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Zoo-based amphibian research and conservation breeding programs

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***Abstract.*—The rapid loss of amphibian species has encouraged zoos to support amphibian research in concert with conservation breeding programs (CBPs). We explore "Zoo-based amphibian research and conservation breeding programs" through conducting a literature review and a survey of research publication with public and subscription search engines. Amphibians are ideal candidates for zoo-based amphibian research and CBPs because of their generally small size, high fecundity, ease of husbandry, and amenability to the use of reproduction technologies. Zoo-based amphibian research and CBPs can include both *in situ* and *ex situ* components that offer excellent opportunities for display and education, in range capacity building and community development, and the support of biodiversity conservation in general. Zoo-based amphibian research and CBPs can also benefit zoos through developing networks and collaborations with other research institutions, and with government, business, and private sectors. Internet searches showed that zoo based research of nutrition, husbandry, reproduction, gene banking, and visitor impact offer special opportunities to contribute to amphibian conservation. Many zoos have already implemented amphibian research and CBPs that address key issues in both *ex situ* and *in situ* conservation; however, to reach its greatest potential these programs must be managed by scientific professionals within a supportive administrative framework. We exemplify zoo-based amphibian research and CBPs through the experiences of zoos of the European Association of Zoos and Aquariums (EAZA), the Russian Federation, and the United States.**

Key words. Zoo research, amphibian, conservation breeding programs, Internet searches, Internet surveys

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Introduction

Official reports estimate more than nearly 158 amphibian species have gone extinct since their description (AmphibiaWeb 2011) and that 30% of the 6726 species of amphibians listed by the IUCN Amphibian Red List (IUCN 2010) are threatened, including 484 Critically Endangered and 754 Endangered species. Over the coming decades threats to amphibians are expected to increase with a corresponding increase in the number of amphibians requiring dedicated management programs (McCallum 2007; Sodhi et al. 2008).

To reduce the rate of biodiversity extinction in general the World Zoo and Aquarium Conservation Strategy (WAZA 2005) committed the world's zoos to include conservation breeding programs (CBPs) supported by research as a key component in their conservation strategies (Baker 2007; Hutchins and Thompson 2008). CBPs prevent species extinction through maintaining genetically representative populations and providing animals for supplementation, rehabitation, or translocation projects (Baker 2009; Shishova et al. 2010; Browne et al. 2011). In 2007 specific support for amphibian CBPs was also provided by the Species Survival Commission of the International Union for the Conservation of Nature (IUCN/SSC) who recommended that CBPs should be implemented where necessary for all critically endangered amphibians (Gascon et al. 2007). To efficiently address the prevention of species loss in 2009 the European Association of Zoos and Aquariums (EAZA) recommended combining CBPs with scientific research, education, and outreach (EAZA 2009).

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Figure 1. Research in zoos, such as this study on tadpole growth and development at Antwerp Zoo, can make substantial contributions to conservation breeding programs. *Image by Robert Browne.*

The number of amphibian species that require CBPs is challenging. However, the World Association of Zoos and Aquariums (WAZA) represent 241 zoos in 48 countries, and globally there are more than 1000 zoos and aquariums in zoo and aquarium associations (WAZA 2009). This number is greater than the total number of Critically Endangered amphibians, some of which do not immediately need CBPs and may be perpetuated through *in situ* initiatives. Therefore, the support of amphibian CBPs by zoos' in concert with other institutions should be able to assure a minimal risk of amphibian extinctions.

To achieve the highest benefit to cost ratio the structure of CBPs preferentially should integrate both international and regional capacities (Reid et al. 2008; Ziegler 2010). CBPs in a species' biogeographical or biopolitical range are generally more economical and sustainable than those out of range, and they also provide the advantages of local scientific expertise, capacity building, and community engagement (e.g., Ziegler and Nguyen 2008; Nguyen et al. 2009). Maintaining rescue populations within regions also reduces the chance of pathogen dissemination (Pessier and Mendelson 2010) or the release of invasive species (NBII 2011). Regional universities, government departments, and NGOs can all provide centers for expertise and facilities combined with academic research.

Amphibian CBPs offer zoos, with limited capacity,

an attractive alternative to those for large mammals and birds, or with zoos, in general, an opportunity for diversification or extension of their conservation programs. The primary goals of CBPs initially include the building of a genetically representative captive population, and then maintaining health, reliable reproduction, and the perpetuation of genetic variation. However, problems with satisfying these criteria for larger vertebrates (Araki et al. 2007) make the management of zoo-based CBPs for these species expensive and difficult (Lees and Wilcken 2009). Baker (2007) showed that since 2000 the success of CBPs for large, thermoregulating vertebrates has declined due to numerous challenges including insufficient founders, poor health and reproduction, and loss of genetic variation (Hutchins and Conway 1995; Baker 2007). In contrast, amphibians are mostly small, adequate numbers of founders may be sampled and held, are amenable to husbandry, and their reproduction and genetic variation can be managed especially when supported by research (Browne and Figiel 2010; Browne et al. 2011).

