

# EAZA Amphibian Taxon Advisory Group Best practice guidelines for *Potomotyphlus* and *Typhlonectes* spp. caecilians

Version 2



Benjamin Tapley<sup>1</sup>, David J. Gower<sup>2</sup>, Christopher J. Michaels<sup>1</sup>, Alberto Barbon<sup>3</sup>, Matt Goetz<sup>3</sup>, Javier Lopez<sup>4</sup>, Adam Bland<sup>4</sup>, Gerardo Garcia<sup>4</sup>, Nathaniel A. Nelson<sup>5</sup>, Mark Wilkinson<sup>2</sup>, Léa Fieschi-Méric<sup>1,6</sup>.

1. Zoological Society of London, Regent's Park, London, NW1 4RY, UK
2. Department of Life Sciences, The Natural History Museum, London SW7 5BD, UK
3. Jersey Zoo, La Profonde Rue, Trinity JE3 5BP, Jersey
4. Chester Zoo, Chester CH2 1LH, UK
5. Sedgwick County Zoo 5555 W Zoo Blvd, Wichita, KS 67212, USA
6. Genetics and Ecology of Amphibians Research Group, Laurentian University, Sudbury P3E2C6, Canada

ATAG Chair: Gerardo Garcia, Chester Zoo, United Kingdom, CH2 1LH. ATAG Vice-chairs: Olivier Marquis, Parc Zoologique de Paris, 51 avenue de Saint-Maurice, 75012 Paris, France and Benjamin Tapley Zoological Society of London, Regent's Park, London, United Kingdom.

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**Preamble**

Right from the very beginning it has been the concern of EAZA and the EEPs to encourage and promote the highest possible standards for husbandry of zoo and aquarium animals. For this reason, quite early on, EAZA developed the “Minimum Standards for the Accommodation and Care of Animals in Zoos and Aquaria”. These standards lay down general principles of animal keeping, to which the members of EAZA feel themselves committed. Above and beyond this, some countries have defined regulatory minimum standards for the keeping of individual species regarding the size and furnishings of enclosures etc., which, according to the opinion of authors, should definitely be fulfilled before allowing such animals to be kept within the area of the jurisdiction of those countries. These minimum standards are intended to determine the borderline of acceptable animal welfare. It is not permitted to fall short of these standards. How difficult it is to determine the standards, however, can be seen in the fact that minimum standards vary from country to country.

Above and beyond this, specialists of the EEPs and TAGs have undertaken the considerable task of laying down guidelines for keeping individual animal species. Whilst some aspects of husbandry reported in the guidelines will define minimum standards, in general, these guidelines are not to be understood as minimum requirements; they represent best practice. As such the EAZA Best Practice Guidelines for keeping animals intend rather to describe the desirable design of enclosures and prerequisites for animal keeping that are, according to the present state of knowledge, considered as being optimal for each species. They intend above all to indicate how enclosures should be designed and what conditions should be fulfilled for the optimal care of individual species.

**Cover image:** *Typhlonectes natans*.

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## Introduction

The information in this Best Practice Guideline has come from a variety of sources including a literature review, the experience of the authors and others in the captive husbandry of *Typhlonectes* spp. and *Potomotyphlus* caecilians; a caecilian husbandry questionnaire that involved both zoological collections, aquariums and keepers from the private sector as well as direct observations of the species in the field.

In this new version, we incorporate the most recent knowledge gathered from the scientific literature published since the release of the original EAZA BPG in 2019. The physiology of *Typhlonectes* caecilians has continued to be broadly investigated, with a special emphasis on their reproductive system and hormonal cycles. A new interest in caecilian' molecular genetics has emerged, with a substantial use of transcriptomics. Advances have been made regarding skin diseases; however, this area needs higher research prioritisation in view of the current context of emerging pathogens that decimate some amphibian populations worldwide. Finally, the knowledge and representation gap among different species within the Typhlonectidae Family is widening: recent scientific publications and *ex-situ* collections are only associated with two Genera out of the five the Family comprises.

Amphibian husbandry is a rapidly evolving field and there are many aspects that require further research. The exact breeding triggers for aquatic caecilians are unknown and further research would be beneficial. Susceptibility to diverse skin diseases needs to be further investigated, to allow the development of appropriate treatments. Lethal dermatitis has recently been reported in captivity, and although the literature suggests that at least some aquatic caecilians are resistant to chytridiomycosis, the epidemiology of other skin diseases requires further research.

### Key husbandry points

1. The provision of appropriate seasonal temperature regimes.
2. Monitoring and management of water quality.
3. Preventing escape.

These guidelines have been reviewed and approved by the Amphibian TAG members. Unless indicated otherwise, all pictures were taken by the authors.

Table of contents

<b>SECTION 1 : BIOLOGY AND FIELD DATA</b> .....	<b>5</b>
1.1. Taxonomy .....	5
1.2. Morphology .....	6
1.2.1. Length .....	6
1.2.2. Weight .....	6
1.2.3. Colouration .....	7
1.2.4. Description .....	7
1.3. Physiology .....	9
1.4. Longevity .....	10
1.5. Zoogeography, ecology and conservation .....	10
1.5.1. Distribution .....	10
1.5.2. Habitat .....	12
1.5.3. Conservation status and population .....	14
1.6. Diet and feeding behaviour .....	15
1.6.1. Food preference .....	15
1.6.2. Feeding behaviour .....	15
1.7. Reproduction .....	15
1.7.1. Developmental stages to sexual maturity .....	15
1.7.2. Age of sexual maturity .....	17
1.7.3. Seasonality of cycling .....	17
1.7.4. Gestation period .....	18
1.7.5. Number of offspring .....	18
1.7.6. Birth details and season .....	18
1.8. Behaviour .....	19
1.8.1. Activity .....	19
1.8.2. Locomotion .....	19
1.8.3. Predation .....	19
1.8.4. Vocalisation .....	19
1.8.5. Social behaviour .....	19
1.8.6. Sexual behaviour .....	20
1.9. Evolutionary research .....	20
<b>SECTION 2 : BIOLOGY AND FIELD DATA</b> .....	<b>21</b>
2.1. Enclosure .....	21
2.1.1. Substrate .....	21
2.1.2. Furnishing and maintenance .....	21
2.1.3. Water .....	22
2.1.4. Temperature .....	23
2.1.5. Filtration .....	23
2.1.6. pH .....	23
2.1.7. Hardness and alkalinity.....	24
2.1.8. Dissolved oxygen .....	24
2.1.9. Lighting .....	24
2.2. Feeding .....	25
2.2.1. Basic diet .....	25
2.2.2. Special dietary requirements .....	25
2.2.3. Method of feeding .....	26
2.3. Social structure .....	26
2.3.1. Changing social structure .....	26
2.3.2. Sharing enclosure with other species .....	26
2.4. Breeding .....	27
2.4.1. Mating .....	27
2.4.2. Gestation period .....	28
2.4.3. Birth .....	28
2.4.4. Development and care of young .....	30
2.4.5. Population management .....	30
2.5. Behavioural enrichment .....	31
2.6. Handling .....	31
2.6.1. Individual identification and sexing .....	31
2.6.2. Catching / restraining .....	32
2.6.3. Transportation .....	33
2.6.4. Safety .....	33
<b>2.7. Veterinary care and specific medical problems: considerations for health and welfare</b> .....	<b>33</b>
2.7.1. Clinical anatomy and physiology .....	33
2.7.2. Physical examination and diagnostics .....	34
2.7.3. Diseases .....	36
2.7.4. Therapeutics .....	38
2.7.5. Anaesthesia and euthanasia .....	39
2.7.6. Surgery .....	41
2.7.7. Recommended research .....	44
Acknowledgements .....	44
References .....	45
Appendix: Post-mortem of female <i>T. natans</i> .....	52

## SECTION 1. BIOLOGY AND FIELD DATA

## 1.1 TAXONOMY

The Neotropical caecilian Family Typhlonectidae includes 14 currently recognised species in five Genera (Wilkinson *et al.*, 2011). Most of these species are quite poorly known, except for three of them, which, along with the giant lungless *Atretochoana eiselti*, seem to form a group of the four most aquatic species in the Family. *A. eiselti* is rarely encountered in the wild and has not been maintained in captivity. Therefore, the mention of “aquatic caecilians” throughout this document is used to refer to this group of three caecilian, distributed along the Genera *Typhlonectes* and *Potomotyphlus* (Table 1).

<b>ORDER: Gymnophiona (Müller, 1832)</b>
<b>FAMILY: Typhlonectidae (Taylor, 1968)</b>
<b>GENUS: <i>Atretochoana</i> (Nussbaum &amp; Wilkinson, 1995)</b> <i>A. eiselti</i> (Taylor, 1968), Eistelt’s caecilian, Atretochoan of Eistelt
<b>GENUS: <i>Chthonerpeton</i> (Peters, 1880)</b> <i>C. arii</i> (Cascon & Lima-Verde, 1994) <i>C. braestrupi</i> (Taylor, 1968), Braestrup’s caecilian <i>C. exile</i> (Nussbaum & Wilkinson, 1987), Bahia caecilian <i>C. indistinctum</i> (Reinhardt & Lütken, 1862), Argentine caecilian <i>C. noctinectes</i> (Silva, Britto-Pereira & Caramaschi, 2003) <i>C. onorei</i> (Nussbaum, 1986), El Reventador caecilian <i>C. perissodus</i> (Nussbaum & Wilkinson, 1987), Minas Gerais caecilian <i>C. tremembe</i> (Maciel, Meite, Silva-Leite, Leite & Cascon, 2015) <i>C. viviparum</i> (Parker & Wettstein, 1929), Santa Catarina caecilian
<b>GENUS: <i>Nectocaecilia</i> (Taylor, 1968)</b> <i>N. petersii</i> (Boulanger, 1882), Upper Amazon caecilian
<b>GENUS: <i>Potomotyphlus</i> (Taylor, 1968)</b> , alternatively spelled <i>Potamotyphlus</i> ★ <i>P. kaupii</i> (Bethold, 1859), Kaup’s caecilian
<b>GENUS: <i>Typhlonectes</i> (Peters, 1880)</b> , Rubber Eel, Caecilian worm, Aquatic caecilian ★ <i>T. compressicauda</i> (Duméril & Bibron, 1841), Cayenne caecilian, le molle (French Guiana). ★ <i>T. natans</i> (Fischer, In Peters, 1880), Rio Cauca caecilian, Anguilla

Table 1. List of the recognised taxa in the Typhlonectidae Family. The focus species of this document, referred to as “aquatic caecilians” are marked by a blue star.

The species *T. cunhai*, although listed by the IUCN (Trefaut Rodrigues *et al.*, 2004), is here considered a synonym of *T. compressicauda* as recommended by Frost (Frost, 2021).



Figure 1. Adult female *Typhlonectes natans*



## 1.2 MORPHOLOGY

Substantial comparative studies of the morphology of aquatic caecilians were provided by Fuhrman (1914) and Wilkinson and Nussbaum (1997).

### 1.2.1 LENGTH (ADULT)

<i>T. natans</i>		250–725mm total length (Duellman & Trueb, 1994).
<i>T. compressicauda</i>	Females	285–800mm total length (Moodie, 1978).
	Males	262–585mm total length (Moodie, 1978).
<i>P. kaupii</i>	Up to	695mm total length (Taylor, 1968).



Figure 2 (left). Adult *Potomotyphlus kaupii*. Figure 3 (right). Highly vascularised skin of *P. kaupii*.

### 1.2.2 WEIGHT

Weights from wild adult *T. natans* of undetermined reproductive status captured in November 2008 (Guarinócito, Colombia) are presented in Table 2. In captivity *T. natans* may double their weight prior to giving birth; Kowalski (2001) reports an adult female *T. natans* weighing 250.0g prior to giving birth. On average captive female *T. natans* were 46% lighter after giving birth at Durrell Wildlife Conservation Trust.

Sex	Total length (mm)	Weight (g)
Female	434.0	52.6
Female	439.0	55.5
Female	413.0	51.1
Female	637.0	172.2
Female	415.0	57.3
Male	374.0	37.9

Table 2. Morphometrics of *Typhlonectes natans* captured in November 2008, Guarinócito, Colombia (Tapley & Acosta, 2010).

There are no published records of the weight of wild *T. compressicauda* or *P. kaupii*. The weights of captive *P. kaupii* at ZSL London Zoo ranged from 60.4–113.6g.

### 1.2.3 COLOURATION

*T. natans* are typically dark purplish/bluish-grey with a paler ventral surface (Fig. 1) and *T. compressicauda* are typically dark (sometimes blackish) grey to dark brown with a slightly paler ventral surface. *T. natans* often appear slightly glossier and with more clearly demarcated annular grooves than the often more velvety appearing *T. compressicauda*. Biofluorescence, widespread amongst amphibians, was recently shown to occur in *T. natans*: when lit with blue light, their skin mucous-like secretions and cloacal region fluoresce in green. This localized biofluorescence in the cloacal region could maybe serve as a visual signal for reproduction (Lamb & Davis, 2020).

*P. kaupii* (Figs. 2 & 3) are dark grey-brown to pale lilac-grey with darker, purplish annular grooves.

### 1.2.4 DESCRIPTION

Like all caecilians, *Typhlonectes* and *Potomotyphlus* are legless and elongate, but unlike other caecilians, they have a longitudinal dorsal fold or fin. Superficially, their bodies are divided into transverse rings, or annuli, separated by annular folds, or grooves. These increase the resemblance between terrestrial caecilians and worms, but are more or less well indicated, and mostly are interrupted by the fin in aquatic caecilians, making them less worm and more eel-like. The eyes are small and covered with more or less translucent skin and the mouth is recessed (below a prominent snout). Terrestrial caecilians have a (probably chemo- and mechanosensory) tentacle on each side of the head, very slightly protruding from an aperture behind and slightly below the small external nostril. In aquatic caecilians, there is a tentacular aperture directly behind the distinctively subtriangular nostrils and the (presumably chemosensory) tentacle is not protrusible and not directly visible.

