

The Dynamics of Re-introduced Kihansi Spray Toad *Nectophrynoides asperginis* and other Amphibians in Kihansi Gorge, Udzungwa Mountains, Tanzania

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Abstract

The dynamics of the reintroduced Kihansi spray toad *Nectophrynoides asperginis* and other amphibian species that occur in the Kihansi gorge, Udzungwa Mountains were studied. The area is a home for the endemic toad that became extinct before individuals bred in captivity were reintroduced. Previous studies on the biology of amphibians conducted in the gorge focused on a few aspects but none has dealt with the dynamics of both the reintroduced Kihansi spray toad (KST) and other amphibians. Information on species richness and the abundance of amphibian species occurring in the gorge have not been dealt with in depth. Time-constrained audio-visual encounter surveys were conducted between February 2015 and May 2017 in the gorge to assess detectability rates of the reintroduced toad and other amphibian species. Sixteen species were detected including the reintroduced Kihansi spray toad. The detection rate of the KST was highest shortly after reintroduction and decreased precipitously during subsequent surveys. The results suggest low survivorship of the reintroduced KST. The detection rates of other amphibian species varied between species, although there was no significant variation in the detection rates between different sampling periods. A few species were encountered once, whereas others were encountered throughout the study period. Detection rates were influenced by the species lifestyle and weather conditions. Further studies are recommended to assess the possible causes of low survivorship of the reintroduced toads.

Keywords: Kihansi gorge, Kihansi Spray Toad, Reintroduction, Survivorship

Introduction

Kihansi gorge is located along Kihansi River in the southern tip of the Udzungwa Mountains, Tanzania. The Udzungwa Mountains form part of the Eastern Arc chain of isolated blocks of mountains stretching from Taita Hills in south eastern Kenya to south central Tanzania in the Udzungwa scarp. The eastern slopes of these mountains are covered by evergreen forests that are renowned for their extraordinary numbers of endemic species of amphibians and other taxa (Burgess et al. 1998, 2007, Channing et al. 2006, Menegon et al. 2008). Tanzania is the third highest amphibian species rich country in Africa after Democratic Republic of Congo and Cameroon with at least

206 species (AmphibiaWeb 2019), the Eastern Arc Mountains (EAM) being the main contributing area for numbers of species. Udzungwa scarp forest reserve in the south eastern slopes of Udzungwa has at least 36 amphibian species (Menegon and Salvidio 2005) and overall, the Udzungwa Mountains have 61 species (Lawson and Collett 2011) making them the amphibian richest EAM block.

Sixteen amphibian species belonging to nine families occur in Kihansi gorge area based on the list compiled from the literature. These include *Arthroleptis stenodactylus* Pfeffer 1893, *Arthroleptis xenodactyloides* (Hewitt 1933) and *Leptopelis uluguruensis* Barbour

and Loveridge 1928 (Family Arthroleptidae), *Sclerophrys maculatus* (Hallowell 1854), *Mertensophryne micranotis* (Loveridge 1925), *Nectophrynoides asperginis* Poynton, Howell Clarke and Lovett 1998 and *Nectophrynoides tornieri* (Roux 1906) (Family Bufonidae), *Afrivalus fornasini* (Bianconi 1849), *Hyperolius mitchelli* Loveridge 1953 and *H. substriatus* Ahl 1931 (formally treated as *Hyperolius puncticulatus* by other workers including Channing and Howell (2006) but *H. substriatus* appears to be the earliest available name (Pickersgill 2007) (Family Hyperoliidae). Other species that occur in the gorge include *Athroleptides yakusini* Channing, Moyer and Howell 2002 (Family Petropedetidae), *Probreviceps macrodactylus* (Nieden 1926) (Family Brevicipitidae), *Xenopus muelleri* (Peters 1844) (Family Pipidae), *Ptychadena anchietae* (Bocage 1868) (Family Ptychadenidae), *Amietia nutti* (Boulenger 1896) (Family Pyxicephalidae) and *Chiromantis xerampelina* Peters 1954 Family Rhacophoridae (Howell et al. 1997, 1998, Howell 1999, Channing et al. 2006, Lee et al. 2006, Ngalason et al. 2016). Only one amphibian species, Kihansi spray toad *N. asperginis* is endemic to the gorge and a few other species including *L. uluguruensis*, *N. tornieri*, *A. yakusini* and *P. macrodactylus* are near endemic to the Udzugwa Mountains.

Kihansi gorge is characterized by a series of waterfalls and rapids such that the activity of the flowing water on the falls and rapids generated a unique microclimate for unique biodiversity. The main upper falls at the head of the Kihansi gorge are over 100 metres high; this with an average water flow of 16 m³/s made the river falls a potential site for generation of hydroelectric power (Poynton et al. 1998). In 1993, the government of Tanzania approved the Lower Kihansi Hydroelectric Power Project (LKHP) resulting to the construction of a 180 MW hydroelectric power facility in the Kihansi River at the head of the Kihansi gorge (Lee et al. 2006). In December 1999, this facility commenced the production of power (Lee et al. 2006) leading to the

diversion of the water flowing down to the Kihansi gorge.

