# amphibian ark Keeping threatened amphibian species afloat

# **Newsletter**

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#### In this issue...

The first experience of reintroduction of the Critically Endangered Golden Mantella frog in Madagascar2
Resources on the AArk web site for amphibian program managers4
Conservation Needs Assessments in Colombia5
First release trials for Variable Harlequin Frogs in Panama6
New children's books by Amphibian Ark8
North America Salamander Biology, Husbandry and Conservation Training Course9
Conservation Needs Assessments for Malaysian amphibians
A European early warning system for a deadly salamander pathogen12
Recent animal husbandry documents on the AArk web site15
Amphibian Ark t-shirts, hoodies and sweatshirts
A future-proofing plan for Papua New Guinea frogs16
Amphibian Ark donors, January 2017 - March 201818





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### The first experience of reintroduction of the Critically Endangered Golden Mantella frog in Madagascar

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The Golden Mantella Frog (*Mantella aurantiaca*), is listed as Critically Endangered in the IUCN Red List (IUCN, 2017) and it has a restricted distribution range in the Moramanga region in eastern Madagascar. This species is most threatened by habitat loss. In 2015, new Protected Areas called The Mangabe-Ranomena-Sahasarotra and The Torotorofotsy wetland added some of their key habitat to their protection, but despite this, some other sites are threatened, especially by illicit gold-mining activities and due to the fact that several populations are situated within the Ambatovy mining site. These major concerns raised the need for a mitigation program. The Ambatovy Mining Company, the first extractor of Nickel and Cobalt in the Ocean Indian, has committed itself to zero extinction of biodiversity within the impacted sites, and the Golden Frog is one of the priority species for protection.

In addition, the widespread presence of the fungus Batra-chochytrium dendrobatidis, which has been detected in the Central Eastern part of Madagascar, challenges any *in situ* actions that prevent this iconic species from extinction. Recent research demonstrated that this fungus has the potential to become more virulent after intense habitat disturbance. As this might relate to the footprint of the Ambatovy site, one of the conservation decisions has focused on the captive breeding and reintroduction at receptor ponds to mitigate extinction vulnerability.

The establishment of the Frog Conservation Breeding Facility in nearby Andasibe was initiated by the IUCN SSC ASG Madagascar, the Malagasy Government, and the local association Mitsinjo, which is the responsible stakeholder for managing and maintaining an assurance population of the Golden Frog in captivity. It received an Amphibian Ark seed grant in 2009 and began operation in 2011.

Initially, 162 individual frogs were collected for the captive breeding and husbandry program in February 2012, from sites within the mine area. Back then, 99 males and 55 females were collected from two breeding ponds (known as MP7 and MP8), located approximately 50 meters from each other. An additional three males and five females were collected in April from a third site (MP5), approximately 160 meters southeast from the other two. They were first held in facilities near their natural area of occurrence and later transferred to a quarantine area at the Mitsinjo facility. As Golden Frogs adapt quite well to captive conditions, acclimatization was successful and soon the first fertile clutches were laid. This has continued since then and every breeding season (roughly from October to February) new progeny was produced by the different breeding groups. In the meantime this species was successfully bred into the F2 generation and extra capacity in the breeding facility was created to house surplus individuals that were planned for release.



Wild caught founder Golden Mantella (*Mantella aurantiaca*) at the age of 5+ years. Photo: Sebastian Wolf.

For the first step of the reintroduction program, a reintroduction protocol was developed by a committee composed by the representatives of the IUCN SSC ASG Madagascar, Malagasy Government officers from the Ministry of Environment, Ecology and Forests, and national associations and universities, using the IUCN Guidelines on the Use of Ex situ Management for Species Conservation (IUCN SSC 2014), IUCN Guidelines for Reintroductions and Other Conservation Translocations (IUCN SSC, 2013), Amphibian Ecology and Conservation, A Handbook of Techniques (Dodd, 2010) and some other publications (Mendelson & Altig, 2016, Polasik et al. 2016).

Then, four release sites where the species occurred historically were selected, monitored and restored mainly by the plantation of fern species *Osmunda regalis* which is a characteristic of ponds occupied by Golden Mantellas. Those sites are fully protected as they are included in two conservation zones managed by the Ambatovy mining company and the local community based associations.

Meantime, about twenty-eight clutches from the first founders were produced in the breeding facility, resulting in 1,522 individuals including 238 adults, 649 froglets, 196 metamorphosed frogs and 439 tadpoles. The frog and froglets were identified using different Visual Implant Elastomer (VIE) tags.



Juvenile Golden Mantella before color change Photo: Sebastian Wolf.

Mesh field enclosure for metamorphosing tadpoles at one of the release sites, in two conservation zones managed by the Ambatovy mining company and the local community-based associations. Photo: Association Mitsinjo.

In April 2017, we transported the different life stages from the breeding facility to the release sites. Some tadpoles were lost during the transportation stage, although the rates were low, with mortality rates of 0.22% for tadpoles and 1.01% for metamorphosed tadpoles. A soft-release method using mesh field enclosures was used for releasing adults, froglets and some of the metamorphosed tadpoles. Tadpoles at earlier life stages were released using a hard-release method. All crucial environmental parameters were collected in situ and ex situ to allow for the acclimation of the frogs during transport and release.

An MSc student from the University of Mahajanga monitored the tadpoles after release, and a PhD student from the University of Antananarivo was in charge of the post release monitoring of the adults and froglets. The surveys that followed the first phase of the release already look promising; the captive-bred individuals have adapted well to their new home. The complete release program will last for two years.

This release of a threatened and endemic amphibian species, which has been successfully bred in captivity, also generated interest among journalists and media in Madagascar who took part in the release procedure.

Stakeholders involved include the General Directorate of Forests (Direction Générale des Forêts), the IUCN SSC ASG Madagascar, the Biodiversity team of Ambatovy Minerals SA, The Association Mitsinjo, Madagasikara Voakajy, and the Universities of Antananarivo and Mahajanga.



