstanding research and development in agriculture, forestry, fisheries and related themes. Candidates need to be less than age 40. The deadline for applications is 29 June 2012. **Link**

Save Our Species - Second SOS Call for Proposals. SOS announces its second call for Threatened Species Grants. Priority is for projects that focus on critically endangered and endangered species in the following groups: threatened tropical terrestrial Asian vertebrates; threatened small marine mammals; threatened cycads; and threatened freshwater African animals. Grants will be made to civil society organizations for field projects. International NGOs are eligible if they work closely with national stakeholders, local communities, and with the required authorizations from governments. The average grant size is expected to be about US$100 thousand. The deadline for proposals is 22 June 2012. **Link**

Social Science Research Council - African Peace Building Network, Research Grants. The Social Science Research Council (SSRC) makes research grants to African researchers, policy analysts, and practitioners working on conflict and peace building in Africa. Support is available for research and analysis on issues that include environmental change and conflict - among other themes. Applicants must be African citizens currently residing in a country of Sub-Saharan Africa. Other eligibility criteria are explained in the grants announcement. SSRC will make up to fifteen grants, each up to a maximum of US$15 thousand. Applications deadline is 15 June 2012. **Link**

Toyota Motor Corporation - Toyota Environmental Activities Grants Program 2012. Toyota makes grants to support environmental activities implemented by nonprofit organizations on environmental themes that vary from year to year. The current themes are "Biodiversity Conservation" and "Global Warming Countermeasures." Grants are made in Japan and overseas (i.e., internationally). The international program is open to applicants in Japan, and international partners in collaboration with Japanese groups. The deadline for applications is 18 June 2012. **Link**

U.S. Agency for International Development - Livelihoods and Management of Arid Lands in Kenya. USAID-Kenya will fund one or more organizations for a program in Kenya, "Resilience and Economic Growth in the Arid Lands - Improving Resilience" (REGAL-IR). The 5-year program is funded at US$49 million to address diversification of livelihood opportunities; community management of natural resources; improving market access; disaster risk reduction; and improving nutritional outcomes. Eligibility for grants is unrestricted. Funding Opportunity SOL-623-12-000008. The closing date for applications is 06 June 2012. **Link**

U.S. Agency for International Development - Supporting Forests and Biodiversity in Cambodia. USAID-Cambodia invites proposals from non-governmental organizations (U.S. and non-U.S.), and/or a consortia, to mitigate climate change and conserve biodiversity in Cambodia. The program aims to reduce greenhouse gas emissions from forests and land use, such as by aiding Cambodia to participate in the framework for REDD+. The level of funding is US$20 million for up to four years. Funding Opportunity USAID-CAMBODIA-442-12-00001-RFA. The closing date for applications is 06 June 2012. **Link**

U.S. Agency for International Development - Vietnam Forest and Deltas. USAID-Vietnam invites proposals from non-governmental organizations (U.S. and non-U.S.), and/or a consortia, to provide technical support to accelerate Vietnam's transition to low-emissions development. The objectives of the program are to reduce net greenhouse gas emissions from forests, and to enhance the resiliency of people, places, and livelihoods in Vietnam's delta regions to short and long-term climate impacts. Program funding will be up to US$30 million for up to five years. Funding Opportunity 440-12-023. Closing date for applications is 15 June 2012. **Link**

U.S. Department of State - International Dialogue on Dams and Water Infrastructure. The Bureau of Oceans and International Environmental and Scientific Affairs (OES) at the U.S. State Department will coordinate a global dialogue on multi-purpose infrastructure and surrounding natural infrastructure at the nexus of water, food, and energy for the developing countries. The OES will fund one or a consortium of nonprofit and non-governmental organizations, or international organizations, to establish and implement the "Nexus Dialogue on Dams and Water Infrastructure Optimization." Funding is up to US$297 thousand for up to two years. Funding Opportunity OES-OWA-11-001. The closing date for applications is 04 June 2012. **Link**

World Agroforestry Center - Ann Stroud Memorial Scholarship for Agroforestry Research in East Africa. The World Agroforestry Center (ICRAF) offers a scholarship to postgraduate students in East Africa to conduct thesis research in collaboration with ICRAF. Applicants should be nationals of East Africa who are 35 years of age or less, and involved in agroforestry research, education, or development. The scholarship is tenable at any university in East Africa. The upper limit of support is US$5 thousand. Applications are due at ICRAF before 30 June 2012. **Link**

