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# FrogLog

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

**REGIONAL EDITION:  
ASIA, RUSSIA AND OCEANIA**

**Salamanders Lost, Salamanders Found,  
Salamanders Saved**

**Do All Threatened Amphibians Belong  
on the Ark?**

**Mapping the Malabar Tree Toad**

**And Much More!**

A calling male Malabar Tree Toad. Photo: Gururaja KV.



# FrogLog

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# Editorial

Dear FrogLoggers,

This final edition of *FrogLog* for 2015 truly does meet two of the broad goals laid out for this magazine. Firstly, this edition shares so many exciting updates that illustrate the incredible progress being made on all fronts of amphibian conservation. You will discover how two rare salamander species that were lost to science for nearly 40 years were not only recently rediscovered, but how the Amphibian Survival Alliance and a consortium of international groups protected some of their last remaining habitat just in the nick of time. You will also read how ASG Chile lead the update for the extinction risk for Chilean amphibians and how this has highlighted a need for local herpetologists to generate data on population ecology that will contribute to the conservation of these species. In addition to this, you will discover how art is not just increasing the public's awareness of the plight of amphibians, but is also highlighting the opportunities that exist for the public to make a difference for amphibians. And on the disease front, you will learn about the launch of the new Global Ranavirus Reporting System, a model for future infectious disease reporting and biosurveillance.

Secondly, this edition highlights some of the key questions that all of us—as amphibian conservationists, researchers, educators and enthusiasts alike—should be asking both ourselves and the community. What are some of the effects of targeted habitat protection on the extinction risk of Threatened amphibians? It's been a decade since the Global Amphibian Assessment so how have the world's zoos responded? When it comes to amphibian conservation breeding programs, do all Threatened amphibians belong on the Ark? What can Lazarus Toads tell us about amphibian conservation? What role can institutional internships play in developing the amphibian husbandry capacity of a country like Madagascar? How can we bridge the gaps between scientists and citizens? Can changing garden management practices help reduce amphibian diseases?

As you flip through the pages of *FrogLog* you will see that we have lots of updates and lots of questions. We hope you enjoy this edition and thank you for making 2015 a fantastic year for amphibian conservations all around the world and let's work together to make 2016 even better!

*Candace M. Hansen-Hendrikx*  
*Editor-in-Chief*

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A Long-limbed Salamander (*Nyctanolis pernix*) spotted by Jeremy Jackson 38 years after he last saw the species. Photo: Robin Moore.

## Salamanders Lost, Salamanders Found, Salamanders Saved

**T**wo rare salamander species lost to science for nearly 40 years have not only been recently rediscovered, but the Amphibian Survival Alliance and a consortium of international groups has protected some of the last remaining forest home of the salamanders just in the nick of time.

Critical habitat of the Finca Chiblac Salamander (*Bradytriton silus*) and the Long-limbed Salamander (*Nyctanolis pernix*) in Guatemala's Cuchumatanes mountain range had been slated for imminent clearing for coffee production.

"The Cuchumatanes mountains are as beautiful as they are diverse—from azure waters of secluded lakes to mist-shrouded forests, their magic is enhanced by the rediscovery of long-lost species and the promise of finding more. To protect this area is to realize the vision of Carlos Vasquez, Marco Cerezo and other Guatemalans who have worked tirelessly towards another inspiring conservation success," said Robin Moore, conservation officer for the Amphibian Survival Alliance.

In the 1970s, a young Paul Elias and Jeremy Jackson entered the cloud forests of the Cuchumatanes and discovered two entirely new salamander species, the Finca Chiblac and Long-limbed Salamanders. These two turned out to be missing links that tied together the evolutionary tree of New World tropical salamanders. The salamanders went unseen for more than three decades. Then, in 2014, Carlos Vasquez, coordinator of the amphibian conservation

program at FUNDAECO, led an international team of scientists that included Elias and Jackson to the site where he had rediscovered them more than 30 years later.

"To see this reserve take shape under the imaginative genius of Carlos Vasquez and partners, and to be able to help that happen in a small way, is the culmination of a forty year dream for me," Elias said. "This incredibly biodiverse cloud forest on the oldest moun-



The Finca Chiblac Salamander, *Bolitoglossa silus*, is a monotypic species found only in the Cuchumatanes mountains. Discovered in 1977, the species went undetected for over three decades before being rediscovered by Carlos Vasquez. Photo: Hussain Aga Khan.

The Cuchumatan Golden Toad, *Incilius aurarius*, from the Cuchumatanes mountains of Guatemala was discovered and described as recently as 2012. Photo: Robin Moore.



tain block in Central America is home to the missing-link species and genera we found there all those years ago. Many other species of great interest have already been found and many more will follow at this exceptional site.”

Along with the Amphibian Survival Alliance, a number of organizations came together to purchase the property, called [Finca San Isidro](#), before it could be cleared for coffee production later this year. Those groups are FUNDAECO, World Land Trust, Global Wildlife Conservation, Rainforest Trust and the International Conservation Fund of Canada. One of the property’s parcels will be named after philanthropist Andrew Sabin, who has supported the conservation of amphibians in Guatemala and worldwide.

“The establishment of the San Isidro Amphibian Reserve as the first Nature Reserve in the Western Highlands of Guatemala is a great conservation success,” said Marco Cerezo, executive director of FUNDAECO, the local NGO that helped identify the 2,000-acre parcel of land and will oversee management of the property. “It marks the beginning of a regional effort to support the protection of forests in the northwest of Guatemala, a region of exceptional biodiversity. Thanks to all our partners that came together to create this sanctuary for unique and endangered amphibians.”

Finca San Isidro is home to a treasure trove of amphibian species, including the recently discovered Cuchumatan Golden Toad (*Incilius aurarius*) and the beautiful Black-eyed Treefrog (*Agalychnis moreletii*). Elias and Jackson discovered Jackson’s Climbing Salamander (*Bolitoglossa jacksoni*) within a few hundred meters of the reserve. The amphibian has evaded detection for 38 years, making it one of the world’s [Top 10 “Most Wanted” Amphibians](#). Ten of the 20 amphibian species that live in or near Finca San Isidro are classified as Critically Endangered or Endangered by the IUCN Red List. The remoteness of the Cuchumatanes mountain range has protected much of the forest to date, but increasing pressures from the coffee industry put these forests at risk. Local and international scientists and conservationists have identified the area as one of the highest priorities for immediate conservation action.



The Black-eyed Leaf frog, *Agalychnis moreletii*, is a Critically Endangered frog that breeds in the pools protected in the new reserve. Photo: Robin Moore.



# ASG Chile Leads Update of the Extinction Risk of Chilean Amphibians for The IUCN Red List of Threatened Species<sup>TM</sup>

By <sup>1</sup>Claudio Soto-Azat, <sup>1,2</sup>Andrés Valenzuela-Sánchez, <sup>3</sup>Juan Carlos Ortiz, <sup>4</sup>Helen Díaz-Páez, <sup>5</sup>Camila Castro, <sup>5</sup>Andrés Charrier, <sup>5</sup>Claudio Correa, <sup>6,7</sup>César Cuevas, <sup>8</sup>Gabriel Lobos, <sup>9,10</sup>Marco A. Mendez, <sup>11</sup>Mario Penna, <sup>1</sup>Alexandra Peñafiel-Ricaurte, <sup>12</sup>Felipe Rabanal, <sup>13</sup>Claudia M. Vélez-R, <sup>14</sup>Marcela A. Vidal & <sup>15</sup>Ariadne Angulo.

**T**welve years have elapsed since the first workshop aimed at assessing the extinction risk of Chilean amphibians occurred, with their resulting classification being published in The IUCN Red List of Threatened Species<sup>TM</sup> (Universidad de Concepción, October 2003). Since then, new and different lines of research have contributed additional knowledge for this taxonomic group (1–4). Much of this work has comprised of updates on distribution ranges, identification of relevant threats, evaluation of taxonomic identity and studies on ecological features for several anuran species inhabiting Chile (salamanders and caecilians are not present in Chile). There are also newly described endemic species that have increased the number of species for the country. Given this and the fact that IUCN Red List assessments have a shelf life of ten years or less, it was imperative to make efforts to update the conservation status of Chilean amphibians so that a valid list was current and useful.

Organized by the Chilean branch of the IUCN SSC Amphibian Specialist Group (ASG), an assessment process to update the conservation status of Chilean amphibians for The IUCN Red List of Threatened Species<sup>TM</sup> was initiated in May 2015. The assessment process followed three steps: 1) compilation of existing published information, 2) a public consultation period to collect any relevant information needed for the extinction risk assessment, and 3) an expert workshop facilitated by two experienced Specialist Group Chairs and the participation of 19 local herpetologists belonging to different institutions, including academia, NGOs and government agencies (Fig. 1). The workshop was hosted by the Universidad Andres Bello (UNAB), Santiago, Chile, on 9–10 July 2015. The event was funded by the Outreach Scheme of the Dirección de Extensión Académica-UNAB, with additional funding kindly provided by the IUCN Species Survival Commission.

At the time of writing this note, the assessments were being prepared for the next step in the assessment process, an external review on the application of the IUCN Red List methodology based on existing documentation. Once this process is completed, species profiles, along with their range maps, will be submitted for publication on the IUCN Red List of Threatened Species<sup>TM</sup>. Although there is a possibility that changes could arise from the review process, workshop results are summarized here.

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Fig. 1: Herpetologists at the workshop, Universidad Andres Bello. From left to right in upper row: Mario Penna, Edgardo Flores, Juan Carlos Ortiz, Claudia Vélez, Camila Castro, Felipe Rabanal, Claudio Correa, Marco Mendez, Sandra Díaz, Mariella Superina, Reinaldo Avilés, César Cuevas, Claudio Soto, Andrés Charrier and Ariadne Angulo. Lower row: Andrés Valenzuela and Charif Tala. Absent from the picture: Marcela Vidal, Helen Diaz and Alexandra Peñafiel. Photo: Claudio Soto-Azat.

Sixty-one species were assessed, encompassing 97% of the native anuran species previously identified for the country (5). One assessment concluded that *Telmatobius peruvianus* is not present in Chile (although it is still present in Peru), as previous records from Putre in northern Chile, have been assigned to *T. marmoratus* based on phylogenetic analyses (6). Also, the recently revalidated *Telmatobius laevis* (7), was not assessed at the workshop given that no new data exist for this species since its original description in 1902 and therefore to date no natural population can be assigned to this species. From the species evaluated, 72% were identified as endemic to Chile. If species having marginal distribution in Argentina are included, this percentage increases to 90%. In addition, four species were assessed for the first time; three recently discovered species: *Alsodes cantillanensis* (8), *Eupsophus altor* (9) and *Telmatobufo ignotus* (10); and one taxonomic re-validation: *Alsodes coppingeri* (11).

Changes in the number of species within each conservation category are shown in Fig. 2. The percentage of Threatened species (*i.e.*, Critically Endangered [CR], Endangered [EN] and Vulnerable [VU]) increased from 38 to 47%. This increase is explained by the addition into the Threatened categories of recently discovered or previously described Data Deficient (DD) species (Fig. 2C). It is important to note that DD does not mean that the species is not of conservation concern, but appropriate data on its distribution and/or population is needed to make a consistent assessment of its risk of extinction. Almost half of the assessed species (45%) experienced a change in category. Seven DD species changed their category as follows: Least Concern (LC; one species), VU (2 spp.), EN (1 sp.) and CR (3 spp.). The species that changed from DD to CR were *Telmatobius dankoi*, *T. frontiensis* and *T. vilamensis*, all

from northern Chile (Fig. 3). When DD species are not considered, changes in categories involve six up-listings and 12 down-listings. Among the latter, only two species were removed from a Threatened status. Two CR species were considered as Possibly Extinct (a tag used in conjunction with the CR category to describe those instances where there is a possibility a species may be extinct): *Rhinoderma rufum* and *Telmatobius pefauri*. In fact, *R. rufum* has not been recorded since 1980, despite numerous attempts to find it (12,13). The reasons for the sudden decline of this species are not fully understood, but the extensive habitat loss across its historical distribution and possibly chytridiomycosis could have played an important role (13). On the other hand, *Telmatobius pefauri* is only known from its holotype, collected in 1976 at the locality of Murruntani in northern Chile (14). This species has not been observed since, in spite of attempts to find it.

The main threats identified for Chilean amphibians (3,4,15) are: a) water scarcity due to anthropogenic modification of natural systems, b) mining activities in northern and central Chile; c) impacts related to agriculture, d) residential development in central and southern Chile, e) exotic tree plantations, and f) anthropogenic fires in southern Chile, which cause loss of habitat and refuges. In addition, livestock pressure and invasive species (particularly sal-



Fig. 3: *Telmatobius vilamensis* from northern Chile. Previously assessed as Data Deficient and now proposed as Critically Endangered as a result of the Chilean IUCN Red List workshop. Photo: Felipe Rabanal.

monids and the African Clawed Frog *Xenopus laevis*) were cited as threats to amphibians across the country.

Following IUCN criteria for assessing species in one of the Threatened categories, most (76%) of the amphibians were categorized by their restricted geographic range (criteria B1 and B2). Another 10% were assessed according to a population size reduction (criteria A) and 14% following very small or restricted populations (criteria C). This overview highlights an imperative necessity for local herpetologists to conduct studies and generate data on population ecology, helping make assessment more informative in the future, with the final purpose of contributing to the conservation of these species.

#### Acknowledgments:

We are very grateful to all those herpetologists who made possible this fruitful assessment process. Special thanks to Dr. Mariella Superina, Chair of the IUCN SSC Anteater, Sloth and Armadillo Specialist Group, who acted as expert facilitator at the workshop. We would also wish to extend our acknowledgements to all the people who supported the assessment process, providing valuable information through the open consultation period, and to the IUCN SSC Amphibian Red List Authority and IUCN Red List Unit for processing these assessments. We believe the workshop results will be an important contribution to the conservation of these amazing animals.

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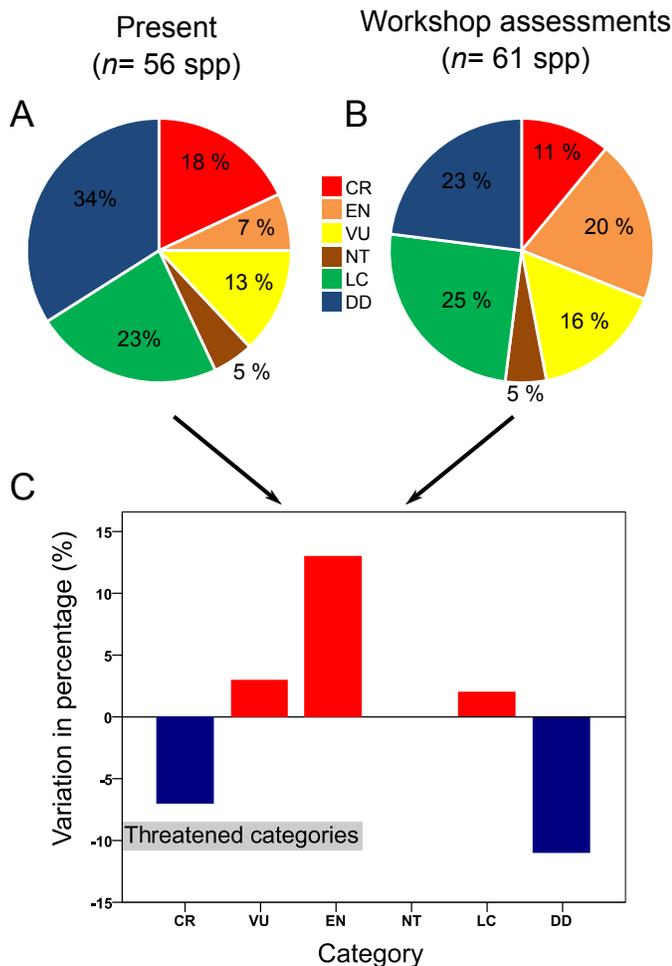


Fig. 2: Percentage of Chilean amphibian species within each conservation category at A) present (IUCN Red List assessments, accessed on 20 August 2015), and B) under the new proposed assessments. The changes in these percentages per category are presented in C), where negative values (blue) indicate a decrease in the percentage of species within the category, while the positive values (red) indicates an increase. CR: Critically Endangered; VU: Vulnerable; EN: Endangered; NT: Near Threatened; LC: Least Concern; DD: Data Deficient.

# Targeted Habitat Protection and its Effects on the Extinction Risk of Threatened Amphibians

By Justin Nowakowski & Ariadne Angulo

The first comprehensive Global Amphibian Assessment (GAA), published in 2004, helped shed light on the scope of the amphibian decline phenomenon and elevated awareness of amphibians as a group in need of targeted conservation intervention. One of the key findings of the GAA was that habitat loss and fragmentation are the most pervasive threats to amphibians globally (1). Informed by the GAA, several multi-stakeholder initiatives have focused on habitat protection with the express purpose of conserving Threatened amphibian species. As a result, new conservation areas (areas protected through various designations, such as private and forest reserves or community stewardship projects) have been established in regions with high concentrations of Threatened amphibians in Guatemala, Colombia, Madagascar, and Sri Lanka. Extinction risk assessments can act as valuable baselines for evaluating and monitoring the long-term success of the new reserves. On the tenth anniversary of the GAA, the Amphibian Specialist Group (ASG) updated extinction risk assessments for 32 Threatened amphibians that occur at these sites, drawing on information about the status of species and conservation areas provided by local biologists and conservation organizations.

In Guatemala, rapid deforestation threatens the Caribbean region of the country, recognized for its diverse fauna that includes an unusually large number of endemic and Threatened amphibian species. Local NGO, Fundación para el Ecodesarrollo y la Conservación (FUNDAECO), along with multiple international organizations, established the Sierra Caral Amphibian Conservation Reserve on the slopes of the Sierra Caral mountains that run along the border with Honduras. The core area of the reserve, which was instrumental for the subsequent expansion of the Reserva Hídrica y Forestal Sierra Caral, encompasses 2,400 ha of wet tropical forest etched with a network of streams and rivers (2)(Table 1). The new protected area harbors nearly a dozen Threatened amphibian species; nine of these species were recently reassessed and include the Endangered Copan Brook Frog, *Duellmanohyla soralia* and Dunn's climbing salamander, *Bolitoglossa dunni*. Sierra Caral not only serves as a key site for local species conservation, but sits between several other protected areas, such as the Copán and Cusuco National Parks in Honduras. Therefore, the protected status will help maintain connectivity among these parks, which are nested within the Mesoamerican Corridor. Acquisition of the land and the subsequent designation of Sierra Caral as a national protected area



*Duellmanohyla soralia*. Photo: Robin Moore.

by Guatemala's congress represents a landmark success for biodiversity conservation in Guatemala and the region. The long-term efficacy of the Sierra Caral reserve, however, depends on balancing the protection of habitat within core areas, with sustainable land use by surrounding communities.

Colombia is a center of remarkable amphibian species richness, second only to Brazil in total number of native species (3). The ASG reassessed the extinction risk of thirteen species that occur in recently created reserves protecting small and medium-sized tracts of megadiverse subtropical forest in the Colombian Andes. These new sites add to a network of 25 reserves that are managed by the Colombian conservation organization ProAves (4). El Dorado Reserve helps conserve a locally-important watershed in the Sierra Nevada de Santa Marta and is home to 14 endemic amphibian species (Table 1). Among these amphibians, several are now listed as Endangered on The IUCN Red List of Threatened Species, such as the harlequin frogs *Atelopus laetissimus* and *Atelopus nahumae*. Farther south, the Ranita Dorada Amphibian Reserve has been lauded as the world's first amphibian-focused conservation area with the express objective of protecting nine Threatened amphibian species occurring within the small forest remnant. In 2006 and 2007, Colombian herpetologists described two new brightly-colored dendrobatid species, *Andinobates dorisswansonae* and *Andinobates tolimense*, thought to only occur within the isolated forest fragment. Located in a re-

Table 1. New conservation areas in four countries providing habitat protection for Threatened amphibians.

Conservation area	Country	Designation	Year established	Area (ha)	Recognized as AZE site?	No. Threatened amphibians present	Founding partners
Sierra Caral	Guatemala	National forest reserve	2012	2,447	Yes	12	1,5,6,7,8, 12,13
El Dorado	Colombia	Private reserve	2006	1024	Yes	5	2, 5, 12
Ranita Dorada	Colombia	Private reserve	2008	120	-	9	2,5,11
La Forzosa	Colombia	Private reserve	2006	1324	Yes	11 <sup>a</sup>	2, 5
Roncesvalles	Colombia	Private reserve	2009	4072	-	≥2	2, 5
Morningside	Sri Lanka	National forest reserve*	-	1000	-	13	3, 5, 6, 7, 9
Fohisokina	Madagascar	Community stewardship	2009	300	-	≥1	4, 5, 6, 10

<sup>1</sup>FUNDAECO, <sup>2</sup>ProAves, <sup>3</sup>Sri Lanka Forest Department, <sup>4</sup>FOMISAME, <sup>5</sup>IUCN SSC Amphibian Specialist Group, <sup>6</sup>Conservation International, <sup>7</sup>International Conservation

Fund of Canada, <sup>8</sup>Global Wildlife Conservation, <sup>9</sup>Wildlife Heritage Trust, <sup>10</sup>Man and the Environment, <sup>11</sup>Dendrobatidae Nederland, <sup>12</sup>American Bird Conservancy, <sup>13</sup>Rainforest Trust,

\*The ownership of this site has transferred to the Sri Lankan Forest Department, but it is awaiting official designation as a reserve

<sup>a</sup>Number of Threatened species known from the immediate area, but not necessarily recorded inside the reserve



*Atelopus nahumae*. Photo: Fundacion ProAves.

gion heavily transformed by coffee production and cattle ranching, the forest was in immediate danger of being cleared, which prompted the rapid purchase of the land through partnerships between ProAves, IUCN Netherlands, Conservation International and Dendrobatidae Nederland.

The IUCN Red List assessments also included species from the La Forzosa reserve located in the cordillera central of Colombia near the town of Anori. La Forzosa was established in large part to preserve habitat for an Endangered bird, the Arrierito Antioqueño, but also extends crucial habitat protection to at least five Threatened amphibian species in the area. In the Andean highlands of the department of Tolima, a mosaic of forest patches and páramo are protected within the Roncesvalles reserve and are home to the



*Atelopus laetissimus*. Photo: Fundacion ProAves.

Critically Endangered harlequin toad, *Atelopus simulatus* and the Endangered Herveo Plump Toad, *Osornophryne percrassa*. ProAves is currently working with local land owners to establish reforestation projects and manage existing forests on private lands near these smaller reserves in an effort to increase connectivity among vital patches of remnant habitat.

On the other side of the globe, 70% of Sri Lanka's amphibian fauna is Threatened with extinction or already extinct, driven largely by clearing of vast swaths of forest for housing and agricultural production (3). Less than 5% of the country's original cloud forest remains and is, therefore, in urgent need of protection. The ASG, along with Conservation International, the International Conservation Fund of Canada, and local partners, including Wildlife Heritage Trust and the Forest Department of Sri Lanka, prioritized protection of an important tract of remaining cloud forest known as Morningside Forest, located adjacent to the Sinharaja World Heritage Site in southeastern Sri Lanka. The ASG re-evaluated the status of nine species of Endangered or Critically Endangered frogs occurring within Morningside. Most of the Threatened species belong to the genus *Pseudophilautus*, a group characterized by direct development, sensitivity to habitat alteration and micro-endemism. Throughout Sri Lanka, 19 of the 67 *Pseudophilautus* species known from the country are considered Extinct, often having been recorded from only a single locality. Morningside Forest is now managed by the Forest Department of Sri Lanka, a crucial step towards conserving this highly threatened amphibian fauna. Current priorities for the site include working with private land owners to increase sustainability of tea and cardamom production on inholdings and on neighboring properties.



*Andinobates doriswainsonae*. Photo: Fundacion ProAves.

Madagascar represents another important hotspot of amphibian species richness, home to numerous Threatened and endemic species that are found nowhere else (3). One of these species, the Endangered *Mantella cowanii*, was the focus of efforts to protect critical savannah and stream habitat at a highland site in Antoetra, Madagascar. In previous decades, the black and orange-colored *M. cowanii* had declined in abundance as a result of widespread habitat loss, frequent brush fires, and overharvesting for the illegal pet trade (5). Through a concerted public awareness campaign, ASG Madagascar, Conservation International and the community organization, FOMISAME, generated local support for protecting an important area of *M. cowanii* habitat (6). FOMISAME has worked to promote ecotourism at the site and to establish sustainable plantations and aquaculture to help relieve pressure on core habitat. Now, as a result of capacity building, local community members patrol and manage the site. Regular monitoring of *M. cowanii* indicates that population densities have increased at Antoetra since its designation as a reserve (7). The project exemplifies the value of pairing habitat protection with information campaigns and capacity building as well as the potential for community stewardship of Threatened habitats and species.

Following the securing of the new conservation areas (achieved at different times over the last decade), the ASG has prioritized reassessment of Threatened species occurring at these sites. Notably, of the 32 reassessed Threatened amphibians that had received habitat protection, none was reassessed at a higher threat



Deforestation in the Sierra Caral of Guatemala. Photo: Robin Moore.

category as a result of genuine change in species status. In fact, two dendrobatid frogs, *A. doriswainsonae* and *A. tolimensis*, were down-listed from Critically Endangered and Endangered, respectively, to Vulnerable, representing a genuine change in their status resulting from the protection of habitat within the Ranita Dorada Forest Reserve. Many of the species changed categories and/or criteria largely as a result of new information on range size or, conversely, a lack of information on population trends or status. The latter scenario highlights a common and crucial need for increased survey efforts to fill large information gaps concerning the distribution and populations status of rare and endemic species, species most likely to be threatened with extinction. The vast majority of species were reassessed under criteria B1ab(iii), reflecting restricted extent of occurrence, severely fragmented populations or reduced number of threat-defined locations, and continuing decline in the extent or quality of habitat.