# Resources on the AArk web site for amphibian program managers

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, <a href="https://www.amphibianark.org/amphibian-husbandry/">www.amphibianark.org/amphibian-husbandry/</a>. 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 150 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 share their husbandry documents, protocols and management plans. If you would like any documents added to this page, please either send them to webmaster@amphibianark.org or email us a link to an online version of the document, 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 the assessment of the conservation needs of 2,957 (36%) of the world's amphibian species through twenty-nine national or regional workshops. The results of these assessments are available on the Conservation Needs Assessment web site, www.ConservationNeeds.org. 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 (see the Species Recommended for Ex Situ Rescue report, www.conservation-needs.org/SpeciesRecommendRescue.aspx) or for ex situ research programs (see the National Recommended Conservation Actions report, www.conservationneeds.org/NationalAssessmentReport.aspx).

Information regarding the likely availability of founder animals and the completion of a relevant taxonomic study is included in the reports 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.

There is also a list of existing rescue and *ex situ* research programs for threatened amphibian species at <a href="https://www.amphibianark.org/progress-of-programs/">www.amphibianark.org/progress-of-programs/</a>.

#### **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 utilized by any institution 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. Suggestions for finding additional resources or advice are made throughout the tool.

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 the number of founder animals

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 whenever possible, 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.

#### **Expert advice**

Many experts in various aspects of managing captive conservation programs are available to offer advice if needed. We have compiled a list of people around the world who are willing to offer advice on subjects such as captive husbandry, habitat restoration, field techniques, reintroduction and population management. Some of these experts can offer advice in English, French, Spanish or Chinese. The list of experts, and the subjects they have expertise in can be found on the AArk web site, (www.amphibianark.org/amphibian-husbandry-experts/).

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.

#### **Conservation Needs Assessments in Colombia**

#### Luis Carrillo, Training Officer, Amphibian Ark

With more than 800 amphibian species, Colombia is considered the second most biodiverse country for amphibian species.

In order to help plan a comprehensive strategy for the conservation of amphibians in Colombia, and taking into account the National Plan for the Conservation of Amphibians (which brought together a significant number of researchers and professionals from NGOs, governmental and academic institutions), a strategic alliance between the Wildlife Conservation Society (USA), Zurich Zoo (Germany), Cali Zoo (Colombia) and several others interested in amphibian conservation was formed to implement the priority actions identified by the plan. In particular this alliance is focusing on complementary conservation actions, both *in situ* and *ex situ*, in priority areas which have a high concentration of threatened amphibians in Colombia.

This alliance is leading the initiative to ensure the conservation of threatened species of amphibians in five national parks (Parque Natural Nacional Selva de Florencia, Parque Natural Nacional Tatamá, Parque Natural Nacional Farallones de Cali, Parque Natural Nacional Munchique and Parque Natural Nacional Sierra Nevada de Santa Marta), which include more than thirty percent of the threatened amphibian species of the country.

Through the coordination of Amphibian Ark, the use of the Conservation Needs Assessment process was proposed to identify the necessary conservation actions for more than 200 species that are distributed in the National Natural Parks system. Using the Conservation Needs Assessment tool (<a href="https://www.Conservation-Needs.org">www.Conservation-Needs.org</a>) a group of experts recommended conservation actions which will now inform conservation strategies for those species.

A small group of field biologists and researchers gathered together at Cali Zoo from March 22nd – 24th to complete assessments for almost 120 species. As result of the assessments, seven species were recommended for *Ex Situ* Rescue, fifty-one species were recommended for *In Situ* Conservation, eighty-six species were recommended for further *In Situ* Research, forty-two species were recommended for *Ex Situ* Research, two species were recommended for Mass Production in Captivity, twenty-nine species were recommended for Conservation Education, seven species were recommended for Supplementation, and seven species were recommended for Biobanking. Definitions for all of these types of conservation actions can be found at <a href="https://www.conservationneeds.org/Help/EN/ConservationActions.htm">www.conservationneeds.org/Help/EN/ConservationActions.htm</a>.

The participants of the workshop also selected two species to develop a holistic conservation program (including both *in situ* 

and ex situ management). One of these, Lehmann's Poison Frog (Oophaga lehmanni), is a species that is currently under pressure from over-collection due to illegal trading for the pet trade, and has been recommended for mass production, supplementation, conservation education and in situ conservation strategies. The Universidad del Valle, in Cali, Colombia, along with the Corporación del Valle del Cauca (the regional government branch in charge of conservation and management of biodiversity in the Department of Cauca) are currently studying the population dynamics for this species, and the Universidad de Los Andes in Bogotá, Colombia, is currently studying the genetic variability of different sub-populations of the species, trying to understand the effect of population decrease on the genetics of the species in the wild due to over-collection. Cali Zoo will be the institution in charge of breeding the species in captivity in a biosecure area so individuals from this assurance population could be translocated to the wild when needed.

The group also identified *Atelopus laetissimus* as a husbandry analog species for mid-altitude (1,900–2,800m above sea level) species from the *Atelopus* genus, such as the Critically Endangered Bogota Stubfoot Toad (*Atelopus subornatus*). For this species the Parque Explora, located in Medellín, Colombia, will develop management and breeding protocols in a biosecure area. Parque Explora already has some experience keeping mid-altitude *Atelopus* species in their facilities.

Finally the group also identified *Colomascitus* (*Hyloscirtus*) *antioquia* as an *ex situ* Rescue species, and Parque Explora will develop a holistic conservation plan for this species. This species was only recently described (in 2013) and is found north of the Cordillera Central of the Departmento de Antioquia, Colombia, where it has been identified in eight localities ranging from 2,500–3,200m above sea level (Rivera-Correa and Faivovich 2013). The population is considered to be severely fragmented, given that most of the population occurs in small creeks, isolated from one another and with no exchange of individuals (Nature-Serve Workshop August 2016); however the population trend is currently considered to be stable.

