Issue 121 (March 2019)

ISSN: 1026-0269 eISSN: 1817-3934

Volume 27, number 1

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

### Haiti's First-Ever Private **Nature Reserve**

The Rediscovery of the Allipacca Water Frog

The Family Pond

... and so much more!

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# FrogLog Contents



**5** Editorial

### NEWS FROM THE AMPHIBIAN COMMUNITY

- 4 Haiti's First-Ever Private Nature Reserve Will Safeguard Treasure Trove of Imperiled Species
- 6 The Family Pond: A Creative Approach to Amphibian Conservation Awareness
- 7 Citizen Conservation: Reinforcing the One Plan Approach
- 8 Jewels Of The Neotropics: A Documentary to Save Poison Frogs
- 10 #10YearsChallenge: A Decade Working Towards Conservation of the Argentinean Marsupial Frogs
- 12 The Rediscovery of the Allipacca Water Frog (*Telmatobius intermedius*) in the Western Andes of Peru
- 15 Unexpected Sources of Food for Amphibians Living Near Humans

- 18 Conservation Strategies and Participatory Monitoring of Threatened Amphibians on Peace Implementation Territories in Southwestern Colombia
- 23 One or Multiple Origins of Midwife Toads in the UK?
- 24 9th CBH: Brazilian Congress of Herpetology Campinas, Sao Paulo
- 25 'Save All Frogs' Initiative Launched
- **26** Best Practices of Amphibian Conservation in the Roman Countryside
- 29 Comments on the Collection Of Buccal Swabs From Amphibians

Recent Publications 31 | Author Instructions 36

### Editorial

Dear friends,

he *FrogLog* team and all of the contributing authors are excited to bring you this latest edition of FrogLog. As always, it highlights recent amphibian research developments, as well as incredible conservation success stories from around the world, all in one place. It is indeed becoming the magazine we have always been working towards: a publication that is both exciting and accessible to not just the scientific community, but also amphibian enthusiasts from all walks (hops?) of life.

As you flip through these pages, you will read about the recent establishment of the first private nature reserve in Haiti, with the final acquisition of more than 1,200 acres on Grand Bois Mountain. This news is particularly exciting for amphibians because this area is home to many species threatened with extinction, including the Critically Endangered Spiny Green Frog (Eleutherodactylus nortoni).

You will also take a step back in time to see how a quick stop in the market of Puquio, a town in the province of Lucanas on the Pacific slope of the Peruvian Andes, led to the rediscovery of the Allipacca Water Frog (*Telmatobius intermedius*). You will go even further back in time to trace the steps towards conserving the missing, forgotten and rediscovered Argentinean Marsupial Frogs (this sounds like a great Indiana Jones storyline, doesn't it?).

Did you know that a simple family activity can have an impact on amphibian conservation and can create a ripple effect across generations? It is an activity that can also make your family more environmentally active and aware, all while having fun together as a family! When you find yourself wondering "What can we do today?" the answer may be as simple as painting a rock.

You will join scientists as they move into the realm of molecular analysis to discover where the different Midwife Toad populations in the UK originated. It had long been assumed that all British Midwife Toad populations have a French origin, but a preliminary analysis suggests otherwise. Intrigued? Read on!

I hope you enjoy this edition, and please do drop me a line if you have an update or story that you would like to share with our readers!

### Candace Hansen-Hendrikx

Editor-in-Chief, FrogLog Director of Operations, Amphibian Survival Alliance



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### NEWS FROM THE AMPHIBIAN COMMUNITY



### Haiti's First-Ever Private Nature Reserve Will Safeguard Treasure Trove of Imperiled Species

### By Lindsay Renick Mayer

n an historic move, Global Wildlife Conservation (GWC) has joined forces with several partners to establish the first private nature reserve in Haiti. The project—in partnership with Rainforest Trust, Temple University, Haiti National Trust and local NGO Société Audubon Haiti (SAH)—launched this week with the final acquisition of more than 1,200 acres on Grand Bois mountain, home to a remarkable 68 vertebrate species. This includes many species threatened with extinction, such as Ekman's Magnolia Tree, the Yellow Cave Frog and the Tiburon Streamfrog.

"With less than one percent of Haiti's original forest left, the country is on the verge of a potential ecological collapse," said GWC Chief Scientist and CEO Wes Sechrest, who is also on Haiti National Trust's board of trustees. "We knew that we needed to take action to protect the country's staggering diversity of unique and threatened species, many of which are found only in Haiti. Global Wildlife Conservation has partnered with Haiti National Trust to directly protect, manage and restore this high-priority conservation site in an effort to begin to turn the tide of centuries of unregulated environmental destruction."

S. Blair Hedges, director of Temple University's Center for Biodiversity, in collaboration with Société Audubon Haiti President Philippe Bayard, led two expeditions to Grand Bois during the last seven years. The team documented 68 species of vertebrates, including 16 amphibian species, giving this area the distinction of being home to one of the largest groupings of frog species anywhere in the Caribbean.

One of the species, the Tiburon Streamfrog, had not been seen in four decades despite intensive search efforts. The Stream frog is a unique lost species that made an evolutionary reversal to an aquatic lifestyle after its ancestors evolved traits for living in the forest. The expedition team discovered three new frog species, all of which the IUCN Red List of Threatened Species will likely classify as Critically Endangered once they have been scientifically described.

Mirroring what is happening across Haiti, Grand Bois's forests are being cut for building materials, slash-and-burn agriculture and charcoal. At least 50 percent of the original forest on Grand Bois, however, is still intact above 1,000 meters. The expedition identified Grand Bois as a biodiversity hotspot and in response, the government of Haiti declared it a national park in 2015, Parc National Naturel de Grand Bois, recognizing it as a priority for conservation and confirming the critical need to acquire and protect the area. The local community has been incredibly supportive of maintaining the natural 'water tower' of the forested mountain,



as nearby peaks have been deforested, resulting in landslides and lack of controlled and clean water in natural forests.

"That Grand Bois and two other areas were named as national parks based on our work has been very gratifying," Hedges said. "Now with funding from Global Wildlife Conservation and Rainforest Trust, we are beginning the process of land purchase and management to build a network of private nature reserves in the country."

Repeated scientific expeditions have identified Haiti's 12 remaining hotspots for wildlife diversity, of which Grand Bois was considered the highest conservation priority because of existing forest habitat and imminent threats. Haiti National Trust is working to implement a forest management and restoration plan for Grand Bois Nature Reserve, with funding from GWC and Rainforest Trust, and to raise support to build the network of private nature reserves.

"When I first landed on Grand Bois mountain with Professor Hedges, I immediately thought that a new strategy had to be found to preserve the plants, animals, and ecosystem services such as clean water and protection from flooding," said Bayard who, together with Hedges, founded Haiti National Trust to preserve the country's natural environment and biodiversity. "The species will never come back if we lose them."

Grand Bois is found in Haiti's Massif de la Hotte mountain range, the number one priority conservation site in the country and one of the most important sites for amphibians in the world. Because 19 Critically Endangered amphibian species are restricted to this single site globally, Massif de La Hotte has been recognized as an Alliance for Zero Extinction site. It is also a Key Biodiversity Area, a nationally identified site of global significance for biodiversity.

This work was made possible by a generous donation from the Sheth Sangreal Foundation.



The Critically Endangered Tiburon Streamfrog, on Grand Bois Mountain, Haiti. Photo: J. Hoppe.

### The Family Pond: A Creative Approach to Amphibian Conservation Awareness

By Thomas Favazza

t can be simple, fun, and rewarding being an ambassador for our often overlooked friends: the amphibians! But with a little creativity, you and your family can make a big splash!

Enjoyable family activities can have an impact on amphibian conservation and can create a ripple effect across generations. It can bring the common interest of conservation into your personal relationships. It can also make your family more environmentally conscientious and contribute to amphibian conservation all while having family fun! When you find yourself wondering, "What can we do today?" the answer may be as simple as painting a rock.

A campaign called *Amphibian Conservation Rocks* takes this idea head-on. This family-oriented campaign was introduced in 2018 by Trinity Favazza, an eleven-year-old with a passion for amphibian conservation and awareness. Trinity has found a unique way to bring families together using a fun and creative approach to spread the word about amphibian conservation. Finding a way to reach and impact the younger generation is the key to sustainability in a movement to protect our natural resources. This campaign brings families together with a simple, enjoyable and effective activity, and it *toad-ally* rocks!

The *Amphibian Conservation Rocks* campaign encourages families to paint amphibian-themed rocks together. It is an excellent activity for those days when the weather keeps you indoors. You can add simple information, such as a specific species status, a fact, or just a word like "conservation" to your rocks. Consider tying cards onto your rocks with more information if the mood strikes you.

The bottom line is to try to share your artistic creations along with some fun and interesting amphibian facts. This will inspire those who find your rocks to tell their families and friends about the cool rock they found! Seeing this painted rocks campaign becoming a trend with families can make for a huge leap forward in the future of amphibian conservation awareness! The idea is to leave your painted rocks around your community for others to find, to remind them of our amphibian resources, to share information with family, and friends and to inspire others to do the same. You may even consider getting your local nature centers, zoos, and/or schools involved.

Taking the time to post photos of your *Amphibian Conservation Rocks* on social media is a great way to make new friends and help introduce this campaign, one rock at a time. Don't be afraid to show your creativity, no matter your skill level...the amphibians will always love what you are doing.

Spending time together, outdoors, hiding your painted amphibian rocks at local hotspots and parks makes for great family time. Invite your friends and their children to paint with you and even suggest it as an activity for your babysitter while you are out. Simply researching the amphibians you are painting and writing a little bit about them will lead you and your children to become ambassadors for amphibian conservation! As your knowledge grows, you will be more driven and better-equipped to share useful information with others. If you instill that passion in others, it will grow exponentially.

The more we share our love and concern for amphibians, the greater the impact we make. Just remember to have fun, paint amphibian rocks, spend time with your family, be creative, and spread the word, ribbit, ribbit, ribbit...

"The Family Pond is a segment that our family will continue to fill with fun and creative Ideas, to reach others... one pad at a time until the pond is full," said Thomas Favazza.





Photos: Thomas Favazza.

6 | FrogLog 27 (1), Number 121 (March 2019)

### Citizen Conservation: Reinforcing the One Plan Approach

By Björn Encke

e all know about the devastating predictions regarding species extinction. And we also know that we'll only succeed in slowing down the grim trend if all stakeholders committed to conservation work together in a cooperative and open-minded way.

From the very beginning, Frogs & Friends has collaborated closely with zoos and private amphibian enthusiasts. These two stakeholders have a wealth of knowledge and experience in captive breeding. Zoos have been working for decades to establish well-coordinated, scientifically led breeding programs for a variety of species, while private experts have built up expertise in fostering a variety of species.

New insights prompt new ideas. In the light of the massive extinction event we're facing, and in the spirit of the One Plan approach, we are convinced that captive breeding programs must be an essential part of any strategy to preserve biodiversity. That is why Frogs & Friends teamed up with the VdZ (Verband der Zoologischen Gärten) and DGHT (German Society for Herpetology) to jointly launch what we see as the next important step: the Citizen Conservation project.

Citizen Conservation aims to help solve two major problems:

First, extinction of species: If we want to save a relevant number of threatened species, we'll have to quickly expand the amount of available space and the number of knowledgeable caretakers. Zoos won't be able to achieve this goal on their own. Second, public misconceptions about captive breeding: Zoos and private breeders are facing significant opposition at both a societal and a legislative level. If we want to safeguard our potential to help solve the existing challenges in the field of biodiversity loss, we have to convince society of the importance of our work.

Citizen Conservation, therefore, has to be two things from the very beginning: a breeding program and a campaign. We have to make it clear that captive breeding is part of the solution, not part of the problem – and we'll have to prove it.