Because of the new habitat protection, the amphibians within the borders of conservation areas are likely better off than they were ten years ago, and they have an advantage over the 50% of all range-restricted amphibian species that currently do not occur within any protected area (8). These efforts represent important amphibian conservation success stories; they demonstrate that, with multi-stakeholder commitment and engagement and diverse habitat protection approaches, it is possible to advance amphibian conservation in the context of very different political and cultural settings. Not only are many described species afforded habitat protection, but it is likely that new species will soon be discovered in these areas of extraordinary endemism. As some of the first conservation areas established, in part, to safeguard Threatened amphibian assemblages, the conservation community will focus attention toward these sites as valuable models that will inform future amphibian conservation action.



Deforestation in Santa Marta, Colombia. Photo: Robin Moore.

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# The Disappearing Frogs Project Leaps into Action to Fund Amphibian Conservation Seed Grants

By Candace M. Hansen-Hendriks, Pam Hopkins & Terry Thirion

In 2015 the Disappearing Frogs Project partnered with the Amphibian Survival Alliance to raise awareness of global amphibian declines, inspire people to take personal action to protect these incredible species, while also providing a unique opportunity for artists to support amphibian conservation, education and research.

Created in 2013 by Charlotte, North Carolina-based artist Terry Thirion, the Disappearing Frogs Project concept brings synergy between artists and scientists to the public, communicating the unprecedented global amphibian decline and potential effects of species extinction. Awareness in the community is being raised, hearts of the public are being touched, and the Disappearing Frogs Project is inspiring people to get involved and to take personal action.

Engaging the senses our exhibitions allow art and science to intersect in a non-threatening way. While being surrounded visibly by the beauty and unique works of art one is also beckoned to listen to the multidisciplinary scientists invited to present real-time research communicating complex ideas as it relates to species loss in a form that is understandable to the public. These dedicated individuals passionately convey the critical issues affecting amphibians globally thus substantiating the efforts of the DFP to raise awareness and to challenge the public to be better stewards of our environment.

Together, the core strengths of the Disappearing Frogs Project and the Amphibian Survival Alliance offers unique synergies that not only help to increase the public's awareness of the plight of amphibians, but most importantly highlight the opportunities that exist for the public to help conserve amphibians and make a difference on the ground.

"By partnering with the Alliance, the Disappearing Frogs Project is truly having a global impact for amphibians around the world. They are helping to protect habitat, support research and engage people around the amazing world of amphibians," said James Lewis, director of operations with the Amphibian Survival Alliance.

Amphibian Survival Alliance seed grants are often seen as a vital funding source to kick start projects and encourage innovative approaches to addressing the amphibian conservation issues of today.



"The work that these grassroots biologists are doing is so vital to the survival of our amphibians and knowing that simultaneously artists are making a difference working in their studios painting, drawing and sculpting makes me smile" said Terry Thirion, founder and artistic director with the Disappearing Frogs Project.

The following three Amphibian Survival Alliance seed grants awardees were funded with support from the Disappearing Frogs Project:

- Sex in the lab: Using a new technique to facilitate breeding in Tree Frogs;
- Returning from the brink: Rebounding amphibian populations in a pathogen enzootic environment; and
- Conservation status of *Telmatobius intermedius* and other amphibians in the Pampa Galeras National Reserve—Barbara D'Achille, Ayacucho, Peru.

"Amphibians are at the intersection of our arts and science exhibitions. It's gratifying to know the effort we put into these events can be directly invested in the dedicated scientists working to enhance the viability of frogs globally. Supporting seed grants gives the Disappearing Frogs Project a unique way to be intimately involved with real-time conservation and research efforts. For example, we are thrilled to offer financial support for the "Sex in the Lab" project because they too use ART to rescue species of amphibians that are experiencing declines and facing extinction. Art comes in many forms. In this case ART—Assisted Reproductive Technologies—represents a technique which uses hormones to stimulate egg and sperm production to aid in breeding," said Pam Hopkins, regional director of communications with the Disappearing Frogs Project.

The Alliance has secured limited funding for this initiative but we are looking to work with other groups to develop collaborative Seed Grants similar to this initiative with the Disappearing Frogs Project. These types of vital projects would not be possible without this type of incredible support and illustrate that successful amphibian conservation outcomes don't always require a substantial investment - a little bit goes a long way. If you are interested in learning more or would like to explore ways in which your organization can get involved with it, please [contact Candace M. Hansen-Hendriks](#), director of communications and partnerships with the Amphibian Survival Alliance.



Pam Hopkins and Terry Thirion at the 2015 Disappearing Frogs Project in Rock Hill, South Carolina. Photo: Robert Fitzpatrick.

# Sex in the Lab: Using a New Technique to Facilitate Breeding in Tree Frogs

By Leah Jacobs

Over the past 30–100 years amphibians have experienced worldwide population declines. With a staggering 32% of the Earth's amphibian species facing imminent decline, the use of captive breeding has emerged to prevent extinction. One method, assisted reproductive technologies (ART), uses hormones to stimulate egg and sperm production to aid in breeding and has been successfully used for years in mammals, fishes, and birds, with more recent applications in frogs. In light of the current amphibian extinction crisis, ART is recognized as an increasingly important avenue for rescuing the 100s of species of amphibians that are experiencing declines and face extinction.

The treefrogs of the Neotropical subfamily Phyllomedusinae, are a large group of colorful leaf frogs distributed throughout Central and South America. I will test two hormone doses to examine which is most effective at inducing spermiation (males) and ovulation (females) among Red-eyed Treefrogs. Previous research using ART indicates that hormone protocols are expected to be similar for closely related species. Although our focal species are not currently declining, making them ideal for large-scale, manipulative experiments, many of their closest relatives are listed on the International Union for the Conservation of Nature (IUCN) Red List as imperiled.



Red-eyed Treefrog. Photo: Leah Jacobs.

Thus, my research will have a direct impact on informing captive breeding and management and the conservation and preservation of 8 imperiled species of leaf frog.

Each individual will be injected with either zero, two or four ug/g body weight GnRH. For males, I will examine the viability of each sperm sample by imaging sperm stained with sperm staining dye (propidium iodide and SYBR 14), under florescent microscopy, allowing for a count of live vs. dead sperm in each sample (see image). I will evaluate spermic urine from each individual every three, seven, 12, 24 hours to produce a sperm response curve for viability and production of each individual. Females will be injected via the same protocol. Comparison of the viability and production of sperm produced by each male, and egg clutch size by each female will be evaluated to compare for each individual dose.

Amphibians are facing an extinction rate (32% of the 7,405 described species) that is greater than that of either birds (12%) or mammals (22%). Since 1980 it is estimated that almost 170 species of amphibians are extinct. Thus, with my research I hope to develop techniques on this species that will give us a clearer understanding of hormone doses and breeding techniques needed to conserve and breed other members of the group (a subfamily consisting of five genera and 59 species, 8 of which are listed as Endangered or Critically Endangered).

*This project has been supported by the Amphibian Survival Alliance's Seed Grant Program, along with a contribution to the program made by the Disappearing Frogs Project. If you would like to further support this project or invest in the Amphibian Survival Alliance's Seed Grant program as the Disappearing Frogs Project did, please contact Candace M. Hansen-Hendrikx, Director of Communications & Partnerships.*



Leah Jacobs, Master's student, California State University, Northridge, holding one of her Red-eyed Treefrogs. Photo: Leah Jacobs.

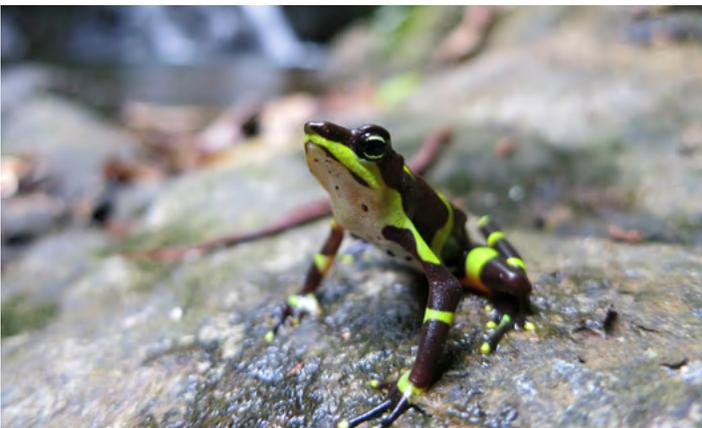


## Returning From the Brink: Rebounding Amphibian Populations in a Pathogen Enzootic Environment

By Alexander Shepack

**A**mphibian declines and disappearances have long captured our attention, but in recent years there have been numerous reports of once “lost” amphibians being rediscovered. Many of these amphibians are from populations or species that are believed to have declined due to *Batrachochytrium dendrobatidis* (*Bd*).

Populations from groups like *Atelopus* or stream-breeding Hyliidae, that were known to be heavily affected by the *Bd* epidemic, have now begun to rebound or become more easily detectable in certain areas. Our focal species (*Rhinella margaritifera*, *Atelopus varius*, *A. limosus*, *Duellmanohyla uranochroa*, *Lithobates vibicarius*)



*Atelopus limosus*. Photo: Alexander Shepack.

all come from areas where they were known to have declined and where *Bd* is present.

With this study we hope to increase understanding of how time since decline, infection dynamics, genetics and changing susceptibility play a role in these rebounding populations. We are using environmental DNA analyses to find additional field sites by screening for genetic material of focal species. Sites are being surveyed to assess the dynamics within rebounding populations and the prevalence of *Bd* in these regions. These populations will be assessed using restriction enzyme associated DNA sequencing (RAD-Seq) techniques to improve analyses of the genetic structure of these populations. Finally, *Bd* susceptibility trials will be conducted to understand rapid adaptation and change in the host-pathogen system.

By increasing our understanding of how these populations have been able to return with *Bd* still present in the environment we can hopefully aid in the conservation of species still affected by *Bd* or similar pathogens in other areas.

*This project has been supported by the Amphibian Survival Alliance's Seed Grant Program, along with contribution to the program made by the Disappearing Frogs Project. If you would like to further support this project or invest in the Amphibian Survival Alliance's Seed Grant program as the Disappearing Frogs Project did, please contact [Candace M. Hansen-Hendrikx](#), Director of Communications & Partnerships.*



Photo: Mauricio Sebastián Akmentins.

## Where is Calilegua's Marsupial Frog?

By Mauricio Sebastián Akmentins

This project is part of a conservation program that is focused on the long-term preservation of the three species of marsupial frogs in the Yungas Andean forest of Northwestern Argentina. Of particular concern is determining the conservation status of Calilegua's Marsupial Frog (*Gastrotheca christiani*) due not only to the sudden lack of registries in the wild since 1996, but also due to evidence of a severe population decline. The Endangered

*Gastrotheca christiani* is in the 250 "lost frogs" list of the IUCN SSC Amphibian Specialist Group. This project aims to determine if the Calilegua's Marsupial Frog is still extant as a keystone for any future conservation effort.

We will perform field search campaigns in the Calilegua National Park and surrounding localities to obtain new registries (photographic/call recordings) of the missing Calilegua's Marsupial Frog. These search efforts, employing visual and aural encounter surveys combined with playbacks at fixed point, will be complemented with the training park rangers of Calilegua National Park in amphibian species recognition and monitoring techniques. In the near future we hope to receive further funding to incorporate passive monitoring techniques with automated recording devices (frogloggers) to increase the detection probability. Moreover, frogloggers may also be very effective as a long term monitoring tool in case of rediscovering the Calilegua's Marsupial Frog.

*This project has been supported by the Amphibian Survival Alliance's Seed Grant Program. If you would like to further support this project or invest in the Amphibian Survival Alliance's Seed Grant program please contact [Candace M. Hansen-Hendrikx](#), Director of Communications & Partnerships.*



One of the last specimens registered of *Gastrotheca christiani* in 1996 near Calilegua National Park, Jujuy province, Argentina. Photo: Marcos Vaira.



# ACRS

Amphibian Conservation Research Symposium

*Sharing research and strategies to empower  
the future of amphibian conservation*

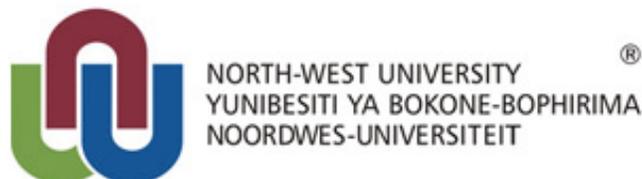
Potchefstroom, South Africa  
18<sup>th</sup> – 21<sup>st</sup> January 2016

Optional field trip to Pilanesberg  
National Park: 22<sup>nd</sup>- 23<sup>rd</sup> January

For ongoing updates please see  
<http://www.amphibians.org/acrs/>

Email: [acrs@amphibians.org](mailto:acrs@amphibians.org)

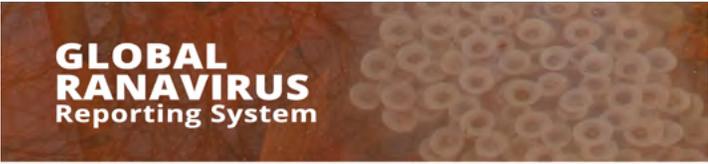
Or sign-up to the ASA mailing list at:  
<http://www.amphibians.org/subscribe/>



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## The Global Ranavirus Reporting System is LIVE!



### GLOBAL RANAVIRUS Reporting System

**W**e are pleased to announce the release of the Global Ranavirus Reporting System (GRRS, <https://mantle.oi/grrs>). The GRRS was built using the EcoHealth Alliance's Mantle platform in consultation with scientists from the US Forest Service, the Global Ranavirus Consortium and EcoHealth. The GRRS is an open-source web platform designed for the storage, sharing, and visualization of world Ranavirus surveillance data, including diagnostics and genetic isolate differences, and is designed to meet the needs of a wide variety of users inclusive of natural resource managers and researchers. Ranavirus scientists in the field or the lab will be able to upload datasets in multiple formats to the system, where they will be stored for easy download and analysis. GRRS users have fine-grained access controls to protect and share their uploaded datasets, and examine datasets in views appropriate to their content (e.g., tables, maps and charts). The GRRS will rapidly advance the scientific community's understanding of ranavirus epidemiology, and help natural resource agencies and other organizations respond intelligently to new outbreaks. The GRRS

will become a model for future infectious disease reporting and biosurveillance.

### QUOTES FROM GRRS BETATESTERS:

"Ranaviruses can have severe impacts on amphibians at the community level and the GRRS provides a great tool to implement better recording of surveillance effort. GRRS has the potential to provide a stronger link between research and wildlife management." Dr. Stephen Price, post-doctoral research associate, University College London.

"Ranavirus is a global problem, much like malaria or AIDS. Mapping its distribution will help preserve biodiversity." Dr. David Lesbarrères, Associate Professor, Department of Biology, Laurentian University.

"The Global Ranavirus Reporting System fills a critical gap in ranavirus research by providing a user friendly platform for data entry and extraction that will be invaluable for researchers and managers seeking to understand ranavirus epidemiology at multiple scales." Dr. Jason Hoverman, Assistant Professor, Department of Forestry and Natural Resources, Purdue University.

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## The Third International Symposium on Ranaviruses

By <sup>1</sup>Patricia Johnson & <sup>2</sup>Amanda Duffus

**R**anaviruses are globally distributed emerging pathogens of lower vertebrates (1). Members of this group of pathogens have been linked to amphibian declines (e.g., 2 and 3), as well as, countless morbidity and mortality events (1).

Following the First International Symposium on Ranaviruses in 2011, the Global Ranavirus Consortium (GRC) was formed. The GRC is an international organization made up of researchers, scientists, managers and veterinarians, and others whose goals include the facilitation of communication and collaboration in the *Ranavirus* research community ([www.ranavirus.org](http://www.ranavirus.org)). In 2013, the Second International Symposium on Ranaviruses was held in Knoxville, TN, USA concurrently with the Wildlife Disease Association's International Meeting. Planning began soon thereafter for the Third International Symposium on Ranaviruses. (Information on previous Ranavirus Symposia can be found at [www.ranavirus.org/symposium](http://www.ranavirus.org/symposium)).

The Third International Symposium on Ranaviruses was held on May 30 – June 2, 2015 at the Hilton – University of Florida, in Gainesville, Florida, USA. Over 70 people from nine different countries attended. The symposium consisted of two days of talks, with over 30 presentations given from experts in all areas of Ranavirus biology. Professor Dr. Richard Whittington (University of Sydney Australia) opened the meeting with an expansive keynote address. During the meeting, attendees had ample opportunities to discuss

urgent research needs during sessions facilitated by leading experts in Ecology, Stressors, and Surveillance, Pathology and Diagnostics, Virology and Immunology, and Evolution, Phylogenetics and Taxonomy. (Session summaries will be available soon at [www.ranavirus.org/symposium](http://www.ranavirus.org/symposium)).

Attendees also took part in a wide variety of workshops and field trips. Importantly, trainings were provided on the design of *Ranavirus* surveillance studies and data analysis (lead by Drs. Gray and Brunner) and sterile sample collection from ranavirus hosts and molecular diagnostics (lead by Drs. Miller and Hick), were held at the Emerging Pathogens Institute at the University of Florida. These workshops provided attendees with an overview of how to properly design disease surveillance studies, collect sterile samples from carcasses and gave them an introduction to the molecular diagnostic methods used to detect *Ranavirus*.

More information on the Third International Symposium on Ranaviruses can be found at [www.ranavirus.org/symposia](http://www.ranavirus.org/symposia)

Once finalized, dates and locations of the 2017 and 2019 symposia will be announced on the GRC website ([www.ranavirus.org](http://www.ranavirus.org)).

### References:

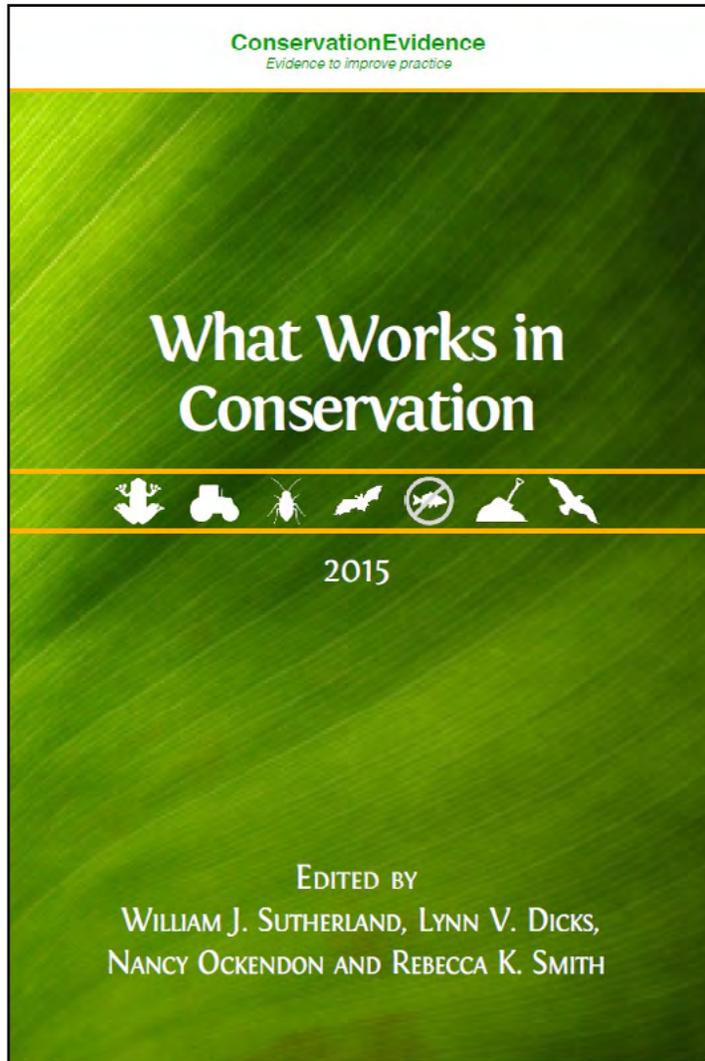
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2. Teacher *et al.* *Anim. Conserv.* **13**, 514 (2010).
3. Price *et al.* *Curr. Biol.* **24**, 2586 (2014).

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<sup>1</sup>Certified Reptile Monitor and Licensed Turtle Rehabilitator, New York, USA; Email: [turtleadvocate@gmail.com](mailto:turtleadvocate@gmail.com) <sup>2</sup>Department of Biology, Gordon State College, Barnesville, Georgia, USA; Email: [aduffus@gordonstate.edu](mailto:aduffus@gordonstate.edu)

# What Works in Conservation

By Rebecca Smith



**A**re you working to conserve a particular group of species or habitat and would like to know the range of different interventions you could carry out? Would you like to know whether those interventions have been tested and have been found to be effective (or not)?

*What Works in Conservation* aims to help answer these questions. The newly published book provides an assessment, by panels of experts, of the effectiveness of a wide range of conservation actions based on the available scientific evidence. These include 98 interventions for the conservation of amphibian populations, ranging from creating ponds to releasing captive bred animals. As well as amphibians, there are chapters covering birds, bats, bees, biodiversity in European farmland and control of invasive freshwater species. The full evidence on which the assessments are based is described in the associated *Conservation Evidence* synopses, including: *Amphibian Conservation – Global evidence for the effects of interventions (1)*. *What Works in Conservation* provides key messages from the summarized evidence and the assessment of effectiveness for each intervention and is freely available, along with all synopses, at [www.conservationevidence.com](http://www.conservationevidence.com).

*What Works in Conservation* is designed for anyone who has to make decisions about how best to support or conserve biodiversity. This includes land managers, conservationists, farmers, campaigners, advisors or consultants, policymakers, researchers or people taking action to protect local wildlife. The resource aims to support decision-making by assessing what evidence there is (or is not) about the effects that your planned actions could have on the target group of species or habitat. It is designed as a starting point and before making any final decisions about implementing interventions it is important that you read the more detailed summarized evidence online at [www.conservationevidence.com](http://www.conservationevidence.com), to assess their relevance for your specific study species or system.

Summarized evidence for 98 conservation interventions within the Amphibian Conservation synopsis was assessed by a panel of 28 amphibian experts. Each intervention was then assigned to a category of effectiveness based on a combination of the assessment of the size of the benefit and/or harm, and the strength of the evidence. An example of a table of effectiveness from *What Works in Conservation*, for interventions for amphibians to mitigate against the threats from transportation and service corridors, is shown below. Each intervention is linked to the summarized evidence online.

An example table from *What Works in Conservation* (Sutherland et al. 2015).

Based on the collated evidence, what is the current assessment of the effectiveness of interventions for amphibians for mitigating the threat of transportation/service corridors?	
<b>Beneficial</b>	
<b>Likely to be beneficial</b>	<ul style="list-style-type: none"> <li>• Close roads during seasonal amphibian migration</li> <li>• Modification of gully pots and kerbs</li> </ul>
<b>Trade-off between benefit and harms</b>	<ul style="list-style-type: none"> <li>• Install barrier fencing along roads</li> <li>• Install culverts or tunnels as road crossings</li> </ul>
<b>Unknown effectiveness (limited evidence)</b>	<ul style="list-style-type: none"> <li>• Signage to warn motorists</li> </ul>
<b>Unlikely to be beneficial</b>	<ul style="list-style-type: none"> <li>• Use humans to assist migrating amphibians across roads</li> </ul>
<b>Likely to be ineffective or harmful</b>	
<b>No evidence found</b>	

For 31 of the 129 interventions listed within the Amphibian Conservation synopsis, we did not find any published evidence of effectiveness and so the interventions could not be assessed. Despite our search effort it is possible that some evidence was missed, but it is also likely that the effects of many conservation projects have not been monitored, or results have not been made widely available. We need you to help this change for the better! Details for submitting case studies on the effects of conservation interventions to the journal *Conservation Evidence* can be found on our website: <http://conservationevidence.com/collection/view>.

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1. Sutherland, W.J., Dicks, L.V. Ockendon, N. and Smith, R.K. (2015) *What Works in Conservation*. Cambridge, UK: Open Book Publishers. [www.conservationevidence.com/](http://www.conservationevidence.com/)

# An Update from the Global Ranavirus Consortium

By <sup>1</sup>Amanda L. J. Duffus, <sup>2</sup>Jesse L. Brunner & <sup>3</sup>Matthew J. Gray

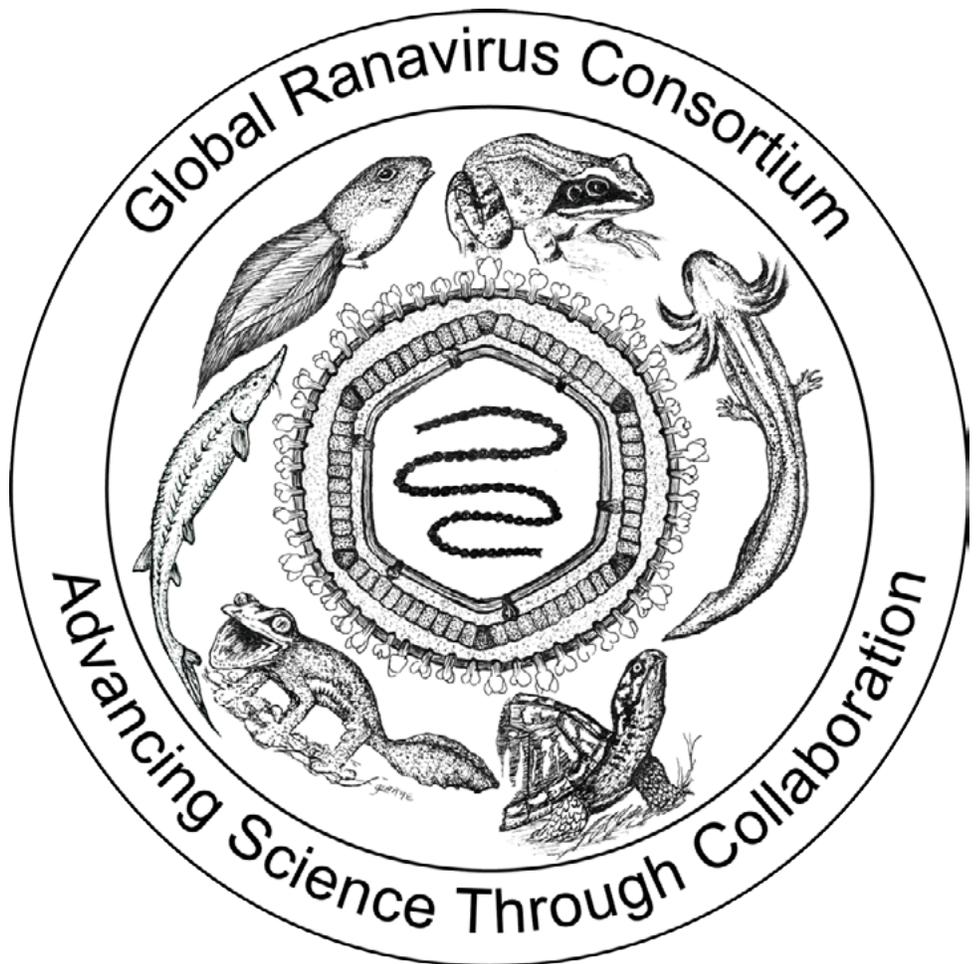
The Global Ranavirus Consortium (GRC), Inc. was formed in 2011 and is a world-wide organization dedicated to advancement of all areas of *Ranavirus* biology. Ranaviruses are emerging pathogens of lower vertebrate species. They have been associated with countless morbidity and mortality events (1), and even population declines of amphibians in some locations (e.g. 2).

