

# Exploration syndromes in the frog *Xenopus (Silurana) tropicalis*: correlations with morphology and performance?

M. Videlier<sup>1</sup>, C. Bonneaud<sup>2</sup>, R. Cornette<sup>3</sup> & A. Herrel<sup>1,4</sup>

<sup>1</sup> UMR 7179, Département d'Ecologie et de Gestion de la Biodiversité, Centre National de la Recherche Scientifique/Muséum National d'Histoire Naturelle, Paris, France

<sup>2</sup> Centre for Ecology and Conservation, College of Life and Environmental Sciences, University of Exeter, Penryn, Cornwall, UK

<sup>3</sup> UMR 7205, Origine, Structure et Evolution de la Biodiversité, Centre National de la Recherche Scientifique/Muséum National d'Histoire Naturelle, Paris, France

<sup>4</sup> Evolutionary Morphology of Vertebrates, Ghent University, Ghent, Belgium

## Keywords

locomotion; behaviour; dispersal; mobility; Anura.

## Correspondence

Anthony Herrel, UMR 7179, Département d'Ecologie et de Gestion de la Biodiversité, Centre National de la Recherche Scientifique/Muséum National d'Histoire Naturelle, 55 rue Buffon, Paris 75005, France.

Email: anthony.herrel@mnhn.fr

Editor: Mark-Oliver Rödel

Received 24 May 2014; revised 28 July 2014; accepted 28 July 2014

doi:10.1111/jzo.12170

## Abstract

Exploration behaviour is a complex trait that may have strong implications for the fitness of individuals and the persistence of populations. Understanding the different exploration strategies is necessary to understand how animals may adapt to changes in their environment including human-induced habitat fragmentation. Behavioural syndromes are often thought to characterize exploration behaviour, and within a population, individual strategies may vary from 'bold' to 'shy'. Although our understanding of behavioural syndromes has increased enormously over the past decade, little is known about the presence of such syndromes in frogs. Yet, frogs are particularly sensitive to changes in their environment because of their ectothermic physiology and low mobility. Here, we investigate the exploration behaviour of wild-caught male frogs under laboratory conditions to test whether distinct behavioural strategies exist. We demonstrate the presence of different behavioural syndromes with two of the syndromes that can be categorized as 'bold' and 'shy', and a third one that is clearly intermediate. These behavioural strategies are, however, independent of variation in morphology and locomotor performance indicating that these two components of mobility (i.e. behaviour and locomotor capacity) are decoupled and could thus respond differentially to selection on mobility.

## Introduction

Exploration behaviour was originally identified as 'an investigative behaviour of a new environment' (Scott, 1956). In natural conditions, exploration behaviour is tightly linked to dispersal and underlies the colonization of novel habitats. Dispersal and migration are important to maintain gene flow and to find reproductive partners, and to find food when resources are scarce. However, the downside of exploration is an exposure to predation (van Oers *et al.*, 2004) and the need to move through a potentially hostile environment in terms of abiotic factors (e.g. temperature, humidity). Consequently, exploration behaviour has a strong impact on fitness and is likely under strong selection in natural populations (Smith & Blumstein, 2008).

Exploration behaviour in animals is often linked to the concept of behavioural syndromes and personality traits (Cote *et al.*, 2010). Exploration syndromes have been identified in many animals (Gosling, 2001; Bell, Hankison & Laskowski, 2009) including invertebrates such as hermit crabs

(Watanabe *et al.*, 2012), mammals (Shillito, 1963; Careau *et al.*, 2008; Uher, Asendorpf & Call, 2008; von Merten & Siemers, 2012), birds (Carere *et al.*, 2005) and fish (Dingemanse *et al.*, 2007). Within this context, two syndromes are typically identified: bold and shy (Dingemanse & de Goede, 2004; Wilson & Godin, 2009). Bold individuals are those individuals that readily explore novel surroundings, show little fear and take risks by moving around. At the opposite, shy individuals do not tend to explore novel surroundings, do not move a lot and avoid risk-taking behaviour. Moreover, these personality traits have been shown to be correlated to fitness and to be variable between populations and species suggesting that they are under natural selection (Smith & Blumstein, 2008).

Thus exploration behaviour is directly related to fitness and selection on an individual's mobility. Mobility is, however, not only composed of behaviour, but is also dependent on the physiology and locomotor performance of an individual. Yet, studies linking performance abilities to personality traits are exceedingly rare (Careau & Garland,

2012) despite being essential to better understand selection on mobility in relation to modifications of the natural habitat such as habitat fragmentation. The current natural environment is becoming exceedingly modified because of global change, inducing an acceleration of the natural cycles resulting in, among others, disturbed rainfall patterns (Beaumont *et al.*, 2010; Zelazowski *et al.*, 2011). Moreover, human-induced habitat fragmentation is common in many tropical areas and selectively impacts biodiversity hotspots and imposes additional selective pressures on natural populations of animals and plants (Ferraz *et al.*, 2003; Dixo *et al.*, 2009). Yet, the effects of climate change and habitat fragmentation are not equal for all taxa. For example, ectothermic species unable to regulate their body temperature and species with low mobility will likely be most strongly affected by the processes of temperature change and habitat fragmentation (Deutsch *et al.*, 2007; Huey *et al.*, 2008; Dillon, Wang & Huey, 2010).