Therefore, zoo-based amphibian CBPs can include direct maintenance of genetically competent populations, as well as their use for education, display, and research. They can also extend to other institutions and private keepers and breeders within the international community (Zippel et al. 2010), while offering support to local communities, preserving habitat, supplying surplus amphibians for the pet market, and reducing wild harvesting (Furrer and Corredor 2008; Zippel et al. 2010). Zoo-based amphibian CBPs can sell surplus amphibians to generate funds directly for conservation, gain valuable publicity, and widen the range of threatened species available to private caregivers.

Zoos are housing an increasing number of exhibits supporting amphibian conservation (Zippel 2009; Amphibian Ark 2010). Amphibians are easily kept in attractive exhibits where their role within ecosystems and the reasons for their decline can be presented. Through public education that demonstrates zoos' role in amphibian conservation and research, zoos can function as ambassadors for contemporary best practice in *ex situ* biodiversity conservation (Reid et al. 2008; Ziegler et al. 2011).

Ex situ research for amphibians can vary over a wide range of disciplines including nutrition and husbandry, display and education, population genetics, and reproduction technologies. *In situ* research includes amphibian biodiversity assessment, ecology, habitat preserva-

Figure 2. *Neurergus kaiseri.* In a pioneering program, Sedgwick County Zoo, Kansas, USA, is breeding for sale the critically endangered Loristan newt (*Neurergus kaiseri*) to support field work and conservation in Iran and to increase stocks with private breeders. *Image by Nate Nelson.*

tion, and identifying threats and their mitigation (Browne et al. 2009). Therefore, amphibian research in zoos can support both *in situ* and *ex situ* conservation of amphibians, contribute to fundamental science, and can develop valuable scientific and conservation collaborations (Furrer and Corredor 2008; Browne et al. 2009).

In situ aspects of amphibian CBPs offer zoos attractive opportunities to integrate their amphibian conservation strategies with those for general biodiversity. These include the establishment of regional facilities, habitat preservation, and community education that provide a focus for biodiversity conservation and ecosystem sustainability (Lawson et al. 2008). Amphibians with aquatic life stages are particularly susceptible to extinction where threats include water borne diseases (Lips et al. 2003), water pollution (Rohr 2008), and introduction of invasive species (M. Bagaturov and K. Mil`to, pers. comm.).

Table 1. The hits for each term, for a scientific field, as a percentage of all hits (years covered, 1900 to 2009). Searches engines; 1) *Google Scholar*, 2) *PubMed*, 3) *Scopus*, and 4) *ISI Web of Knowledge*.

The percentage of "term" hits of total "scientific field" hits from 1900 to 2009					
Search engine	**1**	**2**	**3**	**4**	**Mean**
Scientific field					
Behavior	34	4	19	66	31
Behaviour	9	1	14	21	11
Medicine	21	27	2	7	14
Disease	24	9	8	34	19
Husbandry	7	1	1	1	3
Aquaculture	1	1	1	1	1

Table 2. The hits for each scientific field as a percentage of all hits (for scientific fields: years covered, 1900 to 2009). Searches engines; 1) *Google Scholar*, 2) *PubMed*, 3) *Scopus*, and 4) *ISI Web of Knowledge*.

The percentage of subject hits of total hits from 1900 to 2009					
Search engine	**1**	**2**	**3**	**4**	**Mean**
Scientific field					
Behavior/behaviour	23	6	30	47	27
Physiology	6	70	18	11	26
Medicine/disease	25	3	9	16	13
Reproduction	24	1	8	10	12
Genetics	9	17	11	5	11
Diet	8	1	4	6	5
Population genetics	1	1	8	3	3
Husbandry/aquaculture	4	1	2	1	2
Nutrition	1	1	1	1	1

Consequently, many *in situ* components of amphibian CBPs correspond with the conservation needs of threatened freshwater fish, reptiles, birds, mammals, plants, fungi, microorganisms, and invertebrates, including high risk groups like mussels, crayfish, and aquatic plants (Davic and Welsh 2004). In some cases, due to their aquatic and terrestrial life stages and specialized microhabitats, amphibians may also be important bioindicators through complex ecological interactions (Rohr 2008).

We explore "Zoo-based amphibian research and conservation breeding programs" through a literature review, a survey of research effort through public and subscription Internet search engines, and provide examples of successful programs through the experiences of zoos of the European Association of Zoos and Aquariums (EAZA), the Russian Federation, and the United States.

Methods

A survey of research effort in scientific fields relevant to amphibian CBPs was conducted through two publicly accessible databases on the Internet (*Google Scholar* and *PubMed*), and two subscription Internet search engines (*Scopus* and *ISI Web of Knowledge*, volume 4.7). Searches were conducted over the years covered in the databases between 1900 to 2009. Search dates and data were collected on 27 December 2009 (*Google Scholar*, *Scopus*, and *ISI Web of Knowledge*) and 28 December 2009 (*PubMed*).