For the last three years, we've been busy with preparations, interviewing dozens of captive breeding stakeholders, marketing professionals, and media experts to create a viable framework for the project to succeed. One key insight was that almost all stakeholders suffered from a perceived lack of acknowledgment for their efforts by the public and even by the conservation community. Consequently, reframing this upside-down perception of captive breeding must be the key target of the communication campaign.

With Citizen Conservation #Amphibians we have now started a five-year pilot phase that will help us test and improve our organizational structure and procedures, management and participation guidelines, marketing and communication strategy and, of course, funding. This process of evaluation and adjustments will give us a solid base on which to build as we scale up Citizen Conservation both geographically and taxonomically.

The program currently includes five species. We have plans to expand to between 10 and 15 species within the next four years. Meanwhile, we hope to be able to open up the first Citizen Conservation Centers in zoos. These will serve as training centers, breeding facilities and give volunteers a chance to get involved. It is a quite ambitious agenda. However, we are optimistic that it will soon be clear that the benefits far outweigh the obstacles--and that Citizen Conservation will demonstrate its potential to effectively contribute to our shared task: finding effective ways to slow the extinction crisis.



Showing a stiff upper lip: Captive breeding is part of the solution, and we're doing it. Period. Doris Preininger and Thomas Wampula from Tiergarten Schönbrunn Vienna. Photo: Frogs & Friends/Benny Trapp.



Preparing for an emergency: Uwe Seidel and Philip Gerhard have developed a fire salamander breeding system that optimizes the number of healthy offspring. This knowledge could help save local Fire salamander populations threatened by *BSal*. Photo Frogs & Friends/Benny Trapp.





### Jewels Of The Neotropics: A Documentary to Save **Poison Frogs**

### By Andrés Piolatti

8

s recent research suggests, the skin of many frogs is a potential global pharmacopeia for the treatment of certain medical conditions such as pain or Alzheimer's Disease(1). It is this characteristic, a remarkable skin, that draws attention to anyone who runs into a frog of the Dendrobatidae family. Commonly known as Poison frogs, many of the species in this large family have a very vivid skin color that warns its predators of highly toxic



Ranitomeya imitator climbing the tripod upwards at Río Bosque Mágico, Chazuta, Peru. Photo: Andrés Piolatti.

or, at least, irritant substances present in the outer layer of their bodies. This phenomenon, called aposematic coloration(2), is what hypnotized many people like us, germinating in our interior a real passion and dedication for these delicate beings.

In March 2016, during a trip to Panama in search of the different morphotypes of Oophaga pumilio in their natural environment, we had the opportunity to observe how their habitat was being destroyed. Trees 30 meters high, whose trunks were home to countless lives were being cut down to open new routes through almost virgin forest to build houses to the increasing demand for accommodations for wealthy people who wanted a home in paradise. A paradise that, ironically, was being destroyed to (un)welcome them. With each tree felled, hundreds of species including plants, invertebrates, birds, reptiles and amphibians, were stripped of their true Eden forever. In the depth of that panorama, we continued to hear the cry of the Howler Monkeys (Allouatta palliata) and the claims of the Strawberry Poison Frog (Oophaga pumilio). Our perspective changed during that trip. We could not help feeling the weight of a gray pessimism on our backs. Towards the end of our trip, we realized that there was some hope: a biological station, a recycling plant in the middle of the main island and even an ecological coffee farm whose owner had deliberately increased the number of bromeliads in his lands to favor the reproduction of dendrobatids in the area. Something could be done, and we were



Tadpole at late metamorphosis at Jambatu Center for Amphibian Research and Conservation. Photo: Andrés Piolatti.



Carlos Galvis and his collaborator unpacking just-arrived *Phyllobates terribilis* individuals from another institution. Exchange of individuals guarantees genetic variability of captivity populations. Photo: Andrés Piolatti.

willing to contribute. At that time, we started recording what we saw and what we would like to be changed. Maybe a post on a social network could work, perhaps a campaign on any platform could gain enough strength... but why not to propose a crowdfunded project that unites poison frogs fans and those with initiatives and active projects to save these very specific amphibians? And that's how *Poison Dart Frogs, The Documentary Project* was born. It has now gone further than we could have imagined at that time.

Thanks to the support received by all those people who believed that telling this story could provide that grain of sand so fundamental for changes to be possible, we undertook an unprecedented trip in our lives with the ultimate goal of making an audiovisual project to show how different initiatives are saving dendrobatids: from BioTrade as an alternative to black and grey amphibian markets(3), to *ex situ* and *in situ* conservation at amphibian arks and private reserves, respectively.

#### Acknowledgments

I am grateful to all our 101 Kickstarter backers. Thanks to Brian Kubicki (Director of C.R.A.R.C), Candace M. Hansen-Hendrikx (Director of Operations Amphibian Survival Alliance), Carlos Galvis (Head of the Section of the Biology Area of the Zoological Foundation of Cali), Dendrobates España, Giovanni Chaves (Fundación Ecodiversidad Colombia), Giovanni Onore (Founder of Otonga Foundation), Josh's Frogs, La Y Griega, Lola Guarderas (General Manager of Wikiri), Luis Coloma (Director of Jambatu Center) and Mist King. This project would not be possible without Jane Goodall's reasons for hope.

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Different stages of deforestation at the Peruvian Amazon, near Barrio Florido. Photo: Andrés Piolatti.



### #10YearsChallenge: A Decade Working Towards Conservation of the Argentinean Marsupial Frogs

By Mauricio S. Akmentins<sup>1</sup>, Laura C. Pereyra<sup>1</sup>, Martín Boullhesen<sup>1,2</sup> & Marcos Vaira<sup>1</sup>

t has been ten years since the beginning of this journey, and when we look back, maybe this story could well be entitled like a spaghetti western movie: "the missing, the forgotten and the rediscovered."

An early warning signal of the conservation problems faced by the Argentinean species of marsupial frogs emerged in the year 2000 during the first assessment of conservation status of the Argentinean amphibian species as part of the National Red List (1). Due to their restricted geographic ranges within the montane Yungas Andean forests and the accelerated rate of habitat loss, these three species also were placed in threat categories in the first global amphibian assessment in 2004 (2). An anecdotic report that went unnoticed for a few years, made visible the extirpation of a breeding aggregation of the Calilegua's Marsupial Frog (*Gastrotheca christiani*) near Calilegua National Park (3). This event gained extreme relevance in the following years when specialists realized

<sup>1</sup>Instituto de Ecorregiones Andinas (INECOA), Universidad Nacional de Jujuy – CONICET, San Salvador de Jujuy, Argentina. <sup>2</sup>Programa de Investigaciones de Biodiversidad Argentina (PIDBA), Facultad de Ciencias Naturales e Instituto Miguel Lillo, San Miguel de Tucumán, Argentina. that the last record of this species in the wild was in the year 1996 (4). This lack of registries was also extended to the other two species, *G. chrysosticta*, and *G. gracilis*, which had suddenly vanished since 1993 and 1991, respectively (4).

Considering this background, in 2008 we initiated an intensive fieldworks effort to obtain new registries of the three endemic species of Argentinean marsupial frogs. Thus, we developed a conservation program employing a mixed approach that included scientific research and conservation projects.

The Calilegua's Marsupial Frog is the flag species of the Argentinean Marsupial Frog Conservation Program and one of the "top ten" most wanted lost amphibians of "The Search for Lost Species" initiative of Global Wildlife Conservation. For this reason, we centered our efforts on finding extant populations of *G. christiani*, as a keystone for any future conservation effort. This situation also intensifies our concern about the current conservation status of the species, which is still missing despite the increased search effort. Field search not only include the classic active monitoring techniques of visual and acoustic encounter surveys but also include the use of passive monitoring techniques such as artificial cover ob-

10 | FrogLog 27 (1), Number 121 (March 201

jects/artificial refuges and the deployment of automated recording devices combined with species recognition software, to increase the detection chances of marsupial frogs.

The first remarkable achievement of our conservation program was the rediscovery of La Banderita Marsupial Frog (Gastrotheca gracilis) in the year 2011, and the following detection of more populations of this endangered marsupial frog in Campo de Los Alisos National Park in 2013 (4, 5). Despite the advance that meant to rediscover this species after the absence of registries in the wild for 20 years, not all was good news for this species. In 2014 we reported a new threat for G. gracilis in one of the rediscovered populations in Los Sosa Provincial Reserve due to direct predation and reproductive habitat alteration by domestic pigs (6). After these findings, the species remained relatively unnoticed, and no immediate measures for monitoring or improving its conservation status were performed in the following years. Last year we started new efforts to reverse this situation through an *ex situ* conservation project for population supplementation of G. gracilis in Los Sosa Provincial Reserve. The project is being developed in the Horco Molle Experimental Reserve of Tucumán National University with the Amphibian Ark support. Also, the recent announcement of the creation of the Aconquija National Park could expand the protected geographic range of *G. gracilis*. Nonetheless, we still need to confirm the presence of this species in this new natural protected area.

Another remarkable achievement was the rediscovery of Baritú's Marsupial Frog (*Gastrotheca chrysosticta*) last year after 25 years without registries in the wild (8). After this important finding, we started to work in collaboration with Baritú's National Park authorities for establishing a long-term monitoring project of the rediscovered population and the active search of this species in the historical geographic range to determine its current conservation status.

The publication of the Conservation Action Plan for the Amphibians of Argentina last year gave a framework to our conservation program and other conservation efforts in the country (9). This publication is a watershed for Argentinean amphibian conservation since it offered conservationists and decision makers a guide to conduct and prioritize their conservation actions.

Finally, we want to thank for the continuous support of our partners: Amphibian Ark, Amphibian Survival Alliance, Conservation Leadership Programme, CONICET, Global Wildlife Conservation, Stiftung Artenschutz, National Park Administration of Argentina, The Mohamed bin Zayed Species Conservation Fund, and The Rufford Small Grants Foundation.

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"The missing picture." A representative anuran assemblage of Calilegua National Park, Jujuy province, Argentina, modified from Vaira 2002 (9). First row: Leptodactylus chaquensis, L. elenae, L. fuscus, L. gracilis, L. latinasus. Second row: Boana riojana, Phyllomedusa sauvagii, P. boliviana, Scinax fuscovarius, S. nasicus. Third row: Trachycephalus typhonius, Telmatobius oxycephalus\*, Gastrotheca christiani, Oreobates berdemenos, O. barituensis. Fourth row: Rhinella arenarum, R. gallardoi, Melanophryniscus rubriventris, Rhinella rumbolli, R. dypticha. Fifth row: Pleurodema borellii, P. tucumanum\*, Physalaemus cf. albonotatus, P. biligonigerus\* and Odontophrynus americanus. Photos: Mauricio S. Akmentins and (\*) Laura C. Pereyra.



Adult male of *Gastrotheca chrysosticta* of the rediscovered population in Baritú National Park, Salta province, Argentina. Photo: Mauricio S. Akmentins.



# The Rediscovery of the Allipacca Water Frog (*Telmatobius intermedius*) in the Western Andes of Peru

By Víctor J. Vargas G.<sup>1,2,3</sup>, Pablo Najarro<sup>2</sup>, Marco Rivera<sup>2</sup>, Oscar Chipana<sup>2</sup>, Vladimir Díaz<sup>2</sup>, Kevin Jaico<sup>2</sup> & Alessandro Catenazzi<sup>5,4</sup>

he story of our rediscovery begins in December of 2014, when we were surveying the habitat of the Andean condor (Vultur gryphus), in the Sondondo valley, a community of Chipao in the southern Andes of the Peruvian department of Ayacucho. At the end of our work, and in preparation of our return to Lima, we made a quick stop in the market of Puquio, a town in the province of Lucanas, in the Pacific slope of the Peruvian Andes, and the type locality of the Allipacca water frog (Telmatobius inter*medius*). We met a local seller of frog juice, a concoction of blended and liquified water frogs which is believed to have medicinal properties. We chatted with the seller, and after he showed us the frogs he used as juice ingredients, we realized that the frogs were individuals of Telmatobius, unlike other species we were familiar with. The seller also gave us tips concerning the likely source of these frogs, namely the small creeks of Allipacca near Puquio where children captured the frogs used for his market stand.

