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Preference of Goldfish *(Carassius auratus)* for Dark Places

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A preferência do peixe dourado (*Carassius auratus*) por lugares escuros. Visando determinar a preferência por ambientes claros ou escuros, peixes dourados *(Carassius auratus)* foram submetidos a um teste num aquário em que podiam sair de um compartimento central indo seja para um compartimento preto, seja para um compartimento branco. Registrou-se (1) o primeiro compartimento escolhido, (2) a freqüência de entradas em cada compartimento, e (3) o tempo gasto neles. Os resultados mostram que, nas condições experimentais, *C. auratus* tem preferência significativa pelo escuro, uma característica relevante para o desenvolvimento de um modelo experimental de ansiedade com esta espécie.

Descritores**:** Preferência pelo escuro. Peixe dourado. *Carassius auratus*.

In order to determine their preference for dark or bright environments, goldfishes (*Carassius auratus)* were tested in an aquarium, in which they could swim out of a start compartment either into a black or into a white compartment. The following records were obtained: (1) first compartment chosen, (2) frequency of entries into each compartment and (3) time spent in each compartment. Results indicate that, in such experimental conditions, C. auratus has a significant preference for a dark environment, a feature that might be relevant for the development of an experimental model of anxiety for the study of this species.

Key Words: Dark preference. Goldfish. *Carassius auratus*.

In rat research, dark preference is a feature used to study anxiety-related behaviors and anti-anxiety agents as well. Dark preference constitutes a behavioral model based on conflict and is part of a broader set of models characterized by the use of two opposed-valence stimuli, an appetitive (naturally preferred) and an aversive one (naturally avoided). In a place preference test with rats, there are two contrasting environments, a dark (appetitive) compartment, and a light (aversive) compartment. A preference for dark environments is generally found in rodents, in the open-field test (Aulich, 1976). This preference for dark environments is decreased by repeated exposure of rats to the test situation (Aulich and Spilhofen, 1977).

Fishes have long been used in biological research because they represent the oldest and most diverse classes of vertebrates and live in many different habitats (Bolis et al., 2001), and also because there constitute good models for the study behavior in a laboratory setup. On one hand, fishes do not require large laboratories and sophisticated or expensive apparatuses; on the other hand, they are generally easy to breed and maintain and have a shor life span. As to anxiety, fishes are relevant as experimental subjects: they have an anatomical and functional monoaminergic distribution similar to that of mammalians, enabling comparative studies of the principal drugs used in anxiety therapy (Kah and Chambolle, 1983; Yoshida et al., 1983; Bonn, 1987).

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Goldfish (*Carassius auratus)* are native of Asia and are the most used fish in behavioral research. Studies about their behavior include maze studies, exploratory behavior, stimulus matching and other behavioral paradigms, including or not physiological and pharmacological variables. (Matis et al., 1974; Manteifel and Carelina, 1996; Salas et al., 1996; Cabanac and Lamberge, 1998; Mizukami et al., 1999; Spieler et al., 1999; Talton et al., 1999).

Negative phototaxis, i.e., the preference for dark environments, is found in some fish species. Recently, a preference for dark was demonstrated in zebrafish (*Danio rerio)*, another common laboratory fish (Serra et al., 1999). It is possible that escape from bright environments and permancence in dark ones have a predator-avoidance function. The aim of the present work was to investigate a possible preference for dark in goldfish, a topic about which there is little information available.

Seventeen mature experimentally naïve fish, approximately 4 months old, undetermined sex, weighing between 20 and 40 g were used. The animals were bought in a local pet shop and kept under controlled conditions (temperature 22 ± 2º C and continuous external filtering) and natural dark light cycle, in a 45 x 50 x 25 cm aquarium,in groups of 20 animals. The fish were fed with commercial fish flakes and there was a 2-week acclimatization interval between their acquisition and the start of the experiment.

The glass aquarium used (15 x 10 x 45 cm) was divided into two chambers (22,5 cm long), a white one and a black one. The white and the black compartments were lined with non shining (white and black) adhesive plastic. Two sliding doors delimited a 5-cm area located at the center of the apparatus, half white and half black. This compartment was used as a start compartment and, when the sliding doors were removed, the fish had access to both compartments.

The animals were exposed to only one session in the apparatus. The fish were placed individually in the start compartment and, after a 5 min delay, the two sliding doors were pulled and each fish was observed until it spent 5 min in one particular chamber. The apparatus was rotated from test one to another.The behavioral measures recorded were (1) the first compartment chosen after leaving the start compartment, (2) the frequency of entries into each compartment and (3) the time spent in each one of them. Mean entry time (time spent in each compartment divided by number of entries into that compartment), percentage of animals spending more than 50% of their time in one given compartment and percentage of first choices were calculated and differences evaluated with Student *t*-test for paired samples.

Most fish entered the black compartment first (12 out of 17 cases), and most remained in it more than 50% of the session (14 out of 17 cases). Total time spent in the black compartment (mean 411.8 sec) was higher than time spent in the white one (mean 116.1 sec; $t16 = -3.031$, $p < 0.01$). Mean entry time into the black compartment (mean 183.1 sec) tended to be higher than mean entry time into the white one (mean 65.2 sec; $t16 = -2.266$, $p = 0.038$).

These data indicate a preference for dark environments by *Carassius auratus*. This behavior may express a defense against predation. As the species is originally from shallow waters and suffers predation from birds, dark sites may represent protected environments. The behavior pattern exhibited by the fish may indicate a conflict between a tendency to explore a new environment, where food and sexual partners might be found, and a tendency to seek protection. A similar behavioral conflict has been studied in rats and mice. When observed in the elevated plus-maze, a well validated test of animal anxiety (Pellow et al., 1985; Lister, 1987), rats and mice show a strong preference for the protected parts of the apparatus but also explore the open, more aversive parts of it. There is evidence showing that the behavior of these animals is characterized by a conflict between remaining in the closed protected arms and a exploring the open unprotected arms (Montgomery, 1955; Pellow et al., 1985; Lister, 1987). Drugs that decrease anxiety increase exploration of the open arms (e. g., Pellow et al., 1985).

The behavior of *C. auratus* seems to be the product of a similar approach-avoidance conflict.

If this is true, the experimental model here presented could be used as a test to investigate anxiety and anxiety drugs. Time spent by the fish in the dark chamber could be considered as expressing avoidance of the white (aversive) chamber, as time spent in enclosed spaces of elevated plus-mazes, elevated T-mazes and open-fields is taken as an indication of avoidance of open spaces of the apparatus (Graeff et al., 1993, 1996; Pellow et al., 1985; Zangrossi and Graeff, 1996). The distribution of rat's exploration in these apparatus, in terms of number of entries into unprotected areas and time spent there, is analogous to the distribution of fish localizations obtained in the present study. This analogy qualify this experimental model as possible anxiety test. Further studies, involving pharmacological, physiological and behavioral aspects are necessary for such a validation.

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