Long term environmental assessment and monitoring programme for flora and fauna of the Kihansi gorge were initiated in 1995 shortly after the approval of the Lower Kihansi Hydroelectric Project (LKHP). The programme aimed at investigating the possible long term impacts of the hydropower project on animals, plants and the environment. Some parameters were chosen for assessments and monitoring; these are contained in the detailed ecological monitoring programme for the Lower Kihansi Gorge Ecosystem (LKEMP 2006). Amphibians were among the parameters chosen for the assessments and monitoring due to their sensitivity to habitat alterations. It was during the assessments the Kihansi spray toad *Nectophrynoides asperginis* was first discovered in 1996 and described in 1998 (Poynton et al. 1998). This toad was found only in the Kihansi gorge in a small area of wetlands that received significant amounts of sprays. The species was listed as Critically Endangered species following the International Union for Conservation of Nature and Natural Resources (IUCN) categories and criteria for assessing the conservation status of species. It was also placed in CITES (Convention on International Trade of Endangered Species of Wild Fauna and Flora) Appendix I (Poynton et al. 1998).

Historically, the Kihansi spray toad (KST) was abundant in the gorge with a population of approximately 17,000 individuals (Lee et al. 2006). The construction of the Kihansi hydroelectric power plant and the diversion of water flowing into the Kihansi gorge led to a dramatic reduction in natural sprays and alterations of microclimate in the gorge (Lee et al. 2006). Because of the desiccation of the habitats in the gorge, the numbers of the KST and other amphibian species declined. Following the decline in the numbers of toads in the gorge, the government of Tanzania in collaboration with the Wildlife Conservation Society initiated captive breeding efforts in 2000, to provide a safety net against the

extinction of the species (Lee et al. 2006). This led to the transportation of 499 individual Kihansi spray toads from the gorge to Bronx Zoo, New York in the United States of America (USA) for captive breeding efforts (Lee et al. 2006, Channing et al. 2006). The toads were taken to the USA zoos because at that time there was no existing captive breeding facility for amphibians in Tanzania.

The establishment of captive breeding colony of KST in the USA zoos was accompanied with the installation of artificial sprinkler systems in the Kihansi gorge wetlands between 2000 and 2001. The installation of the artificial sprinkler system was an attempt to restore the desiccated habitats in the gorge. The system greatly improved the wetland habitats leading to the recovering of the KST and other amphibians (NORPLAN 2002). In the late 2003, a sharp decline in the number of KST in the spray wetlands was observed (Weldon 2004). The outbreak of chytridiomycosis, a fungal disease caused by chytrid fungus *Batrachochytrium dendrobatidis* probably had a role in the decline in the numbers of the toads in the gorge. Chytrid fungus is cited as one of the major contributors to the decline of most amphibian species worldwide (Kielgast et al. 2010, Penner et al. 2013). Other factors that can lead to the decline of amphibians include habitat destruction, alterations and fragmentation (Penner et al. 2013). In 2009, the Kihansi spray toad was officially declared extinct in the wild by the IUCN (Channing et al. 2009). The only surviving individuals of Kihansi spray toads were those in captivity. Currently, *Nectophrynoides asperginis* is considered extinct in the wild (Channing et al. 2009, IUCN 2015) because no self-sustaining population is existing.

The genus *Nectophrynoides* has other twelve described species, all found in the Eastern Arc Mountains of Tanzania. These include *N. cryptus*, *N. frontierei*, *N. laevis*, *N. laticeps*, *N. minutus*, *N. paulae*, *N. poyntoni*, *N. pseudotornieri*, *N. tornieri*, *N. vestergaardi*, *N. viviparus* and *N. wendyae*. The toads in the

genus *Nectophrynoides* are ovoviviparous, the females retain eggs in the oviducts but with no maternal nourishment until they give birth to young ones. The eggs are fertilized in the oviducts and the toadlets are born as miniature, grey versions of the adults (Channing et al. 2006). All the species in the genus but *N. asperginis* are found in forests mainly in the leaf litter. *N. asperginis* is exceptional in that, it is the only toad in its genus that depends on sprays of water in the form of mists for its survival.

Successes in captive breeding of the KST in the USA zoos paved the way for the establishment of such a facility in Tanzania with the intention of reintroducing the toads back to their natural environment in the Kihansi gorge. In 2010, a captive colony of Kihansi spray toad from the USA zoos was established in Tanzania at the University of Dar es Salaam captive breeding facility. At present, captive bred colonies of the KST are maintained at several captive breeding facilities including Bronx and Toledo zoos in the USA, at the University of Dar es Salaam, and at the Kihansi captive facility at Kihansi-Morogoro, Tanzania.

A team of conservationists that met in Dar es Salaam in 2010 developed a plan and guidelines for the re-introduction of the Kihansi spray toad to its natural habitats in the Kihansi gorge (Khatibu et al. 2011). This plan led to the reintroduction of the first batch of 2433 captive bred toads in the spray wetlands in the Kihansi gorge on 30th October 2012. Successive re-introduction of captive bred toads in the spray wetlands were conducted on 10th March 2013 (1500 toads), 18th February 2015 (1483 toads), 3rd July 2016 (696 toads), 28th October 2016 (892 toads), 19th November 2016 (105 toads), 11th December 2016 (969 toads) and 15th May 2017 (589 toads). As of May 2017, a total of 8,669 captive bred toads had been released in the Kihansi gorge wetlands.

Few studies have been conducted on the amphibians of the southern Udzungwa Mountains, e.g., Menegon and Salvadio (2005).

The Kihansi gorge has received little attention, and the published studies that exist including Poynton et al. (1998), Channing et al. (2006) have focused on a few aspects of the biology of amphibians. No detailed study exists on the population dynamics of the amphibian species in the gorge following the alterations of the habitats caused by the diversion of water, and the installation of artificial sprinklers in the spray wetlands. In addition, many individuals of the captive bred Kihansi spray toads were re-introduced in the spray wetlands in the gorge but there is no existing published information on the survivorship of these individuals.

