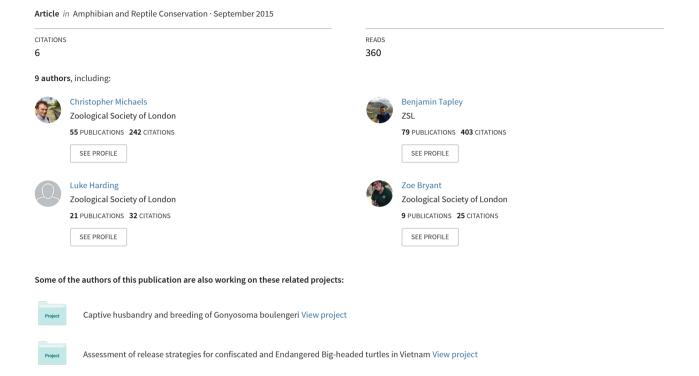
Breeding and rearing the Critically Endangered Lake Oku Clawed Frog (Xenopus longipes Loumont and Kobel 1991).





Breeding and rearing the Critically Endangered Lake Oku Clawed Frog (*Xenopus longipes* Loumont and Kobel 1991)

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Abstract.—The Lake Oku Clawed Frog Xenopus longipes is a Critically Endangered, dodecaploid anuran endemic to Lake Oku in Cameroon. An ex situ population of this species was established at Zoological Society of London (ZSL), London Zoo in 2008, as well as at several other institutions, with the intention of providing data on the biology and husbandry of this species. We report the first captive breeding of the species. Adult frogs maintained under environmental conditions designed to mimic field data produced clutches of 7–300 eggs; eggs measured 1.23 mm in diameter, and were laid singly after a period of 6.5 hours in axial amplexus. Spawning took place only during the day. Tadpoles hatched in 2–3 days and development was very long compared to congeners, lasting 193–240+ days until metamorphosis. Tadpoles grew very large (maximum 79 mm total length), particularly compared with the relatively small adult size (maximum 36 mm Snout to Vent Length [SVL]). Tadpoles proved to be highly sensitive to total dissolved solids (TDS) in the water and only thrived when low levels (20 mg/L) were used. Metamorphosis concluded with an SVL of 19–25 mm and F1 animals began first sexual activity at 5–6 months post metamorphosis. These data will inform future husbandry in captivity as well as illuminating facets of biology previously unknown and difficult to determine in the field.

Key words. Amphibian; ex situ; captive husbandry; water quality; Cameroon; West Africa; field data

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The creation of *ex situ* populations for research and conservation breeding has become an important part of the international conservation response to global amphibian declines (Browne et al. 2011; Gascon 2007; Koute et al. 2012; Wilkinson et al. 2013), which represent one of the greatest conservation challenges in history (Zippel et al. 2011). The requirements of amphibians in captivity are poorly understood and many species are presently difficult to maintain and breed (Antwis et al. 2014; Antwis and Browne 2009; Browne et al. 2006; Dugas et al. 2013; King 2011; Ogilvy et al. 2012; Verschooren et al. 2011). *Ex situ* programs have experienced difficulty in providing conditions under which animals survive (Norris 2007; Gagliardo et al. 2008) or successfully breed (Birkett et al. 1999; Gratwick 2012). Moreover, information on how to

rear tadpoles is particularly lacking in peer reviewed literature (Pryor 2014).

The Lake Oku Clawed Frog *Xenopus longipes* Loumont and Kobel 1991 (Fig. 1) is an entirely aquatic, dodecaploid frog found only in Lake Oku, a high elevation crater lake in the north west region of Cameroon. *Xenopus longipes* is classified as Critically Endangered by the IUCN (Stuart et al. 2008) due to its restricted range and therefore vulnerability to stochastic factors. Between 2006 and 2010 recurring, enigmatic *X. longipes* morbidities and mortalities were observed, but the overall impact of these events is unknown (Doherty-Bone et al. 2013). A captive-breeding program was considered vital in case of a catastrophic collapse of the population due to the potential introduction of fish to the lake as well as habi-

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tat degradation and disease threats (Tinsley and Measey 2004a). *Xenopus longipes* is ranked as the 35th global priority for amphibian conservation on the basis of threat and evolutionary history by the Zoological Society of London's Evolutionarily Distinct and Globally Endangered (EDGE) program (Isaac et al. 2012).

