The effects of phototaxis and thigmotaxis on microhabitat selection by a caecilian amphibian (genus *Ichthyophis*)

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In the field, ichthyophiid caecilians are found in soil and epigeic microhabitats – conditions that provide both the phototactic cue of darkness and the thigmotactic cue of tactile contact. In laboratory experiments, we investigated the use of phototaxis and thigmotaxis on refuge selection in a caecilian of the genus *Ichthyophis*. Refuges that provided light with tactile cues, darkness with tactile cues, or darkness without tactile cues were sufficient in satisfying refuge-seeking behaviour in this species. Tactile stimuli, however, proved to be more preferred than darkness in refuge-seeking behaviour. Our results are consistent with field observations of *Ichthyophis* species. Thigmotactic cues may be preferred because they are more biologically significant for a burrowing vertebrate that has reduced vision.

Key words: Amphibia, Gymnophiona, Ichthyophiidae, darkness cues, refuge selection, tactile cues

INTRODUCTION

Habitat selection is central to understanding the ecology of organisms (Huey, 1991). The choices associated with obtaining shelter, avoiding predators and competitors, locating mates and foraging directly affect performance and ultimately survival. Evaluation of the thermal nature of habitat selection by organisms – especially by ectotherms – has generally received the most attention by biologists (Cowles & Bogert, 1944; Porter et al., 1973; Christian et al., 1983; Riechert, 1985; Huey et al., 1989; Diaz, 1997). In contrast, the roles of phototaxis and thigmotaxis in habitat selection have not been extensively investigated, even though these sensory cues play an important function in some species (Test, 1946; Chiszar et al., 1987; Alberstadt et al., 1995; McGaw, 2001). Clearly, the ability to evaluate multiple variables within the environment is a complex process with profound ecological implications for an animal.

In comparison to other amphibians, little is known of the perceptual mechanisms of caecilians (Gymnophiona). Often considered blind, many caecilians have functioning photoreceptor organs (Wake, 1985). Caecilians of the genus *Ichthyophis* display negative phototaxis (Ross, 1959; Himstedt, 1995), a behaviour that occurs in other amphibians as well (Test, 1946; Ray, 1970; Jaeger & Hailman, 1973). The reduction of visual capabilities implies a greater emphasis on other sense organs in caecilians. Like all amphibians, tactile stimulation is received by receptors in the epidermal layer of the skin (Duellman & Trueb, 1994). Unique among amphibians, however, are paired retractile tentacles situated behind each nostril that are believed to function both in a tactile and chemosensory capacity (Billo & Wake, 1987; Himstedt & Simon, 1995).

We present a preliminary investigation of microhabitat selection in a small burrowing caecilian of the family Ichthyophiidae. Ichthyophiid caecilians have been reported from a variety of habitat associations including forests, open bush, swamps and agricultural areas (Vyas, 2004; Kupfer et al., 2005). Within these habitat types, animals have been found to seasonally inhabit relatively loose, shallow soil and epigeic microhabitats such as under leaf litter, rotting wood and rotting vegetation (Inger, 1966; Nussbaum & Gans, 1980; Gundappa et al., 1981; Vyas, 2004; Kupfer et al., 2005). These objects produce refuges that provide both the phototactic cue of darkness and the thigmotactic cue of tactile contact. When seeking refuge, we assumed that caecilians would prefer refuges that provide these cues over those providing the opposite conditions of no darkness (light) and no tactile cues. We evaluated refuge-seeking behaviour in a caecilian of the genus *Ichthyophis* using six different laboratory choice conditions.

The first question we addressed was whether this species always prefers refuges, even those lacking one cue, over an open area. The three trials to address this question consisted of a refuge versus an open area. The three refuges were: a) both tactile and darkness cues present (DT); b) only tactile cue present (T); and c) only darkness cue present (D). The second question we addressed was the relative preference of phototactic versus thigmotactic cues. We used a series of three choice conditions between two refuges that offered varying combinations of phototactic and thigmotactic cues as follows: a) both cues present versus only tactile cue present (DT vs T); b) only darkness cue present versus only tactile cue present (D vs T); and c) both cues present versus only darkness cue present (DT vs D).