The present study assesses the amphibian species population status in the gorge by comparing detection rates during different sampling periods. The study further assesses the dynamics of the individuals of the captive bred Kihansi spray toads that were re-introduced in the spray wetlands. The information generated will form a basis for informed management and conservation of Kihansi spray toad, other amphibians and their environment in the Kihansi gorge.

Materials and methods

Location of the study area and sites

This study was conducted in Kihansi gorge, Udzungwa Mountains, Tanzania. The gorge is located in Kihansi River (Figure 1). This gorge is about 4 km long and 0.5 km wide with steep sides (Lee et al. 2006). It lies between 300 m and 1150 m a.s.l.; and is located between latitudes 8° 34' and 8° 37' south and longitudes 35° 49' and 35° 51' east (Figure 1). There are five major wetlands in the gorge, including the main falls spray wetlands (MFSW), the upper spray wetland (USW), the lower spray wetland (LSW), Mhalala spray wetland (MSW) and the mid-gorge spray wetland (MGSW) (Figure 1). Artificial sprinklers (Plate 1) were installed in the upper spray wetlands, the lower spray wetlands and in the mid-gorge spray wetlands to imitate the natural sprays that were cut off following the diversion of the water flowing to the gorge for hydroelectric power production.

The wetlands are surrounded by steep sides and forests. The climate in the Udzungwa Mountains is influenced by monsoon winds from the Indian Ocean, causing seasonality in rainfall. The area receives the long rains from March to May, the short rains between October and December, and the driest periods are experienced between June and September. The rainfall in the eastern parts of the mountains is estimated at 2000 mm per year and decreases towards the west to about 800 – 1000 mm (Dinesen et al. 2001).

Sampling technique

Several sampling methods are commonly used to study amphibian communities. These include live-trapping methods using funnel and/or pitfall traps typically associated with a fixed drift fence, transects, plot or quadrat method, time-constrained search and cover boards (Grant et al. 1992, Heyer et al. 1994). In this study, time-constrained audio-visual surveys were used to assess the population status of the re-introduced KST and other amphibians in the Kihansi gorge. This method is one of the standard methods for estimating the relative abundance of amphibians in an area, and is an appropriate technique for both inventory, and monitoring studies (Crump and Scott 1994, Heyer et al. 1994). The results obtained are measured against the time spent in the search (Crump and Scott 1994). Time-constrained audio-visual survey method was selected because it is easy to conduct, causes relatively little disturbance to the fragile spray wetlands, and the amphibians can easily be detected with less efforts. In addition, the results obtained are comparable over long periods of time. All spray wetlands were surveyed by walking along the artificial sprinkler lines, and the surrounding habitats were surveyed by walking in a transect that ran parallel to Kihansi River. The transect ran from the upper to the mid gorge spray wetland approximately 100 metres from Kihansi river.

The surveys were conducted both during the day (between 09:00 and 11:00 hrs), and at night (between 19:30 and 21:30 hours). The

surveys were conducted in February 2015 after the re-introduction of the third batch of 1483 captive bred Kihansi spray toads in the gorge, May 2015 (two months after re-introduction), December 2015 (9 months after re-introduction), April 2016 (one year after re-introduction) and March 2017 after further releases of 696 toads in the upper spray wetland. The toads re-introduced in October 2016, November 2016, December 2016 and May 2017 were released in experimental enclosures constructed in the upper spray wetland. This study did not cover census conducted in experimental enclosures established in the spray wetlands. The results of experimental enclosures are contained in Msuya and Mohamed (2018). All the amphibian species encountered outside the experimental enclosures during the surveys were identified and counted.

To prevent the possibility of spreading pathogens in the wetlands including chytrid

fungus, the footwear and equipment were sterilized following the Weldon (2004) sterilization procedures. Footwear and equipment were first rinsed in water and then immersed in a sodium hypochlorite (NaOCl) solution (100 mg/L) for at least 2 minutes (Weldon 2004). A study conducted by Cashins et al. (2008) showed that treatment of chytrid fungus *Batrachochytrium dendrobatidis* with 6 to 12% NaOCl solution is sufficient to destroy the fungi and prevent contamination. Sterilization was done upon both entering and leaving the wetlands. Laboratory gloves were worn whenever there was a need to handle the toads. The amphibians found during the study were identified by using the amphibian guide books of Channing and Howell (2006) and Harper et al. (2010). The taxonomy of amphibians followed that of Frost et al. (2006), Pickersgill (2007), Barej et al. (2014) and Channing et al. (2016).

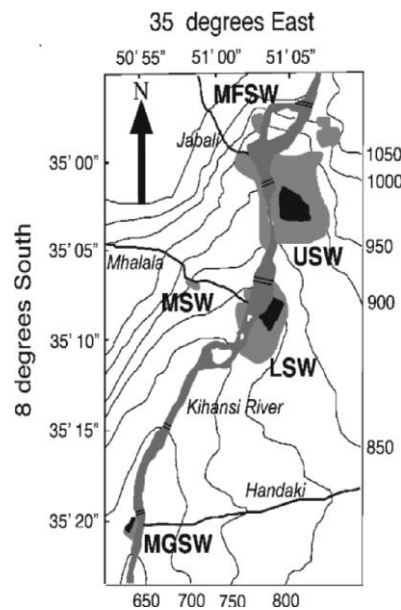


Figure 1: Location of Kihansi gorge and spray wetlands in the Kihansi River. MFSW = Main Falls Spray Wetland, USW = Upper Spray Wetland, LSW = Lower Spray Wetland, MGSW = Mid gorge Spray Wetland and MSW = Mhalala Spray Wetland (After Channing et al. 2006).



