

Aquatic locomotion and behaviour in two disjunct populations of Western Australian tiger snakes, *Notechis ater occidentalis*

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Abstract

Adaptive phenotypic plasticity can be viewed as an evolutionary strategy that enables organisms to match their phenotypes to local conditions. I studied two neighbouring populations of amphibious fresh-water tiger snakes, *Notechis ater occidentalis*, from Western Australia. One mainland population occurs around a lake, feeds primarily on frogs, and is under strong predation pressure, whereas the second population inhabits a small offshore island with no standing water and no known predators, and feeds primarily on chicks. The availability of water and its use as a habitat differ considerably between the two populations. I compared experimentally the locomotor capacities and behaviour of individuals from both populations in water, treating neonates and adults separately. Sex did not significantly influence any trait in both populations, in either adults or neonates. At birth, the two populations exhibited very little difference in swimming speed, stamina, apnoea capacities or behaviour. However, adult mainland snakes had greater apnoea capacities and faster burst swimming speed than island snakes. Mainland adult snakes also spontaneously swam underwater for longer, and a higher proportion filled their lungs while surface swimming compared with island specimens. This study suggests strong behavioural flexibility in tiger snakes depending on habitats.

Introduction

Many studies of vertebrate adaptive evolution have focused on locomotor capacity (reviews in Bennett and Huey 1990; Garland and Losos 1994; Boggs and Frappell 2000; Irschick and Garland 2001). Locomotion is a key component of fitness (Jayne and Bennett 1990; Bauwens *et al.* 1995; Elphick and Shine 1998). Many animals rely on their locomotor apparatus to find and catch their food (Huey *et al.* 1984; Seigel *et al.* 1987), to seek partners and compete with potential rivals (Bonnet *et al.* 1998) and for predator avoidance (Brodie 1989; Losos and Sinervo 1989; Garland and Losos 1994). Thus, locomotor capacity is ecologically relevant and a potential target for natural selection (Huey and Stevenson 1979; Hertz *et al.* 1988). Locomotor capacities have been shown to be highly plastic in vertebrates (Steinhaus 1933; Jones *et al.* 1977; Losos 1990; Parichy and Kaplan 1995; Losos *et al.* 2000; Nelson *et al.* 2003), enabling organisms to match their phenotypes to local conditions (Bradshaw 1965; Stearns 1992; Via *et al.* 1995; Pigliucci 2001). For instance, some lizards show tremendous developmental plasticity, and can adjust the length of their hindlimb to suit the diameters of the perches on which they climb, thus optimising their locomotor performance (Losos *et al.* 2000).

Phenotypic plasticity is likely to be observed in variable environments (see references cited above). This is the case for animals that live at the interface of two environments, for example amphibious animals, as those have to move effectively in a variety of situations that require different locomotor patterns (Gans 1974; Shine *et al.* 2003). An interesting case

occurs in amphibious freshwater snakes (Boulenger 1913; Scribner and Weatherhead 1995), because the degree of aquatic versus terrestrial life may be variable depending on species, individuals, climatic variations over time (flood, drought), location, or because the aquatic habitat is not utilised. In those snakes, one may imagine that due to such variability, natural selection has resulted in a high level of plasticity in order to enhance their locomotor performances, either morphologically or/and behaviourally at terrestrial or aquatic locomotion, or at both (Boulenger 1913; Levins 1968; Via and Lande 1985; Sultan and Spencer 2002).

Despite their superficial similarity, crawling and swimming in snakes differs in many ways (Cundall 1987). In terrestrial lateral undulatory locomotion, the propulsive forces are generated by the lateral surfaces of the body as they push against irregularities on the substrate. In contrast, a swimming snake moves forwards by accelerating portions of the surrounding water; the reaction to this effect produces progression (Gans 1975; Jayne 1985; Cundall 1987). For instance, muscle structures that optimise swimming in snakes may differ from those that optimise terrestrial movement (Jayne 1982). Aquatic habits in snakes have also generated cardiovascular adaptations in response to gravity forces compared with terrestrial or arboreal life styles, including modifications of blood volume and pressure and location of the heart in the body (Lillywhite 1987; Seymour and Lillywhite 1987; Lillywhite *et al.* 1993; Young *et al.* 1997).