Captive colonies of the Critically Endangered X. longipes were established in 2008 at Antwerp Zoo (later moved to Cologne Zoo and one private breeder), Zoological Society of London (ZSL), London Zoo, and more recently in 2013, at the Steinhart Aquarium in the USA, for conservation research purposes (Browne et al. 2009; T. Ziegler pers. comm.; P. Janzen pers. comm.; D. Blackburn pers. comm.). The zoo colonies were intended to be assurance populations for conservation breeding. However, due to concerns over biosecurity and suitability of animals for release to the wild, the ZSL population was assimilated into the main collection with the focus now on conservation research aiming to document the reproductive biology of the species, as little is currently known. Such information is of importance for developing in situ conservation management strategies. Despite repeated attempts in all these institutions, however, efforts to breed and rear this species in captivity have failed, even with the use of artificial reproductive techniques (P. Janzen, pers. comm.; D. Blackburn pers. comm.).





Fig. 1. Male (top) and female (bottom) adults of *Xenopus longipes* in the collection at ZSL London Zoo (ZIMS ID 7441).

Here we report the first captive breeding success of *X. longipes* and the rearing of the tadpoles until metamorphosis.

Methods

In 2008, frogs were collected from Cameroon after consultation with local communities (Permit No. 0742/CO/MINFOF/SG/DFAP/SDVEF/SC and No. 0928/PRBS/MINFOF/SG/DFAP/SDVEF/SC). Lake Oku is considered sacred by the Oku villages and permission had to be granted before any contact with the lake could be made. Thirty-nine founders were housed at Zoological Society of London (ZSL), London Zoo.

Table 1 summarizes the initial and subsequent husbandry used for these frogs between 2008 and 2014. In 2012 the husbandry of X. longipes was reviewed (Table 1) as breeding had not occurred and the temperature regime and water parameters did not reflect conditions in the field (Table 2). Captive management should be informed by field data (Tapley and Acosta 2010; Michaels and Preziosi 2013; Michaels et al. 2014) and replicating field conditions has improved captive breeding success of X. laevis (Godfrey and Sanders 2004). In 2012, all 32 (30.2) remaining founders were sexed; males being smaller, slimmer, and having keratinized nuptial pads (Fig. 2A and C) and females possessing a trio of cloacal papillae (Fig. 2B). These features became more prominent around breeding events, but were noticeable year round. All 30 female frogs were continuously heavily gravid and amplexus was occasionally observed, but without spawning. Additional founders including four more males were imported from Cameroon in July 2012 and after completing their quarantine period were assimilated into the existing *X. longipes* colony.

In June 2013 mixed sex groups varying from 1.6 to 3.3 were transferred to a custom-made system (Fig. 3; Table 1). A new environmental regime based on longitudinal field data collected monthly from Lake Oku by Doherty Bone et al. (2013) was adopted (Tables 1 and 2; Fig. 4). Lake Oku water temperature and pH were simulated initially, and Total Dissolved Solids (TDS) was subsequently added to the parameters being replicated in 2014 (Table 1). Total Dissolved Solids was measured using Micro 800 Optical DO meter (Palintest) and pH using a Micro 600 pH meter (Palintest). The feeding regime was also modified (Table 1) and a more diverse array of food items were offered to compensate for potential dietary deficiencies as knowledge regarding the nutritional requirements of amphibians is lacking (Densmore and Green 2007).

Results

On 20.3.14, two pairs of *X. longipes* spawned naturally and without hormonal induction, followed by a number

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Table 1. Changes in enclosures, life support systems, environmental parameters and diets used for *X. longipes* between 2008 and 2014. Reproduction occurred in 2014.