Plate 1: An artificial sprinkler system in the lower spray wetland, Kihansi gorge (© W. Ngalason, 2nd May 2015).

Analysis

The results of the surveys were presented as the number of individuals of amphibians counted per sampling effort. The sampling effort was expressed as the time spent searching in hours multiplied by the number of persons involved. The method allowed comparison of species richness and relative abundance of amphibian species between different sampling periods, and the results were expressed as detection rates. The detection rates for different amphibian species were compared using a non-parametric Kruskal-Wallis test to test for any significant difference in the detection rates between different species and sampling periods. The statistical analyses were performed using Graphpad package (GraphPad Software Inc, USA) and set to alpha = 0.05.

Results

Amphibian species richness in the gorge

During the study thirteen amphibian species were detected in the Kihansi gorge. These include *Arthroleptis stenodactylus*, *Arthroleptis xenodactyloides*, and *Leptopelis uluguruensis* (Family Arthroleptidae); *Mertensophryne micranotis*, *Nectophrynoides asperginis*, and

Nectophrynoides tornieri (Family Bufonidae); *Afrixalus fornasini*, *Hyperolius mitchelli*, and *Hyperolius substriatus* (Family Hyperoliidae); *Arthroleptides yakusini* (Family Petropedetidae); *Xenopus muelleri* (Family Pipidae); *Ptychadena anchietae* (Family Ptychadenidae) and *Amietia nutti* (Family Pyxicephalidae) (Plates 2 - 14).

These species were found in different habitat types in the gorge. A few species were restricted to certain habitat types, whereas others were widely distributed in the gorge. *Arthroleptis stenodactylus*, *A. xenodactyloides* and *Mertensophryne micranotis* were found in the leaf litter, *Arthroleptides yakusini*, *Nectophrynoides asperginis*, *Xenopus muelleri*, *Ptychadena anchietae*, *Afrixalus fornasini*, *Hyperolius mitchelli*, *H. substriatus*, and *Amietia nutti* were found in the wetlands, *Nectophrynoides tornieri* was found at the edge of the spray wetlands, and *Leptopelis uluguruensis* was found on vegetation overhanging wetlands, and in the surrounding forest. Some males of the leaf litter frog *A. stenodactylus* were observed vocalising on the vegetation one foot above the ground (Plate 15).

The Kihansi spray toad was found in the upper spray wetland only. This is the site where this captive bred toad was reintroduced in 2012 and the consecutive years up to 2017. Other sites in the gorge where the individuals of Kihansi spray toads were released include at Mhalala spray wetland, and at the lower spray wetland. At Mhalala the individuals were released only once in October 2012, and at the lower spray wetlands the toads were housed in experimental cages (Mohamed 2015). No individuals of the KST were found at Mhalala, and at the lower spray wetlands.

Relative abundance

Of the species detected during the study period, some were found only once, including *Amietia nutti*, *Hyperolius substriatus*, *Mertensophryne micranotis* and *Xenopus muelleri*, whereas others were detected throughout the study period. Species that were detected throughout include *Nectophrynoides asperginis*, *Arthroleptis stenodactylus*, *Arthroleptis xenodactyloides*, *Leptopelis uluguruensis*, and *Arthroleptides yakusini* (Table 1). The detection rates varied from one species to another as shown in Table 1. The most abundant species in the gorge was the re-

introduced species *N. asperginis* with a detection rate of 7.98 toads per person-hour. The detection rate was highest shortly after reintroduction and decreased subsequently. The trend showed a decrease in the numbers of toads detected from 28 toads/person-hour in February 2015 to 10 toads/person-hour in May 2015 to 0.42 toads/person-hour in March 2017 (Figure 2). The results of toad counts obtained 24 hours after the re-introduction of 1483 individuals in February 2015 showed that only 20% of the released individuals were detected.

The second most abundant species was the frog *H. mitchelli* with a detection rate of 0.52 frogs/person-hour followed by *A. yakusini* (0.49 frogs/person-hour), *A. stenodactylus* (0.43 frogs/person-hour), *A. xenodactyloides* (0.3 frogs/person-hour) and *L. uluguruensis* (0.25 frogs/person-hour) (Table 1). The least abundant species were *A. nutti* and *X. muelleri* with a detection rate of 0.01 frogs/person-hour each (Table 1). These two latter species were spotted once during the long rains season in April 2016. Generally, there was no significant difference in the detection rates of the amphibian species encountered during different sampling periods (Kruskal-Wallis H = 6.613, $p = 0.0853$).

Table 1: Detection rates of amphibian species in the Kihansi gorge, Udzungwa Mountains.

Species	Detection rates (frogs/toads per person-hour of searching)					Average
	Feb. 2015	May 2015	Dec. 2015	April 2016	March 2017	
<i>Amietia nutti</i>	–	0.05	0	0	0	0.01
<i>Arthroleptis stenodactylus</i>	–	0.11	1.28	0.32	0	0.43
<i>Arthroleptis xenodactyloides</i>	–	0.38	0.33	0.48	0	0.3
<i>Leptopelis uluguruensis</i>	–	0.05	0.83	0.1	0	0.25
<i>Hyperolius mitchelli</i>	–	0.92	0	0.5	0.64	0.52
<i>Hyperolius substriatus</i>	–	0	0.11	0	0	0.03
<i>Hyperolius sp.</i>	–	0.07	0	0.02	0	0.02
<i>Mertensophryne micranotis</i>	–	0.09	0	0	0	0.02
<i>Nectophrynoides tornieri</i>	–	0.05	0	0.05	0	0.03
<i>Nectophrynoides asperginis</i>	28	10	1	0.5	0.42	7.98
<i>Arthroleptides yakusini</i>	–	0.56	0.89	0.17	0.33	0.49
<i>Ptychadena anchietae</i>	–	0.14	0.28	0	0	0.12
<i>Xenopus muelleri</i>	–	0.02	0	0	0	0.01