Search strings for amphibians were based on the following main descriptors: "amphibian [search subject]," "frog [search subject]," "salamander [search subject]," "toad [search subject]." Search strings were chosen for each search engine with a combination of the above descriptors that returned the maximum number of credible hits.

Using the above descriptors, the search subjects of alternative "terms," used to describe "scientific fields," were compared between the numbers of hits from the four search engines (Table 1). For "scientific fields" (alternative terms pooled) we also compared the percentage of hits of each of the total hits from 1900 to 2009 (Table 2).

Results

General: The total number of hits returned for all scientific fields were: *Google Scholar* (1,670), *PubMed* (10,741), *Scopus* (14,528), and *ISI Web of Knowledge* (6,245). *PubMed* indexed the *Medline* database of citations, abstracts, and full-text articles with a total number of indexed citations of more than 19 million. *Scopus* indexed more than 18,000 journals (including 16,500 peer-reviewed), 350 book series, and 3.6 million conference papers. *ISI Web of Knowledge* indexed more than 23,000 journals, 110,000 conference proceedings, and 9,000 websites. *Google Scholar* indexed an undetermined number of full-text articles from most peer-reviewed online journals, as well as citations, websites, and books from the main publishers in Europe and America.

Searches of alternate "terms" for "scientific fields:" Table 1 shows wide and inconsistent differences between search engines in the percentage of hits between alternate "terms" for scientific fields.

Searches of "scientific fields:" Table 2 shows the wide range, in the percentage of hits between search engines, for each term, for each scientific field, between search engines. The percentage of total hits, averaged from all search engines for each term, ranged from 1 to 27%. More than 50% of the average hits were from behavior/behaviour (27%) and physiology (26%), while medicine/disease, reproduction, and genetics comprised about 12% each. Only a small percentage of hits (11%) included diet/nutrition (6%), population genetics (3%), and husbandry/aquaculture (2%).

Discussion

Our Internet search engine survey of amphibian publications showed that search engines varied widely in the number of hits dependent on the terms used to describe the scientific field, and in hits for each scientific field. Therefore, when conducting search engine surveys, alternative subject terms for each scientific field should be compared through an appropriate range of search engines to produce meaningful results (Jansen and Spink 2006; UNEP-WCMC 2009).

There have been relatively few publications on amphibians, compared to other vertebrates, except fish in *Zoo Biology*, where Anderson et al. (2008) showed that from 1982 to 2006 publications mainly concerned mammals (75%), then birds (11%), reptiles (4%), amphibians (3%), fish (2%), and invertebrates (2%).

Anderson et al. (2008) also showed that overall, with vertebrates, some subjects critical to CBPs were poorly represented in zoo research. Publications over all taxa focused on behavior (27%), reproduction (21%), husbandry/animal management (11%), diet and nutrition (8%), veterinary medicine (7%), genetics (6%), anatomy/physiology (6%), and housing enrichment (4%; Anderson et al. 2008). Our Internet search engine survey showed a similar percentage of publication subjects for amphibians as in Anderson et al. (2008) for behavior/behavior and genetics, a higher percentage for medicine/disease, and lower percentages for reproduction, diet, husbandry/aquaculture and nutrition. Our survey also showed that in some fields important to amphibian CBPs, there were relatively few publications concerning medicine/disease, reproduction, and genetics, and even fewer publications on diet/nutrition, population genetics, and husbandry. Therefore, within the needs of CBPs, reproduction, diet, husbandry/aquaculture, nutrition, and genetics offer research subjects of particular value for zoos.

An Internet questionnaire survey of amphibian research efforts in zoos (Browne et al. 2010a) included responses from 89 institutions globally, with 47% of responses from AZA and 10% from each from EAZA, ALPZA, and ZAA/ARAZPA. This survey showed a recent change in emphasis in amphibian research efforts in zoos as a result of zoos' recognition of the value of amphibian CBPs. Research included 23% of institutions supporting wide-ranging research of phylogenetics/taxonomy and 30% supporting research of supplementation, rehabitation, or translocation. *Ex situ* research mainly focused on reproduction (54%), population management and conservation education (40%), diet/nutrition (30%), and disease management (22%). *In situ* research was highest for species conservation assessment (46%) and disease (35%), while 13% investigated each of land/water use, climate change, or introduced species, and 5% of environmental contamination or overharvesting.

Research effort increased over the period from 2008 to 2010, with ~80% of institutions having dedicated research staff and ~50% having space for research or access to museum or university facilities (Browne et al. 2010a). However, only ~35% had dedicated laboratory space or direct research funding, with the majority of funded institutions having less than US$5,000 in research funding. Nevertheless, there was a predicted increased proportion of overall funding in the bracket from US$5,000-50,000 from 2011 to 2013.

The need expressed in the survey for laboratory facilities could be partly satisfied by greater outreach and collaboration with academic institutions. Opportunities for increased scientific collaborations, networking, and provision of projects were also presented as research needs. Sixty percent of respondents had produced popular publications promoting amphibian conservation. There was considerable focus on peer-reviewed publications, with 30% of respondents having published, and 70% currently conducting scientific research for peer-review.

