

amphibian ark

Keeping threatened amphibian species afloat

Newsletter

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Amphibian veterinary training and advisory visits in Colombia

Luis Carrillo, Training Officer, Amphibian Ark

One of the multiple challenges that *ex situ* amphibian conservation managers face is keeping their animals alive and healthy. Health issues related to basic husbandry, nutrition, and disease will, if left unchecked, eventually cause the captive population to collapse, wasting precious time and other resources in the process. Amphibian caretakers must provide proper management to the species in their care from a health and veterinary perspective. To do so, a well-trained veterinary staff must be in place or available to help solve any clinical cases that arise.

Amphibian Ark has developed a program aimed to fulfil this need - the Amphibian Veterinary Outreach Program (AVOP) - an on-site training program with experts in amphibian husbandry, veterinary practice and population management. The AVOP team works with the overall objective to improve regional and local capacity for training, diagnostic and treatment.

Colombia is one of the countries rich in amphibian diversity, with close to 800 species and is also in the highest places in terms of endemism of amphibians. Sadly, Colombia is also in the top five countries in terms of threatened amphibians.

Currently there are five *ex situ* native amphibian programs in Colombia, three in zoos (Parque Zoológico Santa Fé, Zoológico de Santacruz, and Zoológico de Cali), one at an interactive museum (Parque Explora) and one private breeder (Tesoros de Colombia). After communications with the Colombian Zoos and Aquariums Association and sending a survey to explore the needs of those projects, we collectively decided to deliver a veterinary medicine training course followed by an advisory courtesy visit to each of the different programs.

The amphibian veterinary medicine training course was held at the Cali Zoo from November 10-12 with two main objectives: to build capacity among Colombian veterinarians in amphibian medicine; and to create partnerships amongst the participants and their institutions to collaborate and help each other in resolving medical issues with captive amphibians. To deliver the course, a team of experienced veterinarians was selected, including Brad Wilson (private practice and Atlanta Botanical Garden), John Sykes (Bronx Zoo/Wildlife Conservation Society), Carlos Rodriguez (Disney's Animal Kingdom) and Luis Carrillo (Amphibian Ark).

Seventeen participants representing eleven institutions from Colombia and Bolivia were present during the course, which was designed to encourage participation and sharing of knowledge and expertise among participants and trainers, during the lectures and practical sessions and through different group activities.

The course consisted of a mix of lectures, group activities and practical sessions designed to deliver the most relevant information about amphibian veterinary medicine. The lectures delivered critical information and confronted students with real problems that they had to solve. Students also had the opportunity to put into practice what they had learned during the lectures/group work sessions. The students were also able



Above: Amphibian Ark's AVOP team members John Sykes (Wildlife Conservation Society), Brad Wilson (Atlanta Botanical Garden) and Luis Carrillo (AArk) assessing and advising at one of the amphibian programs in Colombia.

Below: Students at the water quality practice during the Amphibian Veterinary Medicine Course. Maintaining water quality is crucial for amphibian health.

Photos: Carlos Rodriguez.





Group photo during the Amphibian Veterinary Medicine Course at Cali, Colombia. Photo: Carlos Rodriguez.

We hope this training and advisory visits helps to improve the husbandry and care of amphibians in Colombia and we will continue to support the current and future programs in any way we can.

to bring real life cases where they presented to the rest of students and instructors their own clinical cases, and how they approached diagnosis and treatment so they could receive feedback on it.

Immediately after the course the team visited the different institutions located in Cali, Medellín and Bogotá to assess the different programs in terms of medical, biosecurity and illness related to husbandry issues. The visits were very productive and the team was able to advise on general issues and also to evaluate animals that were not well at the time and to propose appropriate treatment. A report was produced for each of the institutions with recommendations about the issues observed during each visit.



John Sykes (Wildlife Conservation Society) and Carlos Rodriguez (Disney's Animal Kingdom) giving advice at Parque Explora, Medellín, Colombia. Photo: Luis Carrillo.



AArk T-shirts make great holiday gifts!

Head to the AArk clothing store at www.amphibianark.org/AArk-products.htm and check out our clothing items - if you can't find the design and color combination you're looking for, drop us a line and we'll see if we can create it for you!



Amphibian program resources on the AArk web site

As part of our commitment to building amphibian husbandry expertise and ensuring that best practice husbandry and biosecurity guidelines are followed, Amphibian Ark has developed a number of tools for amphibian program managers over the past few years. We have also greatly expanded our online library of husbandry-related documents, and we're also encouraging others to make their husbandry and management documents available to other program managers.

The primary page on our web site for finding all of these resources is the Amphibian Husbandry page, www.amphibianark.org/amphibian-husbandry/. Some of the most-used documents and tools are highlighted below.

Husbandry documents

The Husbandry Documents page on the AArk web site, www.amphibianark.org/husbandry-documents/, now contains over 140 documents, on a wide range of topics including: Enclosures, Nutrition and feeding, Light and UV, Biosecurity and quarantine, Diseases, Drugs and treatments, and Reproduction. There are also a range of taxon-specific husbandry guidelines and taxon management plans. These species-level husbandry guidelines are a valuable resource when establishing programs for related species which may have similar husbandry requirements.

Additional documents are regularly being added to the page, and we're also encouraging anyone who manages amphibians in captivity to upload and share their husbandry documents, protocols and management plans. Simply use the quick upload form at the bottom of the page, and we'll make your documents available to help other program managers.

Species assessed for *ex situ* programs

Since 2007, Amphibian Ark staff have facilitated assessment of the conservation needs of 3,375 (46%) of the world's amphibian species through 26 national or regional workshops. The results of these assessments are available on AArk's web site, www.amphibianark.org/assessmentresults.htm. Zoos, aquariums and other captive breeding organizations that are considering the implementation of new amphibian conservation programs are strongly encouraged to consider species that have been recommended for urgent rescue (www.amphibianark.org/rescue-species/) or for *ex situ* research programs. There is also a list of existing rescue and *ex situ* research programs at www.amphibianark.org/progress-of-programs/.

Information regarding the likely availability of founder animals and the completion of a relevant taxonomic study is included in the lists of species assessed for rescue, where known. This information may indicate which species are currently appropriate for captive rescue programs, and those which may benefit from additional research in the wild prior to a captive program being established. Species expected to have wild founders available and which have undergone a complete taxonomic analysis in the wild should perhaps be considered ahead of others where information is incomplete. Where possible, AArk strongly recommends that new programs should be established within the range country of the species.