Armed with this information, we hiked to Allipacca and explored a couple of creeks, and quickly discovered tadpoles and frogs of the same Telmatobius species we had seen at the market stand. After comparing these specimens with a description in the literature,

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12

and the type specimens preserved at the natural history museum in Lima, we were able to identify this species as the Allipacca Water Frog *T. intermedius*, a species described by J. Vellard in 1951 and not reported again after that date.

Water frogs in the family Telmatobiidae are typical of the South American Andes, where 61 species are currently recognized of which 25 occur in Peru (2). These frogs are distributed in Chile, Bolivia, Peru, and Ecuador, from about 1800 m a.s.l. (Aguilar, 2010)



Allipacca stream, habitat of *Telmatobius intermedius*. Photo. Victor Vargas.



to the level of Andean glaciers around 5400 m asl. They live in ecosystems such as páramos, Andean subpáramos, the dry or humid puna, cloud forests, and other high elevation habitats. Species of Telmatobius are of semi-aquatic or exclusively aquatic habits (Lehr, 2005.). T. intermedius was previously known only from four specimens collected from its type locality in Allipacca (5). Mr. F. Blancas first collected these frogs in 1950. Because the species had not been reported again following its description, the conservation status of this species was unknown. Therefore, Peruvian legislation (Supreme Decree No. 004-2014-MINAGRI) and the IUCN (until 2018) classified T. intermedius in the category of Data Deficient (DD) (6). Despite our initial rediscovery, we still had an incomplete knowledge of its distribution, the state of its populations and its ecological requirements, although we think the species might be endemic to the Pacific slopes of the Andes of Ayacucho. The presence of a nearby protected area, the National Reserve Pampas Galeras, motivated us to survey the Reserve and surrounding creeks to determine whether *T. intermedius* occurred within protected areas (1).

In 2015, our initiative received support from the Amphibian Survival Alliance's (ASA) (http://www.amphibians.org/ seedgrants/2015garcia/), and we were able to return to Puquio and Allipacca to resurvey the creeks where we had found populations of *T. intermedius*. We also had broadened our goals of assessing population status, identifying the main threats to the species, and estimating prevalence and intensity of infection by the fungus *Batrachochytrium dendrobatidis*. Chytridiomycosis is known to be especially virulent for species of *Telmatobius*. In addition to evaluating the Allipacca stream, we also assessed the streams of the Pampa Galeras National Reserve, about 30 km west of Puquio.

During these surveys starting in 2015, we sampled five streams in Allipacca, near the paved road connecting Puquio to San Andrés, following the protocol of Catenazzi *et al.* (2009). We evaluated transects of 100 m, and searched in all possible tadpole or frog microhabitats (edges of the stream, deep areas, under stones, pools, etc.), we took data of the habitat such as dimensions of pools (width, depth), channel width, current speed, temperature, pH, conductivity, presence of plants, algae, insects, in addition to skin (frog) or mouthpart (tadpole) swab samples to determine the presence and intensity of the chytrid fungus.

Our findings suggest that *T. intermedius* is a threatened species. In the different surveys we carried out from 2015 to 2018, we could find the species in only two streams. The estimated area of occupation is thus very small (+ - 10 km2 approximately). We did not find *T. intermedius* within the Pampa Galeras Reserve. However, at Galeras, we found another species of Telmatobius, which we think may be new to science.

The frogs we captured in Allipacca match the description of the species. Adults of *T. intermedius* measure ~45 mm in snout-vent length. Anatomically, this frog has a wide head dorsolaterally, keratinized skin spicules throughout the body, both on the back and belly, tympanic membrane and tympanic ring absent, and supra-tympanic fold present. It has a variable and uniform gray dorsal coloration, a lighter belly with lower areas of the belly and orange thighs. Males have nuptial spicules scattered on thumbs and chest. The tadpoles have gray coloration with a dark spot at the end of the tail. The eggs are placed in mass, adhered under the stones the current of water, wrapped in a transparent mucilaginous substance.

The two creeks inhabited by *T. intermedius* are located on the western flank of the Andean mountain range, on a moderate slope. These creeks traverse agricultural fields and rural roads where domestic animals and people circulate, is crossed by an asphalted road and is very close to the city of Puquio. The creek channels are



The local market where they sell water frogs for human consumption. Photo. Victor Vargas.

of variable, but narrow width in certain sectors (up to 1.5 meters) and have clear running water that stagnates in moderately deep pools, with a substrate that is muddy, sandy or rocky depending on reach. The riparian vegetation is composed of shrubs and herbs typical of the Peruvian highlands. We found three other species of amphibians along these creeks, the toad *Rhinella spinulosa* and the marsupial frog *Gastrotheca marsupiata*, as well as another species of *Telmatobius* sp., possibly new.

We identified environmental contamination as a threat to these frog populations. The two streams were subject to runoff from the nearby agricultural fields and contamination by solid residues, and during one of our surveys, we found discarded jars of various agrochemicals close to the creek. Furthermore, many of the pools are eutrophic with abundant aquatic plants and algae as a consequence of the use of fertilizers and detergents. Heavily eutrophic pools are unsuitable for tadpoles and adults. Car traffic and rain erosion also promote siltation in both creeks. Overharvesting of adults for local and regional consumption is another threat. In recent years, over 10 thousand water frogs have been confiscated in Peru, mostly originating from southern Peru and all destined to human consumption in cities. It is presumed that climate change will cause the loss of quality and quantity of water due to melting glaciers and changes in local and regional climates. We expect this threat to intensify in the future, considering the current climate already exposes amphibians to extended periods without precipitation during the dry season. Future extremes might include prolonged droughts and flooding from extreme and sudden precipitation events.

### Acknowledgments

Our thanks to Isabella Vargas, Vanessa Loarte, Lady Vargas, André Espinoza and William Ayala. To the Servicio Nacional Forestal y de Fauna Silvestre of Peru and the Dirección Forestal y de Fauna Silvestre of the Gobierno Regional de Ayacucho.

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The research team of the Pro Wildlife Association, Ayacucho, Peru. Photo. Oscar Chipana.

14

### Unexpected Sources of Food for Amphibians Living Near Humans

### By Mikołaj Kaczmarski, Anna Maria Kubicka & Piotr Tryjanowski

any species of amphibians are food generalists, feeding naturally on items that represent the dominant resource at a given moment. More precisely, they can adapt their diet in response to local variations in the frequency of available prey items. Therefore, they are an important link connecting trophic levels, functioning as regulators of fast-growing numbers of the invertebrates (or pests) they eat as well as of predatory vertebrates that feed on amphibians. It turns out, however, that amphibians can transcend the categories of predator or prey.

Recently, the first case of possible mutualistic interaction (or commensalism) between the Marsh Frog *Pelophylax ridibundus* and buffaloes in the Kızılırmak delta (northern Turkey) was described. In the course of a field study, it was discovered that frogs hunted flies in different parts of the bodies of buffaloes (*Fig. 1*). This phenomenon occurs only in autumn when the density of frogs is much higher than during spring (1). Similar interactions involving birds and large mammals, though never frogs, have been described and, as we know, may originate and intensify rapidly under specific local conditions.

However, Pelophylax green frogs (formerly Rana) are generally known for their high degree of plasticity. This feature greatly facilitated the conduct of research by Prof. Leszek Berger in the 1970s. Berger discovered that the edible frog Pelophylax esculenta was not a species, but an interspecies hybrid between the Pool Frog P. lessonae and the Marsh Frog. Thus Berger discovered the process of hybridogenesis, a unique reproductive system in amphibians. Berger conducted research based on crosses between green frogs for 39 years, breeding all 16 taxa of green frogs and obtaining over 800,000 offspring (2, 3). This was made possible by the professor's development of his feeding system for frogs. When Berger started his work with frogs, there were no insects to feed them with, such as live crickets, available in pet shops. Therefore, at first, he used uneaten bits of meat directly from the carnivore catwalks in the zoo in the city center of Poznań (Poland), to which his outdoor frog "farm" was adjacent. Then he invented a simple system. Flies laid eggs on the leftover meat; this was followed by the rapid development of larvae, from which adult insects emerged. Though it may seem unlikely, Berger provided food in this way to the frogs, who ate the flies directly from the pieces of meat (Fig. 2).

This story, combined with reflections on the dietary generalism of amphibians and their plasticity in terms of foraging behavior, led us to consider the question of what other resources might serve them as a source of food. Our attention was drawn by windfalls of fleshy fruit which lie on the ground in high numbers from the end of July until as late as September, *e.g.*, at precisely the same time of year that fresh amphibian metamorphs leave their spawning sites in Central Europe (*Fig. 3A*) and begin feeding and dispersing. Thus, this is a key period in their lives, one that will affect the overall size of adults and ensure greater survivability during hibernation. Taking this into account, it can be assumed that, since fermenting fruit attracts many types of invertebrates, the presence

of fruit trees close to a breeding site might influence foraging behavior and more rapid post-metamorphic growth of amphibians or their dispersion near breeding areas.

We decided to investigate, under experimental conditions, whether the presence of fallen fruit would affect the growth dynamics of fresh metamorphs of the Green Toad Bufotes viridis through ensuring a greater variety of food, such as invertebrates attracted by such fruit (Fig. 3D). Accordingly, 120 juvenile toads were randomly assigned to one of four groups: two experimental groups with fleshy plums and two other groups as controls (without fruit). Each group was kept in an enclosure to which wild invertebrates had free access for 30 days. Every other day, the body mass and length of each juvenile was measured. The fruit used in the experiment was Prunus cerasifera, a very common species in the urban, suburban, and agricultural landscapes near the study site (Fig. 3A, B). Our experiment showed that toads from both enclosures with plums were characterized by more rapid growth than individuals from the control treatments (Fig. 3C). Simultaneously, in the enclosure with fleshy fruit, greater species richness of wild invertebrates was observed (4). The most interesting finding is that this indirect effect (fruit trees - insects - amphibians) may have important implications for the functioning of the altered ecosystem - much more important than we thought at first. Researchers had already demonstrated the role of fruit trees in relation to birds in transformed areas (e.g., urban areas), but nobody had addressed this issue in the context of amphibians.

Some interested individuals argue that fruit trees constitute a problem by causing allergies, creating too much shade, or making it necessary to clean up leaves or rotting fruits. It is worth considering whether the removal of fruit trees may exert a negative impact on some amphibian populations (or wildlife in general) through limiting their available food in the increasingly difficult conditions caused by global urbanization.

### Acknowledgments

We thank the family of Prof. Leszek Berger for permission to use a photo by Z. Pniewski from the family collections.

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Fig. 1: Photograph of the interaction between frogs and buffalos, showing a buffalo in a seated position with many frogs on its fur. Photo: Nizamettin Yavuz.



Fig. 2: Rearing: frogs preying on flies on old meat brought from the zoo. Photo: Z. Pniewski.

16 | FrogLog 27 (1), Number 121 (March 2019)



Fig. 3: Graphics illustrating our experiment. Clockwise: (A) Prunus cerasifera, commonly known as the Cherry Plum or the Myrobalan Plum, often found lying on the ground in large quantities; (B) plums on the tree branch; (C) plot representing the body mass index of toads during the experiment. Red – groups with fleshy fruits (plums), blue – control groups without plums; (D) toadlets of the Green Toad Bufotes viridis prior to the beginning of the experiment. Photo: Mikołaj Kaczmarski. Plot: Anna Maria Kubicka.



### Conservation Strategies and Participatory Monitoring of Threatened Amphibians on Peace Implementation Territories in Southwestern Colombia

By Natalia Bacca-Cortes<sup>1,2</sup>, Alejandro Guerrero-Cupacán<sup>1</sup>, Juan Manuel Daza<sup>5</sup>, Belisario Cepeda<sup>4</sup>, Cristian Flórez-Paí<sup>1</sup> & Nicolás Urbina-Cardona<sup>5</sup>

mplementing biodiversity conservation strategies is one of the biggest challenges for megadiverse countries with sociopolitical conflicts. These countries still have large isolated geographical areas where it is difficult to conduct biodiversity monitoring and environmental education activities, due to, among other factors, the forced displacement of human populations.