Dates	Enclosure type and size	Life support systems and furnishings	Photo- period	Water parameters	Diet
2008– 2012	Acrylic aquaria (Exo Terra, Rolf C. Hagen) 20 L enclosures	Air-stream sponge filter; Plastic plants	10:14	pH c. 8.5 TDS: c. 350 mg/L Temperature: 19–22 °C	Blood worm (<i>Chironomus</i>), Nutrafin Max cichlid sinking capsules, Tetra prima granules and King British blood worm (freeze dried)
2012	Acrylic aquaria 20 L enclosures	Air-stream sponge filter; Plastic plants	10:14	pH c. 8.5 TDS: c. 350 mg/L Temperature: 18–19 °C	Mixed invertebrates: blood worm (Chironomus sp); glass worm (Chaoborus crystallinus); water fleas (Daphnia sp); hatchling crickets (Gryllus bimaculatus and G. assimilis) and worms (Eisenia sp.)
2013	48 L enclosures linked to 100 L sump	TR10 Teco chiller/heater and UV filter; External canister filter (FX6 Fluval). Plastic tubes, plastic and live plants (<i>Vallisneria</i> spp., <i>Echinodorus</i> spp.)	12:12	pH c. 7.5 TDS: c. 150 mg/L Temperature: 17–19 °C, with seasonal variation	Mixed invertebrates (as above)
2014	48 L enclosures linked to 100 L sump	TR10 Teco chiller/heater and UV filter; External canister filter (FX6 Fluval). Plastic tubes, plastic and live plants (<i>Vallisneria</i> spp., <i>Echinodorus</i> spp.)	12:12	pH c. 7.5 TDS: 20 mg/L Temperature: 17–19 °C with seasonal variation	Mixed invertebrates (as above)

of other spawning events (Table 3). In the initial spawning event a single pair in each of two tanks containing 1.2 animals spawned. Audible vocalizations, consisting of metallic clicks typical for *Xenopus* (Tinsley and Kobel 1996) were only heard very infrequently from spawning and non-spawning males and were not closely associated with spawning activity; being heard sporadically during both spawning and non-spawning periods. Amplexus and spawning behavior were only observed throughout the day, with no evidence of spawning occurring over-night. Amplexus was axial and the process of oviposition lasted 6.5 hours from initiation to termination of amplexus. Eggs, numbering seven to 300 per clutch (Table 3), were deposited singly over all available surfaces in aquaria. Egg diameter was 1.23 mm one hour after laying. Occasionally, multiple males attempted to amplex single females, but were dislodged by vigorous kicking on the part of the original male. Laying and non-laying females were observed feeding on the eggs, even during amplexus, so non-amplectant animals were removed immediately. Mating pairs were removed as soon as spawning was complete. Animals could not be individually identified, so it is unclear how many clutches were produced by individual animals. The initial spawning event occurred after increasing the temperature from 17.5 to 19.1 °C. This was done by adding warm tap water to the system, resulting in a pH shift from 7.5 to 8.09 to replicate the seasonal temperature and pH regime in Lake Oku, although being done two months earlier than this shift occurs in the field (Fig. 4). This shift occurred over a period of less than one hour after warm water was added in a single dose. However, as the breeding season is not documented in the field, there is no evidence that this seasonal

change accompanies the initiation of breeding in nature, other than this relationship observed in congeneric species (Kobel et al. 1996). Later spawning events in the following months (see Table 3) were not associated with manipulation of water parameters, but did follow heavy feeds with earthworms (Eisenia sp.). Fertility was highly variable; some clutches were almost entirely infertile, but in most cases fertility rates were close to 100%. Eggs developed and hatched in 2–4 days, with tadpoles initially clinging to hatch sites via the cement gland. Eventually eyes and pigmentation developed before becoming free swimming after 2–4 days. Free-swimming tadpoles initially congregated in areas of slow current, swimming against the water flow. Hatch rate varied between clutches, with later clutches being more consistently successful than earlier clutches.