Program implementation tool

A significant challenge for *ex situ* programs relates to ensuring that all programs are adequately supported for their duration. Establishing facilities and collecting rescue populations is only the first, albeit perhaps the single greatest expense. However, it is insufficient to support only those first-year expenses without operational support for the long term, which may amount to years or even decades. In addition to financial planning, *ex situ* programs should establish at the onset a plan for working with partners to mitigate threats in the wild and, where necessary, releasing animals back into the wild, as well as how to distribute the progeny of captive animals in the interim.

We have developed an easy to use checklist that should be utilised prior to the commencement of any *ex situ* conservation breeding program for amphibians. A new program should be implemented if, and only if, all of the critical program requirements can be met for a species.

The program implementation tool is available to use online (www.amphibianark.org/tools/Program%20implementation%20tool.htm) or it can be downloaded and used offline (www.amphibianark.org/tools/AArk%20Program%20Implementation%20tool.xls). Data should be entered for each species that is being considered for an *ex situ* conservation program.

Determining founder numbers

One of the most important aspects of establishing a new *ex situ* population is obtaining sufficient founder animals (unrelated individuals who help establish a population), but unfortunately, this is often over-looked when new programs are established.

Amphibian Ark recommends that at least twenty pairs of animals (or groups of individuals) are collected as founder animals. Ideally these would be unrelated and will successfully reproduce, but of course that cannot be guaranteed. Realize that many more than this number may have to be captured to ensure that twenty pairs actually survive and successfully reproduce.

We have developed a tool to help calculate the number of founders that should be collected, based on the reproductive biology of the species being considered. The tool uses data from our Amphibian Population Management Guidelines (www.amphibianark.org/pdf/AArk-Amphibian-Population-Management-Guidelines.pdf). After some basic biological values and management types have been selected from the options in the tool, the results will show the number of founder animals that should be collected and the target population sizes for the selected reproductive model.

The founder calculation tool is available for use online (www.amphibianark.org/tools/Founder%20calculation%20tool.htm) or can be downloaded and used offline (www.amphibianark.org/tools/AArk%20Founder%20calculation%20tool.xls). Data should be entered for each species that is being considered for an *ex situ* conservation program.

As always, AArk staff and our dedicated associates are available at any time to provide additional support and guidance when needed. If you would like us to help with any aspect of your captive amphibian programs please feel free to email us at info@amphibianark.org and we will ensure that your questions are answered.

Communicating the international successes of *ex situ* amphibian conservation programs

Noah Shields, Senior Zookeeper, Columbian Park Zoo, Indiana, USA, and Kevin Johnson, Taxon Officer, Amphibian Ark

As we move into the future, the outlook for amphibian survival may seem bleak...newly emerging diseases, rampant habitat destruction, invasive species and climate change. Even though the odds seemed stacked against the entire class, scientists and conservationists alike have devoted their entire careers to aid in the preservation and protection of these species. Professionals around the globe are beginning to work in unison to make important strides in amphibian conservation. As species are evaluated, and management programs are designed and implemented, it becomes increasingly desirable to have data on our progress towards preventing extinctions available to present to the scientific community.

Amphibian Ark is at the forefront of this work and has developed a range of tools and resources available to *ex situ* program managers (www.amphibianark.org/amphibian-husbandry/). One of the projects that Amphibian Ark has been pursuing is the development and maintenance of an online database available to *ex situ* conservation program managers around the world. This database was developed to aid Amphibian Ark's role in monitoring and documenting the global network of captive conservation and research programs for species assessed during an Amphibian Conservation Needs Assessment (www.amphibianark.org/planning-workshops/) as needing urgent *ex situ* rescue or research, and similar programs in countries where an AArk conservation needs assessment has not yet been carried out. The progression of each program through a series of key milestones is documented, with the eventual outcome of each program being phased out, once species are once again safe in their natural habitats. Michael McFadden, Herpetofauna Division Unit Supervisor at the Taronga Conservation Society Australia, utilizes the *ex situ* database to manage *ex situ* conservation programs and considers the database "A great way to bring together projects from around the world and the relevant contacts for those projects. Much of the information relating to these *ex situ* programs and reintroductions is not published so this provides an avenue to make this information available."

The importance of a centralized, current database of *ex situ* program information has become glaringly clear as conservation organizations are continually making efforts to make precious little resources, such as staff time and money, reach the furthest. For many countries, the priority species for *ex situ* conservation programs are known but information regarding which of those species are already being maintained in captive programs and which ones need captive programs designed and implemented still needs to be identified. Additionally, resource needs must be presented to prospective donors efficiently to promote their direct support of *ex situ* programs, and all needs that are identified in the database by program managers are automatically included on the AArk web site, where donors are encouraged to select a program that they would like to support (www.amphibianark.org/supporting-programs/).

An initial part of AArk's role is to evaluate species to aid conservation managers in maximizing the impact of their limited resources by identifying which amphibian species are most in need of particular types of conservation action. To date, over 400 species have been assessed for urgent rescue, in more than 25 countries, but currently, official confirmations of established programs are available for **only 59 of those 400+ species** (www.amphibianark.org/progress-of-programs/). The *ex situ* database allows the AArk team and the international conservation community to identify which programs are in existence and their progress in order to prevent duplicate programs and wasting valuable resources. It also includes lists of species assessed for *ex situ* conservation which have not yet been brought into captive programs, and therefore, should be high priorities when new *ex situ* programs are being considered.

Part of the fulfilment of the Amphibian Conservation Action Plan (ACAP, 2007) can be satisfied from the information and data contained in the *ex situ* program database. The ACAP states that an "advisory board would also conduct regular evaluations of each program to determine its relative success or failure." The most efficient method to achieve this goal is for each manager of an *ex situ* conservation program to regularly provide basic information about the progress of their respective programs. The database also facilitates AArk's responsibility for fulfilling its obligation of maintaining close contact and communication among all facilities in the network, as outlined in the ACAP.

This online database was designed with program managers in mind, with ease of use being one of the top priorities. Program managers are tasked with maintaining their programs, training staff, and seeking funding, among many other distractions. A program database with many hoops and difficulties involved with data input was avoided to streamline the process for program managers. The database was designed to include key milestones in the progression of any *ex situ* program. As managers input data into the database, many of the details can be selected from pre-determined responses, so minimal time and typing is required. According to Devin Edmonds, Amphibian Conservation Director at Association Mitsinjo in Madagascar, "The [online database] interface feels familiar and is a good way to not only share what we're doing in Andasibe but also review similar projects elsewhere. It's a way to connect not only other program managers together but also, and perhaps more importantly, potential donors to projects needing support." AArk staff continues to receive feedback from database users on additional features that would make it even more useful, such as funding resources and permits, and additional enhancements will be made as further input is received, and development resources are available.