– = surveys not conducted

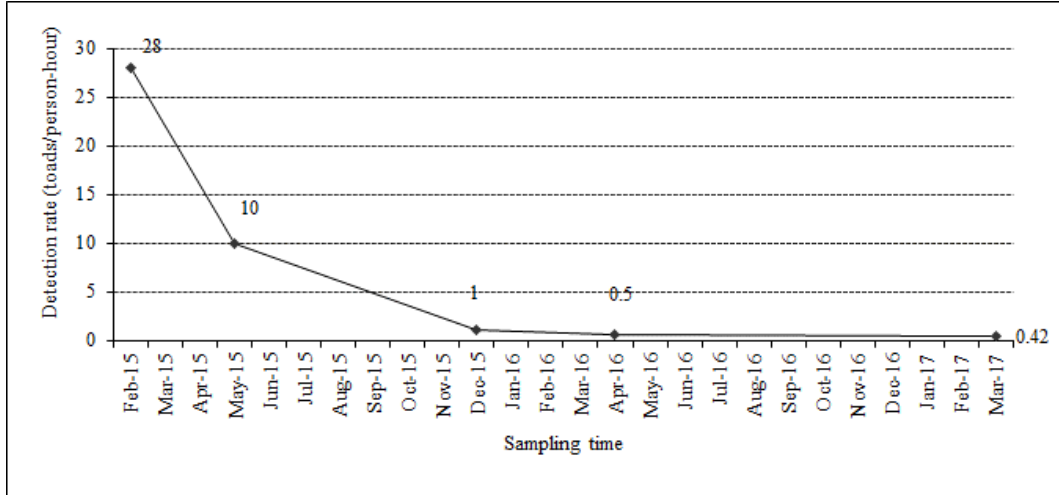


Figure 2: Detection rates of *Nectophrynoides asperginis* in the wetlands in the Kihansi gorge.



Plate 2: *Nectophrynoides asperginis*



Plate 3: *Nectophrynoides tornieri*



Plate 4: *Arthroleptis xenodactyloides*



Plate 5: *Arthroleptis stenodactylus*



Plate 6: *Arthroleptides yakusini*



Plate 7: *Leptopelis uluguruensis*



Plate 8: *Hyperolius mitchelli*



Plate 9: *Hyperolius* sp.



Plate 10: *Hyperolius substriatus*



Plate 11: *Amietia nutti*



Plate 12: *Ptychadena anchietae*



Plate 13: *Mertensophryne micranotis*



Plate 14: *Xenopus muelleri*



Plate 15: *Arthroleptis stenodactylus* on a woody plant

Plates 2 - 15: Some of the amphibian species detected in the Kihansi gorge, Udzungwa Mountains (W. Ngalason, 2015).

Discussion

Species richness

During this study, thirteen species of amphibians were recorded in the Kihansi gorge. Of these, only one species (the Kihansi spray toad *N. asperginis*) is endemic to the gorge. Individuals of this species were reintroduced in the gorge from captive breeding facilities following the extinction of the species in the wild in 2009. The species *N. tornieri*, *L. uluguruensis*, *A. yakusini* and *P. macrodactylus* are endemic to Tanzania in the Eastern Arc Mountains. Other amphibian species that were found in the gorge during the surveys are widely distributed in the forests and the lowland areas of Tanzania including the tree frogs in the genus *Hyperolius*, the leaf litter frogs in the genus *Arthroleptis*, the river frog *A. nutti*, the ridged frog *P. anchietae* and the clawed frog *X. muelleri* (Channing and Howell 2006). The river frog *A. nutti* was formerly grouped with *A. angolensis* before the revision of the genus *Amietia* which separated the species (Channing et al. 2016). *A. angolensis* is now limited to the south-western Africa in Angola. Other amphibian species in the genus, including *A. moyerorum* and *A. tenuoplicata*, are known to occur in the Udzungwa Mountains (Channing et al. 2016), and could possibly occur in the surrounding areas of Kihansi gorge. The East Africa torrent frogs *Arthroleptides* were formerly placed in the genus *Petropedetes* (Scott 2005) before

they were grouped with the genus *Arthroleptides* (Barej et al. 2014). The species *A. yakusini* is endemic to Uluguru, Udzungwa and Mahenge Mountains (Channing and Howell 2006, Menegon et al. 2011).

A few amphibian species that were previously known to occur in the Kihansi gorge were not recorded during the present study. These include *Sclerophrys maculatus* (Family Bufonidae), *Probreviceps macrodactylus* (Family Brevicipitae) and *Chiromantis xerampelina* (Family Rhacophoridae). Reasons for failure to detect the species may include the method of study that was used. Audio-visual encounter survey method can adequately detect active and non-fossorial species. Some amphibian species including *Probreviceps macrodactylus* are leaf litter frogs and or live in underground chambers (Channing and Howell 2006). Most burrowing species emerge from hiding sites mainly during rainy periods. This is due to the fact that terrestrial movements in most amphibians is positively influenced by weather conditions including rainfall and temperature (Vitt and Caldwell 2014). Most leaf litter and fossorial amphibians can be easily obtained by setting pitfall traps, digging underground and searching in leaf litter and substrates, the methods that were not used during this study.