Most populations of the Australian tiger snakes *Notechis scutatus* and *N. ater* (Schwaner 1985) depend on wet environments for food sources (mainly frogs: Shine 1977, 1987; Shine and Shetty 2001; Aubret *et al.* 2004; occasionally tadpoles and fish: Mirtschin and Davis 1992). They also use water as an escape route (Shine 1977, 1987). However, the relative dependence on water (degree of aquatic life) in those populations is highly variable. For instance, Kreft's tiger snakes (*Notechis ater ater*) are highly aquatic and feed on tadpoles that they chase underwater (Mirtschin and Davis 1992). In contrast, other populations lack permanent standing water, such as the population on Carnac Island (*N. ater occidentalis*) (Bonnet *et al.* 1999, 2002; Aubret *et al.* 2004). Because of their large distribution (southern regions of Australia), tiger snakes are exposed, both within and among populations, to a relatively wide range of environments but they are genetically very similar (Scott *et al.* 2001). Thus, tiger snakes provide an opportunity to evaluate the degree of plasticity exhibited in aquatic locomotor performances and antipredator behaviour by comparing populations that live in radically different environments.

I studied two populations of tiger snakes from Western Australia (*Notechis ater occidentalis*). One population (Herdsman Lake; HL) occurs around a lake, feeds mainly on frogs, and is under strong predation pressure, whereas the other population occurs on a small offshore island (Carnac Island; CI), where there is no free water, frogs are absent and there are no known predators (Bonnet *et al.* 2002; Aubret *et al.* 2004). I tested the swimming abilities and behaviour in both naïve animals (at birth) and adults between the two populations. Under the hypothesis that habitats may have a significant effect on phenotypes (including behaviours), it is expected that HL adults will be faster swimmers, will be able to remain underwater for longer periods, and will utilise water as an escape route.

Methods

Study sites

Carnac Island (32°07'S, 115°39'E) is located 10 km off the coast of Perth, Australia. The island (total area of 16 ha) was formed ~6000 years ago by rising sea levels (Seddon 1972). There are no fresh water sources

available to the snakes except rainfall or dew. No frogs occur on Carnac, and skinks and mice constitute the major diet components of neonate and juvenile snakes while adults feed mostly on silver gull (*Larus novaehollandiae*) chicks (see Bonnet *et al.* 1999 and Aubret *et al.* 2004 for details). There are no known predators on CI, at least for adults (Bonnet *et al.* 2002).

Herdsmen Lake (31°55'44"S, 115°48'19"E) is a nature reserve in Perth (25 km away from CI). This perennial lake constitutes a favourable habitat for several frog species (*Litoria moorei*, *Crinia insignifera*; Bush *et al.* 1995). *Litoria moorei* is a tree frog that occurs in damp places such as swamps and watercourses. In Perth, it is generally resident around suburban lakes, in flooded melaleucas, lawns and in areas of dense reed cover. *Crinia insignifera* is found in permanent and seasonal wetlands and is considered to be terrestrial. Those two species are largely consumed by HL tiger snakes (Aubret *et al.* 2004: frogs comprised 62.5% of prey items). Kookaburras, domestic and feral cats, and humans (Aubret *et al.* 2004) are the main predators of HL snakes. A three-year study on this site found that tiger snakes are often spotted basking at the edge of the water or swimming, but just as many are found in the surrounding forests, which extend a substantial distance (several hundred metres) from the lake (unpublished data). In order to avoid potential confounding effects (swimming skills could be at least partially learnt, for instance, or subpopulations may also genetically diverge), all HL tiger snakes involved in the present study were captured along the edge of the lake.

Individuals from the two populations have been recently sampled for DNA analyses (1999, 2000). The results failed to detect any divergence (less than 0.3% in a matrix of genetic distances (5 mitochondrial genes, 4825 base pairs) between CI and other Western Australian mainland tiger snakes (including HL): Scott *et al.* 2001). Despite their genetic similarities, the populations live in very different environments and exhibit marked phenotypic divergences (Bonnet *et al.* 2002), suggesting a strong effect of phenotypic plasticity.