Depending upon which cue is preferred, the following predictions can be made. First, if phototactic cues are preferred over thigmotactic cues, we would expect that the tactile cues would be generally ignored if the darkness cue (a preferential cue) was present. Second, if thigmotactic cues are preferred over phototactic cues, we

Table 1. Predictions of refuge choice dependent upon preference of cues.

Hypothesis of cue preference	Choice a DT vs T	Choice b D vs T	Choice c DT vs D
Phototactic cue preferred	DT>T	D>T	DT=D
Thigmotactic cue preferred	DT=T	D<T	DT>D
Neither cue preferred	DT>T	D=T	DT>D

would expect the darkness cue to be generally ignored if the tactile cue (a preferential cue) was present. Third, if neither cue is preferred, then we would expect the caecilians to always choose a refuge with two cues (darkness and tactile) over a refuge that has only one cue (darkness or tactile). Outcomes for each choice are given in Table 1. Our results should provide a better understanding of the role of phototaxis and thigmotaxis in refuge seeking by ichthyophiid caecilians.

MATERIALS AND METHODS

Nine field-collected unsexed adult caecilians of the genus *Ichthyophis* (36.5–59.1 g) were obtained from Thailand through a dealer in December 2000 and March 2001. Because of the high diversification and taxonomic uncertainty of many striped ichthyophiid caecilians from Thailand (Gower et al., 2002), we refer to our specimens simply as a striped *Ichthyophis*.

Specimens were maintained at two facilities (hereafter referred to as "facility 1" and "facility 2") in groups of 2-3 in glass enclosures measuring 61 x 32 x 43 cm. Half of the substrate consisted of a mixture of potting soil and peat moss and the other half consisted of pea gravel. Specimens could be individually identified by variations in the lateral yellow stripe characteristic of many ichthyophiid caecilians. All caecilians were maintained on a photoperiod of 12L:12D at 27°C and were fed earthworms (*Lumbricus* spp.) on a weekly basis.

Specimens were individually tested with six conditions between July and December 2002. The testing enclosures consisted of five 86 x 40 x 31 cm clear plastic storage boxes. The enclosures were partially covered with clear acrylic panels that allowed air exchange at both ends but maintained humidity (>95%). The bottom of each enclosure was covered by a 1–1.5 cm layer of washed pea gravel and 1 cm of dechlorinated water. Two 40-watt fluorescent tubes positioned 20 cm above each enclosure provided light. Illuminance was measured using a General Electric Type 214 exposure meter at the substrate surface level of the enclosures (1398 lx). All enclosures, substrates, and testing apparatus were vigorously cleaned prior to and between testing with a commercial detergent and thoroughly rinsed with dechlorinated water. Specimens were not fed during testing.

We introduced specimens into the centre of each testing enclosure at 1400 the day prior to testing. Over the next five days, we made observations at 0800, 1100, 1400 and 1600 at facility 1. At facility 2, a remote video camera (Burle model TC652EA) was mounted directly above three testing enclosures. All tests were recorded on videotape and reviewed later. We recorded a specimen as being in a

hide box if the head of the specimen was underneath a hide box. We returned all specimens to their home enclosures for 8–12 days between testing. Order of testing conditions and of refuge location (left or right side of enclosure) was determined randomly in advance.

We used three types of refuges that provided different combinations of cues. *Ichthyophis* spp. are found under objects that provide the condition of both darkness and tactile cues. The DT refuge was a black opaque acrylic refuge (29 x 23 x 1.5 cm) that offered both conditions. Its construction facilitated entrance and exit by having two open ends. The height of the refuge (1.5 cm) created a crevice-like condition so that a specimen would receive tactile cues from both the substrate and the ceiling of the refuge as well as the two closed ends. The T refuge provided tactile cues but not the darkness cue. It was identical to the first refuge except that it was constructed of clear rather than opaque acrylic. The D refuge provided the darkness cue but no tactile cue. To achieve this, we suspended a flat, black opaque acrylic refuge (29 x 23 cm) 3 cm above the floor of the testing arena. Thus, the refuge provided the condition of darkness, yet did not come in direct contact with the specimens, and therefore provided little if any tactile stimulation. A fourth condition that was not technically a refuge consisted of the open area of the testing enclosure. This area provided the conditions of light and no tactile cues.