Externally and superficially, it can be difficult to tell *T. natans* and *T. compressicauda* apart. *T. compressicauda* has a low, more or less prominent (even varying in life in single individuals), laterally compressed dorsal fin situated along at least the caudal third of the body, this fin is usually far less developed in *T. natans*. In aquatic caecilians, the vent is situated within a cloacal disc close to the terminus of the body (Wright & Whitaker, 2001). A true tail is absent (Taylor, 1968) although there some vertebrae posterior to the level of the vent (Wilkinson, 1989), with only a very short distance between the vent and the posterior terminus of the body. The most reliable characteristic to differentiate *T. natans* and *T. compressicauda* externally is the form of the disc surrounding the vent. The anterior half of the 'cloacal' disc is divided into 5 sections (denticulations) in *T. compressicauda* and into 4 in *T. natans* (Fig. 4 & 5). The posterior half of the cloacal disc is divided into five sections in both *T. natans* and *T. compressicauda*. Tooth crown morphology differs between *T. natans* and *T. compressicauda*, the tooth crowns of the latter are broadly dilated and this is more prominent in neonates (Fig. 6). The number of annuli, vertebrae and lung lengths may also be used to differentiate between *T. natans* and *T. compressicauda* (Wilkinson, 1991).

The external colouration alone makes separating *Typhlonectes* from *Potomotyphlus* simple. A review of *P. kaupii* was recently published (Ledesma, 2019). *P. kaupii* may possess a low, middorsal ridge as well as a dorsal fin. The head, collar and anterior of the body are smaller than in *Typhlonectes*, and they have relatively numerous narrow pointed teeth. The cloacal disc has a distinctive key-hole shape with an elongate anterior part. Unlike *Typhlonectes*, the choanae (internal nostrils) are partially sealed with a concealed aperture (Wilkinson and Nussbaum, 1997).

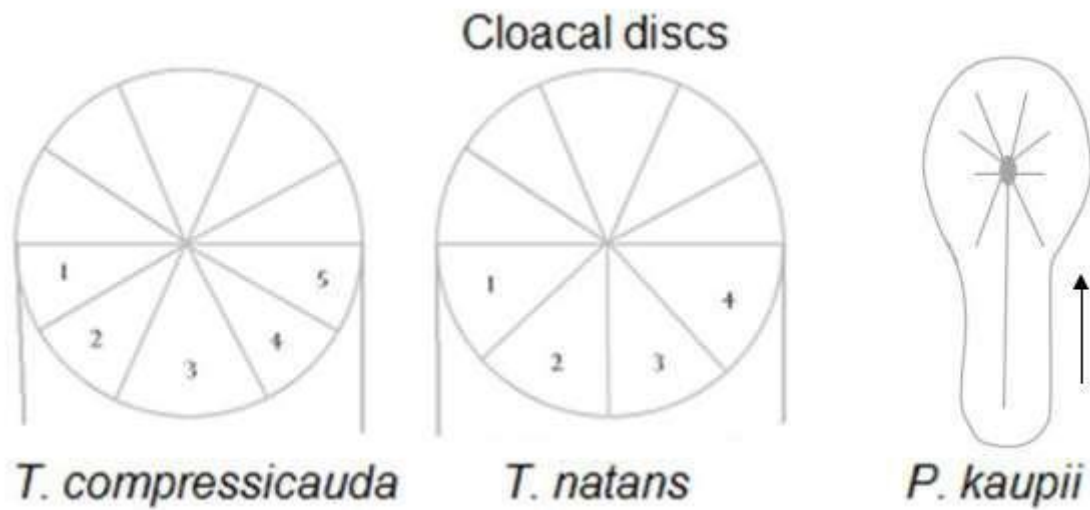


Figure 4. Cloacal discs (region surrounding the vent) showing the differences in anterior denticulations forming the vent in *Typhlonectes natans*, *T. compressicauda* and *Potomotyphlus kaupii*. Arrow points posteriorly.



Figure 5. Cloacal disc in *Typhlonectes compressicauda* © Ian Stephen (top) and *T. natans* (bottom).



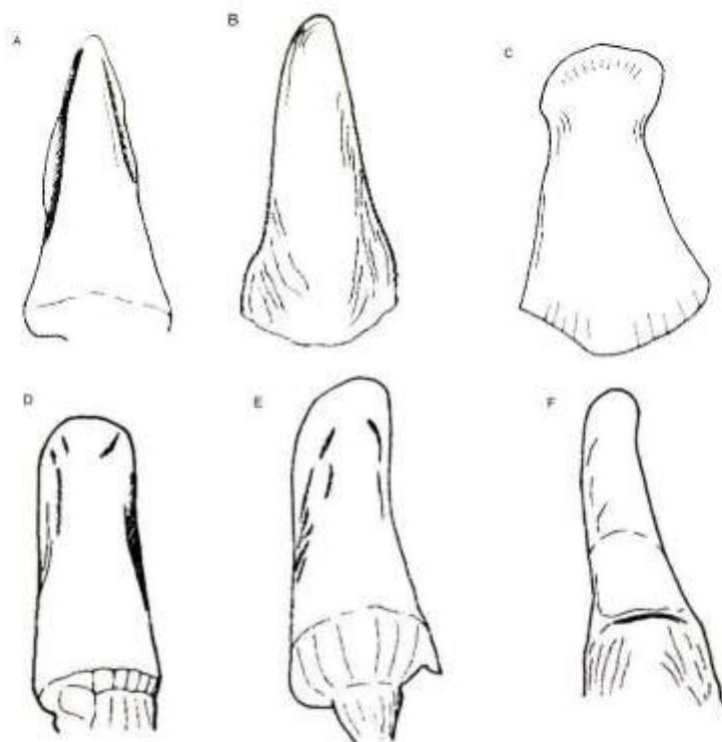


Figure 6. Dentition of (A-B) *Typhlonectes natans*, and of (C) newborn and (D-F) adult *T. compressicauda*. Taken from Wilkinson, 1991.

### 1.3 PHYSIOLOGY

#### RESPIRATORY RATE

Frequency of lung ventilation for *T. natans*  $6.33 \pm 0.84$  breaths  $\text{hr}^{-1}$  (Prabha *et al.*, 2000). Caecilians use a buccal pump, with cyclical lowering of the floor of the mouth to suck in air through their nostrils and raising of the floor of the mouth to pump this air into the lungs. Exhalation is passive and facilitated by the elasticity of the lungs and their intrinsic cartilages. Multiple iterations of the buccal pump, as many as thirty in *T. natans* (Wilkinson, 1980), are required to fill the lungs. There is surprisingly little information on the frequency of lung ventilation, and the duration and number of pulses of the buccal pump that are involved in each episode of lung ventilation.

#### GAS EXCHANGE

94% of gaseous exchange for *T. compressicauda* is derived from pulmonary respiration (Sawaya, 1947). They are capable of drowning.

In *P. kaupii* the left lung is shorter than the right and it is exceptionally broad and sac-like, lacking any respiratory epithelium. It presumably serves as a hydrostatic organ, more important in the control of buoyancy than in respiration (Wilkinson & Nussbaum, 1997). *P. kaupii* exhibit a concomitant reduction in the capacity of its buccopharyngeal force pump, associated with their relatively small heads, indicating a reduction in the importance of pulmonary gas exchange and increased reliance on cutaneous gas exchange (Wilkinson & Nussbaum, 1997). *P. kaupii* can appear pinkish in life and the skin appears to be highly vascularised (Fig. 3), this may be an adaptation due to an increased reliance on cutaneous gas exchange.

## HOMEOSTASIS

In caecilians, individuals can adapt their body size adjusts along humidity gradients to maximise hydric homeostasis. This supports the water conservation hypothesis: increasing precipitations lead to a body size reduction (Pincheira-Donoso *et al.*, 2019). The hypothalamic expression of diuretic/anti-diuretic hormones and the expression of their receptors in the kidney is in fact modulated by seasonality in *T. compressicauda*, leading to an increased diuresis during the rainy season (Yousef *et al.*, 2018).

### 1.4 LONGEVITY

The maximum recorded age for *T. natans* is 14 years, in a captive individual from Jacksonville Zoo, USA (Mendyk & Rost, 2018). *P. kaupii* obtained as adults in 2008 by Sedgwick County Zoo are still alive at the time of writing and are estimated to be at least 16 years old. Captive *T. compressicauda* can live for at least 10 years (individual still living; M. Wilkinson, pers. obs.).

## FIELD DATA

### 1.5 ZOOGEOGRAPHY / ECOLOGY

#### 1.5.1 DISTRIBUTION

*T. natans* - Cauca and Magdalena drainages of Colombia and north-eastern Venezuela (Fig. 7). Tapley & Acosta (2010) hypothesise that *T. natans* may occur further south in the Magdalena valley (High Magdalena) than Figure 7 implies: this region requires further investigation to ascertain the presence or absence of this species in the area. They also suspect that *T. natans* may occur further south to the source of the Urrá river.

One individual, likely a released or escaped pet, was also recently detected in Florida, where environmental conditions are in favour of the prospering the species. This represents the first record of caecilian in the United States (Sheehy *et al.*, 2021).

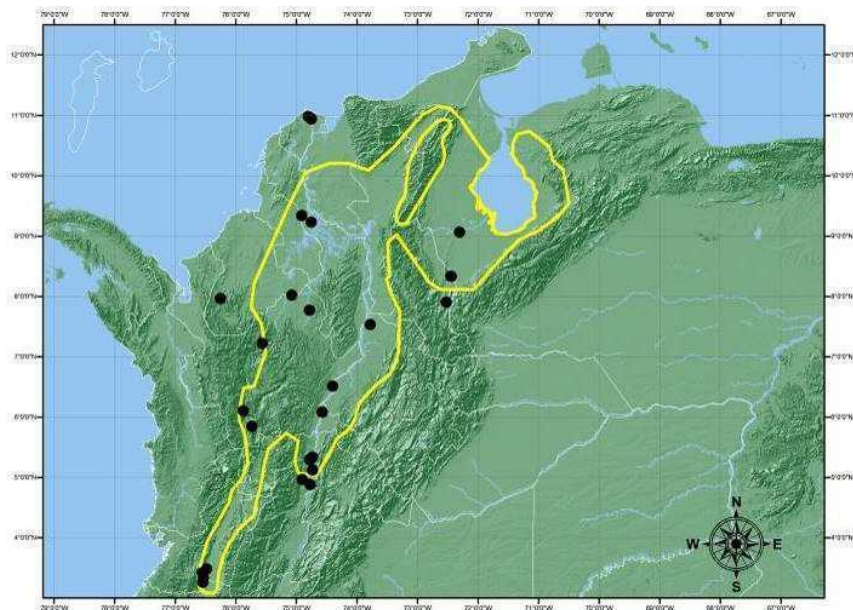


Figure 7. The native distribution of *Typhlonectes natans* after Tapley & Acosta, 2010. The yellow line corresponds its distribution according to the IUCN Red List assessment published in 2009 (Mijares *et al.* 2009), and the black spots to known localities (back in 2010). The species was reassessed in 2020 but the reassessment map still does not reflect the known range of the species (points on the Caribbean coast are missing as well the point outside of the yellow polygon to the left).

*T. compressicauda* - Rivers of the Guianan shield and the Amazon basin from the eastern coast of Brazil to the Atlantic slopes of the Peruvian and Colombian Andes (Fig. 8). Its range was recently extended to the Jari River basin in Brazil (Oliveira *et al.*, 2019).

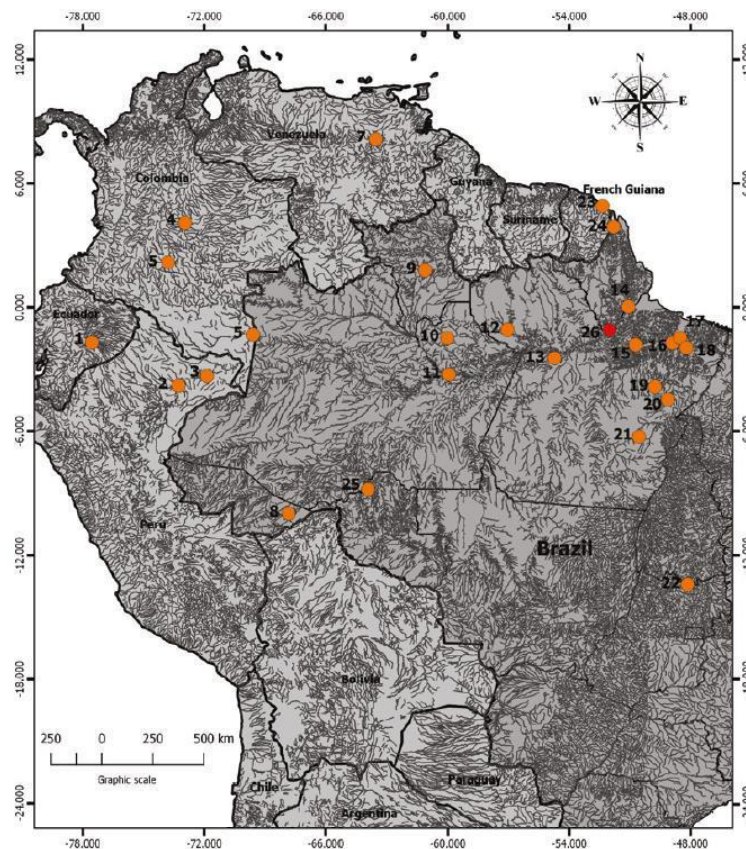


Figure 8. The distribution of *Typhlonectes compressicauda* from Oliveira *et al.*, 2019 (map courtesy of the authors, ©Oliveira). The red dots represent the new site described by the authors, and the orange dots represent sites described in prior literature.

*P. kaupii* – Distribution covering the drainage systems of the Orinoco and Amazon (Fig. 9) (Oliviera *et al.*, 2012) below 500m asl. (Wilkinson *et al.*, 2010), including the countries of Colombia, Ecuador, Peru, Venezuela, Brazil, and the territory of French Guiana. Could also be present in Suriname and Guyana (Ledesma, 2019). The most southern extent of its distributional range is poorly known (Marty *et al.*, 2007 Wilkinson *et al.*, 2010).