Anderson et al. (2008) showed that there was little direct collaboration between zoos and other institutions on research publications, with only 9% of articles coauthored between zoos and universities. The recent development of zoo research reliant upon professional staff may account for the greater emphasis on collaborative scientific publications. An aspect of zoo-based CBPs and research not investigated by Anderson et al. (2008) or (Browne et al. 2010a) was the embracing of authorship from regions of high amphibian biodiversity. Previous limitations in the breadth of authorship of articles (Newman 2001) are being addressed globally through the Internet, which offers expanding potential for both networking and communication (Olsen et al. 2008).

Six major challenges need to be overcome to achieve successful CBPs: 1) maintaining good husbandry techniques, 2) controlling reproduction, 3) maintaining genetic variation, 4) success in rehabitation, supplementation, or translocation, 5) providing oversight by professional scientific personnel, and 6) the fostering of career development through exchanges, meetings, and training of keepers and amphibian managers. These goals all appear achievable within zoo-based amphibian CBPs with the support of research.

Hutchins and Thompson (2008) found with rehabitation programs, mainly for mammals, that only 12% had established self-sustaining populations. In contrast, amphibian rehabitations were much more successful, where Griffiths and Pavajeau (2008) showed a success rate of 52% between 1991 and 2006. Similarly, Germano and Bishop (2009) found increased success of amphibian rehabitations between 1991 and 2009 in comparison to those before 1991 (Dodd and Siegel 1991). Although these achievements are impressive, Hutchins and Thompson (2008) suggested that further improvements could be made in CBPs through increased long-term research commitments.

In 1986, Soulé et al. published the need for CBPs for thousands of threatened mammal, bird, and reptile species. Due to low founder numbers, large body size restricting the numbers in captive populations, low fecundity, poor health, and difficulties in arranging suitable pairings, few of the established CBPs for mammals, birds, and reptiles are maintaining genetic variation (Baker 2007). Lowered genetic variation results in poor health and reproduction, which reduces the viability of the captive population and the production of competent individuals for release (Baker 2007; Akari et al. 2007; Allentoft and O'Brien 2010).

The small size of amphibians and recent advances in genetics, husbandry, and reproduction technologies, offer zoos the opportunity to develop CBPs with healthy and reproductive amphibians populations, the perpetuation of their genetic variation, and the ultimate goal of providing competent individuals for rehabitation, supplementation, or translocation (Browne and Zippel, 2007a; Burggren and Warburton 2007; Browne and Figiel 2011). The increasing use of gene banking, and particularly the use of cryopreserved sperm, enable the cost efficient and reliable perpetuation of amphibians' genetic variation. Additional cost benefits of gene banking are reduced numbers of individuals required for CBPs (Shishova et. al 2010; Browne and Figiel 2011, Mansour et al. 2011). Zoos are now in an excellent position to facilitate or directly develop reproduction technologies for amphibians (Browne and Figiel 2011; Browne et al. 2010; Shishova et al. 2010). Some zoos and supporting institutions can also now develop gene banks for threatened amphibians that store a range of samples including sperm, cells, and tissues (Browne and Figiel 2011).

However, although fertilization was first achieved with cryopreserved amphibian sperm in 1996 (Kaurova et al. 1996), sperm banks are only now being established

Figure 3. Hellbender sperm sampling. A team led by Dale McGinnity, Nashville Zoo at Grassmere, Tennessee, USA, is creating the first genetically representative gene bank for any amphibian put forth using the hellbender (*C. alleganiensis*). *Image by Sally Nofs.*

that represent the natural genetic variation of any amphibian species. For example, the North American giant salamander (*Cryptobranchus allegianensis*), most commonly called the hellbender (CNAH 2011), is suffering from very low or negligible recruitment over much of their range and only older adults remain. In response, Nashville Zoo at Grassmere, USA, has recently pioneered the sampling of semen over the range of *C. allegianensis* and developed techniques for its sperm cryopreservation and gene banking (National Geographic 2010; Michigan State University 2010). Zoos have played a significant role in the use of hormones to induce reproduction in both male and female amphibians (Browne et al. 2006a, b), and these technologies now promise the reliable reproduction of many species (Trudeau et al. 2010).

Diet and nutrition have a major effect on amphibian health, lifespan, and reproductive output (Li et al. 2009). Historically, research of amphibian diet and nutrition has mainly tested the benefit of dusting feeder insects with vitamin/mineral powder. However, the natural diet of amphibians includes insects with a wide variety of micronutrients. Recent research in zoos has included reviews of Vitamin D_3 deficiency (Antwis and Browne 2009), nutritional metabolic bone disease (King et al. 2010), and the supplementation of feeder insects to avoid vitamin and other micronutrient deficiencies (Li et al. 2009).

To reach their greatest potential, amphibian CBPs should extend to areas where amphibian biodiversity faces the greatest threats (Lötters 2008; Bradshaw et al. 2009). These areas are generally in developing countries of tropical regions where there is high growth in human population (United Nations 2004) and corresponding loss of native vegetation and wetlands (Wright and Muller-Landau 2006a, b), including much of Africa (Lötters 2008).

