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# ETHOLOGICAL ASPECTS OF *Hoplias curupira* (ERYTHRINIDAE) IN A CONFINED ENVIRONMENT, UNDER DIFFERENT VISUAL STIMULI

## Aspectos etológicos sobre *Hoplias curupira* (Erythrinidae) em ambiente confinado, sob diferentes estímulos visuais

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## ABSTRACT

The species of Giant trahira that inhabit the Amazon region have adaptation to different aquatic environments and unique predatory behavior. *Hoplias curupira*, is found mainly in the Araguaia-Tocantins and Xingu basins. It belongs to the Erythrinidae family and was recently described by Oyakawa & Mattox in 2009. It has morphological and genetic characteristics distinct from other trahira, and mainly seeks headwaters and fast waters. Its dark brown patterns disguise stony environments, allowing it to feed on a variety of prey. The study of fish behavior is essential for their *ex-situ* maintenance, where it interferes with their natural behavior, making it necessary to provide adequate conditions. This study investigated how changing environmental colors affect camouflage behavior and pattern. Where the tests were carried out using different colors of cardboard as the background of the aquarium, visual contrasts, tones and hues were explored. Aiming to analyze behavioral responses to these chromatic stimuli.

**Keywords:** Characiformes, feeding behavior, adaptability, camouflage.

## RESUMO

As espécies de trairão que habitam a região amazônica possuem adaptação a diferentes ambientes aquáticos e comportamento predatório singular. *Hoplias curupira*, é encontrada principalmente nas bacias Araguaia-Tocantins e Xingu, pertence à família Erythrinidae e ordem Characiformes, e foi recentemente descrito por Oyakawa & Mattox em 2009. Tendo com características morfológicas e genéticas distintas das demais traíras, e busca principalmente as cabeceiras e águas rápidas. Seus padrões marrom-escuro dissimulam ambientes pedregoso, possibilitando alimentar-se de uma variedade de presas. O estudo do comportamento em peixes é essencial para sua manutenção *ex situ*, onde interfere em seu comportamento natural, sendo necessário fornecer condições adequadas. Este estudo investigou como a alteração das cores do ambiente afetam o comportamento e o padrão da camuflagem. Onde os testes foram realizados utilizando diferentes cores de papel cartão como fundo do aquário, exploraram contrastes visuais, tons e matizes. Objetivando analisar as respostas comportamentais a esses estímulos cromáticos.

**Palavras-chave:** Characiformes, comportamento alimentar, adaptabilidade, camuflagem

## INTRODUCTION

The fish studied here, *Hoplias curupira* Oyakawa & Mattox, 2009, is a species widely distributed in the north of South America, popularly known as “giant trahira” and strictly living in freshwater, it belongs to the family Erythrinidae, order Characiformes (Sassi et al., 2021). This organism, recently separated from *H. macrophthalmus*, has aroused interest among researchers, aquarium enthusiasts and aquaculturists, due to its adaptation in different environments and predatory behavior (Guimarães et al., 2022a).

Such organisms can be found in the basins that form the Araguaia-Tocantins hydrographic complex, where the holotype was collected, as well as the others that form the drainage network of the Amazon River (Ramos et al., 2022). Its occurrence has been recorded in several rivers, mainly in areas with exposed rocks, rapids, and clear waters, sometimes in streams with temperatures lower than the region's standard (Saviato et al., 2020).

Considered a predatory fish, medium to large (Saviato et al., 2021), with its dark appearance, suitable for life near the rocky bottom (Salaro et al., 2022), it facilitates the capture of food by lurking (Rodrigues et al., 2022). And thus, it is an essential component for the aquatic community (Quéméré et al., 2021), as its ecological attributes help to balance the food chain (Oliveira et al., 2019). Likewise, their presence is associated with good quality environments, as they are sensitive to environmental disturbances, such as pollution and environmental degradation (Saviato et al., 2022a).

Therefore, the study of fish behavior is important to understand their needs, and thus, enable the creation of subsidies for good maintenance practices appropriate in confined environments (Reis et al., 2019). Likewise, understanding the natural behavior of these organisms is essential to ensure that, in the laboratory, an environment is stimulated that meets their requirements and promotes the animal's well-being (Rodrigues et al., 2021).

In this sense, ethology studies cover a wide variety of experiential aspects, which include feeding, reproduction, establishment of social hierarchy, communication, and swimming patterns (Kolmann et al., 2021), as these characteristics eventually leave it exposed in the environment (Lopes et al., 2022). In the same way, reproductive displays highlight the animal in the middle and among the others (Fernandes et al., 2021).