Animal collection

In January and February 2002, 75 adult snakes were captured (24 males and 17 females from CI; 16 males and 18 females from HL). Snakes were sexed by eversion of the hemipenes, measured in snout-vent length (SVL; precision ± 0.5 cm) and body length (BL), and their body mass (BM; precision ± 0.1 g) weighed with an electronic scale.

Between 17 March 2002 and 18 May 2002, another 13 wild-caught females (9 from HL and 4 from CI) gave birth in the laboratory. All were maintained until parturition in a controlled-temperature room (27°C by day and 20°C by night). Neonates were weighed and measured rapidly after birth (same measurements as above) and placed in individual plastic boxes with water container, shelter and paper towel. I randomly chose 89 neonates for experiments: 54 from HL (9 different litters) and 35 from CI (4 different litters).

All experiments were performed at 27°C, as the average body temperature chosen by both HL and CI snakes in captivity is $26.8 \pm 0.7^\circ\text{C}$ (Ladyman and Bradshaw 2003). In all tests, water temperature was 27°C.

Although video-taping is best suited for studies on locomotor performances, this option was not available. However, special attention was paid to overcome accuracy issues by systematically performing test replication with two recorders.

Swimming performances and behaviour in neonates

Swimming tests started when neonates were 10 days of age and were spread over 5 days in the same order for each individual. Neonates were not fed until the completion of the tests.

- (1) Free swimming trial. Snakes were removed from their plastic container by hand and dropped from 5 cm above the water level in a circular track (average diameter of 94 cm) containing 15-cm-deep water. Snakes started to swim instantaneously around the track. From this point, snakes were free to swim or rest in the track for a trial of 2 min. Two observers independently recorded the number of laps completed, and the time spent swimming versus resting (percentage of activity). In many cases, the snake's bodies were subject to shaking while swimming, presumably due to balance or muscular coordination problems, or perhaps due to difficulties in maintaining appropriate buoyancy (see Jayne 1985). I therefore recorded the frequency of shaking behaviour, as well as the level of buoyancy exhibited by the snakes while surface swimming (filled lungs or not): snakes normally fill up their lungs while swimming to adjust their buoyancy (parts of the lung are used as an hydrostatic organ in sea snakes: Heatwole 1978), though in the present study, many did not, resulting in snakes swimming head and neck in the air with their body underwater. As swimming underwater may be a way to flee from predators (Weatherhead and Robertson 1992) it was also noted when snakes deliberately swam underwater during the trial.

- (2) Forced swimming trial. Two days later, the test was repeated. This time, neonates were kept swimming by touching their tail with a pencil every time the snake stopped. Number of laps completed was recorded and swimming speed was calculated.
- (3) Sprint swimming speed. Snakes were picked up by the head and then dropped in the water at one end of a linear swimming track (glass aquarium, 120 cm long by 12 cm wide). This mimicked a situation where a snake had to flee a predator under maximum stress. Time was recorded along an 84-cm section of the pool to make sure full speed was reached. For accuracy, two observers independently recorded the time taken to cross the section and a mean value was recorded. This test was done three times on three consecutive days for each neonate and the best performance was kept for analysis. Burst swimming speed was calculated for each individual, and underwater swimming noted.
- (4) Voluntary apnoea. I used an opaque PVC tube (10 cm length and 2.5 cm diameter), closed at one end. The diameter of the tube was large enough to allow easy movement of the snake inside it. Snakes were removed from their cage, and the open end of the PVC tube was presented to them. Snakes always spontaneously entered the tube as they normally feel safe in such confined environments. Time started when the tube was fully submerged in the pool, and stopped as snakes chose to reach the surface. The observer was directly in sight of the snake above the water so this test aimed at putting the snake under predation pressure. The test was repeated on three consecutive days, and the best score kept for analysis.

Swimming performances and behaviour in adults

As pregnancy may influence locomotor performances in squamates (Shine 1980; Seigel *et al.* 1987), no reproductive females were tested (the presence of eggs was checked by gentle palpation of the abdomen). Food (dead mice) was provided approximately every 3 weeks. As ingested food may also influence locomotor abilities in squamates (Garland and Arnold 1983; Shine and Shetty 2001), I waited for snakes to have fully digested prey prior to experiments.

