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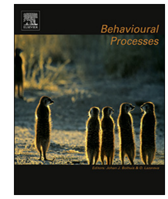

Short report

Lower light intensity reduces larval aggression in matrinxã, *Brycon amazonicus*

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ABSTRACT

Brycon amazonicus shows a high frequency of aggressive behavior, which can be a limiting factor in intensive farming systems. Environmental changes can modulate the social interactions of fish and reduce aggression during the different stages of production. Groups of three larvae at 12 h after hatching (HAH) were subjected to different levels of light intensity: low (17 ± 3 lx), intermediate (204 ± 12.17 lx) and high (1,613.33 ± 499.03 lx), with eight replicates for each level. The lower light intensity reduced the frequency of aggressive interactions and locomotor activity exhibited by the animals. Based on these results, light intensity modulates aggression in *B. amazonicus* larvae. Manipulation of this factor could improve the social conditions of this species during farming and contribute to the development of new production technologies.

1. Introduction

Aggression in fish has been a major obstacle in farming systems, as it compromises the growth and survival of the animals. According to Kaspersson et al. (2010) and Manley et al. (2014), aggressive interactions may be exacerbated in production systems due to confinement and the high density of individuals.

During aggressive displays in fish, there is an intense expenditure of energy that changes hematology and biochemistry, reduces growth, and alters behavior (Wendelaar-Bonga, 1997). In addition, aggressive interactions may result in lesions and bodily injuries that increase susceptibility to disease and weaken the animals, making them more vulnerable to predation or death (Huntingford et al., 2006). The stress caused by aggression can also compromise the welfare of fish and, consequently, reduce the yield of the production system (Conte, 2004).

A number of factors modulate the frequency and intensity of aggressive behavior in fish, including group size (Andries and Nelissen, 1990; Quinn et al., 1996), developmental phase (Holder et al., 1991; Jaroensutasinee and Jaroensutasinee, 2003), hormone levels (Villars, 1983; Trainor and Hofman, 2006), previous social experience (Nelissen and Andries, 1988), gender (Carvalho and Gonçalves-de-Freitas, 2008) and environmental parameters (Nicieza and Metcalfe, 1999; Sloman et al., 2001). Naumowicz et al. (2017) describe the state of knowledge about the neuroendocrine and immune modulation of aggressive behavior, with an emphasis on the impact of neuromodulators (i.e., serotonin, cortisol and neuropeptide Y) on the aggressiveness in fish.

Several studies have sought to minimize aggressive behavior in fish through the use of different types and sizes of feed (Miki et al., 2011), probiotic supplementation (Dias et al., 2012), thyroid hormones (Leonardo et al., 2013), tryptophan (Wolkers et al., 2014), changes in the shape and color of the aquariums (Pedreira et al., 2006) and reductions in stocking densities (Van-de-Nieuwegiessen et al., 2009). However, the effect of light intensity on fish behavior is poorly understood, especially its potential modulation of aggressive interactions in larvae.

The matrinxã, *Brycon amazonicus* (Spix and Agassiz, 1829), is a species of fish native to the Amazon basin that has proved successful in breeding systems in Brazil and other South American countries. It is the second most highly produced species in Northern Brazil because it exhibits rapid growth relative to the provision of artificial feed as well as desirable feed conversion and zootechnical performance (Zaniboni-Filho et al., 2006). However, *B. amazonicus* exhibits a high frequency of aggressive behavior and cannibalism (Ferraz and Gomes, 2009; Wolkers et al., 2012; Sampaio-Nakauth et al., 2016), and evidence indicates that the aggressive behavior begins at the larval stage (e.g., Senhorini et al., 1998; Souza et al., 2014). According to Senhorini et al. (2002), the hatching of matrinxã faces substantial technological barriers because larvae loss can reach 90% due to aggressiveness and cannibalism at this

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stage. This can be a limiting factor in intensive breeding systems.