Amphibian Ark will follow the development of these actions plans and will provide help whenever required.



# First release trials for Variable Harlequin Frogs in Panama

Brian Gratwicke, Panama Amphibian Rescue and Conservation

Once common along highland streams in western Costa Rica and Panama, the Variable Harlequin Frog (*Atelopus varius*) is endangered throughout its range, decimated by a disease caused by the amphibian chytrid fungus. On January 17, researchers from the Smithsonian Conservation Biology Institute released approximately 500 frogs at First Quantum Minerals' concession site in Panama's Colon province as a first step toward full-scale reintroduction of this species.

The Variable Harlequin Frog takes its name from the variety of neon colors - green, yellow, orange or pink - juxtaposed with black on its skin. In order to monitor the released frogs over time, thirty are wearing miniature radio transmitters. The scientific team

for release into the wild in Panama. Photo: Brian Gratwicke.

also gave each frog an elastomer toe marking that glows under ultraviolet light to mark individuals as part of a population monitoring study.

"Before we reintroduce frogs into remote areas, we need to learn how they fare in the wild and what we need to do to increase their chances of survival in places where we can monitor them closely," said Brian Gratwicke, international coordinator of the Panama Amphibian Rescue and Conservation project (PARC) at the Smithsonian Conservation Biology Institute. "Release trials may or may not succeed but the lessons we learn will help us to understand the challenges faced by a frog as it transitions from captivity into the wild."

Variable Harlequin Frogs are especially sensitive to the amphibian chytrid fungus, which has pushed frog species to the brink of extinction in Central America. PARC brought a number of individuals into the breeding center between 2013 and 2016 as chytrid continued to impact wild populations.

"The Variable Harlequin Frog is one of the closest relatives of Panama's iconic Golden Frog (*Atelopus zeteki*), another target species in our captive breeding program," said Robert Ibáñez,

The field release team (left to right): Blake Klocke (GMU-SCBI),
Heidi Ross (STRI), Jorge Guerrel (STRI), Orlando Ariel
(STRI), Eric Klaphake (Cheyenne Mountain Zoo),
Elliott Lassiter (SCBI), Brian Gratwicke
(SCBI) and Roberto Ibanez (STRI).
Photo: Brian Gratwicke.

Number 42, March 2018



Above: Newly metamorphosed Variable Harlequin Froglets bred in captivity.

Below: Heidi Ross, Director of the El Valle Amphibian Conservation Center facility at the Nispero Zoo in Panama, with baby Variable Harlequin Frogs, that were used in the release trial. Photos: Brian Gratwicke.

PARC project director at the Smithsonian Tropical Research Institute in Panama. "We'll be monitoring the surrounding amphibian community and the climate at this site, and comparing this to the amphibian community at another control site. This kind of intensive monitoring will help us to understand disease dynamics in relation to the release trials".

PARC members hope to secure the future for this and other endangered amphibians by reintroducing animals bred in captivity according to an action plan developed with Panama's Ministry of the Environment and other stakeholders.

"It took us several years to master how to successfully breed these frogs in captivity," said Ibáñez. "As the number of individuals we have continues to increase, it provides new research opportunities to understand factors influencing survival that will ultimately inform long-term reintroduction strategies."

The release trial is made possible with the support of First Quantum Minerals, National Geographic Society, Mohammed bin Zayed Species Conservation Fund, the Smithsonian Women's Committee and the WoodTiger Fund. PARC is a partnership between the Cheyenne Mountain Zoo, the Houston Zoo, the Smithsonian Tropical Research Institute (STRI), the Smithsonian Conservation Biology Institute (SCBI) and Zoo New England. It has two facilities in Panama: the Gamboa Amphibian Rescue and Conservation Center at STRI and the El Valle Amphibian Conservation Center at El Níspero Zoo. Combined, these facilities have a full-time staff caring for a collection of twelve endangered species.

A recently-released female Variable Harlequin Frog exploring her new home in the wild. Photo: Brian Gratwicke.

#### New children's books from Amphibian Ark

The Unite for Literacy team works with its partners to develop a wide range of free, online children's books, narrated in multiple languages, to celebrate language, culture and a love of reading. These short books feature wonderful images, and short pieces of text on each page, and along with the narration, help children and their families who are learning to speak English, or other languages.

Amphibian Ark's Community Education Officer, Rachel Rommel-Crump, developed the text for five new books about amphibians, which are beautifully illustrated with photos from our photo competition, and others which have been generously supplied by some of our partners. The first five books can be found on AArk's "bookshelf" on the Unite for Literacy web site, www.uniteforliteracy.com/aark/arkbooks. The new books are:

**Amazing Amphibians** – Introduces these amazing animals, providing some basic biological information, and encouraging readers to "Bring a map as you explore your neighbourhood and mark the places where amphibians might live."

**Fantastic Frogs** – Shares information about frogs from different countries, and different breeding strategies, suggesting that you can "Visit a pond and look for tadpoles swimming close to the edge, or frogs sitting in the water."

**Super Salamanders** – Explains where salamanders live and how they can regrow damaged limbs and recommending that you "Learn the name of a salamander or frog that lives near you."





**Secret Caecilians** – Introduces these lesser-known amphibians and suggests that children can "Lie on your stomach, put your arms against your body, and try to move like a caecilian."

Amphibian Heroes – Talks about the threats facing amphibians, and how amphibian heroes in Madagascar, Argentina, Japan and the US are helping to save them. It also recommends that you "Join an effort to protect wildlife habitat in your community, or start one with your friends."

These books include English narration, and Spanish will be available soon. We hope you enjoy them!

Special thanks to Amphibian Ark's Anne Baker, Luis Carrillo and Kevin Johnson who provided creative and editorial support throughout the project. Thanks to Paul Crump and Joe Mendelson for reviewing the final books and providing helpful comments. Also, thanks to Andy Gluesenkamp and Tariq Stark for confirming factual information about some specific species.