A variety of combinations of conditions were used in attempts to rear tadpoles (see Table 4). However, we only had success by maintaining tadpoles in water with a very low TDS of 20 mg/L (measured at roughly weekly intervals) and without any live plants or accumulation of humic detritus, and only in aquaria isolated from the adult system possibly as a result of secretions from adults or toxins from PVC pipework used in the aquatic system. Mortality of tadpoles remained high until the TDS of the systems fell below 80 mg/L, with tadpoles becoming weak, opaque, and finally sinking to the floor of the aquaria before dying. Following gradual replacement of high TDS water with low TDS reverse-osmosis water, surviving tadpoles began to feed, swim normally, and to develop. Doherty-Bone et al. (2013) report a TDS of <10 mg/L (See Table 2), but our value of 20 mg/L was the lowest possible output from the RO system in use (Pen-

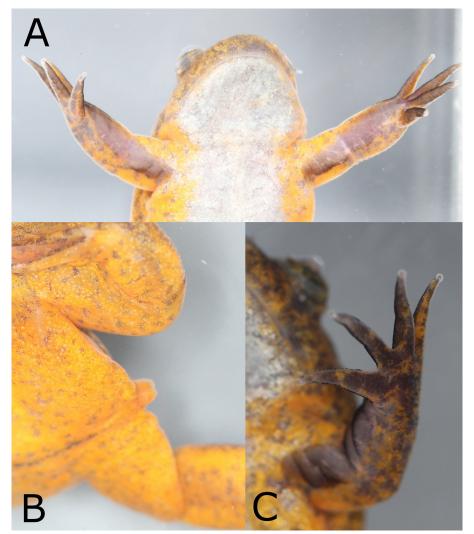


Fig. 2. Keratinized nuptial pads on the inside surfaces of the front limbs of male (A and C) and cloaca of a female *X. longipes* (B); note the cloacal papillae, which are absent in male frogs.

tair PRF; Fileder) and appears to be adequate for larval rearing.

Tadpole enclosures were glass aquaria measuring 50 × 36 × 30 cm (L × W × H) held in a temperature controlled room with water temperature at 18–20 °C. Between three and 15 tadpoles were housed per aquarium (maximum density of one tadpole per 3.6 litres). Aquaria were filtered with air-stream sponge filters set to the minimum effective flow to reduce turbulence, which would have disturbed the swimming and foraging behavior of tadpoles. Tadpoles were fed 2–4 times throughout the day on a suspension consisting initially of blanched and blended spinach or nettle, commercial *Xenopus* tadpole food, SERA Micron powdered food, and *Spirulina* alga, which was strained prior to use to remove larger plant

fragments. After several weeks, the diet was changed to only include commercial *Xenopus* tadpole food, SERA Micron (SERA), and *Spirulina* (3:1:1 by mass, suspended in water before adding to aquaria) to avoid the high oxalate content of spinach (Noonan and Savage 1999), which may interfere with calcium metabolism (Rosol et al. 1995). Food was added throughout the day dependent on the rate at which food was consumed in a given aquarium, with food density of 5.3 mg/L aquarium water provided immediately after feeding; density reduced gradually as food was consumed by tadpoles. Uneaten suspended food accumulated on the bottom of aquaria, where tadpoles were unable to consume it. Additionally, the low carbonate content of the water reduced the capacity for biological filtration. Consequently, nitrogenous

Table 2. Water temperature, pH, and TDS measured at the Lake Oku shoreline (modified from Doherty-Bone et al. 2013).

Parameter	Mean Value	±	Units
Water temperature	17.27	4.17	Celsius
рН	7.58	0.24	-
Total Dissolved Solids	8.72	2.27	Ppm



Fig. 3. Aquarium for *X. longipes*, set within a custom built, centrally filtered system (inset photograph) at ZSL London Zoo. Life support system and sump not shown – see text for details.

waste (measured using Photometer 7100 [Palintest]) was difficult to manage and tadpoles were briefly exposed to high levels of ammonia (>1 mg/L) and, later, nitrite (up to 2.4 mg/L) without mortality. A regime of 10% water changes in the morning and 50% water changes in the afternoon, both accompanied by removal of uneaten food on the bottom of tanks by siphon and thorough cleaning of sponge filters in aquarium water, helped to suppress nitrogenous waste to more acceptable, but still detectable levels (Ammonia: <0.1 mg/L; Nitrite: <0.5 mg/L) for most of the tadpole rearing period.