In Colombia, one of the world's megadiverse countries, the rate of deforestation has accelerated over the last 20 years, mainly in lowland forests in the south of the country (Nariño Department). This constitutes a threat to the existence of different species of fauna that depend on forest cover (1). In Nariño, the increase of illicit crops are promoting the transformation of the territory, enhancing deforestation rates and changing the social and economic dynamics of the communities in this border region with Ecuador (2). At the end of 2016, Colombia signed a peace treaty with the FARC guerrillas and its implementation has commenced, which allowed for the reactivation of biodiversity conservation initiatives in the southern basins of the country.

In the Andean-Coast Piedmont of Nariño Department (*Fig. 1*), the binational basin of the Mira and Mataje Rivers, few biological expeditions have been conducted focusing mainly on amphibians.

<sup>1</sup>Fundación FELCA; <sup>2</sup>Estudiante Maestría en Conservación y Uso de Biodiversidad, Facultad de Estudios Ambientales y Rurales, Pontificia Universidad Javeriana; <sup>3</sup>Profesor, Instituto de Biología, Universidad de Antioquia; <sup>4</sup>Grupo de Investigación en Ecología de Páramos y Ecosistemas Andinos, Universidad de Nariño; <sup>5</sup>Regional chair for Colombia, Amphibian Specialist Group (ASG), Species Survival Commission (SSC), International Union for Conservation of Nature (IUCN). E-mail: ncbacca@ gmail.com This region shares amphibian species between Colombia and Ecuador (3, 4). This region is part of the biogeographic Chocó, which is an important biodiversity hotspot (5) and serves as a refuge for several threatened amphibian species, such as *Atelopus* aff. *elegans*, *Rhaebo colomai*, *Oophaga sylvatica*, *Epipedobates narinensis*, *Centrolene peristictum*, *Bolitoglossa medemi*, *Pristimantis verecundus* and *P. colomai*, (*Fig.* 2). Despite of being a region with high species diversity, it faces anthropogenic activities such as deforestation, expansion of the agricultural frontier and illicit crops, ecosystem fragmentation and mining (6), which synergistically may imply a greater risk for the biodiversity persistence (7).

Amphibians are one of the most vulnerable vertebrate groups in the world ( $\vartheta$ ), and Colombia is one of the most amphibian-rich countries worldwide. Unfortunately, the country also has the highest number of threatened amphibians, with nearly 170 species under some threat category ( $\vartheta$ ). The Ecological Foundation Los Colibríes de Altaquer (FELCA), with the support of the Critical



Fig. 2: Bolitoglossa medemi (left), Rhaebo colomai (right). Photo: Alejandro Guerrero-Cupacán.

18 | FrogLog 27 (1), Number 121 (March 2019



Fig. 3: First course on "Conservation and Participatory Monitoring of Threatened Amphibians of the Coastal Andean Piedmont, Río Ñambí Natural Reserve.

Ecosystem Partnership Fund (CEPF), is building the "Binational Strategy for the Conservation and Participatory Monitoring of Threatened Species (plants, amphibians, birds, and mammals) of the Key Biodiversity Areas in the Tropical Andes (EBC, by its initials in Spanish)". In this EBC strategy, our main goal is to establish strategic alliances for research, monitoring, capacity building and biodiversity conservation processes in southwestern Colombia. We attempt to contribute to the knowledge of amphibians, and work in collaboration with local organizations. In so doing, we expect to reduce the loss of forest cover, strengthen the social governance of the communities to successfully implement conservation plans along the binational border of the Mira and Mataje River basins.

As part of the EBC's environmental education and local capacity building activities, the first course on "Conservation and Participatory Monitoring of Threatened Amphibians of the Coastal Andean Piedmont" was held in the Río Ñambí Natural Reserve (RNRÑ) (Fig. 3 and 4). Both, theoretical and practical goals of the course were designed to strengthen local capacities to involve the community with initiatives for participatory monitoring of threatened amphibians. The course was led by biologists, with extensive research experience in the area of herpetology and biodiversity monitoring, from the Universidad de Antioquia, Universidad del Valle, Universidad de Nariño and Pontificia Universidad Javeriana all Colombian academic institutions. Scholarships were awarded to local organizations, nearby nature reserves staff and community representatives to take part in the course. Among the participants were staff members from reserves such as La Planada, Tirapuentes, Bangsias and El Quinde Lago Reserva; as well as key actors from the areas of influence of RNRÑ such as Altaquer, Nembí, El Barro and Tajada localities. Lastly, our EBC initiative had support



Fig. 4: First course on "Conservation and Participatory Monitoring of Threatened Amphibians of the Coastal Andean Piedmont, Universidad de Nariño.

and collaboration from allied international organizations with decision-making capacity on the Mira y Mataje binational basin such as the Fundación Altrópico and the World Wildlife Fund (WWF).

As a closing activity of the course, the "1st Festival of the Poisonous Diablito Frog" event was held and coordinated by a local primary school. The "Diablito" Frog (*Oophaga sylvatica*) is categorized as Near Threatened (NT) (10), because like many other poisonous frogs (family Dendrobatidae) from South America, it has aposematic coloration to warn predators (Fig. 5), which also makes it appealing to illegal traders. Precisely, in this region, there were dynamics of use that involved episodes of massive extraction of *Oophaga sylvatica* to supply chains of illegal wildlife trafficking. In response to these pressures, the local community strengthened networks to create this festival, an initiative that is being led by the village community of San Francisco (Junín District, Nariño Department) and students from the San Francisco Educational Institution.

The "1st Festival of the Poisonous Diablito Frog" event is a good example of how a united and organized community seeks to generate income by carrying out sustainable activities, and seeking resources to preserve threatened frogs and its habitat. Consequently, this "Festival of the Diablito Frog" was an activity that brought together more than 140 people from the surrounding communities such as Junín, Barbacoas, Ricaurte, Altaquer and El Palmar. They participated in the socializations, field trips and painting contests with the children, who, as an example of direct interaction with the species, drew and artistically expressed their observations of the diablito frog (*Fig. 6, 7 and 8*).

The "1st Festival of the Poisonous Diablito Frog" event is articulated to the conservation processes of southwestern Nariño region, in which diverse actors are working for the conservation of amphibians. The San Francisco School students and their teacher formed the "Grupo Ecológico para la Conservación de la Rana Diablito (GERD)" (Ecological Group for the Conservation of the "Diablito" Frog), and with the collaboration of the FELCA Foundation and the Universidad de Nariño (Biology undergraduate program professors and "Andean and Paramo Ecosystems Conservation Research Group" researchers), are currently developing projects that seek the conservation of the species *Oophaga sylvatica* ("diablito" frog). All the activities are supported by the "Alianza Sur Sostenible" formed by the Small Seed Grants Program of the Global Environment Facility (GEF), the Government of Nariño and SENA Emprende Rural.



Fig. 5: Oophaga sylvatica (The Diablito Frog). Photo: Natalia Bacca-Cortes.

Thus, the EBC initiative has made possible to initiate environmental education processes and support conservation initiatives in territories that historically has suffered by socio-political conflicts. We highlight the articulated work of the community, the local organizations, the nearby natural reserves and all the people who joined to support this initiative, as they have brought to consideration new goals and social challenges, by projecting a change of perspective for this region. In addition, it constitutes a basis for the strengthening of both the sense of community belonging, and the exchange and appropriation of knowledge.

The winds of post-armed conflict environment in Colombia, offers opportunities in these territories to develop participatory conservation processes and seek alternatives for local development making sustainable use of biodiversity. At the same time, it is important to articulate research projects and inventories in these vulnerable regions to the new interactive platforms, as they are key tools for monitoring species in strategic sites. We hope to continue supporting these efforts to strengthen and consolidate the EBC strategy through community involvement and academic research.

### Acknowledgements:

We would like to thank all the organizations that have facilitated the management of resources to make these processes become a reality: Monica Orjuela (CEFP), Ana Beatriz Barona and Ana Cristina Enriquez (PPD), Yina Pantoja (UNDP) and Luis Fernando Gómez (WWF). To the professors of the Universidad de Nariño, Guillermo Castillo, Belisario Cepeda and Maria Elena Solarte, for the support from the academy. To Juan Manuel Daza (Universidad de Antioquia), Wilmar Bolívar (Universidad del Valle) and especially to Pontificia Universidad Javeriana for trusting in the process and sharing their knowledge with us. To the school teacher Libia Fajardo and her students at the San Francisco School, for being the main actors and for their commitment to conservation. To the FELCA Foundation team for their hard work in the development of these initiatives. To the staff members and personnel of the Rio Ñambí Natural Reserve, for providing their facilities for the development of the course.

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Fig. 6: Socializations during First Festival of the Diablito Frog.





Fig. 7: Painting contests with the children, First Festival of the Diablito Frog.



Fig. 8: Field trips, First Festival of the Diablito Frog.

### Calling the Rains With the Help of the Lake Titicaca Frog

By Roberto Elías<sup>1,2</sup>, Enrique Ramos<sup>1</sup>, James García<sup>1</sup>, Matt Herbert<sup>1</sup> & Jhazel Quispe<sup>3</sup>

n 2013, the Lake Titicaca or "K'ayra" Frog (Telmatobius culeus) was declared a species of regional interest by the Regional Government of Puno given its ecological and sociocultural importance for the region. It is currently considered to be Critically Endangered at both the global and national levels due to pollution problems in Lake Titicaca and the unregulated collection and trade for different purposes. One of the reasons for collection is for the preparation of "medicinal extracts" which are sold in popular markets in the cities of Lima, Arequipa, and Cusco. The frogs have also been observed in soups on the



Uros islands where food is sold. However, these are not the only reasons why the frog is collected.

Although there is not much literature in this regard, the frog or K'ayra is considered by some local populations as a connection between them and the gods. The locals use the K'ayra in sacred ceremonies aimed at bringing the rains that assure them a good harvest. One of the places that still maintains this tradition in the region is the populated center of Perka-Norte, located on the shores of Lake Titicaca (3,815 m.a.s.l.) in the district of Platería.

Platería is one of 15 districts of the province of Puno and located in the region of the same name. The main economic activities of the district are agriculture, livestock farming, and fishing. More recently, efforts have been made to develop experiential eco-tourism, highlighting biodiversity, traditional customs and practices.

Amongst the ancestral practices that have endured over time is the "rain ritual" that local inhabitants have inherited from their ancestors and have mixed with some modern objects and practices. We describe herein a "calling of the rain" ceremony observed in September 2017.

The ritual begins in the early morning with the meeting of some locals with the "Paco" or shaman responsible for carrying out the ceremony. With the help of other men from the area, all enter the waters of Lake Titicaca in search of K'ayra, which are captured and placed into a clay pot that is an important part of the activity. The entire ritual is accompanied by a musical band that plays local music in front of a group of wives, who remain seated while watching the men perform their work.

After collecting the K'ayra, all participants are directed to the top of a hill or "Apu" which has been previously chosen because it is sacred to the community. A campfire is lit and maintained with dung, and the Paco prepares to carry out the ceremony. The cer-

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emony begins when the Paco and his assistant finish accommodating all their materials and kneel on a blanket woven by them, called "Lliklla." After making several prayers and showing offerings, they ask all participants to make a prayer or personal request by raising five coca leaves and then placing one on top of the other on the "Lliklla." Each participant delivers the bundles of coca leaves by each participant to the Paco. After the ritual, all offerings are thrown into the campfire while the Paco and his assistant continue with their prayers and keep the clay pot nearby. The entire ceremony lasts about nine hours, beginning at 8 am and finishing at about 5 pm. At close to 11 am participants are already at the Apu and will stay there until the end of the ceremony.