Question 1: Refuge use
The objective of these tests was to determine whether specimens preferred refuges over an open area. For each condition we placed one of the refuges at one end of the testing enclosure. The conditions were: 1a) DT refuge with both cues; 1b) T refuge; 1c) D refuge.

Question 2: Preference for phototactic versus thigmotactic cues
The objective of these tests was to determine whether one cue was preferred during refuge seeking behaviour in simultaneous choice conditions. The three choice conditions were: 2a) DT refuge versus T refuge; 2b) D refuge versus T refuge; 2c) DT refuge versus D refuge.

Statistical analysis
Because the animals usually stayed in a chosen refuge all day, the scans within a day were not independent samples of refuge choice. To accommodate this and to avoid pseudoreplication, we used each day as an independent trial. If an animal changed locations, we used the location where it was found most frequently as its choice. We analysed refuge use in conditions 1a, 1b and 1c using G-tests (Sokal & Rohlf, 1981). Wilcoxon signed-rank tests were used to evaluate the difference between refuge preference during conditions 2a, 2b and 2c using SYSTAT (version 8.0) software. All tests were two-tailed and differences were considered significant when $P<0.05$.

RESULTS

Question 1: Refuge use
The caecilians were almost always seen under a refuge during the day. The one individual that was seen in the open died soon afterwards. Rather than perform indi-
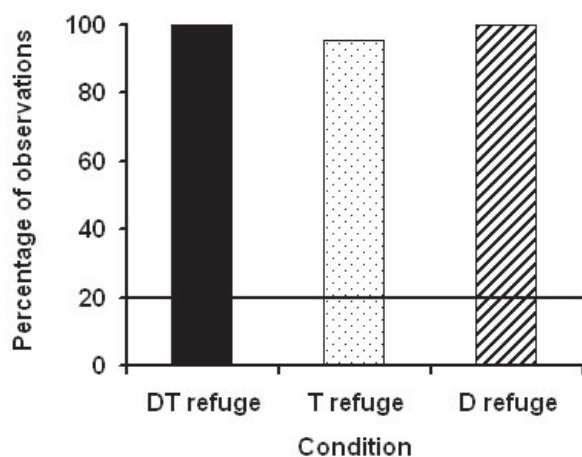
Fig. 1. Percentage of trials that *Ichthyophis* individuals were observed in refuges during conditions 1a (open vs DT refuge), 1b (open vs T refuge) and 1c (open vs D refuge). The horizontal line at 20% indicates the expected value if the choice was random.

Fig. 2. Percentage of trials that *Ichthyophis* individuals were observed in refuges during conditions 2a (DT vs T refuge), 2b (T vs D refuge) and 2c (DT D refuge).
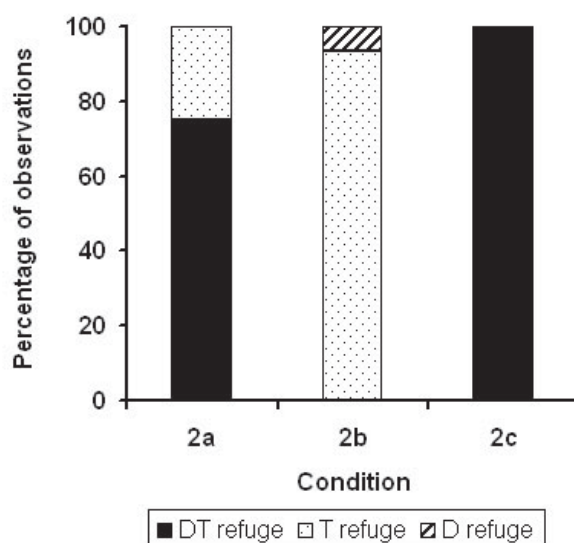
vidual chi-square goodness-of-fit tests on each animal, we used the more powerful heterogeneity G-test (Sokal & Rohlf, 1981) to determine whether the caecilians were seen significantly more times under the refuge than would occur by random chance. The refuge covered 20% of the floor area of the testing chamber. Therefore, if the caecilians occupied random areas in the chamber, they should be under the refuge during 20% of the trials. For all three conditions, the caecilians strongly preferred to be under the refuge versus in the open area (1a: GT=128.7, $P<0.001$, $n=8$; 1b: GT=132.6, $P<0.001$, $n=9$; 1c: GT=128.7, $P<0.001$, $n=8$, Fig. 1).