Specific threats to amphibians that could be incorporated into zoo-based *in situ* research include the loss and fragmentation of wetlands and forests (Bradshaw et al. 2009), emerging diseases (Dazak et al. 1999; Pessier 2008; Skerratt et al. 2007), pollutants and climate variability (McDonald and Sayre 2008; Foden et al. 2008), and unregulated harvest (Mohneke and Ródel 2009). In general, essential *in situ* research components of amphibian CBPs include surveys of range and distribution, pathogen assessment, DNA sampling and population genetics, microhabitat assessment, and autecology (Browne et al. 2009). Relict montain rainforests in tropical regions often provide the only remaining natural habitat for much biodiversity, and these forests are often subject to ongoing vegetation clearance (Lötters 2008; Bradshaw et al. 2009). Zoo research integrated with direct financial support, of the conservation of these relict habitats, could be particularly cost effective.

Many of these conservation initiatives are incorporated into Cologne Zoo's amphibian CBPs within a framework of long-term amphibian biodiversity research and nature conservation (Ziegler 2007; 2010). An Amphibian Breeding Station was established and founded by the Vietnamese and Russian Academies of Sciences at the Institute of Ecology and Biological Resources (IEBR) in Hanoi, Vietnam. Research supported by Cologne Zoo at the breeding station has focused on the ecology, reproduction, and larval identification, development of data-deficient and threatened amphibians, and the commercial breeding of selected species to both decrease over harvesting and provide financial support to help the station become self-supporting. Fourteen out of 21 species have successfully reproduced.

Cologne Zoo and their Vietnamese partners, including the Vietnam National University, Hanoi and IEBR, since 1999 have also conducted long-term biodiversity research at a UNESCO World Heritage Site, Phong Nha-Ke Bang National Park, Vietnam. This project works in concert with forest protection, ranger support, and wildlife rescue. In the past decade, thirteen new amphibian and reptilian species have been described from a small area of 86,000 ha and more than 40 new amphibian species have been described since 1980 (Ziegler et al. 2006, 2010; Ziegler and Vu 2009). Cologne Zoo also supports a CBP for amphibians at their aquarium in Cologne where 16 species have been reproduced in the past decade (Ziegler et al. 2011).

Many other zoos in EAZA have supported programs to develop regional capacity for amphibian conservation, where Durrell Wildlife Conservation Trust, UK, leads a major program for the conservation of the Montserrat mountain chicken frog (*Leptodactylus fallax*; Martin 2007; Garcia et al. 2007). A consortium of zoos and institutions in Europe, Canada, and the USA are building both *ex situ* and *in situ* capacity and research for the critically endangered Lake Oku clawed frog (*Xenopus longipes*; Browne and Pereboom 2009). A similar CBP is established for the critically endangered Kurdistan newt (*Neurergus microspilotus*) and Loristan newt (*N. kaiseri*) between European and USA institutions with Razi University, Iran (Browne et al. 2009).

Figure 4. *Trachycephalus nigromaculatus.* The black-spotted casque-headed treefrog (*Trachycephalus nigromaculatus*) is an excellent display species because it is large (10 cm), spectacular, and sits in the open. These frogs are very popular pets in the Russian Federation. *Image by Mikhail Bagaturov.*

Durrell Wildlife Conservation Trust, UK, has head-started Agile frogs (*Rana dalmatina*) in a successful program for their recovery. These skills were then transferred to an *ex situ* and *in situ* program for the Iberian frog (*Rana iberica*) and the Midwife toads (*Alytes obstetricans* and *A. cisternasii*; G. Garcia, pers. comm.). Perth Zoo, Australia, has established a CBP and rehabitation for the White-bellied frog that involves both *ex situ* and *in situ* components (*Geocrinia alba*; Read and Scarparolo 2010). These are only a few examples of the many similar programs being developed globally.

The recently established (2009) Department of Invertebrates and Amphibians in Leningrad Zoo (St. Petersburg, Russia) has developed an amphibian collection of over 80 species. Their *ex situ* programs focus on the reproduction of Asiatic amphibians and has succeeded in reproducing and raising to adulthood over 10 amphibian species, including such rare and threatened species as *Paramesotriton laoensis*, *Rhacophorus feae*, *R. orlovi*, *R. annamensis*, *Theloderma* spp., American species of Dendrobatidae, and several amphibian species of former USSR territories (e.g., *Bombina variegata*; Bagaturov 2011a, b). This work is supported through collaboration

Figure 5. Fea's tree frog (*Rhacophorus feae*) from SE Asia, possibly the largest species of tree frog in the world. Found in high montane forests and recently captive bred for the first time at Leningrad Zoo. *Image by Mikhail F. Bagaturov*.

with the Department of Ornithology and Herpetology of the Zoological Institute of the Russian Academy of Sciences.