After making the offerings, the pot is kept on the top of the hill for eight days so that the frogs can call the rains. After the eight days have passed the K'ayra are returned to the lake and released.

No K'ayra mortalities were observed during this particular ceremony.





### One or Multiple Origins of Midwife Toads in the UK?

By Steven J. R. Allain, Rob Gandola & John W. Wilkinson

s some of you may be aware, we've been working to closely monitor the population of Midwife Toads (*Alytes obstetricans*) that have been introduced to Cambridge, UK. So far this has been in the form of disease screening and trying to establish a rough population estimate (1). Now we aim to move into the realm of molecular data to find out where the different midwife toad populations in the UK have originated from. It has long been assumed that all British midwife toad populations have a French origin (2), but preliminary analysis suggests otherwise; that the Cambridge population originated from a source population in northern Spain. We will be employing a similar analysis utilized in previous studies to investigate the origins of Smooth Newts (*Lissotriton vulgaris*) in Australia and the Asian Common Toad (*Duttaphrynus melanostictus*) in Madagascar (3,4).

Therefore this project is very similar to the projects mentioned above, and we are just working with a very different species. However, the acquisition of our samples is entirely in the hands of volunteers, although we may have to make a few visits towards the end of the project to ensure collection from some of the more sensitive sites. Another factor that makes this project unique in terms of citizen science is that the toads mainly live in urban areas, particularly in people's gardens. Thankfully in Cambridge, we have been granted permission from the homeowners to access their gardens and, as a bonus, we've managed to get them involved too! We've expanded on this model of local involvement and taken it one step further for our national project, with interested citizen scientists being sent swabbing kits in the post with everything they need to collect samples themselves. Interestingly, some of the toads have gone full circle and have ended up back in captivity; these individuals have also been swabbed to illuminate the origins of the populations they were sourced from.

Although it may sound peculiar, even though this is a common strategy to obtain DNA, taking buccal swabs from these non-native toads will essentially allow us to conduct a phylogeographic test. This information is beneficial to us as not only can we find out where all of the different populations originated, but we can also work out how many different introductions there have been. The toads were once a favorite pet due to their unusual breeding behavior as well as a common laboratory animal so it's likely there would have been escapees or intentional releases. To help fund the project we have set up an online crowdfunding campaign which can be found here. We'd appreciate it if you could share the link with any potentially interested parties as well as taking a look at yourself. This project is possible thanks to an active collaboration between The Herpetological Society of Ireland, the Amphibian and Reptile Conservation Trust and Amphibian and Reptile Groups UK. We aim to add regular project updates on the GoFundMe fundraiser page highlighting how and where everyone's donations are helping us to answer this herpetological enigma.

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### 9th CBH: Brazilian Congress of Herpetology Campinas, Sao Paulo

### By Janaína de Andrade Serrano<sup>1</sup> & Luís Felipe Toledo<sup>1</sup>

e are proud to announce that the 9<sup>th</sup> edition of the Brazilian Herpetology Congress will be held in Campinas, São Paulo, from the 22<sup>nd</sup> to 26<sup>th</sup> July 2019. This year's theme is Inclusive Herpetology. We want to invite you to join us to celebrate such great biological, and cultural diversity only found in Brazil! Amongst the speakers, we will have great names, as Célio Haddad, Paula Eterovick, Ana Prudente, Christine Strüssmann, Marcio Martins, Karen Warkentin, David Blackburn, Mike Ryan, and Dan Rabosky.

There will be a series of workshops, short courses, roundtables and symposia in trending topics, as well as photography and art contests. For the first time, we'll also exhibit the Science Slam presentations in local pubs! If you are interested in Herpetology, don't miss this opportunity! Don't miss the discounts on registration fees and keep posted on our Website, Facebook, Twitter, and Instagram! See you soon! Bem vindos!





24 | FrogLog 27 (1), Number 121 (March 2019)

### 'Save All Frogs' Initiative Launched

By Matt Ellerbeck

he most diverse form of herpetofauna in the province of Ontario is frogs, boosting more species than turtles, snakes, or salamanders. However, there is no outreach education effort solely devoted to these amphibians within the province.

This is unfortunate as many frogs species are threatened with extinction. Furthermore, the endangerment of frogs is not exclusive to regions outside of Ontario. Several of the province's species are in serious decline. The Great Lake/St. Lawrence population (east and north of Toronto) of the Western Chorus Frog (*Pseudacris triseriata*) is listed as Threatened under the federal Species at Risk Act. The Fowler's Toad (*Anaxyrus fowleri*) is even more at risk, being listed as Endangered. Worse still, the diminutive Northern Cricket Frog (*Acris crepitans*) is considered extinct from Ontario.

More of Ontario's frogs could also be disappearing, as many species have not yet been properly assessed. This is what inspired me to launch my *Save All Frogs* project. With this effort, I will be educating individuals throughout the province on why frogs are disappearing, what roles they play in the environment, and most importantly how they can help. I will be emphasizing as I visit schools, camps, conservation areas and other venues that individuals can become involved with the recovery of frogs via behavioral changes, informed decision making, environmental stewardship actions, and habitat management efforts.

Education has been noted as an effective conservation tool by numerous groups and organizations. The Amphibian and Reptile Conservancy (ARC) states that it recognizes the need to increase awareness, appreciation, and understanding of amphibians, reptiles and their habitats, which can then enhance conservation actions and stewardship practices. The Amphibian and Reptile Conservation Trust also proclaims that education is one of the most important tools in the long-term conservation of amphibians and reptiles. By raising awareness, enhancing knowledge and encouraging people to take action, real steps can be made towards conserving amphibian and reptile species.

This is why I am committed to educating the public on the plight of frogs!

This issue is near and dear to my heart, as I have always loved amphibians. When I was a small child my grandfather and his friends would use frogs as bait when they went fishing. I was extremely upset by this and articulated this with great passion. So much so that many of them stopped using frogs, including my grandfather. www.saveallfrogs.com



Photo: Matt Ellerbeck.

## Best Practices of Amphibian Conservation in the Roman Countryside

By Antonio Pizzuti Piccoli<sup>1</sup>

he Roman landscape is represented by a mosaic of contiguous environments. Excluding urban environments, we find the agricultural ecosystem as the main element, which, in some places, still maintains features of high naturalness and biodiversity, intermingled with residual areas of natural environments such as mixed forest, Mediterranean scrub and wetlands.

These elements combine to form a unique landscape that is in serious decline and needs to be preserved.

Due to the recent environmental degradation, the survival of amphibians is extremely compromised. Below we present some good practices which we are employing to contribute to the conservation of amphibians.

The experiences are proposed in the areas of the "Castel di Guido" Farm and in the "Bosco di Palo" Natural Park. Both areas are included in the European Network "Nature 2000" as Sites of Community Importance and are established as National Herpetological Relevance Areas - ARER by the *Societas Herpetologica Italica*.

### THE RESTORATION OF ANCIENT FOUNTAINS

In the Roman landscape, we find many fountains, structures built for providing water to the numerous animals that were bred outdoors or in the wild.

The existing fountains are those built from 1600 to today and are largely abandoned or unused. This abandonment is due to the significant social changes and processes that have characterized the Roman countryside in the last 50 years. Amphibians are in strong decline today on the regional territory; in the agricultural ecosystem the amphibians have adapted over time and survive, above all, thanks to the presence of artificial water supply (tanks for irrigation, fountains, artificial pools) (1,2).

The study area for this experience is the public farm of "Castel di Guido," between the 16th and the 20th km of the state road "Aurelia." The farm extends for 1,966 hectares (including the 250 hectares of the Natural Park managed by LIPU – ONG, the Italian League for the Protection of Birds) and is characterized by hilly areas degrading towards the coastal plain. The maximum altitude reached is 80 meters, while the minimum altitude is about 10 meters. Since 1978, the farm has been owned and managed by the Municipality of Rome. In the farm you will find cultivated cereals and bovine fodders, as well as bred sheep and cows, both in the stable (Italian Friesian cow) and in the wild ("Maremmana" cow) (3).

Climatically, the area is part of the Mediterranean Transitional Region, characterized by hot and dry summers and mild winters. The climate is particularly mild for the proximity of the sea. The study area is characterized by an evident vegetational complexity and by a great floristic richness, present in the different habitats (4).

The landscape, with a strong vocation for pastoral activities, is dotted with numerous fountains, used to provide water to the domestic animals. These water collections, although of artificial origin, often constitute real habitats, with the presence of plants and algae that favor the growth of a remarkable community of aquatic

<sup>1</sup>"Bosco di Palo" Natural Park - Via Monteroni 1265, I – 00055 Ladispoli (Rome) Italy; Email: info@fattoriapertutti.it



Fig.1: Common Toad *Bufo bufo* egg-laying in one of the fountain in the "Castel di Guido" farm. Photo: Antonio Pizzuti Piccoli.

animals.

The numerous springs, ditches and artificial water collections (including fountains), even temporary ones, make the territory of "Castel di Guido" a precious site for the conservation of amphibian communities.

The various scientific experiements that have been carried out in the farm, have led to important data being collected on the phenology and on the structure of the local populations of amphibians (5).

The amphibians of the area are represented by five species: the common toad *Bufo bufo* (L., 1758), the European Green Toad *Bufotes balearicus* (Boettger, 1880), the Italian Tree Frog *Hyla intermedia* Boulenger, 1882, the Green Frog *Pelophylax bergeri* (Gunther, 1986) / *Pelophylax kl. hispancus* (Bonaparte, 1839) and the Smooth Newt *Lissotriton meridionalis* (Boulenger, 1882).

Currently, in the agricultural ecosystem, one of the few sources



Fig.2: The "Fountain of the viper" whose restoration is planned. Photo: Antonio Pizzuti Piccoli.

**26** | FrogLog 27 (1), Number 121 (March 2019)



Fig.3: Access ramps (wooden ramps, dry stone walls, embankments) are planned on the edge of the fountains. In the photo a grassy embankment realized in the fountain of "Animal farm" Environmental Education Center near Rome. Photo: Antonio Pizzuti Piccoli.

of water is made up of artificial fountains and these represent often the only site for amphibian egg laying (*Fig. 1*).

In this study, we proceeded to identify and map the existing fountains, have described their state of conservation, designed their recovery where they are no longer functional (water restoring, restoration and maintenance of the wall part), always maintaining the original architectural aspects (*Fig.* 2).

The project aims, after a first phase of structural restoration, to identify functional improvements to the use by amphibians, in order to benefit colonization and presence. After the first phase of reactivation of the fountains, specific measures were planned to favor the access and exit of amphibians during the reproductive time (6, 7).

In this regard, ramps for exit will be arranged (*Fig.* 3) and uncultivated grass will be preserved at least on one side of the structure, in continuity with the surrounding shrubs and arboreal plant formations, to facilitate the movement of individuals (*Fig.* 4).

Furthermore, guidelines have been elaborated for the management of the fountains to guarantee the survival of amphibians and, contemporarily, the use for sheep and cows breeding. these guideline, reported below, will contribute to minimize the habitat disturbance, primarily during the reproductive period of amphibians.

- Do not empty the fountains, to allow the completion of the reproductive cycles of amphibian species, keeping the water levels relatively constant.
- Monitoring of the chemical-physical and biological quality of water, in order to periodically check the possible concentration of biocides or anoxia situations, that could cause considerable damage to the reproductive cycles of these animals.
- Avoid "cleaning" the fountains during the period from January to August. This practice consist in the removal of vegetable mass from fountains to keep water clean for domestic animals; it should be avoided or be less invasive, a low impact cleaning can be performed in periods away from the breeding season.
- Maintaining a natural growth strip of vegetation near the fountains, avoiding drastic mowing methods.
- Protection of wooded areas adjacent to wetlands, allowing

them to mature, aiming to increase the forming of microhabitats and shelters suitable for terrestrial phase of amphibians.

• Controlling the expansion of aliens species, with particular reference to fishes and Crustaceans, lethal presences for eggs and larvae of amphibians (also by removing specimens where present).