A rectangular swimming pool 370 cm long, 64 cm wide, and 100 cm deep was lined with black plastic. The 64 cm width was adjusted to allow normal swimming waves of the largest snake involved in the study (i. e. amplitude of waveform less than 64 cm). Water depth (40 cm) was constant in all experiments, and prevented the largest snake from touching the bottom. Two observers recorded all parameters, placed at opposite ends of the pool. Each test was separated by 2 days and performed in the same order for each individual, as follows:

- (1) Free swimming trial. Snakes were gently dropped from ~15 cm above the water level at one end of the pool. Each snake would start to swim towards the opposite end. During a 5-min trial, I recorded the total time spent swimming (versus resting; percentage of activity) and the distance travelled by each snake (cumulated number of length and width of the pool). No snakes swam on random trajectories in the pool, but all showed stereotypic patterns: fast swimming along the length of pool and slow turns at each ends. Thus, records of numbers of length and width travelled provided relatively accurate estimation of the total distance swum by each snake. Intermediate times of each length swum were also recorded. As for neonates, I recorded the level of buoyancy exhibited by snakes, the occurrence of shaking and the occurrence of underwater swimming.
- (2) Forced swimming trial. The 5-min trial was repeated but snakes were kept swimming by gently touching their tail with a stick each time they stopped. Cumulated shaking times and cumulated underwater swimming times were recorded.
- (3) Sprint swimming speed. As for neonates, maximum swimming speed was recorded by putting the snakes in a situation where they had to flee a predator under maximum stress: snakes were picked up by the head and then dropped in the water at one end of the pool. For accuracy, two observers independently recorded the time taken to cross a 280-cm section of the pool and a mean value was calculated, and underwater swimming was also noted.
- (4) Voluntary apnoea. I used the same protocol for adults as described for neonates, using a larger PVC tube though (40 cm long with a diameter of 8 cm).

Statistics

A body condition index was calculated for each snake in adults and neonates separately, using the residual values of the linear least-squares regression of $\log(\text{BM})$ against $\log(\text{SVL})$. Such an index provides accurate estimates of body reserves (fat, muscle) in snakes (Bonnet and Naulleau 1994).

The assumption of normal distribution was tested for each variable using Kolmogorov–Smirnov tests, and all were normally distributed after log-transformation. Therefore, parametric tests were used. However, results were unchanged with non-parametric tests.

Among the 75 adults used for the swimming trials, 3 males and 4 females from HL had tail injuries. Because tail length may influence swimming speed (Jayne and Bennett 1989; Shine and Shetty 2001), those animals were excluded from the analysis of all traits (except for apnoea time and underwater swimming time, in which any effect of tail injury is unlikely).

Because neonates were obtained from few pregnant females, I used mixed-model ANOVA and ANCOVA to compare both morphological and swimming traits in neonates, in order to assess the ‘mother’ effect as a variance component. ‘Mother’ was thus used as a random effect. All statistics were performed using STATISTICA 5.0.

The experimental design I used did not enable corrections for multiple comparisons to be performed. There are no simple solutions to correcting for multiple comparisons since there is a structural trade-off in the two types of errors made (α -errors versus β -error) as one adjusts the P -values used as the criteria for statistical significance. Consequently, I acknowledge that some of my conclusions may be affected by the absence of corrections for multiple comparisons. However, even if some results may not remain significant after corrections, the main results for which strong effects were found (no difference between neonates versus many differences between adults) are not likely to be altered.

Results

Morphological comparison between HL and CI

CI neonates were larger at birth than HL neonates in BM and BL but did not significantly differ in body condition (Table 1). As previously reported (Bonnet *et al.* 2002; Aubret *et al.* 2004), CI adult snakes were, on average, larger than HL adult snakes (see Table 2).

Influence of sex on swimming performances in neonates

There was no significant effect of sex (two-way ANOVAs with sex and origin as factors, $77 < n < 80$, all $P > 0.15$) in the percentage of activity, distance swum (body length travelled), swimming speed (body length travelled per second) in the free swimming trial; in the swimming speed (body length travelled per s) in the forced swimming trial; in the apnoea time relative to BM; and in the burst swimming speed (body length per second).