A group of animals particularly affected by global change and habitat fragmentation are amphibians. This group is characterized by a low overall mobility and a temperature dependence of their physiology and performance, thus often resulting in a tight adaptation to their local environment (Ernst, Linsenmair & Rodel, 2006; Hillers, Veith & Rödel, 2008). How selection on mobility because of habitat fragmentation and global change may affect amphibians, and more precisely their mobility, remains largely unknown. However, studies on the invasion of *Rhinella marina* in Australia have shown that strong selection for mobility at the invasion front resulted in changes in both behaviour and performance with subsequent profound impacts on morphology and life-history traits (Phillips, Brown & Shine, 2010; Tracy *et al.*, 2012). This suggests that selection on mobility may have large-scale cascading effects, and that mobility is an important trait.

Here, we study the exploration behaviour in wild-caught male *Xenopus (Silurana) tropicalis* under laboratory conditions to test whether different behavioural strategies exist. This species is of interest not only because it is a model system in biology, but more specifically because its natural habitat in the West African rain forest belt is becoming increasingly fragmented (Hillers *et al.*, 2008). Here, we decided to study males more specifically because in many frog species, males are more mobile than females and will move during the breeding season to find sexual partners (Wells, 1977). We analyse the movements of individuals during the exploration of a novel environment and test for the presence of behavioural syndromes. Moreover, by correlating behavioural data to data on morphology and performance, we test whether these behavioural syndromes are driven by variation in underlying physiological performance (Careau & Garland, 2012). If behaviour is decoupled from performance, then this may, for example, allow animals to circumvent constraints on the evolution of locomotor capacity (i.e. because of the presence of physiological trade-offs between burst performance and endurance capacity; Wilson, James & Van Damme, 2002; Herrel & Bonneaud, 2012a).

## Materials and methods

### Animals

We focus on mobility in *Xenopus (Silurana) tropicalis*. Individuals of three sub-populations of *X. tropicalis* were caught in Western Cameroon in 2009. Animals were transported to France and housed at the Muséum National d'Histoire Naturelle (MNHN) in Paris. Animals were maintained in 21-L tanks mounted on three-shelf stand alone *Xenopus* frogs racks (Aquaneering, Inc., San Diego, CA, USA) with the water temperature set at 24°C. This temperature is close to the optimal performance temperature of *Xenopus* (Herrel & Bonneaud, 2012a) and similar to temperatures measured under field conditions for ponds in the forest (Careau *et al.*, 2014). Animals were fed with beef heart and mosquito larvae twice weekly. All individuals were pit-tagged (Nonatec, Rodange, Luxembourg) allowing unique identification of each individual. Thirty-seven male frogs were used in the exploration behaviour experiments.

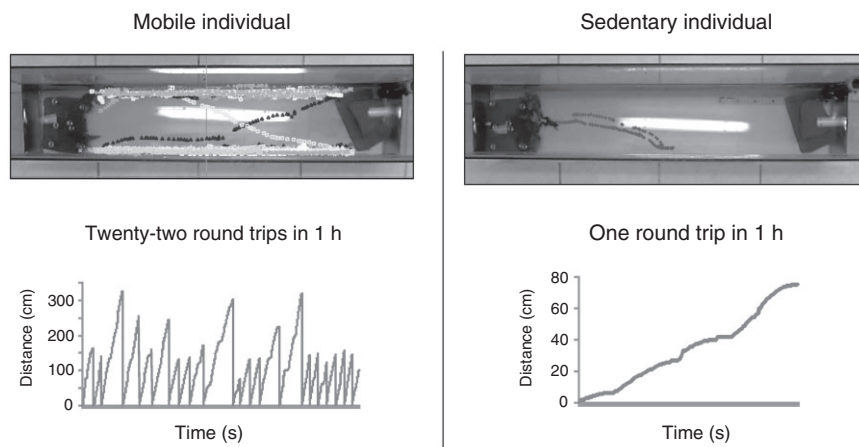
### Morphology and performance

Morphological and performance data were taken from previously published measures of the same individuals (Herrel & Bonneaud, 2012a,b; Herrel *et al.*, 2012). Performance measures included swimming velocity and acceleration, as well as terrestrial endurance capacity (time and distance jumped until exhaustion).

### Behavioural analysis

Frogs were filmed for 60 min with a Quickcam Pro 500 (Logitech, Inc. at Romanel-sur-Morges, Switzerland) set at 15 frames per second in a rectangular container (height: 0.98 m, length: 0.40 m, width: 0.20 m) with a water level of 0.20 m maintained at  $24 \pm 2^\circ\text{C}$  (Fig. 1). Animals were introduced in the tank and left quietly for 5 min before the onset of the recording. Shelters were placed at the two extremities to provide a hiding place. Each individual was tested three times at different times of the day in a randomized way (morning: 09:00 AM to 12:00 PM; early afternoon: 12:00–04:00 PM; late afternoon: 04:00–08:00 PM). This allowed us to test the repeatability of behaviour across different activity periods.

