

2014 ANNUAL REPORT

PANAMA AMPHIBIAN RESCUE AND CONSERVATION PROJECT



A project partnership between: Cheyenne Mountain Zoo, Houston Zoo, Smithsonian's National Zoological Park, Smithsonian Tropical Research Institute, and Zoo New England.



MISSION

Our mission is to rescue and establish assurance colonies of amphibian species that are in extreme danger of extinction throughout Panama. We will also focus our efforts and expertise on developing methodologies to reduce the impact of the amphibian chytrid fungus (Bd) so that one day captive amphibians may be re-introduced to the wild.

VISION

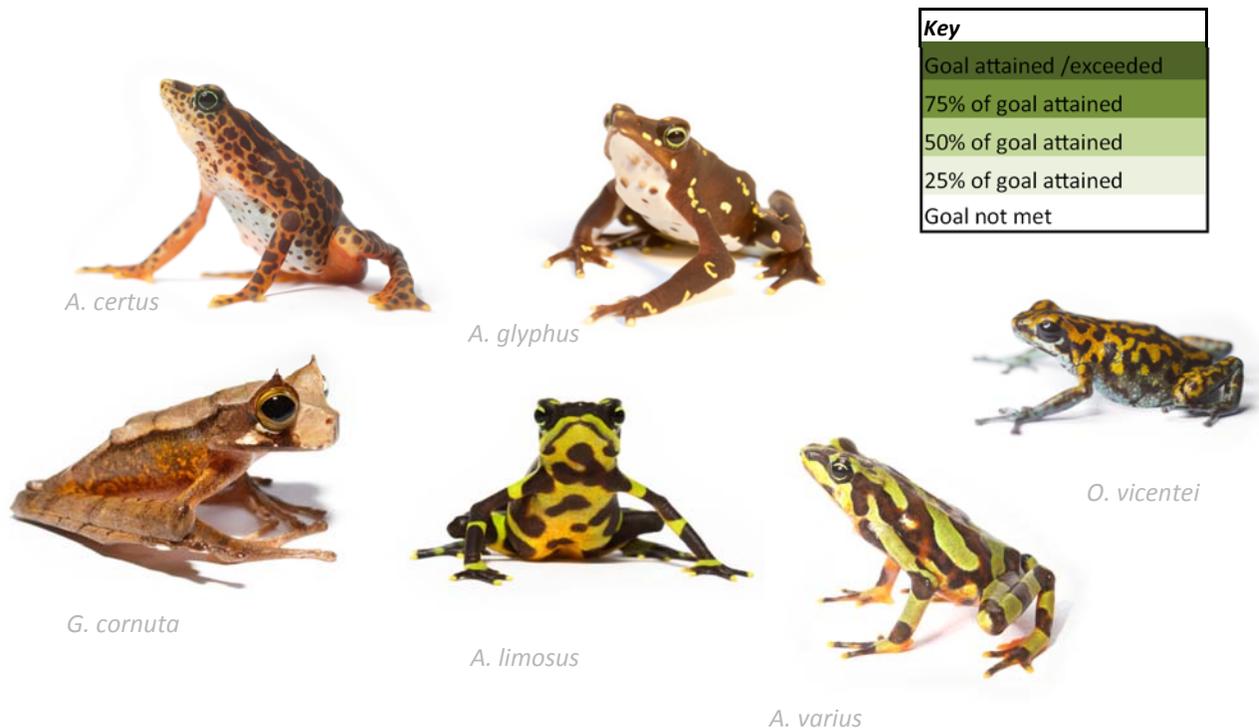
The Panama Amphibian Rescue and Conservation Project will be a sustainably financed, Panamanian-led organization that has stemmed the tide of extinctions caused by amphibian chytrid fungus and other threats to amphibians. We will lead successful recovery programs for Panama's endangered amphibians and serve as an exemplary model that can be replicated to address the threat of chytridiomycosis to the survival of amphibians worldwide.