In general, these conservation measures of artificial breeding sites will ensure a coexistence between amphibians and human activities on the territory.

### TEACHING AMPHIBIANS TO CHILDREN AS AN INVESTMENT FOR THEIR FUTURE CONSERVATION

People who live in the Roman territory, often completely ignore the presence of animal species, and in particular amphibians. To bridge this gap, in the last ten years, in the "Bosco di Palo" Natural Park, laboratories have been created to illustrate the autochthonous amphibians of the territory.

The "Bosco di Palo" Natural Park is a protected area located in the coast near Ladispoli (41 ° 56 'N, 12 ° 05'E) in the north of *Latium* (Central Italy). We have mesomediterranean climate, with soft winter and a summer period of dry. Environments present are the Mediterranean bush, wood and prairie. The wood, with its characteristic presence of temporary ponds, consists of a mixed forest of deciduous oaks of about 60 hectares, with a predominance of *Quercus ilex* L., *Quercus cerris* L., *Quercus pubescens* Willd. and *Ulmus minor* Miller (8).

In the Park, many scientific studies have been carried out from 1995; they have led to collection of important data on the phenology and on the structure of the local populations of amphibians (9, 10).

In this area, four amphibians species were recorded: the Common Toad *Bufo bufo* (L., 1758), the Italian Tree Frog *Hyla intermedia* Boulenger, 1882, the Green Frog *Pelophylax bergeri* (Gunther, 1986) / *Pelophylax kl. hispanicus* (Bonaparte, 1839) and the Smooth Newt *Lissotriton meridionalis* (Boulenger, 1882).

In the last ten years, park management plan has transformed the park into an environmental education center dedicated to amphibians. In 2018 over 3,000 children (primarily students) visited the Park to participate in environmental education activities diesnged to improve the knowledge of the amphibians. Specialized person-



Fig. 4: In the fountains near natural vegetation areas it is planned to create environmental continuity favoring the presence and growth of shrubs until the board of fountains. In the picture the "fountain of the bulls" included in the project. Photo: Antonio Pizzuti Piccoli.



Fig. 5: Artificial pond to observe the larval stages of the amphibians realized at the entrance of the park. Photo: Antonio Pizzuti Piccoli.

nel explain to young visitors the importance of a well-preserved habitat in which all the animal and plant species are present. To facilitate educational activities, artificial ponds have been created to observe the main species present during their aquatic phase (Fig.



5). In recent times, "city science" activities have been developed, involving the people (students of the neighboring schools) in collecting data for the numerous scientific researches in process in the park. Furthermore, for increasing people awareness of the world of the amphibians, "warning panels" have been installed at the focal points of road crossing during amphibians migration (*Fig. 6*).

The scope of the educational action is contributing to the knowledge of amphibians characteristics bring local people to understand the importance of their respect and conservation.

In conclusion, the models proposed in the farm of "Castel di Guido" and in the "Bosco di Palo" Natural Park, in the Roman countryside, can be considered good practices of managing of the territory, and aims at the conservation of animal biodiversity and particularly of populations of amphibians.

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Fig. 6: A road sign warning of toads crossing on the secondary country roads. Photo: Antonio Pizzuti Piccoli.

**28** | FrogLog 27 (1), Number 121 (March 2019)

# Comments on the Collection Of Buccal Swabs From Amphibians

By Steven J. R. Allain

inimally invasive DNA collection techniques are one of the marvels of the modern scientific age due to increased sensitivity and precision of the associated sequencing equipment. This is of course due to advances in technology and our growing understanding of the specific areas of science that deal with genetics and DNA. No longer do we need to collect tissue samples using questionable methods (which may make your institution's ethics board cringe). Recently I've been involved with a project with my old friend the Common Midwife Toad (Alytes obstetricans) here in the UK, in terms of collecting buccal samples in order to answer a very important question: where did they come from? As a non-native species we are interested in how many different introductions have been made and whether or not the toads are all the same species. We've already been swabbing them for disease as you may have read in previous issues and so far the news is good, the population I'm intimately investigating is free from chytrid!

Now back to the whole reason for this short comment, so far the genes within the mitochondrial genome have been sequenced from this population and compared with those in reference libraries such as GenBank; this has allowed myself and the rest of the team to slowly piece this puzzle together. It is going to take us another year or so to collect samples from the other known populations and build the complete picture. In the meantime I thought that I would just highlight some hurdles we've had along the way to help others completing similar studies in the future.

Buccal swabs have been an effective tool for collecting DNA from amphibians for quite some time (1), as I'm sure anyone who has seen CSI can agree. The previous widely used was that of toe-clipping but this wasn't always the most ethically responsible option and often had unwanted side effects (2, 3) despite it's sup-

posed minimal impact. Of course tissue samples are still a valuable source of DNA but I would urge to only collect these if necessary and preferably from deceased individuals. Thankfully technology has caught up and buccal swabs are the preferred technique which have been shown to be perfect for reliable microsatellite sequencing (4). This can in turn help with species delimitation, look at inbreeding within a population or as I am looking at the phylogeography of an introduced species from many multiple unknown sources.

Now comes the tricky part as you can't exactly ask the frog, toad or salamander in question to open its mouth and voluntarily give you a sample like in the TV crime dramas. Instead you have to for ibly open the little guy's mouth to collect the mucosal cells needed to complete the DNA analysis. Now this can be done safely in a number of ways but the easiest ways that I've found to do so are to either use a guitar pick, a disposable plastic spoon or similar instrument (5). Both of these are of course quite blunt and shouldn't cause any harm to the fragile oral cavity of the amphibian in your hand - their use is needed just to convince the subject to open its mouth so you can quickly get a sample and then release them at the point of capture. When swabbing it is vital that the 'cheeks' of the amphibian are swabbed as is the roof of the mouth. Do take care when working in this latter area as you may end up poking the poor amphibian in the eye from the inside of its oral cavity. Be sure to twirl the swab between your index finger and thumb between 10-12 times in each area to ensure enough cells are collected to provide a viable sample.

It is important to note at this point that if you're going to complete your own similar study that sterile gloves will need to be worn at all times. The swabs used by myself and others with such projects are Medial Wire's MW-100 dry swabs, which handily come packaged sterile and can be stored in a fridge or freezer until the

> time when the analysis is going to be completed. For more guidance please read the literature cited but I'm confident that I've covered most of the main problem areas that our volunteers have been having.

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### Amphibian Conservation Research Symposium 2019 27-28th April 2019

Manchester Metropolitan Business School, Manchester, UK

### **Keynote speakers:**

Helen Meredith (Amphibian Survival Alliance) Brad Wilson (Atlanta Botanical Gardens) Luis Coloma (Centro Jambatu) Jamie Voyles (University of Nevada)

Early bird registration closes 28th February 2019 Normal registration closes 29th March 2019

Abstract deadline for a talk or poster 29th March 2019

For more information and to register, visit: www.manchesteramphibianresearch.com

The Future Leaders of Amphibian Conservation award is returning to ACRS 2019. Visit the above website for more information.

### **RECENT PUBLICATIONS**



Introduced Alpine Newt (*Ichthyosaura a. alpestris*) from Larzac (France). Photo: Mathieu Denoël.

Provenance of *Ichthyosaura alpestris* (Caudata: Salamandridae) introductions to France and New Zealand assessed by mitochondrial DNA analysis

### Jan W. Arntzen, Tania M. King, Mathieu Denoël, Iñigo Martínez-Solano & Graham P. Wallis

he last century has seen an unparalleled movement of species around the planet as a direct result of human activity, which has been a major contributor to the biodiversity crisis. Amphibians represent a particularly vulnerable group, exacerbated by the devastating effects of chytrid fungi. We report the malicious translocation and establishment of the Alpine Newt (Ichthyosaura alpestris) to its virtual antipode in North Island of New Zealand. We use network analysis of mitochondrial DNA haplotypes to identify the original source population as I. a. apuana from Tuscany, Italy. Additionally, a population in southern France, presumed to be introduced, is identified as I. a. alpestris from western Europe. However, the presence of two differentiated haplotypes suggests a mixed origin. This type of analysis is made possible by the recent availability of a phylogenetic analysis of the species throughout its natural range. We discuss the particulars of both introductions.

J. W. Arntzen, T. M. King, M. Denoël, I. Martínez-Solano, G. P. Wallis, Herpetol. J., 26, 1 (2016): 49–56. http://hdl.handle. net/2268/181073

Drought-mediated extinction of an arid-land amphibian: insights from a spatially explicit dynamic occupancy model

### Erin R. Zylstra, Don E. Swann, Blake R. Hossack, Erin Muths & Robert J. Steidl.

E ffective conservation of species with patchy distributions requires an understanding of how natural and anthropogenic processes, like climate change, affect metapopulation dynamics. Few applications of metapopulation theory have explored temporal variation in extinction and colonization rates, despite evidence that demographic rates and dispersal patterns vary in response to a wide array of biotic and abiotic processes. We extended a spatially explicit dynamic occupancy model to better explore spatiotemporal variation in colonization and extinction rates. Our model requires only binary presence-absence data, accounts for imperfect detection, and provides a flexible framework for modeling dispersal processes. We used this framework to assess metapopulation dynamics of Lowland Leopard Frogs (Lithobates yavapaiensis), a species that inhabits arid mountain canyons in the Sky Island region of the southwestern U.S., based on 22 years of survey data. We found that both colonization and extinction rates varied over time with hydrologic conditions. Local extinctions were associated with drought conditions, especially at sites that lacked deep pools with groundwater inputs. Unoccupied sites had a higher probability of being recolonized when sites in close proximity were occupied in the previous season and when larval or dispersal periods were wetter than normal. One of the two metapopulations we studied was extirpated during a period of severe drought. Frogs persisted in the other metapopulation, likely because surface water was more reliable at many of the sites they inhabited and because sediment levels, associated with erosion events following high-elevation wildfire, were low. Application of our model provided valuable information about how climate and wildfire-related processes affected the distributional dynamics of a threatened arid-land amphibian. More broadly, this analytical framework could be used to explore factors governing metapopulation dynamics of a wide array of taxa that inhabit patchy environments and consequently inform conservation and management efforts for imperiled species.

E. R. Zylstra, D. E. Swann, B. R. Hossack, E. Muths, R. J. Steidl, *Ecological Applications* (In press). DOI: 10.1002/eap.1859

Niche shift and resource supplementation facilitate an amphibian range expansion

### Sarah J. Davies, Matthew P. Hill, Melodie A. McGeoch & Susana Clusella-Trullas

A im: To determine whether recent range expansion of small-bodied arboreal frogs, *Hyperolius marmoratus* Rapp, is accompanied by changes in species– environment relationships and whether its historical range was constrained by climate, availability of water bodies or topographic variables. We test if artificial water bodies in the novel range have facilitated niche shift by increasing available habitats for frog establishment. Location: Western Cape Province, South Africa, with reference to the broader species range in southeastern Africa.

Methods: We build species distribution models using occurrence data from the historical and novel ranges and reciprocally project them to highlight areas of putative niche change. We test for niche shift through ordination-based approaches to disentangle how species–environment relationships may have altered and whether climate or landscape features (artificial water bodies and topography) are more strongly associated with the identified change. We further decompose niche change into areas of expansion and unfilling to quantify niche shift and describe potential future spread.

Results: We observed niche expansion into novel environmental space, with 21% of niche space in the invaded range composed of environments that were not occupied in the native range. We also observed 16% niche unfilling, signifying range disequilibrium and potential for further spread. Mean annual precipitation and proximity to water bodies were more influential in models constructed in the novel range than in historical or combined range models, suggesting that presence of artificial water bodies in the landscape ameliorates novel range conditions. Together, these metrics suggest that range expansion may be ongoing based on climate and water body availability.

Main conclusions: Our analyses identify a realised niche shift that has allowed painted reed frogs to occupy drier and more thermally variable habitats in their novel (invaded) range. This shift may be mediated by artificial water bodies that provide additional buffered habitats, a key resource supplement for these small-bodied tropical frogs.