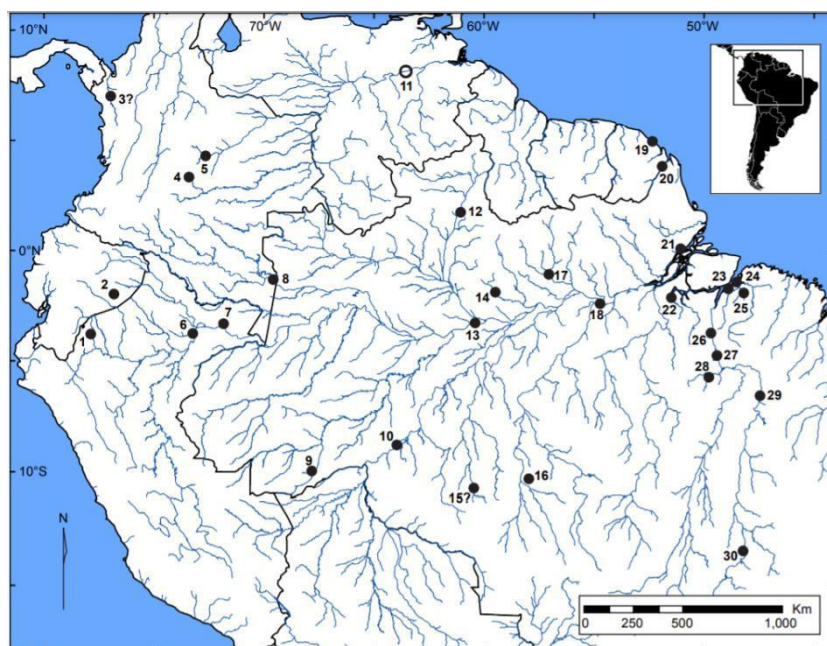


Figure 9. The distribution of *Potomotyphlus kaupii* from Ledesma 2019 (map courtesy of the author, ©Ledesma). The locality “3?”, corresponding to the Atrato River system (Colombia), was described as needing confirmation.



### 1.5.2 HABITAT

*T. natans* - Is an aquatic species, living in rivers, marshes and lakes (Fig. 10), usually in open areas, and is only rarely found on land (IUCN *et al.*, 2008). *T. natans* has been recorded in eutrophic waters in synanthropic and urbanised habitats in Venezuela (Gower & Wilkinson, 2005).

In Colombia, *T. natans* is associated with alluvial flood plains of the Magdalena and Cauca rivers. *T. natans* appear to migrate to the flood plains, against the flow of water during the wet season. (Tapley & Acosta, 2010) and is commonly encountered in floating mats of aquatic vegetation (Figs 10 & 11). In Colombia there are seasonal changes in water temperature (Fig. 16). In Colombia, during the wet season the water in creeks inhabited by *T. natans* were soft and slightly acidic (Table 3).

*T. compressicauda* is an aquatic species, inhabiting permanent rivers, ponds and marshes (Figs. 12-14), mainly in the lowland forest zone (IUCN *et al.*, 2008). Moodie (1978) reports that *T. compressicauda* inhabits water filled burrows. Water parameters were recorded in a pond near Cayenne (Fig. 12), French Guiana where *T. compressicauda* are known to occur, water pH 5.5 - 6; water temperature, 28.0°C; 0.9dKH (D.J. Gower & M. Wilkinson, unpublished data). Water parameters have also been reported from the Colombian Amazon where, in the month of July, a single individual was found in a cavity within a submerged log in a clear water stream: water pH 6.16; water temperature 25.4°C; conductivity 30 µS / cm; and total dissolved solids 15 ppm (Acosta-Galvis *et al.*, 2014).

*P. kaupii* - Little is known about the ecology of *P. kaupii* (Wilkinson & Nussbaum, 1997). This probably reflect the difficulties of collecting in rivers and the low frequency of herpetological collections in such habitats (Lynch, 2006). *P. kaupii* is an aquatic species that occurs in rivers, streams, large and small lakes, and flooded plains (Wilkinson *et al.*, 2010). It is unknown whether this species burrows. Specimens have been collected beneath rocks in shallow water (Wilkinson & Nussbaum, 1997). Water parameters were recorded at Saut Maripa (Fig, 15), French Guiana where *P. kaupii* are known to occur: pH 6.8; 26.7°C; 0.3-0.6dKH (D. Gower & M Wilkinson, unpublished data). Observations from captivity suggest that this species uses more oxygenated waters than *Typhlonectes* species (C. Michaels, pers. obs).



Figure 10 (top left). Habitat of *Typhlonectes natans*, Victoria, Colombia. Figure 11 (Top right). Habitat of *T. natans*, Victoria, Colombia. Figure 12 (middle left). Habitat of *T. compressicauda*, pond near Cayenne, French Guiana. Figure 13 (middle right). Habitat of *T. compressicauda*, slow moving part of the river Oyapock between French Guiana and Brazil. Figure 14 (bottom left). Habitat of *Potomotyphlus kaupii*, pool near Saut Maripa, Oyapock river, French Guiana of Saut Maripa. Figure 15 (bottom right). Habitat of *P. kaupii*, Saut Maripa, French Guiana.



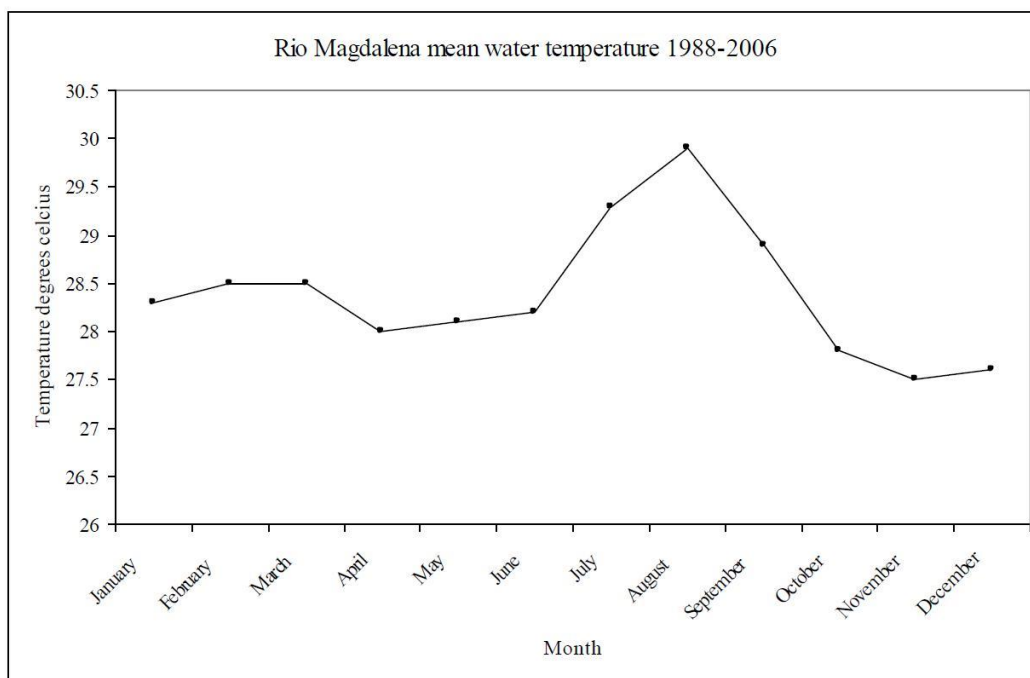


Figure 16. The mean temperatures of the Rio Magdalena at Honda from 1988 – 2006, taken from Tapley & Acosta 2010.

Parameter	min	max
Water temperature (°C)	28.1	30.7
pH	6.17	7.06
Ammonia (ppm)	0.00	0.25
Nitrite (ppm)	0.00	
Nitrate (ppm)	0.00	10.00
GH (ppm)	71.6	107.4
KH (ppm)	71.6	107.4

Table 3. Water parameter ranges from the Rio Magdalena in Colombia, November 2008 (Tapley & Acosta, 2010).

### 1.5.3 CONSERVATION STATUS & POPULATION

*T. natans* - Assessed as Least Concern in view of its wide distribution, tolerance of at least some habitat degradation, and presumed large and stable population (IUCN SCC Amphibian Specialist Group, 2020). Note, however, that the species can be found online for pet trade in Europe (Kaczmarek & Kolenda, 2018). It is unknown if this trade could threaten wild populations.

*T. compressicauda* - Assessed as Least Concern in view of its wide distribution, presumed large population, and because it is unlikely to be declining fast enough to qualify for listing in a more threatened category (La Marca *et al.*, 2021). However, the status of this species urgently needs to be reevaluated, not only because the assessment is more than 10 years out-of-date but also because recent predictive models, incorporating different scenarios of climate change, deforestation, and hydroelectric dams’ construction, anticipate that 25% of the species' range will be exposed to anthropogenic threats by 2030, and almost 50% by 2050 (Silva *et al.*, 2018).

*P. kaupii* – Assessed as Least Concern in view of its wide distribution, presumed large population, and because it is unlikely to be declining to qualify for listing in a more threatened category (Wilkinson *et al.*, 2010). Needs to be reassessed for the same reasons given above for *T. compressicauda*, given their similar distributions.

## 1.6 DIET AND FEEDING BEHAVIOUR

### 1.6.1 FOOD PREFERENCE

Hofer (2000) notes that *Typhlonectes* can be particularly abundant around fishing villages where they have been observed feeding on the discarded entrails of fish. This has been widely noted and supports the idea that wild *Typhlonectes* might be scavengers as well as predators (Kupfer *et al.*, 2006).

*T. compressicauda* – In the wild juveniles of this species consume oligochaete worms, aquatic invertebrates, terrestrial invertebrates, and anuran larva and eggs (Verdade *et al.*, 2000). Gut contents of wild specimens also contained large amounts of detritus and plant matter which could have been ingested accidentally or indicate partial detritivorous habits (Verdade *et al.*, 2000). *T. compressicauda* has been observed feeding on dead fish caught in nets (Exbrayat & Delsol, 1985) and have been caught in fish traps baited with dead fresh and tinned fish (Kupfer *et al.*, 2006) and hauled in with dead fish on rope in French Guiana. In Manaus the stomach contents of adult *T. compressicauda* included shrimp and arthropods, local fishermen reported that the species also feeds on small fish and they have been captured by hand after being attracted to fish scraps thrown into the water (Moodie, 1978).

Little is known about the wild diet of *T. natans*. Tapley & Acosta (2010) observed *T. natans* feeding on the entrails of fish which had been discarded in the water in Colombia, and the same is reported by local fisherman for populations in Lago Maracaibo, Venezuela.

Little is known about the wild diet of *P. kaupii*, but they have been captured in aquatic traps using cooked shrimp and raw fish (Rendle *et al.*, 2015).

### 1.6.2 FEEDING BEHAVIOUR

*T. compressicauda* is carnivorous and generalistic. Juvenile, wild specimens have been observed actively foraging in shallow water at night. Analysis of gut contents found prey items associated with the bottom of the aquatic habitat as well as items associated with the surface; perhaps encountered when caecilians surface to breathe (Verdade *et al.*, 2000).

Observations from captivity show that feeding is restricted to items either on the substrate or potentially lying on mid-water structures such as branches or plants. Swimming or floating foods are generally not consumed as the animals cannot find them. Animals detect food through olfaction as soon as it is present in the surrounding water and begin to move actively around on the substrate, sweeping the heading side to side in short arcs, systematically searching for food. Once located, the food item is rapidly swallowed whole if possible. If very large, for example a fish carcass, a mouthful is taken and then the animal rolls along its long axis and uses tugging motions to separate the mouthful from the food item (C. Michaels, per. obs.).

## 1.7 REPRODUCTION

### 1.7.1 DEVELOPMENTAL STAGES TO SEXUAL MATURITY

Viviparity is characteristic of all aquatic caecilians. Development of the embryo is supported by yolk reserves (Wake, 1977). These yolk reserves are rapidly depleted.

Embryonic *T. compressicauda* may increase their length six times by the time their yolk is fully absorbed (Stebbins & Cohen, 1995). Once the yolk is depleted the embryos emerge from their egg membranes and uncurl, foetuses are able to freely move along the length of the oviduct and they align themselves lengthways in the oviduct (Pough *et al.*, 2002) or they may fold back along themselves.

The foetuses are sustained by lipid rich epithelial walls of the oviduct. Foetuses have well developed dentition, which is very different to the dentition of the adults (Fig. 17). The mandibular teeth in foetal aquatic caecilians are numerous, not arranged in regular rows and extend external to the buccal cavity. These specialised teeth appear to be used to scrape the oviductal epithelium, which possibly stimulates the formation of the nutrient rich and hypertrophied epithelium (Wake, 1976). The snout is less prominent in foetal aquatic caecilians (Fig. 8), this probably facilitates the scraping manner in which they feed (Wilkinson, 1991). The specialised, deciduous teeth of foetal caecilians have been termed 'vernal' teeth because similar teeth are seen also in hatchlings of some oviparous caecilians (*i.e.* species that lack a foetal stage) (San Mauro *et al.*, 2014).

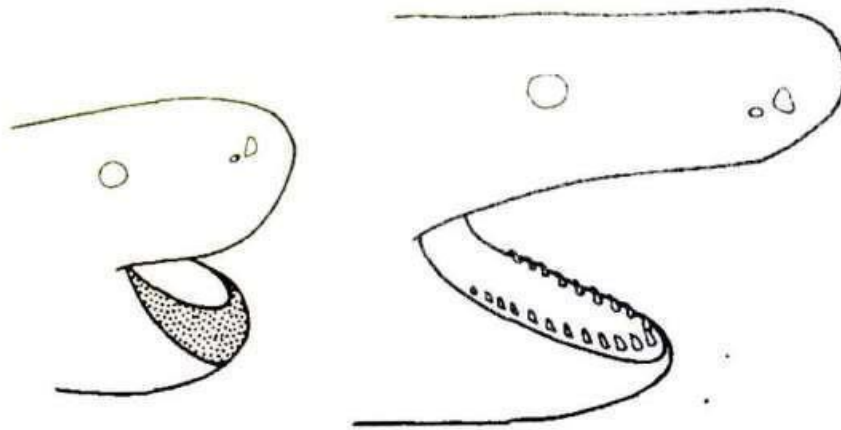


Figure 17. Mandibular tooth disposition and head shape in foetal (left) and adult (right) aquatic caecilians. Taken from Wilkinson, 1991.