The program database stands to serve as an important tool for Amphibian Ark and the conservation community. Cybele Lisboa is the Curator of Reptiles, Amphibians and Invertebrates at the São Paulo in Brazil, and she uses the online program to share the Zoo's experiences with their *ex situ* research and rescue programs. "A lot of information from this online database about *ex situ* programs will be available to program managers so they have examples that can be applied for current and future *ex situ* programs." Cybele says. As more and more program managers participate in this project, the information and data gathered from the database will become even more encompassing and provide a broader and more complete understanding of amphibian programs. With an increased understanding, AArk officials and other conservation entities can begin to make more concerted and choreographed efforts in amphibian conservation.

All managers of *ex situ* amphibian rescue and research programs are encouraged to use the online database to communicate their progress with others, and to connect with managers who are running programs for similar species. Logins have been provided to all known program managers and distributed via email - if you have forgotten your login details, or have not yet added your programs to the database, please contact AArk staff at exsituprogress@amphibianark.org. To update your program details in the database, please visit <http://progress.amphibianark.org/>.

Reference

Gascon, C., Collins, J. P., Moore, R. D., Church, D. R., McKay, J. E. and Mendelson, J. R. III (eds). 2007. *Amphibian Conservation Action Plan*. IUCN/SSC Amphibian Specialist Group. Gland, Switzerland and Cambridge, UK. 64pp. www.amphibianark.org/pdf/ACAP.pdf.

Be a salamander and newt hero!

Get your pets screened for *Batrachochytrium salamandrivorans*

If you have a pet salamander or newt, or are simply an amphibian lover, then chances are you have heard about the very worrying news recently about a new fungal disease that is killing off salamanders in Europe.

Batrachochytrium salamandrivorans (*Bsal*) seems to have originated in Asia where it co-exists with local salamander species and was probably accidentally introduced by humans into Europe. With North America being so important for salamanders we need to protect both wild and captive salamanders and newts as quickly as possible. As we work with our partners to address the threats to wild populations we also want to help you keep your pets as safe as possible. To help with this process we highly recommend the following three steps:

Bleach waste water

Before you dispose of any waste water that might have come in contact with your pet salamanders/newts be sure you add a little bleach before you dispose of it. Although the science is still out on the concentrations needed, the best evidence points towards a ratio of about 1 part bleach to 10 parts water. Using this ratio should ensure any of the deadly *Bsal* fungus is killed before leaving your home.

Don't release captive salamanders into the wild

More important than ever, please don't release your pets into the wild. If a captive salamander or newt has *Bsal* and ends up in the wild it could be devastating for wild populations. If you have a salamander that you are no longer able to look after try contacting your local herpetological society, humane society or zoo.

Get tested

Finally get your pet salamander and newts tested. The easiest way to be sure that your salamander is healthy and does not pose a risk to wild populations is to get it tested for *Bsal*. For a limited time only the Amphibian Survival Alliance (ASA, www.amphibians.org) is offering to test the first 500 swabs that they receive from pet salamanders and newts in the Contiguous US. Sign up and the ASA will send you a FREE SAMPLE KIT and let you know the

results as soon as they have them. You can Sign up now for your free kit while supplies last at <http://eepurl.com/7-Xwv>.

By doing all this you are not only protecting your salamanders but also the wild salamanders that we all enjoy so much.

Please share this information with your friends, online through social media and your favorite forums, local herpetological societies and pet stores. You can also read the letter (www.amphibianark.org/pdf/lt-Ashe-Bsal-final.pdf) from Amphibian Ark, the Amphibian Survival Alliance, the Center for Biological Diversity, the Center for Invasive Species Prevention, Defenders of Wildlife, and the Natural Areas Association to the US Fish and Wildlife Service, calling for urgent and immediate action to keep *Bsal* out of the US.

More information about keeping salamanders and other amphibians is available on the Amphibians as Pets page on our web site, www.amphibianark.org/amphibians-as-pets/.



The Southern Two-lined Salamander (*Eurycea cirrigera*) and the Eastern Newt (*Notophthalmus viridescens*) are just two of many species of newts and salamanders that potentially face decimation if the new fungal disease *Batrachochytrium salamandrivorans* (*Bsal*) arrives in the US. Photos: Robert Hill.



National Amphibian Conservation Coordinator for the Philippines

Cindy Lucero, ASG Pilipinas' National Amphibian Conservation Coordinator

During the amphibian Conservation Needs Assessment workshop that was held in Manila in July this year, the Amphibian Specialist Group (ASG) Pilipinas members discussed how best to coordinate the development of a National Amphibian Conservation Action Plan for the Philippines. It was decided that hiring a person in a half-time position, dedicated to the development of the Action Plan, and helping to coordinate future amphibian conservation activities would be of great benefit. A position description was drawn up, and thanks to the generous support of the Virginia Zoo in Norfolk, Virginia USA, funding has been secured for the first twelve months of this position (see articles in the September edition of the AArk Newsletter, www.amphibianark.org/Newsletters/AArk-newsletter-28.pdf). A Memorandum of Understanding has been drawn up between the Virginia Zoo and the ASG Pilipinas, and in October, I was appointed to the part-time position of National Amphibian Conservation Coordinator for the Philippines. My goal is to update the existing database of species presence in the Philippines and assist in creating an action plan for the Philippine amphibians.

I finished my bachelor's degree in Biology and worked as a project research assistant for the World Wide Fund Philippines. I have also volunteered in various civic organizations. My previous working experience has been a tremendous advantage for this position, as understanding the science is essential, but also being able to develop programs, manage, and communicating with a team of people with dynamic backgrounds is extremely important with my position in ASG Pilipinas.

I have always enjoyed the people and the diversity of work I do. The role of National Amphibian Conservation Coordinator is a very interesting job, as conservation is now being focused on what were previously thought of as the least priority species in our country. I personally had not known that our frogs are on the thin line of extinction.



The Luzon Striped Stream Frog (*Hylarana similis*) was assessed as one of the highest priority species for various conservation actions during the Conservation Needs Assessment for the Philippines earlier this year.