Question 2: Preference for phototactic versus thigmotactic cues

Because this question focuses on the choice between refuges, we only used observations in which an animal was using a refuge. During only one trial did an animal switch refuges. For this animal, we used the refuge it was seen in most as the refuge of choice for that trial. For each animal, the number of trials during which an animal was seen in a refuge was used as the measure of preference. In condition 2a, the caecilians were under the DT refuge more often than under the T refuge, but not significantly ($Z=-1.869$, $P>0.05$, $n=8$; Fig. 2). In condition 2b, the caecilians were under the T refuge significantly more than the D refuge ($Z=2.751$, $P=0.006$, $n=9$; Fig. 2). In condition 2c, the caecilians were under the DT refuge significantly more than the D refuge ($Z=2.828$, $P=0.005$, $n=8$; Fig. 2).

DISCUSSION

The results of conditions 1a–c indicated that refuges that provided at least one cue (darkness or tactile) were sufficient to satisfy refuge-seeking behaviour in these striped *Ichthyophis*. These results were consistent with field observations that have documented *Ichthyophis* species as being found under surface objects or in loose, shallow soil (Inger, 1966; Nussbaum & Gans, 1980; Gundappa et al., 1981; Vyas, 2004; Kupfer et al., 2005).

Having provided evidence that the caecilians preferred refuges over open areas during daylight hours, we tested the preference between phototactic and thigmotactic cues for refuge choice. When both refuges provided tactile cues, the caecilians did not show a preference between the refuge with and the one without the darkness cue. When both refuges provided the darkness cue, the caecilians strongly preferred the refuge with the tactile cue versus the one without this cue. When the caecilians had to choose between refuges that only offered one preferential cue, they strongly preferred the refuge with the tactile cues. These results fit our predictions of thigmotactic cues being more important than phototactic cues.

The preference of these caecilians for thigmotactic cues over phototactic cues is not a common result in other studies investigating refuge-seeking behaviour in ectotherms. In amphibians, thigmotaxis has not been found to play an important role in refuge-seeking behaviour. *Heleioporus eyrie*, for instance, displays a positive tactile response only under conditions of severe dehydration (Packer, 1963). Although *Plethodon cinereus* is usually found under cover objects, laboratory experiments find that dorsal contact is coincidental rather than preferential (Test, 1946). In reptiles, however, tactile stimuli influence snapping turtle (*Chelydra serpentina*) distribution (Sexton, 1958). Conversely, red spitting cobras (*Naja mossambica pallida*) prefer darkness over tactile cues in cover-seeking experiments (Chiszar et al., 1987). When given a choice, though, cobras prefer the condition of tactile stimuli and darkness over either stimulus alone.

From an evolutionary perspective, the importance of sensory mechanisms in gymnophiones may have an adaptive significance. Reductions in the visual, auditory

and electroreceptive systems generally correspond with a fossorial lifestyle (Wake, 1992). Accordingly, the emphasis on tactile and chemical receptors may conceivably have increased in importance as other sensory systems have regressed. For example, *I. kohtaoensis* can effectively localize prey by chemical orientation alone (Himstedt & Simon, 1995). We suggest the importance of thigmotaxis in refuge selection is strongly correlated with the burrowing and cover-seeking lifestyle of ichthyophiid caecilians.