The foetuses of *Typhlonectes* species develop very large, highly vascularised leaf like gills (Fig. 18 & 19) which are probably used for gas exchange across the maternal uterine wall (Wells, 2007).



Figure 18. *Typhlonectes natans* newborn with external gills (left) and the temporary indentation (scar) that is left once the gills drop off (right).

Oxygen affinity of foetal blood in *T. compressicauda* is higher than that of adult blood (Garlick *et al.*, 1979). It has also been suggested that the gills may uptake nutrients during foeto-maternal exchanges, in a pseudo-placentation (Exbrayat & Hraoui-Bloquet, 1992). Gills are lost within several hours after birth (Parkinson, 2004), and their transparency is greater immediately after the birth (Fig. 19, M. Wilkinson, pers. obs). New-born *T. compressicauda* may measure 40% of the female's total length (Stebbins & Cohen, 1995).



Figure 19. *Typhlonectes compressicauda* newborn with very transparent external gills.

*P. kaupii* are born with external gills similar in structure to those of *Typhlonectes*, although the gills are more transparent (N. Nelson pers. obs.). In *P. kaupii* gills are often lost during birth although some have retained the gills for a couple of hours after birth (N. Nelson pers. obs.). Embryonic limb buds can be observed until stage 28 in *T. compressicauda*, but the lengthening of the body and organs is seen throughout development (Exbrayat, 2018a).

### 1.7.2 AGE OF SEXUAL MATURITY

All aquatic caecilians continue to grow after sexual maturity.

In captivity, *T. compressicauda* attain sexual maturity at 16 months (Exbrayat & Delsol, 1985). The age at which *T. natans* reach sexually maturity in the wild is unknown. In captivity, a female was recorded being sexually mature at approximately 4 years of age (Ilze Duncce pers. com.). The age at which *P. kaupii* reach sexually maturity in the wild or in captivity is unknown.

### 1.7.3 SEASONALITY OF CYCLING

The reproductive biology of *T. compressicauda* in captive animals has been studied in detail both in captivity and in the wild (Exbrayat *et al.*, 1983; Exbrayat, 1986). One study on *T. compressicauda* in French Guiana found that males have a yearly cycle of spermatogenesis, which occurs from June to July; copulation occurs between February and May (Exbrayat & Delsol, 1985). Females have a biennial cycle (Fig. 20). During the first year, females are vitellogenic from October to February. This period is when ovulation occurs and gestation begins. Parturition occurs between July and October. Vitellogenesis occurs once again but very quickly. During the second period of reproduction, oocytes are not mature, and they degenerate becoming atretic follicles (Exbrayat & Delsol, 1985). The cyclical development of the genital tract is dependent on sex and pituitary hormones, with a direct control by the pituitary gland (Brun *et al.*, 2021). A short review on the endocrine regulation of the reproduction in caecilians, encompassing *T. compressicauda*, was recently published (Exbrayat, 2018b).

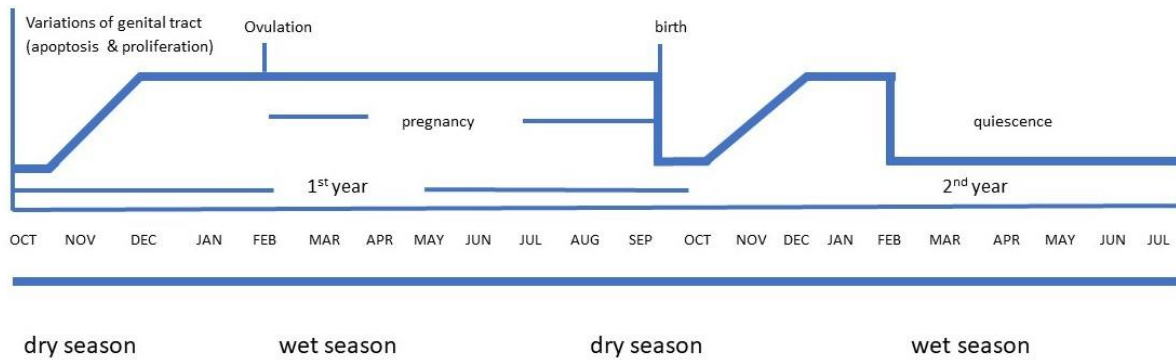


Figure 20. The biennial sexual cycle of female *Typhlonectes compressicauda* (after Raquet *et al.*, 2017).

The reproduction of *T. compressicauda* is directly related to seasonal variation in rainfall (Yousef, 2018). During the wet season, there is an increased supply of food and this is when spermatogenesis, copulation and gestation occur (Exbrayat & Delsol, 1985).

Some authors suggested that reproduction could not take place if was not preceded by a dry, terrestrial phase, during which the animals could bury in the mud (Yousef, 2018) but this contradicts observations made in captivity, where the species were successfully bred without providing a terrestrial phase (B. Tapley, pers. obs.).

*T. natans* is reportedly a seasonal breeder, breeding occurs during the wet season (Herman, 1994). The anatomy of their oviducts suggests that females could have the ability to store sperm (Dymek *et al.*, 2018).

The reproductive biology of *P. kaupii* is unknown other than that the species is viviparous (Wilkinson *et al.*, 2010; San Mauro *et al.*, 2014).

#### 1.7.4 GESTATION PERIOD

In the wild gestation in *T. compressicauda* lasts six months (Exbrayat & Delsol, 1985). In captivity the gestation period of *T. natans* is approximately 7 months (Reinhard, 2014) and *P. kaupii* gestation period is unknown.

#### 1.7.5 NUMBER OF OFFSPRING

In the wild *T. compressicauda* give birth to on average 4 young (Exbrayat & Delsol, 1985). In captivity an average of 3 young are born (Exbrayat & Delsol, 1985), but as many as eight young have been born to a single mother (M. Wilkinson, pers. obs.).

The number of offspring produced by *T. natans* and *P. kaupii* in the wild is unknown.

#### 1.7.6 BIRTH DETAILS AND SEASONS

Young of *T. compressicauda* are born at the beginning of the dry season *i.e.* when water levels are still high (Exbrayat & Delsol, 1985).

Birth details and seasons of *T. natans* and *P. kaupii* in the wild are unknown.



## 1.8 BEHAVIOUR

### 1.8.1 ACTIVITY

Activity patterns of aquatic caecilians in the wild are largely unknown. *T. compressicauda* have been observed foraging in shallow water at night (Verdade *et al.*, 2000) as well as in deeper water (D. Gower and M. Wilkinson, pers. obs.). In captivity, they can be active and forage at any time.

### 1.8.2 LOCOMOTION

Lateral undulation; can also be supported by some worm-like concertina-motion when on rough ground above the water surface or when squeezing through small gaps both under and above water (M. Goetz, pers. obs.).

### 1.8.3 PREDATION

The electric eel *Electrophorus voltai* is a predator of *T. compressicauda* (Oliveira *et al.*, 2019). Local fisherman report that *T. compressicauda* are also consumed by *Arapaima gigas* (Moodie, 1978). Other caecilians, including aquatic ones, are known to be eaten by snakes and potentially birds (*e.g.* Taylor, 1968; Kupfer *et al.*, 2003; Bittencourt-Silva & Wilkinson, 2018; Gonzalez *et al.*, 2018).

### 1.8.4 VOCALISATION

In captivity, a squeaking sound has been reported in aquatic caecilians when they surface to breathe (Stebbins & Cohen, 1995). It is probably involuntary, caused by movement of air.

### 1.8.5 SOCIAL BEHAVIOUR

In captivity, aquatic caecilians are gregarious (Figure 21) and share refugia. Refugia sites are perhaps marked with chemical signals, and individuals are attracted to chemicals emanated by conspecifics (Warbeck *et al.*, 1996), though this has not been studied in detail.



Figure 21. Young *Typhlonectes compressicauda* congregating on a raft, with attending mother.

A captive study on *T. natans* showed breeding males in captivity may be aggressive towards one another but not necessarily territorial (Warbeck & Parzefall, 2001). Males have also been observed to chase and bite females in captivity (G. Garcia, pers. obs.). A laboratory study showed that non-reproductive females preferred chemical cues from other females over male chemical cues. Reproductive males preferred cues from receptive females to

unreceptive females and did not discriminate between related and unrelated females (Warbeck & Parzefall, 2001). Parental care in the forms of egg-guarding and feeding is observed in other families of caecilians, but it has not yet been reported in Typhlonectidae (Schulte *et al.*, 2020).

### 1.8.6 SEXUAL BEHAVIOUR

There are not many records of mating in caecilians. One observation of *T. natans* showed that males entwine themselves around females vent to vent. Other reports indicate that there is often little/no entwining in *T. natans*, at least after an initial period and that the female continues to swim freely, dragging the male around by his phallus. Similar dragging of the male by the female has been observed in *T. compressicauda* (M. Wilkinson, pers. obs.). Fertilisation is internal; males having a copulatory organ or phallus formed by an evertible posterior chamber of the cloaca, the phalloseum (Fig. 22 & 23). Mating initially is frenzied but activity soon subsides. The pair can remain entwined for several hours (Parkinson, 2001). Murphy (1978) noted that copulation continued for three hours in *T. compressicauda* after it was first observed.



Figure 22 (right). A preserved male *Typhlonectes natans* illustrating the phalloseum in its everted state.  
Figure 23 (left). Everted phalloseum in a sick *T. natans*.

### 1.9 EVOLUTIONARY RESEARCH

Wilkinson (1989), Wilkinson and Nussbaum (1997), San Mauro *et al.* (2014) and Maciel *et al.* (2016) have provided hypotheses of phylogenetic relationships among aquatic caecilians and discussed aspects of their morphological evolution.

The very first putative Typhlonectidae caecilian fossil, also the first documented caecilian fossil from the Oligocene, was recently discovered in the Taubaté basin in Brazil (Santos, 2020), and a review of all caecilian fossil records to date sheds light on their systematics (Santos *et al.*, 2020). Comparisons of the crania of several extant caecilians highlighted morphological differences shaped by different phylogenetic, allometric, and ecological effects (Bardua *et al.*, 2019).

The extensive use of comparative transcriptomics allowed the identification of some of the genes that may have enabled caecilians to adapt to various habitats and habits (Torres-Sánchez *et al.*, 2019a). They also suggested vertebrate homologues for several of their functional genes (Torres-Sánchez *et al.*, 2019b), and revealed differential skin functions depending on body regions (Torres-Sánchez *et al.*, 2020). For example, the high expression

of claudin transmembrane proteins in the posterior skin could prevent uncontrolled loss of water in this body region highly abundant in granular glands (Torres-Sánchez *et al.*, 2020).

## SECTION 2. MANAGEMENT IN CAPTIVITY

*This section has been completed with the help and support of private keepers, Zoological institutions, museums and Aquaria. Questionnaires were sent out to many caecilian keepers. We received 12 responses. There was a great deal of knowledge held by all sectors and their responses to the questionnaire have been invaluable when compiling these guidelines.*

Note that *Typhlonectes* and *Potomotyphlus* needs are fundamentally different: the former should be provided with muddy, relatively low-oxygen environments, while the enclosures of the latter should replicate stony, well oxygenated streams.

### 2.1 ENCLOSURE

The enclosure must be aquatic and is recommended to have a small, wet land area; although a large piece of floating cork bark or dense mat of floating vegetation will suffice. The larger the enclosure the better. A larger volume of water is especially important to buffer against fluctuations in water parameters, which are sub optimal. A 180 L aquarium would be suitable to a trio of *T. compressicauda* or *P. kaupii* and a 250 L aquarium would be suitable to a trio of *T. natans* provided large external canister filters were provided for filtration. The land area should ideally have crevices in which the animals can lie.

#### 2.1.1 SUBSTRATE

For easy maintenance and monitoring, a substrate is not essential when keeping aquatic caecilians, but its absence may have welfare implications through preventing burrowing behaviour. If substrate is required, aquarium sand and aquarium pea gravel have both been used successfully. Thick sand beds may produce toxic gases such as hydrogen sulphide. Caecilians may spend significant amounts of time buried in the sand if this particular substrate is used, and for this reason it should be recommended.

#### 2.1.2 FURNISHINGS AND MAINTENANCE

Refugia should be provided, to allow animals to congregate beneath or inside it. PVC pipes, rocks, bog wood and terracotta can be used. Rough edges of refugia will facilitate skin shedding, these edges should not be sharp, however. Refugia should provide tight crevices, into which the caecilians can squeeze. For aesthetically pleasing exhibits aquatic plants can be added; however, plants are often uprooted or damaged during shedding or feeding frenzies so free-floating plants might be the better option. *Ceratophyllum* species has been reported by one keeper to be an excellent shedding aid. In the field *T. natans* have been found in floating mats of aquatic vegetation. If this is recreated in captivity *T. natans* and *T. compressicauda* will spend considerable amounts of time in the vegetation mat and are easy to see. Floating mats of vegetation may make exhibiting this species in zoos and aquaria easier. Watercress (*Rorippa nasturtium*) has been used successfully for this purpose. Importantly, live aquatic plants will also reduce nitrate, nitrite and ammonia levels in aquaria. These nitrogenous waste products are toxic to aquatic animals.

As mentioned above, the provision of a wet land area is recommended for *Typhlonectes* (*Potomotyphlus* do not require this as they are not known to leave the water). A floating section of cork bark or dense mat of emergent vegetation is commonly used. Water parameters and animal health should be checked if caecilians are found to be on this terrestrial area for long periods of time because this might indicate problems with water quality or temperature.

Partial water changes of 10 – 20% should be carried out on a regular basis (the frequency would be dependent on the volume of water in the aquarium, amount of live plants used, amount of constant fresh-water influx, stocking density). Smaller, densely stocked aquaria will often require more water changes than larger aquaria with fewer animals. If tap water is used it should be treated with a water conditioner and allowed to stand at aquarium temperature, while being well aerated, for 24 hours before it is added to the aquaria to neutralise or remove toxic contaminants; chlorine removal can be facilitated and expedited by inserting an aeration stone. Water should also be preheated to match the water temperature of the enclosure it is to be added to in order to prevent temperature shock. If parameters of the tap water are unsuitable for aquatic caecilians, re-mineralised rainwater or remineralised reverse osmosis (RO) water can be used.