Photo: Arvin Diesmos.

Under the supervision of ASG Pilipinas, my work as National Amphibian Conservation Coordinator is to develop a plan of action. This includes formulating ways to monitor the amphibian wildlife populations, human-animal interaction and conservation status. I am also responsible for gathering, classifying, and disseminating the available information to the team. Accordingly, I may seek assistance from government and private institutions and agencies in order to complete the action plans. Lastly, I may help in writing the National Amphibian Action Plan as needed.

Captive breeding of three species of Costa Rican moss salamanders

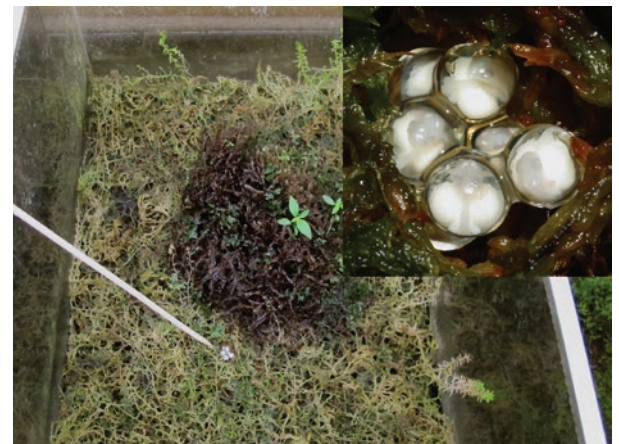
Brian Kubicki, Costa Rican Amphibian Research Center, Costa Rica

Costa Rica is home to 200 species of amphibians within a tiny national territory of approximately 51,000 km². Nearly a quarter of Costa Rica's amphibian diversity is made up of three genera and forty-nine species of salamanders. Unfortunately most of Costa Rica's salamanders are still surrounded by mystery in regards to their general biology and actual distributions.

In 2013 the Costa Rican Amphibian Research Center (CRARC) was awarded \$4,700 through an Amphibian Ark Seed Grant for a project titled *Ex situ* methodology building for Neotropical caudates, with a special emphasis on three species of Costa Rican Moss Salamanders of the genus *Nototriton*. This *ex situ* aspect is part of a larger project by the CRARC to obtain a better understanding of Costa Rican salamanders through research efforts both in the wild and captivity.

A principal focus of the CRARC salamander work has been dedicated to the poorly known and understood moss salamanders of the genus *Nototriton*. Costa Rica is home to eight species of moss salamanders, and all are currently known to be endemic to the country. The CRARC has dedicated hundreds of hours in the field to gather more information on the general biology and distributions of Costa Rican moss salamanders and these field studies have been crucial in providing the knowledge necessary to allow the designing of terrariums that provide the specific needs these animals require.

During 2013 the CRARC collected several species of Costa Rican moss salamanders to form captive colonies. The salamanders that were collected adapted very well to their specially designed terrariums but despite their adapting well to the terrariums, no breeding was observed during 2013. During the period of June to October of 2014 several egg masses were discovered in the terrariums at the CRARC. These breeding events represent the first known captive breeding for any species of *Nototriton*. The three species of *Nototriton* that bred in the CRARC lab are *Nototriton abscondens*, *N. gamezi*, and *N. major*. Successfully raising the tiny captive bred moss salamander neonates presented some initial difficulties, but modifications to their food items and enclosures have shown positive results thus far.



An egg mass of *Nototriton abscondens* within a terrarium in the CRARC lab. Insert in the upper right corner shows the details of the developing embryos.

Photo: Brian Kubicki.



A captive-bred individual of *Nototriton abscondens* from another egg mass at four months old. Photo: Brian Kubicki.

Working together to develop Madagascar's amphibian captive breeding capacity

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The Amphibian Conservation Action Plan (ACAP, 2007) highlighted captive programs as being an essential component of integrated amphibian conservation plans and central to this long-term success was the establishment of captive operations in range countries (Gascon et al. 2007). Doing this however is easier said than done, especially in countries lacking infrastructure, resources and capacity to easily implement them. One such country is Madagascar.

A mega-diverse island nation, Madagascar has a tremendously rich diversity of amphibian species, with four families, twenty-five genera and potentially upwards of 400 species, of which over 99% are endemic and many are yet to be described. Along with much of Madagascar's other unique fauna its amphibians face a number of threats, most notably habitat loss but also the ongoing effects of climate change, over-harvesting for the international pet trade and the recent discovery of chytridiomycosis in exported frogs is a new potentially devastating threat.

It was the threat of chytridiomycosis and its potentially catastrophic consequences that the 2008 Sahonagasy Action Plan used to urge the need for investment in *ex situ* breeding programs (Andreone et al. 2008). Now that it has been discovered in exported Malagasy frogs immediate attention is therefore warranted to develop the infrastructure and personnel in Madagascar needed to enact *ex situ* conservation programs as an important component in safeguarding Madagascar's threatened amphibian species.



The recently-renovated frog and insect breeding facility at Parc Ivoloïna in Madagascar. Photo: Devin Edmonds.

ums) funded Amphibian Conservation Husbandry training course in 2012. This included Malagasy participants from eight institutions, including Madagascar Flora and Fauna Group's (MFG) Parc Ivoloïna, a zoo and forestry station located near the eastern coastal city of Toamasina.

Parc Ivoloïna is now the site of a second small amphibian captive breeding facility, and was the recipient of an Amphibian Ark Seed Grant in 2014. Completed in October 2013, staff have since been culturing fruit flies, developing biosecurity protocols, and finally maintaining a captive group of the Common Reed Frog (*Heterixalus madagascariensis*) as a practice species. It is imperative that any new captive breeding facility or initiative begins on the correct foundation on which to build and develop. Parc Ivoloïna is in the fortunate position of having the technical expertise and a successful model in country with Mitsinjo.

Collaborative actions

As a key component of Durrell's Madagascar amphibian program Durrell are facilitating a series of training exchanges between Association Mitsinjo and MFG Parc Ivoloïna. These exchanges will enable the specialized amphibian technicians at Mitsinjo to share their expertise with the MFG team, working together to trade ideas and to improve the park's newly developed facilities.