There are several organisms that could change their color at specific times, such as situations related to stress, reproduction, or feeding, characterized by a feeding frenzy. Furthermore, this chromatic mutability can also be triggered by pressures originating from the scenic compositions of the environment (Culbert et al., 2020). At the same time, communication between individuals is established through visual, olfactory and sound signals, with direct repercussions on social interactions and the defense of territories (Kohda et al., 2022), and may even correlate with swimming patterns in cohesive schools, up to the inclination towards solitude (Silva & Gramkow, 2019).

When placed in restricted environments, such as laboratories or entertainment aquariums, these beings face a series of obstacles that impact their natural attitudes (Guimarães et al., 2021b). In this sphere, variables such as the size of the space, the quality of the water, the presence of ornaments, the nuances of the environment and coexistence with other individuals emerge as preponderant influences on their behavior and level of tension (Gneiding et al., 2019). These elements provide refuges, encourage exploratory behaviors, and can catalyze spontaneous interactions between specimens (Agrillo et al., 2020).

Because, in these places, the absence of space and environmental stimuli, or an excess of these, not only affects their eating habits and well-being (Saviato et al., 2022b), but can also lead to limited social interactions, culminating in starvation or attempts at evasion (Campelo et al., 2019). Combined with these factors, water quality is a preponderant element for the comfort of these captive organisms (Mariano et al., 2021), since inadequate conditions, such as oxygen levels, pH and concentration of toxic compounds, have the potential to impact them adversely (Saviato et al., 2023). It is therefore imperative to monitor and maintain water quality in accordance with parameters specific to the demands of each species in confined environments (Wolff & Donatti, 2016).

Therefore, the present study aimed to investigate how changing colors in the confinement environment affect the behavior and color pattern of *H. curupira*.

## METHODOLOGY

The experiment took place between 03/2017 and 12/2019, lasting 32 months. During this period, changes in the behavior of the species *Hoplias curupira* Oyakawa & Mattox, 2009, were observed in the biology laboratory of the SESI School. The individual chosen was a male that was already 40 cm long at the beginning of the test, having been acquired legally. During this study, it was housed in a rectangular aquarium, designed to provide an environment that simulates nature and is conducive to its development (Figure 1).



**Figure 1.** Male specimen of *Hoplias curupira* Oyakawa & Mattox (2009), kept in the laboratory during the experiment.

The enclosure where it was kept is an aquarium with common patterns and dimensions of 80 cm long, 50 cm high and 60 cm wide. Natural decoration remained constant throughout the experiment and consisted of natural trunks, low plants, and background creepers, including *Hygrophila difformis* (L.fil.) Blume and *Hydrilla verticillata* (L.f.) Royle. The substrate used was coarse, inert river sand, typical and found in the region, which was washed and properly sterilized.

Temperature and light conditions were controlled to provide a stable environment for the fish. The aquarium temperature was maintained at a constant 28°C using a thermostat. Lighting was provided by 4 fluorescent LED lamps of 20W each, programmed to provide 12 hours of light per day. These environmental conditions did not suffer external interference.

An external Hang On filtration system, with a flow of 850 liters/hour, was used in the aquarium. Media for fixing denitrifying bacteria were added to the filtration system to ensure water quality. The parametric conditions of the water were periodically monitored through manual reading and chemical titration. Parameters such as pH, ammonia, nitrite, nitrate, and temperature were evaluated to ensure that water conditions were suitable for the well-being of the fish and did not interfere with the objective of the study (Figure 2).



**Figure 2.** Decoration and filtration of the enclosure for the experiment, presenting plants and natural trunks collected in the region and used in the aquascape composition.

Environmental color exposure tests were carried out, in which the color of the aquarium's back wall was changed. Each test lasted 24 hours and an interval of three days was provided between exposures to allow the fish to rest. During this interval, the fish was fed with 4 to 5 individuals of *Astyanax novae* Eigenmann, 1911, provided as live food.