The tadpoles of *X. longipes* are described separately (Tapley et al. 2015). Development in the most rapidly developing tadpole (Fig. 5) lasted 193 days between hatching and metamorphosis. We report development using Gosner (1960) stages, as it was impossible to accurately apply the more detailed Nieuwkoop and Faber (1994) stages for Xenopus laevis development to live tadpoles without restraining them. This would likely have proven fatal for these delicate and Critically Endangered tadpoles, though could be employed in future offspring once captive population growth has been assured. However, developmental rates were highly variable and the more slowly developing tadpoles had not yet metamorphosed at the time of writing. A maximum total length of 68 mm was reached in the first tadpole to metamorphose (Fig. 5), and the largest tadpole reached a maximum total length of 79 mm. Metamorphs measured 19–25 mm SVL and captive-bred males began to exhibit amplexus six months post metamorphosis, by which time they had nearly reached adult size. Further details of tadpole development are provided by Tapley et al. (2015). Once front limbs emerged from the operculum, tadpoles were separated by placing them in identical systems with

Table 3. Spawning dates and clutch sizes for *X. longipes*.

F					
Date	Clutch number	Clutch size			
20.03.14	1	93			
	2	115			
21.03.14	1	190			
22.03.14	1	40–50			
	2	40–50			
05.04.14	1	40			
25.08.14	1	50			
04.09.14	1	Not counted			
16.09.14	1	20			
17.09.14	1	120			
18.09.14	1	80			
20.09.14	1	Not counted			
29.09.14	1	50			
04.10.14	1	120			
05.10.14	1	300			

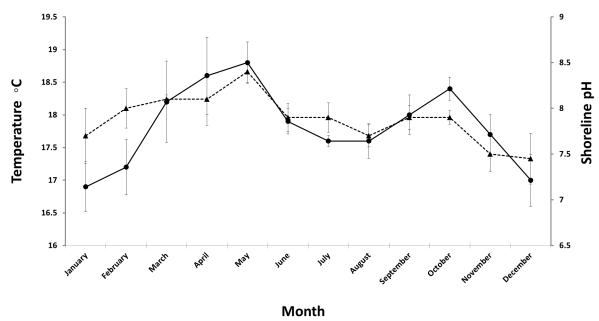


Fig. 4. Monthly water temperatures (circles) and pH (triangles) recorded from the shoreline of Lake Oku between 2008/2009 and 2013. Error bars represent SEM.

Table 4. Combinations of conditions used to rear *X. longipes* tadpoles, and the outcome in terms of tadpole survival.

Water TDS (mg/L)	Refugia (live plants)	Detritus	Lighting	Tannins	Isolated from adult system?	Tadpoles survived?
	-	-	-	-	+	+
	-	-	+	-	+	
	-	-	-	-	-	
	-	-	+	-	-	
	+	-	+	-	-	
20	+	+	+	-	-	
20	-	-	-	+	-	
	-	-	+	+	-	
	+	-	+	+	-	
	+	+	+	+	-	
	+	+	+	-	+	
	+	+	+	+	+	
	-	-	-	-	-	
	-	-	+	-	-	-
	+	-	+	-	-	
	+	+	+	-	-	
	+	+	+	+	-	
	-	-	-	+	-	
150	-	-	-	-	+	
	-	-	+	-	+	
	+	-	+	-	+	
	+	+	+	-	+	
	+	+	+	+	+	
	-	-	-	+	+	

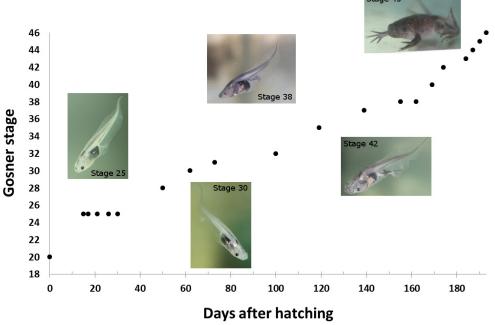


Fig. 5. Gosner stage progression of the most rapidly developing *X. longipes* tadpole. Hatching to metamorphosis took 193 days, but smaller tadpoles had only reached stage 35 by this point.

sponge filters that had been matured in the system housing the adult frogs, but with a shallower water depth of 15 cm to facilitate access to the surface for breathing. Metamorphosis from this point took around seven days to complete. Froglets fed on a similar range of prey items to adults.