Mitsinjo made an initial visit to Ivoloïna at the beginning of May 2014. Following this trip a plan for the coming months was drafted which includes minor infrastructure improvements - for example the installation of an isolated quarantine room - and then training exchanges whereby staff from Parc Ivoloïna work alongside Mitsinjo in Andasibe and vice versa. An improved version of the curriculum used to train Mitsinjo technicians during the first six month period in 2010-2011 when the facility in Andasibe was being constructed will be used with Ivoloïna staff. Not only will this be useful for keepers at Ivoloïna, but it will also provide an opportunity for Mitsinjo's technicians to become teachers of captive amphibian husbandry themselves, building upon their more than three years existing experience working with captive frog populations in Andasibe.

Building on solid foundations

Fortunately this process is not starting from the complete beginning. In early 2011, through a collaborative effort between Association Mitsinjo, the Amphibian Specialist Group of Madagascar, and the Direction Générale des Forêts a breeding facility was developed in Andasibe, east-central Madagascar. Currently operating a breeding program for the Critically Endangered Golden Mantella (*Mantella aurantiaca*), the biosecure facility now manages nine species, all of which are native to the local area, in addition to sustaining populations of a range of live food cultures. These species comprise a variety of breeding and life history types which provide information and guidelines for future threatened species.

The breeding facility is operated by the locally-run conservation organization Association Mitsinjo and is staffed by a team of eight residents from the Andasibe area. This impressive operation is testament to what can be achieved in Madagascar and is an exemplary model for other breeding facilities to follow.

To help this initiative and expand on the existing knowledge developed in Andasibe, Durrell Wildlife Conservation Trust (Durrell) jointly ran an EAZA (European Association of Zoos and Aquariums) funded Amphibian Conservation Husbandry training course in 2012.

This included Malagasy participants from eight institutions, including Madagascar Flora and Fauna Group's (MFG) Parc Ivoloïna, a zoo and forestry station located near the eastern coastal city of Toamasina.

These training exchanges are being supported by additional activities to guide future captive breeding initiatives. Mitsinjo and MFG with assistance from Durrell and Amphibian Ark are applying the AArk Amphibian Conservation Needs Assessment process (AArk, 2014) to the species in the Andasibe and Betampona areas respectively. Doing so will provide information on which species to prioritize for future captive breeding and allow the necessary preparations to be made. This could involve taking in a common analog species so as to develop husbandry guidelines and protocols prior to bringing in threatened species when needed.

Following on from this assessment, collecting crucial life history and ecological information on priority species is critical for their future captive breeding. Durrell will be working with Mitsinjo to do just this for five Data Deficient species that have been identified as being at high risk from chytridiomycosis in the Andasibe area (Lötters et al. 2011). Through this Mitsinjo staff and local community members will be trained in amphibian survey and monitoring techniques, again improving the local capacity for amphibian conservation.



Raising *Boophis* tadpoles at Mitsinjo. Photo: Devin Edmonds.

We are hopeful that through these training exchanges, further capacity will be built in Madagascar to allow rapid *ex situ* action to take place for the unique and highly endemic amphibian fauna of the island. Importantly we also hope this will be the beginning of a broader goal to build a national captive breeding network for Madagascar. This will enable the sharing of ideas, techniques and skills amongst the centres and develop an integrated collaborative ethos within the amphibian captive breeding community which will ultimately benefit the threatened amphibian fauna of Madagascar.

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Taking the first steps together at Parc Ivoloïna

Devin Edmonds, Amphibian Conservation Director, Association Mitsinjo, Madagascar

Parc Ivoloïna, northeast Madagascar, 12 November 2014, 19:00 - Mampionona and Nirina squat near the ground and shine their lights under fallen palm fronds and debris on the side of the dirt path. It's sticky humid, the air is thick. Frogs call in the distance, snoring drones of *Mantidactylus* at the water's edge and rapid notes like gunfire further above from *Heterixalus* reed frogs. Although trained in amphibian biology and captive husbandry, the two Mitsinjo technicians aren't looking for frogs tonight. Instead, they're on the hunt for frog food.

In a woven basket they carry several plastic containers, each one containing a handful of crinkled paper and live crickets sorted by species. Some of the insects look familiar, perhaps are even the same as the ones they culture at Mitsinjo's captive breeding facility further inland near Andasibe, but others are completely new and we don't know whether or not we'll be successful setting up colonies at Parc Ivoloïna. It's this first step that we're focused on, establishing live food cultures, and as anyone who has spent time managing large populations of frogs in captivity already knows it is not only the most time-consuming aspect of the job but also perhaps the most important.

Importing live insects to Madagascar was not a viable option when we set up our captive breeding facility in Andasibe in 2010. We didn't want to risk potentially introducing a new pest or foreign species to the island. Instead, Mampionona and Nirina worked with me and four other Mitsinjo members to catch local crickets already abundant around Andasibe village, which we had to learn how to culture ourselves. For nearly six months prior to keeping our first frogs we cleaned plastic boxes stuffed with egg carton, incubated cricket eggs, recorded how long it took for them to hatch, and attempted to identify what species we were producing. A Russian entomologist eventually identified one of our crickets as *Malgasia marmorata*, a species unknown for more than a century and finally redescended this year using stock from our insect colonies.



After four days of searching, Mampionona from Mitsinjo, and Rakoto and Pascal from Parc Ivoloïna had found half a dozen different kinds of crickets in quantities large enough to attempt to start breeding colonies. Photo: Devin Edmonds.

With a 2014 Seed Grant from Amphibian Ark and further support leveraged through Durrell Wildlife Conservation Trust, Parc Ivoloïna recently finished renovations on a small building set aside specifically for amphibians. New walls were put up, an isolated quarantine room constructed, shelving units and terrariums built, and ventilation improved so as to better control the temperature of the structure. Biosecurity measures were also put in place - a separate entrance room to change into dedicated clothing and footwear, protocols developed for cleaning and disinfecting equipment, lists of prohibited materials that are unsafe to bring inside were written. The progress moved along at a steady pace. By November it was time to start culturing live foods.

After four days of searching, Mampionona and Nirina had found half a dozen different kinds of crickets in quantities large enough to attempt to start breeding colonies. Bernard Lambana Richardson, responsible for managing the zoological park, brought in several dozen plastic boxes for cricket housing from the nearby city of Toamasina. Parc Ivoloïna's staff learned how we operate our cricket production room and we discussed some of the difficulties we have been through in Andasibe and how best to avoid them. We weren't starting from scratch, but rather taking established protocols from Mitsinjo and applying them to Parc Ivoloïna.