Water changes should ideally be carried out after feeding so that uneaten food and faeces are removed from the aquaria as quickly as possible before they have the chance to decay. The filter media should be cleaned regularly in water taken from the aquarium to preserve beneficial filtration bacteria within which commonly die when flushed with water of different parameters and/or temperature again, the schedule of filter maintenance will depend on filter type and size, stocking density, and feeding regime. There is a tendency for aquatic caecilians to squeeze into filtration tubes and the filters themselves and this can result in severe injury or drowning. Filter tubes and filters should be well protected to prevent such access.

### 2.1.3 WATER

Aquatic caecilians are commonly kept in tap water. If tap water is to be used, the municipal water provider should be contacted to get detailed information on water parameters, as the parameters will vary dramatically both geographically and temporally. Water parameters from tap water can vary greatly on a day-to-day basis. Re-mineralised rainwater can be collected. However, pure reverse osmosis water or pure rainwater should be used with caution with amphibians, and not at all with aquatic caecilians, because it is too pure and may interfere with the osmotic balance of the caecilian.

The water should be deep enough to provide the animals with adequate space to swim and to reach the surface without losing contact with the substrate, plants or larger hides. The lowering of the water level to 18cm was one of the factors attributed to the breeding of *T. natans* by one breeder (Parkinson, 2004).

“You can-not manage what you do not measure”. Water quality should be tested on a regular and frequent basis (ideally weekly as routine); this will allow the performance of filtration systems to be assessed, it can identify one-off problems or recurring issues and assess corrective measures. The most important routine tests to perform are those for nitrogenous waste (ammonia, nitrite and nitrate), pH, and hardness/alkalinity.

Fluctuating water parameters can be very stressful for aquatic organisms. It is better to have slightly suboptimal parameters than wildly fluctuating parameters. At Jersey Zoo the tap water supply (well / borehole water) was unsuitable for aquatic caecilians as it was

contaminated with nitrates from agricultural run-off. Reverse osmosis (RO) water was reconstituted with a remineralisation solution (sodium bicarbonate) which matched the water parameters presented in Table 3 and an alkalinity of c. 70mg/L. A test for general hardness was used periodically to check that salts were not becoming concentrated as the tank water evaporated (dGH range between 3-8).

Historically, at ZSL London Zoo RO water was used for *T. natans*, *T. compressicauda* and *P. kaupii*. This was remineralised with sodium bicarbonate to achieve an alkalinity of 60mg/l and pH of 7.8. However, the plant growth in this water was suboptimal and parameters were unstable. Tap water diluted with RO water is used (1:9) and tannins are added to achieve an alkalinity of c. 70mg/L and a pH of about 6.5 (the tannins suppress the pH).

#### 2.1.4 TEMPERATURE

Water temperature should be between 24 – 30°C for *Typhlonectes* spp. (see Fig. 16 for details of seasonal fluctuations) and between 26 – 29°C for *Potomotyphlus kaupii*. Aquarium heaters can be used for this purpose but ensure that heater guards are used; aquatic caecilians often lie wrapped around heaters and having a guard can prevent the risk of burns. The use of inline heaters avoids this risk. Ideally, more than one heater should be used to allow for redundancy.

When the temperature regime presented in Fig. 16 was implemented at Jersey Zoo, breeding of *T. natans* was achieved. However, it is unproven whether or not the breeding was due to a change in temperature regime or whether the specimens had by that time simply acclimatised to captive conditions. As mentioned in 1.7.3, the complex cycle of reproduction of aquatic caecilians seems heavily influenced by foregone conditions, and endogen cycles (Exbrayat & Laurent, 1986).

#### 2.1.5 FILTRATION

Aquatic caecilians are messy feeders and shed their skin often. It is easy for detritus to accumulate in aquaria and therefore, large filters are vital. Internal box filters are only sufficient for neonates and small juvenile animals, or for adult tanks at low density and/or heavy plant growth. External canister filters are more suitable for adults because they provide a larger surface area for biological filtration. Large air-driven sponge filters are also suitable. Filter media should be cleaned on a regular basis (depending on the size of the filter and the stocking density), media should be cleaned in tank water rather than tap water because the change in parameters can be detrimental to the nitrifying bacteria, that are vital for biological filtration. Live plants, provided that they are growing and not simply slowly dying, can also help to improve and stabilise water quality as well as remove toxins from the water column.

Juvenile and adult animals can easily find their way into the internal mechanisms of filters. For this reason, it is vital that any way into the filter is blocked up with mesh or sealed with silicone. External filter intakes and outlets must be well covered with a firmly affixed strainer. This should be physically tied or similar to the intake/outlet as caecilians may detach push-fit strainers.

#### 2.1.6 pH

Water pH does not appear to be of great significance in the husbandry of aquatic caecilians; however, it is advisable to avoid extremes, rapid or constant fluctuations.



It has been reported that a drop in pH from pH 5-6 to 5 was one of the factors attributed to the breeding of *T. natans* (Parkinson, 2004). pH 5 is extremely acidic and probably not optimal when maintaining *T. natans* in captivity. Tapley and Acosta (2010) reported pH in the range of 6-7 in the Magdalena valley of Colombia in November (Table 2). Where possible, parameters from the field should be replicated in captivity.

In the Natural History Museum, *T. compressicauda* survived an unintentional increase in acidity to pH ca.4, but health problems were not apparent until directly moving animals from pH ca.4 to pH ca.6–7 resulted in skin problems and, in some cases, death. At Sedgwick County Zoo *P. kaupii* are maintained at a pH of 7.8-8.2.

In the wild, *Typhlonectes* often inhabit blackwater environments with high tannin content. Tannins act as antimicrobials and also suppress pH and seem to contribute positively to the husbandry of this Genus in captivity. Tannins may be added to water through commercial additives (e.g. Tropol, produced by JBL), or through inclusion of tannin-rich substances such as dead oak leaves, peat or bog wood in aquaria. Tannins may be used to achieve acidic pH while still including some mineral content in the water (at ZSL an alkalinity of c. 70mg/L, which would normally have a pH of c. 7.5, is maintained with a pH of 6.5 through the use of Tropol at a concentration much higher than the dose recommended by the manufacturer: ca. 2ml per L water). Whichever route is taken, effects on pH should be carefully measured. At Paignton Zoo, a piece of bogwood released so much tannin into an aquarium that it suppressed pH below 5, causing health issues with *T. natans*.

### 2.1.7 HARDNESS AND ALKALINITY

Kowalski (2001) reports that *T. natans* may die if the hardness of the water exceeds 5.6dH (100PPM).

At Sedgwick County Zoo a breeding group of *P. kaupii* are maintained in relatively alkaline water ranging from 82-227 mg/L. Hardness (as CaCO<sub>3</sub>) ranges 120-130 mg/L.

### 2.1.8 DISSOLVED OXYGEN

*P. kaupii* have a greater reliance on cutaneous gas exchange and thus it is very likely that they have a requirement for more dissolved oxygen than *Typhlonectes* species. Air stones are used at ZSL London Zoo and Sedgwick County Zoo to increase the dissolved oxygen content of the water. At ZSL London Zoo the outflow of the filter was positioned at the surface of the water as this also increases the oxygen content of the water. Moderate water flow, provided that refugia from current are available, may be beneficial in preventing the formation of hypoxic pockets of water.

### 2.1.9 LIGHTING

Standard aquarium lighting with a 12:12 hour photoperiod seems to be sufficient for the maintenance of aquatic caecilians and is appropriate due to their tropical distribution. All species prefer dark conditions, so if lighting is included for plant growth and/or aesthetics/public viewing, the animals must have access to dark refuges.

The UVB requirement of aquatic caecilians is unknown. However, as the animals were frequently found resting on half-submerged floating cork bark at Jersey Zoo, adult specimens were exposed to UVB radiation (70 µW/cm<sup>2</sup> at the surface of the water) using a Zoo Med ReptiSun<sup>®</sup> 5.0 with reflector for 14 months on a 12:12 light cycle, X-rays pre

and post UVB exposure revealed no changes in bone mineralisation. Bone mineralisation was considered good to begin with and all of the specimens were of captive bred origin and had never been exposed to UVB emitting lights.

## 2.2 FEEDING

Feeding of aquatic caecilians is recommended 2 to 3 times a week. In their first year, juvenile animals should be offered food at least three times a week. Aquatic caecilians probably locate their food via olfaction. Juvenile *T. natans* have been observed swimming up and down an enclosure with jaws open and pressed down against the substrate, the jaws close immediately when they encounter the food item (B. Tapley, pers. obs.). Aquatic caecilians can be conditioned to hand feed which can help to ensure all specimens are fed suitable amounts. When possible, live food should be offered, because it does not break down immediately and so causes fewer issues with water quality in the short term. However, aquatic caecilians do not appear to be active vertebrate hunters so although live invertebrates including worms and insect larvae will be consumed live, vertebrate components of the diet (especially fish) will typically not be captured live, and dead items must be offered.

### 2.2.1 BASIC DIET

A huge variety of food will be readily accepted by aquatic caecilians including both live and dead land invertebrates (e.g. crickets, earthworms, beetle larvae etc), thawed fish (both freshwater and saltwater fish), crustacea, molluscs, pinkie mice, shrimp blood-worm as well as commercially available amphibian pellet. Many groups of fish are high in thiaminase, which breaks down vitamin B<sub>12</sub> even under frozen conditions. Thiaminase is present in both freshwater and marine fish; among freshwater fish (to which aquatic caecilians diets should be limited due to salt content), cyprinid (carp Family) fish are especially rich in thiaminase and should only form part of a varied diet. Salmonid fish are low in thiaminase, but extremely high in oils, and so should not be used as a standard diet, either. The risk of ill health associated with dietary deficiencies and hypovitaminosis can be decreased by offering captive specimens a varied diet and by storing foodstuffs appropriately.

### 2.2.2 SPECIAL DIETARY REQUIREMENTS

Adding dietary supplements to the food of aquatic animals can be difficult. At Jersey Zoo, Nanovits produced by Vetark were trialled over a three-month period. 0.2mls were injected into a food item and given to each caecilian once a month for three months. It was noted that the vitamins leached out into the water from the injection site, the effectiveness of the supplement could not be quantified, and staff did not know if the Nanovit could be an irritant to the caecilians if it were to go into solution. Because the nutritional requirements of caecilians are unknown, it is probably better to offer a wide variety of food items as a varied diet containing fresh prey that has been fed a natural and balanced diet.

The mouths of *P. kaupii* are relatively smaller than the mouths of *T. compressicauda* and *T. natans*. Keepers at Sedgwick County Zoo offer *P. kaupii* smaller prey items relative to the prey items they offer *T. natans*. However, aquatic caecilians are typically capable of tearing off pieces of food from larger items.

### 2.2.3 METHOD OF FEEDING

Food can be placed in the aquaria in different locations to ensure all the inhabitants get the chance to feed. Larger animals tend to consume food items more quickly than smaller ones and so can potentially deprive smaller animals of their share. Animals can accept food offered to them on forceps and this may be beneficial in ensuring that all specimens have the opportunity to feed. At Chester Zoo, dead crayfish or fish were submerged and attached to a branch where caecilians spent significant amounts of time feeding, pulling at pieces of flesh. This not only provided visitors with the opportunity to see how aquatic caecilians feed, but also increased their visibility during feeding times (G. Garcia, pers. com.)

## 2.3 SOCIAL STRUCTURE

*T. natans* do not appear to be territorial (Warbeck & Parzefall, 2001); however, during the breeding season males may become aggressive to one another. They appear to prefer to share refugia with other animals and are anecdotally reported to do better when housed in colonies of multiple animals, rather than singly. They may bite each other when hurriedly trying to find and ingest food.

### 2.3.1 CHANGING GROUP STRUCTURE

Introducing a new male to an enclosure which already houses both males and females during the breeding season should be avoided.

### 2.3.2 SHARING ENCLOSURE WITH OTHER SPECIES

Because *T. natans* have been reported to feed on small fish, it is best to avoid adding these to an enclosure housing aquatic caecilians. Piscivory appears to be largely limited to dead and dying fish, however, and exhibits containing *Typhlonectes* and a variety of small fish of appropriate geographic origins, e.g. tetras (Characiformes) are often very successful. There were no reported problems when keeping aquatic caecilians with larger fish, providing the fish are not predatory. In the wild, Moodie (1978) noted that *T. compressicauda* was toxic to some fish. *T. natans* have been kept successfully alongside *T. compressicauda* (M. Wilkinson, pers. obs), *P. kaupii* and pipid frogs *Pipa pipa*. At Sedgwick County Zoo, stressed *P. kaupii* released a skin secretion after being handled that killed fish maintained in the same aquarium. Be aware though, that aquarium fish commonly harbour Mycobacteria which have the potential to affect amphibians and human skin.

A variety of fish species including *Aequidens pulche*, *Astyanax magdalenae*, *Cyphocharax magdalenae*, *Hyphessobrycon inconstans*, *Pimelodus blochii*, *Poecilia caucana*, *Prochilodus magdalenae* and *Triportheus magdalenae* are reported from the fishing village of Guarinócito where several *T. natans* were observed by Tapley & Acosta (2010).

Housing aquatic caecilians alongside species originating from different geographical origins could expose specimens to novel pathogens, or result in one or both species being exposed to suboptimal conditions. This should be avoided in captivity.

## 2.4 BREEDING

In captivity *T. natans* are relatively frequently bred. Some zoological institutions in the United States have bred *T. natans* to F5. *T. compressicauda* have also been bred in captivity (Exbrayat & Delsol, 1985; Mark Wilkinson pers. comm.). Sedgwick County Zoo have successfully bred *P. kaupii* (Fig. 24) on multiple occasions (N. Nelson pers. obs.).

Although captivity does not seem to affect the male reproductive cycle, it leads to delayed vitellogenesis in *T. compressicauda*. Litter size, duration of the embryonic development and offspring survival are also reported to be reduced in captivity (Exbrayat & Laurent, 1986).