Table 1. Morphological traits and swimming performance comparison between neonate tiger snakes from Carnac Island and Herdsman Lake

Mean values \pm s.d. are given; mixed-model ANOVAs were used. BL, body lengths

	Carnac Island	Herdsman Lake	F	d.f.	P
Traits					
Body mass (g)	6.62 \pm 1.02	4.22 \pm 0.86	32.5	1,10.3	0.018
Body length (cm)	24.35 \pm 1.51	19.86 \pm 1.60	14.9	1,10.5	0.003
Body condition index	-0.001 \pm 0.044	0.001 \pm 0.047	0.1	1,7.1	0.93
Burst speed (cm s ⁻¹)	26.91 \pm 7.46	21.16 \pm 6.50	2.2	1,7.8	0.17
Burst speed (BL s ⁻¹)	1.09 \pm 0.30	1.06 \pm 0.31	0.1	1,7.5	0.78
Free swimming trial					
Distance swum (m)	2.82 \pm 1.95	3.09 \pm 1.64	0.6	1,5.3	0.47
No. of BL travelled	11.51 \pm 7.78	15.58 \pm 8.46	2.5	1,5.0	0.18
Activity (%)	54.20 \pm 24.14	52.01 \pm 26.10	0.1	1,6.5	0.84
Mean swimming speed (cm s ⁻¹)	4.22 \pm 2.01	5.59 \pm 2.94	2.0	1,5.6	0.21
Mean swimming speed (BL s ⁻¹)	0.17 \pm 0.08	0.28 \pm 0.14	8.2	1,4.8	0.037
Forced swimming trial					
Mean swimming speed (cm s ⁻¹)	7.35 \pm 2.28	4.75 \pm 1.59	8.0	1,6.8	0.026
Mean swimming speed (BL s ⁻¹)	0.30 \pm 0.09	0.24 \pm 0.08	4.3	1,5.1	0.10

Table 2. Body size and swimming performances in adult tiger snakes from Carnac Island and Herdsman LakeMeans \pm s.d. are given (adjusted means \pm s.d. when a covariable was used). BL, body lengths

	Carnac Island	Herdsman Lake	<i>F</i>	d.f.	<i>P</i>
Traits					
Body mass (g)	377.17 \pm 132.63	202.97 \pm 65.07	38.0	1,73	0.0001
Body length (cm)	86.17 \pm 11.78	75.42 \pm 9.68	16.1	1,73	0.0001
Body condition index	0.039 \pm 0.078	-0.039 \pm 0.067	25.4	1,73	0.0001
Burst swimming speed (cm s ⁻¹)	60.56 \pm 16.34	63.52 \pm 17.34	0.4	1,59	0.54
Burst swimming speed (BL s ⁻¹)	0.60 \pm 0.16	0.72 \pm 0.19	5.8	1,59	0.019
Free swimming trial					
Distance swum (m)	56.97 \pm 21.02	43.09 \pm 20.99	5.0	1,55	0.029
No. of BL travelled	57.59 \pm 23.38	50.78 \pm 26.12	1.4	1,55	0.23
Activity (%) ^A	80.20 \pm 19.62	66.38 \pm 27.77	7.6	1,55	0.008
Mean swimming speed (cm s ⁻¹)	23.50 \pm 6.73	19.47 \pm 5.08	4.9	1,55	0.030
Mean swimming speed (BL s ⁻¹)	0.23 \pm 0.073	0.23 \pm 0.069	0.1	1,55	0.71
Forced swimming trial					
Shaking time (s) ^A	23.06 \pm 20.10	9.18 \pm 11.64	9.5	1,62	0.003
Underwater swimming (s)	26.72 \pm 33.13	88.01 \pm 95.16	4.9	1,68	0.031

^ARelative to BL.

That is, the way in which sex affected swimming performances did not significantly differ in HL and CI populations.

However, this analysis did not take into account the effect of the 'mother' variable. I therefore conducted separate analyses within each population to evaluate this potential effect using mixed-model ANOVA and ANCOVA (with Sex as fixed factor, Mother as random factor, the different swimming traits as dependent variables, and BL or BM as covariate when applicable). Within HL neonates, the analysis revealed a tendency in females to swim further than males in the free swimming trial (absolute value, $F_{1,3,9} = 6.01$, $P = 0.071$), and also in body lengths travelled (respectively 20.62 ± 9.23 versus 12.65 ± 6.67 , $F_{1,3,8} = 7.52$, $P = 0.054$). For all other swimming traits, there was no significant effect of sex on locomotor abilities (all $P > 0.09$).