GOAL 1: Create assurance colonies of Panama’s most vulnerable amphibian species

Our objective is to establish ex-situ colonies of 20 species at EVACC and Gamboa, and to demonstrate success with 14 species by the end of 2015. We have attempted to establish founding populations of 16 species. To date we have acquired 20 pairs or bred 10 pairs, whichever ratio is greater, for 3 species. We are close to achieving this goal for an additional four species.

Table 1: Progress towards captive population management goals at EVACC and Gamboa.

	Proportion of required founder population*	Pairs bred to F 1 (goal 10 pairs bred)	Pairs bred to F2 (goal 10 pairs bred)
<i>Atelopus certus</i>	1	11	0
<i>Atelopus glyphus</i>	1	4	0
<i>Oophaga vicentei</i>	1	1	0
<i>Gastrotheca cornuta</i>	0.8	9	2
<i>Atelopus limosus (chevron)</i>	0.75	5	0
<i>Atelopus varius</i>	0.8	3	0
<i>Craugastor evanesco</i>	0.7	0	0
<i>Anotheca spinosa</i>	0.5	5	7
<i>Strabomantis bufoniformis</i>	0.525	1	0
<i>Hemiphractus fasciatus</i>	0.375	2	0
<i>Hylomantis lemur</i>	0.2	1	0
<i>Atelopus zeteki</i>	0.2	2	0
<i>Atelopus limosus (brown)</i>	0.2	2	0
<i>Andinobates geminisae</i>	0.275	1	0
<i>Colostethus panamansis</i>	0.15	0	0





We studied aggressive interactions in co-housed male Atelopus. Frogs initially interacted aggressively, but after four weeks together, levels of stress and aggressive interactions declined. We concluded that animals could safely be co-housed in same-sex groups over the longer term.

Prioritization

One species (*Hyloscirtus colymba*) was discontinued. Despite having collected a full founding population, breeding attempts were unsuccessful and all founders collected primarily between 2008 and 2010 had died. After 6 years of experience and lessons learned, we realized that our programmatic deviations from the original Amphibian Ark species prioritization process were driven primarily by the ability to collect founding animals, ability to breed the animals successfully in captivity and the probability that the animals would become extinct in the wild. We surveyed 25 Panamanian amphibian experts and used this model to estimate the probability of avoiding species extinction through captive breeding programs to help refine our prioritization efforts.

We partnered with the IUCN redlist to help reevaluate all Panamanian amphibian species and have conducted literature reviews of all publications since 2004. In 2014, we consulted with the Panamanian Amphibian Specialist Group and updated literature reviews and fast-tracked updates for 110 species of amphibians in Panama that are Least Concern. We also began soliciting opinions on the remaining species using an inaturalist redlisting forum.

Husbandry

This year we continued to improve and update our record-keeping using the ZIMS software program. This was facilitated at EVACC through a generous donation of satellite internet from Odebrecht. We updated all of our husbandry manuals and now have facility-specific husbandry



manuals for EVACC, Gamboa and the Punta Culebra Nature Center. We began a tadpole nutrition research project to investigate the role of protein content in the development of tadpoles in conjunction with the University of Derby and the Waltham Center for Pet Nutrition, funded by the Waltham foundation. We published our findings on the stress effects of group housing on *Atelopus* (Cikanek et al 2014) and on the effects of captivity on the skin microbiome of Panamanian golden frogs (Becker et al 2014).

Atelopus varius at EVACC © Joel Sartore, PhotoArk

Veterinary care



Dr. Della Garelle works with Heidi Ross to examine a frog at the El Valle Amphibian Conservation Center.

2014 was challenging for our veterinary team, as many of the frogs of all four target species that were brought in from the field had *Bd* infections. This was further complicated by a novel fungus detected primarily in *A telopus varius* that was associated with unusual symptoms not encountered in this species before. The fungus was contained in quarantine but prophylactic itraconazole or heat treatments did not clear the infection and it was associated with high mortality rates. We are still investigating the new fungus, trying to develop PCR tools to detect it, and investigate potential treatment or management options.

We are collaborating with the San Diego Zoo's Alan Pessier who conducted necropsies on amphibians from this project that were published this year (Pessier et al. 2014). We identified a unique kidney pathology that is associated with edema problems in *A telopus* both at EVACC and Gamboa facilities, but the cause remains unknown and requires further investigation.