Mitsinjo took something back from the trip as well - vermiculture techniques. As part of a demonstration garden used for training local people in sustainable agricultural practices Parc Ivoloïna cultures earthworms. While useful for a variety of agriculture purposes, earthworms are also an extremely healthy food source for captive amphibians and one Mitsinjo has considered looking into for some time. Mampionona and Nirina took photos and notes about the worm bins and discussed the process with park staff, with future plans to install a similar worm culturing operation in Andasibe to supply our captive frogs with a new additional food source.



As part of a demonstration garden used for training local people in sustainable agricultural practices Parc Ivoloïna cultures earthworms, which are an extremely healthy food source for captive amphibians. Photo: Devin Edmonds.

We still have a lot to do. Once live food colonies are established at Parc Ivoloïna, amphibian husbandry expertise will need to be developed and eventually proficiency in captive population management built up. Fortunately, Madagascar Fauna and Flora Group, the organization that oversees the operation of the zoological park, already has the right relationships in place to bring in this specialized knowledge, both what already exists in country through Mitsinjo and from within the broader international zoo community. From our side, we're looking forward to watching Parc Ivoloïna's new cricket colonies grow, with hopes that soon there will be frogs inside terrariums to practice feeding. This is, after all, about the frogs, and the sooner we are prepared to enact *ex situ* conservation breeding programs at another site in Madagascar the better off they will be.



The start of live food cultures in the recently renovated amphibian building at Parc Ivoloïna. Photo: Devin Edmonds.

Can captive-reared tadpoles re-establish Crawfish Frog populations?

Rochelle Stiles, Indiana State University and Michael Lannoo, Indiana University School of Medicine, USA



Metamorphosed Crawfish Frog (*Lithobates areolatus*). Tadpoles are raised from hatching to Gosner stage 25-41 in 100-gallon tanks at the Detroit Zoo and are then released. Photo: Rochelle Stiles.

Crawfish Frogs (*Lithobates areolatus*) belong to the most endangered anuran clade in North America. Of the four species in this clade, Mississippi Gopher Frogs (*Lithobates sevosus*) are listed in the IUCN Red List as Critically Endangered, while Crawfish Frogs and Gopher Frogs (*Lithobates capito*) are listed as Near Threatened globally. Researchers are currently developing techniques to conserve these imperilled frogs. Our collaborative research focuses on Crawfish Frogs, a species where we have demonstrated unusually high mortality (> 1% of the larval population dies daily) in the larval stage, which causes a population bottleneck. Therefore, one key to both maintaining existing populations and restoring or establishing new populations appears to be to reduce larval mortality. A successful, large-scale repatriation effort across their former range, backed by a captive-rearing program has the potential to recover this species.

In cooperation with Detroit Zoological Society, we captive-reared and released 510 juvenile Crawfish Frogs in 2013 and 2,998 juveniles in 2014. Both years, we used egg masses from Indiana's largest breeding aggregation - Hillenbrand Fish and Wildlife Area-West in Greene County, Indiana, USA. In 2013, we raised tadpoles from hatching to Gosner stage 25-41 in 100-gallon tanks at the Detroit Zoo and released these late-stage tadpoles into an enclosure at Hillenbrand Fish and Wildlife Area-West. Upon metamorphosis, juveniles exited the enclosure and were captured by a drift fence and pitfall traps. Prior to final release,

we measured, weighed, and marked (toe clipped) juveniles and documented malformations. In response to shrew predation, we augmented our drift fence with mammal-proof hardware cloth and transferred remaining tadpoles to reptariums (aquatic mesh cages) for their protection. In 2014, we raised tadpoles from hatching to pre-Gosner stage 25 in custom, shallow pools at Detroit Zoo and moved tadpoles to reptariums at Hillenbrand Fish and Wildlife Area-West. Upon metamorphosis, we processed these juveniles in the same way as animals raised in 2013.

Our results suggest Crawfish Frogs are negatively affected by partial cannibalism (i.e., exhibit reduced toes or limbs) when held at high densities in late larval stages and are sensitive to environmental conditions during development. Of the 510 frogs released in 2013, 155 (30%) were normal, while 355 (70%) were malformed. Of the 2,998 frogs released in 2014, 1,872 (62%) were normal, while 1,123 (38%) were malformed. However, unlike 2013 where almost all malformed exhibited partial cannibalism, only 180 (6% of metamorphosed juveniles) malformations resulted from cannibalism. In 2014, we also observed scoliosis (609 animals, 20%), knee/hip dysplasia (i.e., non-normal limb angles, 199 animals, 7%), and a combination of some or all malformations previously stated (135 animals, 5%). Lastly, in 2014, survivorship during the later developmental stages was reduced by disease; of the 4,951 tadpoles raised at Hillenbrand Fish and Wildlife Area-West, the majority of 1,953 animals (39%) likely died from Ranavirus.



Detroit Zoological Society and volunteers assisting with the transfer of Crawfish Frog tadpoles to reptariums at Hillenbrand Fish and Wildlife Area-West, Greene County, Indiana. Photo: Marcy Sieggreen.

We will continue to monitor these captive-reared Crawfish Frogs as they return for the first time to breed in 2015–2016 and plan an additional year of rearing tadpoles with further modifications. As the first Crawfish Frog repatriation attempt, this work will contribute to a better understand of and guidelines for augmentation and re-establishment techniques for this threatened species locally and nationally.

Project Lemur Frog

Claes Andrén, Scientific Director, Nordens Ark, Sweden

The Lemur Leaf Frog (*Agalychnis lemur*) is a Critically Endangered Phyllomedusine frog from Central America. Populations of Lemur Leaf Frog have severely declined over all its former range in recent years, due at least in part to the chytrid fungus *Batrachochytrium dendrobatidis*. In Costa Rica the species is now only known from one or two localities. There is a significant difference between the Costa Rican and the Panamanian populations, similar to that found between other different *Agalychnis* species. This has indicated the necessity to manage populations of these frogs from Costa Rica and Panama separately for conservation purposes. A collaborative project is now running with the aim to secure the species' future through a combination of specific research, education, and focused support for both *in situ* and *ex situ* conservation.

An international collaboration between several committed individuals and institutions has been developed, aimed at conserving Costa Rican Lemur Leaf Frogs. The project is managed by Andrew Gray at Manchester Museum (UK) in close cooperation with Claes Andrén at Foundation Nordens Ark (Sweden), Tim Skelton at Bristol Zoo (UK), Brian Kubicki at The Costa Rican Amphibian Research Center (Costa Rica) and Robert Jehle at Salford University (UK). Project Lemur Frog aims to support conservation of the last known wild population of the frog in Costa Rica, through habitat protection, restoration, and promoting *in situ* conservation work. Another important component of the project is to develop husbandry protocols for best practice in maintaining the species *ex situ*, through specific research to evaluate natural requirements and assess influences on captive frogs.