Videos were analysed using the ProAnalyst software (Xcitex, Inc., Cambridge, MA, USA) by tracking all the movements of the frogs during their exploration of the environment for 1 h (Fig. 1). Coordinates of the snout-tip were extracted and used to quantify exploration behaviour. Behavioural variables included: (1) total distance moved in 1 h (cm); (2) average, minimum and maximum speed of the movement extracted from the video ( $\text{cm s}^{-1}$ ); (3) latency to the first and second movements, and the time of the last movement (s); (4) average, minimum, and maximum time of a round trip (s); (5) time of all movements, of all movements without pauses, and the total and the average time spent hidden between two round trips (s); (6) number of complete round trips, total number of movements, and the number of pauses; (7)



**Figure 1** Figure illustrating the swimming tank (top) and the tracking of the swimming movements for 1 h for a mobile (left) and a sedentary individual (right). Note that on each side of the tank shelters are provided for the animal. Below are illustrated the displacements quantified for each of the two individuals.

frequency of movement; and (8) number of movements away from the wall of the aquarium. In total, 19 variables were extracted for each video.

### Statistic analyses

Before analyses, all data were  $\log_{10}$ -transformed to conform to assumptions of normality and homoscedasticity for parametric analyses. The repeatability of each variable was tested using Pearson correlations, we exclude five parameters that were not repeatable (i.e. no correlation across at least two of the three trials: the minimal time of a round trip, the latency to the second movement, the total time spent moving without pauses, the total and average time spent hidden between two round trips, and the number of pauses). Two outliers representing individuals with extreme behaviours were detected in an initial exploration of the dataset. Both showed no exploration behaviour at all and were consequently removed from the dataset. However, all individuals were in good health and were still alive at the time of the submission of the paper and showed no weight loss.

To classify individuals with similar exploration behaviour, a Gaussian mixtures model analysis (Banfield & Raftery, 1993) was used using the individual average of each repeatable behavioural variable. The number of groups set to two given that two types of exploration behaviour are typically recognized among animals ('shy' and 'bold'), and group membership was saved. A Gaussian mixtures analysis is well suited to detect groups based on biological data that show a multivariate normal distribution (Banfield & Raftery, 1993; Baylac, Villemant & Simbolotti, 2003). The validity of the assignment of individuals to groups was tested using a cross-validation test with a  $k$ -nearest neighbours (with  $k = 1$ ) assignment based on the training set determined by the Gaussian mixtures approach (Ripley, 1996). The same procedure was then run with three groups to test whether three groups gave a better classification than just two. In both cases, the same two individuals were misclassified suggesting that two or three groups represent the structuring of the data equally well. Based on an exploration of the raw data, we decided to retain three groups

for our subsequent analysis as group three was behaviourally distinct from the two other ones. However, analyses based on two or three groups gave highly similar results (i.e. no differences in morphology or performance). In the two-group analysis, the individuals from group three were classified as belonging to group two. All clustering analyses were performed in *R* using the *Mclust* and *Class* packages (R Development Core Team, 2013).

To test which variables differed between the clusters identified, a multivariate analyses of variance (MANOVA) coupled to univariate analyses of variance ANOVAs and *post hoc* tests with Bonferroni correction were performed (Table 1) (Hochberg, 1988). Finally, we tested whether behavioural groups differed in morphology and performance using MANOVA. All analyses were performed using IBM-SPSS (V. 15.0, SPSS, Inc., Chicago, IL, USA).

## Results

### Exploration behaviour

Male *X. tropicalis* explore their environment with a mean latency to the first movement of 592.4 s (range: 3.9–3291.0 s). While doing so they cover a distance of 15.6 m in 1 h, on average, ranging up to 76 m for the individual that moved most. In contrast, one of the individuals moved only 66 cm, illustrating strong differences in exploration behaviour among individuals. Note that two individuals that did not move at all were excluded from the dataset. On average, animals moved 35 min out of the 1 h recorded and stopped moving after 45 min. The average swimming speed during exploration is  $12.8 \text{ cm s}^{-1}$  (range:  $1\text{--}60 \text{ cm s}^{-1}$ ), remaining well below the maximal swimming speed of this species ( $1.0\text{--}2.5 \text{ m s}^{-1}$ ; see Herrel & Bonneaud, 2012b).

### Behavioural analysis

A clustering analysis using Gaussian mixtures performed on the average behavioural data for each individual retained

three significant groups. The first group is composed of 17 individuals, the second group of 15 individuals and the third group of three individuals. A MANOVA performed on the average behavioural data detected significant differences between the groups (Wilk's lambda = 0.03,  $F_{28,38} = 6.42$ ,  $P < 0.001$ ). Subsequent univariate ANOVAs showed that groups were different for most variables except for the mean, maximal and minimal speeds, and the time of the last movement (all  $P > 0.05$ ; see Tables 1 and 2). The time of a round trip, the total number of movements, the total distance moved, the total time moved without pauses and the frequency of movement were significantly different among the three groups (Table 3). Whereas the average time of a round trip and the number of movements away from the wall of the tank were similar for groups one and two, the total movement time, the number of movements away from the wall, the latency to first movement, and the maximal time of a round trip were similar for groups two and three (Table 3). In general, the first group was characterized by a high number of round trips, a large number of movements, a greater total distance moved, a shorter latency to the first movement and a higher frequency of movement. Whereas group three showed opposite characteristics, group two was generally intermediate between the two with a longer latency than group three, but a later occurrence of the last movement.