In addition, the species previously recorded in the gorge but not in this study, including the foam-nest frog *Chiromantis xerampelina*, were

thought to have been introduced accidentally during the construction work of installation of artificial sprinklers in the wetlands (NORPLAN 2002). It could be that the species failed to re-establish in the wetlands due to unfavourable environmental conditions. The foam-nest frog is adapted to live in savanna areas (Channing and Howell 2006) rather than in forests. The study conducted by Menegon and Salvidio (2005) in the southern Udzungwa scarp forest also did not record this species, suggesting that it is not a forest dependent species. Other species including *Amietia nutti* and *Xenopus muelleri* were detected only once during the study period (Table 1) suggesting that these species occur in low density in the gorge.

Relative abundance

The Kihansi spray toad had on average higher detection rate than other amphibian species in the gorge. This is due to the fact that hundreds of individuals of the toad were previously reintroduced in the gorge. The study showed the numbers of individuals detected during subsequent surveys decreased (Figure 2). These observations suggest low survivorship of the captive bred toads reintroduced in the spray wetlands in the Kihansi gorge. The reasons for low survivorship could not be established, but factors such as adaptation, diseases, parasites, predation and food availability may have contributed. The toads released in the gorge were bred in captivity where the environmental conditions, and food were controlled. It is likely that the toads had some difficulties coping with the natural conditions. Challenges individual toads could encounter in their wild environment

include difficulties in locating food, avoiding predators, finding suitable sites and finding suitable mates. Other amphibian reintroduction programmes elsewhere have encountered problems such as desiccation, predation and competition (Griffiths et al. 2008). In the gorge there are potential predators such as snakes and other amphibian species (Ngalason et al. 2016). Factors that previously caused the extinction of the Kihansi spray toad including amphibian diseases, water quality and microclimatic conditions might have also contributed to the low survivorship. The findings obtained in this study coincide with those of Msuya (2013) and Tuberville et al. (2014) which showed the numbers of re-introduced toads that survived decreased precipitously after the reintroduction (Figure 3).

The findings presented in Table 1 showed that some amphibian species were detected once, e.g., the river frog *Amietia nutti*, whereas others were detected throughout the study period, e.g., the torrent frog *Arthroleptides yakusini*. Amphibian detectability is affected by a number of factors including their lifestyle and their density (Johana et al. 2016). Species such as *A. nutti* and *X. muelleri* spend most of their time in water bodies (Channing and Howell 2006), and others occur in low density in the study area, thus were difficult locating by visual encounter method. In this study, there was no significant difference in the detectability of the amphibians between different sampling periods, probably because the surveys were conducted during the rainy seasons. Studies show that rainfall has strong influence on the activity and detectability of amphibians (Ocock et al. 2016).

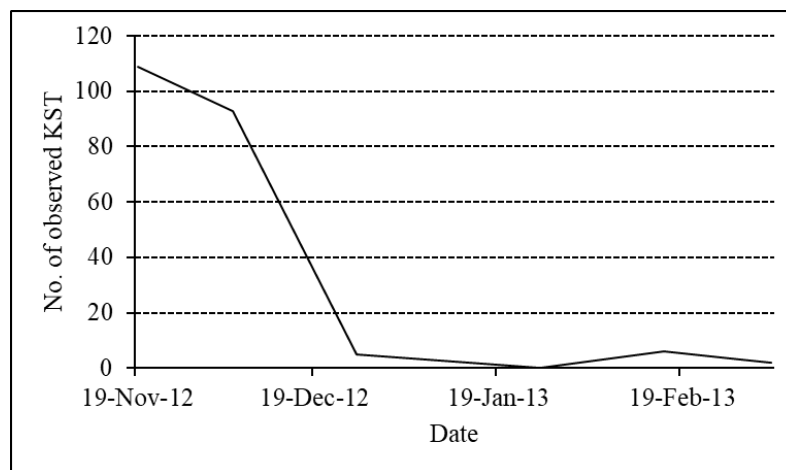


Figure 3: The number of Kihansi spray toads counted in the gorge shortly after the first re-introduction in October 2012.

Conclusion and recommendations

This study assessed amphibian species richness and detectability rates during different sampling periods in the Kihansi gorge. Sixteen amphibian species were detected, one of the species (the Kihansi spray toad *Nectophrynoides asperginis*) is endemic to the gorge, and four others are near endemic. The toad is currently considered extinct in the wild. The toads surviving in the gorge were the individuals reintroduced from captivity. The study showed the decrease in the numbers of individuals reintroduced suggesting low survivorship. Some amphibian species were detected once whereas others were detected throughout the sampling periods. Differences in amphibian lifestyles may have affected the detectability rates. Further studies are recommended to assess the possible causes of low survivorship of the reintroduced captive bred toads.