We would not have had such engaging photographs to share with children if it wasn't for the generosity of Todd Pierson, Dave Huth, Candace Hansen-Hendrikx, Arturo Muñoz, Ben Tapley, Paul Crump, Brian Gratwicke, Norhayati Ahmad, Federico Kacoliris, Melina Velasco, Aiko Taguchi, Devin Edmonds, Cassidy Johnson, Wikicommons contributors, and the AArk calendar contest photo entry participants. Finally, thanks to Unite for Literacy staff member Holly Hartman for her expertise, and who made creating the books such a fun and easy process.



Hopping over to Japan, heroes like Yuki and friends work to save the Japanese Giant Salamander. These are among the largest amphibians in the world. They need clear, cold, fast-flowing rivers with large rocks to hide under.

# North America Salamander Biology, Husbandry and Conservation Training Course

Luis Carrillo, Training Officer, Amphibian Ark

The appearance of the salamander chytrid fungus (*Batrachochytrium salamandrivorans*) has raised concerns among the amphibian conservation community. Discovered recently, it is potentially an important threat to salamanders and newts in Europe and America, having already caused significant declines in the wild population of Fire Salamanders (*Salamandra salamandra terrestris*) in the Netherlands.

In the North-American sub-continent (Mexico, United States and Canada) there are 304 salamander species which represents 54% of all the salamander species of the world. Due to the potential threat this new disease has for the salamander species in this sub-continent, the Amphibian Ark is looking forward to building capacity among *ex situ* amphibian conservation community professionals, so both the people and institutions will be better prepared to create assurance colonies if and when needed.

From September 18th – 22nd, 2017 Amphibian Ark and Zoo Atlanta, USA, organized and delivered the second North America Salamander Biology, Husbandry and Conservation Training Course. Ten participants representing nine institutions (zoos, universities and research institutions) from the US, Canada and Belgium were present at the course.

The course consisted of a mix of lectures, group activities, practices, and field trips designed to deliver the most relevant information about salamander husbandry and also to put into practice what the students learned during the lectures/group work sessions.



Carolina Spring Salamander (*Gyrinophilus* porphyriticus dunni), a common species around Wharton Center, Georgia.

Photo: Luis Carrillo.

The course content was designed to:

- inform students about the critical situation of many salamanders species in the US
- deliver updated salamander husbandry and breeding methods and techniques
- inform students about infectious diseases and biosecurity in captive assurance colonies
- provide the students with information and skills to successfully breed and maintain different species of North American salamanders
- encourage the participation and sharing of knowledge and expertise among instructors and students, providing spaces to do so in a comfortable environment of camaraderie.

A survey was sent to all the students as a way to evaluate the effectiveness of the course in an indirect way. One hundred percent of the participants said that the information and knowledge acquired during the course was very good to excellent; 100% of the participants said that the course met their expectations; and 100% said that they took advantage of the experiences of their classmates and instructors.

Ten participants and faculty from zoos, universities and research institutions in the US, Canada and Belgium attended the salamander husbandry and conservation course in Atlanta in September 2017. Photo: Luis Carrillo.



Renowned *in situ* and *ex situ* salamander biologists and veterinarians comprised the faculty. Ninety-five percent of the participants said the course faculty was good to very good.

Quotes from some of the students:

"The course was very in depth and well planned out. The instructors, leaders, and fellow classmates were amazing and I loved networking with everyone! Great class! Well worth it!"

"I came to this course with some background knowledge of salamanders and walked away with more information than I could imagine. Not only was this course useful but I also gained new friends who I can count on to help further my knowledge of salamander husbandry, behavior, and conservation."

"The North American Salamander Biology, Husbandry and Conservation Training Course went above and beyond my expectations. Hearing about first-hand experiences from the true experts in the field was extremely helpful and informative, and restored my hope in salamander conservation for the future. The course was well-organized although laid-back, and all of the instructors were knowledgeable and approachable. I would recommend this course for anybody working with any or all Caudata species in any way, shape, or form!"



Students from the 2017 North American Salamander Biology, Management and Conservation Training Course looking for salamanders in a stream transect during the field practice at Wharton Center. Photo: Luis Carrillo.

A Southern Gray-cheeked Salamander (*Plethodon shermani*) spotted at Wharton Center during the 2017 North American Salamander Biology, Management and Conservation Training Course's night field trip. Photo: Luis Carrillo.



#### **Conservation Needs Assessments for Malaysian amphibians**

#### Kevin Johnson, Taxon Officer, Amphibian Ark

Malaysia is home to 265 species of amphibians, with 144 of these being endemic to the country, 46 species previously listed as threatened (three Critically Endangered, ten Endangered and thirty-three Vulnerable), thirty species listed as Data Deficient and sixty-one species not previously assessed for the Red List. In 2012, 169 Malaysian amphibian species were assessed for their conservation needs, however these assessments were made using an early version of the assessment process, which was not as reliable as the current version, and were in need of re-assessment

In January this year, seven Malaysian amphibian experts met with Programme Officers from the IUCN Amphibian Red List Authority and Amphibian Ark, to undertake Red List Assessments and Conservation Needs Assessments for these species. This joint assessment workshop was a great opportunity for both organizations to see each other's processes, as well as the outcomes from each set of assessments, and to make the best use of resources, and of course, the participants' time. Red List assessments (www.iucnredlist.org) determine the relative risk of extinction, highlighting those species that are facing a high risk of extinction, while the Conservation Needs Assessments (www.conservationneeds.org) produce prioritized recommendations for a range of conservation actions, which will hopefully help to prevent further extinctions.

The workshop was hosted by the School of Biological Sciences, Universiti Sains Malaysia, in Penang, and was funded by Rainforest Trust and Amphibian Ark. We worked in two groups, split geographically, with one group focussing on species from Peninsular Malaysia, and the other group working on species from Malaysian Borneo. The focal species were those which had either not previously been Red Listed, species whose Red List assessments were quite out of date, or those where significant changes had occurred in the wild populations since the last Red List Assessment was made.