### Morphology and performance

Behavioural clusters were not significantly different in overall body size (Wilk's lambda = 0.77,  $F_{4,62} = 2.15$ ,  $P = 0.09$ ). Indeed, neither body mass ( $F_{2,32} = 0.12$ ,  $P = 0.89$ ) nor snout-vent length ( $F_{2,32} = 1.93$ ,  $P = 0.16$ ) were different between groups. Moreover, behavioural clusters were not different in head size (Wilk's lambda = 0.83,  $F_{8,58} = 0.69$ ,  $P = 0.70$ ), forelimb dimensions (Wilk's lambda = 0.67,  $F_{10,56} = 1.26$ ,  $P = 0.28$ ) and hind limb dimensions (Wilk's lambda = 0.74,  $F_{10,56} = 0.91$ ,  $P = 0.53$ ). Finally, no significant difference in locomotor per-

formance were detected among behavioural clusters (Wilk's lambda = 0.80,  $F_{10,56} = 0.65$ ,  $P = 0.76$ ).

## Discussion

### Exploration behaviour

All variables retained in the analysis were repeatable across trials despite the fact that animals were tested on different

**Table 2** Results of Bonferroni *post hoc* tests testing for differences in behaviour among groups identified using the Gaussian mixture model analysis

	Groups	Mean	Mean difference	<i>P</i> -value
<b>Number of complete round trips</b>	<b>1–2</b>	1.02	0.53	0.00
	<b>1–3</b>	0.49	1.07	0.00
	<b>2–3</b>	−0.04	0.54	0.00
<b>Total number of movements</b>	<b>1–2</b>	1.56	0.45	0.00
	<b>1–3</b>	1.11	0.82	0.00
	<b>2–3</b>	0.74	0.37	0.00
<b>Total distance</b>	<b>1–2</b>	3.36	0.44	0.00
	<b>1–3</b>	2.91	0.84	0.00
	<b>2–3</b>	2.51	0.40	0.00
Average speed	1–2	1.10	0.00	1.00
	1–3	1.09	0.01	1.00
	2–3	1.08	0.01	1.00
Maximal speed	1–2	1.28	0.19	1.00
	1–3	1.26	0.06	0.95
	2–3	1.22	0.04	1.00
Minimal speed	1–2	0.87	−0.36	1.00
	1–3	0.90	−0.01	1.00
	2–3	0.88	0.02	1.00
<b>Average time of a round trip</b>	<b>1–2</b>	1.91	0.15	0.29
	<b>1–3</b>	1.77	−0.47	0.01
	<b>2–3</b>	2.38	−0.62	0.01
<b>Maximal time of a round trip</b>	<b>1–2</b>	2.58	0.36	0.00
	<b>1–3</b>	2.22	−0.02	1.00
	<b>2–3</b>	2.60	−0.38	0.07
<b>Latency of the first movement</b>	<b>1–2</b>	2.20	−0.71	0.00
	<b>1–3</b>	2.91	−0.70	0.00
	<b>2–3</b>	2.90	0.01	1.00
Latency of the last movement	1–2	3.44	0.15	1.00
	1–3	3.42	0.10	0.30
	2–3	3.34	0.08	0.48
<b>Time of all movements</b>	<b>1–2</b>	3.41	0.20	0.00
	<b>1–3</b>	3.21	0.30	0.00
	<b>2–3</b>	3.10	0.10	0.74
<b>Time of all movements without pauses</b>	<b>1–2</b>	2.52	0.35	0.01
	<b>1–3</b>	2.17	1.10	0.00
	<b>2–3</b>	1.42	0.75	0.00
<b>Number of movements away from wall</b>	1–2	0.59	0.05	1.00
	<b>1–3</b>	0.54	0.35	0.03
	<b>2–3</b>	0.24	0.29	0.08
<b>Frequency</b>	<b>1–2</b>	−2.37	0.41	0.00
	<b>1–3</b>	−2.78	0.81	0.00
	<b>2–3</b>	−3.18	0.40	0.00

**Table 1** Results of analyses of variance testing for differences between the three behavioural groups

Behavioural variable	<i>P</i> -value	$F_{2,32}$
<b>Number of complete round trips</b>	0.00	43.90
<b>Total number of movements</b>	0.00	75.41
<b>Total distance moved</b>	0.00	66.89
Average speed	0.96	0.04
Maximal speed	0.58	0.56
Minimal speed	0.74	0.30
<b>Average duration of a round trip</b>	0.00	8.42
<b>Maximal duration of a round trip</b>	0.00	9.19
<b>Latency to the first movement</b>	0.00	25.84
Latency of the last movement	0.25	1.44
<b>Duration of all movements</b>	0.00	11.96
<b>Duration of all movements without pauses</b>	0.00	18.07
<b>Number of movements away from the wall</b>	0.03	3.79
<b>Frequency of movement</b>	0.00	59.18

Bold variables are those that are significantly different between groups.

Bold variables are those that are significantly different between groups.