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References

- AmphibiaWeb 2019; <https://amphibiaweb.org> University of California, Berkeley, CA, USA. Accessed 7 Aug 2019.
- Barej MF, Rödel M, Loader SP, Menegon M, Gonwouo NL, Penner J, Gvoždík V, Günther R, Bell RC, Nagel P and Schmitz A 2014 Light shines through the spindrift – Phylogeny of African torrent frogs (Amphibia, Anura, Petropedetidae). *Molec. Phylogenet. Evolut.* 71: 261–273.
- Burgess N, Fjeldså J and Botterweg R 1998 Faunal importance of the Eastern Arc

- Mountains of Kenya and Tanzania. *J. East Afr. Nat. Hist.* 87: 37–58.
- Burgess N, Butynski TM, Cordeiro NJ, Doggart NH, Fjeldså J, Howell KM, Kilahama FB, Loader SP, Lovett JC, Mbilinyi B, Menegon M, Moyer DC, Nashanda E, Perkin A, Rovero F, Stanley WT and Stuart SN 2007 The biological importance of the Eastern Arc Mountains of Tanzania and Kenya. *Biol. Conserv.* 134: 209–231.
- Cashins SD, Skerratt L and Alford RA 2008 Sodium hypochlorite denatures the DNA of the amphibian chytrid fungus *Batrachochytrium dendrobatidis*. *Dis. Aquat. Organ.* 80: 63–67.
- Channing A and Howell KM 2006 Amphibians of East Africa. Cornell University Press, Ithaca, New York.
- Channing A, Finlow-Bates KS, Haarklau SE and Hawkes PG 2006 The biology and recent history of the critically endangered Kihansi Spray Toad *Nectophrynoides asperginis* in Tanzania. *J. East Afr. Nat. Hist.* 95(2): 117–138.
- Channing A, Dehling JM, Lötters S and Ernst R 2016 Species boundaries and taxonomy of the African river frogs (Amphibia: Pyxicephalidae: *Amietia*). *Zootaxa* 4155: 1–76.
- Channing A, Howell KM, Loader SP, Menegon M and Poynton JC 2009 *Nectophrynoides asperginis*. The IUCN Red List of Threatened Species. Version 2014.3. <www.iucnredlist.org>. Downloaded on 31 December 2019.
- Crump ML and Scott Jr NJ 1994 Visual encounter surveys. In: Heyer WR, Donnelly MA, McDiarmid RW, Hayek LG and Foster MS 1994 (eds) *Measuring and Monitoring Biological Diversity: standard methods for amphibians*. Smithsonian Institution Press, Washington and London.
- Dinesen L, Lehmsberg T, Rahner MC and Fjeldså J 2001 Conservation priorities for the forests of Udzungwa Mountains, Tanzania, based on primates, duikers and birds. *Biol. Conserv.* 99: 223–236.
- Frost DR, Grant T, Faivovich J, Bain RH, Haas A, Hadad CFB, De Sa RO, Channing A, Wilkinson M, Donnellan SC, Raxworthy CJ, Campbell JA, Blotto BL, Moler P, Drewes RC, Nussbaum RA, Lynch JD, Green DM and Wheeler WC 2006 The Amphibian Tree of Life. *Bull. Am. Museum Nat. Hist.* 2006(297): 1–291.
- Grant BW, Tucker AD, Lovich JE, Mills AM, Dixon PM and Gibbons JW 1992 The use of cover boards in estimating patterns of reptile and amphibian biodiversity. In: Siegel R and Scott N (eds) *Wildlife 2001*. Springer, Dordrecht, pp. 379–403.
- Griffiths RA, Garcia G and Oliver J 2008 Re-introduction of Mallorcan midwife toad, Mallorca Spain. In: Soorae PS (ed) *Global re-introduction perspectives: re-introduction case-studies from around the globe* (pp.54–57). IUCN/SSC Re-introduction Specialist Group, Abu Dhabi, UAE.
- Harper EB, Measey J, Patrick DA, Menegon M, Vonesh JR and Swilla 2010 Field guide to the amphibians of the Eastern Arc Mountains and Coastal Forests of Tanzania and Kenya. Camerapix Publishers International, Nairobi, Kenya.
- Heyer WR, Donnelly MA, MacDiarmid RW, Hayek LC and Foster MS 1994 Measuring and monitoring biological diversity. Standard methods for amphibians. Smithsonian Institution Press, Washington D. C.
- Howell KM 1999 Continued search for the Kihansi spray toad. Report on fieldwork carried out in March 1999. Long term environmental monitoring project. Report produced for NORPLAN AS and the Tanzania Electric Supply Company Ltd (TANESCO), Dar es Salaam, Tanzania.
- Howell KM, Msuya CA and Kihale PM 1997 Kihansi fieldwork report: 21 Oct.–10 Nov. 1997. Small mammals, amphibians and reptiles. Report prepared for NORPLAN/TANESCO, Dar es Salaam, Tanzania.