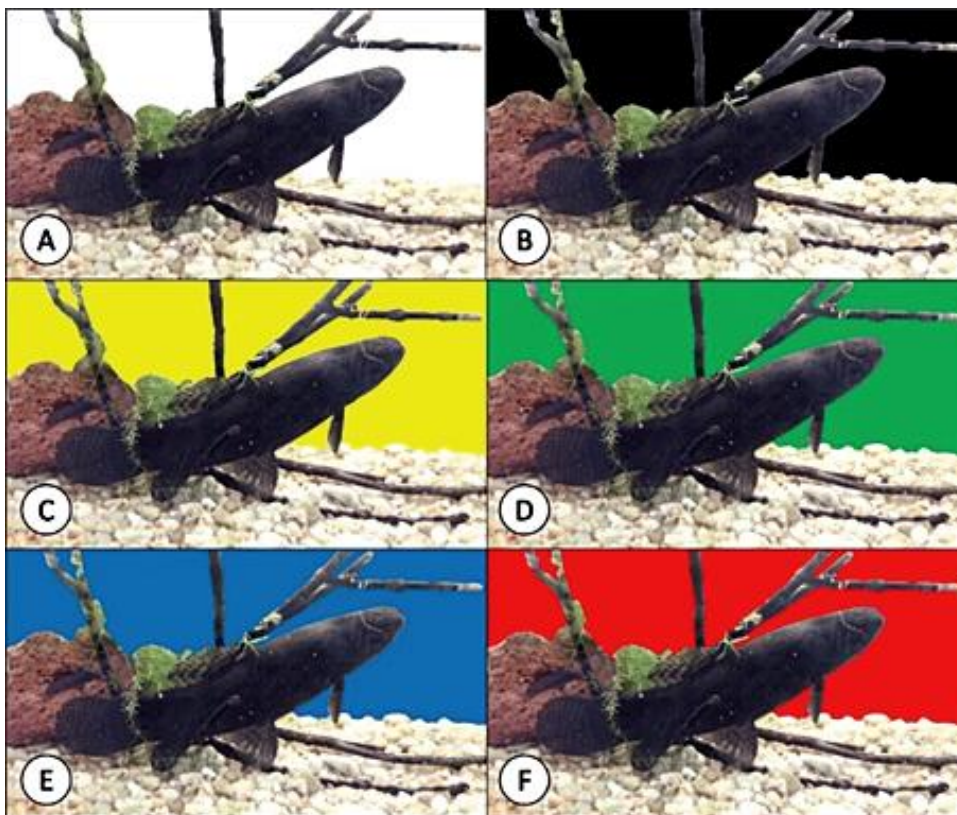
In this experiment, it was decided to use cardboard sheets in different shades as the backdrop for the aquarium. The colors chosen included white, black, green, blue, yellow, and red, being distributed randomly, but with the same incidence for each one. This selection was based on their relevance, aiming to explore possible behavioral reactions and adaptations associated with these chromatic stimuli (Escobar-Camacho et al., 2019).

White and black cardboards were included to represent light and dark extremes, allowing the fish's response to visual contrasts to be assessed (Geller et al., 2020). White tends to reflect more light, while black tends to absorb it, and this can influence the fish's visual perception and behavior (Cullbert et al., 2020). The choice of green, blue and yellow aimed to explore the effect of different tones and hues. Green and blue are commonly associated with natural aquatic environments, such as rivers and lakes, while yellow can represent elements such as aquatic plants or potential prey (Eördegh et al., 2022).

In this way, such colors can trigger specific responses in fish, related to camouflage, visual communication, search for food or territory recognition (Kohda et al., 2022). In the same way, changes in the color patterns of the natural environment, sometimes due to their displacement in the Home range, sometimes due to changes in the colors of the water due to climatic events.

However, red was included due to its importance in previous studies with other fish, demonstrating that many species present distinct responses to this stimulus (Maximova et al., 2021). And so, the presence of this hue can trigger aggressive behaviors, making its investigation relevant in this context (Parker et al., 2020).

Therefore, by using this variety of colors, we sought to cover a broad chromatic spectrum and explore possible behavioral responses of the fish in relation to different visual stimuli. The random fixation of colors and the equal number of repetitions for each color aimed to minimize possible experimental biases and guarantee the validity of the results obtained (Figure 3).



**Figure 3.** Examples of how the color changes to the back wall of the aquarium occurred: A) White cardboard; B) black; C) yellow; D) green; E) blue; and F) red.

During each test, the behavior of the *H. curupira* individual was carefully observed and noted. Performances were subdivided into color change, predatory action, apparent stress, or peaceful and inactive behavior. These observations allowed a detailed assessment of the fish's responses to different background colors in the aquarium.

After each environmental color exposure test, the data collected were tabulated and statistically analyzed. Where the frequencies of the different observed behaviors were recorded and compared between the different background colors. Likewise, this study was carried out in accordance with ethical guidelines for animal research and all precautions were taken to ensure the well-being and health of the fish used in the present experiment.