**Table 3** Results of the MANOVAs and subsequent ANOVAs testing for differences in morphology and performance between groups

	MANOVA			ANOVA	
	Wilks' lambda	F	P-value	F	P-value
<b>Body size</b>	0.77	2.16	0.85		
	Snout-vent length			1.93	0.16
	Mass			0.12	0.89
<b>Head dimensions</b>	0.83	0.69	0.70		
	Head length			0.07	0.93
	Head width			0.18	0.84
	Head height			1.02	0.37
	Lower jaw length			0.62	0.55
<b>Forelimb segments</b>	0.67	1.26	0.28		
	Humerus			0.76	0.48
	Radius			0.01	0.99
	Hand			0.05	0.95
	Longest finger			2.15	0.13
	Forelimb length			0.90	0.42
<b>Pelvic girdle</b>	0.10	0.12	0.98		
	Ilium length			0.16	0.85
	Ilium width			0.10	0.90
<b>Hind limb segments</b>	0.74	0.91	0.53		
	Femur			1.92	0.16
	Tibia			0.40	0.68
	Foot			0.38	0.69
	Longest toe			1.39	0.26
	Hind limb length			1.23	0.31
<b>Performance</b>	0.802	0.652	0.763		
	Mean swimming velocity			0.62	0.55
	Maximal swimming velocity			0.37	0.70
	Maximal swimming acceleration			1.92	0.16
	Endurance (maximal time jumped)			0.20	0.82
	Endurance (maximal distance jumped)			0.04	0.96

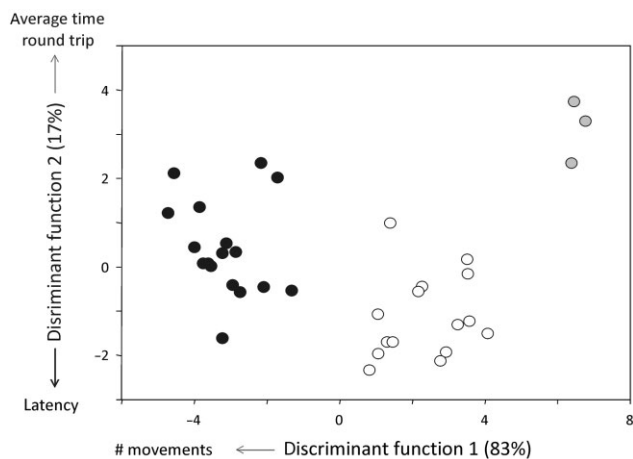
ANOVA, analysis variance; MANOVA, multivariate analyses of variance.

days and at different times of the day. The average behaviour thus represents a good proxy for an individual's behavioural strategy. Three significant behavioural clusters were identified in *X. tropicalis* male frogs freely exploring a novel environment. Animals in cluster one moved often and did so at high frequency. Moreover, animals in cluster one explored with limited pauses resulting in round trips of shorter duration. Animals grouped in behavioural cluster two displayed an average number of movements of relatively short duration, yet began to move later. The few individuals grouped together in the third cluster move very little, at low frequency and stay hidden more, resulting in round trips of longer duration.

The three behavioural clusters were identified using clustering methods that take into account all behavioural variables under the assumption that each behavioural strategy shows a normal distribution in a population. The retrieved clusters were then cross-validated using an independent clustering method, which showed that the grouping was robust (Fig. 2). Interestingly, the grouping of individuals in either two or three clusters was equally robust with the reassignment of individuals showing the same two individuals being misclassified. The three individuals comprising the third cluster, although being

classified as belonging to the second cluster if only two groups are predefined, show a distinct and unique behaviour compared with the entire sample characterized by extremely low levels of exploration. In summary, three groups differing in the amount of exploration and the time to the onset of exploration were detected.

van Oers *et al.* (2004) showed the importance of the latency of the first movement when investigating avian exploration syndromes as this reflects the willingness of individuals to take risks. Our analysis demonstrated differences among the three clusters in the latency to the first movement. Indeed, animals in cluster one start moving earlier than individuals in group two, with the maximal latency observed for individuals in cluster two. Generally, frogs moved close to the walls of the tank and did not explore the centre much. This behaviour involving exploration close to a physical structure such as a wall reduces visibility to predators and provides some shelter while exploring. Similar behaviours where animals disperse and explore using landscape elements have been demonstrated for other taxa (Baguette *et al.*, 2013). Among our behavioural clusters, animals in cluster three showed significantly less movement away from the walls of the cage compared with the animals in the other two clusters.



**Figure 2** Results of a discriminant function analysis performed on the behavioural data to illustrate the three clusters retrieved in the clustering analysis. In black are indicated animals that move a lot at relatively low latency. The white circles represent animals that move less and have longer latency. Finally, the cluster of grey circles represents animals that move very infrequently and take a very long time to complete a round trip.