Figure 24. Recently born *Potomotyphlus kaupii* at Sedgwick County Zoo.

### 2.4.1 MATING

In captivity, *T. natans* mate throughout the year. Parkinson (2004), noted that mating commenced in July when water levels were lowered to 18 cm, and the pH decreased to pH5 (from pH 5-6) and nitrates levels were increased (from dH3). Parkinson made these changes to the water chemistry in an attempt to replicate dry season conditions with low water. pH can be lowered by filtering water through peat moss (Bailey *et al*, 2005) although the sustainability of peat moss should be established when it is sourced (C. Michaels, pers. com.). Kowalski (2001) reports that the lowering of total hardness of aquarium water can induce breeding and recommends the hardness should be halved providing that hardness does not drop below 1 dH (17.8PPM). Hardness can be lowered with the addition of distilled or reverse osmosis water. At Jersey Zoo, *T. natans* bred when the temperature regime in Fig. 16 was implemented.



Figure 25. A rare observation of copulating *Typhlonectes natans*. The animals remained motionless for at least 20 minutes, after being found in this position.



At Sedgwick County Zoo and ZSL London Zoo, *P. kaupii* are kept and bred without any seasonal changes of water temperatures or chemistry (N. Nelson, pers. obs.). Indeed, stability in conditions both in terms of physical housing (refuges, individuals in the group and the actual tank used) as well as chemical (water chemistry and quality) are often associated with successfully breeding colonies of aquatic caecilians, and changes to any of these factors might be detrimental to the health and reproduction of historically successful and stable colonies.

#### 2.4.2 GESTATION PERIOD

A gestation period of 43 weeks has been reported for *T. natans* in captivity (Parkinson, 2004). Young can be detected via ultrasound from early stages of development (< 2 months). Once further developed, fetuses can be observed via ultrasound moving within the oviduct of the female, the sac like gills can also be observed. During mid to late stages of development (> 5 months), once the skeleton of the fetuses has become calcified, a radiograph image may clearly show the developing fetuses within the adult (Fig.26). This method is particularly useful for estimating the number of fetuses present.

A gestation period of 24 – 30 weeks has been reported for *T. compressicauda* in captivity (Bruins *et al*, 1999; Exbrayat & Delsol, 1985).

The gestation period of *P. kaupii* is unknown. At Sedgwick County Zoo gestation period is estimated to be between 6-10 months with females first appearing gravid in the early autumn and giving birth late autumn / early winter (N. Nelson, pers. obs.). At Sedgwick County Zoo, water temperatures are increased to 29°C for the duration of the gestation period because this improved survival rate of neonates (N. Nelson, pers. obs.). At ZSL London Zoo, females first appeared gravid (*i.e.* became noticeably rotund) in early spring and gave birth in mid-June.

#### 2.4.3 BIRTH

*T. compressicauda* give birth to a recorded maximum of 8 young over a period of several days (M. Wilkinson, pers. obs.) which measure between 100 – 140mm (Bruins, 1999).

*T. natans* give birth to 1 – 11 young, this process may take place over multiple birthing events during up to seven days in which one to three young are born at a time. Birth may take place during the night or early morning. Some evidence suggests that in captivity environmental change may trigger birth in *T. natans*, although experiences with this have been conflicting (Chester Zoo). In one instance, a heavily pregnant female known to be carrying seven young as identified on radiograph to be of appropriate size for birth, was removed from an aquarium with a water depth of 60 cm with fast flowing filtration, and placed in an aquarium with a water depth of 17 cm. The aquarium had a water temperature of 26-28°C (PH 6.8 – 7.0) and contained sunken leaf litter and gentle filtration, after five days four young were born overnight, the following three were born 24 hours later.

On a separate occasion, a heavily pregnant female which was transferred to shallow water but failed to give birth after one month in these conditions, upon returning to an aquarium with increased depth and water flow, all young were born within 48 hours. Although conflicting, this may suggest that some form of environmental manipulation may influence the release of young. In more than one instance, females that were not provided with environmental change were found dead within aquaria, one containing seven large (<20 cm) fully developed young that had died within the female. A post mortem of this individual showed developing ova alongside fully developed young, some

of which had already begun decomposition, which was the likely cause of death in the female. This may also support the benefit of some form of environmental stimulus to encourage birth.



Figure 26. A radiograph of a gravid *Typhlonectes natans*. Note the obvious PIT tag towards the posterior end of the body.

Two authors (Wake, 1994; Parkinson 2004) have observed female *T. natans* carrying young on their backs to the water surface for their first breaths of air. There have been reports of adult animals attempting to eat neonates (Kowalski, 2001).

In captivity *P. kaupii* give birth to 1-9 young. One gravid female that died contained 14 well developed young (N. Nelson, pers. obs.).

#### 2.4.4 DEVELOPMENT AND CARE OF YOUNG

Young should not be separated from the adults after birth given their tendency to aggregate with one another. Although there are reports of adult animals attempting to eat neonates (Kowalski, 2001), this has not been observed by the authors and the risk is considered low. Additional material should be added (*e.g.* floating plants, cork bark, twigs etc.) to the enclosure to aid as young are weaker swimmers and may drown. It may be several weeks before young will take food but once they begin feeding, they should be provided food more frequently than adult specimens. The young of *T. natans* nearly double their weight at birth within 3 months (Fig. 27) and will accept a variety of food items including many also used for adults, fish carcasses are readily accepted, even large fish can be used for groups of young which will actively carcass feed, care must be taken when using such food items to maintain water quality.

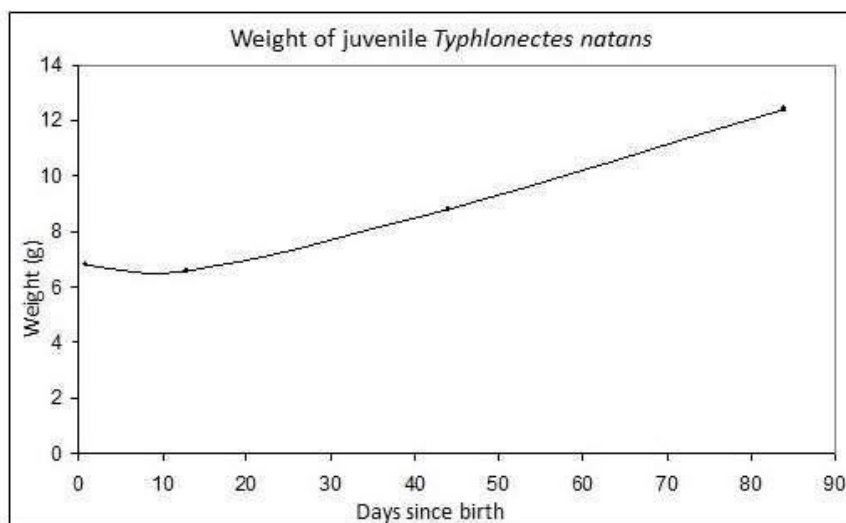


Figure 27. Average weight of juvenile *T. natans* after birth.

#### 2.4.5 POPULATION MANAGEMENT

As in many viviparous species, reproduction is associated with complications (Barbon *et al.*, 2017) and high female mortality in aquatic caecilians. However, preventing females from reproducing could impact their welfare, as is observed in other species. Therefore, until a studbook for population management is established, we recommend that institutions take into account their ethical policies to decide on whether to keep sexes apart or let them reproduce regularly and dispatch the offspring. For that latter case, see the section “Anesthesia and Euthanasia”.

Captive populations of aquatic caecilians are not formally managed. The populations of *T. natans* and *T. compressicauda* in EAZA collections are monitored by the EAZA Amphibian Taxon Advisory Group. The validation of standardised marking techniques would assist the establishment of EEPs should this be desired in future.

As of August 21<sup>st</sup>, 2021, 433 individuals in the Genus *Typhlonectes* are held in zoos and aquaria across Europe and North America, and 42 *Potomotyphlus* are divided across North American *ex-situ* institutions. No other genera in the Typhlonectidae Family are reported to be maintained in captivity (Species360, 2021).

## 2.5 BEHAVIOURAL ENRICHMENT

Behavioural enrichment is an important component of captive husbandry and can enhance the welfare of individuals. Behavioural enrichment has been understudied in amphibians (Michaels *et al.* 2014) and this may be a result of a lack of understanding of amphibian cognition in general. Prolonging feeding times by offering small, scattered food items or tethering a large food item to enclosure furnishings may be potentially enriching for aquatic caecilians.

## 2.6 HANDLING

Caecilians should be handled as little as possible, and they often thrash around when grabbed. Gloves should always be used when handling amphibians to avoid disease transmission and damage and injuries to the animals themselves. Research (Cashins & Alford, 2008) suggests that latex and nitrile gloves can have lethal effects on *Xenopus* tadpoles. The potential lethal effect of gloves on caecilians has not yet been investigated.

### 2.6.1 INDIVIDUAL IDENTIFICATION AND SEXING

It is difficult to individually identify aquatic caecilians. Visible implant elastomers (VIE) have successfully been used to distinguish terrestrial *Herpele squalostoma* individuals for 191 days on average; however, this same technique did not work in terrestrial but smaller and darker pigmented *Microcaecilia unicolor* (Tapley *et al.*, 2019). The outcome of this procedure likely depends on skin properties, which may differ between caecilian families and genera. Note that the implantation of VIE requires anaesthesia because unanaesthetised caecilians are extremely difficult to manually restrain.

Gower *et al.* (2006) successfully used Alpha-numeric fluorescent tags to mark the terrestrial caecilian *Gegeneophis ramaswamii*. Passive integrated transponder (PIT tag) has also been used with success on adult individuals maintained at Chester Zoo; an implanted PIT tag can be seen in the radiograph image presented in Fig. 26.

Freeze branding has also been used to mark caecilians (Gower *et al.*, 2006) although this technique may not be ethically acceptable in many institutions. To insert the PIT tags, animals need to be anaesthetised because they can be difficult to restrain. Transponders should then not be implanted parallel to the main axis of the body, but transversally and maintained within the anterior and posterior edges of the annuli (at least in smaller individuals). Immersions in a 0.1% MS222 bath at a pH of 7.2 until righting reflex is lost should give enough time for marking (Javier Lopez, pers comm.).

The sexes can only be externally reliably determined in mature animals. Males have relatively larger, more bluntly rounded posterior ends with somewhat wider cloacal discs (Fig. 28) (Stebbins & Cohen, 1995). The cloacal discs of females appear more slit like and elongated (Wilkinson, 1989). In *Potomotyphlus* (Fig. 28) and *T. compressicauda*, the cloacal disc is especially large and the flat circular end of the body is very apparent from a dorsal aspect of the animal.





Figure 28. Cloacal disc of male *Typhlonectes natans* (top left) and female *T. natans* (top right). Cloacal disc of male *Potomotyphlus kaupii* (bottom left) and female *P. kaupii* (bottom right).

### 2.6.2 CATCHING / RESTRAINING

Restraint can be difficult due to the large amounts of mucus secreted by aquatic caecilians when handled. Nets should be avoided where possible as they can strip the mucus layer from caecilians and leave the skin prone to infections. Instead, they should be encouraged into a solid container. Anaesthesia may be necessary for marking individuals or for delicate veterinary procedures. Immersions in a 0.1% MS222 bath at a pH of 7.0-7.4 until righting reflex is lost should give enough time for minor procedures such as microchipping, ultrasound scanning, etc. For longer procedures such as surgery, keep in bath for 2-3 more minutes and then remove, rinse and place in fresh water. Depth of anaesthesia is monitored by response to a skin pinch, if there is a reaction, it needs to be returned to the anaesthetic bath until response disappears again. Water bath used during prolonged anaesthetics should be aerated with oxygen to increase levels solution as thus increase diffusion through skin.

### 2.6.3 TRANSPORTATION

Aquatic caecilians can be transported in ventilated boxes on wet / water soaked paper towel (between two pads of paper) or moss; sphagnum moss should be avoided, however, because it is extremely acidic. In temperate climates care should be taken that the animals are not transported in cold periods.

### 2.6.4 SAFETY

Aquatic caecilians do not pose a threat to keepers. Contrary to some other caecilian families, *T. compressicauda* are reported to have tooth-related venom glands only in the lower jaws. They are unknown to bite and are not a threat to keepers (Mailho-Fontana *et al.*, 2020). However, Moodie (1978) found that skin secretions of *T. compressicauda* were toxic to the coexisting predatory fish *Hoplias malabaricus*. When handled, *P. kaupii* can secrete a substance from their skin that is capable of killing tetra fish that are housed in the same enclosure (N. Nelson, pers. obs.). If the mucus of aquatic caecilians comes in contacts with the eyes or with cuts, it causes a burning sensation (Stebbins & Cohen, 1995). It has been reported that prolonged exposure to the mucus of *T. compressicauda* can lead to a tingling sensation in the hands (Mark Wilkinson, pers. comm.). As with all aquatic vertebrates, there is the potential of *Mycobacterium* infections, which can also affect human skin.

## 2.7 VETERINARY CARE AND SPECIFIC MEDICAL PROBLEMS: CONSIDERATIONS FOR HEALTH AND WELFARE

### 2.7.1 CLINICAL ANATOMY AND PHYSIOLOGY.

The integument is vascular and permeable to water (Clayton & Mylniczenko, 2015), detailed skin structure has been described in the terrestrial-to-semiaquatic (as adults) ichthyophiids *Ichthyophis tricolor* and *Uraeothyphlus oxyurus*, showing an epidermis and dermis containing mucous and granular glands (Arun *et al.*, 2018). In some caecilian species these glands that can produce substances that can produce irritation to mucous membranes (O'Reilly *et al.*, 1996) and may have an antiprotozoal activity (Pinto *et al.*, 2014). More recently, dermal glands of *T. natans* were described extensively (Zuwala *et al.*, 2018). Like many amphibians, *T. natans* are biofluorescent: their cloacal region and skin mucous-like secretions fluoresce in green when lit with blue light (Lamb & Davis, 2020).