- Howell KM, Msuya CA and Kihale PM 1998 LKHP Environmental Monitoring—Small mammals, amphibians and reptiles, October 1998. Report prepared for NORPLAN/TANESCO, Dar es Salaam, Tanzania.
- IUCN SSC Amphibian Specialist Group. 2015 *Nectophrynoides asperginis*. The IUCN Red List of Threatened Species 2015: e.T54837A16935685. <http://dx.doi.org/10.2305/IUCN.UK.2015.2.RLTS.T54837A16935685.en>. Downloaded on 31st December 2019.
- Johana N, Muzzneena AM, Grismer LL and Norhayati AI 2016. Species composition, diversity and relative abundance of amphibians in forests and non-forest habitats on Langkawi Island, Peninsular Malaysia. AIP Conference Proceedings Vol. 1784, No. 1 p. 060035 doi: 10.1063/1.4966873.
- Khatibu FHA, Church DR, Moore RD, Lewis JP, McAloose D, Magurisha HB, Hanley C, Pessier A, Misinzo G, Malago JJ, Howell KM, Kalokola P, Ngalason W, Pramuk J, Newmark B, Weldon C, Odum A, Gibbs J, Mturi F, Mbuta C, Masunzu C, Sabuni F, Ferdinard F, Pilla J, Jeswald U, Manusa M, Shayo C, Mwikila D, Sylvester B, Maembe A, Kibassa J, Kihale J, Kaoneka B, O'Connor M, Soorae P and Msuya C (eds) 2011 Kihansi spray toad re-introduction guidelines. IUCN/SSC Amphibian Specialist Group.
- Kielgast J, Rödder D, Veith M and Lötters S 2010 Widespread occurrence of the amphibian chytrid fungus in Kenya. *Anim. Conserv.* 13:36–43.
- Lawson LP and Collett L 2011 Herpetofauna of montane areas of Tanzania. 1. Results from two amphibian surveys of Malundwe Mountain, Mikumi National Park. *Fieldiana Life Earth Sci.* 4: 74–80.
- Lee S, Zippel KC, Ramos L and Searle J 2006 Captive breeding program for the Kihansi Spray Toad (*Nectophrynoides asperginis*) at the Wildlife Conservation Society, Bronx, New York. *Int. Zoo Yearbook* 40(1): 241–253.
- LKEMP (Lower Kihansi Environmental Management Project) 2006 An Ecological Monitoring Program for the Lower Kihansi Gorge Ecosystem. Dar es Salaam, Tanzania.
- Menegon M and Salvidio S 2005 Amphibian and reptile diversity in the southern Udzungwa Scarp Forest Reserve, south-eastern Tanzania. In: Huber BA Sinclair BJ and Lampe KH (eds) *African Biodiversity: Molecules, Organisms, Ecosystems*. Proceedings of the 5th International Symposium on Tropical Biology. Museum Koenig, Bonn. Pp. 205–212.
- Menegon M, Doggart N and Owen N 2008 The Nguru Mountains of Tanzania, an outstanding hotspot of herpetofaunal diversity. *Acta Herpetol.* 3: 107–127.
- Menegon M, Bracebridge C, Owen N and Loader SP 2011 Herpetofauna of montane areas of Tanzania. Amphibian and reptiles of Mahenge Mountains, with comments on biogeography, diversity and conservation. *Fieldiana Life Earth Sci.* 4: 103–111.
- Mohamed N 2015 *A study on the population dynamics of re-introduced Kihansi spray toads Nectophrynoides asperginis in artificially maintained habitat at the Kihansi gorge, Kihansi, Tanzania*. MSc thesis, Department of Zoology and Wildlife Conservation, University of Dar es Salaam.
- Msuya CA 2013 Monitoring of reintroduced population of Kihansi spray toad at Kihansi gorge wetlands. Unpublished report for Lower Kihansi Environmental Management Project, Dar es Salaam, Tanzania.
- Msuya CA and Mohamed N 2018 Semi-annual report on reintroduction of the Kihansi spray toad in the Kihansi gorge wetlands by using acclimatization cages. Unpublished report for Kihansi Catchment Conservation and Management Project (KCCMP), Dar es Salaam, Tanzania.
- Ngalason W, Kalenga PP and Ugomba RS 2016 Amphibian species monitoring in the Kihansi Gorge, Southern Udzungwa

- Mountains, Tanzania. Unpublished report for Kihansi Catchment Conservation and Management Project (KCCMP), Dar es Salaam, Tanzania.
- NORPLAN 2002 Lower Kihansi Hydropower Project. Immediate Rescue and Emergency Measures (IREM). Final specialist report: amphibian studies. Unpublished report produced for the Tanzania Electric Supply Company Ltd. (TANESCO), Dar es Salaam, Tanzania.
- Ocock JF, Kingsford RT, Penman TD and Rowley JLL 2016 Amphibian abundance and detection trends during a large flood in a semi-arid floodplain wetland. *Herpetol. Conserv. Biol.* 11: 408–425.
- Penner J, Adum GB, McElroy MT, Doherty-Bone T, Hirschfeld M, Sandberger L, Weldon C, Cunningham AA, Ohst T, Wombwell E, Portik DM, Reid D, Hillers A, Ofori-Boateng C, Oduro W, Plötner J, Ohler A, Leache AD and Rödel MO 2013 West Africa - A safe haven for frogs? A Sub-continental assessment of the chytrid fungus (*Batrachochytrium dendrobatidis*). *PLoS One* 8(2): p. e56236. doi:10.1371/journal.pone.0056236.
- Pickersgill M 2007 Frog search: results of expeditions to southern and eastern Africa. Edition Chimaira Andreas S. Brahm Frankfurt am Main Germany.
- Poynton JC, Howell KM, Clarke BT and Lovett JC 1998 A Critically Endangered new species of *Nectophrynoides* (Anura: Bufonidae) from Kihansi Gorge, Udzungwa Mountains, Tanzania. *Afr. J. Herpetol.* 47: 59–67.
- Scott E 2005 A phylogeny of ranid frogs (Anura: Ranoidea: Ranidae), based on a simultaneous analysis of morphological and molecular data *Cladistics* 21: 507–574.
- Tuberville TD, Buhlmann KA and Mohammed N 2014 Unpublished final report on Kihansi spray toad re-introduction efforts during 2012-2013, including progress report for December 2012 – July 2013. A report submitted to Dr. Cuthbert Nahonyo, University of Dar es Salaam.
- Vitt LJ and Caldwell JP 2014 *Herpetology: an introductory biology of amphibians and reptiles*. 4th ed, Academic Press, San Diego, California, USA.
- Weldon C 2004 Chytridiomycosis risk assessment in the Kihansi and Udagaji gorges with special reference to Kihansi spray toad. Unpublished report for LKEMP, Dar es Salaam, Tanzania.