July 2012

African-Eurasian Waterbird Agreement - Small Grants 2012. AEWA makes grants to support waterbird conservation in African countries of the African-Eurasian Flyways. Grants are up to €25 thousand for projects of up to two years in AEWA countries, and up to €15 thousand for projects in countries of non-contracting parties. The program is open to government organizations and NGOs. The application deadline is 27 July 2012. **Link**

Belmont Forum - International Opportunities Fund. The International Opportunities Fund will provide about €20 million to support global research on (i) coastal vulnerability, and (ii) freshwater security. The countries in this initiative are Australia, Brazil, Canada, France, Germany, India, Japan, Russia, South Africa, UK, and USA. Some of these countries may also support participation from developing countries. Consortia will consist of partners from at least three participating countries - and bring together natural scientists, social scientists, and research users. Projects will be funded at €1 million to €2 million for two or three years. The deadline for pre-proposals is 20 July 2012. **Link**

Netherlands Organization for Cooperation in Higher Education (NUFFIC) - MENA Scholarships 2012. The government of the Netherlands funds the MENA Scholarship Program to contribute to capacity building in seven countries of the Middle East and North Africa: Algeria, Iraq, Lebanon,Morocco, Oman, Syria, and Tunisia. The grants are to mid-career professionals who are employed by organizations and institutions in professional fields that include water management; agriculture, forestry, and fisheries; and geographic information science (among others). Each year, the program aims to support about 120 professionals, half of whom are women. The application deadline is 02 July 2012. **Link**

Regional Fund for Agricultural Technology-AgriculturalInnovations in Latin America. The Regional Fund for Agricultural Technology (Fondo Regional de Tecnología Agropecuaria, FONTAGRO) promotes agricultural research and technology in Latin America and the Caribbean. In 2012, FONTAGRO invites case studies of successful innovations in small-scale agriculture in the region. Awards will be made for leading examples in the private sector (i.e., producers associations, processing and marketing organizations, NGOs); national R&D organizations and universities; and international organizations. FONTAGRO will award prizes ranging from US$5 thousand to US$10 thousand, plus travel to Washington DC for the presentations. Submissions are due no later than 16 July 2012. **Link**

Society of Environmental Journalists - Grants for Environmental Journalism 2012. The Society of Environmental Journalists (SEJ) makes grants for environmental journalism projects and entrepreneurial ventures. Journalists working independently, or on the staffs of either for-profit or nonprofit news organizations, are eligible to apply. There are no nationality restrictions. Funding is up to US$3,500 per grant. The application deadlines are 15 July 2012 and 15 November 2012. **Link**

UNESCO with L'Oreal Corporate Foundation - Young Women in Life Sciences 2012. The UN's Educational, Scientific, and Cultural Organization (UNESCO) and L'Oreal co-sponsor the annual "Fellowships for Young Women in Life Sciences." A maximum of three young women (under age 35), from each of five geo-cultural regions of the world, are awarded research grants in biology, biotechnology, agriculture, and other life sciences. Special attention is given to applications from the least-developed countries. The initial fellowship is for up to 12 months, for a maximum sum of US$20 thousand. The closing date for applications is 15 July 2012. **Link**

August 2012

U.S. National Science Foundation - International Research Experiences (IRES) for Students 2012. The NSF supports U.S. scientists and engineers in the early stages of their careers 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 21 August 2012. **Link**

Zayed International Prize for the Environment - Sixth Cycle. The Zayed International Prize for the Environment offers cash prizes to recognize and promote pioneering contributions in environment and sustainable development. There are three categories: (i) Global leadership - US$500 thousand; (ii) Scientific and technological achievements - US$300 thousand; and (iii) Environmental action leading to positive changes in society - US$200 thousand. The prizes are open to individuals and organizations worldwide. The nomination deadline for the Sixth Cycle of the Prize is 31 August 2012. **Link**

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 bimonthly 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 - South America
- March - Europe, North Africa and West Asia
- May - North and Central America and the Caribbean
- July - Sub Saharan Africa
- September - Mainland Asia
- November - Maritime Southeast Asia 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/resources/asg-members/. If for some reason this cannot be done, contributions will be reviewed by at least one individual within the ASG. FrogLog is not a peer reviewed publication and the onus for submitting accurate information remains with the authors.

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James P. Lewis (jplewis@amphibians.org)

Editorial Committee

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Additional reviewers will be requested as require

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 James Lewis at jplewis@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

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

Books

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

Technical reports

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

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

Published Online Only

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

Web site

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

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

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Coming up in FrogLog103

Sub Saharan Africa

Updates from Sub Saharan Africa

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Grants

and much more...

July 2012

Photo: James Lewis