The refuge-seeking behaviour of caecilians does not differ markedly from other organisms in purpose. The amphibian integument is a highly permeable membrane susceptible to water loss by evaporation. As such, refuge-seeking behaviour in a terrestrial environment is of adaptive value in reducing the danger of desiccation. While other amphibians behave similarly to reduce evaporative loss (Heatwole, 1960, 1962; Packer, 1963), the physiological benefits gained by seeking shelter for moisture are probably widespread (McGaw, 2001). The integumentary secretion of caecilians may serve as a defence against predation (Moodie, 1978; Jared et al., 1999), but the avoidance of predators altogether would have a greater selective advantage to caecilians. Thus, the ability to seek and find suitable refuges would also benefit caecilians by reducing the risk of predation.

It is unlikely that caecilians rely exclusively on phototactic and thigmotactic stimuli when assessing a potential refuge site. Nonetheless, our results indicate that these sensory cues play an important but variable role in refuge selection by ichthyophiid caecilians. While this study examined the responses of specific sensory systems on short-term refuge use, it did not examine the role of temporal features on refuge selection. Clearly, further studies are needed with other caecilian species in order to provide a better understanding of the sensory systems and their influence in the ecology of gymnophiones.

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REFERENCES

Alberstadt, P.J., Steele, C.W. & Skinner, C. (1995). Cover-seeking behavior in juvenile and adult crayfish, *Orconectes rusticus*: effects of darkness and thigmotactic cue. *Journal of Crustacean Biology* 15, 536–541.

Billo, R. & Wake, M.H. (1987). Tentacle development in *Dermophis mexicanus* (Amphibia, Gymnophiona) with an hypothesis of tentacle origin. *Journal of Morphology* 192, 101–111.

Chiszar, D., Radcliffe, C.W., Boyer, T. & Behler, J.L. (1987). Cover-seeking behavior in red spitting cobras (*Naja mossambica pallida*): effects of tactile cues and darkness. *Zoo Biology* 6, 161–167.

Christian, K.A., Tracy, C.R. & Porter, W.P. (1983).

Seasonal shifts in body temperature and microhabitats by Galapagos land iguana (*Conolophus pallidus*). *Ecology* 64, 463–468.

Cowles, R.B. & Bogert, C.M. (1944). A preliminary study of the thermal requirements of desert reptiles. *Bulletin of the American Museum of Natural History* 82, 265–296.

Diaz, J.A. (1997). Ecological correlates of the thermal quality of an ectotherm's habitat: a comparison between two temperate lizard populations. *Functional Ecology* 11, 79–89.

Duellman, W.E. & Trueb, L. (1994). *Biology of Amphibians*. Baltimore: John Hopkins University Press.

Gower, D.J., Kupfer, A., Oommen, O.V., Himstedt, W., Nussbaum, R.A., Loader, S.P., Presswell, B., Krishna, S.B., Müller, H., Boistel, R. & Wilkinson, M. (2002). A molecular phylogeny of Ichthyophiidae (Amphibia: Gymnophiona): out of India or out of Eurasia? *Proceedings of the Royal Society of London Series B* 269, 1563–1569.

Gundappa, K.R., Balakrishna, T.A. & Shakuntala, K. (1981). Ecology of *Ichthyophis glutinosus* (Linn.) (Apoda, Amphibia). *Current Science* 50, 480–483.

Heatwole, H. (1960). Burrowing ability and behavioral responses to desiccation of the salamander, *Plethodon cinereus*. *Ecology* 41, 661–668.

Heatwole, H. (1962). Environmental factors influencing local distribution and activity of the salamander, *Plethodon cinereus*. *Ecology* 43, 460–472.

Himstedt, W. (1995). Structure and function of the eyes in the caecilian *Ichthyophis kohtaoensis* (Amphibia, Gymnophiona). *Zoology (Jena)* 99, 81–94.