In the past three years, the GRC has been busy. In 2013, the Second International Symposium on Ranaviruses was held in Knoxville, TN USA in association with the 62nd International Meeting of the Wildlife Disease Association. The outcomes of this meeting, which can be found at [www.ranavirus.org/symposium](http://www.ranavirus.org/symposium), were instrumental in planning future GRC activities, as well as, influencing the direction of ranavirus research in many areas. At the symposium, the newly elected GRC board met for the first time and set out a list of tasks to be accomplished by the 2015 symposium.

The GRC has a new website, [www.ranavirus.org](http://www.ranavirus.org), that is filled with important information on ranaviruses, a reference list of most articles that have been published on ranaviruses, a list of labs that can diagnose Ranavirus infections, and many other resources that are of use to anyone who is studying amphibians and may come across ranavirus-infected animals. In 2015, the GRC also published the first comprehensive text on ranaviruses, edited by Dr. Matthew Gray and Dr. V. Gregory Chinchar. This text is open access and is available through Springer Online at <http://link.springer.com/book/10.1007/978-3-319-13755-1> (hard copies may also be purchased through this site). In 2015, the GRC also offered charter membership to support the collaborative, interdisciplinary mission of the GRC. You can become a member for a nominal annual fee at <http://www.ranavirus.org/get-involved/>. Funds raised are used to maintain the website, various outreach activities, and support of the biennial symposia. Please consider joining!

The Third International Symposium on Ranaviruses was held in May in Gainesville, FL USA, as part of the Aquatic Animal Health 2015 meeting. The GRC Board met again and the GRC held its first general business meeting. The GRC passed the bylaws that had previously been drafted and approved by the GRC Board. The GRC is currently working on its web presence in social media and are working to design a Facebook page. Its Twitter handle is @RanavirusGRC.

In conjunction with the GRC Regional Representatives, several outreach activities were identified, including a possible online



course on ranaviruses and a Common Midwife Toad Virus summit in Europe in 2016. The GRC recently released a call for proposals to host the International Symposium on Ranaviruses in 2017 and 2019. More information about GRC activities is at: [www.ranavirus.org](http://www.ranavirus.org).

Inquiries about the GRC can be directed to Dr. Amanda Duffus, the Secretary/Treasurer at [aduffus@gordonstate.edu](mailto:aduffus@gordonstate.edu).

#### References:

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2. Price *et al.*, *Curr. Biol.* 24, 2586 (2014).

<sup>1</sup>Department of Biology, Gordon State College, Barnesville, GA USA.

<sup>2</sup>School of Biological Science, University of Washington, Pullman, WA

USA. <sup>3</sup>Center for Wildlife Health, University of Tennessee, Knoxville, TN USA.

# Saving Salamanders with Citizen Science



*Plethodon cinereus*. Photo: Jonathan E. Kolby.

By Jonathan E. Kolby

**A** newly discovered salamander disease is spreading around the world and we need your help to find out where it's going! Please join our citizen science project to save the salamanders: <https://www.inaturalist.org/projects/saving-salamanders-with-citizen-science>

If you find a dead or sick salamander in the wild, please take pictures and upload them to this iNaturalist project as soon as possible! Here are some important project details:

Species identification is NOT necessary. If you cannot identify the type of salamander because they are too long dead, or simply because you don't know salamanders, that's ok. All dead salamander photos are important records!

If your salamander is not dead, but has skin sores or other unusual marks, we also want you to take a photo and report these.

Your photos don't have to be pretty and you don't need a fancy camera! Feel free to use the camera on your phone. A blurry picture is always better than no picture.

Record this simple information along with your photo:

- Date
- Location
- Number of dead salamanders you saw (*i.e.*, if you find a pond with 20 dead salamanders, you might only take a picture of a few, but can report 20)
- Species (your best guess is great, but it's ok if you have no idea, just call it a salamander)
- Suspected cause of death (*i.e.*, was it hit by a car, stepped on, partially eaten by an animal? etc.). Please always make a comment in the description box about this observation. If you tell us there was no obvious reason why it was dead, this is very helpful because we can rule out non-disease factors like road-kill, predation, etc.

The emerging infectious disease we are specifically worried about is caused by a newly discovered species of chytrid fungus (*Batrachochytrium salamandrivorans*, or "*Bsal*"). The faster we can detect its arrival in a new region, the greater our chances to protect the salamanders from disease, decline and extinction.

It appears that *Bsal* originated in Asia long ago, and recently started to spread around the world by the international trade in salamanders. It recently invaded Europe, and is now causing alarming declines in Fire Salamander populations. According to recent surveys, it has not yet arrived in the USA—the global hotspot of salamander diversity—but an outbreak may happen at any moment. Outside Asia, *Bsal* has so far only been detected in Europe (the Netherlands, Belgium and the UK). To help monitor its continued spread, we are seeking records of dead salamanders found ANYWHERE.

With your help, we might be able to detect salamander disease outbreaks much faster than with traditional field surveys alone. So next time you go for a short walk in the park or a long hike in the wilderness, keep your eyes peeled and camera (phone) ready for salamanders! Whether you see just one dead salamander or a bunch, everything you see is important. So please join this project and help us save the salamanders!

Please note: If you suspect you might have visited an area experiencing a disease outbreak, you should sterilize your boots with a 10% bleach solution when you get home to make sure you do not accidentally spread disease to the next place you go for a hike.

Please contact us at [DeadSalamanders@gmail.com](mailto:DeadSalamanders@gmail.com) with any questions or concerns.

# Crowdfunding for Chytrid 2.0 (*Batrachochytrium salamandrivorans*) in Belgium

By Tariq Stark, Sara Viernum, Steven Allain, Myra Spiller & Veronica Reeves

That amphibians are the most imperilled group of terrestrial vertebrates is a fact we are all well aware of in the amphibian conservation community. Over 40% of all species are endangered in their existence and one of the greatest threats are emerging infectious diseases (1). The single celled fungus *Batrachochytrium dendrobatidis* (*Bd*) causes the disease chytridiomycosis and has already affected over 500 species ([www.bd-maps.net](http://www.bd-maps.net)). High elevation species in Central America and Australia, but also in various other locations, have been particularly hard hit by this disease (2). Vredenburg *et al.*, noted: "The effect of chytridiomycosis on amphibians has been described as the greatest loss of vertebrate biodiversity attributable to disease in recorded history" (3). Recently we learned that *Bd* is not alone. In 2013 a new chytrid species called *B. salamandrivorans* (*Bsal*) has been described, originating from Asia and has entered Europe via the pet trade and caused the near extinction of a population of Fire Salamanders (*Salamandra salamandra terrestris*) in the Netherlands (4). Not long after *Bsal* was identified in the Netherlands it was found in two Fire Salamander populations in Belgium causing mortalities and population declines. *Bsal* has also been confirmed in Alpine Newts (*Ichthyosaura alpestris*) in Belgium where it has caused mortality in one population of Alpine Newts. A study published in 2014 showed that all European salamanders and newts were highly susceptible to *Bsal* in the laboratory and died soon after infection, as do some North American species (5). In Europe we could lose up to 44 species and even more in the Americas (more than 300)! This is no longer a problem limited to just the Netherlands and Belgium but could become a global problem very soon! A recently published paper by Yap (*et al.*,) warning about the potential biodiversity crisis if *Bsal* is introduced in North America highlights this very real threat (6).

Can the fungus be stopped or halted? We think that with the results of a study we'd like to perform in the Belgian province of Wallonia we will have a better understanding of the ecology of this disease in the wild. This study will attempt to find out where *Bsal* occurs, how fast it spreads, how it spreads, which host species are affected and how it impacts salamander populations. The results of this study can then be applied to other locations when and where outbreaks occur in the future. Wallonia is a very important place



Tariq Stark and Carlijn Laurijssens swabbing a Fire Salamander (*Salamandra salamandra terrestris*). Photo: Peter de Koning.

to study this fungus for it is the gateway to large fire salamander populations (and other species) in France, Germany and Luxembourg. We plan to collect non-invasive skin samples in the field from salamanders in locations where *Bsal* has not been documented yet or is suspected to be. The swab samples will be sent to Ghent University in Belgium for analysis. The scientists that originally discovered the fungus, Dr. An Martel and Dr. Frank Pasmans, will perform the analysis.

Unfortunately very little funds are available for this study and this is where you can help! Amphibian Survival Alliance (ASA) partner The Wandering Herpetologist initiated a crowdfunding campaign in order to raise money for this much needed study! Our campaign can be found on crowdfunding page and ASA partner WorthWild. You can make a "pledge" or follow our campaign by subscribing. Lots of updates will be uploaded in the next few weeks including a brand new video! We have yet to go live but making a pledge makes all the difference in the world and shows that the amphibian conservation community supports this initiative! No donation is too little or too much, all is welcome! Questions about this campaign? Please email Tariq Stark and Sara Viernum ([wanderingherpetologist@gmail.com](mailto:wanderingherpetologist@gmail.com)) or take a look on our website [www.wanderingherpetologist.com](http://www.wanderingherpetologist.com). You can find our campaign at: <http://www.worthwild.com/prelaunches/17>. This campaign is supported by Ghent University, Nature conservation organization Natagora (Belgium), Reptile, Amphibian and Fish Conservation the Netherlands (RAVON) and the ASA. By donating you actively contribute to this study and salamander conservation in Europe. Thank you!

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How we like to see them: a healthy and gorgeous Fire Salamander found in Wallonia! But for how long? Photo: Tariq Stark.



*Alytes muletensis* - Mallorcan Midwife Toad, one amphibian species that has seen a genuine improvement in status due to the conservation action of zoos Photo: Dawn Fleming.

## A Decade on From the Global Amphibian Assessment: How Have the World's Zoos Responded?

By Jeff Dawson

**T**arget 12 of the Aichi Biodiversity Targets states that, "By 2020 the extinction of known threatened species has been prevented" (<http://www.cbd.int/sp/targets/>). If we are to meet this target then the huge conservation challenge posed by global amphibian declines, brought to the world's attention by the 2004 Global Amphibian Assessment (GAA), must be addressed.

Key actions and issues required to address the crisis were outlined in the subsequent Amphibian Conservation Action Plan one of which was *ex situ* captive breeding and the need to establish multiple captive amphibian programs to safeguard those species most at risk (1) The global zoo and aquarium community (hereafter zoos) represent one of the most influential and important groups of institutions to undertake such programs (2,3). Globally, zoos have contributed substantially to the recovery of 17 out of 68 vertebrate species including at least one amphibian species, *Alytes muletensis* (4,5).

Ten years on from the GAA, the overall feeling in the conservation community has been one of disappointment at the slowness of the response to the amphibian crisis with many conservation organizations still not addressing the issue (6,7). A similarly slow response had been suggested amongst the zoo community, with amphibians being seriously underrepresented in both collections and *in situ* projects supported by zoos (8,9).

As part of Durrell's Saving Amphibians From Extinction (SAFE) Programme we set out to assess what the response within the international zoo community had been; identifying areas of success but also gaps. Using information from the International Species Information System (ISIS) zoo network, we examined trends in global zoo amphibian holdings across species, zoo region and species geographical region of origin from 1994 to 2014. These trends were compared before and after the 2004 GAA to assess whether any changes occurred and whether zoo amphibian conservation effort had increased. The full results of this study have recently been published in *Conservation Biology* and presented below is a short synopsis of the principle findings.

### SUMMARY OF FINDINGS

Over the last 20 years it appears that zoos at the global level have put more effort into globally threatened species (GTS) than non-globally threatened species, which is reflected in a number of metrics. Firstly, the proportion of amphibian holdings that were GTS increased from 17.2% in 1994 to 23.9% in 2014. Secondly, the proportion of all amphibian individuals held that were GTS increased much faster than the proportion of holdings that were GTS, from 16.2% in 2004 to 43.9% in 2014. This is also reflected in the proportion of GTS with metapopulations (*i.e.*, the total number of

individuals held across all zoos) greater than 250 which increased significantly more than the corresponding proportions of non-GTS.

While very positive and encouraging, the absolute numbers and proportions of GTS held in zoos in 2014 was still very low with only 121 species or 6.2% of all globally threatened amphibians being held. This is a much smaller figure than for birds 15.9%, mammals 23% and reptiles 38% (data from 8). Additionally while 23.9% of all amphibians held by zoos were GTS an estimated 41% of those in the wild are threatened with extinction. To reach a similar composition in zoos, as would be expected if a random global sample were to be taken, would take a further 21 years to reach at current rates of change.

Perhaps even more surprising and worrying are those figures relating to the 801 species assessed by Amphibian Ark through their Conservation Needs Assessments as being *ex situ* priority species. Containing both GTS and non-GTS, only 76 of these AArk species were held in zoos over the last 20 years with no difference in holdings in years before or after their assessment year.

Clear differences were found in holdings of GTS based on the species region of origin. The best represented *i.e.*, regions with the highest proportion of all GTS held in zoos globally, in 2014 were North America (45.6%), Oceania (23.6%) and Europe (15.6%) while

South America (2.1%) and Asia (2.6%) were the most poorly represented. When looking at the proportion of species held from a specific region that were globally threatened then Oceania and Caribbean saw the greatest increase, especially in the last 10 years, indicating that zoos holding species from those regions have changed their collections significantly in favor of GTS.

Differences were also found in the holdings at the regional zoo level. European zoos held a lower proportion of GTS in 2014 (17.6%) than zoos in North America (24.4%) and the Rest of the World (20.8%) and unlike the other two regions this had not increased since 2004.

## MAIN CONCLUSIONS

Encouragingly, zoos have put increased efforts into amphibians over the last 20, and in particular the last 10 years. Whether this is a direct response to the amphibian crisis or simply reflects a change in general policy however is unclear. Should similar changes also be seen among bird and mammal holdings then it likely indicates the latter. What is apparent though is that more focus is needed on *ex situ* conservation priority species and clear gaps exist in efforts in relation to certain regions.

It is therefore crucial to understand the barriers to increasing numbers of globally threatened and *ex situ* priority species in zoos and to understand why certain regional faunas are severely under-represented. With this knowledge, measures can be undertaken to increase numbers and proportions of globally threatened and conservation priority amphibians held, such as improving the level of husbandry expertise.

The study also highlights another key issue; the lack of accessible and complete information on amphibian holdings. Although ISIS is the most comprehensive database available, it is not complete and there are potentially many other amphibian captive programs not being publically recorded. This complete data set is critical if the full response to the crisis is to be assessed, identifying gaps and opportunities within these efforts and further developing an evidence-based approach to amphibian conservation planning at a global level, helping achieve Aichi Target 12 and prevent the amphibian crisis becoming a catastrophe.

Full paper reference: J. Dawson, F. Patel, R. A. Griffiths, R. P. Young, Assessing the global zoo response to the amphibian crisis through 20-year trends in captive collections, *Conservation Biology* DOI: 10.1111/cobi.12563 (2015).

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Biosecure breeding unit for the Critically Endangered Mountain Chicken *Leptodactylus fallax* at Durrell Wildlife Park, Jersey Photo: Matt Goetz/Durrell.



*Eleutherodactylus nortoni* listed as Critically Endangered and an Amphibian Ark *ex situ* Research Species that is not in any zoos Photo: Jeff Dawson/Durrell.



## Amphibians and Conservation Breeding Programs: Do All Threatened Amphibians Belong on the Ark?

By <sup>1</sup>Christopher J. Michaels, <sup>1</sup>Benjamin Tapley, <sup>2</sup>Kay Bradfield & <sup>3</sup>Mike Bungard

In 2005, the Amphibian Conservation Action Plan (ACAP) was produced by the IUCN/SSC to outline the threats faced by amphibians worldwide and the conservation steps necessary to protect them (1). A key component of this document, and the one that caught the attention of zoos, conservationists and the public globally, was the concept of conservation breeding programs (CBPs) and ark populations. In the face of rapid, enigmatic and catastrophic declines, populations of amphibians could be collected from the wild and held in breeding centers until the coast was clear for reintroductions to take place. The Amphibian Ark was launched to co-ordinate captive programs in zoos, aquaria and academic institutions around the world, and the 2008 Year of the Frog focussed media attention on the *ex situ* management of threatened amphibians.

However, despite a few notable *ex situ* successes, the hype around CBPs has not translated into a multitude of successful programs and many CBPs have met with little or no success even after more than a decade of efforts in some cases (2). Problems have included

<sup>1</sup>Zoological Society of London, UK; <sup>2</sup>Perth Zoo, Australia; <sup>3</sup>Paignton Zoo, UK.

basic husbandry and captive reproduction, making meaningful progress against threats in the field, particularly emerging infectious diseases, and the management of disease transfer between wild and captive populations, particularly in programs based outside the natural range of focal species. All of these issues led to a recent reconsideration of the *ex situ* model as a means to conserve threatened amphibian species and the widely held notion that all amphibians are suitable for CBPs.

In a recent paper in the journal *Biodiversity and Conservation*, authors from ZSL and Paignton Zoo in the UK and Perth Zoo in Australia critically appraise each of the key factors often cited as predisposing amphibians for CBPs. These include supposed general attributes of amphibians such as small body size and associated low space requirements, high fecundity, applicability of reproductive technologies, short generation time, lack of parental care, hard-wired behavior, low maintenance requirements, the cost effectiveness of such programs compared with programs for other vertebrates, the success of several amphibian CBPs and reintroductions and because capacity exists in the private and zoo sectors (3-7). Tapley *et al.* demonstrate not only how many species fall outside of

these generalized assumptions, but also that the process of choosing which species to include in CBPs is a complex one that requires detailed insight into the biology as well as the conservation needs of individual species. Given that the capacity for species CBPs in the conservation community is an order of magnitude smaller than the number of species requiring such programs for their short term survival (8), it is critical that species are properly assessed not only for their need for CBPs, but also their suitability for such programs.

#### SMALL SIZE AND LOW SPACE REQUIREMENTS

Many amphibian species are small, but there is huge variation across the class; Asian Giant Salamanders, Goliath Frogs and Mountain Chicken Frogs, to name just a few, are by no means small and the space required to hold viable captive populations may therefore be prohibitive. Irrespective of body size, the behavior and locomotion of many amphibian species necessitates a large captive environment. Many anuran species can clear several meters in a single jump, and are highly prone to physical injury and stress if contained within too small a space. Still other species are highly territorial and, particularly when combined with large body size, therefore have large space requirements for their size. These features do not necessarily preclude these species from involvement in CBPs, but highlight the fact that in many cases large and therefore costly facilities will be required to maintain the suggested minimum breeding population size of 20 pairs of animals, particu-

larly when there must be capacity to hold animals separately at times, and to house all life stages appropriately.

#### HIGH FECUNDITY AND SHORT GENERATION TIME

Large clutch size and rapid maturation have been suggested to be beneficial for CBPs as large numbers of individuals can be produced in a short period of time. Although many anurans (and particularly lowland, tropical species) fit this generalization, there is huge variation in clutch size across the amphibia, ranging from a single egg per clutch to more than 45,000 in some anuran species. Not only do some amphibians have small clutch sizes even in comparison with mammals or birds, but those with smaller numbers of offspring may be more likely to need *ex situ* intervention (9). A large proportion of amphibians actually have a relatively long generation time, with temperate species in particular sometimes taking years or even decades to reach reproductive age. Species with such slow maturation rates may require substantially more time and resources to see a project through to completion as they will take proportionately longer to generate cohorts of captive bred animals for release. Conversely, large clutch sizes and short generation time may create difficulties in effective population management and particularly in reducing the effect of selection for captive environments, which can lead to animals maladapted for reintroduction to the wild. In either case, appropriate facilities, timescales and population management strategies will be different and thus



Mountain Chicken Frog. Photo: Benjamin Tapley / ZSL

require specific planning prior to the commencement of the program as well as adequate funding.

### **HARD-WIRED BEHAVIOR**

The role of learning in amphibian behavior is becoming more widely accepted (10), hence the CBPs cannot necessarily rely on instinct to program behaviors necessary for survival in the wild. While mammals and birds rely much more heavily on social learning to develop their behavioral repertoires, the effects of learning with real-world consequences for survival are now known in amphibians; e.g., larval Hellbender Salamanders were trained to avoid the chemical cues of Rainbow Trout, an introduced predator to which naive salamanders exhibit no aversive behavior (11).

### **EASY TO MAINTAIN AND BREED**

Amphibians are highly sensitive to their environment and as such are extremely demanding in captivity, often requiring very specific conditions in order to survive and reproduce. The vast majority of amphibian species have not been maintained in captivity before and, for the hundreds of highly threatened and poorly known species that may be considered for CBPs, any husbandry protocols would be a matter of informed guesswork. Fundamental husbandry issues have been the main cause of failure for a number of otherwise well-resourced programs (see 2 for review). Aside from the specific environmental requirements of individual species, a number of broad areas of amphibian husbandry are still poorly understood. Nutritional disorders are still common in many captive amphibians, as animals are limited to a handful of readily available prey items and the optimal dosage for supplements is not known. Research into the relationships between different aspects of lighting, notably UVB provision and amphibian health is also in its infancy. In many cases, recent research has only opened up further questions.

Assisted Reproductive Techniques (ART) have been proposed to overcome the “captive breeding crisis” created by the difficulty in stimulating many amphibians to reproduce in captivity. Such artificial treatments can also be used to control paternity and potentially select for disease resistance, as well as to provide gametes for transfer between facilities instead of adult amphibians (12–14). Although in some cases such techniques may be appropriate, in general failure to breed in captivity is the result of improper husbandry and a lack of appropriate reproductive stimuli. ART in these cases simply treats the symptom rather than the cause, and may have long term detrimental effects on fitness through both direct effects and by influencing sexual and natural selection (15–17). Perhaps more importantly, although protocols for ART are to an extent predicted by phylogeny (18), precise dosages can be difficult to determine (17; 19) and improper dosages can have detrimental or lethal effects (20). CBPs should therefore not rely on these techniques as a quick-fix to gaps in husbandry knowledge; an understanding of reproductive cues is an important aspect in determining the chances of successfully maintaining a species in captivity.

The quality of captive bred stock is an important determinant of reintroduction success. Therefore, once breeding techniques have been developed, care must be taken to ensure that the captive environment does not produce animals that cannot survive in nature as a result of either environmental or inherited effects. Amphibians can adapt rapidly to a changing physical environment; European Moor Frogs (*Rana arvalis*), for example, quickly adapt through both genetic and non-genetic inherited means to water sources of dif-

ferent pH (21–23). Genetic adaptation to captivity can impact ability to survive with other species; captive bred Mallorcan Midwife Toad tadpoles (*Alytes muletensis*) lose their anti-predator behavior after being reared in the absence of predators for several generations (26). The problem of adaptation to captivity requires a greater depth of information about the precise biotic and abiotic conditions experienced by a population in the wild, and potentially about how those conditions may change before the species is reintroduced.

### **COST EFFECTIVENESS**

Amphibian CBPs will generally require less funding than those for large avian or mammalian species, however, the high technological requirements to successfully maintain most species in captivity, particularly outside of their natural range, can lead to substantial costs. Many amphibians require completely different environments at different stages of their life cycle and a number of different types of enclosure may be needed in tandem to maintain breeding populations. In the case of very large species or those requiring large amounts of space, the costs could exceed those of programs for some middle-sized mammals or birds.

Due to the time required to ameliorate many threats in the wild, and to produce release cohorts of slow-reproducing species, amphibian CBPs may need to run for significant periods of time. Captive populations of the Kihansi Spray Toad (*Nectophrynoides asperginis*) were established in the US in 2000 (24), the first releases happened in 2012 and the project still continues, 15 years later, in an effort to secure the species in the wild. Similarly, frogs of the 20 species involved in the Panama Amphibian Rescue and Conservation Project (formerly EVACC) were collected between 2001–2005 and as yet there have been no releases (25).

The costs of an amphibian CBP usually are not insurmountable, but any project must take into account both the full requirements to house species appropriately and also the longitudinal commitment for funding required to see a project through from initiation to securing a species in the wild and eventual redundancy of a facility.

### **REINTRODUCTIONS CAN WORK**

The eventual goal of most CBPs is to create self-sustaining wild populations of target species by releasing captive bred individuals. This requires an appropriately designed and funded captive breeding facility, subject to the caveats already discussed above, but also a concrete plan of how to ensure that releases happen and are successful. It is one thing to “rescue” animals from a rapidly declining population and quite another to design a CBP with a well-defined end point, both in terms of goals and a timeline. CBPs must work in tandem with *in situ* projects designed to ameliorate threats in the wild so that reintroduced animals do not simply succumb to the threats that necessitated *ex situ* intervention. This means that the nature of threats must be carefully factored into the process of prioritizing species for CBPs. Some of the most dramatic amphibian declines have been precipitated by emerging infectious diseases, often in association with climate change. Both of these have proven very difficult to tackle in the wild and many programs dealing with disease-threatened species have no defined timeline or end point as the means to tackle these pathogens has not yet been developed. Such species may end up in a captive limbo, with no way to return to the wild while resources are diverted away from other species that could be reintroduced within a relatively short timescale.

## CAPACITY EXISTS

The original model for amphibian CBPs was to use the resources and skill sets available in zoos and aquaria to run *ex situ* programs. This model largely involved moving species across the world from where threat levels are typically highest and where resources, skill sets and organization (*i.e.*, capacity) are typically lowest, to institutions in the US and Europe, where capacity is greater, but conservation need is lower. Even where species were to stay in country, they would typically be moved to an existing institution outside of their actual geographic distribution.