### Behavioural syndromes

Behavioural syndromes (bold vs. shy) are typically recovered in studies analysing exploration behaviour (Dingemanse & de Goede, 2004; Wilson & Godin, 2009). Bold individuals are defined, in this context, as those that show curiosity and a willingness to explore; they move a lot at high frequency and take risks by moving away from walls or other structures that provide shelter. On the opposite end, shy individuals stay hidden long, explore little and use landscape elements during exploration to avoid open space. When analysing the behavioural clusters discovered in our data, it becomes evident that animals in cluster one can be characterized as bold, those in cluster three as shy and those in cluster two as intermediate. Indeed, our data show a large group of male *X. tropicalis* that show a distinct, yet intermediate behaviour (cluster 2), that cannot be characterized as neither shy nor bold: animals wait longer than the shy ones before starting to explore their environment, yet move frequently, similar to bold individuals. Animals in this group may decrease risk-taking by waiting longer before starting the exploration of the novel environment. This allows for an *a priori* analysis of the environment, and once deemed safe, exploration starts. The consequence of the longer wait before the onset of exploration may cause missed opportunities to encounter potential food resources or sexual partners compared with bold individuals. Thus, rather than characterizing exploration behaviour into two groups, we here suggest that three strategies may better describe the exploration behaviour in *X. tropicalis*. When defining only two groups, animals from clusters two and three group together resulting in one group of shy (clusters two and three) and one group of bold individuals (cluster one).

Male *X. tropicalis* from clusters one and three that conform with the classical descriptions of behavioural syndromes can be characterized as bold and shy, respectively. Bold individuals are mobile, allowing them to encounter food resources or reproductive partners more frequently, yet expose themselves to an increased risk of predation (Dingemanse & Réale, 2005). At the opposite end, shy individuals may come across less resources or reproductive partners, but are less exposed to predation, which may increase longevity. The overall fitness of these two behavioural syndromes should be equal over medium to long time spans as frequency-dependent selection likely operates on such a two-strategy system (Wolf & Weissin, 2012). However, bold animals may colonize new areas more rapidly, may recover faster from stress, show increased levels of inducible morphological defences and may learn more quickly (e.g. Bridle *et al.*, 2014; Hulthén *et al.*, 2014). Yet, our data show that other intermediate strategies may also exist.

Given a scenario of habitat fragmentation as in the case of *X. tropicalis*, however, bold individuals may be selected for, given that they are likely to explore their environment more, and thus may encounter new ponds and reproductive partners more readily. As such, they may ensure gene flow between fragmented populations. This does not mean, however, that shy animals are incapable of exploring novel environments (Wolf & Weissin, 2012), just that the time needed to do so is greater. However, in the case of continuous and extensive habitat fragmentation, shy individuals may not be able to keep up with the rate of fragmentation and ultimately may be selected against over the long term. Whereas gene flow is assured by mobile individuals, sedentary individuals run the risk of inbreeding, which may result in local extinction (Dixo *et al.*, 2009).

### Is behaviour driven by variation in morphology and performance?

*Xenopus tropicalis* is an aquatic pipid frog that spends most of its time in water. Yet, like most frogs, *X. tropicalis* can both swim and jump (Emerson & de Jongh, 1980; Nauwelaerts, Stamhuis & Aerts, 2005) and the species shows overland migrations during the rainy season (Rödel, 2000). Although behavioural syndromes are the result of a behavioural characterization of complex behaviours, they also underlie the evolution of mobility. However, mobility is also dependent on locomotor performance. Consequently, to understand mobility, both behaviour and performance need to be studied (Careau & Garland, 2012). Indeed, in some cases, correlated evolution of both behaviour and performance has been demonstrated in cases of strong selection on mobility. For example, cane toads on the invasion front in Australia are characterized by both a more directional exploration of their environment as well as morphological adaptations resulting in greater locomotor performance (Phillips *et al.*, 2010).

Our data suggest that exploration behaviour is decoupled from locomotor performance and morphology in *X. tropicalis*. This is important as it implies that these parameters can be under selection without affecting the other. Thus,

whereas selection on exploration behaviour may take place, this need not affect locomotor performance *per se* and may allow animals to maintain levels of performance adequate for both predator escape (aquatic burst performance) and the exploration of novel areas (i.e. endurance). This pattern may also suggest differences in the underlying genetic architecture and the absence of pleiotropy between these traits. Whereas the heritability of exploration behaviour has been demonstrated for birds (Pulido, Berthold & Van Noordwijk, 1996; Pulido *et al.*, 2001; Dingemanse *et al.*, 2002; Drent, van Oers & van Noordwijk, 2002; van Oers *et al.*, 2004), this remains to be demonstrated for frogs such as *X. tropicalis*. Understanding the genetic basis of variation in behaviour will greatly increase our understanding of how selection may act on animals in highly fragmented populations and is currently under investigation.

## Conclusion

We demonstrated stable, repeatable patterns of exploration behaviour in male *X. tropicalis*. Moreover, in addition to the two classic behaviours, 'shy' and 'bold', we identified an intermediate strategy. In this species, behavioural strategies are decoupled from morphology and locomotion performance. As habitat fragmentation imposes strong selection on mobility animals may respond by taking advantage of two complementary and independent strategies: behaviour and performance.

## Acknowledgements

We would like to thank L. N. Gonwouo and E. Fokam for their valuable help in the field. This research was supported by l'Agence Nationale de la Recherche MOBIGEN (ANR-09-PEXT-003 to A.H. and C.B.), a Muséum National d'Histoire Naturelle Action transversale du Muséum (MNHN ATM) grant of the programme 'Biodiversité actuelle et fossile' to A. H., and a Marie Curie Reintegration grant to C. B. (FP7-PEOPLE-IRG-2008 #239257).