This joint approach to assessments was very successful, and the

participants were happy to see recommendations being made for future conservation actions. Some of the assessments require additional input from experts who were not present during the workshop, and this will hopefully be completed within the next couple of months. The Red List Assessments will be reviewed within the next few months, with the final Red List category being determined, and the assessment information being published online. The Conservation Needs Assessments will also be reviewed, and once approved, will also be available online.

Collectively, we managed to complete 126 Conservation Needs Assessments, with another fourty-one assessments which require some additional input from other experts currently being reviewed and updated. Since the meeting, we have reviewed all the assessments, corrected any grammatical issues and spelling mistakes. The 126 completed assessments are now available on the Conservation Needs Assessment web site (www.ConservationNeeds.org), and the assessment data is also be available in all reports on the site.

Of the 126 assessments completed so far, four threatened species were recommended for urgent ex situ rescue (Leptolalax kecil, Ingerophrynus kumquat, Ansonia smeagol and Abavorana nazgul). Three of these species have never been held in captivity before and more common, but related analog species have been recommended that can potentially be used to develop husbandry and breeding protocols prior to the threatened species being brought into captivity. AArk staff will work with our colleagues in Malaysia to investigate potential captive programs for these species, however additional field work is required to determine the suitability of establishing rescue programs for these species.

We hope that the Amphibian Red List Authority and AArk staff have the opportunity to follow this joint approach to assessments again in the future, and both groups are currently reviewing their priority countries and regions to determine potential overlap with future assessments.



### A European early warning system for a deadly salamander pathogen

Tariq Stark, An Martel, Frank Pasmans, Valarie Thomas, Maarten Gilbert, Annemarieke Spitzen

#### Introduction

The chytrid fungus Batrachochytrium salamandrivorans (Bsal) was described in 2013 after the rapid decline of a Fire Salamander (Salamandra salamandra) population in the south of the Netherlands (Spitzen et al., 2013; Martel et al., 2013). This population experienced a 99.9% decline over a seven-year period with no signs of recovery. Soon after the description of Bsal more outbreak sites with massive population crashes were discovered in Belgium, Germany and in additional sites in the Netherlands (Spitzen et al., 2016; Dalbeck et al., 2018). In most cases Fire Salamander populations were affected but Alpine Newts (Ichthyosaura alpestris) and Smooth Newts (Lissotriton vulgaris) also suffered mortalities (Spitzen et al., 2016; Stegen et al., 2017). In 2017 Bsal was also detected on Great Crested Newts (Triturus cristatus) and Palmate Newts (Lissotriton helveticus) in Germany (Dalbeck et al., 2018) but without obvious clinical signs. Bsal is deadly to nearly all European urodelans (newts and salamanders) and poses a massive threat to European urodelan diversity (Martel et al., 2014).

Bsal originated in Asia and likely arrived in Europe via the trade in Asian salamanders and certain Asian anurans (frogs and toads) (Martel et al 2014; Laking et al., 2017; Ngyuen et al., 2017; Yuan et al., 2018). From these vectors it is believed Bsal spilled over to European salamanders and newts, which are naive to the pathogen, and most die soon after being infected. Recently Stegen et al (2017) found that Alpine Newts with low Bsal infection intensity

may persist with the fungus, and even clear the infection. This means that these newts can vector the fungus, as can some anuran species. Bsal has also been detected in populations of captive urodelans in Germany, the Netherlands, Spain and the United Kingdom (Cunningham et al., 2015; Sabino-Pinto et al., 2015; Fitzpatrick et al., 2016; Actieplan – België, 2017).

Legislation has anticipated halting the spread of the fungus via trade. The USA banned the interstate transport and import of salamanders and newts in 2016, Switzerland has banned the import, as has Hungary, and the European Union has recently announced animal health protection measures for intra-Union trade in salamanders in order to prevent further introduction of Bsal (http://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX: 32018D0320&from=NL).

The current knowledge of the distribution of Bsal outside of its native range is incomplete. It might be present in more European countries than currently known. Early detection of Bsal induced population declines is therefore very important. Urodelans with small ranges and/or small population sizes are especially at risk. In order to stem the tide of amphibian population declines and prevent mass extinctions, the European Commission issued the tender ENV.B.3/SER/2016/0028, Mitigating a new infectious disease in salamanders to counteract the loss of European biodiversity.

## BsalEurope Mitigating Batrachochytrium salamandrivorans in Europe Home What is Bsal? Report cases Bsal Diagnostic Centres Public Awareness Material Early Warning System Publications Contact Salamanders and newts at risk Tweets by @BsalEurope be put in place. Besides preventing a further entry of this amphibian pathogen into Europe, these measures Annemarieke should aim at minimizing the impact of Bsal on European urodelan species. Any Bsal abatement plan requires a clear overview of the current extent of the Bsal distribution in Europe. Therefore, the first and @chytrid EU legislation for intra-Union trade in salamanders and the introduction into the Union of such animals in relation to the fungus Bsal goo.gl/3s5GAA second objectives of this contract are to delineate the current *Bsal* range in Europe and to establish an early warning system that should allow rapid detection of novel *Bsal* outbreaks. Efficient mitigation requires apt measures both in the short and the longer term. Therefore, the third and fourth objectives of this study are to develop an emergency action plan for the short term and to provide proof of concepts for sustainable long term mitigation measures. In order to stem the tide of amphibian population declines and prevent mass COMMINION DEFINION DECISION (IL) REQUID of 29 reference 2019 on comin mixed both procession ensures for insure trains made in administrate and the introduction into the Union of such animals or relates to the forges Returbably vision internal commission of the Com extinctions, the European Commission(EC) issued the tender ENV.B.3/SER/2016/0028 (Mitigating a new infectious disease in salamanders to counteract the loss of Europ Having regard to Control Distraction 90(31/SEC of 26 June 1990 concerning varietiesy and assemblesical checks applicable to intro-Community trade in contact live entirals and products with a steer to the completion of the instead creditor (5), and to previolate Perfect (10) (about).