Claes Andrén is the Scientific Director at Nordens Ark conservation center in Sweden, and in 2013 this foundation decided to support DNA research carried out by Salford University to describe microsatellite markers for the species, which was fundamental for establishing a stud-book for captive Costa Rican Lemur Frogs. This will also allow for the assessment of genetic diversity in wild populations. From 2014, Nordens Ark has also been a committed partner in the Lemur Leaf Frog conservation project in other ways, and will soon house an assurance *ex situ* population under strict quarantine conditions with defined blood-lines. Nordens Ark has prepared a special building for keeping exotic endangered amphibian assurance populations, and one part of the building is specially equipped for Lemur Leaf Frogs, including a rearing facility for the prey insects. Currently Nordens Ark has non-program frogs to help it align its procedures and personnel. In early 2015, selected individuals will arrive from Bristol Zoo which will constitute the real *ex situ* population. Claes recently visited Costa Rica to see for himself how the funding provided by his conservation-oriented institution can further support this species.

Andrew Gray is the Curator of Herpetology at Manchester Museum, at the University of Manchester, England. Andrew leads Project Lemur Frog and is dedicated to helping facilitate all aspects of the program. He collected the founder stock for the *ex situ* population established in captivity, and he has specialist scientific interest in Neotropical frogs of the sub-family Phyllomedusinae, to which the Lemur Leaf Frog belongs.

Tim Skelton is the Curator of Reptiles at Bristol Zoo and his skills in breeding the Lemur Leaf Frog in captivity have been crucial to Project Lemur Frog. Founder animals were provided to Bristol Zoo from Manchester Museum and the zoo has been highly supportive of the conservation of this particular species. Tim has developed husbandry guidelines for the species and he has developed a purpose-built high-tech facility that allows for the adjustment of specific temperature, humidity and day lengths to create the perfect conditions to encourage the frogs to breed.

Brian Kubicki is the founder of the Costa Rican Amphibian Research Center (CRARC), a private reserve created in an attempt to further biological and conservational efforts directed specifically at Costa Rican amphibians, including the Lemur Leaf Frog. He is a major collaborator and key person within Project Lemur Frog. The area where the CRARC is based is now one of the last known places the Lemur Leaf Frog is known to exist within Costa Rica. Brian is conducting research, biological surveys and conservation work such as reforestation, habitat creation, and habitat rehabilitation.



Maintenance of one of the *ex situ* populations of Lemur Leaf Frogs (*Agalychnis lemur*). An international collaboration between several committed individuals and institutions has been developed, aimed at conserving Lemur Leaf Frogs from Costa Rica. Photo: Manchester Museum.



Robert Jehle is a population biologist with an unexplainable fondness for amphibians, ponds and rainforests. He arrived at Salford University in 2008, and his main research area revolves around the ecology, behavior and evolution of amphibians at the population level. One of his main focuses covers the documentation of spatial and temporal population processes using genetic markers, and the use of genetic information to document mating systems. His involvement and support in the research aspects of Project Lemur Frog is crucial to its success.

For more information about the Lemur Leaf Frog and the Lemur Frog Project with videos, photos and facts, please see our web site, www.lemurfrog.org.

Claes Andrén, Scientific Director at Nordens Ark conservation center in Sweden, with a Lemur Leaf Frog. Photo: Manchester Museum.

First rescue center for threatened amphibians in Argentina

Federico Kacolaris and Jorge Williams, Museo de La Plata, Consejo Nacional de Investigaciones Científicas y Técnicas, Argentina

With support from an Amphibian Ark Seed Grant and with collaboration with the Museum of La Plata and Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET, National Scientific and Technical Research Council), we are creating the first rescue and captive breeding center for threatened amphibians in Argentina. This project seeks to establish captive breeding and survival colonies of highly threatened and endemic Argentinian species, including Calchaquies Valleys' Frog (*Telmatobius pisanoi*) and Valcheta's Frog (*Pleurodema somuncurensis*). Due to the restricted distribution of these species, their small population sizes and the deterioration of natural habitats for human use, we are also working on the development of *in situ* conservation activities to reduce the threats to their habitats. However the existence of external factors such as climate change and the presence of chytrid fungus in both species has prompted us to develop the captive rescue center as a concrete alternative that will ensure the long-term persistence of these species.



Assurance colonies for threatened Argentinian species including Valcheta's Frog (*Pleurodema somuncurensis*) will be managed in the new amphibian breeding center. Photo: Federico Kacolaris.

Our goal is to work in parallel with both *in situ* and *ex situ* conservation programs to achieve long-term viability of the wild populations. In this context, the rescue center will allow us to establish breeding groups of both the Calchaquies Valleys' Frog and the Valcheta's Frog as assurance colonies in the case of extreme weather events or emerging diseases. Furthermore, the development of management tasks aimed at reducing threats will allow us to recover suitable habitat for both species and so we can plan for future reintroduction projects to eventually stabilize the natural populations.

We have begun the construction and development of the rescue and breeding center, which is located within the herpetology lab at the Museum of La Plata (La Plata, Buenos Aires). So far we have assembled the shelves, built some aquariums and added water circulation systems for aerated and filtered water. The center is now ready to receive the first batch of frogs for quarantine and they will then be moved to their final terrariums. Parallel to these activities we have been collecting data about the water and microhabitat in which these species occur in the wild, so we can replicate these environmental parameters in the rescue center and offer an optimal environment that hopefully allows individuals to thrive.

The next step will be to collect and transport some breeding pairs to the rescue center and to start the quarantine process and treatment for chytrid. We will begin this stage with Valcheta's Frog as it is a species for which we have sufficient background information. After the process of quarantine and acclimation of these breeding pairs is complete, we will attempt to collect some Calchaquies Valleys' Frogs.

Since this is the first amphibian rescue center in Argentina, and we also aim to disseminate and promote these types of conservation projects throughout the country, we are developing an outreach program to publicize our goals. The outreach efforts are part of a recently-created program that was baptized with the name "Cururú" which combines various conservation projects, both *in situ* and *ex situ* (such as this project) which are focused on threatened amphibian species in Argentina.