Himstedt, W. & Simon, D. (1995). Sensory basis of foraging behaviour in caecilians (Amphibia, Gymnophiona). *Herpetological Journal* 5, 266–270.

Huey, R.B. (1991). Physiological consequences of habitat selection. *American Naturalist* 137, S91–S118.

Huey, R.B., Peterson, C.R., Arnold, S.J. & Porter, W.P. (1989). Hot rocks and not-so-hot rocks: retreat-site selection by garter snakes and its thermal consequences. *Ecology* 70, 931–944.

Inger, R.F. (1966). The Amphibia of Borneo. *Fieldiana: Zoology*, 52, 1–402.

Jaeger, R.G. & Hailman, J.P. (1973). Effects of intensity on the phototactic responses of adult anuran amphibians: a comparative survey. *Zeitschrift fur Tierpsychologie* 33, 352–407.

Jared, C., Navas, C.A. & Toledo, R.C. (1999). An appreciation of the physiology and morphology of the caecilians (Amphibia: Gymnophiona). *Comparative Biochemistry and Physiology A: Comparative Physiology* 123, 313–328.

Kupfer, A., Nabhitabhata, J. & Himstedt, W. (2005). Life history of amphibians in the seasonal tropics: habitat, community and population ecology of a caecilian (genus *Ichthyophis*). *Journal of Zoology* 266, 237–247.

McGaw, I.J. (2001). Impacts of habitat complexity on physiology: purple shore crabs tolerate osmotic stress for shelter. *Estuarine, Coastal and Shelf Science* 53, 865–876.

Moodie, G.E.E. (1978). Observations on the life history of

the caecilian *Typhlonectes compressicaudus* (Dumeril and Bibron) in the Amazon basin. *Canadian Journal of Zoology* 56, 1005–1008.

Nussbaum, R.A. & Gans, C. (1980). On the *Ichthyophis* (Amphibia: Gymnophiona) of Sri Lanka. *Spolia Zeylanica* 35, 137–154.

Packer, W.C. (1963). Dehydration, hydration, and burrowing behavior in *Heleioporus eyrei* (Gray) (Leptodactylidae). *Ecology* 44, 643–651.

Porter, W.P., Mitchell, J.W., Beckman, W.A. & Dewitt, C. B. (1973). Behavioral implications of mechanistic ecology: thermal and behavioral modeling of desert ecototherms and their microenvironment. *Oecologia* 13, 1–54.

Ray, A.J., Jr. (1970). Instrumental avoidance learning by the tiger salamander *Ambystoma tigrinum*. *Animal Behaviour* 18, 73–77.

Riechert, S.E. (1985). Decisions in multiple goal contexts: habitat selection of the spider, *Agelenopsis aperta* (Gertsch). *Zeitschrift für Tierpsychologie* 70, 53–69.

Ross, D.M. (1959). The response to light in *Ichthyophis* (Amphibia – Apoda) from Ceylon. *Proceedings of the Zoological Society of London* 132, 83–98.

Sexton, O.J. (1958). The relationship between the habitat preferences of hatchling *Chelydra serpentina* and the physical structure of the vegetation. *Ecology* 39, 751–754.

Sokal, R.R. & Rohlf, F.J. (1981). *Biometry*. New York: W.H. Freeman.

Test, F.H. (1946). Relations of the red-backed salamander (*Plethodon cinereus*) to light and contact. *Ecology* 27, 246–254.

Vyas, R. (2004). Notes on the distribution and natural history of *Ichthyophis bombayensis* (Gymnophiona: Ichthyophiidae). *Hamadryad* 28, 130–136.

Wake, M.H. (1985). The comparative morphology and evolution of the eyes of caecilians (Amphibia, Gymnophiona). *Zoomorphology* 105, 277–295.

Wake, M.H. (1992). "Regressive" evolution of special sensory organs in caecilians (Amphibia: Gymnophiona): Opportunity for morphological innovation. *Zoologische Jahrbücher für Anatomie und Ontogenie der Tiere* 122, 325–329.