Within CI neonates, there was no significant effect of sex on locomotor abilities or any of the swimming traits (all $P > 0.62$). Sexes were therefore pooled for further analysis.

Influence of body size and origin on swimming performances in neonates

Apnoea time, mean swimming speeds, distance swum or percentage of activity (free swimming trial) were not correlated with BM and BL (Spearman's rank correlations, $n = 82$, all $r < 0.12$, all $P > 0.26$). However, swimming speed in the forced swimming trial was positively correlated with BM ($n = 85$, $r = 0.59$, $P < 0.0001$) and BL ($n = 82$, $r = 0.58$, $P < 0.0001$). Burst swimming speed was positively correlated with BM ($n = 82$, $r = 0.40$, $P < 0.0002$) and BL ($n = 82$, $r = 0.39$, $P < 0.0003$). Maximum apnoea time reached 34 min and 13 s in HL neonates and 32 min and 47 s in CI neonates. Considering all neonates, underwater swimming was significantly faster than swimming at water level in the sprint swimming test (respectively 1.32 ± 0.30 BL per second versus 1.05 ± 0.37 BL per second, $F_{1,48} = 4.67$, $P < 0.036$).

The locomotor performances of neonates of both populations were very similar (Table 1). However, HL neonates exhibited a significantly higher mean swimming speed in

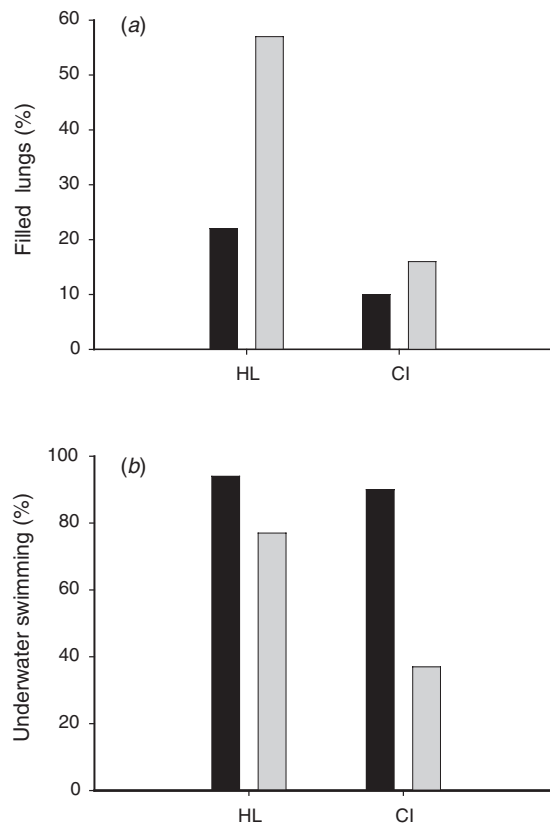


Fig. 1. Graphs show (a) the proportions of snakes that fled underwater during the burst swimming speed test, and (b) filled their lungs while surface swimming (thus excluding trials in which individuals swam underwater). The proportion of snakes that swam underwater during the sprint tests differed in HL snakes between adult (grey bars) and neonates (black bars) ($\chi^2_{(df=1)} = 7.42$, $P < 0.007$), but not in CI snakes ($\chi^2_{(df=1)} = 0.01$, $P = 0.97$). Finally, most neonates and adults from HL filled their lungs while surface swimming ($\chi^2_{(df=1)} = 1.87$, $P = 0.17$). In CI snakes, the proportion significantly decreased in adults ($\chi^2_{(df=1)} = 18.32$, $P < 0.0001$).

the free swimming trial (BL per s), and CI neonates were faster swimmers in the forced swimming trial (significantly in absolute values, but not relative to BL; see Table 1). No significant difference was found in respect to the behaviours recorded: similar proportions of snakes deliberately swam underwater during the free swimming trial (20% in HL versus 7% in CI, $\chi^2_{(df=1)} = 0.43$, $P = 0.51$), and in the sprint swimming test ($\chi^2_{(df=1)} = 2.67$, $P = 0.10$) (Fig. 1a). In all, 54% of HL neonates versus 69% of CI neonates were subject to shakings while swimming ($\chi^2_{(df=1)} = 0.39$, $P = 0.53$), and filled their lungs while surface swimming ($\chi^2_{(df=1)} = 0.13$, $P = 0.71$) (Fig. 1b).