We have much work ahead, but we are confident that the development and implementation of this rescue and breeding center will have a vital role in the conservation of both of the target species and will promote local initiatives for the protection of other species of threatened amphibians.

Some of the aquariums that have been established in Argentina's first rescue and captive breeding center for threatened amphibians. Photo: Federico Kacolaris.



Ex situ conservation program for the Ecuadorian Tiger Frog

Katty Garzón, Herpetological Fundación Gustavo Orcés and Diego Almeida Reinoso, SARgrillo Insect Breeding Farm, Quito, Ecuador

The Ecuadorian Tiger Frog (*Hyloscirtus tigrinus*) was first reported in Ecuador in 2012, where it was found in restricted cloud forests in northern Ecuador. Due to its limited range and accelerated destruction of its habitats, this species has been categorized as Endangered (EN) in the IUCN Red List. Captive conservation programs in Ecuador amphibians have not included this species in the past, so the Herpetological Fundación Gustavo Orcés decided to begin *ex situ* management of this species, in order to maintain a genetically viable population that would hopefully eventuate in a reintroduction program for the species.

Thanks to a seed grant of \$ 5,000 awarded by the Amphibian Ark in July 2013, the Center for Amphibian Conservation (CCA-FHGO) was created, and we began to assemble terrariums and the necessary equipment to handle the species: automatic water sprinkler system, drainage, lighting and UVB water filtration. At the same time, we conducted fieldwork, where exhaustive samplings were performed using the Survey for Visual Encounters methodology in sites around the town of Santa Barbara, where the species was first reported. During our surveys, larval stage individuals at different stages were reported, but adult individuals were not found. We presume that this species is affected by chytrid fungus and that most of the tadpoles, which are long-lived, were not developing to become adults. Since the tadpoles of this species are long-lived they are exposed to chytrid fungus for a prolonged period at their most sensitive development stage. Appropriate evidence of the presence or absence of the fungus during future surveys will give us answers to questions that arise in the way we are managing and understanding the development of this interesting species.

The captive Ecuadorian Tiger Frog program began with twenty-three tadpoles and one young individual, and our management program aims to breed the species and be able to maintain a genetically viable population under controlled conditions. Eventually we would like to begin a reintroduction program back to the species' natural habitat, however the main focus at this time is to stabilize an *ex situ* population. Little is known about this species so studying these animals in captivity will also allow us to compile biological information so we can better understand the species, and hopefully understand the causes of the decline of their populations in the wild. As the program progresses, we hope to significantly increase the captive population size, and we will continue our field work to collect additional founder animals for the captive management program.

Captive management

Since arriving at the CCA - FHGO, animals have been kept individually in an aquarium with constant oxygenation, and with an average water temperature of 16.5 °C. They were fed SAR type 1 (Super Food for Tadpoles) an exclusive formula for herbivores and detritus-feeding tadpoles - this diet contains 25.16% crude protein, 13.08% crude fiber, 49.08% total carbohydrates, 0.84% calcium and 0.034% phosphorus.



Terrariums at the Center for Amphibian Conservation, where the Ecuadorian Tiger Frog tadpoles were raised. Photo: Jorge Valencia.



The Ecuadorian Tiger Frog (*Hyloscirtus tigrinus*) was first reported in Ecuador in 2012, where it was found in restricted cloud forests in northern Ecuador.
Photo: Diego Almeida.

When the metamorphs emerged from the water they were transferred to a terrarium where they could continue to reabsorb their tails. Each terrarium has a false floor, water reservoir, sphagnum moss substrate, rocks, a small branch and a plant. The terrariums receive three sprays of water daily to provide sufficient humidity, and additionally we maintain a cooler in the room to lower the ambient temperature, accompanied by a fog effect during the afternoon and night.

The frogs are fed three times a week, and their diet includes a variety of insects produced at the SARgrillo farm: two species of crickets (*Gryllus assimilis* and *Acheta domestica*), (*Porcelio scaber*) "chanchitos" moisture and moths (*Galeria mellonella*). The crickets are dusted with calcium and vitamins A and D3 twice a week before being fed to the frogs.

Diet modification

After approximately eight months in captivity 40% of the animals began to show signs of chytridiomycosis, with excessive skin



A young metamorph Ecuadorian Tiger Frog at nine months of age, and measuring 410mm. Photo: Katty Garzón.

peeling being observed, and chytrid was later confirmed with PCR testing, which showed positive results for *Batrachochytrium dendrobatidis* (Bd) fungus. So we decided to immediately start treatment with Itraconazole solution 0.01%. To avoid too much stress on the frogs, a variation of the traditional treatment was used, which consisted of the frog baths being placed inside the terrariums, with a drip solution for five minutes for a period of ten days being performed. Thus we did not need to actually touch the animals at any time. The terrariums were sprayed with the solution of Itraconazole for the duration of the treatment. Any possible excess Itraconazole was removed each day with daily water spraying, and fortunately the animals have responded well to the treatment.

Conclusions

Being the only conservation effort for the Ecuadorian Tiger Frog in Ecuador at this time, we are keeping twenty developed individuals in good conditions. Although all of the tadpoles arrived at the same time and the tadpoles and frogs were kept in the same facilities, two of them were in the tadpole stage for longer than the others, reaching a larger size in comparison to the others. These two tadpoles have already reached their sub-adult phase and continue their growth.

It is important to mention that Ecuadorian Tiger Frog tadpoles are found in temperatures of less than 10 °C in their natural habitat, yet they have managed to adapt to higher temperature conditions (where the average room temperature is 16 °C) where they can stay until they have reached the subadult and juvenile phase.

We can say preliminarily that one of the threats to the species is the presence of Chytridiomycosis, as we confirmed the presence of chytrid by PCR testing when they were starting their sub-adult phase when they were in the final stages of reabsorbing their tails.

Individuals were only found in their larval stage, with no adults being reported during our comprehensive field work, and no vocalizations of adults indicating their presence. It is likely then that the early stages of the frog's development are when the species is more vulnerable to chytrid fungus, and they are not developing to the adult stage. This could be one reason why we were not able to find adult frogs.

We have observed that the treatment we administered to counteract the fungus in our frogs, despite having been administered in a slightly different way to the traditional treatment, has been successful, as the frogs have continued to progress well, and we have managed to minimize the stress on then individual animals.



Terrariums at the Center for Amphibian Conservation, where the Ecuadorian Tiger Frog tadpoles were raised. Photo: Katty Garzón.

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