Discussion

Although the husbandry of adult X. longipes is largely similar to that established for other *Xenopus* species (Green 2012), and adult frogs are able to survive a range of water parameters, the tadpoles are more sensitive. The dietary requirements of tadpoles are similar if not identical to those of X. laevis and X. tropicalis, but tadpoles appear to be more sensitive to the mineral/solute content of water. Tadpoles maintained in water with a TDS higher than around 80 ppm died rapidly and tadpoles developed well with a TDS of around 20 ppm. Total Dissolved Solids represent the total amount of dissolved mobile charged ions, including minerals, salts or metals and is closely related to hardness, but includes a broader range of dissolved substances. Typically, very low solute content of aquarium water can lead to osmotic imbalances in amphibians, but species may adapt evolutionarily to relatively pure water (Odum and Zippel 2008). Sensitivity to hardness or TDS values in *Xenopus* is not without precedent. The tadpoles of the softwater specialist X. gilli from the Cape also appears to be intolerant of hard, alkaline conditions (Rau 1978), while the reproductive success of captive X. laevis is improved by matching the hardness of their wild environment (Godfrey and Sanders 2004).

Other amphibians including the Hellbender Cryptobranchus alleganiensis have been shown to be reproductively sensitive to TDS levels (Ettling et al. 2013). The closely related X. amieti has been reproduced with hormonal induction in captivity (Xenopus Express pers. comm.), and the tadpoles of this species were maintained in hard water with success. However, there are no field data for water quality in its wild range and the larger distribution of X. amieti, which is not restricted to a single lake (Tinsley and Measey 2004b), may have led to the evolution of less specific environmental requirements. Our combinations of environmental conditions, summarized in Table 3, were not fully exhaustive and so the effects of some parameters (particularly tannins) cannot be fully elucidated based on these data. Tannins are thought to be important in reducing the frequency of fungal infections in the tadpoles of some anuran species (e.g., Theloderma corticale; Rauhaus et al. 2012), but there are no data concerning the tannin levels in Lake Oku. The forested shores of Lake Oku do produce inputs of leaf litter (so far unquantified) suggesting some levels of tannins, but this needs to be confirmed. Underwater photographs of the lake suggest relatively clear water (T. Doherty-Bone pers. obs.), which may mean that tannins are unimportant or potentially harmful in this species.

Amplexus and egg-laying behavior is similar to other *Xenopus*, although we did not observe calling in close association to spawning. Indeed, calls were very rare in general and we were unable to record them despite repeated efforts. Amplexus and oviposition were exclusively diurnal, in comparison to the often nocturnal habit of *X. laevis* (Green 2012) and the apparent strictly nocturnal amplexus, calling, and spawning reported

from hormone induced *X. amieti* (Xenopus Express pers. comm.). Specific triggers involved in stimulating spawning activity remain unclear. In a species from a habitat that is relatively stable year round (Fig. 4), and with no periods of drought and pond drying, it is possible that reproduction can take place year round and strong environmental stimuli are not required. Although initial spawning was associated with a change in temperature and pH, the breeding season is not documented in the wild and there is no evidence that this seasonal change accompanies the initiation of breeding in nature. Our observations suggest that heavy feeding may contribute to spawning activity, and so breeding may be more linked to a threshold in body condition than to external triggers. Kobel et al (1996) have suggested that some *Xenopus* species breed following first rains, when nutrients in the water have increased and secondary productivity of invertebrates is thus stimulated. Our observations indicated the initial stimuli of changing temperature, but correlated more strongly with increased availability of food. These speculations merit further investigation.

Clutch size (7–300; Table 3) was smaller than that produced by X. laevis (500–30,000 eggs Green, 2012) or X. tropicalis (1,000–3,000 eggs; Green, 2012). This may partly reflect the smaller body size of X. longipes, but may also be a function of breeding in a more stable lake system habitat, where there may be advantage in producing a smaller number of larger eggs. The fact that egg size is similar for X. laevis and X. longipes (1.3 mm [Brown 2004] and 1.23 mm, respectively), as well as for a number of other *Xenopus* species much larger than *X*. longipes (Kobel et al. 1996) supports this hypothesis. The pattern of small clutch size and relatively large eggs is continued in the very large tadpoles of this species (maximum total length 79 mm), particularly compared with adult size (32-36 mm snout-to-vent length [SVL]; Loumont and Kobel 1991); see Tapley et al. (2015). The closely related X. amieti, which has larger adults than X. longipes, has a tadpole of only 40 mm total length (Channing and Rodel 2012), while the very large X. laevis has tadpoles of 80 mm compared with adults of over 140 mm SVL (Green 2012). The metamorphs of X. longipes are correspondingly large relative to adult size, being similar in size to the metamorphs of X. laevis despite a fivefold difference in adult size between the two species (see Tapley et al. (2015), for further discussion of larval size).