Leningrad Zoo also works with cooperative *in situ* programs for the reintroduction of the regionally threatened Great crested newt (*Triturus cristatus*). The Moscow Zoo and institutions from the Republic of Georgia support CBPs for the endangered, Caucasian parsley frog (*Pelodytes caucasicus*), and the breeding and rehabitation of other anuran and Caudata species, including *N. kaiseri*, as well as *Megophrys nasutus*, *Tylototriton* spp., and *Cynops* spp. (M. Bagaturov, pers. comm.)

Exhibition design for amphibians (Kreger and Mench 1995; Swanagan 2000) has not received a high

Figure 6. Visitor experience. An interactive educational amphibian exhibit at St. Petersburg Zoo, Russian Federation, not only informs, but also provides tactility to increase fun and experience retention. *Image by Mikhail Bagaturov.*

research priority (Hurme et al. 2003; Quiguango-Ubillús and Coloma 2008). Amphibian CBPs offer new possibilities for the scope of amphibian displays through using critically endangered species as examples of both amphibian biology and of conservation needs. The Internet is ideally suited to exchanging the information needed to create the most effective displays for threatened species.

The exhibition of amphibians arranged in some zoos (e.g., amphibian exhibition in Leningrad Zoo consists of over 30 species of Caudata and Anuran species) accompanied by information desks displaying their biology, reproduction, decline, and how the public may contribute to their conservation. Terraria with amphibians that are decorated in a natural way serve not only the role of attractive exhibitions for visitors but also to display the amphibian's natural habitat (Bagaturov 2011a, b). These and other educational materials make major contributions to the conservation conscience of the zoo's visitors, especially with children.

Direct academic supervision can be very beneficial to amphibian CBPs. Nordens Ark, Sweden, has maintained a foundation that supports amphibian CBPs of threatened species as part of a progressive scientific society with close contacts to universities. Nordens Ark also appointed an academic conservation biologist as scientific leader so that science could inform, management, and implement successful strategies. This initiative has resulted in successful CBPs including reintroduction for the Green toad (*Pseudepidalea viridis*) and the Fire-bellied toad (*Bombina bombina*). Research projects that include undergraduate students from neighboring universities are also proving popular by providing students with a direct, hand's on approach to supporting conservation (Innes 2006).

There are considerable cultural, intellectual, and funding benefits from collaborations for amphibian research between zoos and other institutions, including increased animal welfare, scientific status, conservation commitment, display, and education (Benirschke 1996). Broad cultural collaborations can also increase the impact of exhibitions and educational programs, funding opportunities, as well as providing mutually beneficial intellectual scrutiny and stimulation (Benirschke 1996). Funding bodies can encourage the promotion of projects for both education and the inspiration of future scientists and conservationists (Anderson et al. 2008). CBPs with amphibians have provided many successful research collaborations between zoos, universities, and other entities. For examples, Chester Zoo has many valuable international research collaborations in their CBPs (Chester Zoo 2010).

Collaborations between zoos and private collectors offer a major opportunity to increase the conservation support for many threatened amphibians (Hassapakis 1997). The numbers of species successfully reproduced by private breeders far outweighs those in zoos, and many popular species are now semi-domesticated, including threatened species of anurans and salamanders (Janzen 2010). Caecilians have received less attention, although several aquatic species are bred by private collectors and some zoos (Riga Zoo). Durrell Wildlife Conservation Trust has been involved in a successful joint project with private breeders for the conservation of the Sardinian brook salamander (*Euproctus platycephalus*) using husbandry guidelines developed from private experience. Similarly, the husbandry guidelines for the two critically endangered Iranian newts, the Kurdistan newt (*Neurergus microspilotus*; Browne et al. 2009) and Loristan newt (*N. kaiseri*), were largely developed through the experience of private breeders. Many other species, including some now successfully kept in zoos, these examples of CBPs were formerly bred and distributed via private researchers. Consequently, it is important to not underestimate the contribution of private keepers to amphibian CBP's and to encourage collaboration with private keepers and their organizations wherever possible.

Anderson et al. (2010) conducted a 57-part questionnaire with 210 professionals at AZA zoos and aquariums that were involved in research programs. Support from the chief executive officer and specialized personnel employed to conduct scientific programs were judged as the two most important factors contributing to success. Successful collaboration between zoos and academic institutions required recognition of their different research emphasis. Zoos tend to focus research on animal welfare, conservation, display, and education, while academic institutions focus on description, experimentation, modeling, and specific aspects of animal biology and behavior. Mainly referring to mammals and birds, Fernandez and Timberlake (2008) showed that the main fields of collaboration between zoos and universities were the control and analysis of behavior, conservation and propagation of species, and the education of students and the general public. The latter two are particularly important to amphibian CBPs.

Formal collaboration between institutions can be established by Memorandums of Understanding (MOU), and these should clarify objectives, outcomes, responsibilities, finances, and authorship (Fernandez and Timberlake 2008; Anderson et al. 2010). Innes (2006) considered that many zoos needed an improved communication network between direct research outcomes and animal management.