Expeditions

The Gamboa facility and EVACC facility coordinated eight field trips to collect founding populations of priority species, which are located in the Donoso region of Panama targeting *A telopus varius*, *Craugastor evanescens* which are both highly sensitive to *Bd* as well as the newly described *Andinobates geminisae* and *Oophaga vicentei* that have highly restricted distributions and are vulnerable to habitat modifications. We continued our collaboration with project *A telopus* led by Cori Richards-Zawacki and Jamie Voyles searching for *A telopus varius* and *A. zeteki* at historical sites. The team found some persisting populations of *A. varius*, but no *A. zeteki*.





GOAL 2: Re-establish healthy wild populations of Panamanian amphibians

Finding tools to manage *Bd* in reintroduced amphibians

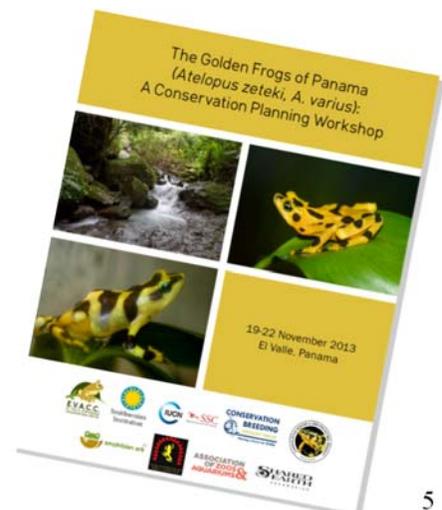
We found that probiotic inoculations generate a significant immune response from frogs, which may help to explain why it is difficult to manipulate frog skin microbiomes (Küng et al. 2014). We analyzed data on the survivors of our 2013 probiotics experiment and found that the golden frogs that survived and cleared *Bd* infections had a unique skin microbiome that was unconnected with the probiotic treatments. There was no clear genetic signal associated with survival, likely because samples were collected long after the frogs that had cleared an infection and were no longer expressing the genes that were part of the immune response.

The finding that there can be differential *Bd*-infection outcomes in Panamanian golden frogs is significant for this highly sensitive species. It offers new avenues of research into the mechanisms of survival, and if we had an ability to predict survival it would be extremely useful for reintroduction purposes. These questions are being actively pursued by Smithsonian Post-doctoral Fellow Matthew Becker with support from the Golden Frog Species Survival Plan and a grant from the USFWS and the Smithsonian.

In 2014, the wider research community made incremental progress to better understand *Bd* with 163 peer-reviewed papers on the topic published in 2014. The first ever vaccination method involving live *Bd* that conferred a small, but significant survival benefit on two species of amphibians (McMahon et al. 2014). This may have applications in the future.

Reintroduction planning

The IUCN Conservation Breeding Specialist Group published “The Golden Frogs of Panama (*Atelopus zeteki*, *A. varius*): A Conservation Planning Workshop Final Report” this year. The report lays out a detailed framework of actions that will ultimately lead to the reintroduction Panamanian Golden Frogs in the wild. A formal implementation group meets periodically to coordinate implementation of the plan among different stakeholders.



GOAL 3: Build capacity in Panama to safeguard the nation’s amphibian fauna over the long-term

We met our goal of 5,000 sq ft of ex-situ conservation space and are on target to exceed that goal by 60% by the end of 2015 with more than 8,000 sq ft dedicated to ex-situ conservation of amphibians in Panama.

Table 2: Ex situ amphibian conservation space built, or under construction in Panama.

Ex situ conservation space	Complete	Incomplete	Completion date
EVACC main building	2,400		2008
EVACC Minera annex	500		2013
EVACC Golden frog pod		400	2015
Gamboa ARCC Phase 1	2,800		2013
Gamboa ARCC Phase 2		2,200	2015
Total sq ft	5,680	2,600	

This year we had planned to focus on getting the EVACC golden frog pod up and running, but termite issues in the main building required that we prioritize our attention to replace shelving and other wooden furnishings there. EVACC is now 7 years old and several maintenance issues require urgent attention and funding. The Gamboa site phase 1 is operational, but is part of an active construction site. Workflows will be much improved once the main building and research lab is complete in 2015.