Caecilians do not have vestigial limbs, thoracic or pelvic girdles, the ribs do not support the body (O'Reilly *et al.*, 1996). The liver, kidney, spleen and thymus have a haematopoietic role as caecilians lack functional bone marrow (Wright, 2001).

In many caecilian species, only the right lung is developed and functional (Prabha *et al.*, 2000) and while one species of aquatic caecilians does not possess lungs (*Atretochoana eiselti*) the others have at least a well-developed right lung (Wilkinson & Nussbaum, 1997). The ventilatory cycle consists in a long expiration and a series of short inspirations aided by buccopharyngeal pumping, the respiratory rate in *T. natans* is 4-7 breaths per hour (Prabha *et al.*, 2000).

Gas exchange occurs by a combination of pulmonary, buccopharyngeal and cutaneous mechanisms, oxygen uptake occurs through pulmonary while carbon dioxide elimination takes place through the skin (Gardner *et al.*, 2000).

Terrestrial caecilians appear to be uricotelic while aquatic species are ammoniotelic (Stiffler & Talbot, 2000). A bilobate urinary bladder is present in some species (Clayton & Mylniczenko, 2015).

Eyes are rudimentary, covered by skin or bone. They are, however, functional and adapted to dim light conditions. Caecilians possess only one photoreceptor cell type (common to other amphibian orders) which expresses the red rhodopsin pigment RH1, providing them with scotopic vision (Mohun & Davies, 2019). External and middle ears are absent in all caecilian species. Olfaction, aided by chemosensory and tactile external tentacles connected to vomeronasal organ and nervous system in some species, is the main method to locate prey (Clayton & Mylniczenko, 2015).

Presence of prominent fat bodies in the caudal third of the body in *T. natans* needs to be considered for diagnostic imaging purposes and surgery. (Barbon *et al.*, 2017).

In *T. natans*, the female reproductive system anatomy consists of longitudinal, paired ovaries incorporated with fat bodies, paired oviducts and the cloaca. The ovaries are arranged in germinal nests and can contain oocytes in various stages of development. The wall of the oviduct consists of three layers: the mucous membrane, layers of muscles and serous membrane (Dymek *et al.*, 2018).

### 2.7.2 PHYSICAL EXAMINATION AND DIAGNOSTICS

Water quality assessment should be a basic step while assessing the health status in this species including parameters such as temperature, pH, ammonia, nitrite, nitrate and dissolved oxygen.

Visual examination in the enclosure may reveal non-specific clinical signs such as abnormal swimming, spending time and head bobbing in and out of the water. Visual assessment of body condition should consider fluid accumulation in the coelomic cavity in presumptive overweight animals.

Skin examination can be carried out in and outside of the water as certain abnormalities may change appearance significantly. Impression smears and skin scrapes can be carried out from lesions such as ulcerations or areas with mucous strips. The cloacal disc should be examined closely as erythema in this area is a non-specific sign of disease.

Faecal parasitology including direct examination and flotation can be carried out, specific species of nematodes such as *Pharyngodon Boulengerula* have been described in caecilians (Ubelaker, 1965), a survey of faecal parasitology results in captive caecilians (*T. natans*, *Dermophis mexicanus*, *Schistometopum thomense*, *Ichthyophis kohtaoensis*) showed presence of unidentified flagellates, ciliates, coccidia, nematodes and cestodes, including parasites from food items such as *Monocystis* sp. trophozoites from earthworms or mites from rodents (Mylniczenko, 2006).

Fine needle aspirate from lesions, organs of fluid in coelom can be carried out for diagnostic purposes.

Blood sampling can be done by cardiocentesis under general anaesthesia, the heart can be located easily in the cranial third of the body by doppler or ultrasound. A limited range of haematological and biochemistry parameters in *Dermophis mexicanus*, *T. natans*, *T. compressicauda* and *Boulengerula taitanus* are available in Clayton & Mylniczenko (2015).

The blood parasite *Hepatozoon* sp. has been observed in blood smears of wild (non-aquatic) caecilians in the Seychelles islands without any reported clinical implications (Harris *et al*, 2014).

Radiography can be carried out with the animals conscious by placing them in a rigid clear plastic tube; radiographs have been used for the diagnosis of pregnancy.

Ultrasonography can be carried out with the animal submerged in water or wrapped in wet material.



Figure 29. Cloacal disc erythema in a *Typhlonectes natans* at post-mortem

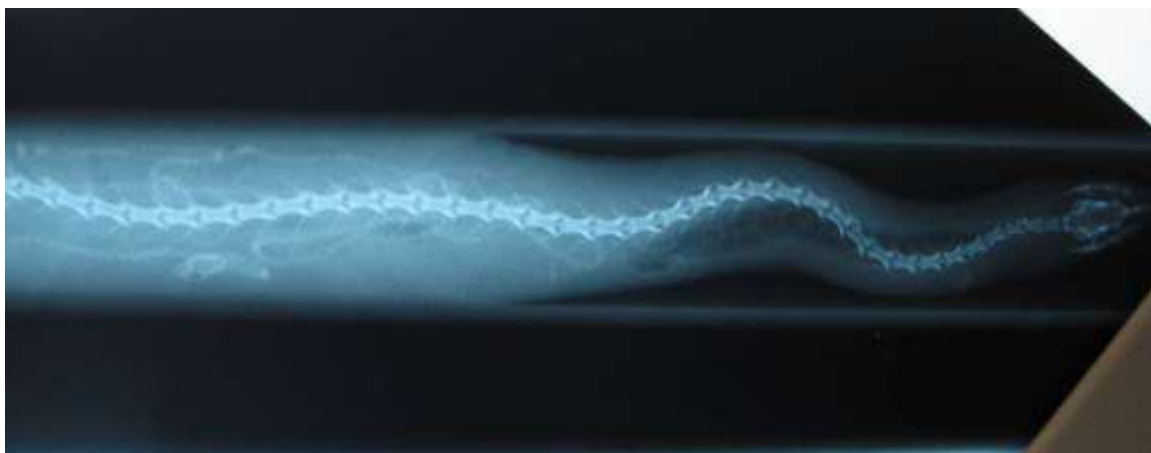


Figure 30. Radiographs to diagnose pregnancy, notice how far anterior in the mother's body the foetuses can be displaced.



### 2.7.3 DISEASES

Dermatitis has been reported as the most common medical problem encountered in captive caecilian species as a cause of morbidity and mortality, associated with *Saprolegnia* sp and subcutaneous nematode migration (Mylniczenko, 2006; Flach *et al.*, 2020, Kane *et al.*, 2021). Nematode migration will cause localized subcutaneous swellings and white plaques on the epidermis surface (O'Reilly, 1996). Nematodes may break the skin causing open skin wounds (Wake, 1994) and haemorrhage (Carsten Kirkeby, pers. comm.). Mites, cestodes, and achlorophyllous algae have also been detected in skin lesions in deceased aquatic caecilians (Flach *et al.*, 2020). Hartigan *et al.* found parasitic myxozoan infections in *Typhlonectes* (Hartigan *et al.*, 2016). Kowlaski (2001) reports that neonate *T. natans* are more susceptible to fungal infections at temperatures lower than 25.6°C. The fungal dermatitis has been observed as multiple raised, white cotton wool like spots on the surface of the skin. Fungal dermatitis can be diagnosed by microscopic examination of a fresh skin scrape (Javier Lopez, pers. comm.). Fungal dermatitis has been treated successfully in *T. natans* using itraconazole.



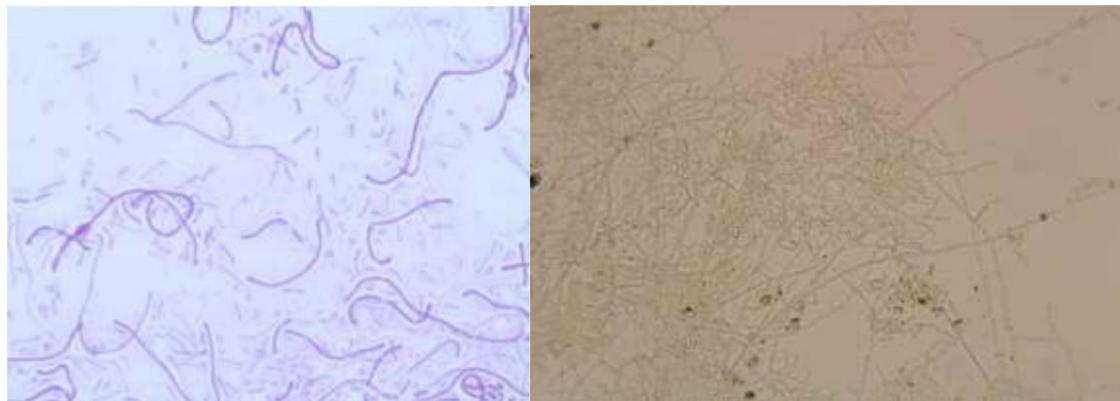
Figure 31. White cotton-like cutaneous lesions associated with fungal organisms, noted ante-mortem in a *Typhlonectes natans*.

A variety of bacteria have been also isolated from cutaneous lesions in captive caecilians including *Acitenobacter johnsoni*, *A. junii*, *A. sobria*, *Aeromonas hydrophila*, *Chromobacterium violaceum*, *Citrobacter freundii*, *Comamonas testosteroni*, *Fusarium* sp, *Moraxella nonliquafaciens*, *Pseudomonas aeruginosa*, *P. alcaligenes*, *P. fluorescens*, *Streptococcus* sp. and *Vibrio alginolyticus* (Mylniczenko, 2006; Flach *et al.*, 2020).

Septicaemia has been described as a cause of morbidity and mortality, specimens showed ventral erythema, especially noticeable in the cloacal disc, and lethargy ante mortem. Bacteria isolated from coelomic cavity and blood included *Aeromonas hydrophila*, *Bacillus* sp., *Brevimundis vesicularis*, *Citrobacter freundii*, *Enterobacter cloacae*, *Enterococcus* sp, *Escherichia coli*, *Morganella morganii*, *Providencia* sp, *Staphylococcus epidermidis* and *Vibrio alginolyticus* (Mylniczenko, 2006). Septicaemia has been treated with a variety of parenteral antibiotics; the bacteria mentioned may provide a guide for the specific antibiotic selection in absence of culture and sensitivity results.



Figures 32. Dermatitis post-mortem with intralesional fungi and bacteria observed in histology.



Figures 33. Stained and non-stained fungal hyphae, suspected to be *Saprolegnia* sp obtained from a skin scrape in an animal affected by dermatitis.

Neotropical caecilians, including aquatic ones, are known to be sometimes infected with the amphibian chytrid fungus *Batrachochytrium dendrobatidis* (*Bd*) in nature (Rendle *et al.*, 2015; Lambertini *et al.*, 2017), but lethal chytridiomycosis has not been proven and the effects of this pathogen on this Family of caecilians have not been established. *T. compressicauda* and *P. kaupii* specimens were found *Bd*-positive and it has been hypothesised that aquatic caecilians might be *Bd* reservoirs (Lambertini *et al.*, 2017; Flach *et al.*, 2020). Mortality associated with chytridiomycosis infection has been documented in aquatic caecilians (Raphael and Pramuk, 2007; Churgin *et al.*, 2013; Rendle *et al.*, 2015), and in other caecilian families at low genomic equivalent values (Gower *et al.*, 2013). Temperatures of 32.2°C for 72 hours cleared chytrid infection in *T. natans* (Churgin *et al.*, 2013). Itraconazole treatment successfully cleared chytrid infection in the dermophid caecilian *Geotrypetes seraphini* (Rendle *et al.*, 2015). Newly imported *P. kaupii* that had not been in contact with any other amphibians between the time of collection and screening have tested positive for *Batrachochytrium dendrobatidis* (Rendle *et al.*, 2015) and *P. kaupii* that underwent a prophylactic 30-minute immersion in a 0.01% solution of the antifungal itraconazole for a period of 11 days did not exhibit any discernible side effects (Rendle *et al.*, 2015). Chytrid has also been identified in asymptomatic wild species, routine quarantine and chytrid screening should be part of animal transfers between institutions.

The so-called “salamander chytrid fungus” (Martel *et al.*, 2013), *Batrachochytrium salamandrivorans* (*Bsal*) is believed to attack Urodela only, and was not detected when screened for in a captive *T. natans* (n=1, Marquis *et al.*, 2019). Moreover, an exposure experiment on a reduced number of *T. compressicauda* (n=2) demonstrated that the species is resistant to *Bsal* (Martel *et al.*, 2014). A phylogenetic ancestral state reconstruction subsequently suggested that all caecilians might be resistant to *Bsal* (DiRenzo *et al.*, 2021).

Toxicity has been reported associated with some therapeutic agents including malachite green, methylene blue (Clayton & Mylniczenko, 2015) levamisole, flubendazole (see Therapeutics). Environmental toxicity due to ammonia and copper has been described (Mylniczenko, 2006).

Lids of aquaria must be heavy and extremely tight fitting. Aquatic amphibians are escape artists: they are prone to trying to squeeze through small gaps and in certain materials, such as nylon fishing line used to attach plants to wood or rocks, or apertures in items with projections that might act as barbs, animals may become stuck and damage their skin. Damaged skin is, like burns, extremely susceptible to infection. Moreover, if they escape, they may rapidly desiccate and die. Recovered escapees may require intracoelomic fluid administration, placement in shallow baths with electrolytes (*e.g.* Amphibian ringers) and administration of antimicrobials to prevent secondary infections due to the damage of the cutaneous barrier.

Burns from animals wrapping around exposed aquarium stick heaters are a common issue in these animals if heaters are not properly guarded. Burns are extremely susceptible to infection and in many cases invasion of *Saprolegnia* and environmental bacteria rapidly overwhelm and kill the animal even if the burn appears superficial (C. Michaels, pers. com.).

Trauma caused by conspecifics or enclosure furniture, has been described as part of captive caecilian morbidity (Mylniczenko, 2006). Wounds may take several weeks to heal by second intention, requiring an increased hygiene and prophylactic antimicrobial treatment to prevent secondary bacterial infections. Kowalski (2001) reports that temperatures as high as 32.2°C can cause thermal stress in *T. natans*. Data in Table 3 should be used as a guide when maintaining *T. natans* in captivity.