Scientific knowledge generated from minimally invasive research is more likely to make its way into zoo husbandry and veterinary procedures and provide favorable publicity. Minimally invasive practices can lead to the development of innovative research methods that expand rather than restrict research potential. For instance, noninvasive molecular techniques improve our knowledge of population genetics (Moritz 2008), and assays of hormones improve reproduction and health (Goncharov et al. 1989; Browne et al. 2006; Iimori et al. 2005). Similarly, information systems and databases for amphibian conservation provide the opportunity for extensive analysis of existing data (Melbourne and Hastings 2008), and noninvasive methods such as ultrasound, X-ray, thermal, and photographic digital imaging can address many unsolved research questions. For instance, Nashville Zoo at Grassmere is using ultrasound to determine the reproductive status of the American giant salamander (*C. alleganiensis*) in both their *ex situ* and *in situ* conservation program (D. McGinnity pers. comm.).

Conclusions

Conservation resources for amphibians in many zoos are still largely devoted to display and education and not translated into significant conservation outcomes for specific threatened species. Greater support for conservation can be achieved by zoos also adopting CBPs for threatened amphibian species. Amphibian CBPs and research in zoos can include both *in situ* and *ex situ* components of and preferably should be conducted in concert with in range institutions and programs. Amphibians are ideal subjects for zoo-based research because of the economical provision of their facilities and husbandry and their relatively low maintenance under a variety of research and display conditions. Direct benefits to zoos of amphibian CBPs include the ability to maintain genetically significant numbers, the provision of competent individuals for rehabitation, supplementation, or translocation, the relatively low cost of amphibian research, education, and display, and opportunities for increased outreach and collaboration.

The primary goals of amphibian research in zoos are improved husbandry, health, reproduction, and the perpetuation of genetic variation. Zoos can also provide amphibians to other institutions, such as universities, for conservation-based studies. Research is particularly productive when integrated into CBPs with species that are novel to husbandry, which can then provide significant scientific discoveries. These activities can strengthen all segments of the conservation network between zoos, captive breeding populations, field research, and habitat preservation.

A scientific program with administrative support and dedicated facilities will attract qualified candidates for research and education positions. To maximize the productivity and quality of "Zoo-based amphibian research and conservation" qualified researchers with academic affiliations should be employed. Within this framework, institutions can design a science-based management structure for research that is tailored to their institutional capacity and amphibian collection (Hutchins 1988).

Amphibian research in zoos offers opportunities to form research collaborations with universities and other institutions, both regionally and internationally (Fernandez and Timberlake 2008; Lawson et al. 2008). Through their capacity for fund raising, grants, organizational capacity, and academic affiliations, zoos can develop projects of international stature through CBPs for threatened species (Lawson et al. 2008; Reid et al. 2008). Amphibian research in zoos can offer students and young conservation scientist's attractive opportunities to participate directly in amphibian welfare and to directly contribute to amphibian conservation through research projects of short duration (Kleiman 1996).

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ROBERT BROWNE has worked as an investment manager, builder, design draftsman, video producer, professional photographer and he has now found his true vocation, Conservation Biologist and Collaborative Researcher.

Robert has completed an Honour's degree in Aquaculture at the Key Center for Aquaculture, Australia, and then obtained a Ph.D. (1998) in Conservation Biology from the University of Newcastle, Australia.

Robert's science employment has included consultancy with biotechnology corporations and in response to the global biodiversity conservation crisis has focused on amphibian conservation and sustainability. Working with zoos in Australia, the USA, Europe, and as Research Officer for the IUCN has led Robert to work with collaborative conservation programs in the USA, Peoples Republic of China, Australia, Russian Federation, Islamic Republic of Iran, and Cameroon.

Robert has experience in a wide range of research fields supporting herpetological conservation and environmental sustainability. He has published in the scientific fields of nutrition, pathology, larval growth and development, husbandry, thermobiology, reproduction technologies, and facility design.

Robert's Ph.D. in the late 1990s was seminal to the development of gene banking to preserve genetic diversity of threatened species. Since then his research with reproduction technologies has led to major advances in the use of hormones to promote amphibian reproduction. This was responsible for the first use of artificial fertilization, to produce tadpoles for release, of the critically endangered amphibian, the Wyoming toad (*Bufo baxteri*). These techniques have since been adopted for a number of other critically endangered amphibian species. Robert's recent collaborative work with Nashville Zoo at Grassmere, USA, and international organizations on the North American giant salamander (*Cryptobranchus alleganiensis*), commonly known as the Hellbender, has fostered the development of the first genetically representative gene bank for any amphibian.

KATJA WOLFRAM focused her undergraduate studies on marine biology, zoology, and genetics and graduated with a Diplom in biology at Bremen University, Germany. In her graduation thesis, she addressed population genetics as well as physiology, and genetics, of the respiratory pigment in the Common European cuttlefish *Sepia officinalis*. Currently, she is completing her Ph.D., thesis at Antwerp Zoo's Centre for Research and Conservation (Antwerp, Belgium), researching the genetic background of mate choice in the Eurasian black vulture, *Aegypius monachus*, a species of conservation concern.