We now have 13 full time employees at Gamboa and EVACC. It has been difficult to recruit a biologist position to live in El Valle de Anton, but the addition of the Minera Annex apartment to EVACC has been a valuable benefit that has allowed us to attract more qualified candidates. As a result, staff retention has improved markedly over the last few years and has helped us to solidify the capacity-building gains in the training area. We are actively working to build a support base of potential contributors and supporters in Panama that have toured our facilities extensively in 2014. Our grant from Minera Panama is being renewed for 2015 and 2016 and together with core partner-provided funding this will continue to provide funding for staff salaries, training, and animal care over the longer term that will be supplemented by project-specific grants and external collaborations.

In 2014, Matthew Becker a US PhD student graduated from VA Tech University. Two of our former Panamanian project staff members Angie Estrada and Daniel Medina began their PhDs at VA Tech in the Belden Lab. We also have Gina DellaTogna, a Panamanian PhD student finishing up her PhD at the University of Maryland.



Dr. Roberto Ibáñez receiving the BBVA award for conservation ceremony in Spain.

In 2014 we were awarded a prize for €250,000 from BBVA in Spain, in recognition of our outstanding contributions to conservation in Latin America. Roberto Ibáñez and Heidi Ross travelled to Spain to receive the prestigious award. Brian Gratwicke was awarded the Defenders of Wildlife Spirit of Defenders award for science in recognition of his contributions to amphibian conservation.

GOAL 4: Cultivate and foster an appreciation for amphibians in the public mindset

Fabulous Frogs of Panama exhibit and informal curriculum

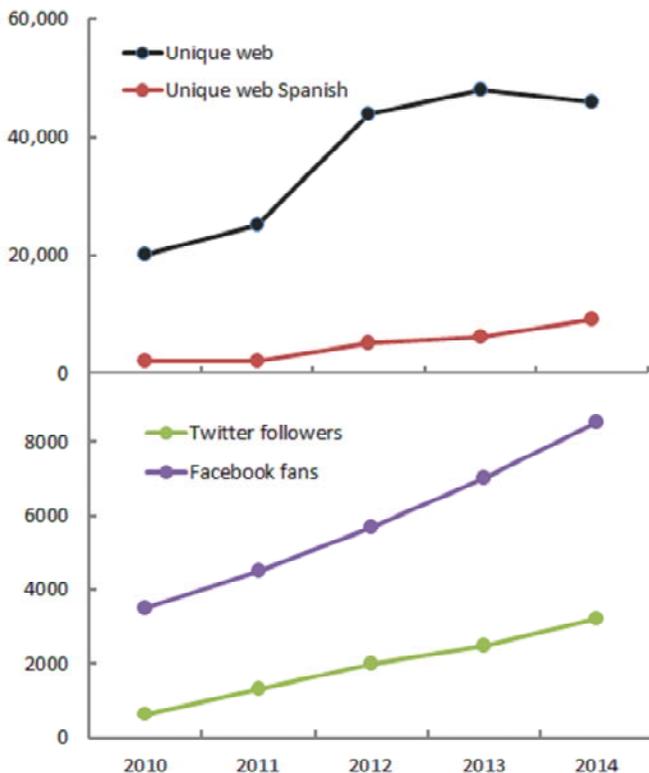
This year was our biggest and most focused year with education activities, thanks in large part to the contributions of Hannah Arney, a Peace Corps volunteer who acted as a full time education coordinator for this project. We opened a significant new frog exhibit “Fabulous Frogs of Panama” at the Punta Culebra Nature Center in Panama. This Smithsonian exhibit pulled together by Sharon Ryan, director of public programs at STRI. The exhibit covers the biodiversity of frogs, threats to frogs, frogs in Panamanian culture and highlights Panama’s natural treasures. We also developed an informal learning curriculum and trained more than 100 teachers to use it. Each module features a Panamanian amphibian species; adaptations for different age groups; explicit links to the national school’s curriculum; link to the amphibian rescue project; and highlights an action that participants can take to directly help the Panama Amphibian Rescue and Conservation Project fulfill its mission. Explore the curriculum here <http://amphibianrescue.org/education/>