Larval development was slower in *X. longipes* than congeners. Larval duration was 193 days at 17–19 °C for the fastest developing larva, in comparison to the faster development of *X. laevis* (42–56 days (Green 2012); 53 days at 18 °C; Gomez-Mestre et al. 2010) or *X. tropicalis* (21–42 days; Green 2012). Several healthy tadpoles of *X. longipes* remained untransformed at 240 days post hatching. This may, again, be linked to a relatively stable breeding habitat at higher altitude, where very low seasonal variation in environmental parameters (Fig. 4), lower temperatures, and no risk of the water body drying

out may select for a longer larval phase (Werner 1986). In *X. gilli*, which is found in more temperate lowland habitat in the extreme south of the African Cape, lower temperatures comparable with those measured in Lake Oku are also associated with the long developmental duration of this species (120 days; Rau 1978), albeit still shorter than for *X. longipes*.

The observations presented herein provide the first insight into the behavior, development, and captive requirements in X. longipes. This is of particular note as to the best of our knowledge the tadpoles of this species have never been observed alive in the field and so nothing is known of their habits in nature. In particular, the high sensitivity to mineral content and smaller clutch size of this species than in commonly maintained Xenopus may make X. longipes more susceptible to aquatic pollution and less able to recover quickly from declines. Moreover, this characteristic highlights the limitations of the "analogue species" concept (Preece 1998; Michaels et al. 2014), whereby common relatives of a threatened species are used as models to develop husbandry strategies before working with target, usually Critically Endangered, species. The relative ease of breeding and rearing X. laevis in captivity does not entirely transfer to X. longipes, particularly where water TDS for tadpoles is concerned

Our findings will hopefully improve success with this species in other institutions, and contribute to the longterm viability of captive colonies. This includes attaining reproduction from the first generation of captive bred X. longipes. Once reproduction is achieved regularly, a studbook should be developed to ensure that a viable population of this species is maintained in captivity longterm, both for conservation breeding and for research purposes. A studbook would require individual marking techniques as X. longipes do not have distinctive skin markings. Such marking techniques has not yet been trialled in this species. *Xenopus longipes* is one of only two vertebrates known to be dodecaploid (the other being X. ruwenzoriensis) and so there is considerable interest in this species as a model laboratory organism. Inclusion of X. longipes in research captive colonies may help to secure the future of this species in captivity.

Although the current captive populations of *X. lon-gipes* are not managed under strict enough biosecurity controls to be suitable for reintroduction efforts (IUCN/SSC 2014), laboratory techniques for other *Xenopus* exist to generate "clean" animals (e.g., Kay and Peng 1991). There is therefore potential to use these techniques to create biosecure cohorts that could safely be used for reintroduction should it be required. Moreover, husbandry protocols can also be distributed to Cameroonian specialists so that conservation breeding facilities can be developed in country if necessary; this option is often preferable due to reduced risk of disease transmission and reduced cost. More work is required to fully un-

derstand and control the reproduction of this species in the laboratory as well as the field.

Conclusions

Although superficially similar to other *Xenopus* species better established in captivity, the husbandry and captive breeding of *X. longipes* differs in several important aspects. The breeding triggers are poorly defined and less obvious than for many other species, which often breed in response to large water changes with cool water. Clutches are small and eggs are relatively large for the adult body size compared with other *Xenopus* species. The tadpoles are also very large and take a very long time to develop in comparison with other species. Moreover, they are highly sensitive to dissolved solids. These characters may reflect adaptation to a single volcanic lake with a stable environment.

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