## References

- Baguette, M., Blanchet, S., Legrand, D., Stevens, V. & Turlure, C. (2013). Individual dispersal, landscape connectivity and ecological networks. *Biol. Rev.* **88**, 310–326.
- Banfield, J.D. & Raftery, A.E. (1993). Model-based Gaussian and non-Gaussian clustering. *Biometrics* **49**, 803–821.
- Baylac, M., Villemant, C. & Symbolotti, G. (2003). Combining geometric morphometrics with pattern recognition for the investigation of species complexes. *Biol. J. Linn. Soc.* **80**, 89–98.
- Beaumont, L.J., Pitman, A., Perkins, S., Zimmermann, N.E., Yoccoz, N.G. & Thuiller, W. (2010). Impacts of climate change on the world's most exceptional ecoregions. *Proc. Natl. Acad. Sci. USA* **108**, 1306–2311.
- Bell, A.M., Hankison, S.J. & Laskowski, K.L. (2009). The repeatability of behaviour: a meta-analysis. *Anim. Behav.* **77**, 771–783.
- Bridle, J.R., Buckley, J., Bodsworth, E.J. & Thomas, C.D. (2014). Evolution on the move: specialization on widespread resources associated with rapid range expansion in response to climate change. *Proc. R. Soc. Lond. B.* **281**, 1776–20131800.
- Careau, V. & Garland, T. Jr. (2012). Performance, personality, and energetic: correlation, causation and mechanism. *Physiol. Biochem. Zool.* **85**, 543–571.
- Careau, V., Thomas, D., Humphries, M.M. & Reale, D. (2008). Energy metabolism and animal personality. *Oikos* **117**, 641–653.
- Careau, V., Biro, P., Bonneaud, C., Fokam, E. & Herrel, A. (2014). Individual variation in thermal performance curves: swimming burst speed and jumping endurance in wild-caught tropical clawed frogs. *Oecologia* **175**, 471–480.
- Carere, C., Drent, P.J., Privitera, L., Koolhass, J.M. & Groothuis, T.G.G. (2005). Personalities in great tits, *Parus major*: stability and consistency. *Anim. Behav.* **70**, 795–805.
- Cote, J., Clobert, J., Brodin, T., Fogarty, S. & Sih, A. (2010). Personality-dependent dispersal: characterization, ontogeny and consequences for spatially structured populations. *Phil. Trans. R. Soc. B.* **365**, 4065–4076.
- Deutsch, C.A., Tewksbury, J.J., Huey, R.B., Sheldon, K.S., Ghalambor, C.K., Haak, D.C. & Martin, P.R. (2007). Impacts of climate warming on terrestrial ectotherms across latitude. *Proc. Natl. Acad. Sci. USA* **18**, 6668–6672.
- Dillon, M.E., Wang, G. & Huey, B. (2010). Global metabolic impacts of recent climate warming. *Nature* **46**, 704–706.
- Dingemanse, N.J. & de Goede, P. (2004). The relation between dominance and exploratory behavior is context-dependent in wild great tits. *Behav. Ecol.* **6**, 1023–1030.
- Dingemanse, N.J. & Réale, D. (2005). Natural selection and animal personality. *Behaviour* **142**, 1165–1190.
- Dingemanse, N.J., Both, C., Drent, P.J., Van Oers, K. & van Noordwijk, A.J. (2002). Repeatability and heritability of exploratory behaviour in great tits from the wild. *Anim. Behav.* **64**, 929–938.
- Dingemanse, N.J., Wright, J., Kazem, A.J.N., Thomas, D.K., Hickling, R. & Dawna, N. (2007). Behavioural syndromes differ predictably between 12 populations of three-spined stickleback. *J. Anim. Ecol.* **76**, 1128–1138.
- Dixo, M., Metzger, J.P., Morgante, J.S. & Zamudio, K.R. (2009). Habitat fragmentation reduces genetic diversity and connectivity among toad populations in the Brazilian Atlantic Coastal Forest. *Biol. Conserv.* **142**, 1560–1569.
- Drent, P.J., van Oers, K. & van Noordwijk, A.J. (2002). Realized heritability of personalities in the great tit (*Parus major*). *Proc. R. Soc. Lond. B.* **270**, 45–51.
- Emerson, S.B. & de Jongh, H.J. (1980). Muscle activity at the ilio-sacral articulation of frogs. *J. Morphol.* **166**, 129–144.
- Ernst, R., Linsenmair, K.E. & Rodel, M.O. (2006). Diversity erosion beyond the species level: dramatic loss of functional diversity after selective logging in two tropical amphibian communities. *Biol. Conserv.* **133**, 143–155.