C. Davies, S. J., Hill, M. P., McGeoch, M. A. and S. Clusella-Trullas, *Diversity and Distributions* 25, 154–165. https://doi. org/10.1111/ddi.12841

Factors influencing tadpole deposition site choice in a frog with male parental care: An experimental field study

### Johana Goyes Vallejos, T. Ulmar Grafe & Kentwood D. Wells

Parents have evolved a variety of strategies to minimize risks to their offspring, choosing rearing sites based on different abiotic and biotic factors, which affect offspring survival. Because availability and quality of these sites are variable, parents may have to choose between lowquality rearing sites or extended search time. In frog species with larval transport, parents are known to select bodies of water that are free of predators, cannibals, intra and/or interspecific competitors. We

experimentally tested if abiotic factors and the presence of predators and conspecifics affect tadpole deposition behavior in a population of the Smooth Guardian Frog of Borneo Limnonectes palavanensis. In this species, females lay their eggs on land and after fertilization, males guard them until they hatch; tadpoles are then transported on the male's back to small pools of water on the forest floor. We estimated the abundance of natural tadpole rearing sites and conducted experiments in the field using artificial pools to test if abiotic characteristics of these pools affect the probability of larval deposition. We also experimentally tested whether males of L. palavanensis avoid pools with conspecific tadpoles or predators. The abundance of natural deposition sites was low, and males readily used artificial pools for tadpole deposition. Males were less likely to deposit tadpoles in areas were pool permanency was compromised. Males did not avoid depositing tadpoles in pools with conspecifics or with predators. Interestingly, males exhibited clutch-partitioning behavior, dividing tadpoles between adjacent pools. Pool availability, rather than the presence of potential competitors or predators may be the main factor affecting parental decisions in this species.

J. Goyes Vallejos, T. U. Grafe, & K. D Wells, *Ethology*, **125**, 29–39 (2019). http://doi. org/10.1111/eth.12820



Logistic growth models for each *Bd* genotype by temperature. *Bd* life history traits (carrying capacity, time to fastest growth and exponential growth rate) were most dissimilar among genotypes at lower temperatures and converged as temperature increased.

Diverse genotypes of the amphibian killing fungus produce distinct phenotypes through plastic responses to temperature

#### Carly R. Muletz-Wolz, Samuel E. Barnett, Graziella V. DiRenzo, Kelly R. Zamudio, L. Felipe Toledo, Timothy Y. James & Karen R. Lips

Phenotypes are the target of selection and affect the ability of organisms to persist in variable environments. Phenotypes can be influenced directly by genes and/or by phenotypic plasticity. The amphibian-killing fungus *Batrachochytrium dendrobatidis* (*Bd*) has a global distribution, unusually broad host range, and high genetic diversity. Phenotypic plasticity may be an important process that allows this pathogen to infect hundreds of species in diverse environments. We quantified phenotypic variation of nine Bd genotypes from two Bd lineages (Global Pandemic Lineage [GPL] and Brazil) and a hybrid (GPL-Brazil) grown at three temperatures (12, 18 and 24°C). We measured five functional traits including two morphological traits (zoospore and zoospo- rangium sizes) and three life history traits (carrying capacity, time to fastest growth and exponential growth rate) in a phylogenetic framework. Temperature caused highly plastic responses within each genotype, with all Bd genotypes showing phenotypic plasticity in at least three traits. Among genotypes, Bd generally showed the same direction of plastic response to temperature: larger zoosporangia, higher carrying capacity, longer time to fastest growth and slower exponential growth at lower temperatures. The exception was zoospore size, which was highly variable. Our findings indicate that Bd genotypes have evolved novel phenotypes through plastic responses to temperature over very short timescales. High phenotypic variability likely extends to other traits and may facilitate the large host range and rapid spread of *Bd*.

C. R. Muletz-Wolz, S. E. Barnett, G. V. DiRenzo, K. R. Zamudio, L. F. Toledo, et al., *J. Evol. Bio.* Early view (2019). © 2019 European Society For Evolutionary Biology. Journal of Evolutionary Biology © 2019 European Society For Evolutionary Biology https://doi.org/10.1111/jeb.13413

A global meta-analysis of the ecological impacts of alien species on native amphibians

#### Ana L Nunes, Jennifer M Fill, Sarah J Davies, Marike Louw, Alexander D Rebelo, Corey J Thorp, Giovanni Vimercati & John Measey

he exponential increase in species introductions during the Anthropocene has brought about a major loss of biodiversity. Amphibians have suffered large declines, with more than 16% considered to be threatened by invasive species. We conducted a global meta-analysis of the impacts of alien species on native amphibians to determine which aspects of amphibian ecology are most affected by plant, invertebrate, fish, amphibian, reptile or mammal introductions. Measures of fitness were most strongly affected; amphibian performance was consistently lower in the presence of alien species. While exposure to alien species caused a significant decrease in amphibian behavioural activity when compared to a no species control, this response was stronger towards a control of native impacting species. This indicates a high degree of prey naïveté towards alien species and highlights the importance of using different types of controls in empirical studies. Alien invertebrates had the greatest overall impact on amphibians. This study sets a new agenda for research on biological invasions, highlighting the lack of studies investigating impacts of alien species on amphibian terrestrial life-history stages. It also emphasises the strong ecological impacts that alien species have on amphibian fitness and suggests that future introductions or global spread of alien invertebrates could strongly exacerbate current amphibian declines.

A. Nunes, J. Fill, S. Davies, M. Louw, A. Rebelo, C. Thorp, G. Vimercati, G. J. Measey *Proceedings of the Royal Society*, **228**, 20182528 (2019). https://doi.org/10.1098/ rspb.2018.2528



A Bombina pachypus (Apennine Yellow-bellied Toad) adult in unkenreflex. Photo: Lorenzo Talarico.

Genetic drift shaped MHC IIB diversity of an endangered anuran species within the Italian glacial refugium

the Italian glacial relugium

Lorenzo Talarico, Wieslaw Babik, Silvio Marta & Marco Mattoccia

ighly polymorphic genes of the major histocompatibility complex (MHC) encode proteins involved in the immune response that protect vertebrates from parasites and pathogen infections. MHC variation is shaped by a complex and poorly understood interplay of selective and demographic forces. Studies of MHC variation provide biologically meaningful insights useful for conservation of threatened taxa such as amphibians, which are globally declining partly due to the impact of emerging infectious diseases (e.g. chytridiomycosis). Here we characterized MHC class IIB variation in the Apennine Yellow-bellied Toad (Bombina pachypus), an endangered anuran, across its whole distribution range within the Italian peninsula. We compared MHC IIB diversity between refugial (REF) and post-glacial expansion (PGE) areas and tested for the correlation between MHC and neutral variation to quantify the role of drift in shaping MHC diversity. Overall MHC polymorphism was limited compared to other amphibian species. Despite the clear evidence of historical positive selection acting on antigen-binding sites, we found a significant correlation between the diversity of MHC and that of putatively neutral microsatellite markers, which suggests that genetic drift has contributed extensively to shaping MHC variation. MHC diversity was higher in the REF populations and decreased northwards into the PGE area, in accordance with the "southern richness and northern purity" pattern. Past demographic events likely determined the reduction of MHC variation in the PGE area, in particular the northern populations appeared to survive for a long time with a depleted MHC variation. We discussed recent population declines of *B. pachypus* in the light of our findings.

L. Talarico, W. Babik, S. Marta, M. Mattoccia, J. Zool. **307**, 61–70 (2019)



An alpine lake historically inhabited by Alpine Newts (*lchthyosaura alpestris*) but since fish introductions, newts subsist only in the nearby pools (La Pianca, Switzerland). Photo: Mathieu Denoël.

Newt life after fish introduction: extirpation of paedomorphosis in a mountain fish lake and newt use of satellite pools

#### Mathieu Denoël, Patrick Scimè & Nicola Zambelli

 ${f F}$  ish introduction is one of the main causes of amphibian decline worldwide. It affects particularly rare aquatic phenotypes such as paedomorphs, which retain gills during the adult stage. In this context, we determined whether small wetlands, such as pools surrounding fished and fishless lakes, could sustain paedomorphic and metamorphic newts. To this end, we surveyed lakes known historically to sustain Alpine Newts (*Ichthyosaura alpestris*) as well as 35 nearby pools. On the basis of the published records, the only known population exhibiting paedomorphosis in the Swiss Alps was found to be extirpated by salmonid introductions. However, the metamorphs persisted in peripheral pools, paedomorphosis was discovered at a new locality, and overwintering larvae were still present in one of the lakes. These results show the importance of conserving varied aquatic habitats such as pools in mountainous environments where the main resources can become unsuitable for amphibians because of fish introductions. Pools may

also function as reservoirs in maintaining newt populations until programs to remove fish from lakes can be carried out. It is not known if paedomorphs could reappear after fish removal. However, the combined resilience of amphibians after fish removal and the genetic basis for paedomorphosis highlighted in other taxa by previous studies suggest that there is the potential to maintain this intraspecific case of diversity even after its disappearance.

M. Denoël, P. Scimè, N. Zambelli, *Curr.* Zool. 62, 1 (2016): 61–69. http://hdl.handle. net/2268/180066



Why have a pet amphibian? Insights from YouTube

#### John Measey, Annie Basson, Alex Rebelo, Ana Nunes, Giovanni Vimercati, Marike Louw& Nitya P. Mohanty

The desire to own a pet amphibian L is growing, and with it a growth in amphibian trade and in negative impacts on native populations, including disease transmission and invasive amphibian populations. We know very little about how or why people choose amphibians as pets, but amphibian owners share large numbers of videos on freely accessible platforms, such as YouTube. We aimed to use videos of captive amphibians to determine which species are kept, their life-history stage and the types of videos uploaded. We watched and categorized 1162 videos by video type, type of amphibian behavior and amphibian taxonomy (superfamily, family and species). We used data on the amphibian trade from the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES), on conservation status from the International Union for Conservation of Nature (IUCN) red list, and on potential environmental impact from published Environmental Impact Classification of Alien Taxa (EICAT) records, to determine potential conflicts of owning pet amphibians. We recorded 173 captive species in 847 videos with a taxonomic overrepresentation of salamandroids and pipoids, and an underrepresentation of ranoids and plethodontoids. When compared to videos of wild amphibian species, videos of captive animals featured disproportionate amounts of adults feeding, being handled and moving. The videos watched had a smaller proportion of threatened amphibian species, but a higher proportion of invasive species, than would be expected by chance, with the proportion present in CITES appendices (18%) being non-significant. We suggest that such data can be used to profile potential pets for trade and attempt to avoid conflicts with threatened and highly impacting alien species.