Unfortunately, this model has several fundamental issues. Firstly, the removal of animals from their range country and the running of projects by foreign institutions and individuals raises ethical and legal issues. Exporting animals, when this is even a legal possibility, may disenfranchise key stakeholders *in situ* and compromise the ability to ameliorate threats in nature. Secondly, facilities in the US and Europe, as well as the cost of transporting animals and personnel between these regions and range states, are comparatively expensive, which can compromise long term funding for projects. Lastly, zoos and aquaria, wherever in the world they exist, are potential sources of pathogen transfer and other forms of contamination, including hybridization. Holding animals for release alongside cosmopolitan animal collections creates a very real threat of pathogen acquisition and the potential to do more harm than good when animals are released. This scenario played out in Mallorca, when captive bred Midwife Toads were released along with *Bd*, previously unknown on the island (27). The fact that the fungus was unknown to science at the time stresses the importance of biosecurity in the face of both known and unknown pathogens. The private sector can offer a huge range of highly specialist skills to CBPs, with large numbers of hobbyists, particularly in the US and Europe, able to maintain and breed a vast array of species. As such, these individuals can provide vital information on the husbandry and biology of conservation targets. In some cases, private individuals may be involved in CBPs, but the same biosecurity risks apply to these cases as to the zoo and aquaria setting. The high turnover of staff and changing interests in both the zoo and private sectors can also jeopardize CBPs that may need to run for decades.

The CBP model now adopted by Amphibian Ark is to host dedicated facilities within the country and ideally within the distributional range of the target species. This minimizes the risk of disease transfer both to and from captive animals. Dedicated programs may also be staffed by individuals with skill sets honed to target species. This type of in-country program also facilitates the involvement of range-state stakeholders and the combination of *in*- and *ex-situ* aspects of conservation. However, many areas of high conservation interest have limited capacity, with poor infrastructure, funding availability, access to specialist technology, skill sets and organization. Moreover, some areas may actually be politically unstable or unsafe. 92% of Haiti's amphibian fauna is threatened with extinction, for example, but political unrest and extreme poverty in the country make it impractical to obtain funds and safely establish a CBP there.

All of these issues must be taken into account when a species is considered for a CBP and, if dedicated facilities with appropriate capacity cannot be established within the range of a particular species, the difficult decision not to develop an *ex situ* program may be the most appropriate option.

## THE BIGGER PICTURE

Amphibian CBPs can be a critical and useful part of amphibian conservation, as shown by those projects that have underpinned or at least contributed to the safeguarding of species. However, for too long they have been considered a panacea for amphibian declines. Currently, resources may be channelled to species that are not suitable candidates for CBPs on the basis of particular characteristics and/or where threats in the wild cannot be tackled, in particular infectious disease, and therefore for which no goals and end-points can be defined. Moreover, the practicality of meeting husbandry and biosecurity needs for the potentially long duration of a CBP, including financial implications, is often not explicitly considered in assessing the appropriateness of a proposed program. By considering these aspects, resources can be directed to those species which both require *ex situ* intervention to survive and for which there is a likelihood of a CBP ultimately succeeding.

Source article: Tapley, B., Bradfield, K., Michaels, C.J. and Bunnard, M. (2015). Amphibians and conservation breeding programmes: do all threatened amphibians belong on the ark? *Biodiversity and Conservation*. doi: 10.1007/s10531-015-0966-9. Accessible at: <http://link.springer.com/article/10.1007/s10531-015-0966-9>

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## Lazarus Toads: What Can They Tell Us About Amphibian Conservation

By Michael Roy

CREA, a Marin based conservation non-profit and Amphibian Survival Alliance partner became the subject of *New Scientist* magazine's first photo driven feature in its 60-year history. CREA's work to save the beautiful, endangered Harlequin Toad (*Atelopus limosus*) is offering new hope for the species' recovery in the face of a devastating disease.

Amphibian populations around the globe have been in free-fall for the last few decades. The culprit, a deadly fungus known as *Batrachochytrium dendrobatidis* (*Bd* for short) and identified only as recently as 1997, has been responsible for wiping out many species seemingly overnight.

But in a handful of locations there are, it turns out, a few surprising survivors of this deadly disease.

Conservation through Research Education and Action (CREA), which undertakes amphibian research at their Cocobolo Nature Reserve in Central Panama, recently made a wonderful discovery. *Atelopus limosus*, one species that is virtually extinct in the wild due to *Bd*, was reported not only to be surviving the *Bd* wave but also to be breeding. *New Scientist* (Aug 15<sup>th</sup> 2015) recently published an article on these *Lazarus* Frogs and on the research that is taking place at Cocobolo into why this population, like a few others in Costa Rica, may have survived.

Dr. Michael Roy, CREA's founder, noted that "uncovering the mechanisms by which these populations survive may be critical for creating a conservation plan for wild amphibian populations and planned reintroduction efforts, in the face of *Bd*. This discovery has presented us with a golden conservation opportunity but we desperately need funding to take advantage of it and expand our research. The outcome of our work has the potential not only to save *A. limosus*, but also aid in the management of endangered frogs and toads all over the world."

"The fact that some of these species are reappearing years or even decades after they were last seen is enormously encouraging," says Robin Moore, conservation officer with the Amphibian Survival Alliance. "After decades of witnessing rampant declines, these glimmers of hope are much-needed morale boosters."

CREA is currently seeking international partners to collaborate on research and education programs at the Cocobolo Nature Reserve, especially those that support conservation efforts for *Atelopus*. Visit [www.crea-panama.org](http://www.crea-panama.org) to learn more.

# Developing Madagascar's Amphibian Husbandry Capacity with Institutional Internships

By Jeff Dawson

**A** core component of Durrell's Saving Amphibians From Extinction (SAFE) Program ([www.durrell.org/safe](http://www.durrell.org/safe)) is aiding the development of *ex situ* conservation activities in areas and for species where captive breeding is needed and appropriate. Madagascar is one such place where the development of in-country captive breeding capacity is most definitely needed. As well as the primary threat of habitat loss, the new and yet unknown threat of chytrid in the country means that for a number of species captive breeding may become a necessity to help ensure their survival. As part of Durrell's SAFE program work in Madagascar we have been working with local NGO's Association Mitsinjo and Madagascar Fauna and Flora Group (MFG) to develop amphibian captive breeding capacity in country.

Of course, it is highly fortunate that Mitsinjo, based near Andasibe, already manage a fantastic community run bio-secure amphibian conservation center. This is an ideal model for both establishing and running other such centers within Madagascar and as an in-country resource expertise and knowledge. Previously Durrell has helped utilize this knowledge by facilitating a number of training exchanges between Mitsinjo and a new breeding center at Parc Ivoloina, run by MFG, to help ensure enclosure set up and protocols are properly established. As and when more centers are planned in Madagascar Mitsinjo staff will be at the forefront leading training activities and helping develop a network of in-country captive breeding expertise.

Durrell itself also has great experience in the captive breeding and management of amphibians through our Wildlife Park in Jer-

sey and an important part of Durrell's philosophy is to integrate and link Park based activities with our field conservation work. We want to do this through the SAFE Program to develop capacity and skills and as such, earlier this year we ran the first of what we hope is a regular series of internships with Durrell's Herpetology team. One of Mitsinjo's amphibian technicians Jeanne Soamiarimampionona—or Mampy as she prefers to be known—spent a month working with the team at the Durrell Wildlife Park in Jersey.

This was an invaluable opportunity for Mampy, who had never before left Madagascar—to get experience working for an extended period of time in a world leading herpetology department and develop her own knowledge and skills. Mampy spent time working alongside Durrell staff and volunteers in all areas and programs, including learning the different food cultivation methods and working in our biosecure breeding facility for the Critically Endangered Mountain Chicken (*Leptodactylus fallax*). Importantly it also gave her constant exposure to the application of all the various protocols involved in the running of the department including biosecurity and detailed daily record keeping. In addition, her time at Durrell allowed her to see animals that she had never seen before not only amphibians but Gorillas, Fruit Bats and Komodo Dragons. In Mampy's own words, "I'm very satisfied and proudly going back to my country with the large knowledge I have got from Durrell Wildlife Conservation Trust and apply it to our amphibian conservation program in Andasibe Madagascar."

Of course, the true impact of any training intervention is how trainees subsequently utilize the knowledge and implement the



Mampy working in Durrell's biosecure Mountain Chicken breeding room. Photo: Durrell.

skills learned. It is therefore fantastic to hear that since returning to Andasibe, she has been doing just that. According to Devin Edmonds, Mitsinjo's Amphibian Conservation Director, upon her return Mampy instigated some experimental cultures for springtails, one of the food sources raised, based on what she had learned in Jersey. These have been so successful that they are now planning on switching all their springtail cultures to this new method.

Spending time working in a facility such as Durrell's with so many strict protocols, especially around biosecurity, means new knowledge learned becomes instilled in the learner. Since returning Mampy has been able to strengthen and further instill this ethos amongst the team at Mitsinjo. This perhaps reflects one of the most important aspects that internship style interventions like this can deliver, improving an individual's confidence and self-belief. Having the confidence to pass on knowledge, influence others and implement changes is hugely important if the training undertaken by an individual is to be disseminated through an organization. Indeed, as Devin reports even though it was just one individual who went on the training trip it has given a lift to the whole team, boosting motivation which is really encouraging to hear.

Looking to the future the hope and plan is that the team at Mitsinjo, bolstered by such training interventions will be able to further share their skills and knowledge with other captive centers in Madagascar to develop a network of skilled technicians and practitioners within the country. As mentioned Parc Ivoloina is the first of these and Durrell were very pleased to be able to host Parc Ivoloina's Director Bernard Iambana for two weeks in June in between meetings in the US and UK.



Mampy with Durrell herpetology staff Tom Wells and Dan Lay. Photo: Durrell.



Mampy's favourite amphibian in the collection - Strawberry Poison-dart Frog, *Oophaga pumilio*. Photo: Matt Goetz.

During this time Bernard was able to spend a week working with the Herpetology team and get first-hand insights into what protocols are used and their importance. Having this knowledge and understanding will hopefully enable Bernard to be better able to guide the development of the captive breeding center at Parc Ivoloina, including identifying key areas to focus on.

At Durrell, we hope that Mampy will be the first of a regular series of amphibian captive husbandry internships. Not only will this help develop in-country capacity in this area but from an institutional perspective will help link our park based staff in with our conservation programs and utilizing the full range of expertise that Durrell as an organization has. On a wider scale this type of intervention, could also be a highly productive way for zoos to contribute to amphibian conservation that utilizes their expertise and engages their park staff.



Mampy working at the Mitsinjo facility in Andasibe. Photo: Jeff Dawson.

# Amphibians in a Changing World: A Global Look at Their Conservation Status

By <sup>1</sup>Rafael Loyola, <sup>1</sup>Priscila Lemes, <sup>2</sup>Nicolás Urbina-Cardona, <sup>3</sup>Diego Baldo, <sup>4</sup>Julián Lescano & <sup>4</sup>Javier Nori

Currently, more than 40% of extant amphibian species are threatened with extinction and a quarter of them still lack information to be classified as threatened, being therefore enlisted as Data Deficient by the International Union for the Conservation of Nature (IUCN) (1,2). Furthermore, amphibians are the greatest underrepresented group in the global network of protected areas (PAs) worldwide.

A decade ago, some studies showed that around 17% of amphibian species lived completely outside of protected areas (3). Obviously, the underrepresentation of amphibians in protected areas is much higher for range-restricted species that inhabit highly human-modified landscapes. Even in face of this worrying scenario, since 2004 not a single update has been published showing amphibian species represented inside PAs at the global scale. Actually, there is a large gap of information, especially if we consider that today data on the distribution of many amphibian species are available, and the areas covered by PAs has increased over the last ten years from 11% to more than 13% worldwide (4).

We have recently filled this gap by publishing a new and comprehensive overview on the ability of the global network of PAs to protect amphibian species (5). We also offered new information about the overlap of species' distributions with different types of human land-use around the globe. In this study, we considered different amphibian taxa and geographic regions, making a particular distinction between gap species (*i.e.*, those completely outside PAs) and range-restricted species (*i.e.*, those with geographic distributions smaller than 10,000 km<sup>2</sup>) (5).

Our analyses revealed that almost 25% of all amphibians, which is more than 1,500 species, still remain totally outside protected areas. Moreover, 1,119 species have less than 5% of their geographic distribution represented in protected areas. Although we have more protection (about 10% more area designated as protected and 13,000 additional reserves), the proportion of amphibian species falling outside these protected areas has also increased. In reality, only a few designated reserves perform by avoid species loss or reducing species' extinction risk.

While this situation seems to be paradoxical, it was actually expected due to how protected areas are selected. Within governments and all administrative levels authorities tend to establish residual reserves, that is, reserves located in places where human interests are minimal. These places play a minor role in protecting biodiversity, given that threatened species are precisely where human impacts are higher.

This is why continents harboring a large proportion of gap species (such as Latin America, Asia and Africa) are being highly im-

acted by human activities. On average, 65% of every gap-species' distribution is now inside human-modified landscapes. Africa has the largest proportion of species affected by human impacts with only 16% of gap species' free from human influences. In several key

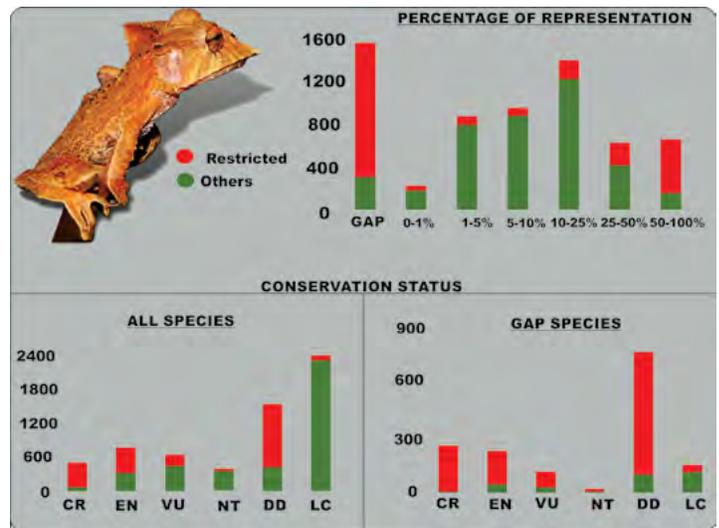


Fig. 1: Histograms showing the percentage of the distribution of the species included in PAs for each species and the number of species assigned to each IUCN status when considering all species and only the gap species. All histograms discriminate range-restricted species. Amphibian species: *Hemiphractus bubalus*.

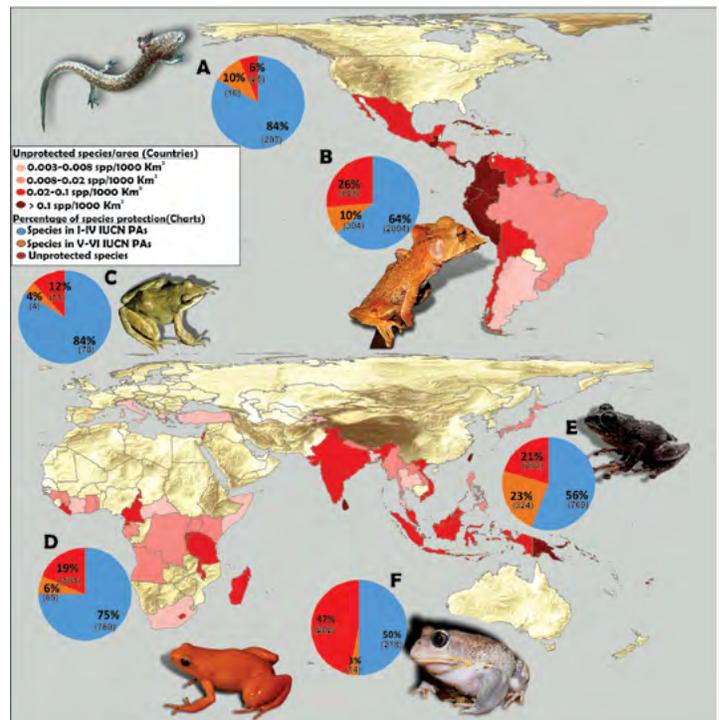


Fig. 2: Map showing the number of unprotected species per unit area in the world's countries and pie charts with the percentage of species occurring in different protected area management categories. This figure is illustrated with an amphibian species from each continent: *Eurycea latitance* (North America), *Hemiphractus bubalus* (Latin America), *Rana pyrenaica* (Europe), *Mantella aurantiaca* (Africa), *Heliophorus australiacus* (Oceania) and *Philautus umbra* (Asia).

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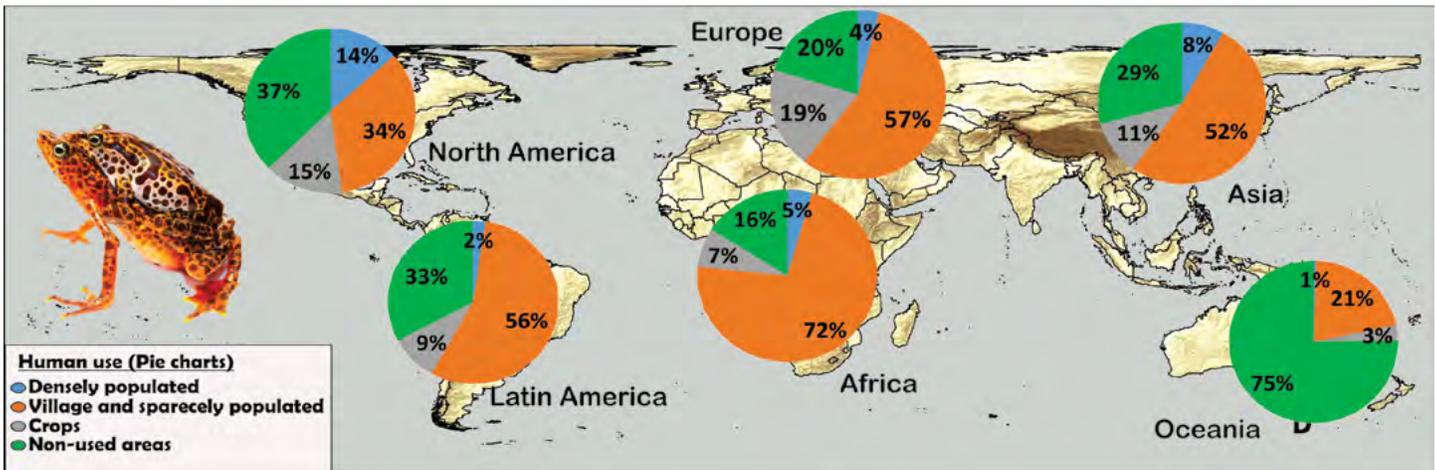


Fig. 3: Percentage of spatial overlap between species' geographic distribution and different types of human land use in the world's continents. Amphibian species: *Atelopus certus*.

regions (such as part of tropical Andes, Southeast Asia and central Africa), the combined effect of low levels of protection and a steady human influence will inevitably aggravate the current crisis scenario for amphibians by further declines and extinctions. Tenacity must be shown by the public and the scientific community to urgently implement conservation policies, including governmental and social initiatives aimed at strategically expanding the current network of Protected Areas for greater conservation purposes.

More than area protection, we need reserves that make a difference for conservation. That is, new protected areas should be established where they would produce the largest impact on amphibian conservation. By impact, we mean an explicit evaluation (or simulation) of what would have happened if there had been no conservation intervention or establishment of protected areas (6). Only with such evaluation we will be able to estimate the real impact new reserves would have on amphibian conservation. In addition, growing the size of the global network of protected areas will not be enough, as we observed.

Furthermore, it is important to note that 45% of gap species are currently classified as Data Deficient by the IUCN. Many of these species inhabit highly disturbed environments. Data Deficient species are usually ignored or considered as species of least concern in conservation policies, plans and recommendations (2). Hence, it is essential to increase our knowledge on many biological aspects of these species, such as taxonomy, systematics, demography, ecology, natural history and threats, in order to generate adequate conservation policies.

This brief overview highlights important issues, which can potentially increase the current crisis faced by amphibians, but points out several challenges and opportunities towards creating more comprehensive amphibian conservation strategies in the next decade. It is essential to consider amphibians when developing conservation policies that lead to the implementation and management of new protected areas. It is critical to increase funding for scientific research to expand our knowledge of amphibian species, especially on those tropical key topics mentioned above.

Finally, we need to start planning for positive impacts of conservation intervention, carefully measured and monitored, so we can foster the establishment of protected areas that will make a real difference in avoiding amphibian species loss and reducing their extinction risk. With that in mind, and the new and improved protected areas strategies established, we are hopeful that amphibian conservation will reach a vastly improved level of animal species conservation and protection worldwide.

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# Frog eat Frog

By <sup>1</sup>Giovanni Vimercati & <sup>2</sup>John Measey



Cannibalism in the African Bullfrog *Pyxicephalus adspersus* in Polokwane, Limpopo Province, S. Africa. Photo: Les Minter.

**F**rogs can be voracious predators, and we usually think of their prey as insects and other small invertebrates. Generations of herpetologists have extracted stomach contents to see what frogs eat. The results are not what we might have expected however, as their capacity to feed on relatively large items such as reptiles, birds or mammals is surprising (1). Not least among these larger prey items are other frogs. Some species are notorious frog eaters, such as the South American Horned Frogs (genus *Ceratophrys*), the African Bullfrogs (genus *Pyxicephalus*) and the North American Bull Frog (*Lithobates catesbianus*). But are these the only frogs eating frogs? What variables are influencing this behavior?

A common hypothesis is that bigger frogs are more likely to consume other frogs. However, this has yet to be tested across taxa and maybe there are other characteristics that are strongly associated with frogs that eat other frogs. We decided to investigate the extent of anurophagy (literally “feeding on frogs”; from Latin prefix an-, “not” + Ancient Greek ourá, “tail” and from Ancient Greek-phagia, from phagein, “eat”) at the population level to ask how widespread it is in frogs. In addition, we wanted to determine the influence of some key variables: habitat, diversity and invasiveness. To accomplish this we conducted a literature review of post-metamorphic diet in Anura (2). The ease of stomach content analyses through dissection or stomach flushing has produced an extensive literature on frog diet. From each paper we extracted the species name, total prey items, total anurans eaten (eggs, larvae and post-metamorphics), location and mean body size. Moreover, we also considered for each record: species taxonomic position at family and superfamily level, anuran species diversity at the study site, habitat, cannibalism occurrence and if the studied population was native or invasive. In total we analyzed data from 355 cases in 323 papers representing 228 species. Our results show that anurophagy is not uncommon, with the predation on eggs, tadpoles or post-metamorphic frogs reported in more than 20% of cases. Ranoidea and Pipoidea were observed feeding on other frogs more frequently than other super-

families, showing how the phylogenetic position is correlated with anurophagy. Correcting for this taxonomic effect, we confirmed the size hypothesis, with large frogs more likely to feed on other frogs. For every additional millimetre in the body size, the likelihood of observing frogs in the diet increased 2.8%. We also found that habitat and anuran diversity play a role in determining whether a frog species showed anurophagy. More specifically, generalist species consume significantly more frogs than forest, shrubland and grassland species, and frogs from sites with high anuran species diversity were more likely to consume frogs. On the other hand, cannibalistic species (*i.e.*, species that had conspecifics among their prey items) were not observed to have more frogs in their diet if compared with non-cannibalistic species. Last but not least, invasive anurans were 40% more likely to consume frogs than non-invasive ones.

While the positive effect of body size on the capacity to prey on other frogs is fairly straightforward to interpret, other factors such as habitat or anuran diversity are more difficult to put into context. Generalist species should have the capacity to use a larger spectrum of microhabitats and show a more flexible behavior, having a higher possibility to come across other frogs to feed on. For analogous reasons, anuran diversity could act as a proxy of higher frog abundance in the ecosystem or determine a more diversified niche partitioning—both elements that should cause higher encounter rate of one anuran with another (especially when one is generalist). These areas seem ripe for further investigation. Our finding that invasive species were more likely to be predators of other frogs, even after accounting for the effect of body size, is an important result. However, dietary data for invasive species was limited and we encourage more research on this topic. From a conservation perspective, it has to be noted that native frog populations are currently declining across the globe (3) and introduced amphibians are at least partially driving this decline (4). Since the amphibian trade is potentially causing new frog introductions (5) and some countries are currently compiling list of species that should not be traded, we suggest that large generalist species, and especially ranids and pipids, should be of particular concern because of their tendency to feed on other frogs, especially in areas characterized by high anuran diversity.

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An adult African Clawed Frog *Xenopus laevis* regurgitates a Clinking Stream Frog *Strongylopus grayii*. Photo: John Measey.



*Pelophylax perezii*. Photo: E. Fasola.

## Genetic Erosion: Menace for Amphibian Species Viability?

By <sup>1</sup>E. Fasola, <sup>2</sup>R. Ribeiro & <sup>1</sup>I. Lopes

**A**lmost half of all known amphibian species are threatened with extinction and it is hypothesized that a decrease in the genetic variability of their natural populations may worsen this scenario (1-3). Indeed, the maintenance of genetic variability is important to prevent the loss of the evolutionary potential of populations/species and, therefore, the loss of their capacity to handle environmental changes (4-12). The genetic structure of natural amphibian populations arises from past events and present evolutionary processes. The former are shaped by biogeography of the single species, while the latter occur due to micro- or macroevolution (6). Furthermore, since amphibian populations usually hold a small number of individuals, which will contribute to the gene pool each mating season, it is expected that they would be easily affected by inbreeding enhanced by genetic erosion (*sensu* loss of genetic variation in a population), which could drive local populations to extinction (4,12).