## RESULTS

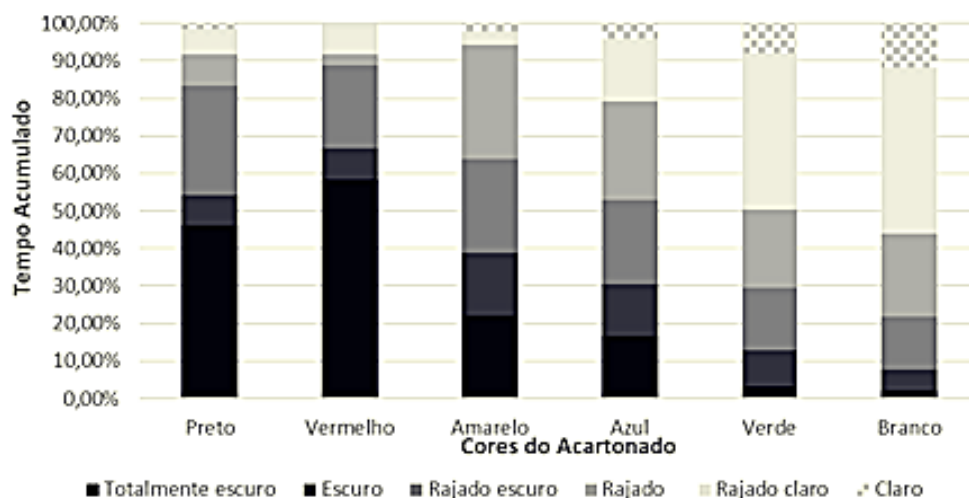
Observation of the behavior and coloring of *Hoplias curupira* in a controlled environment showed changes in its color as the color of the bottom wall of the aquarium changed. During the experiment, it was possible to notice that the animal spent most of its time in a specific position within the enclosure, positioning itself just below the branches in the center of the aquarium, with its head facing the surface.

Its color varied widely, presenting tones that varied from very dark, almost black, to lighter colors, allowing the visualization of the large, decorated scales and the opercular spot, distinctive characteristics of the Giant trahira group.

Thus, during this study, it was observed that the fish darkened its color when exposed to backgrounds in red, black, and yellow, while it exhibited lighter tones in response to backgrounds in blue, green and white. The animal's dark color persisted almost 80% of the time it was exposed to the red background. Likewise, when presented with a green background, it behaved more peacefully and displayed a light color practically all the time (Table 1 and Figure 4).

**Table 1.** Relationship of colored cardboard plates and the accumulated time for each chromatic change reaction of *Hoplias curupira*.

Animal coloring	Aquarium background					
	Black	Red	Yellow	Blue	Green	White
Totally dark	46,11%	58,33%	22,22%	16,67%	3,06%	2,22%
Dark	8,06%	8,33%	16,67%	13,89%	9,72%	5,56%
dark streak	29,17%	22,22%	25,00%	22,22%	16,67%	13,89%
Gust	8,33%	2,78%	30,56%	26,39%	20,83%	22,22%
Light burst	6,94%	8,33%	3,33%	16,67%	41,67%	44,44%
Clear	1,39%	0,00%	2,22%	4,17%	8,06%	11,67%



**Figure 4.** Relationship between the accumulated time of the experiment with the permanence of the chromatic reaction in the studied animal (*Hoplias curupira*) (totally dark; dark; dark striped; striped; light striped; and light), and exposure to cardboard colored with the colors used.

Such results are of great relevance for understanding the behavior and adaptation of *H. curupira* in a controlled environment. The fish's ability to change its color in response to different visual stimuli highlights the importance of the environment in the expression of its behavior and can provide valuable information to improve its management and well-being in captivity (Figure 5).



**Figure 5.** Main color patterns of *H. curupira*, observed during the experiment and recorded immediately after removing the colored background: A) Totally dark; B) Dark streak; C) Light burst; and D) Clear with details on the scales.

Likewise, during this experiment, it was observed that the animal exhibited predominantly static behavior, showing little or no movement. This characteristic indicates that the organism is an ambush predator, which passively waits for its prey instead of being an active hunter. Furthermore, it was found that the fish's feeding behavior was restricted to capturing prey, followed by returning to the resting position, most of the time.

## DISCUSSION

The analysis of actions and chromatic transformations in fish not only emerges as a tool to investigate their adaptations and interactions with the habitat (Mazzoni & Barros, 2021), but also proves to be relevant for understanding the behavior of *H. curupira*. Which is distributed in neotropical basins of the Amazon region (Mata et al., 2023). The exploration of its pigmentation changes, as well as its behavior sheds light on these and the relationship with ecological pressures, the state of well-being and the complex behavioral activities of this organism (Anjos et al., 2022).