#### **General objectives**

Bsal has devastating effects on European urodelans and measures to protect them from these are of the utmost importance. The general objectives of this project can be summarized as followed:

- 1. Delineate the current range of Bsal in Europe
- 2. Create an Early Warning System which allows the rapid detection of novel Bsal outbreaks
- 3. Development of an emergency action plan (short term)
- 4. Provide proof of concepts for sustainable long-term mitigation measures.

This article focusses mostly on the first and second objective. More on the third (www.bsaleurope.com/emergency-actionplans/) and fourth (www.bsaleurope.com/ mitigation-and-reporting/) objectives can be found on www.BsalEurope.com.

### **Early Warning System**

#### Website

Hering regard no Council Distraction 91/496/EEC of 13 July 1993 Juping down the principles governing the experiments of vestimary checks on animals emering the Community from third countries and amending Distraction 85(645/EEC 09475/EEC 09476-1951) and no particular heavier 1980 (1980).

□

Annemarieke

We created an online platform www. BsalEurope.com (Figure 1) in order to educate the European public about Bsal, with the aim of detecting novel Bsal outbreaks throughout Europe. This website replaces the previous website (https://bsalinfoeurope.wixsite.com/eubsalmitigation2017). The

Figure 1. Home page of the BsalEurope web site (www.BsalEurope.com).

Universität Trier

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BsalEurope web site provides general information on *Bsal* (www.bsaleurope.com/b-salamandrivorans/), from pathogen characteristics, clinical signs and hosts, to the European distribution of *Bsal*, options for treatment and prevention. The map of the current known distribution of *Bsal* in Europe will be kept up to date. In addition, several public awareness materials (www.bsaleurope.com/public-awereness-material/) were created which aid in the early detection of *Bsal* outbreaks. Reporting dead and moribund animals is very important, therefore regional hotlines (www.bsaleurope.com/report-cases/) and *Bsal* diagnostic centres (www./bsaleurope.com/laboratories/) are listed on BsalEurope. Comprehensive lists of scientific and popular scientific papers can also be found on the website (www.bsaleurope.com/sample-page/) in addition to more helpful resources. BsalEurope will be updated on a regular basis, so be sure to bookmark it!

#### **Public awareness materials**

On BsalEurope several public awareness videos and documents can be found that were specifically created for this project: three videos and several informative leaflets. The first of the animated videos (www.youtube.com/watch?v=kss8B7V zAA&t=1s) shows where Bsal originates from, outlines its effect on European urodelans and where you can report dead and moribund animals. The second animation (www.youtube.com/watch?v=-WgYZME-GA9Y) outlines best practices for captive populations: biosecurity measures, quarantining of newly acquired specimens and a call to report dead and moribund animals (figure 2). The third video (www.youtube. com/watch?v=i-WJtmPdkng) stresses the importance of adherence to a field hygiene protocol in order to reduce the risk of human-mediated pathogen dispersal. Both animated clips have subtitles in sixteen

European languages in order to reach a large (European) audience. The clips can be found on the Reptile, Amphibian and Fish Conservation Netherlands YouTube channel (www.youtube.com/channel/UCdzWX2q4OhU0KGx\_kRcixig). The other public awareness materials are three easy-to-use leaflets on recognition of *Bsal* in urodelans (www.bsaleurope.com/recognize-sick-animals/, including FAQ's for fieldwork and captive collections), disinfection protocols for fieldwork (www.sossalamander.nl/LinkClick.aspx?fileticket=wilFeby6by8%3d&portalid=17&language=nl-NL) and heavy machinery (www.sossalamander.nl/LinkClick.aspx?fileticket=FAncbfR0tck%3d&portalid=17&language=nl-NL, figure 3).

#### **Report Cases**

In order to detect *Bsal* outbreaks and hence delineate the current range of the pathogen in Europe an Early Warning System is of the essence. If you have found a salamander or newt that is not the obvious victim of traffic or predation, then please report it to your nearest regional hotline (www.bsaleurope.com/report-cases/). Eight hotlines have now been established in eight European Union countries. The network of diagnostics centres (www.bsaleurope.com/laboratories/) in the European Union currently consists of fourteen laboratories in eleven countries. Ghent University centralizes all data on *Bsal* outbreaks and monitoring. Additionally, a European network of stakeholders that will report urodelan population declines is being developed.

If you notice suspicious deaths or sick animals, please take the following steps:

- · Do not handle sick or dead amphibians with your bare hands
- Use your mobile phone or other device to take multiple photos of the animal (from all sides)
- Be sure to include photos of any obvious lesions which you notice on its body
- Make a note of the location, date, time, and the number of animals which are sick or dead
- In Europe it is prohibited to collect wildlife (alive or dead) from the environment, therefore please contact the relevant hotline as soon as possible for further action.



In Europe 38 species of newts and salamanders are susceptible to fatal infection with this fungus. We might completely lose these species!

Figure 2. Screenshot of one of the animated videos.

#### **Monitoring**

The detection of disease-induced population declines relies heavily on long-term monitoring schemes. Several European NGO's developed programs where individual amphibian species, populations and communities are followed in long-term monitoring schemes. Long-term studies are extremely valuable to assess population trends and can act as an Early Warning System when populations are declining (especially rapid declines). Sharp declines can be a sign of the involvement of a pathogen, like *Bsal*, and follow-up research can quickly be deployed.

Are you interested in participating in such a program? Please contact one of the regional hotlines.

#### Contact and follow us

An Early Warning System only works when many organisations, professionals and volunteers work together. Questions about this project can be directed to Prof. Dr. An Martel of Ghent University via email (An.Martel@ugent.be) or via the contact form on the web site (www.bsaleurope.com/contact/). The project also has its own Facebook Page (@Bsalamandrivorans) and Twitter account (@BsalEurope). Be sure to follow us!