Genetic erosion may be promoted by factors like: drift, including bottlenecks and mutational meltdown (6,8,9,11). Thus, studying and understanding the influence of environmental changes in the

genetic diversity of amphibian populations is of utmost importance to plan for accurate conservation strategies and prevent further species loss. Chemical contamination may impact the genetic diversity of exposed populations by causing genetic erosion, which in turn may lead to the loss of alleles in the population (6,8,9). Genetic erosion can also increase the susceptibility to other stressors. It is possible to differentiate genetic erosion from other evolutionary processes if we identify a population in which sensitive genotypes are absent in a polluted site and tolerant genotypes are present at both polluted and reference sites. This genetic erosion at the impacted site should have been caused by contaminant-driven natural selection (6,8,9). Genetic erosion can also be due to contaminant-driven random genetic drift (including bottleneck effects), which may lead to inbreeding (6). This is crucial for amphibians as their populations commonly have an effective size ( $N_e$ ) lower than 100. When compared with census size ( $N_c$ ), this makes them very susceptible to genetic depletion (4,12).

Amphibians are also susceptible to a wide range of stressors and are considered to be highly sensitive to environmental alterations. Habitat fragmentation, climate change, diseases, introduction of allochthonous species, pollution and water acidification have been described, among others, as factors responsible for amphibian declines (1-3). Decreased genetic variation can cause reduced fitness and lack of adaptability to such varying and changing environ-

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ments. Genetic erosion can impact amphibian populations causing: 1) reduction of fitness, 2) reduced phenotypic plasticity, 3) failing of protective co-tolerance effects, 4) tradeoffs between fitness and genetically determined tolerance mechanisms, and 5) increased susceptibility to pathogens.

In the first of these scenarios, genetic-fitness-correlations are explored. In some amphibian species, heterozygous individuals show increased fitness in comparison to homozygous ones suggesting that individual genetic variability should be correlated with a higher fitness (6,13–19). The second point analyzes how organisms cope with changing environments with “phenotypic plasticity” capabilities. These represent the ability of a certain genotype to generate different phenotypes. Phenotypic plasticity helps individuals to face abrupt habitat variations because each genotype can retain adequate fitness components for future use in populations. However plasticity has a genetic basis and genetic variation for plastic responses is observed. Thus, genetic erosion may affect plastic responses too (6,15,20–25).

The third point tackles how a single-stressor perspective is inadequate because individuals, populations and ecosystems are affected by multiple stressors occurring simultaneously (6,26–29). Amphibian populations may be forced beyond their tolerance limits and genetic erosion may occur. Individuals use co-tolerance mechanisms to face these situations decreasing genetic variability lowering the adaptive potential towards multiple stressors (6,30). In these difficult scenarios, trade-offs occur when the ability of an organism to perform in one ecological scenario suffers at the expense of its abilities to perform in other different situations.

In addition, genetic erosion can bear fitness costs associated with altered physiological processes. These costs can affect population viability or reproductive processes lowering effective size ( $N_e$ ) or increasing inbreeding (6,18,31).

Finally, in the fifth scenario, inbred populations could be tolerant to one pathogen, but are possibly susceptible to most other unrelated pathogens. Furthermore, an allele that provides tolerance to

an infectious disease could be negative when the pathogen is absent, because of possible pleiotropy effects. Hence, heterozygosity is very important for the functioning of the immune system. Actually, the loss of genetic diversity decreases tolerance to pathogens. Therefore, it would be crucial to monitor amphibian populations’ genetic variability in a world in which they are exposed to many infectious agents like *Ranavirus* or *Batrachochytrium dendrobatidis* (6,32).

In conclusion, amphibian populations represent good indicators to assess the impacts of contaminant-driven genetic erosion. There is some evidence correlating lower genetic diversity with decreased: fitness, environmental plasticity and tolerance mechanisms towards pollution or pathogens (6). However focused research is needed to understand the structure of genetic erosion in microevolutionary processes.

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*Pelophylax perezi*. Photo: E. Fasola.

# Garden Management Could Help Reduce Amphibian Disease: Citizen Science in the UK

By Alexandra Catherine North

Our recent study uses a long-term citizen science dataset to identify a range of variables associated with disease in wild Common Frogs (*Rana temporaria*) in the UK (1). Emerging diseases are one of many threats facing amphibians across the globe (2,3). *Ranavirus*, caused by a double stranded DNA virus (4), is one disease causing notable die-offs of amphibians across Europe, North America and Asia (5–10). The disease has been implicated in the decline of amphibian populations in Europe (9,10), including declines of over 80% in Common Frogs in the UK (9). *Ranavirus* was thought to have been first introduced into the UK through the international pet trade (11). Unsolicited reports of unusual frog mortalities in the thousands led to a nation-wide campaign to better understand the spread and drivers of the disease. This campaign was named the Frog Mortality Project, and resulted in a dataset that now spans two decades. The dataset has been administered by *Froglife*, with each mortality event listing signs of disease, and details about the garden, pond ecology and management. In the UK, amphibians are commonly found in urban and suburban garden ponds lending themselves well to pond owner population monitoring. Using population size estimates, recorded signs of disease and a range of environmental variables, we used this dataset to better understand the ecology of the disease and to determine if there were associations between garden management and the incidence (how likely a population was to get infected) or prevalence (how many frogs within each population became infected) of *Ranavirus* (1).

Our results suggest that the reduction of common garden chemicals and limiting the introduction of non-natives such as ornamental fish, could help reduce *Ranavirus* prevalence in Common Frog populations. We also provide insight into the complexities of transmission dynamics of *Ranavirus* in the wild with frog population density and the presence of potential hosts increasing *Ranavirus* prevalence; this highlights the generalist nature of the virus and how it can be an infector of whole amphibian communities. The negative impacts of anthropogenic chemicals for wildlife are well documented (12–14) and our study contributes to this body of research, suggesting the use of herbicides and slug pellets increases disease prevalence. Pesticides are known to cause immunosuppression in amphibians (15) and have been shown to increase *Ranavirus* susceptibility in salamanders in controlled conditions (16,17). Pesticide and herbicide free gardens will encourage a healthier ecosystem, with healthy slug and snail eating amphibian populations negating the need for use of these pesticides.

Fish presence increased the prevalence of *Ranavirus*, though the exact mechanism behind this pattern is currently unknown. We hypothesize that ornamental fish could be increasing the density of susceptible hosts, amplifying environmental levels of virus or influencing immune function through energy trade-offs. Fish species are known to be susceptible to *Ranavirus* with the ability to infect amphibian species in experimental conditions (18). Indeed, in the wild *Ranavirus* has been extracted from fish and amphibian species that live in the same place (19), suggesting that transmission is likely to occur *in situ* too. Alternatively, the presence of fish could cause reduced immune function if for example, predator presence reduces foraging opportunities. Fish are known predators of tadpoles (20) and tadpoles have been shown to exhibit reduced immune function

in the presence of another predator, the dragonfly larvae (21). The impact of fish presence on Common Frog immune function however requires further investigation.

Our study highlights the immense value of citizen science projects identifying large-scale trends useful for conservation efforts. The importance of amphibians for ecosystem functions (22), coupled with knowledge that amphibians are the most threatened taxonomic group (23), makes identifying positive management actions as important as ever. We hope that the enthusiasm of UK citizens in contributing to the Frog Mortality Project demonstrates their dedication to amphibian conservation efforts. We are optimistic that our findings could encourage the implementation of wildlife friendly gardening approaches to reduce *Ranavirus* prevalence and to improve the health of ecosystems as a whole.

If you find an amphibian that is unwell in the UK, you can now report it to the [Garden Wildlife Health Project](#).

## Acknowledgments:

For further details on statistical approaches and all variables tested, please see the original paper at PLOS ONE: A.C. North, D. J. Hodgson, S. J. Price, A. G. F. Griffiths. *PLOS ONE*. DOI: 10.1371/journal.pone.0127037. (2015). We would like to thank charity *Froglife* for providing the Frog Mortality Project dataset, and to pond owners for submitting their records to the project.

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Barton Springs Salamander (*Eurycea sosorum*). Photo: Matt Charnock.

## Part One: Within The Public Water Column: *Eurycea sosorum*

By Matt Chanrock

The melodious rumble emanating from my car's exhaust now rests silent, suffocated by the left-turn of an ignition switch. Headlights off, the warm early morning sunbathes the opaque plastics in a washed yellow. Still not yet fully awake, despite the twenty-minute drive snaking through downtown Austin, I clamber from my antiquated, well-worn driver's seat, fogged by confusion. "Okay, now do I have everything? Do I have my swimsuit and towel?" I question myself with little reassurance. "Nope, I left the towel in the hatch." The mental checklist I'd simultaneously created now crossed-off, I coax my stiff legs from their pedal-resting confines onto the adjacent cold asphalt, dampened by the morning's dew. With a somewhat conscious ease, I make it through the weathered gates, past the antiquated "Welcome!" sign, down the limestone steps, eventually releasing my nylon duffel bag onto the graveled rock lining the creek bed. All is quiet, the eerie silence coddled by situational solitude. No persons have yet stirred the mirror-like water, and no stray waterfowl could be seen floating through the veiling mist. But despite my waning hypnotic state, I meticulously take off my black-hooded sweater, unlace my double-knotted sneakers, pluck the no-show ankle socks from my now bare

feet, and slowly dip my yet acclimated lower body into the cold spring waters. Amassing any lingering morale: "You've done this a hundred times. Well, maybe not a hundred, but at least ten." Again, I repeat—and with bated breath. But the unnerving plunge is inevitable, a gesture of returned dignity. "How is this seventy-degrees? Whose thermostat is leaking mercury?" But this is Baton Springs, after all; a place where man and amphibian intermingle with little acknowledgement of the other.

Because I honestly believe each of the aquifer's endemic salamanders needs an equal, encompassing piece, this excerpt is one the first of a three-part installment, focusing around the vulnerable Barton Springs Salamander (*Eurycea sosorum*).

Hatched in the secluding shadows of the spring's rocky crevices, these enigmatic, small salamanders begin life as mirror-images of their adult ilk—just packed in a smaller, opaque epidermis. Again, while we know every little about either their reproductive cycles or maturation patterns, one factoid is all too clear: from the millisecond they emerge from their encasings, Barton Spring Salamanders rely heavily on well-stocked populations of amphipods. Not only has the salamander's affinity for this dietary cornerstone been

observed in the wild, but also in CB/CBB (Captive Bred/Captive Bred Born) individuals as well (1). And, should the elements and habitat preservation be in their favor, the larval salamanders will, in time, develop into neotenic adults, capable of spawning in the late-fall and early-winter; researchers have observed developing egg clusters in their niche habitats as early as September until late January (2).

Abiotic factors hold no biotic prejudice, no predisposed connotations; they exist absent minded of their surroundings. But, that being said, the latter subject doesn't quite hold the same mantra for the former—and these translucent quadrupeds are no exception.

Like every other amphibious poikilotherm known to science, Barton Spring Salamanders lean on the homeostatic, consistent conditions of their aquatic realms (3). Occupying the secluded, somewhat still shadows of the aquifer, they lurk, mostly unnoticed, amongst their crystal-clear aquatic confines. However, despite their geological crutch, these “canaries in the mine” do rely heavily on both the health and abundance of the creek’s aquatic vegetation; researchers observed a sharp decline in population counts when such floral growth was scant. This correlation between healthy plant growth and well-established populations of the salamanders is likely the byproduct of ideal dissolved oxygen levels produced by the copious amounts of photosynthetic verdure. But that’s not to say all growth, regardless of taxonomic designation, is welcome.

Chytridiomycosis has already suffocated a slew of amphibian species out of existence and left many teetering. And, unfortunately, the Barton Springs Salamander didn't elude the invasive fungus completely—or successfully. Seven specimens were collected and, out of the small sample pool, only one individual tested positive for *Bd*; each animal's DNA was extracted and put through PCR amplification with two primer codes known for their affinity for chytrid fungus (*Batrachochytrium dendrobatidis*) code primer Bd1a (5' CAG TGT GCC ATA CAC G-3') and Bd2a (5'-CAT GGT TCA TAT TCG TTC AG-3'). But, fortunately, this mycelium alien hasn't woven the fungal noose that's knotted much of their other, more susceptible kin (4).

Lungless salamanders, as a conglomerate whole, are tightly woven into a water body's flow regime, a variable sometimes eclipsed by a discharge. But where those animals decide to live within the water column is crucial, is essential to their livelihood. And, while a system's discharge may, on an office spreadsheet, look unchanged, the passing water five feet below may have drastically shifted from .15-feet/per-second to .24-feet/per-second in the same timespan, rendering it uninhabitable for otherwise lethargic biotic factors. In



Barton Springs Salamander (*Eurycea sosorum*). Photo: Matt Charnock.

order to truly grasp the health and future sustainability of such environmentally dependent animals—and Barton Spring Salamanders tote that moniker proudly—micro data and analysis must be at the forefront, placing macro assimilations in the still laudable periphery (5, 6).

And then the ecological teeter-totter becomes unbalanced; the ecology of that microhabitat now skyward in heavy disproportion.

Fully submerged, my hair now reminiscent of a damp suburban rodent, I rocket back to the surface in thermal revolt. “OK, the hard part's over,” validating my now cherry-red complexion. “And it's only a 20-minute swim today, anyways. I got this.” But, just as I'm about to kick my legs in forward population, a peculiar presence populates my foresight. A small apparition of sorts—or so it initially appeared. Scaling the adjacent rock carpeted by silt-free algae, its translucent body combs the vegetation for a satiating meal. “It's a Barton Springs Salamander! Wow, it really is one!” And, with the enigmatic ease that graces such small amphibians, it slips almost unnoticed into the crevice created by a neighboring rock. All is, again, still, danced only by the passing bits of vascular plants; both man and beast carry on in shared harmony.

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Photo: Timothy J/Flickr.

# Mapping the Malabar Tree Toad—a Citizen Science Initiative in Conserving an Endangered Toad in the Western Ghats of India

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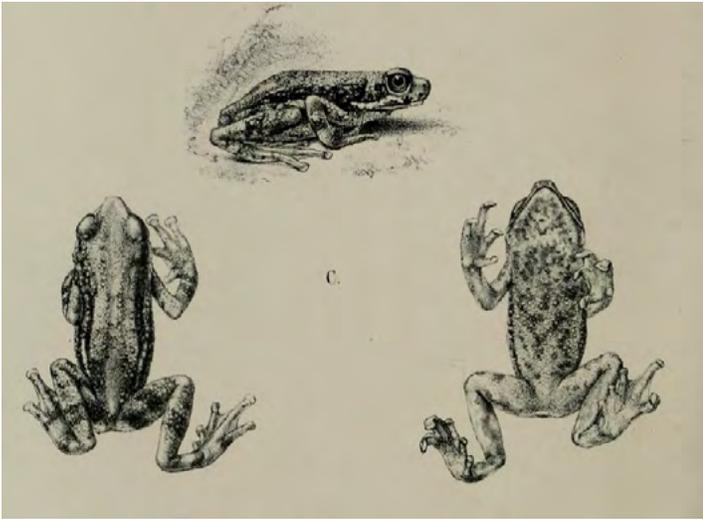


Fig. 1: Malabar Tree Toad: Original description by Gunther 1875.

With the arrival of monsoon, the Western Ghats of India have transformed into a lush green heaven for frogs and toads. Among them is a unique species of toad. Unlike its fellow toads that live on land, it lives in the forest canopy of evergreen forests across the Ghats—the Malabar Tree Toad, *Pedostibes tuberculosus* Gunther, 1876. From a systematics perspective it is the only species of *Pedostibes* found in the Western Ghats, while the other four congeners are from North East India and Malaysia.

The Malabar Tree Toad was described way back in 1875 by Dr. Albert Gunther based on a collection by Lieutenant Colonel Beddome (Fig. 1) but lacked precise locality information (1). It had been known from a few locations like Kalakkad, Ponmudi, Silent Valley National Park, Cotigao Wildlife Sanctuary, and Charmadi Ghats of Karnataka (2). Apart from these sporadic observations and call records, there is little information about this species, leaving a great chasm of knowledge waiting to be filled (Fig. 2).

In a first, scientists at Gubbi Labs have assembled citizens to bridge gaps in knowledge about this species. The citizen science initiative called “Mapping the Malabar Tree Toad” started on June 6, 2015 and has already collected over 25 unique observations from various parts of the Ghats in the past three months (Fig. 3). With this information the habitat of this toad appears severely fragmented; the distribution of this species is limited and the population is suspected to be declining. These factors led the Global Amphibian Assessment Team to classify the toad as Endangered (3) on the Red List of the International Union for Conservation of Nature (IUCN).

Scientists are constrained by time and resources to survey the entire Western Ghats for such a rare species. However, for a nature enthusiast living in and around the forests, they can simply observe what is around them and report it on a common platform, thus showing the unique power of Citizen Scientists at work. This

will not only help local inhabitants to enjoy and appreciate their surroundings better but will contribute to strengthening the body of knowledge needed to conserve species and their habitats.

Furthermore, this effort will enable us to bridge knowledge gaps in the species’ ecology itself. So far, we know they breed in the month of June but we do not know where they go for the rest of the time. Do they climb trees, burrow into soil, or are they simply hiding in plain sight as no one is looking for them? These are some questions the initiative will be able to answer given the effort needed.

Critiques of citizen science initiatives often cast doubts about the credibility of citizen based initiatives. But in this case, with Gubbi Labs, a private research collective, India Biodiversity Portal ([http://indiabiodiversity.org/group/frog\\_watch/userGroup/show](http://indiabiodiversity.org/group/frog_watch/userGroup/show)) and EarthWatch Institute India as joint knowledge partners, there is a strong scientific support to help gather the necessary information into a useful format. Through this process, citizens, amateur naturalists, nature photographers and the like can be transformed into the role of a scientist and begin to systematically document natural observations otherwise, too costly and time consuming for scientists to gather. Such efforts are not new to biodiversity and its conservation. In the past several other taxa like birds, trees, bears, and other groups have caught citizens’ attention and they have helped map distribution, with varying levels of success (4-7).

Such initiatives serve two purposes: 1) they enable citizens to appreciate the importance and beauty of scientific documentation, and 2) it reduces the alienation of nature in their hearts and minds, bringing them closer as a society to life around them. With much of India’s biodiversity being found outside of protected areas and among the general public, conservation efforts are greatly enhanced and many might falter without the public’s support.

Scientists at Gubbi Labs have been actively working in the Western Ghats and have come up with several publications and com-



Fig. 2: A calling male Malabar Tree Toad. Photo: Gururaja KV.

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puter/smart phone applications (apps) like the first pictorial guide to frogs and toads of the Western Ghats (8); *FrogFind* a mobile application (9) to identify area frogs and recently, an acoustic guide to the frogs and toads of Western Ghats called *Mandookavani* (10).

Will we pay heed to the voice of frogs? Will we listen to what they have to say and take the necessary action? It remains to be learned what can be accomplished through citizen based initiatives. Citizen programs like Mapping the Malabar Tree Toad are a brazen attempt to bring these wonderful creatures of the night one leap closer to our hearts. Each one of us could potentially be part of this process of discovery by reporting our sightings of this rare toad on <http://tinyurl.com/malabartreetoad>.

### HOW TO REPORT A SIGHTING?

Step 1: Download *Frog Find* or log into India Biodiversity Portal (IBP) to get familiar with the tree toad.

Step 2: On seeing the toad, record (take) a picture/record video/record calls.

Step 3: Note the location and other pertinent observations.

Step 4: Report sighting using *Frog Find* by simply logging into IBP and filling out the proper form(s).

### Acknowledgements:

We thank all participants, who shared and contributed to the knowledge about Malabar Tree Toad < [http://indiabiodiversity.org/group/frog\\_watch/show](http://indiabiodiversity.org/group/frog_watch/show)>. We also extend a wonderful thanks to our knowledge partners Indiabiodiversity.org and EarthWatch Institute India.

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Fig. 3: Citizen scientists searching the toad. Photo: Gururaja KV.



Photos: Ramya Badrinath.

## Bridging Gaps between Scientists and Citizens: Uncovering the World of Frogs and Toads in Honey Valley, Coorg, Karnataka, India

By Ramya Badrinath

**L**ike previous workshops, Bamboo Rustles, an organization which organizes nature education and awareness programs, hosted “Scientists and Citizens:” Amphibian workshop with Dr Gururaja KV. The workshop was in its 3<sup>rd</sup> year and was scheduled to take place in Chingara, Honey Valley, Coorg, from 10–12<sup>th</sup> July 2015. As I expressed my desire to join the group, Dr. Gururaja gave me an unforgettable opportunity—this time, I will be giving an introductory presentation about amphibians, their global presence, and describing our own Western Ghats and its amazing diversity and citizen science initiatives to a bunch of amphibian enthusiasts. This was officially the first time, I was addressing a group of people who were from diverse backgrounds—business, commerce, engineering, advertising and many others but had converged to uncover the interesting world of frogs and toads. I was enthusiastic and equally nervous, but was looking forward to this exciting exchange nonetheless.

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We reached Honey Valley in the afternoon of July 10<sup>th</sup> and my talk was scheduled after lunch. A funny thought about participants falling asleep during my talk did cross my mind and I was getting all the more nervous; it was about time I started off. Well, assembling the setup for my presentation was at its creative best and took a good 45-minutes time. After setting up, I began giving a general introduction about amphibians, their local and worldwide distribution, about a few commonly occurring and highly endemic species, their behavior, current IUCN status of some species, threats posed to them and citizen science initiatives. Rants become monotonous, but thankfully, the group got eager and enthusiastic and the participants were posing great questions which eventually led to avid discussions. The session was gripping enough. No one seemed to be bothered about the cold, chilly weather outside—everyone was so engrossed in the discussion. I was delighted to see participants opening up in spite of coming from such diverse backgrounds, their knowledge about wildlife was immense. Our discussions got inter-



Photos: Suma H / Ramya Badrinath.

esting, one thing led to another and there was a great outpouring of enthusiasm that I didn't realize how the time flew. By the time I wrapped up the introductory session, I realized it was already time to set out to the field for another adventure in amphibian biology!

Dr. Gururaja joined us later that evening and we set out in search for frogs. It was indeed an audio visual treat for all of us, for we got to hear the amazing symphony of bush and stream croakers and even see a few species. The participants were engrossed in observing the behavior of the species they saw and diligently making notes. I was more drawn towards recording the frog songs. The lush green thicket of Honey Valley seemed more like an acoustically engineered concert hall with operas, choirs and symphonies from frogs, birds and insects around. It was nothing short of euphoria. We spent close to six hours in the field that night.



Sound recording in progress. Photo: Gururaja KV.

The next day began with an extended presentation by Dr. Gururaja KV and some amazing inputs about amphibians and his experiences from his research career that span fifteen years. We then set out to Tadiandamol, the highest peak in Coorg district. As we hiked along, we could spot a variety of bush croakers besides hearing the symphony. Dr. Gururaja gave some amazing natural history insights. It was great to see the participants' eagerness to learn new things. Their excitement knew no bounds. It seemed to flow beyond its capacity. Dr. Gururaja led the team during field work and we trekked all along. We didn't climb till the peak of Tadiandamol. All that mattered to us was to make random stops, look for frogs on either sides of the trekking pathway, get into dense patches on the way and scout around for bush and stream frogs. All the participants were instructed not to catch the frogs barehanded, for there is a possibility of infections spreading. Also, the ethics of photographing a species in its natural habitat was instructed. We were glad that none of the participants breached the rules. Be it morning or late night, all the participants, Srikanth, Suma, Amatya, Nagesh and Raja were enthusiastic to set out to the field and make notes of whatever they observed or heard. Time didn't stop their excitement to observe frogs! Srikanth and Amatya were professional photographers, but others weren't less either. All of them got great record shots of the species they saw.

The next morning, we went near a private waterfall owned by Chingara Resorts Pvt. Ltd., to look for dancing frogs. Dancing frogs, as they are called, have a spectacular foot flagging behavior. We could see one or two species of dancing frogs, calling out to their counterparts, displaying their white vocal sacs which were in contrast to their morphological color.

Apart from the frogs, we saw a couple of non-venomous snake species too. Sandeep, our snake expert was sharing his immense knowledge on snakes with us. Amatya also has an amazing knowledge on snakes. As a bonus, while we were returning in jeeps from Honey Valley to Kabbinkaad junction where our vehicle was parked, Sandeep spotted a Coral Snake gulping in a caecilian! That was quite a sight. One group (including me) which had already reached Kabbinkad missed the sighting; the other group was fortunate enough to witness it. This was indeed a great natural history record.

Our two-day workshop ended with that note, but was a great beginning for all the participants to embrace themselves into the wilderness. The to and fro journey was a sheer contrast of sorts. We behaved like aliens- not knowing how to break the ice and what to talk while coming, but while going back, it was completely opposite. All of us had eventually immersed ourselves into intense scientific discussions that we crossed a host of towns during our journey in almost no time.

Here's a list of the frogs and toads we encountered during the two-day workshop at Honey Valley:

1. *Duttaphrynus melanostictus*, Common Indian Toad
2. *Ghatophryne ornate*, Malabar Torrent Toad
3. *Indosylvirana intermedius*, Rao's Intermediate Golden Backed Frog
4. *Micrixalus elegans*, Elegant Dancing Frog
5. *Micrixalus saxicola*, Wayanad Dancing Frog
6. *Nyctibatrachus grandis*, Wayanad Night Frog
7. *Nyctibatrachus minimus*, Miniature Night Frog
8. *Nyctibatrachus sanctipalustris*, Coorg Night Frog
9. *Raorchestes chromasynchisii*, Confusing Colored Bush Frog
10. *Raorchestes glandulosus*, Glandular Bush Frog

11. *Raorchestes luteolus*, Yellow Bush Frog
12. *Raorchestes nerostagona*, Kalpetta Yellow Bush Frog
13. *Raorchestes ponnudi*, Ponnudi Bush Frog
14. *Raorchestes tuberohumerus*, Knob Handed Bush Frog
15. *Rhacophorus malabaricus*, Malabar Gliding frog
16. *Zakerana caparata*, Cricket Frog
17. *Zakerana kudremukhensis*, Kudremukh Cricket Frog
18. *Ichthyophis* sp.
19. *Gegeneophis* sps.



Photos: Ramya Badrinath.