Influence of sex on swimming performances in adults

There was no significant effect of sex (two-way ANOVAs with Sex and Origin as factors, $53 < n < 78$, all $P > 0.34$) in the percentage of activity, burst swimming speed (body length per second), distance swum (number of body lengths), mean swimming speed (body length per second), apnoea time (relative to BM), underwater swimming time, and shaking time (relative to BL). Therefore sexes were pooled for subsequent analysis.

Influence of body size and origin on swimming performances in adults

For all snakes pooled, apnoea time was negatively correlated with BM ($r = -0.26$, $n = 73$, $P < 0.022$) and SVL ($r = -0.25$, $n = 73$, $P < 0.030$). HL individuals spent significantly longer time in apnoea than CI snakes (see Table 2) even when BM or SVL effects were taken into account (ANCOVA with Origin as factor, apnoea time as dependent variable and

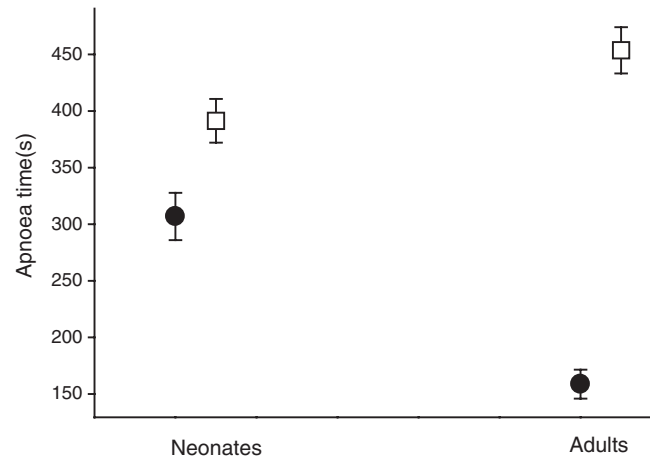


Fig. 2. Graph shows the comparison of apnoea performances at birth and adulthood in HL and CI tiger snakes. BM weighed mean values (\pm s.e.) are plotted. Mixed-model ANCOVA were applied to data for neonates to control maternal effects. Apnoea performances were similar at birth between HL snakes (open squares; 458.25 ± 368.72 s) and CI snakes (black circles; 400.77 ± 374.10 s) ($F_{1,5,6} = 0.23$, $P = 0.64$). However in adults, apnoea scores were significantly higher in HL individuals (391.39 ± 372.85 s) than in CI individuals (158.71 ± 164.55 s) ($F_{1,71} = 7.29$, $P < 0.009$).

BM as covariate, $F_{1,70} = 4.49$, $P < 0.038$; and SVL as covariate, $F_{1,70} = 4.67$, $P < 0.034$). Maximum value reached 23 min and 13 s in HL snakes versus only 10 min and 53 s in CI snakes. Shaking time was positively correlated with BM ($n = 65$, $r = 0.38$, $P < 0.0016$) and BL ($n = 65$, $r = 0.37$, $P < 0.0025$). No other swimming traits were significantly correlated with BM or BL (Spearman correlations, $57 < n < 62$, all $r < 0.20$, all $P > 0.10$).

Performances comparisons are presented in Table 2 and apnoea scores in Fig. 2. HL snakes sprinted faster than CI snakes (relative to BL), and swam underwater for longer. Over the 10 first lengths swum in the track, there was no significant difference in swimming speed between HL and CI snakes (repeated-measures ANOVA, with Origin as factor, and swimming speeds (BL per second) as the repeated variable, Wilks $\lambda = 0.61$, $P = 0.23$). However, there was a significant effect of time ($F_{9,279} = 13.79$, $P < 0.0001$): as expected, swimming speed decreased over 10 successive lengths swum.