#### **Partners**

This project is a collaboration of Ghent University (Belgium: Flanders), Natagora (Belgium Walloon), The Spanish National Research Council (Spain), Reptile, Amphibian and Fish Conservation Netherlands (the Netherlands), Centre d'Ecologie Fonctionnelle et Evolutive (France), Genoa University (Italy), Trier University (Germany), and Zoological Society of London (United Kingdom). This tender (ENV.B.3/SER/2016/0028 Mitigating a new infectious disease in salamanders to counteract the loss of European biodiversity) was issued by the European Commission.

Figure 3: Example of one of the leaflets on hygiene protocols.

# Disinfection protocol fieldwork

















# **Background**

This document provides simple but effective measures that can help limit the spread of fungi and viruses pathogenic to amphibians in disease free areas. The advice listed below only encompasses "standard" field research methods. In case of reintroductions, translocation of animals, etc. stricter hygienic requirements are in order.

Many emerging infectious diseases, among which the chytrid fungi Batrachochytrium salamandrivorans and B. dendrobatidis, but also ranavirosis, currently pose a significant threat to amphibians in Europe. Anthropogenic spread of pathogens has been identified as a considerable threat to amphibian health. We encourage all biologists, researchers and volunteers to disinfect their field material.

This way, we can reduce the spread and 'buy' time while both field- and laboratory trials are run in order to counter/mitigate the effects of these disease agents.

# **Advice**

- Only handle amphibians when absolutely necessary. There are no limitations in the field as long as precautionary measures are taken in account.
- Also take precautionary measures in account when you work with freshwater fish, aquatic invertebrates or aquatic plants.
- Always return amphibians to the exact location<sup>1</sup> where they were caught.
- When handling amphibians one needs to wear disposable (powderless) gloves. Nitril gloves are recommended. Nonperfumed hand sanitizer (which contains ethanol) is also effective for disinfecting your hands afterwards.
- All materials used on a location<sup>1</sup> need to be disinfected before using them at another site.
- Boots and wading suits that have been in direct contact with water or muddy soil need to be disinfected thoroughly.
- Park your vehicle preferably on paved road and not in soft, muddy soil or vegetation.
- Dead and sick amphibians can pose a high ecological risk. Only handle them with disposable gloves, report them to the proper authorities and if possible - and legally allowed to - take them with you (dead animals). Transport dead animals in two plastic bags in order to prevent leakage. Report dead and sick salamanders directly to your research institute.



Help to prevent mass mortality.

#### Recent animal husbandry documents on the AArk web site

The Husbandry Document library on the AArk web site (<a href="www.amphibianark.org/husbandry-documents">www.amphibianark.org/husbandry-documents</a>/) currently contains over 150 articles with additional articles being added regularly. Four new documents have been added recently:

#### **Pool Frog Species Action Plan (English)**

This document outlines the actions required to conserve the Pool Frog (*Pelophylax* (*Rana*) *lessonae*) in Britain and provides a framework for all conservation work. More detail of each action is provided in the implementation guide. Each action has been assigned the appropriate level for implementation i.e. United Kingdom, country, region or local. Key actions have been identified through a 'signposting' exercise; these statements are shown in two shades of green and the corresponding signposted actions are marked with a similarly coloured signpost symbol.

Author: Amphibian and Reptile Conservation Trust

Publication: July 2009

www.amphibianark.org/pdf/Pool-Frog-Species-Action-Plan.pdf

#### Batrachochytrium salamandrivorans (Bsal) (English)

This leaflet can be used to recognize *Bsal* in the amphibian host. Important: the symptoms are variable and can be difficult to detect at an early infection stage. It is often that lesions become evident at a relatively late stage of infection with *Bsal*.

Publication: February 2018

www.amphibianark.org/pdf/Bsal-Flyer-DEF.pdf

#### Disinfection protocol fieldwork (English)

This document provides simple but effective measures that can help limit the spread of fungi and viruses pathogenic to amphibians in disease free areas. The advice listed below only encompasses "standard" field research methods. In case of reintroductions, translocation of animals, etc. stricter hygienic requirements are in order.

Publication: February 2018

www.amphibianark.org/pdf/Disinfection-protocol-fieldwork-DEF.

#### Disinfection protocol heavy machinery (English)

This document provides simple, but effective measures in order to minimize the spill over of infectious amphibian pathogens to disease free areas. The advice listed is meant for heavy machinery that is used for work in and around water bodies like ponds, canals and streams that are home to amphibians. This includes (for example) tractors, excavators, loaders, mowers, harvesters, dredgers, etc. For ecological fieldwork we refer to our disinfection protocol fieldwork.

Publication: February 2018

www.amphibianark.org/pdf/Disinfection-protocol-fieldwork-DEF.





We're continuing to help support amphibian conservation programs for threatened species by raising awareness and resources, with the sale of AArk clothing. Please join us and check out our new T-shirt designs featuring some of your favourite frog species, or show your support by proudly wearing our new AArk Rescue Team t-shirts

We've recently added many different designs and colors, in men's women's and children's sizes.

Some of the items feature species from our partners' breeding programs, and all profits from these shirts will go directly to supporting amphibian conservation programs.

Head to the AArk clothing store at www. amphibianark.org/AArk-products.htm and check out our clothing items!

Your continued support is helping to save the most threatened amphibians!



### A future-proofing plan for Papua New Guinea frogs

Chris Banks, Zoos Victoria, Australia; Dr Simon Clulow, Macquarie University, Australia; Michelle McGeorge, Port Moresby Nature Park, Papua New Guinea; and Dr Deborah Bower, James Cook University, Australia

#### **Background**

The amphibian chytrid fungus (*Batrachochytrium dendrobatidis*) is now recognised as the most significant example of an emerging infectious disease worldwide. New Guinea is the world's largest tropical island and remains the last major centre of amphibian biodiversity that may be chytrid-free. This is significant because the island is megadiverse for frogs - home to >6% of the world's amphibian species in less than 1% of the world's terrestrial surface, with more than 400 described species and many more yet to be described. New Guinea is particularly vulnerable to the introduction of parasitic chytrids because of its proximity to Asian pet markets and infected sites in both eastern Indonesia and Australia, and the increasing movement of people between these regions.