The main aim of the workshop was to get all the participants interested in the world of amphibians. Their full participation at every step gave a deep sense of satisfaction that we had achieved what we intended to do. By the time the workshop ended, irrespective of the backgrounds they came from, they had all turned to citizen scientists by then. The citizen science initiative, started by Gubbi Labs and India Biodiversity Portal is slowly gaining impetus and is having active participation from both scientists and citizens all over India. People are actively contributing their observations which eventually would help in gaining insights to amphibian distribution, their behavior, threats and IUCN status. One such initiative which has gained full momentum is the Mapping Malabar Tree Toad initiative (Mapping the distribution of *Pedostibes tuberculatus* – Malabar Tree Toad). This initiative started and led by Dr. KV Gururaja, along with Gubbi Labs, India Biodiversity Portal and Earthwatch India is progressing well with great contributions from users.



A coral snake swallowing a caecelian. Photo: Gururaja KV.

# Identification of Tadpoles of an Endemic Genus *Nyctibatrachus* from Central Western Ghats India

By H. <sup>1,2</sup>Priti, K.V. <sup>3</sup>Gururaja & <sup>1</sup>G. Ravikanth



Fig. 1: Myristica swamp forest, central Western Ghats, India. Photo: Gururaja KV.

India's Western Ghats is known for its high amphibian diversity. In the last decade more than 100 new frog species were discovered from this region (1) and many more are still waiting to be discovered from this amazing biological treasure trove. While much is known about adults, the larval life forms of frogs from Western Ghats are less studied with few descriptions dating back to the 19<sup>th</sup> century (2).

The frog genus *Nyctibatrachus* (late Cretaceous in origin) is endemic to Western Ghats of India. These are stream dwelling frogs found in torrent streams or leaf litter on forest floor (3). The adults are nocturnal. While working on adults we realized that we know very little about the tadpoles. Understanding larval ecology is important as they reflect the natural history (4) and can provide useful information about evolution of amphibians (5).

Our study involved identifying *Nyctibatrachus* tadpoles for understanding their ecology. The study was conducted in streams and *Myristica* swamps of evergreen forests (Fig. 1) of Sharavathi and Aghanashini river basins. These river basins are two of the major west flowing rivers of the Western Ghats and form a part of Aghanashini Lion tailed Macaque Conservation Reserve. Myristica swamps are unique ecosystems that are relict in origin (6). These are freshwater swamps that provide perennial source of water. They also harbor many endemic flora and fauna. We conducted the study in six streams and sampling was carried out from upstream to downstream. The downstream areas comprised of swamp forests while the upstream areas consisted of agricultural fields. Previous inventory of amphibians from these Myristica swamps included

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three *Nyctibatrachus* species *N. kempholeyensis*, *N. jog* and *N. kumbara*. The tadpoles of these three species co-occur in the streams. They were seen at the edges of the stream where the water is shallow and slow flowing. The microhabitat is mainly made of sand, small gravel and leaf debris. The tadpoles could be seen scraping leaf debris and when approached hid under the substrate. These tadpoles are diurnal and we did not observe any cannibalistic or schooling behavior.

Morphologically the tadpoles of *N. kempholeyensis*, *N. jog* and *N. kumbara* look similar with brownish dorsal color. However, there were significant differences in their size (Table 1). Although *N. kempholeyensis* could be distinguished easily from other two species, it was difficult to identify *N. jog* and *N. kumbara* morphologically (Fig. 2). Therefore we made use of molecular methods like DNA barcoding for better identification. DNA barcoding using standard 16S rRNA gene was helpful in distinguishing the tadpoles of three species. Besides identification, we could also understand that among the tadpoles so far

described from Western Ghats, tadpoles from this genus lack labial teeth, a trait that is unique to this genus and could have evolutionary implications (7).

The tadpoles of *Nyctibatrachus* inhabit streams of evergreen forests and Myristica swamps. Many of the Myristica swamps have been converted into areca plantations or into agriculture fields. These Myristica swamps once were found throughout the water-courses of Western Ghats but now are fragmented. Such changes can also alter the microhabitats used by the tadpoles and can affect both tadpoles and adults. We think that batrachologists and people involved in amphibian conservation must also study tadpoles. Such studies can be helpful in conservation planning as tadpoles remain in their habitat for a longer period of time than adults. In India, there has not been much research on tadpoles. Studies on tadpoles of over 350 species can not only increase our understanding about their ecology but also to appreciate conservation and evolutionary perspectives.

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Characters	<i>N. jog</i>		<i>N. kempholeyensis</i>		<i>N. kumbara</i>	
	Mean±Sd	Range	Mean±Sd	Range	Mean±Sd	Range
<i>BL</i>	11.2 ± 0.6	(10.4 - 12.1)	6.2 ± 0.5	(5.2 - 6.8)	16.6 ± 1.9	(13.8 - 20.2)
<i>BH</i>	5.1 ± 0.2	(4.7 - 5.3)	2.6 ± 0.4	(2.1 - 3.2)	7.2 ± 1.1	(5.7 - 8.7)
<i>BW</i>	6.8 ± 0.7	(5.7 - 7.6)	3.5 ± 0.4	(3.0 - 4.0)	10.5 ± 2.0	(7.9 - 13.3)
<i>TL</i>	31.6 ± 2.6	(28.4 - 36.2)	18.5 ± 1.8	(16.4 - 20.6)	47.0 ± 5.8	(39.0 - 56.9)
<i>TAL</i>	20.5 ± 2.0	(18.0 - 24.1)	12.3 ± 1.4	(10.6 - 13.9)	30.4 ± 4.1	(24.5 - 36.7)
<i>ED</i>	0.9 ± 0.1	(0.8 - 1.1)	0.5 ± 0.1	(0.4 - 0.6)	1.4 ± 0.3	(0.9 - 1.8)
<i>IOD</i>	3.3 ± 0.3	(3.0 - 3.7)	1.8 ± 0.2	(1.5 - 1.9)	4.9 ± 0.7	(3.7 - 6.0)
<i>IND</i>	2.0 ± 0.2	(1.8 - 2.3)	1.2 ± 0.1	(1.0 - 1.4)	2.7 ± 0.3	(2.1 - 3.1)
<i>ESD</i>	3.6 ± 0.3	(3.3 - 4.2)	2.3 ± 0.3	(2.0 - 2.6)	5.2 ± 0.5	(4.4 - 5.9)
<i>TMH</i>	3.8 ± 0.3	(3.3 - 4.1)	2.3 ± 0.2	(2.0 - 2.6)	5.9 ± 1.1	(4.6 - 7.8)
<i>TMW</i>	2.6 ± 0.4	(2.1 - 3.2)	1.3 ± 0.1	(1.1 - 1.4)	4.3 ± 0.7	(3.1 - 5.5)
<i>MTH</i>	5.1 ± 0.6	(4.5 - 6.3)	2.7 ± 0.2	(2.4 - 3.0)	7.9 ± 1.4	(5.9 - 10.1)
<i>DF</i>	1.5 ± 0.2	(1.4 - 2.0)	0.8 ± 0.1	(0.7 - 0.9)	2.4 ± 0.5	(1.6 - 3.1)
<i>TMHM</i>	2.5 ± 0.4	(2.1 - 3.2)	1.3 ± 0.2	(1.1 - 1.5)	3.9 ± 0.6	(3.2 - 4.9)
<i>VF</i>	1.1 ± 0.3	(0.7 - 1.6)	0.6 ± 0.1	(0.5 - 0.6)	1.5 ± 0.4	(0.9 - 2.2)
<i>ODW</i>	1.9 ± 0.2	(1.5 - 2.0)	1.0 ± 0.1	(0.8 - 1.1)	2.7 ± 0.5	(2.0 - 3.3)

Table 1. Morphometric variations in tadpoles (stage 25) of *Nyctibatrachus jog* (n = 8), *N. kempholeyensis* (n=6) and *N. kumbara* (n = 7). Measurements in mm.



Fig. 2: Tadpoles of *Nyctibatrachus kumbara*. Photo: Dr. Gururaja.

# Rhacophorid Frogs Breeding in Bamboo: Discovery of a Novel Reproductive Mode from Western Ghats

By Seshadri K S

Reproduction is central to progression of all life forms. While processes like budding are straightforward, things start to get complex when sexual reproduction is examined. Amphibians are model organisms where reproduction—the all-important life history trait—reaches bewildering complexity. The diversity so high, that we refer to them as reproductive strategies or more simply, reproductive modes. Anurans exhibit a staggering 42 different reproductive modes, by far the highest among all vertebrates. The nearest contender, mustering a mere 20 odd modes lays rather expectedly, in water—fish.

## ANURAN REPRODUCTIVE MODES

There is no single answer to what is a reproductive mode. Over the century, our thinking of what exactly is a reproductive mode has evolved (1). A simple means of classifying a reproductive mode is based on three aspects of an anurans life history: the egg deposition site; what kind of an egg and whether or not parental care exists (2). If one examined the reproductive modes of all anurans, the pattern of increasing terrestriality and decreasing dependence on water becomes apparent as over 20 modes are aquatic. It seems only natural to think that anurans, with their aquatic ancestors would have reproductive modes that are largely aquatic too. But molecular evidence and increasing natural history information have made

us question the notion that anuran reproduction has progressed further from aquatic dependence to increased terrestriality (3).

## DISCOVERY FROM A RE-DISCOVERY: THE INTRIGUING CASE OF *RAORCHESTES CHALAZODES*

It was the middle of 2009. I was working on acoustic monitoring canopy dwelling frogs in the Kalakad Mundanthurai Tiger Reserve (KMTR) in Southern Western Ghats. Sprawling over 900 sq kilometers, the upper reaches of this reserve are covered with the largest contagious tropical wet evergreen forests in all of Western Ghats. In the monsoons, I spent extended periods of time studying frogs among other things. It was during one such stint that I witnessed an exhilarating sight. Dr. Ganesh, Prashanth and I were heading up into the field station in Upper Kodayar. We decided to stop by a place called the “Wooden Bridge” named so after a bridge opened in the October of 1942 by the British. This road across the origins of River Manimutharu leads to a tea estate owned by the Bombay Burmah Trading Corporation. It was June and the rains had begun to lash out relentlessly. That night, the rains ceased momentarily. We walked into the bamboo clump along the stream and homed in on calls of *Raorchestes chalazodes* (Fig. 1). It was not an onerous task. We saw an adult male calling on a bamboo stalk. I grabbed my camera and began to record. As we stood there watching it, the frog did



Fig. 1: *Raorchestes chalazodes* from Upper Kodayar, Kalakad Mundanthurai Tiger Reserve. Photo: Seshadri KS.

something that left us all in awe. It squeezed itself into the bamboo stalk via a small opening and that too with great difficulty (Fig. 2).

We were the first to observe this behavior and capture footage of it. *Raorchestes chalazodes* is an enigmatic frog belonging to the tree frog family Rhacophoridae. For several years, it was thought to be “lost” until an expedition comprising of Drs. Biju, Ganesan and myself stumbled upon it on bamboo clumps along the road in Upper Kodayar (4). After this, I had spent several nights trying to locate this frog and had narrowed down to bamboo clumps. I had carefully split the bamboo and observed that the frogs breed inside and usually the male frog would be with the egg. Over two monsoon seasons, I managed to get a fair hold on what was happening. Several nights and man-hours of searching for these frogs in forests away from bamboo and streams did not yield results.

After seeing the frog strenuously enter bamboo, I undertook further explorations and studies as part of my Ph.D. This resulted in observations of an egg clutch, developmental stages, breeding phenology and habitat preferences some in depth, some cursory but nonetheless, one tiny leap towards unveiling the momentary truth of knowledge. While several modes were described earlier, there was no such mode described where the frog would enter bamboo via a small opening and lay direct developing eggs in internodes devoid of water. In addition, there was the male taking care of the developing embryos.



Fig. 2: *R. chalazodes* entering bamboo- a sequential screen grab. (Video url: [amphibiaweb.org/species/4399](http://amphibiaweb.org/species/4399)). Photo: Seshadri KS.



Fig. 3: *Raorchestes ochlandrae* male from Coorg, Karnataka. Photo: Seshadri KS.



Fig. 4: *Ochlandra travancorica* habitat where *R. chalazodes* breeds. Photo: Seshadri KS.

In this process, we realized that another species of frog from the Western Ghats, *Raorchestes ochlandrae* (Fig. 3) also showed similar behavior. Eggs were laid inside bamboo internodes and the adult male would stay with eggs indicating the presence of parental care. Having now gathered sufficient evidence, we realized that we had discovered a novel reproductive mode among all frogs and described the same (5).

### WHY BREED IN BAMBOO?

These frogs, measuring between 20.6–25.2 mm from snout to vent breed inside hollow bamboo internodes; they enter and exit bamboo via small openings. Stems where we found egg clutches ( $n = 2$ ) and having froglets ( $n = 1$ ) were on average  $26.5 \pm 6.6$  mm in outer diameter and  $16.4 \pm 5.02$  mm in the inside. The openings were  $39.0 \pm 32.88$  mm in length and  $3.4 \pm 2.77$  mm in width. The egg clutches were always observed inside internodes where the openings were towards the base and eggs would be on the upper end. The internode lengths ranged from 400–630 mm and the upper end of opening to nearest egg in each egg ranged from 77–450 mm. The clumps of bamboo comprised of 13–86 stalks and grew in dense clusters along streams (Fig. 4).

Eggs were spherical and transparent with creamy white yolks. They were attached to the inner walls of the bamboo by means of a mucilaginous strand (Fig. 5). Multiple clutches were separated by only a few millimeters and averaged  $1.5 \pm 1$  clutches per internode ( $n = 6$ ). Average clutch size was  $6.7 \pm 1.2$  egg/clutch ( $n = 4$ ) and eggs were  $5.73 \pm 0.66$  mm in diameter ( $n = 28$  from five clutches; Fig.



Fig. 5: Egg clutches of *R. chalazodes* inside internode. Photo: Seshadri KS.

6). The froglets would remain inside the internode upon hatching. Coloration would be similar to the adult except the eye ornamentation (Fig. 7).

Bamboo breeding appears to be a common trajectory taken by several frogs in the Western Ghats. There is another group of frogs like *R. manohari* and *R. uthamani* which are distinctly smaller and geographically spaced out and yet, breed in smaller bamboo stalks. Recent molecular evidence also does suggest that the bamboo breeding frogs from a distinct clade within the larger Rhacophorid group of *Raorchestes* (6). E.O Wilson in 1975 (7) attributed four main drivers for the evolution of life strategies in amphibians. Adaptations like parental care (Fig. 8) evolved when organisms were in stable and structured habitats; or in unusually stressful environments; depended on scarce or specialized food resources; and lastly, faced considerable predation pressures. In both *R. ochlandrae* and *R. chalazodes* the bamboo breeding seemed to be an adaptation being driven by some force.

What transpires inside the bamboo internodes? What animal makes openings on bamboo? How do these frogs find and keep track of these limited ephemeral resources? These are questions with answers of varying certainty. Perhaps when we find out more, we would know why this novel reproductive mode evolved.

#### CONSERVATION AND MANAGEMENT

This frog, listed as Critically Endangered, also inhabits habitats that are considered unimportant or are commercially viable and exploited. *Ochlandra travancorica* the Indian flute bamboo grows luxuriantly along streams in the evergreen forests of the Western Ghats. Several species of this bamboo are also used for manufacture of paper and pulp. A few years ago, a charismatic frog *Raorchestes manohari* was described after Robin Abraham and his team heard a new frog call from a truck carrying *Ochlandrae* reeds to a paper factory (8)! My ongoing work has yielded interesting outcomes on novel behavior and insights into ecological constraints. These are

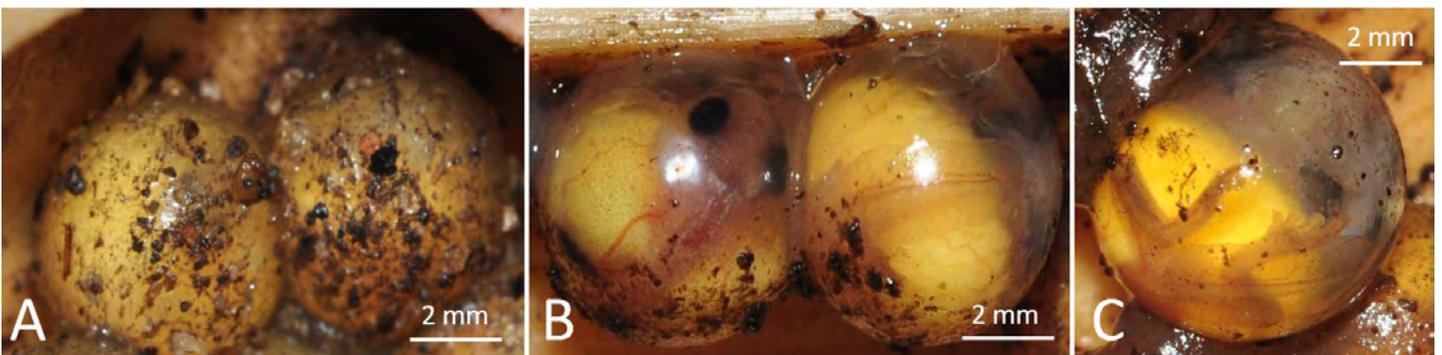


Fig. 6: Egg development inside bamboo progressing from A to C. Photo: Seshadri KS.

vital to determine key drivers for evolution of reproductive strategies. The studies will feed into management plans were bamboo reeds are harvested. Intervention measures to conserve these frogs could range from simply not harvesting bamboo during the frogs' breeding season.

### IMPORTANCE OF NATURAL HISTORY

Studies on the reproductive modes in anurans have come a long way. In a seminal synthesis of anuran reproductive modes, Marty Crump (1) writes, "The concept of reproductive mode has evolved from descriptive natural history to an integration of developmental biology, genetics, systematics, evolution, ecology, behavior, and physiology that frames our thinking about the transition of vertebrates from water to land and about anuran reproductive adaptations to diverse environments today...The next decade is certain to witness significant advances in our understanding of anuran reproductive modes."

True to this, a novel reproductive mode was discovered from a fanged frog *Limnonectes larvaepartus* (9). In the Western Ghats alone, yet another reproductive mode was discovered in late 2014. The mud packing potter frog *Nyctibatrachus kumbara* was found to be covering non-aquatic eggs with mud (10). Our own discovery of novel reproductive mode in *R. ochlandrae* and *R. chalazodes* was another addition. These discoveries all have one thing in common—natural history observations. It is the perseverance of people who have spent time in the field doing nothing but listening to what frogs have to say.

In light of our new discovery, says Dr. David Bickford based at the National University of Singapore "It is 2014, and we are still making discoveries like these; natural history is sexy—always was and always will be." He adds, "No matter what the molecular and genomic revolutions have accomplished for us in the biological sciences, nature is still the ultimate source for everything we do in biology."

India is home to an astonishing variety of life forms and the Western Ghats is a well-known hotspot for amphibian radiation. While amphibians are on the decline everywhere, several novelties have been uncovered from the Western Ghats. While taxonomy and systematics have gone leaps and bounds; natural history and evolutionary ecology of amphibians has, in general, been lagging behind. This gap can and surely will be filled by good science stemming from patiently documenting natural history. There is much ground to be covered for fully unraveling nature's mysteries



Fig. 8: Adult male of *R. chalazodes* (A) and *R. ochlandrae* (B) guarding eggs inside bamboo, an example of parental care. Photo: Seshadri KS

around anurans and us will lead us forward.

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Fig. 7: Hatched froglets at varying stages of development. D is inside internode and E is after it emerges out. Photo: Seshadri KS.

# Rapid Decline and Extinction of a Montane Frog Population in Southern Australia Follows Detection of *Bd*

By <sup>1</sup>G. R. Gillespie, <sup>2</sup>D. Hunter, <sup>3</sup>L. Berger & <sup>4</sup>G. Marantelli

Identifying the relative impact among different threatening processes is critical to understanding the causes of population declines. However, determining the proximal cause of declines of wildlife populations is often difficult because multiple factors may be involved and demographic population data pre- and post-decline are often inadequate (1,2).

Researchers have identified the amphibian pathogen *Batrachochytrium dendrobatidis* (*Bd*) as a major cause of amphibian declines globally (3). However, few studies have documented the real-time changes in host population dynamics and pathogen prevalence during the arrival of *Bd* and decline of amphibian populations (4). Consequently, in many instances the role of *Bd* in the decline of many amphibian species is only inferred, rather than demonstrated.

We report the decline and extinction of a population of a threatened temperate montane frog species, the Spotted Tree Frog (*Litoria spenceri*), in southeastern Australia. The Spotted Tree Frog is a stream-breeding species from the southern highlands of eastern Australia, and is listed as Critically Endangered by the IUCN Red List of Threatened Species (5). In 1992 scientists began intensive research and population monitoring to examine the species' ecology and factors responsible for its decline. Two causes of decline were subsequently implicated as primary causes of decline (6-8): habitat degradation from forestry and historic gold mining, and predation of tadpoles by introduced trout. However, in 1996 we observed a precipitous decline of one population that we were studying, in Kosciusko National Park, which ultimately went extinct. This population was unique: it was restricted to a trout-free stream, was one of few populations in protected areas; and it had a high population density (6,9). Unlike most declining species at the time, the ecology and population demography of the Spotted Tree Frog were well understood, and a monitoring program was in place, enabling prompt detection of the decline and evaluation of its cause.

Researchers had used phalange-clipping for mark-recapture studies prior and during the decline. Historically all phalanges had been histologically mounted for skeletochronological age estimation of individual frogs in the population (8). After the observed population crash, scientists scanned the large set of histological samples for *Bd*. Bayesian modelling of the pattern of change in detection of *Bd* in the phalanges (4) showed that the decline was strongly linked to the arrival and increased prevalence of *Bd*, estimated to have emerged in the population within 39 days of first detection. Our extensive ecological knowledge of this species, combined with the demographic data on this population, enabled us to confidently discount alternative explanations for the observed population extinction.

These findings are the first real-time observation of a mass die-off and subsequent population decline in a temperate Australian species, and the first precise estimate of *Bd* arrival in a frog population in Australia. The historical population declines, resulting from trout



Spotted Tree Frog *Litoria spenceri* at Bogong Creek, Kosciusko National Park in 1993. Photo: Graeme Gillespie.

predation, restricted the Spotted Tree Frog to a small geographic area at the environmental margins of its natural range, rendering it vulnerable to environmental and demographic stochastic extinction processes—in this case a disease outbreak. Therefore these findings not only provide compelling evidence that *Bd* has contributed to amphibian declines, but they also demonstrate how *Bd* may work in concert with other threatening processes, resulting in extinction.

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## Conservation and Ecology



The Critically Endangered Mountain Chicken Frog (*Leptodactylus fallax*), a species where conservation breeding is, at the moment, the most realistic hope for the species. Photo: Benjamin Tapley, ZSL.

Amphibians and conservation breeding programs: Do all threatened amphibians belong on the ark?

Benjamin Tapley, Kay S. Bradfield, Christopher Michaels & Mike Bungard

Amphibians are facing an extinction crisis and conservation breeding programs are a tool used to prevent imminent species extinctions. Compared to mammals and birds, amphibians are considered ideal candidates for these programs due to their small body size and low space requirements, high fecundity, applicability of reproductive technologies, short generation time, lack of parental care, hard wired behavior, low maintenance requirements, relative cost effectiveness of such programs, the success of several amphibian conservation breeding programs and because captive husbandry capacity exists. Superficially, these reasons appear sound and conservation breeding has improved the conservation status of several amphibian species, however it is impossible to make generalizations about the biology or geo-political context of an entire class. Many threatened amphibian species fail to meet criteria that are commonly cited as reasons why amphibians are suitable for conservation breeding programs. There are also limitations associated with maintaining populations of amphibians in the zoo and private sectors, and these could potentially undermine the success of conservation breeding programs and reintroductions. We recommend that species that have been assessed as high priorities for *ex situ* conservation action are subsequently individually reassessed to determine their suitability for inclusion in conservation breeding programs. The limitations and risks of maintaining *ex situ* populations of amphibians need to be considered from the outset and, where possible, mitigated. This should improve program success rates and

ensure that the limited funds dedicated to *ex situ* amphibian conservation are allocated to projects which have the greatest chance of success.

B. Tapley, K. Bradfield, C. Michaels, M. Bungard, *Biodivers. Conser.* (2015).



A male Red-eyed Coquí (*Eleutherodactylus antillensis*) courting a female coqui in St. Thomas, U.S. Virgin Islands. The Red-eyed Coquí is a terrestrial, nocturnal frog endemic to the Puerto Rican Bank (Puerto Rico and numerous islands and cays off its eastern coast), in the eastern Caribbean Sea.

Photo: Brittany S. Barker.

Climate as a driver of tropical insular diversity: Comparative phylogeography of two ecologically distinctive frogs in Puerto Rico

Brittany S. Barker, Javier A. Rodríguez-Robles & Joseph A. Cook

The effects of late Quaternary climate on distributions and evolutionary dynamics of insular species are poorly understood in most tropical archipelagoes. We used ecological niche models under past and current climate to derive hypotheses regarding how stable climatic conditions shaped genetic diversity in two ecologically distinctive frogs in Puerto Rico. Whereas the Mountain Coquí, *Eleutherodactylus portoricensis*, is restricted to montane forest in the Cayey and Luquillo Mountains, the Red-eyed Coquí, *E. antillensis*, is a habitat generalist distributed across the entire Puerto Rican Bank (Puerto Rico and the Virgin Islands, excluding St. Croix). To test our hypotheses, we conducted phylogeographic and population genetic analyses based on mitochondrial and nuclear loci of each species across their range in Puerto Rico. Patterns of population differentiation in *E. portoricensis*, but not in *E. antillensis*, supported our hypotheses. For *E. portoricensis*, these patterns include: individuals isolated by long-term unsuitable climate in the Río Grande de Loíza Basin in eastern Puerto Rico belong to different genetic clusters; past and current climate strongly predicted genetic differentiation; and Cayey and Luquillo Mountains populations split prior to the last interglacial. For *E. antillensis*, these patterns include: genetic clusters did not fully correspond to

predicted long-term unsuitable climate; and past and current climate weakly predicted patterns of genetic differentiation. Genetic signatures in *E. antillensis* are consistent with a recent range expansion into western Puerto Rico, possibly resulting from climate change and anthropogenic influences. As predicted, regions with a large area of long-term suitable climate were associated with higher genetic diversity in both species, suggesting larger and more stable populations. Finally, we discussed the implications of our findings for developing evidence-based management decisions for *E. portoricensis*, a taxon of special concern. Our findings illustrate the role of persistent suitable climatic conditions in promoting the persistence and diversification of tropical island organisms.