J. Measey, A. Basson, A Rebelo, A. Nunes, G. Vimercati, M. Louw & N. P. Mohanty. *Frontiers in Ecology And Evolution* (2019) doi: 10.3389/fevo.2019.00052



Adult male of alien Bufo japonicus formosus from a toadinvaded region of Hokkaido, Japan. Photo: Evangelia Kazila

Foraging traits of native predators determine their vulnerability to a toxic alien prey

### Evangelia Kazila & Osamu Kishida

he invasion of alien species can have severe effects on the survival of native fauna. We investigated the interactions and possible impacts of hatchlings of an alien toad species (Bufo japonicus formosus) on native frog tadpoles (Rana pirica) and native salamander larvae (Hynobius retardatus) in Hokkaido, Japan. Native salamander larvae are carnivorous and readily consume native frog tadpoles if their gape size enables them to do so. Native frog tadpoles exhibit a more omnivorous and opportunistic feeding behavior, grazing on plant and animal organic matter. The invasion of alien toads in ponds across the native habitat of native frogs and salamanders in Hokkaido could alter the dynamics of the native amphibian predator-prey system, as toad hatchlings are small enough to be considered as potential prey by native amphibians, but also carry toxins that could prove lethal for prospective predators. We designed a mesocosm and a laboratory experiment, to test the effects of alien toad hatchlings on native amphibians in a range of predation regimes and in near-natural conditions. Indeed, the native salamander larvae preyed on alien toad hatchlings as expected. Occasionally, native frog tadpoles were seen feeding on alien toad hatchlings, as well. Following alien toad consumption, both native species

experienced reduced survival. However, native frog tadpoles were impacted to a much greater extent compared to the native salamander larvae, suggesting that the former are less resistant to the alien toads' toxins. What came as a surprise though, was that native frog tadpoles experienced a disproportionately large reduction in their survival in relation to the amount of toad hatchlings they had consumed (i.e., consumption of one alien toad hatchling led to more than two native frog tadpoles being killed). To gain a better understanding of the specific processes of toxic impact, we proceeded by running a series of supplementary laboratory experiments. We found that omnivorous traits of native frogs such as food-sharing between conspecifics and feeding on toxic carcasses (i.e., native amphibians that had died as a result of alien toad hatchling ingestion) contributed vastly to their vulnerability. This study highlights the importance of investigating foraging traits of native species to determine their vulnerability and decide the course of action in cases of alien species invasion. Finally, it calls for further investigation of the actual impacts of the so far understated toxic alien toad invasion on native amphibians and other endemic fauna in natural systems in Hokkaido.

E. Kazila, O. Kishida, *Freshwater Biology*. **64**(1), 56-70 (2019).



Palmate Newt (*Lissotriton helveticus*) and Goldfish (*Carassius auratus*) during the night. Infrared photo: Laurane Winandy.

Temporal habitat shift of a polymorphic newt species under predation risk

### Laurane Winandy, Mélanie Colin & Mathieu Denoël

The temporal partitioning hypothesis suggests that the evolution of different diel activity rhythms in animals might facilitate the coexistence between prey and predators. However, the temporal shift of habitat use induced by predation has rarely been observed. The study of such a mechanism is particularly relevant for introduced species because it might explain how native species can persist or decline in response to the presence of alien species. The introduction of fish into ponds inhabited by amphibians has severe consequences

for their occurrence and abundance. Fish particularly affect an alternative newt phenotype, the paedomorph, which does not undergo metamorphosis and maintains larval traits such as gills at the adult stage. In a laboratory design, we assessed the diel patterns of habitat use in the 2 distinct morphological phenotypes of Palmate Newt (Lissotriton helveticus) in the presence or absence of Goldfish (Carassius auratus). Both newt phenotypes avoided a risky habitat more in the presence than in the absence of fish. This habitat shift was more pronounced during the daytime (i.e., when the risk could be considered higher for the newts) than during nighttime. However, in contrast to metamorphs, paedomorphs showed less adaptive changes according to temporal risk and remained in their shelter for most of the time. Temporal and habitat partitioning at the diel scale between native and alien species might promote their coexistence, but diel change can also imply a cost in the overall reduction of the time allocated to essential activities, showing that species interactions remain complex.

L. Winandy, M. Colin, M. Denoël, *Behav. Ecol.* 27, 4 (2016): 1025–1032. http://hdl. handle.net/2268/193100



Goldfish (*Carassius auratus*), an alien species in many ponds (Larzac, France). Photo: Mathieu Denoël.

The importance of phenotypic diversity in conservation: Resilience of palmate newt morphotypes after fish removal in Larzac ponds (France)

### Mathieu Denoël & Laurane Winandy

**R** esilience of organisms after threat removal is an essential feature to justify conservation efforts. Amphibians are particularly threatened with a worldwide decline, showing a low resistance to invaders such as fish. Previous research has shown that they could recover after fish extirpation due to metamorphosed colonizers. However, not all amphibian phenotypes are able to persist to fish introduction and disperse. In many species of newts and salamanders, paedomorphs retain gills in the adult stage, which makes them fully aquatic. A proposed way to conserve this phenotype would be to remove introduced fish from their habitats. However, because paedomorphosis is usually not expressed in the presence of fish, it is unknown whether fish removal could allow the resilience of paedomorphs. This would be possible only if progenies of metamorphosed individuals could become paedomorphic in restored habitats. Through a quantitative survey in three types of ponds, including control ponds without fish, ponds in which fish were extirpated, and fish ponds, we determined abundances of paedomorphic and metamorphic Palmate Newts (Lissotriton helveticus). The results show that paedomorphosis resilience is possible and even highly frequent, as paedomorphs were found in 80% of ponds where fish disappeared. Abundances were similar between these ponds and control ponds whereas fish ponds had almost no newts, indicating a very low resistance to invaders. This shows that conserving common phenotypes can help to preserve endangered phenotypes, as paedomorphs were produced through the reproduction of metamorphs. There is thus hope of maintaining intraspecific biodiversity though conservation action involving threat removal.

M. Denoël, L. Winandy, *Biol. Cons.* 192 (2015): 402–408. http://hdl.handle. net/2268/187526

Surface-water availability governs survival of an amphibian in arid mountain streams.

### Erin R. Zylstra, Don E. Swann & Robert J. Steidl.

mphibian declines have been linked A to a number of factors, including disease, habitat loss or degradation, and climate change. Changes in precipitation and temperature have the potential to affect several demographic parameters, particularly for species that are active yearround. We studied survival of juvenile and adult Lowland Leopard Frogs (Lithobates yavapaiensis) in mountain canyons in the Sonoran Desert of the southwestern U.S., where surface water levels in bedrock-lined pools vary greatly among seasons and years. We surveyed each of six stream reaches in two catchments 33-74 times between May 2013 and November 2015. We used photographs taken in situ to document observations of individuals based on their unique spot patterns and used Cormack-Jolly-Seber models to assess how surfacewater availability and other environmental factors influenced survival of juvenile and adult frogs. Annual rates of apparent survival were low (mean = 0.11). Monthly survival was associated positively with surface-water availability and also varied with ambient temperature, dew point, vegetation cover along pool perimeters, and year. Monthly rates of apparent survival were high ( $\geq 0.88$ ) when pools were full

or nearly so, but dropped precipitously when pools were < 50% full. Decreases in juvenile and adult survival during a severe drought in 2014 likely contributed to the subsequent extirpation of frogs from one catchment in 2015. Persistence of Lowland Leopard Frogs in the desert southwest over the long-term may be less likely if droughts become more frequent or severe, as predicted under climate change scenarios. Droughts are likely to decrease survival of larval and post-metamorphic individuals and limit the ability of frogs to disperse among increasingly rare sources of reliable surface water.

### E. R. Zylstra, D. E. Swann, R. J. Steidl, Freshwater Biology 64, 164–174 (2019). DOI: 10.1111/fwb.13204



Percent of ponds with each species (a), species richness at ponds (b), percent of ponds with ranavirus infected hosts for each species (c), and mean percent of hosts infected with ranavirus per pond (with 95% confidence intervals) of those collected of each species (d) in amphibian assemblages in the East Bay region of California in 2013. Numbers above bars indicate number of ponds with each species or species richness (n = 76). For plots (a), (c), and (d): *Ambystoma californiense*, California Tiger Salamander; *Anaxyrus boreas*, Western Toad; *Rana catesbeiana*, American Bullfrog: *Hyliola regilla*, northern Pacific Tree frog; *Rana draytonii*, California Red-legged Frog; *Taricha granulosa*, Rough-skinned Newt; *Taricha torosa*, California Newt.

The influence of landscape and environmental factors on ranavirus epidemiology in a California amphibian assemblage

Brian J. Tornabene, Andrew R. Blaustein, Cheryl J. Briggs, Dana M. Calhoun, Pieter T. J. Johnson, Travis McDevitt-Galles, Jason R. Rohr & Jason T. Hoverman

A fundamental goal of disease ecology is to determine the landscape and environmental processes that drive disease dynamics at different biological levels to guide management and conservation. Although ranaviruses (family Iridoviridae) are emerging amphibian pathogens, few studies have conducted comprehensive field surveys to assess potential drivers of ranavirus disease dynamics. We examined the factors underlying patterns in site-level ranavirus presence and individual-level ranavirus infection in 76 ponds and 1,088 individuals representing 5 amphibian species within the East Bay region of California. Based on a competing-model approach followed by variance partitioning, landscape and biotic variables explained the most variation in site-level presence. However, biotic and individual-level variables explained the most variation in individuallevel infection. Distance to nearest ranavirusinfected pond (the landscape factor) was more important than biotic factors at the site-level; however, biotic factors were most influential at the individual-level. At the site level, the probability of ranavirus presence correlated negatively with distance to nearest ranavirus-positive pond, suggesting that the movement of water or mobile taxa (e.g., adult amphibians, birds, reptiles) may facilitate the movement of ranavirus between ponds and across the landscape. Taxonomic richness associated positively with ranavirus presence at the site-level, but vertebrate richness associated negatively with infection prevalence in the host population. This might reflect the contrasting influences of diversity on pathogen colonization versus transmission among hosts. Amphibian host species differed in their likelihood of ranavirus infection: American Bullfrogs (Rana catesbeiana) had the weakest association with infection while Roughskinned Newts (Taricha granulosa) had the strongest. After accounting for host species effects, hosts with greater snout-vent length had a lower probability of infection. Our study demonstrates the array of landscape, environmental, and individual-level factors associated with ranavirus epidemiology. Moreover, our study helps illustrate that the importance of these factors varies with biological level.

B. J. Tornabene , *et al. Freshwater Biol.* **63**, 639–651 (2018). https://doi.org/10.1111/ fwb.13100

Assessing habitat quality when forest attributes have opposing effects on abundance and detectability: A case study on Darwin's frogs

#### Andrés Valenzuela-Sánchez, Benedikt R. Schmidt, Catalina Pérez, Tania Altamirano, Verónica Toledo, Ítalo Pérez, Sebastián Teillier, Andrew A. Cunningham & Claudio Soto-Azat

The understanding of species-habitat relationships is vital to inform the protection and management of imperilled species and their habitats. In the context of habitat protection and management, it is vital that the relationships between the proxies for habitat quality and the focal species abundance are based on robust data and inference. For instance, the inadequacy of relative abundance (i.e., raw counts) as a proxy of true abundance has been largely acknowledged: due to imperfect detection, true abundance can be underestimated to an unknown degree following this approach. By taking advantage of recently developed stratified capture-recapture spatially models, in this study we evaluated the effects of stand-level forest attributes on detection probability and local abundance for the endangered Southern Darwin's Frog (Rhinoderma darwinii). This fully terrestrial amphibian is endemic to the South American temperate forest. Our results support our hypothesis that forest structural attributes are an important component of the habitat of this forest-specialist frog. Namely, an increase of stand basal area and a decrease of daily microclimatic fluctuation were positively associated with the local abundance of R. darwinii. This evidence indicates that local abundance of R. darwinii in our study area is higher in old-growth, more structurallycomplex forest stands in comparison to earlier successional stages. These stand-level forest attributes also explained the amongpopulation variation in detection probability, although the relationships were opposite to those for abundance. Therefore, if raw counts are used in this specific case, abundance will tend to be underestimated to a larger degree in sites with good habitat quality than in those with poorer habitat quality, hampering the detection of appropriate habitat quality surrogates. Our results provide further support to previous claims that raw counts of individuals should not be used, generally, as a proxy of abundance in species inhabiting forest ecosystems and elsewhere. More importantly, the opposite effect of forest attributes on abundance and detectability observed in our study highlights the need to use methods that quantify species-habitat relationships in a robust way and which take habitat-specific imperfect detection into account.

A. Valenzuela-Sánchez, B. R. Schmidt, C. Pérez, T. Altairano, V. Totedo, et al., Forest Ecol. Manag. 432, 942–948 (2019).

### **INSTRUCTIONS TO AUTHORS**

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*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.

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

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A male southern Darwin's frog (Rhinoderma darwinii) found in an old-growth forest located at the Reserva Biológica Hullo Huilo, southern Chile. Photo credit: Andrés Valenzuela-Sánchez