- Ferraz, G., Russell, G.J., Stouffer, P.C.R., Bierregaard, O., Pimm, S.L. & Lovejoy, T.E. (2003). Rates of species loss from Amazonian forest fragments. *Proc. Natl. Acad. Sci. USA* **24**, 14069–14073.
- Gosling, S.D. (2001). From mice to men: what can we learn about personality from animal research? *Psychol. Bull.* **127**, 45–86.
- Herrel, A. & Bonneaud, C. (2012a). Trade-offs between burst performance and maximal exertion capacity in a wild amphibian, *Xenopus tropicalis*. *J. Exp. Biol.* **215**, 3106–3111.
- Herrel, A. & Bonneaud, C. (2012b). Temperature dependence of locomotor performance in the tropical clawed frog, *Xenopus tropicalis*. *J. Exp. Biol.* **215**, 2465–2470.
- Herrel, A., Gonwouo, L.N., Fokam, E.B., Ngundu, W.I. & Bonneaud, C. (2012). Inter-sexual differences in body shape and locomotor performance in the aquatic frog, *Xenopus tropicalis*. *J. Zool. (Lond.)* **287**, 311–316.
- Hillers, A., Veith, M. & Rödel, M.O. (2008). Effects of forest fragmentation and habitat degradation on West African leaf-litter frogs. *Biol. Conserv.* **22**, 762–772.
- Hochberg, Y. (1988). A sharper Bonferroni procedure for multiple tests of significance. *Biometrika* **75**, 800–802.
- Huey, R.B., Deutsch, C.A., Tewksbury, J.J., Vitt, L.J., Hertz, P.E., Alvarez Perez, H.J. & Garland, T. Jr. (2008). Why tropical forest lizards are vulnerable to climate warming. *Proc. R. Soc. Lond. B.* **276**, 1939–1948.
- Hulthén, K., Chapman, B.B., Nilsson, P.A., Hollander, J. & Brönmark, C. (2014). Express yourself: bold individuals induce enhanced morphological defences. *Proc. R. Soc. Lond. B.* **281**, 1776–20132703.
- von Merten, S. & Siemers, B.M. (2012). Exploratory behaviour in shrews: fast-lived *Sorex* versus slow-lived *Crocidura*. *Anim. Behav.* **84**, 29–38.
- Nauwelaerts, S., Stamhuis, E. & Aerts, P. (2005). Swimming and jumping in a semi-aquatic frog. *Anim. Biol.* **55**, 3–15.
- van Oers, K., Drent, P.J., de Goede, P. & van Noordwijk, A.J. (2004). Realized heritability and repeatability of risk-taking behaviour in relation to avian personalities. *Proc. R. Soc. Lond. B.* **271**, 65–73.
- Phillips, B.L., Brown, G.P. & Shine, R. (2010). Evolutionarily accelerated invasions: the rate of dispersal evolves upwards during the range advance of cane toads. *J. Evol. Biol.* **23**, 2595–2601.
- Pulido, F., Berthold, P. & Van Noordwijk, A.J. (1996). Frequency of migrants and migratory activity are genetically correlated in a bird population: evolutionary implications. *Proc. R. Soc. Lond. B.* **93**, 14642–14647.
- Pulido, F., Berthold, P., Mohr, G. & Querner, U. (2001). Heritability of the timing of autumn migration in a natural bird population. *Proc. Biol. Sci.* **268**, 953–959.
- R Development Core Team. (2013). *R: a language and environment for statistical computing*. Vienna: R Foundation for Statistical Computing.
- Ripley, B.D. (1996). *Pattern recognition and neural networks*. Cambridge: Cambridge University Press.
- Rödel, M.O. (2000). *Herpetofauna of West Africa. Amphibians of the Western Savannah*. Vol. 1. Frankfurt am Main, Germany: Chimaira Buchhandelsgesellschaft mbH.
- Scott, J.P. (1956). The analysis of social organization in animals. *Ecology* **37**, 213–221.
- Shillito, E.E. (1963). Exploratory behaviour in the short-tailed vole *Microtus agrestis*. *Behaviour* **21**, 145–154.
- Smith, B.R. & Blumstein, D.T. (2008). Fitness consequences of personality: a meta-analysis. *Behav. Ecol.* **19**, 448–455.
- Tracy, C.R., Christian, K.A., Baldwin, J. & Phillips, B.L. (2012). Cane toads lack physiological enhancements for dispersal at the invasive front in Northern Australia. *Biol. Open* **1**, 37–42.
- Uher, J., Asendorpf, J.B. & Call, J. (2008). Personality in the behaviour of great apes: temporal stability, cross-situational consistency and coherence in response. *Anim. Behav.* **75**, 99–112.
- Watanabe, N.M., Stahlman, W.D., Blaisdell, A.P., Garlick, D., Fast, C.D. & Blumstein, D.T. (2012). Quantifying personality in the terrestrial hermit crab: different measures, different inferences. *Behav. Processes* **91**, 133–140.
- Wells, K.D. (1977). The social behaviour of anuran amphibians. *Anim. Behav.* **25**, 666–693.
- Wilson, A.D.M. & Godin, J.G.J. (2009). Boldness and behavioral syndromes in the bluegill sunfish, *Lepomis macrochirus*. *Behav. Ecol.* **20**, 231–237.
- Wilson, R.S., James, R.S. & Van Damme, R. (2002). Trade-offs between speed and endurance in the frog *Xenopus laevis*: a multi-level approach. *J. Exp. Biol.* **205**, 1145–1152.
- Wolf, M. & Weissin, F.J. (2012). Animal personalities: consequences for ecology and evolution. *Ecol. Evol.* **8**, 452–461.
- Zelazowski, P., Malhi, Y., Huntingford, C., Sitch, S. & Fisher, J.B. (2011). Changes in the potential distribution of humid tropical forests on a warmer planet. *Phil. Trans. R. Soc. B. A.* **369**, 137–160.