B. S. Barker, J. A. Rodríguez-Robles, J. A. Cook, *Ecography*, 38, 769–781 (2015).

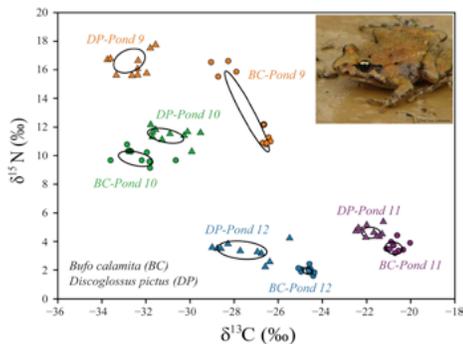
Amphibian conservation, land-use changes and protected areas: A global overview

Javier Nori, Priscila Lemes, Nicolás Urbina-Cardona, Diego Baldo, Julián Lescano & Rafael Loyola

Amphibians are undergoing a global conservation crisis, and they are one of the most underrepresented groups of vertebrates in the global network of protected areas (PAs). In this study, we evaluated the ability of the world's PAs to represent extant amphibian species. We also estimated the magnitude of the human footprint along the geographic distributions of gap species (*i.e.*, those with distributions totally outside PAs). Twenty-four percent of species ( $n = 1,535$ ) are totally unrepresented, and another 18% ( $n = 1,119$ ) have less than 5% of their distribution inside PAs. Nearly half of all species with ranges under 1,000 km<sup>2</sup> do not occur inside any PA. Furthermore, more than 65% of the distribution of gap species is in human-dominated landscapes. Although the Earth's PAs have greatly increased during the last ten years, the number of unprotected amphibians has also grown. Tropical countries in particular should strongly consider (1) the importance of using amphibians to drive conservation policies that eventually lead to the implementation and management of PAs, given amphibians' extinction risk and ability to act as bioindicators; (2) the effectiveness of national recovery plans for threatened amphibian species; and (3) the need for increased funding for scientific research to expand our knowledge of amphibian species. Meanwhile, data-deficient amphibian species should receive a higher priority than they usually receive in conservation planning,

as a precautionary measure. Throughout this paper, we point out several challenges in creating more comprehensive amphibian conservation strategies and opportunities in the next decade.

J. Nori *et al.*, *Biol. Conserv.* **191**, 367-374 (2015).



δ<sup>13</sup>C and δ<sup>15</sup>N values and standard ellipse areas for *B. calamita* and *D. pictus* in the four ponds where the species coexist (A–D). *Discoglossus pictus*. Photo: Olatz San Sebastián.

Trophic strategies of a non-native and a native amphibian species in shared ponds

Olatz San Sebastián, Joan Navarro, Gustavo A. Lorente & Alex Richter-Boix

Invasive species are, together with habitat degradation and pollution, one of the major threats to amphibians. One of the pivotal factors for understanding the successful establishment and impact of invasive species and their potential impact on native species is a thorough knowledge of how these species manage trophic resources. It is known this special importance for amphibians that usually occupy ephemeral ponds. When ponds dry larval density increase and an efficient management of temporally-limited trophic resources are important to faster development. The introduction of invasive amphibians that are generally better competitors can trigger a trophic displacement of native species to underexploited resources with consequences over their fitness. Two main trophic strategies for resource acquisition have been described, competition and opportunistic hypothesis. In order to identify the main trophic strategies of the non-native amphibian *Discoglossus pictus* and native amphibian *Bufo calamita*, in the present study we investigated whether *D. pictus* exploits similar trophic resources to those exploited by the native *B. calamita* (*competition hypothesis*) or alternative resources (*opportunistic hypothesis*). To this end, we analyzed stable isotopic values of nitrogen and carbon in larvae of both species sampled in natural ponds inhabited by both species and in ponds only inhabited by one species. Isotopic approach has achieved great advances in trophic ecology studies, providing an integrated view of resource

consumption, identifying food strategies and trophic levels of species. We also conducted a laboratory controlled-diet experiment to calculate the isotopic trophic discrimination factors for each species in order to correct interpretation of the fieldwork experiments. The similarity of the δ<sup>15</sup>N and δ<sup>13</sup>C values in the two species coupled with isotopic signal variation according to pond conditions and niche partitioning when they co-occurred indicated dietary competition. The invasive amphibian was located at higher levels of trophic niches than the native species. Also, *B. calamita* suffered an increase in its isotopic trophic niche width when it shared ponds with *D. pictus*. Moreover, invasive species showed a broader isotopic trophic niche than native species in all conditions, indicating increased capacity to exploit the diversity of resources; this may indirectly favor its invasiveness. The results of this study corroborates a previous laboratory hypothesis (the *competition strategy* by invasive species), reporting the first evidence of this species' competition ability in the field, and support the high success of this species in selected habitat by this species in its invaded range.

O. San Sebastián, J. Navarro, G. A. Lorente, Á. Richter-Boix, *PLoS ONE* **10**(6), e0130549 (2015).

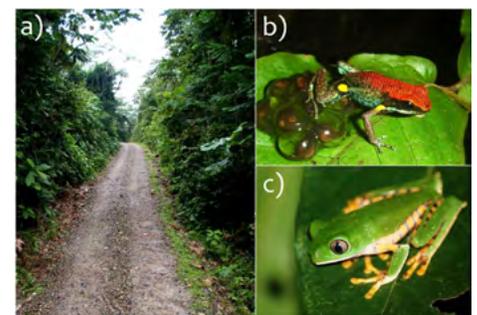
### Southern Toads alter their behavior in response to red-imported fire ants

Andrea K. Long, Daniel D. Knapp, Lauren Mccullough, Lora L. Smith, L. Mike Conner & Robert A. Mcclurey

We used the Southern Toad (*Anaxyrusterrestris*) as a model species to explore how an invasive species, the red-imported Fire Ant (*Solenopsis invicta*; hereafter RIFA), influences amphibian predator avoidance and movement behaviors. Our objective was to determine if toads spent less time near and moved more frequently in the presence of RIFAs compared to Pyramid Ants by comparing behavioral reactions of toads to RIFAs versus a control and pyramid ants versus a control. Laboratory experiments involved three treatments including no ants, RIFAs, and native pyramid ants (*Dorymyrmex burenii*) within an experimental arena. We randomly placed ants into one of two containers located at each end of the arena. For each trial we placed a toad into the experimental arena, allowed the toad to acclimate and then recorded its behavior. We calculated the proportion of time the toad spent near ants and the number of movements completed by each toad. Comparing the RIFA treatment to the pyramid ant treatment, toads spent 35% less time on the half of the experimental arena near RIFAs ( $P = 0.0304$ ). Toad movements were 1.5 times more frequent in trials with RIFAs than Pyramid Ants ( $P = 0.0488$ ). We

propose that southern toads associate RIFAs either with increased predation risk or risk of injury compared to Pyramid Ants. Although the behaviors we observed might lessen the direct effects of RIFAs on southern toads via predation and injury, the indirect effects of increased movement and avoidance of RIFAs could also influence toad fitness by decreasing reproductive and foraging success. The original copyright is given to the publication in which the material was originally published with permission from Springer Science+Business Media.

A. K. Long, D. D. Knapp, L. Mccullough, L. L. Smith, L. M. Conner *et al.*, *Biol. Invasions*, **17**, 2,179–2,186 (2015).



a) The unmarked road running through the study area (Photo: Andrew Whitworth); b) *Ameerega bilinguis*, male with eggs (Photo: Christopher Beirne); *Phyllomedusa tomopterna* (Photo: Andrew Whitworth).

### The response of faunal biodiversity to an unmarked road in the Western Amazon

Andrew Whitworth, Christopher Beirne, Jasmine Rowe, Fraser Ross, Caroline Acton, Oliver Burdekin & Philip Brown

Roads are an increasingly common feature of forest landscapes all over the world, and while information accumulates regarding the impacts of roads globally, there remains a paucity of information within tropical regions. Here we investigate the potential for biodiversity impacts from an unmarked road within a rainforest protected area in Western Amazonia. We focus on three key taxonomic groups; amphibians, butterflies and birds, each of which have been shown to be both sensitive and reliable indicators of forest disturbance. In total, 315 amphibians of 26 different species, 348 butterflies of 65 different species, 645 birds representing 77 different species were captured using mist netting and 877 bird records representing 79 different species were recorded using point counts. We provide evidence to show that the presence of a small unmarked road significantly altered levels of faunal species richness, diversity, relative abundance and community structure. This was true to a varying degree for all three taxa, up to and potentially beyond 350 m into the forest

interior. Responses to the road were shown to be taxon specific. We found increasing proximity to the road had a negative effect on amphibian and understory bird communities, while butterfly and overall diurnal bird communities responded positively. We show that the impact on biodiversity extends up to at least 32% of the whole reserve area; a serious impact under any scenario. This work provides support for recently voiced calls to limit networks of unmarked roads in order to realistically and effectively preserve natural levels of tropical biodiversity.

A. Whitworth *et al.* *Biodiversity and Conservation*, **24**, 1,657–1,670 (2015).

### Assessing the global zoo response to the amphibian crisis through 20-year trends in captive collections

Jeff Dawson, Freisha Patel, Richard A. Griffiths & Richard P. Young

**G**lobal amphibian declines are one of the biggest challenges currently facing the conservation community, and captive breeding is one way to address this crisis. Using information from the International Species Information System zoo network, we examined trends in global zoo amphibian holdings across species, zoo region and species geographical region of origin from 1994 to 2014. These trends were compared before and after the 2004 Global Amphibian Assessment to assess whether any changes occurred and whether zoo amphibian conservation effort had increased. The numbers of globally threatened species (GTS) and their proportional representation in global zoo holdings increased and this rate of increase was significantly faster after 2004. North American, European and Oceanian GTS were best represented in zoos globally, and proportions of Oceanian GTS held increased the most since 2004. South American and Asian GTS had the lowest proportional representation in zoos. At a regional zoo level, European zoos held the lowest proportions of GTS, and this proportion did not increase after 2004. Since 1994 the number of species held in viable populations has increased with these distributed among more institutions. However, as of 2014, zoos held 6.2% of globally threatened amphibians, a much smaller figure than for other vertebrate groups and one that falls considerably short of the number of species for which *ex situ* management may be desirable. Although the increased effort zoos have put into amphibian conservation over the past 20 years is encouraging, more focus is needed on *ex situ* conservation priority species. This includes building expertise and capacity in countries that hold them and tracking existing conservation efforts if the evidence-

based approach to amphibian conservation planning at a global level is to be further developed.

J. Dawson, F. Patel, R. A. Griffiths, R. P. Young, *Conservation Biology* DOI: 10.1111/cobi.12563 (2015)



Breeding Western Toads, *Anaxyrus boreas*. Photo: Steve Corn, USGS.

### Trends in Rocky Mountain amphibians and the role of beaver as a keystone species

Blake R. Hossack, William R. Gould, Debra A. Patla, Erin Muths, Rob Daley, Kristin Legg & Paul Stephen Corn

**D**espite prevalent awareness of global amphibian declines, there is still little information on trends for many widespread species. To inform land managers of trends on protected landscapes and identify potential conservation strategies, we collected occurrence data for five wetland-breeding amphibian species in four national parks in the U.S. Rocky Mountains during 2002–2011. We used explicit dynamics models to estimate variation in annual occupancy, extinction, and colonization of wetlands according to summer drought and several biophysical characteristics (*e.g.*, wetland size, elevation), including the influence of North American Beaver (*Castor canadensis*). We found more declines in occupancy than increases, especially in Yellowstone and Grand Teton National Parks (NP), where three of four species declined since 2002. However, most species in Rocky Mountain NP were too rare to include in our analysis, which likely reflects significant historical declines. Although beaver were uncommon, their creation or modification of wetlands was associated with higher colonization rates for 4 of 5 amphibian species, producing a 34% increase in occupancy in beaver-influenced wetlands compared to wetlands without beaver influence. Also, colonization rates and occupancy of Boreal Toads (*Anaxyrus boreas*) and Columbia Spotted Frogs (*Rana luteiventris*) were  $\geq 2$  times higher in beaver-influenced wetlands. These strong relationships suggest management for beaver that fosters amphibian recovery could counter declines in some areas. Our

data reinforce reports of widespread declines of formerly and currently common species, even in areas assumed to be protected from most forms of human disturbance, and demonstrate the close ecological association between beaver and wetland-dependent species.

B.R. Hossack, W.R. Gould, D.A. Patla, E. Muths, R. Daley, K. Legg, P.S. Corn, *Biol. Cons.* **187**, 260 (2015).



A male of Palmate Newt (*Lissotriton helveticus*) with conspicuous secondary sexual traits (Larzac, France). Photo: M. Denoël.

### Expression of sexual ornaments in a polymorphic species: phenotypic variation in response to environmental risk

Laurane Winandy & Mathieu Denoël

**S**econdary sexual traits may evolve under the antagonistic context of sexual and natural selection. In some polymorphic species, these traits are only expressed during the breeding period and are differently expressed in alternative phenotypes. However, it is unknown whether such phenotypes exhibit phenotypic plasticity of seasonal ornamentations in response to environmental pressures such as in the presence of fish (predation risk). This is an important question to understand the evolution of polyphenisms. We used facultative paedomorphosis in newts as a model system because it involves the coexistence of paedomorphs that retain gills in the adult stage with metamorphs that have undergone metamorphosis, but also because newts exhibit seasonal sexual traits. Our aim was therefore to determine the influence of fish on the development of seasonal ornamentation in the two phenotypes of the Palmate Newt (*Lissotriton helveticus*). During the entire newt breeding period, we assessed the importance of phenotype and fish presence with an information-theoretic approach. Our results showed that paedomorphs presented much less developed ornamentation than metamorphs and those ornamentations varied over time. Fish inhibited the development of sexual traits but differently between phenotypes: in contrast to metamorphs, paedomorphs lack the phenotypic plasticity of sexual traits to environmental risk. This study points out that internal and external parameters act in complex ways in the expression of seasonal sexual ornamentations and

that similar environmental pressure can induce a contrasted evolution in alternative phenotypes.

L. Winandy, M. Denoël, *J. Evol. Biol.* **28** (2015): 1,049-1,056. <http://hdl.handle.net/2268/180029>

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Mean body sizes of amphibian species are poorly predicted by climate

Alex Slavenko & Shai Meiri

Climate is thought to be a strong driver of animal body size evolution. Climatic gradients in body size have been documented for many terrestrial vertebrate taxa, including amphibians. However, the patterns uncovered for amphibians generally change with examined taxon and the method used in the study. Therefore, there is still disagreement on whether body sizes of amphibians display climatic clines. We examined the relationship between amphibian body size and several climatic variables, using two methods, to discern which climatic variables, if any, affect amphibian size evolution.

We collected mean body sizes of 356 amphibian species out of the 360 extant species in Europe, the USA and Canada, and tested how they are related to temperature, precipitation, primary productivity and seasonality. We used two methods. In the first, we compared the median body sizes of the amphibian assemblages inhabiting equal-area grid cells (of 96.3 km × 96.3 km). We also generated randomized assemblages to test if the observed body size distributions were likely under random assemblages. In the second method, we examined the relationship between mean species body size and the environmental predictors across their ranges, using an updated amphibian phylogeny (based on Pyron and Wiens, 2013) accounting for phylogenetic effects.

Median body sizes of amphibian assemblages in grid cells were positively correlated with temperature in urodeles and negatively in anurans. However, the observed amphibian body size distributions across grid cells were mostly statistically indistinguishable from distributions generated by random assignment of species to cells, meaning the observed size clines could simply be generated as a spurious effect of richness clines with climate. Furthermore, the phylogenetic analysis revealed opposite trends in relation to temperature in both amphibian orders, and most of the other examined climatic variables were not associated with size. What few significant relationships were retained in the models were very weak.

Richness has good explanatory power in the grid-cell analysis, and climate has low explanatory power in the interspecific analysis. Given that the interspecific analysis

probably better informs us on actual size evolution within clades, our results suggest that spatial patterns in amphibian body size likely reflect climatic diversity gradients, and climate affects amphibians more as a buffer to their distribution and not as a driver of evolution of body size.

A. Slavenko, S. Meiri. *J. Biogeogr.* **42**, 1,246-1,254 (2015).



Rock turning survey for Chinese Giant Salamanders in Fanjingshan National Nature Reserve. Photo: Benjamin Tapley ZSL.

Failure to detect the Chinese Giant Salamander (*Andrias davidianus*) in Fanjingshan National Nature Reserve, Guizhou Province, China

Benjamin Tapley, Sumio Okada, Jay Redbond, Samuel Thomas Turvey, Shu Chen, Jing-Cai Lü, Gang Wei, Min-Yao Wu, Yuan Pan, Ke-Feng Niu & Andrew Alexander Cunningham

The Chinese Giant Salamander, *Andrias davidianus*, is the world's largest amphibian. It is endemic to China and is currently listed as Critically Endangered by the IUCN. Wild populations of this species are threatened and some have already become extinct. Population declines have been attributed to habitat loss and fragmentation, and especially hunting for luxury food markets and potentially to stock salamander farms. We surveyed two river systems in Fanjingshan National Nature Reserve, Guizhou province. The reserve was established in 1978 specifically to protect *A. davidianus* as well as other threatened species. We used a variety of survey methods including night-time surveys, wading, turning substrate, netting, snorkelling, nocturnal spotlighting, and baited traps in our search for salamanders. Despite a cumulative 1,388 trapping hours, 62.7 person hours of day-time wading, turning substrate, netting and snorkelling, and 66 person hours of night-time spotlighting and snorkelling, we failed to encounter *A. davidianus* in either of the surveyed river systems. We found evidence of ongoing hunting pressure on *A. davidianus* within the reserve. Our failure to detect *A. davidianus* and the presence of ongoing poaching of this protected species within a protected area highlights the need for radically improved and strengthened

conservation management of *A. davidianus* in the reserve and potentially elsewhere in China. We suggest that this is achieved through raising the profile of *A. davidianus* in communities within the range of the species and amongst tourists visiting protected areas with historical or existing *A. davidianus* populations, as well as through regular night-time patrols of the river systems that contain *A. davidianus* by protected area staff.

Tapley *et al.*, *Salamandra* **51**, 206–208 (2015).

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High genetic connectivity in Wood Frogs (*Lithobates sylvaticus*) and Spotted Salamanders (*Ambystoma maculatum*) in a commercial forest

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

We characterized the genetic structure of two pond-breeding amphibian species in a commercial forest to evaluate population connectivity and investigate whether landscape features and timber harvest influenced dispersal and gene flow. We sampled 20 Wood Frog (*Lithobates sylvaticus*) populations and 23 Spotted Salamander (*Ambystoma maculatum*) populations across an area of 40 × 52 km. We estimated genetic diversity and differentiation, and used both a Bayesian clustering approach and a spatial autocorrelation analysis to evaluate genetic structure. We used a least-cost path analysis to examine dispersal and gene flow within each species. In both species, we found high genetic diversity and low differentiation across the study area, and the Bayesian clustering analysis identified a single genetic cluster for each species. The spatial autocorrelation analysis indicated there was greater spatial genetic structure in Spotted Salamanders than Wood Frogs. None of the landscape features measured were significantly related to genetic distance in Wood Frogs, and lakes impeded dispersal in Spotted Salamanders. We attribute the findings of high genetic connectivity in both species to a combination of abundant forest and wetlands with minimal anthropogenic disturbance. These findings suggest that current silviculture practices in the study area do not significantly impede dispersal and gene flow of pond-breeding amphibians.

S. S. Coster, K. J. Babbitt, A. I. Kovach, *Herpetol. Conserv. Biol.* **10**, 64–89 (2015).

## Evaluation of two individual identification techniques for Spotted Salamanders (*Ambystoma maculatum*)

F. Whitner Chase, Benjamin E. Hardie, Maximilian M. Kern, Leigh Anne Harden, Shannon E. Pittman & Michael E. Dorcas

Capture-mark-recapture studies are valuable to conservation decision-making as they allow for the evaluation of demographic parameters of a population. In capture-mark-recapture studies, spotted salamanders (*Ambystoma maculatum*) were marked with visible implant elastomers (VIEs), allowing for individual salamanders to be identified upon recapture. However, this elastomer coding system is expensive, invasive, and offers a finite number of codes, making it unsuitable for a long-term study. Thus, we have developed a new coding system that identifies spotted salamanders based on individuals' unique spot patterns. This study compared the two coding systems to determine the effects of both identification method and observer on identification accuracy. Over one breeding season we monitored *A. maculatum* entering and leaving a 0.5 ha ephemeral wetland in the North Carolina Piedmont using a 400 m drift fence containing 40 aquatic funnel traps and pitfall traps at 10 m intervals. Several Davidson College Herpetology Laboratory students blindly identified each individual *A. maculatum* using both spot pattern codes and VIE codes. Photographs were also taken of each individual for verification of spot pattern codes when recaptured. Identification accuracy was compared among observers, between coding systems and among captures. Our study provides recommendations for use of an identification system using spot pattern so that it can be implemented in long-term studies of *A. maculatum* and potentially adapted for use in other species.

F. W. Chase *et al.*, *Herpetol. Rev.* 46, 2 (2015).

## DNA barcoding survey of anurans across the Eastern Cordillera of Colombia and the impact of the Andes on cryptic diversity

Carlos E. Guarnizo, Andrea Paz, Astrid Muñoz-Ortiz, Sandra V. Flechas, Javier Méndez-Narváez & Andrew J. Crawford

Colombia hosts the second highest amphibian species diversity on Earth, yet its fauna remains poorly studied, especially using molecular genetic techniques. We present the results of the first wide-scale DNA barcoding survey of anurans of Colombia, focusing on a transect across the Eastern Cordillera. We surveyed 10 sites between the Magdalena Valley to the west and the eastern foothills of the Eastern Cordillera, sequencing portions

of the mitochondrial 16S ribosomal RNA and cytochrome oxidase subunit 1 (CO1) genes for 235 individuals from 52 nominal species. We applied two barcode algorithms, Automatic Barcode Gap Discovery and Refined Single Linkage Analysis, to estimate the number of clusters or "unconfirmed candidate species" supported by DNA barcode data. Our survey included ~7% of the anuran species known from Colombia. While barcoding algorithms differed slightly in the number of clusters identified, between three and ten nominal species may be obscuring candidate species (in some cases, more than one cryptic species per nominal species). Our data suggest that the high elevations of the Eastern Cordillera and the low elevations of the Chicamocha canyon acted as geographic barriers in at least seven nominal species, promoting strong genetic divergences between populations associated with the Eastern Cordillera.

C. E. Guarnizo, A. Paz, A. Muñoz-Ortiz, S. V. Flechas, J. Méndez-Narváez, A. J. Crawford, *PLoS ONE* 0(5), e0127312. doi: 10.1371/journal.pone.0127312.

## Disease and Toxicology

### Differential uptake of endosulfan in the South American toad under sublethal exposure

Gabriela V. Svartz, Damián Marino, Alicia Ronco, & Cristina S. Pérez Coll

On July 2015, we published a paper in the Archives of Environmental Contamination and Toxicology journal. The aim of this study was to evaluate the uptake of environmentally relevant concentrations of endosulfan and its correlation with differential sensitivity in the early development stages of the Common South American Toad, *Rhinella arenarum*. Agroecosystems are usually polluted with a wide variety of contaminants, with pesticides being very frequently detected. Endosulfan, an organochlorine pesticide, has been identified as a persistent organic pollutant (POP) due to its persistence, bioaccumulation, long-range transport and adverse effects to human health and aquatic ecosystems. For these reasons, the United Nations Association in 2011 decided to promote the ban of the use of endosulfan worldwide. However, despite regulations and restrictions, it is still largely used, particularly in some developing countries such as Argentina where it has been phased out just recently. Endosulfan has been shown to cause both lethal and sublethal effects on aquatic organisms such as amphibians and especially on early developmental stages. In this context, we exposed *R. arenarum* embryos and larvae to sublethal concentrations of endosulfan for several periods of exposure.

Bioconcentration factors (BCFs) for embryos significantly decreased with exposure time and concentration ( $p < 0.05$ ) reaching a maximal BCF of 1679 exposed to 1 mg endosulfan L-1 at 96 h. BCFs for larvae significantly increased with exposure time ( $p < 0.05$ ) obtaining a maximum of 40 at 504 h. In our previous study, we have reported that embryos were less sensitive than larvae to endosulfan, associated with the main tendency of embryos to bioconcentrate endosulfan as observed also in this study. The results obtained confirm the important potential uptake of endosulfan in *R. arenarum* embryo-larval development and are in line with the decision to restrict and promote the ban of its worldwide use.

G. Svartz, D. Marino, A. Ronco, C. Pérez Coll, *Arch Environ Contam Toxicol.* 69, 104 (2015).



The detection of *Bd* in the Yellow Toad (*Incilius luetkenii*) from the Honduran Isla del Tigre (Dpt. Valle) represents the first record in this species. Photo: A. Gutsche.

### New records of the chytrid fungus *Batrachochytrium dendrobatidis* in Honduran frogs

Alexander Gutsche, James R. McCranie, Torsten Ohst & Leonardo Orellana Valdés

The chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) has been implicated as a reason for the amphibian decline in Honduras. However, knowledge about the pathogens presence within the country is still poor, and up to now, it is known only in seven frog species from two localities in northern Honduras. During the rainy seasons in 2006, 2008, 2009 and 2010, we examined 23 species of frogs, three salamander and one caecilian species in 23 different localities in Honduras for the presence of *Bd*. We took skin samples which were tested by using a modified quantitative real-time (qRT) TaqMan PCR assay. Positive *Bd* records occurred in 7 of 23 (30%) localities sampled, with elevations that ranged from 10 m to 1,850 m. The new records extend the known distribution of *Bd* more than 200 km southwards in Honduras from the northern Caribbean coast. We found *Bd* in 16 of 114 (14%) specimens tested, which represented ten frog species. No positive *Bd* records occurred in the six salamanders and the one caecilian. *Bd* was detected for the first

time in the following frog species: *Craugastor laevisimus*, *C. lauraster*, *Dendropsophus microcephalus*, *Lithobates forreri*, *L. vaillanti*, *Incilius luetkenii*, *I. valliceps* and *Smilisca baudinii*. The detection of *Bd* in *Rhinella marina* represents the first Honduran record in this species. The two infected *Craugastor* species are listed as “Endangered” in the IUCN Red List of Threatened Species because of their fragmented and restricted distributions. Most of their populations have declined or disappeared at elevations above 950 m during recent years and chytridiomycosis has been assumed a possible cause. Our data confirm for the first time that *Bd* occurs in populations of both species at the upper elevations of their respective altitudinal ranges. Also, the widespread frog *Lithobates maculatus* has disappeared in recent years from certain localities, and deforestation and resulting water pollution are probably associated with this decline. We suggest adding chytridiomycosis as one potential threat, because several studies, including ours, have confirmed the widespread presence of *Bd* in this species.