MIKHAIL F. BAGATUROV formerly a professional lawyer, was always a wild fauna collector and researcher traveling to the Middle Asia, Caucasus, Crimea, Siberia, Baltic region, Carpathians, and most of the former USSR territories with exception of the Russian Far East. An exotic animal keeper and breeder all his life Mikhail now works at the Leningrad Zoo (Saint Petersburg, Russia) as a zootechnist in the Department of Insectarium and Amphibians.

Mikhail is a member of the Russian Nikolsky's Herpetological Society at Russian Academy of Sciences and has been a terrarium animal keeper for over 30 years (one of the most experienced animal keepers in the former USSR).

In 2009, Mikhail began contributing to programs of study on the biodiversity of herpetofauna in Vietnam under the auspices of the Department of Herpetology, Zoological Institute of the Russian Academy of Sciences, St. Petersburg, Russia (Profs. Profs. Natalia Ananjeva and Nikolai Orlov).

Since 2010, Mikhail has been a member of Conservation Breeding Specialist Group (CBSG), Species Survival Commission (SSC), International Union for Conservation of Nature (IUCN), which is dedicated to saving threatened species by increasing the effectiveness of conservation efforts worldwide.

Since 2011, Mikhail had been a member of IUCN/SSC Amphibian Specialist Group (ASG).

While a large part of Mik's work is with amphibians and reptiles, he is also working on developing techniques for captive management of a variety of invertebrate groups with special focus on Theraphosid spiders (Tarantulas). Mikhail is further working on international programs on invertebrate husbandry and conservation under the guidance of the Terrestrial Invertebrates Advisory Group, European Association of Zoos and Aquariums (TITAG-Europe).

Mikhail has present plans to start a Ph.D. program at the Department of Herpetology, Zoological Institute, Russian Academy of Sciences, with research focusing on the reproductive biology of amphibians.

GERARDO GARCÍA was born in Barcelona (Spain) and has been Head of the Herpetology Department at Durrell Wildlife Conservation Trust, based in Jersey, United Kingdom (UK), since 2003. His herpetological career began at Barcelona Zoo in 1992 becoming involved in the early years of the Recovery Programme for the Mallorcan midwife toad (*Baleaphryne muletensis*) and at the Science Museum of Barcelona (CosmoCaixa) up until 1996, when he moved for work to Thoiry Zoo (Paris, France).

Gerardo's work with amphibians since 1992 has involved captive breeding programs of reptiles and amphibians in several institutions, linking *ex situ* with *in situ* conservation in Jersey (*Rana dalmatina*, *Bufo bufo*), Montserrat/Dominica (*Leptodactylus fallax*), Madagascar (*Erymnochelys madagascariensis*, *Pyxis planicauda*, *Astrochelys yniphora*), Spain (*Alytes obstetricans*, *Rana iberica*), and Mauritius (*Nactus coindemirensis*, *Gongylomorphus fontenayi* sp.). During the last few years he has been involved in various training initiatives for amphibians around the world (France, Germany, Sweden, Spain, South Africa, Mexico, Madagascar, India, Sri Lanka, Colombia, Venezuela, Montserrat, and Dominica), improving the husbandry protocols of captive colonies and diverse *in situ* programs such as the Montserrat mountain chicken frogs, genus *Alytes* and *Rana* in Spain and the amphibians of Jersey.

Gerardo completed a Ph.D. at the Institute of Conservation and Ecology (DICE), University of Kent on the "Ecology, human impact, and conservation of the Madagascan side-necked turtle (*Erymnochelys madagascariensis*) at Ankarafantsika National Park," where he lived for two years during his data collection and field work in Madagascar. Gerardo analyzed his data and began to write his thesis at the Laboratoire des Reptiles et Amphibiens, Muséum d'Histoire Naturelle of Paris, moving to Jersey in 2001.

Gerardo has been actively involved in the European Association of Zoos and Aquariums (EAZA) as chair of the Amphibian Taxon Advisory Group (ATAG) and vice-chair for the Reptile Taxon Advisory Group (RTAG). His major goal is to bring *in situ* conservation and research for these programs into the core activities of the EAZA. Gerardo was actively involved in the development of the amphibian campaign for the *Year of the Frog 2008* and co-directed the first amphibian conservation courses in Europe for Zoos and Aquariums in 2006 and 2008.

Gerardo also takes a great interest in raising the profile of the herpetological programs within both specialist groups and the general public. In his spare time, he assists affiliate zoological institutions in the development of their animal collections, design exhibits, and off show facilities for reptiles and amphibians, and in the development of new conservation programs.

ZJEF J. J. M. PEREBOOM is head of the Center for Research and Conservation and coordinator of Behavioral Research, Royal Zoological Society of Antwerp, Antwerp, Belgium. His research interests include behavioral and evolutionary ecology of primates, birds, and social insects, and the ethology of zoo animals with a link to conservation biology and animal welfare. Zjef is particularly interested in sexual selection processes and how they affect e.g., captive breeding programmes in particular, and population management measures in general.