In this way, the present experiment outlined the movement of *H. curupira* in the enclosure, preferably with an inert posture, seeking shaded places in the aquarium, characteristic of ambush feeding (Ferraz et al., 2019), a typical strategy for predator species in this group. (Lopes et al., 2022).

Therefore, the nuances observed in the color of *H. curupira*, in response to changes in the color of the aquatic bottom, present recurring variations (Pires & Zuanon, 2021). Where, the palette of tones, from dark to light, reveals patterns found in their natural environment, such as rocks and submerged trunks, and highlighting the pink scales and the opercular spot, distinctive elements of the traitor group (Marques et al., 2021).

During the experiment, *H. curupira* showed a chromatic adaptation, where each background color revealed different body tones for the animal (Honorato et al., 2021). Therefore, during exposure to red, black, and yellow cards, the fish presented darker tones and apparent opacity (Fernandes et al., 2022). Where the darker tone, during exposure to red cardboard, proved to be more persistent than the others (Wilson et al., 2021; Davis et al., 2020), in 80% of the time observed (Green et al., 2019).

In contrast to this, the blue, green and white backgrounds caused the organism to display lighter and brighter tones (Santos & De-Carli, 2018). This result suggests that there is a more pronounced chromatic

response to the red background than to the others (Silva & Canova, 2019), an expression that may be related to behavioral adaptation or an intrinsic visual communication of the species (Santos et al., 2021).

On the other hand, the phenomenon of pink tones, in its lighter and slightly darker color, stands out as clashing with other organisms in the Erythrinidae family (Andrea et al., 2019). Therefore, such inferences, corroborated by previous studies, present color plasticity in freshwater fish, most of the time correlated to environmental conditions (Sousa et al., 2023).

Thus, these chromatic variations, in addition to standing out as visual elements, play an important role in social dynamics (Montoya-Ospina et al., 2020), in individual identification and selection of partners (Fernandes et al., 2021).

However, this study, despite its relevance, is just one of the many characteristics of the organism, where other variables such as predators, food supply and seasonal factors (Saviato et al., 2023), have an influence on the chromatic reflections of this species (Giao et al., 2020). And in short, these results are lectures on the ability to adapt to different scenarios, an understanding for the conservation of freshwater fish species (Luz et al., 2019). Where, these results become important tools for understanding and preserving aquatic ecosystems (Reichenthal et al., 2019).

Therefore, such observations are essential for understanding the behaviors and feeding strategies of these organisms (Lima et al., 2019). And, when we consider the role of *H. curupira* as an ambush predator, we envision a camouflage strategist, as an efficient instrument in the search for food (Soares et al., 2023), also considering, energy savings, minimum effort for maximum gain (Silva et al., 2023).

However, these inferences are not immutable or end the interpretative possibilities of this experiment, as the deepening of these hunting and feeding strategies, together with the exploration of environmental (Soares et al., 2020), and seasonal influences on these behaviors, require further studies focusing on environmental control and segregation of possible environmental influences on the organism's behavior (Fernandes & Moron, 2020).

Finally, this study revealed a relationship between the variation in aggressiveness of this fish and the lunar phases, mainly during the full moon. It is another behavioral observation that, although not used as a metric for this test, needs to be explored further (Pessoa et al., 2019). Where such a relationship brings with it new questions and the need to understand the mechanisms underlying this behavior (Chiao & Hanlon, 2019).

## CONCLUSIONS

In conclusion, the observation of behavior and color changes in *Hoplias curupira* played a fundamental role in understanding their adaptations and interactions with the environment. Thus, as it is a fish widely distributed in neotropical Amazonian regions, it revealed that its color varies according to the color of the background of the environment. This variation is related to the fish's adaptive responses to different visual stimuli. However, during the experiment, the animal adopted behavior related to predatory stalking behavior.

Where it was possible to observe a clear change in the color of the fish in response to different color conditions. And so, when exposed to backgrounds in bright, dark colors, the fish darkened its color, while on backgrounds in lighter, brighter colors, it acquired lighter tones. These results are in line with previous studies that emphasize the plasticity of coloration, highlighting its ability to change appearance according to environmental conditions. However, these studies were not related to the animal in question, emphasizing its originality and potential for scientific contribution.

Furthermore, viewing dark or light colors cannot be interpreted as indicating stress. Normally, the color of fish in the environment varies from dark to dark striped more frequently, with lighter shades being found only during the night. More research is needed to better understand the relationships between fish color and their well-being.

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