Golden Frog Festival 2014

The Golden Frog Festival in 2014 has grown rapidly and been enthusiastically embraced by the people of Panama. The 2014 festival was marked by events throughout the week and a golden-frog themed lottery ticket drawing. In El Valle de Anton, the Ministry of the Environment assumed leadership in organizing the 2014 Golden Frog Day Parade and we organized a fun family day at Rey Supermarkets over the

weekend. Caminando Panama hosted the first ever La Dorada 5k/15k Trail Run. In Panama City we opened the “Fabulous Frogs of Panama” and hosted a frog-themed family day at the Punta Culebra Nature Center catered by La Rana Dorada food truck. We also had a ‘science in the city’ series of pub talks hosted by the American Trade Hotel and La Rana Dorada Pub. All in all, we had 80 volunteers, 2,000 festival participants (excluding the parade count) and were covered in 10 TV spots.



Media

Our project was covered in 17 independent English and Spanish news stories in 2014, and our online constituency continued to grow strongly (Fig 1). Unique web visitor counts were lower than in 2013 because we produced less original content than in previous years due to the loss of key communications team volunteers who developed and translated content for us, but we had sustained growth of Spanish language visitors to our website.



New amphibian exhibit at Punta Culebra Nature Center



Golden frog day festivities



Captive-bred *Strabomantis bufonifamis* at EVACC

FINANCIAL REPORT FOR CALENDAR YEAR 2014

	FUNDING SOURCE	2014 expenses
EVACC		
Staff salaries and supplies	<i>Houston Zoo and EVACC donors</i>	\$ 82,200
	<i>SCBI and donors</i>	\$ 8,860
Gamboa ARCC		
New facilities	<i>USAID</i>	\$ 93,345
	<i>NSF</i>	\$ 317,518
Staff salary and supplies	<i>SCBI and donors</i>	\$ 21,242
	<i>USAID</i>	\$ 79,799
	<i>Zoo New England</i>	\$ 20,000
	<i>Cheyenne Mountain Zoo</i>	\$ 20,000
	<i>Minera Panama</i>	\$ 331,974
	<i>Friends of the National Zoo (online)</i>	\$ 1,720
Education		
Exhibit at Punta Culebra	<i>NSF</i>	\$ 18,000
	<i>USAID</i>	\$ 3,210
	<i>STRI and donors</i>	\$ 20,000
Informal curriculum	<i>Disney Worldwide Conservation Fund</i>	\$ 4,000
Golden frog festival 2014	<i>STRI and donors</i>	\$ 13,000
Research		
Sperm freezing	<i>Wood Tiger Foundation</i>	\$ 29,000
Tadpole nutrition	<i>Waltham foundation</i>	\$ 20,000
Golden frog research	<i>SCBI and donors</i>	\$ 55,000
SCBI staff salary and travel	<i>SCBI and donors</i>	\$ 100,000
TOTAL		\$ 1,238,868



2014 Golden Frog Day Festival Celebrations
in El Valle de Anton

2014 Scientific Publications Related to the PARC Project

Becker, M. H., Richards-Zawacki, C. L., Gratwicke, B. & Belden, L. K. (2014). The effect of captivity on the cutaneous bacterial community of the critically endangered Panamanian golden frog (*Atelopus zeteki*). *Biological Conservation* 176, 199.

Cikanek, S. J., Nockold, S., Brown, J. L., Carpenter, J. W., Estrada, A., Guerrel, J., Hope, K., Ibañez, R., Putman, S. B. & Gratwicke, B. (2014). Evaluating Group Housing Strategies for the Ex-Situ Conservation of Harlequin Frogs (*Atelopus spp.*) Using Behavioral and Physiological Indicators. *PLoS one* 9, e90218.

Estrada, A., Gratwicke, B., Benedetti, A., DellaTogna, G., Garrelle, D., Griffith, E., Ibañez, R., Ryan, S. & Miller, P. S. (2014). The Golden Frogs of Panama (*Atelopus zeteki*, *A. varius*): A Conservation Planning Workshop Final Report) Apple Valley, MN: IUSN/SSC Conservation Breeding Specialist Group.