A. Gutsche, J. R. McCranie, T. Ohst, L. Orellana Valdés, *Herp. Rev.* **46**(2), 202–205 (2015).



Common Frogs (*Rana temporaria*) are frequently found in urban and suburban ponds in the UK allowing citizen population monitoring. Photo by Alexandra North.

Anthropogenic and ecological drivers of amphibian disease (Ranavirosis)

Alexandra C. North, David J. Hodgson, Stephen J. Price & Amber G. F. Griffiths

Ranaviruses are causing mass amphibian die-offs in North America, Europe and Asia, and have been implicated in the decline of Common Frog (*Rana temporaria*) populations in the UK. Despite this, we have very little understanding of the environmental drivers of disease occurrence and prevalence. Using a long term (1992–2000) dataset of public reports of amphibian mortalities, we assess a set of potential predictors of the occurrence and prevalence of *Ranavirus*-consistent common frog mortality events in Britain. We reveal the influence of biotic and abiotic drivers of this disease, with many of these abiotic

characteristics being anthropogenic. While controlling for the geographic distribution of mortality events, disease prevalence increases with increasing frog population density, presence of fish and wild newts, increasing pond depth and the use of garden chemicals. The presence of an alternative host reduces prevalence, potentially indicating a dilution effect. Ranavirosis occurrence is associated with the presence of toads, an urban setting and the use of fish care products, providing insight into the causes of emergence of disease. Links between occurrence, prevalence, pond characteristics and garden management practices provides useful management implications for reducing the impacts of *Ranavirus* in the wild.

A. C. North, D. J. Hodgson, S. J. Price, A. G. F. Griffiths, *PLOS ONE.* **10**(6), e0127037 (2015).



A pair of Green Tree Frogs (*Litoria caerulea*) in amplexus in Southeast Queensland, Australia. Photo: Michel Ohmer.

Skin sloughing rate increases with chytrid fungus infection load in a susceptible amphibian

Michel E. B. Ohmer, Rebecca L. Cramp, Craig R. White & Craig E. Franklin

Amphibian chytridiomycosis, caused by the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*), is responsible for the greatest disease-driven loss of vertebrate biodiversity in recorded history. Understanding drivers of host susceptibility to this cutaneous disease is hindered by gaps in our knowledge of the host–pathogen relationship. One such overlooked aspect of susceptibility is variation in skin maintenance processes, particularly skin turnover via routine sloughing. It has been suggested that sloughing plays a role in immune defense, by removing skin-associated microbes. Thus, skin sloughing may play an important role in the pathogenesis of chytridiomycosis. To determine the relationship between skin sloughing and disease progression, we exposed adult Australian Green Tree Frogs (*Litoria caerulea*) to a local *Bd* strain and monitored sloughing rates and

individual infection loads on a naturalistic cycling temperature regime (15–23 °C). We determined sloughing rates in real-time by using an array of infrared video cameras to film frog behavior and monitored infection load before and after sloughing by swabbing and analysis with quantitative PCR. We found that sloughing rate increased with *Bd* infection load in infected frogs, but sloughing itself did not affect *Bd* load on the ventral skin surface. Furthermore, *Bd* infection did not affect the duration of characteristic sloughing behavior, and sloughing retained rhythmicity even at high infection loads. Although an increased sloughing rate might be considered advantageous for *Bd*-infected animals, it does not appear to curb the progression of disease and may actually contribute to the loss of physiological homeostasis seen in terminally ill frogs by further inhibiting water and electrolyte transport across the skin. By measuring sloughing rates directly for the first time, our results shed light on how *Bd* interacts with the physiological processes of the skin and indicate that variation in skin sloughing frequency may play a role in the observed inter- and intraspecific variation in susceptibility to disease. © 2014 British Ecological Society

M. E. B. Ohmer, R. L. Cramp, C. R. White, C. E. Franklin, *Func. Eco.* **29**, 674–682 (2015).



Adult male *Lithobates yavapaiensis* from the geothermal spring locality Muleshoe Ranch in Arizona, USA. Photo: Anna Savage.

Linking genetic and environmental factors in amphibian disease risk

Anna E. Savage, Carlos G. Becker & Kelly R. Zamudio

A central question in evolutionary biology is how interactions between organisms and the environment shape genetic differentiation. The pathogen *Batrachochytrium dendrobatidis* (*Bd*) has caused variable population declines in the Lowland Leopard Frog (*Lithobates yavapaiensis*); thus, disease has potentially shaped, or been shaped by, host genetic diversity. Environmental factors can also influence both amphibian immunity and *Bd* virulence, confounding our ability to assess the genetic effects on disease

dynamics. Here, we used genetics, pathogen dynamics and environmental data to characterize *L. yavapaiensis* populations, estimate migration and determine relative contributions of genetic and environmental factors in predicting *Bd* dynamics. We found that the two uninfected populations belonged to a single genetic deme, whereas each infected population was genetically unique. We detected an outlier locus that deviated from neutral expectations and was significantly correlated with mortality within populations. Across populations, only environmental variables predicted infection intensity, whereas environment and genetics predicted infection prevalence, and genetic diversity alone predicted mortality. At one locality with geothermally elevated water temperatures, migration estimates revealed source-sink dynamics that have likely prevented local adaptation. We conclude that integrating genetic and environmental variation among populations provides a better understanding of *Bd* spatial epidemiology, generating more effective conservation management strategies for mitigating amphibian declines.

A. E. Savage, C. G. Becker, K. R. Zamudio, *Evol. Appl.* 8, 560–572(2015).



A male Common Mistfrog (*Litoria rheocola*) calls from a rock beside a rainforest stream in Queensland, Australia. Photo: Angus McNab.

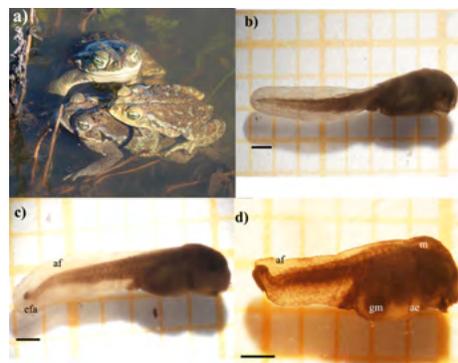
### Condition-dependent reproductive effort in frogs infected by a widespread pathogen

Elizabeth A. Roznik, Sarah J. Sapsford, David A. Pike, Lin Schwarzkopf & Ross A. Alford

To minimize the negative effects of an infection on fitness, hosts can respond adaptively by altering their reproductive effort or by adjusting their timing of reproduction. We studied effects of the pathogenic fungus *Batrachochytrium dendrobatidis* on the probability of calling in a stream-breeding rainforest frog (*Litoria rheocola*). In uninfected frogs, calling probability was relatively constant across seasons and body conditions, but in infected frogs, calling probability differed among seasons (lowest in winter, highest in summer) and was strongly and positively related to body condition. Infected frogs in

poor condition were up to 40% less likely to call than uninfected frogs, whereas infected frogs in good condition were up to 30% more likely to call than uninfected frogs. Our results suggest that frogs employed a pre-existing, plastic, life-history strategy in response to infection, which may have complex evolutionary implications. If infected males in good condition reproduce at rates equal to or greater than those of uninfected males, selection on factors affecting disease susceptibility may be minimal. However, because reproductive effort in infected males is positively related to body condition, there may be selection on mechanisms that limit the negative effects of infections on hosts.

E. A. Roznik, S. J. Sapsford, D. A. Pike, L. Schwarzkopf, R. A. Alford, *Proc. R. Soc. B.* 282, 20150694 (2015).



a) Adults of the Common Toad, *Rhinella arenarum*. (b-d) Malformed *R. arenarum* larvae as a result of Nonylphenol exposure (Stereoscopic Microscopy): (b) Control. Embryos become larvae while they are continuously exposed from the blastula stage for 168 h (c) 0.25 mg NP/L. (d) 0.75 mg NP/L. Observe the reduced body size, axial flexures (af), microcephaly (m), gut miscoiling (gm), abdominal edema (ae) and the extrusion of the fin axis (efa). Scale: 1 mm. Photos: Carolina M. Aronzon.

### Developmental toxicity and risk assessment of nonylphenol to the South American Toad, *Rhinella arenarum*

Carolina M. Aronzon, Paola A. Babay & Cristina S. Pérez Coll

On August 2014, we published a paper in *Environmental Toxicology and Pharmacology*. The aim of the study was to assess the toxicity of Nonylphenol (NP), an emerging pollutant, on two different developmental periods (embryos and larvae) of the Common South American Toad, *Rhinella arenarum*. NP is one of the major degradation products of Nonylphenol polyethoxylate, a surfactant with exceptional performance and widely used in industrial, commercial and household applications such as detergents, emulsifiers, wetting and dispersing agents, antistatic agents, demulsifiers and solubilisers. NP was stage and time dependent, as larvae were almost

six times more sensitive than embryos. The Median Lethal Concentrations (LC50) for acute (96 h), short-term chronic (168 h) and chronic exposure (336 h) were 1.06; 0.96 and 0.17 mg NP/L for embryos (exposed from early blastula), whereas for early larvae, LC50 remained constant at 0.37 mg NP/L from 96 h to 168 h, decreasing to 0.11 mg NP/L at 336 h. The No Observe Effect Concentration (NOEC)-168 h of NP exposure for embryos was 0.025 mg NP/L. The Teratogenic Potential (NOEC-lethality/NOEC-sublethal effects) was 23 times higher than 1.5, the threshold value, to be considered a high risk for embryos to be malformed in the absence of significant lethality representing a threat to the species conservation. Some of the main and non-specific sublethal effects observed in the study were delayed development, reduced body size, microcephaly, underdeveloped gills, axial flexures, different kinds of edemas, malformed mouth and adhesive structures, and gut miscoiling. The exposure to NP also caused an atypical extrusion of the fin axis. By comparing with other amphibians, the early development of *R. arenarum* was very sensitive to NP. The results of this study are very important for Argentina and other developing countries with large agricultural areas because nonionic surfactants are commonly included as wetting agents and dispersants in pesticide formulations. Despite that some active constituents of pesticides are reported of low toxicity, the additive surfactant components may be a health risk to aquatic fauna as this study shows. Moreover, the results also highlight the relevance of extending the exposure time and look for the most sensitive stage of a species for conservation purposes.

C. Aronzon, P. Babay, C. Pérez Coll. *Environ. Toxicol. Pharmacol.* 38, (2), 634–642 (2015).

# General Announcements

## Events

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

### January 2016

18–22 Amphibian Conservation Research Symposium, Potchefstroom, South Africa

### August 2016

15–21 8th World Congress of Herpetology, Hangzhou, China

### September 2016

1–10 IUCN World Conservation Congress, Honolulu, Hawai'i

## Internships & Employment

### MG1: Natural Resources

Wildlife Resources Division Headquarters, Walton County, GA. (Posted to PARC 09/17/15, Closing September 29, 2015)

### Desert Tortoise Recovery Biologist, GS-401/486-11/12

Las Vegas Fish and Wildlife Office/Desert Tortoise Recovery Office, Las Vegas, NV. (Posted to PARC 09/15/15, Closing September 24, 2015)

### M.Sc./Humboldt State University: Trophic Interactions of Frogs, Trout, and Snakes

Humboldt State University, Arcata, CA. (Posted to PARC 09/9/15, Closing September 30, 2015)

### Biological Monitoring & Sustainable Agriculture Volunteers

The Amazon Conservation Association, Andes-Amazon. (Posted to PARC 08/4/15, Open Until Filled)

### Research Fellow in bioacoustics with an interest in alligator research

Department of Natural Sciences at the University of South Carolina Beaufort, Bluffton, SC. (Posted to PARC 08/4/15, Open Until Filled)

### Biology/Hefner Museum of Natural History: Director

Oxford, OH. (Posted to PARC 07/30/15, Open Until Filled)

### The Department of Wildlife Ecology and Conservation at the University of Florida Fort Lauderdale Research and Education

## Center

Fort Lauderdale, FL. (Posted to PARC 07/6/15)

### Agricultural Conservation Coordinator, AFWA

Washington, DC. (Posted to PARC 07/2/15, Open Until Filled)

### Salamander Field Technician

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

### Department of Wildlife Ecology and Conservation at the University of Florida Fort Lauderdale Research and Education Center, Fort Lauderdale, FL

(Posted to PARC 01/08/15, Deadline for applications is 6 weeks before corresponding start date (Multiple))

### Rock Iguana Volunteer Field Assistants

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

## Funding Opportunities

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

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

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

### October 2015

### U.S. Environmental Protection Agency -- International Network for Environmental Compliance and Enforcement (INECE).

The U.S. EPA invites proposals in support of INECE, an informal international partnership promoting compliance and

enforcement of domestic and international environmental laws through networking, capacity building, and enforcement cooperation. EPA anticipates awarding one cooperative agreement from this announcement, subject to availability of funds and the quality of proposals received. The award amount is US\$750 thousand over four years. The application deadline is 05 October 2015. [Link](#)

### United Nations Development Program (UNDP) -- Public Awareness on Climate Change.

The UNDP announces a climate change storytelling contest to raise public awareness on the negative impacts of climate change as well as on the opportunities and solutions in actions by individuals and governments in vulnerable developing countries. The contest provides young journalists in developing countries a unique opportunity to contribute to the global debate on climate change in advance of COP21 in Paris later this year. The contest is open to qualified journalists 35 years of age and under from developing countries vulnerable to the impact of climate change. The closing date for entries is 11 October 2015. [Link](#)

### U.S. Department of State -- Fulbright/Clinton Fellowship 2016-2017.

The J. William Fulbright-Hillary Rodham Clinton (Fulbright-Clinton) Fellowship is a component of the Fulbright U.S. Student Program. The fellowships provide professional experience and research opportunities in public policy areas that include agriculture, energy, environment, and others. Host countries in 2016-2017 are the African Union, Burma (*Myanmar*), Cote d'Ivoire, Guatemala, Haiti, Kosovo, Malawi, Nepal, Peru, Samoa, Timor-Leste, and Ukraine. Eligibility for the fellowships is limited to U.S. citizens. The application deadline is 13 October 2015. [Link](#)

### Oklahoma City Zoo & Botanical Garden -- Conservation Grants 2015.

The Oklahoma City Zoo and Botanical Garden (USA) manages "Conservation Action Now" as a program of small grants for conservation education, scientific research, and species preservation anywhere in the world. Grants are up to US\$2,500. The application deadline is 16 October 2015. [Link](#)

### U.S. Fish and Wildlife Service -- Program for Mexico 2015.

In "Wildlife Without Borders, the U.S. Fish and Wildlife Service (USFWS) partners with Mexico's Ministry of Environment and Natural Resources (SEMARNAT) to invite projects that build Mexico's capacity for biodiversity

conservation. Applications are invited from government agencies, non-profit organizations, educational institutions, private-sector entities, and individuals. The approximate amount of funding is US\$500 thousand for an anticipated 15 awards. The deadline for applications (*English, Spanish*) is 16 October 2015. [Link](#)

**Conrad Foundation -- Spirit of Innovation Challenge 2016.** The Spirit of Innovation Challenge is a worldwide competition for youth ages 13-18 to create commercially viable products or services to address issues of global sustainability. Categories include "Energy and Environment" (*among others*). Ideas are submitted by student teams, which can be international, if desired. There is a fee for team registration. The winning teams are offered seed money to continue the development of their ideas. The application deadline is 19 October 2015. [Link](#)

**Explorers Club -- Grants for Exploration and Youth Activities 2016.** The Explorers Club invites applications for its (i) Youth Activity Fund; and (ii) Exploration Fund. The Youth Activity Fund Grant supports high school students and college undergraduates to foster a new generation of explorers. The Exploration Fund Grant is for graduate, post-graduate, doctorate and early-career post-doctoral students. The Explorers Club considers proposals in disciplines that include climate change, marine science, plants and molds, animals, conservation science, and others. There are no restrictions by nationalities or country of residence. Awards in both programs range from US\$5 hundred to US\$5 thousand. The application deadline is 19 October 2015. [Link](#)

**American Forests -- Tree Planting 2016.** Since 1990, the program "Global ReLeaf" has supported the planting of about 50 million trees in the USA and internationally for long-term environmental, economic, and social benefits. The program invites applications for tree-planting projects in 2016. Proposals must be submitted by non-profit organizations or public agencies that have expertise and experience. The program favors applicants that are able to provide matching resources. Most grants range from US\$3 thousand to US\$30 thousand. The closing date for applications is 27 October 2015. [Link](#)

**African Union -- Kwame Nkrumah Scientific Awards 2015.** The African Union (AU) honors outstanding African scientists through the Kwame Nkrumah Scientific Awards for fields that include agricultural

sciences, environmental sciences, and energy innovation (*among others*). The program seeks to recognize outstanding science at the continental level for which it awards a prize of US\$100 thousand. It also makes regional scientific awards to African women of US\$20 thousand each. The application deadline for regional scientific awards for African women is 30 October 2015. The application deadline for the continental scientific award is 15 November 2015. [Link](#)

**Technische Universität Dresden (TUD) -- Support for Masters Studies in Tropical Forestry 2016.** Germany's DAAD (the German Academic Exchange Service) will fund a limited number of scholarships for applicants from developing countries to enroll in TUD's 2-year masters program in tropical forestry. Applicants must hold a university degree in forestry, agriculture, horticulture, or other related field. The deadline to apply for DAAD scholarships is 30 October 2015. [Link](#)

**University of St. Andrews -- St. Andrews Prize for the Environment 2016.** The annual St Andrews Prize for the Environment recognizes significant contributions to environmental conservation in the developing world. Projects that have won in the past include subjects such as water management; agriculture and food security; by-products from waste; renewable energy; and others. The Prize consists of an award of US\$100 thousand and a medal. Awards of US\$25 thousand are presented to each of two other finalists. The deadline for applications is 31 October 2015. [Link](#)

**Whitley Fund for Nature -- Whitley Awards 2016.**

The Whitley Fund for Nature (WFN) offers "Whitley Awards" to outstanding biodiversity conservation leaders in developing countries around the world. The awards are both an international prize and a form of project funding, currently £35 thousand over one year. Whitley Awards are open to biodiversity conservation leaders working in countries or regions of which they are nationals, and that are not defined as high-income economies by the World Bank (with exceptions). The application deadline is 31 October 2015. [Link](#)

**November 2015**

**Aspen Institute -- New Voices Fellowship 2016.** The New Voices Fellowship is a year-long program in media skills, communication, and leadership for top development professionals in the developing world. Fellows are expected

to have both a record of significant professional achievement and a desire to share their perspectives on global development with a broader international audience. The Aspen Institute aims to select 20 fellows who will write opinion articles, participate in interviews with local and international media, and speak at international conferences. Applications are welcome from all developing countries, and from subject areas including all those relevant in the Terra Viva Grants Directory. A particular priority for 2016, among others, is food security. The deadline for nominations is 01 November 2015. [Link](#)

**Field Museum -- Visiting Scholarships and Graduate Fellowships.** The Field Museum (Chicago, USA) supports basic research on its collections by interested students and scholars throughout the world. The Museum offers a modest number of grants and fellowships, including funding for short-term visits of up to three months for collection-based research studies. Grants to examine specimens in the Museum's collections are open on a competitive basis to all individuals in the national and international scholarly community working on problems related to natural history. The deadline to apply for the visiting scholarships is 01 November 2015. The deadline for graduate fellowships is 30 January 2016. [Link](#)

**Morris Animal Foundation -- Wildlife Health and Welfare 2016.** The Morris Animal Foundation supports research on animal health and welfare, including wildlife/exotics. The Foundation invites proposals in several categories (i.e., established investigator; first award; fellowship training; pilot study). The application deadline for wildlife/exotics is 18 November 2015. (*Note: The Foundation also manages a wildlife rapid response fund that has no calendar deadlines.*) [Link](#)

**Scottish Government -- International Development Small Grants 2016.** The Scottish Government's International Development Small Grants Programme provides project funding in support of the government's International Development Policy. Applications for grants are invited from incorporated not-for-profit organisations which have a presence in Scotland and an annual turnover of less than £150 thousand. Projects must focus in the following priority countries: Malawi, Rwanda, Tanzania, Zambia, Pakistan, Bangladesh and the Indian States of Bihar, Madhya Pradesh and Orissa. Awards are a maximum of £60 thousand for project grants over three years, or a maximum of £10 thousand for feasibility and capacity

building grants over one year. The application deadline is 25 November 2015. [Link](#)

#### December 2015

**International Zoo Educators Association -- Sponsored Delegate Program.** The International Zoo Educators Association funds conservation educators from developing countries to attend its biennial conference. A limited number of professional development grants to attend the IZE conference are available every other year. Each conference grant includes support for conference registration, airfare, accommodations, some meals, and IZE membership for two and half years. The application deadline is 01 December 2015. [Link](#)

#### January 2016

**Conservation, Food, and Health Foundation (CHF) -- Grants for Grassroots Development.** The CFH Foundation makes grants to nonprofit organizations worldwide for projects in conservation, sustainable agriculture, and health in developing countries. The average grant is approximately US\$20 thousand. The deadlines for concept applications are 01 January and 01 July of each year. [Link](#)

**Wild Gift -- Fellowship Support for Young Social Entrepreneurs on Climate Change.** The Wild Gift Network invites applications for a new class of social entrepreneurs to join its network. The program is open to applicants ages 21-35 in the USA and Canada. The selected leaders will be supported for projects anywhere in the world on the themes of climate adaptation and climate change mitigation. Grants are up to US\$10 thousand for projects of 16 months. Applicants must be able to participate in a three-week wilderness orientation and training session in Idaho, USA. The application deadline is 01 January 2016. [Link](#)

**World Wide Fund For Nature (WWF) -- Prince Bernhard Scholarships for Nature Conservation 2016.** WWF supports professional training and formal studies of individuals working in disciplines directly relevant to nature conservation. Eligibility extends to mid-career nationals from Africa; Asia and Pacific; Latin America and Caribbean; Eastern Europe; and the Middle East -- including WWF staff, or candidates working as partners with WWF. The maximum grant is CHF 10 thousand for studies or training lasting one year or less. The deadline for applications (*English, French, Spanish*) is 05 January 2016. [Link](#)

**Harvard University -- Environmental Fellowships 2016.** Harvard University's Center for the Environment will award six environmental fellowships for 2016. The fellowships enable recent doctorate recipients to use Harvard's resources to tackle complex environmental problems. Eligibility for funding extends to candidates with a doctorate or equivalent in any subject area from any university in the world. Moreover, candidates may propose research projects in any discipline. The fellowships are US\$62 thousand per year, in addition to other benefits. The deadline for applications is 13 January 2016. [Link](#)

**Alexander von Humboldt Foundation -- Georg Forster Research Award.** The Georg Forster Research Award supports the work of accomplished researchers who are expected to continue to develop research-based solutions to specific challenges facing transition and developing countries. Nominees must be nationals of a developing or transition country, excluding the People's Republic of China and India. The Foundation particularly encourages the nomination of qualified female researchers. The award amount totals €60 thousand. Additionally, award winners are invited to conduct a research project of their own choosing in Germany in close collaboration with a specialist

colleague. To support the collaboration, the Foundation may grant additional funding of up to €25 thousand. The deadline for nominations is 15 January of every year. [Link](#)

**Maypole Fund -- Small Grants to Women.** The Maypole Fund takes an international perspective in supporting women in non-violent and politically expressive projects in subject areas that include environmental issues, among several others. The Fund gives priority to small women's groups and individual women. Grants are up to £750. The application deadlines are 31 January and 30 June of each year. [Link](#)

#### February 2016

**American Philosophical Society -- Lewis and Clark Fund for Exploration and Field Research 2016.**

The Fund supports doctoral students to collect specimens and data in disciplines relying heavily on field studies, e.g., including biology, ecology, geography, and others. Applicants from the USA may use the grants for research anywhere in the world. Applicants from other countries must be based at an institution in the USA, or carry out their work in the USA. The grants are up to US\$5 thousand. The next closing date for applications is 01 February 2016. [Link](#)

## FrogLog Schedule

January – Special Edition

April – The Americas

July – Africa, West Asia, Madagascar, Mediterranean and Europe

October – Asia, Russia and Oceania



## INSTRUCTIONS TO AUTHORS

### Background

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

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

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

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

### PUBLICATION

*FrogLog* is published online at: [www.amphibians.org](http://www.amphibians.org) and is Open Access.

### REVIEW

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

### PRODUCTION EDITOR

Candace M. Hansen-Hendrikk: [cmhansen@amphibians.org](mailto:cmhansen@amphibians.org)

### EDITORIAL COMMITTEE

Candace M. Hansen-Hendrikk  
Craig Hassapakis  
Lindsay Renick Mayer

Additional reviewers will be requested as required.

### SUBMISSION OF MANUSCRIPTS

Manuscripts can only be received as electronic files. Text should be submitted in MS Word format and may contain tables, but figures should be sent as a separate attachment where possible. All documents should be sent to [froglog@amphibians.org](mailto:froglog@amphibians.org). Each file should be labeled in a style that illustrates clear association, i.e., authors\_name\_ms and authors\_name\_figure1.

### GUIDELINES FOR AUTHORS

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

### TITLE

Titles should ideally be no more than 15 words.

### AUTHORS

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

### MAIN BODY OF TEXT

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

### AUTHOR DETAILS

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

### FIGURES

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

### TABLES

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

### CITATION OF LITERATURE

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

#### Journals/Periodicals

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

#### Books

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

#### Technical reports

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

#### Paper presented at a meeting

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

#### Published Online Only

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

#### Web site

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

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

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

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