Cloacal prolapse has been described in caecilians, leading in some cases to mortality due to additional complications (Mylniczenko, 2006).

Renal (tubular ectasia, glomerulonephritis, nephritis and polycystic kidneys) and hepatic pathology (hepatic lipidosis, hepatitis) have been described during post-mortem examination (Mylniczenko, 2006). Renal disease was cited to be the most common cause of death in captive *T. natans* at three different institutions in the United States (Kane *et al.*, 2021)

Uterine rupture during pregnancy leading to secondary septicaemia has been observed in a *T. natans*, oxytocin was used ante-mortem in this specimen (Barbon *et al.*, 2017).

#### 2.7.4 THERAPEUTICS

No pharmacokinetic and pharmacodynamic studies have been carried out in caecilian species. Caecilians are reported to have lower metabolic rates than other amphibian species (Stiffler & Talbot, 2000), this may need to be taken into consideration while extrapolating dosages from other species.

A variety of antibiotics has been used without any obvious side effects including amikacin 5 mg/kg every 72 hours intramuscularly (IM) and ceftazidime mg/kg 20 mg/kg IM.

Subcutaneous injections of enrofloxacin at 10mg/kg have been used for both for normal and broken-down surgical wounds. Daily 6-hours enrofloxacin baths (8 mg/litre) for three weeks has been used for treatment of dermatitis but this dose shows a significant discrepancy with dose suggested in the literature; 500mg/litre every 6-8 hours once a day, although it is necessary to point out that water baths containing antibiotics may not provide as consistent distribution as parenteral administration (Whitaker & McDermott, 2018)

Itraconazole 0.01 % baths once a day 10-30 minutes/day for 10-11 days has been used successfully for treatment of fungal dermatitis and chytridiomycosis.

Meloxicam 0.2 mg/kg IM has been used intramuscularly post operatively.

A product containing essential oil from the Cajeput tree (*Melaleuca leucadendra*) (Melafix, API) has been as an antibacterial at just over half the recommended dose (following manufacturer's guidelines) with *T. natans* with open wounds with no reported problems.

Levamisole 200-300 mg/L immersion bath for over 12 hours caused flaccid paralysis in two specimens in a group of six *Typhlonectes natans*, but shorter exposure times did not have any deleterious effects (Mylniczenko, 2006).

Mortality was observed in a group of *T. natans* when they were placed in a tank that had previously been treated with flubendazole, despite the tank having two 90% water changes before the addition of the animals. Toxicity was suspected, although conclusive evidence was not found in post-mortem examination.

Fungal dermatitis has been observed in aquatic caecilians. Kowlaski (2001) reports that neonate *T. natans* are more susceptible to fungal infections at temperatures lower than 25.6°C. The fungal dermatitis has been observed as multiple raised, white cotton wool like spots on the surface of the skin. Fungal dermatitis can be diagnosed by microscopic examination of a fresh skin scrape (Javier Lopez, pers. comm.). Fungal dermatitis has been treated successfully in *T. natans* using Itraconazole baths (3 parts Itraconazole to 100 parts water). The specimen concerned was bathed for five minutes daily for eleven days.

Traumatic wounds can take several months to heal. Daily 6-hour baths of Enrofloxacin for a period of three weeks have been successful in treating such injuries.

### 2.7.5 ANAESTHESIA AND EUTHANASIA

Chemical immobilization is reported to be achieved with buffered (to a neutral pH) tricaine methanesulfonate at 1-3 grams per litre for induction and maintained with 100-200 milligrams per litre. (Whitaker *et al.*, 1999).

Anaesthesia to carry out three caesarean sections in two *T. natans* used only 1 gram per litre for induction and no anaesthetic for maintenance (Barbon *et al.*, 2017). Three baths were prepared prior to the surgery; one with 1 gram of tricaine methanesulfonate per litre of tank water, buffered at 7.2 pH with sodium bicarbonate. A second bath was similarly prepared with 0.5 g in 1 litre of water and one containing amphibian ringer solution (1 L of distilled water, 6.6 g NaCl, 0.15 g KCl, 0.15 g CaCl<sub>2</sub>, and 0.2 g NaHCO<sub>3</sub>). Baths were prepared in trays containing about 1 cm depth of fluid and maintained at 26-28 C. Oxygen at 1 litre per minute was perfused into the baths.

Anaesthesia induction was achieved by placing animals in a clear plastic bag with anaesthetic solution at 1 gram per litre. Loss of the righting reflex occurred between 6 and 8 minutes, following induction animals were kept in amphibian Ringers' bath without anaesthetic agent.



The initial exposure to anaesthetic agent produced surgical anaesthesia for 20-40 minutes; in one case pain response was noted while closing the body wall, which required placing the animal in the 0.5 gram per litre bath while the wound was being sutured. The animals recovered the righting reflex between 70 and 180 minutes from the initial exposure to anaesthetic bath.



Figure 34. Surgical set up with multiple baths containing different anaesthetic concentration and no anaesthetic.

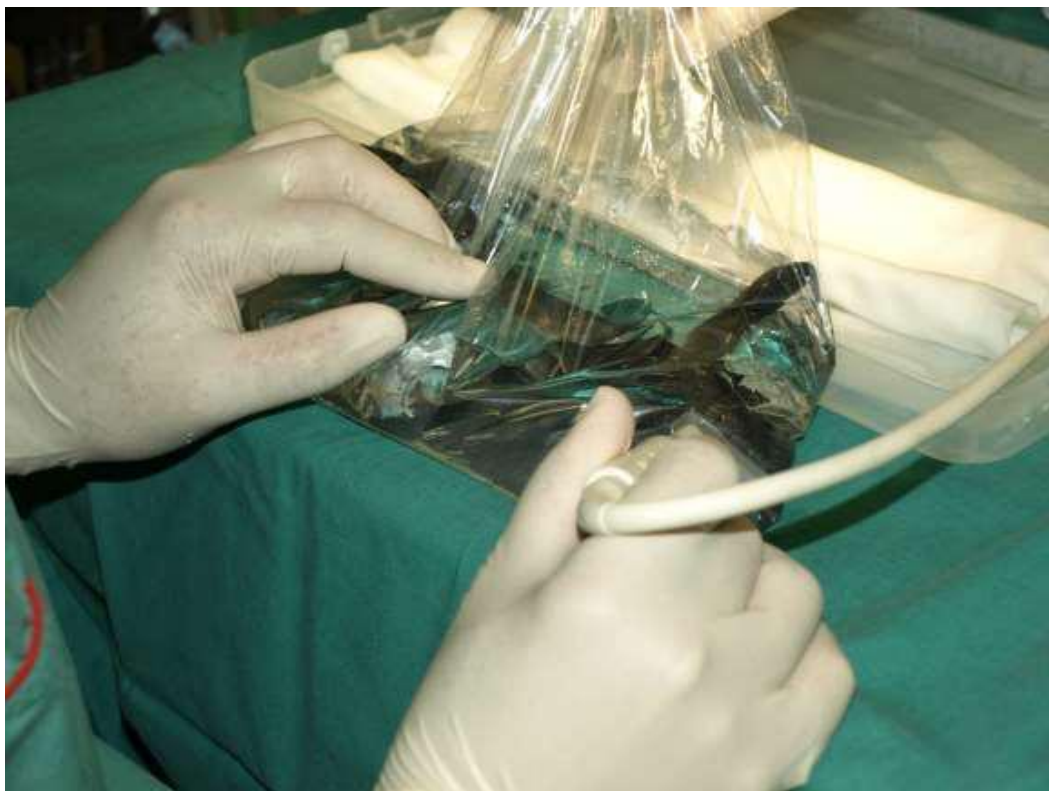


Figure 35. Determination of basal heart rate prior to anaesthesia using ultrasound.



Figure 36. Heart rate monitoring during anaesthesia using ultrasound.

Basal heart rate was between 72 and 75 beats per minute, while during anaesthesia to 50-55 beats per minute. No spontaneous respirations were observed during these procedures; in *T. natans*, the respiratory rate has been reported to double in response to aerial hypoxia while aquatic hypoxia did not have any effects, aquatic hypercapnia tripled the respiratory rate (Gardner *et al.*, 2000), additional research into the anaesthetic effects and physiological responses in this species is required.

The chemical tricaine methanesulfonate (Tricaine Pharmaq 1000 mg/g, Pharmaq LTD, Fordingbridge, SP6 1PA, United Kingdom) described before for anaesthesia, has been used by the authors as an agent for euthanasia and is considered an acceptable agent for euthanasia in aquatic amphibians (Wright, 2001). This chemical, dissolved in tap or distilled water is acidic, and it is recommended to neutralize the solution to a 7-pH using sodium bicarbonate before (Conroy *et al.*, 2009).

The animals can be immersed in a 1% bath of tricaine methanesulfonate for 15-30 minutes followed by an intracoelomic or intracardiac 250 mg/kg dose using the same 1% solution (Wayson *et al.*, 1976). This chemical does not appear to create artifacts or hinder histological examination of tissues (Baier, 2006). Heart activity termination can be checked visually, using a doppler or ultrasound.

### 2.7.6 SURGERY

Coeliotomy has been performed successfully to excise a portion of mineralized necrotic fat (Sykes *et al.*, 2006), caesarean sections (Barbon *et al.*, 2017) and partial enterectomy-anastomosis (Clayton & Mylniczenko, 2015).

Access to coelom is done through a paramedial incision, prominent fat bodies are present in *T. natans*, in the caudal aspect of the body that may make the access to the organs in this section of the body difficult. Polydioxanone has been used for the closure of the coelomic membrane and uterus while polydioxanone and nylon has been used to close the skin (Barbon *et al.*, 2017, Sykes *et al.*, 2006). Partial skin wound breakdown post operatively was noted in one case in which polydioxanone was used to close the skin, suspected to be caused by faster absorbable suture material degradation in an aquatic environment (Barbon *et al.*, 2017).

Wound surgical management has been described for the treatment of a tail wound in a caecilian (Poll, 2009).



Figure 37. Incision in the caudoventral portion of the body, about 10 centimetres cranial to the cloaca allowing exposure of the uterus to perform a caesarean. Notice the presence of fat bodies.

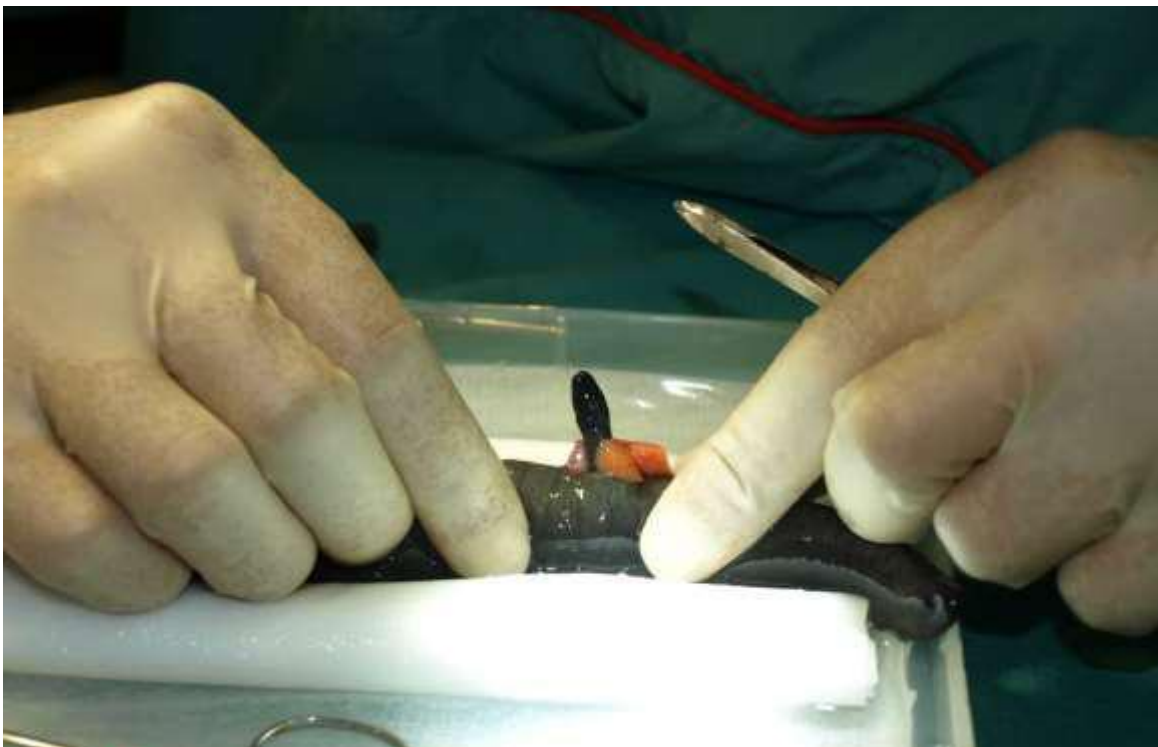


Figure 38. Applying pressure in both sides of the uterine incision.





Figure 39. Uterus closure following caesarean section, notice stay sutures on the sides of the uterine incision.



Figure 40. Skin closure using an everting pattern.



Figure 41. Neonate after extracting from uterus, notice gills that will be shed naturally, shallow water and substrate is provided to facilitate the animal taking breaths.

## 2.8 RECOMMENDED RESEARCH

Although they are more familiar to keepers and in some cases exposed to more scientific research than other caecilians, relatively little is known about the biology of aquatic caecilians. Accurate record keeping is essential and wherever possible morphometric measurements as animals mature should be taken. There is conflicting information on the reproductive triggers for aquatic caecilians in captivity. Moreover, considering the commonness of dermatitis in captivity, the epidemiology of skin diseases as well as treatment protocols require further research. Aquatic caecilians have tested positive for *Batrachochytrium dendrobatidis*, one of the causative agents of the disease chytridiomycosis, and its treatment has had mixed success. Our current state of knowledge, based on scarce evidence, leads to presume that *Bsal* is not a threat to aquatic caecilians. We strongly recommend publication of novel findings about Typhlonectidae biology and husbandry.

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APPENDIX – Post-mortem of female *T. natans*