Küng, D., Bigler, L., Davis, L. R., Gratwicke, B., Griffith, E. & Woodhams, D. C. (2014). Stability of Microbiota Facilitated by Host Immune Regulation: Informing Probiotic Strategies to Manage Amphibian Disease. *PLoS ONE* 9, e87101.

Perez R, Richards-Zawacki CL, Krohn AR, Robak M, Griffith EJ, Ross H, Gratwicke B, Ibañez R & Voyles, J (2014). Field surveys in Western Panama indicate populations of *Atelopus varius* frogs are persisting in regions where *Batrachochytrium dendrobatidis* is now enzootic. *Amphibian & Reptile Conservation* 8, 30.

Pessier, A. P., Baitchman, E. J., Crump, P., Wilson, B., Griffith, E. & Ross, H. (2014). Causes of mortality in anuran amphibians from an ex situ survival assurance colony in Panama. *Zoo Biology* 10.1002/zoo.21166.



This *Atelopus varius* was found to persist at some sites in central Panama where *Bd* is endemic Photo © Jamie Voyles, Project Atelopus

Donors

In addition to the contributions from project partners, we are grateful to the following donors who have made additional contributions to the project directly or via the Houston Zoo: Anele Kolohe Foundation, Baton Rouge Zoo, Buffalo Zoo, Dawn and Bob Bermingham, Disney Worldwide Conservation Fund, Canadian Embassy in Panama, Conservation Breeding Specialist Group, Dickerson Park Zoo, Friends of the National Zoo, La Rana Dorada Restaurant, Los Mandarinos Hotel, Minera Panama, National Science Foundation, North Face, Odebrecht, Towerbank, Taffy Stern, Sedgwick County Zoo, George and Mary Rabb Foundation, Roger Williams Park Zoo, Shared Earth Foundation, Undercover Rain Boots, USAID, US Fish and Wildlife Service, Waltham Foundation, Wood Tiger Foundation, Zoo Miami.

Online Contributions

Our sincere thanks to the following individuals who contributed \$25 or more online: Bethany Baldwin, Rob Heath, Daemon Hobbs, Charlene Mangi, Caroln Miller, Matt Parisi, Bruce Potter, Anthonie Ravensbergen, Hugo Saunders-Camposanto, Johanna Schmidt, Aaron Zorn.

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El Valle Amphibian Conservation Center Director – Heidi Ross.

Technical Staff – Milagro Gonzalez, Yeisson Muñoz, Fernando Sánchez.

Minera Panama support staff – Diana Troetsch, Abileth González, Edgardo Griffith.

Steering Committee

Bob Chastain, President and CEO *Cheyenne Mountain Zoo*;

Rick Barongi, Executive Director *Houston Zoo*;

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Matthew Larsen, Director *Smithsonian Tropical Research Institute*;

John Linehan, President and CEO *Zoo New England*.

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Heidi Ross *Smithsonian Tropical Research*

Institute; Dr. Eric Baitchman *Zoo New*

England; Dr. Brad Wilson.



2014 Volunteers

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2014 Golden Frog Festival

Organizing Committee: Sharon Ryan, Roberto Ibáñez, Jorge Alemán, Nelly Florez, Sonia Tejada, Álvaro González, Hannah Arney, Rigoberto Díaz, Lidia Valencia, Adrián Benedetti, Ana Matilde Ruíz, Ana Endara, Sean Mattson, Carlos Celis, Ana Lucrecia Arosemena, Heidi Ross, Dayra Navarro, Lanki Cheucarama, Dara Wilson. **Volunteers:** Lillian Ayala, Anna Frogge, Jean Glaxy, Katherine González, Esteban Guerra, Johany Lau, Marlyn Montenegro, Joanna Pérez, Kyara Ortega, Iliana Quintero, Quiriatjaryn Ortega, Andreina Ríos, Marie Ciel Rodríguez, Marie Sheznar Rodríguez, Carol Simon.

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