I found significant differences between populations in the proportion of snakes that filled their lungs while surface swimming in the free swimming trial ($\chi^2_{(df=1)} = 8.05$, $P < 0.005$) (Fig. 1b), and that swam deliberately underwater ($\chi^2_{(df=1)} = 11.05$, $P < 0.001$). The difference in snakes exhibiting shaking behaviour was close to the conventional level of significance ($\chi^2_{(df=1)} = 3.79$, $P = 0.051$). Finally, the proportion of snakes that swam underwater during the sprint test (i.e. under stress) was also significantly different ($\chi^2_{(df=1)} = 5.44$, $P < 0.019$) (Fig. 1a).

Discussion

This study showed that measured locomotor capacities were similar at birth between two disjunct populations of tiger snakes. However, numerous significant differences were found between adults. Although preliminary, these results suggest that from birth to adulthood,

habitat-dependent behavioural plasticity (possibly learning: Pigliucci 2001) occurs in tiger snakes.

Differences between the sexes

Faster locomotion in males relative to females seems to be widespread in many terrestrial vertebrates, including snakes (Kelley *et al.* 1997), mirroring a trend for greater relative muscle mass in male snakes than in conspecific females (Bonnet *et al.* 1998). However, some authors have suggested that the difference in locomotion between males and females may not be as great in aquatic locomotion (Scribner and Weatherhead 1995; Shine *et al.* 2003). The current results on tiger snakes support such observations, with no differences observed between males and females in locomotor performances.

Differences between habitats

Very little difference was found between the two populations at birth, neither in swimming abilities nor in the behaviour exhibited in the water. HL neonates exhibited faster routine swimming speed (relatively to BL). However, HL neonates were smaller in body size and smaller animals commonly have faster relative speeds, within a single population or ontogenetically (Jayne 1985; Jayne and Bennett 1990; Shine and Shetty 2001). Therefore, I may consider any difference in adult populations as a potential reflection of the influence (or absence) of water as locomotion venue.

A striking difference was found in apnoea time: HL adults stayed underwater more than twice as long as CI adults. It may be advantageous to remain underwater for long periods to escape predation or to enhance hunting success (frogs are the main prey of most tiger snakes populations: Shine 1977, 1987; Aubret *et al.* 2004). Furthermore, the results also suggest that apnoea performances may change during ontogeny in tiger snakes (Fig. 1), seemingly in opposite directions for HL and CI snakes. This finding suggests an ability to develop apnoea time relative to the degree of usage of the water habitat. An experimental follow-up of a number of individuals from birth to adulthood would be necessary to document that possibility. Finally, HL adult snakes exhibited faster burst swimming speed (but again, this could be a direct consequence of their smaller size) and shorter shaking time, and spontaneously swam underwater for a longer time than did CI adult snakes.

Where most neonates of both populations filled their lungs while surface swimming, many more HL adults did so than CI adult snakes. Finally, the proportion of snakes choosing to swim underwater as an escape route during the sprint swimming test (mimicking a fleeing behaviour) increased from birth to adulthood in HL snakes, whereas it decreased in CI snakes.

This study suggests high levels of behavioural flexibility in tiger snakes. Further studies need to confirm these results, although there are several limitations to this work. Notably, additional causations remain to be assessed: the existence of predators in HL but not on CI may result in the selection of better aquatic performances or water-related behaviour in HL snakes. For instance, because underwater burst swimming was faster than at water level in neonates, individuals that do not flee underwater may face a greater risk of predation. This potentially affects a number of traits measured here, such as swimming speed, the ability to swim and remain underwater, or simply the use of water as an escape route.

Finally, results and interpretations are based on only two populations, and more populations need to be sampled. This study leaves open the possibility that factors other than the presence or absence of aquatic habitats influence the variations in aquatic

locomotion and behaviour observed here. Future experimental studies in controlled environments may allow us to tease apart such confounding issues.

Thus, future work may confirm that freshwater snakes, such as *Notechis*, can develop a precise tuning of plastic responses to a semiaquatic life. The functional and behavioural significance of such plasticity remains to be supported by field data, but is likely to involve improved survival and feeding success, via decreased detection by predators and improved mechanisms of escape as well as increasing hunting efficiency. Beneficial (sometimes termed 'adaptive') plasticity is being increasingly recognised (see Schlichting and Pigliucci 1998), although few studies have provided empirical evidence to support the assumption that locomotor capacities are critical to Darwinian fitness (Garland and Losos 1994).

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