Climatic modelling reveals that large areas of the central highlands of New Guinea have a climate favourable for the fungus. Many of the Australian frog species that have declined since the 1970s have close evolutionary and ecological affinities with the New Guinea frog fauna, strongly suggesting that, as in Australia, dramatic declines will occur when the pathogen arrives. Patterns of frog declines in Australia suggest that more than one hundred frog species across New Guinea could be at risk from the amphibian chytrid fungus.

A recent publication in Science calls for "unified, international, multidisciplinary action to prevent or slow the spread of chytrid to, and within, New Guinea and prepare for its arrival". The ultimate goal is to secure New Guinea's amphibian fauna against the likely catastrophic impacts of amphibian chytrids. With the regular

movement of people, animals and goods between New Guinea and neighbouring countries, amphibian chytrids reaching New Guinea is considered a matter of when, not if.

New Guinea is a large island, with rugged terrain and a poor road system that prevents easy access to habitats where many of the frog species occur. Moreover, it is subject to the laws and management of two national governments, Papua New Guinea (PNG) in the east and Indonesia in the west. In light of the long and varied relationships between PNG and Australia, and existing herpetological links by Australian scientists with PNG, the first phases of this program will focus on PNG.

Developing and implementing an island-wide program is a longterm endeavour, requiring multiple avenues of research and development with many partners for success. In PNG, government capacity for implementing wildlife protection policies is limited. Similarly, in-country husbandry and research capabilities are in their infancy and heavily reliant on international support.

As such, immediate priorities are developing captive management capabilities for PNG frogs, in PNG, along with the capacity

Stakeholders in the Papua New Guinea frog plan (left to right):
Michelle McGeorge (Port Moresby Nature Park), Jon Birkett
(Melbourne Zoo), Nikko (Conservation & Environment Protection
Authority), Barnabas Wilmot (Conservation & Environment
Protection Authority), Chris Banks (Zoos Victoria), Dr Simon
Clulow (Macquarie University), Dr Deborah Bower (James Cook
University) and Brett Smith (Port Moresby Nature Park).



for genome storage of gametes and other tissues and the associated assisted reproductive technologies required for managing the genetic diversity of these populations in perpetuity. While the ideal solution for managing declining populations is to arrest the cause(s) of decline, this is not always possible in the timeframe required to prevent the erosion of genetic diversity, as is almost certainly the case with the amphibian chytrid fungus when it arrives in PNG.

#### Establishing policy frameworks and initiating management & research protocols for PNG frogs

Following extensive discussions during 2017, and a meeting in Port Moresby in December, phase 1 is well underway:

- Agreement between all stakeholders on the program's rationale and policy framework, including the PNG Conservation & Environment Protection Authority (CEPA).
- Port Moresby Nature Park will be the physical base of the program and has allocated space and initial infrastructure. The Park is the only zoo in PNG with the necessary capacity and commitment to frog biodiversity conservation.
- Zoos Victoria, under the banner of its sister zoo partnership with the Nature Park, has a long-term commitment to staff support
  and training. This includes sharing its frog husbandry and breeding expertise to develop and sustain a captive management plan
  for PNG frogs.
- Three Australian universities (Newcastle, James Cook and Macquarie) will establish a genome storage facility at the Nature Park to manage genetic diversity of declining populations and to enable disease research, collection and storage of genetic material on-site, and the use of assisted reproduction in managing genetic diversity.
- The captive husbandry program will develop in line with growth of staff skills at the Nature Park. Recognizing that these are currently low, the program will commence with two or three common species naturally occurring on the Nature Park grounds. The Cane Toad (*Rhinella marina*) and White-lipped Tree Frog (*Litoria infrafrenata*) are resilient species that will allow development of husbandry capacity with terrestrial and arboreal species. The Pond Frog (*Papurana daemeli*) is an additional species under consideration.
- As management skills develop with the aforementioned species over the next twelve to eighteen months, appropriate species for
  the following phase will be identified and collection approvals negotiated. These will be chosen from the microhlyids, which comprise the most extensive group of frogs in PNG, with a huge radiation of species across numerous ecological niches. Most are
  very small and only found in the highlands, with unknown captive requirements. Their high diversity of ecology and morphology
  suggests that some are also likely to be vulnerable to the amphibian chytrid fungus and are therefore the highest priority.

These plans complement two additional activities, which are already in place and ongoing:

- Expand permanent research transects and survey sites throughout PNG (currently established in the Kikori Basin and Eastern highlands (Goroka and surrounds) to monitor for disease arrival.
- Biodiversity surveys with local landowners to establish baseline data sets, monitor frog populations, and increase knowledge of species diversity, in areas where disease surveillance has been established.

#### **Moving forward**

The December meeting in Port Moresby allowed the team to agree on priorities for 2018:

- Site visit by Zoos Victoria invertebrate specialist to Port Moresby Nature Park in June-July to identify live food collection and production opportunities and establish best options.
- Fit-out of two forty-foot insulated shipping containers on-site now at the Nature Park in September-October.
- Collect first groups of frogs at the Nature Park in November-December and establish in the first container.
- Establish genome capture and storage capability in the second container.

This is a very exciting project and we look forward to bringing in other stakeholders over coming years to secure a positive future for PNG frogs.



The White-lipped Tree Frog is already a feature at the Port Moresby Nature Park.

### Amphibian Ark donors, January 2017 - March 2018

The work of AArk is possible due to the generous support of the following individuals and institutions:

#### Up to \$60,000



**Bernard & Nancy Karwick** 

George Rabb, in honor of Mary Sughrue Rabb



Up to \$10,000

**Anne Baker & Robert** Lacy in memory of George Rabb





**Louis Schauer** 



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Kate Woodle, The Kuehlthau Family Foundation

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