In aquaculture systems, physical environmental factors are commonly manipulated to improve the production of commercial species (Franke et al., 2013). For example, light intensity greatly influences biological rhythms and modulates diverse physiological functions in different species of fish, which may be reflected in survival (Mukai et al., 2013), food intake (Puvanendran and Brown, 2002; Kitagawa et al., 2015), metabolic rate (Porter et al., 2001), weight gain (Perkin et al., 2014; Biswas et al., 2016), locomotor activity (Trippel and Neil, 2003; Navarro et al., 2014), body pigmentation, sexual maturation and reproduction (Fujii et al., 1992; Song et al., 2016). Environmental changes trigger the release of chemical substances such as melatonin that may interfere with the social interactions of the animals.

Variation in the release of melatonin might mediate the effect of light on aggressive behavior, as light intensity and photoperiod modulate aggression in fish (Carvalho et al., 2012, 2013) and the release of melatonin is reduced in response to light exposure (Ekström and Meissl, 1997; Falcón et al., 2010). We hypothesize that increasing light intensity increases aggression in *B. amazonicus*. This hypothesis is based on the finding that melatonin levels are reduced during periods of high luminosity (Martinez-Chavez et al., 2008) and the fact that reductions in this hormone increase aggressiveness in fishes (Munro, 1986). Understanding the modulation of aggressive behavior through environmental factors, such as light intensity and/or photoperiod, is extremely relevant to improving the performance, profitability and sustainability of *B. amazonicus* farming practices.

2. Materials and methods

2.1. Rearing conditions

The experiment was performed at the Center for Technology, Training and Aquaculture Production located in Presidente Figueiredo (AM), Brazil. The larvae were produced by induced reproduction using crude carp pituitary extract as described by Romagosa et al. (2001). After extrusion, the oocytes were fertilized and stored in 60-L incubators with constant water renewal at a temperature of 27.0 ± 0.3 °C, a pH of 6.6 ± 0.3 and a density of 1 g of eggs per L of water (1 larva/mL)

2.2. Experimental design

The influence of light intensity on aggression in matrinxã larvae was evaluated by comparing groups of three larvae subjected to different levels of light intensity: low (17 ± 3 lx), intermediate (204 ± 12.17 lx) and high (1,613.33 ± 499.03 lx). Each treatment was replicated eight times.

The average light intensity for each experimental condition was maintained by sampling three points within the incubator. The low light level was obtained by using standard lighting in the laboratory with fluorescent bulbs (λ = 400–700 nm). The intermediate and high light levels were obtained by using standard lighting in the laboratory and one 5 W or 13 W fluorescent bulb placed 10.0 cm from the water surface. Light intensities were measured using a portable digital lux meter (LD-511, ICEL, Manaus, Brazil) to check the maintenance of the tested light levels. A treatment of 204 ± 12.17 lx was close to the light level in the production system.

Time zero was the point at which 50% of the larvae had hatched (Romagosa et al., 2001), and the larvae were observed for 12 h after hatching (HAH) because the frequency of aggression is higher at this stage of development (Souza et al., 2014). For each experimental treatment, three specimens were collected from the incubator and transferred simultaneously to a glass bowl (2 cm × 1.5 cm) with 3 ml of water (as performed by Souza et al., 2014), which was equivalent to the density in the incubator (1 larva/mL). The water temperature was maintained at 26.9 ± 0.5 °C.

Because body size affects aggressive behavior (Beeching, 1992), groups were formed using fish of standard length and similar body weight (ANOVA, $F < 1.70$; $P > .21$).

2.3. Behavior analysis

The fish were acclimated for 10 min, and behavior was then recorded for a period of 20 min. The aggressive interactions were quantified based on the ethogram described by Souza et al. (2014): approach, attack, chase, flight, mouth fight, frontal display and threat. We considered the total contest, corresponding to the sum of all the behavioral units.

The locomotion rate was quantified based on the frequency of movement during each observation period (Olla et al., 1978; Sabate et al., 2008). A sheet of graph paper was placed under each glass bowl to improve the analysis method. The type of motor activity was classified as one of the following: circular swimming – larvae move from one place to another in a circle – and linear swimming – animals move from one square to another in a straight line. The sum of swimming (Total) was also taken into account, as described by Souza et al. (2014).

2.4. Data analysis

The data were analyzed to identify discrepant values and were then tested for normality and homogeneity by the Shapiro-Wilk test and the Fmax test, respectively. The frequency of aggressive interaction was compared between experimental treatments by using the Kruskal-Wallis test. Locomotor activities under the different light intensities were compared by ANOVA. The LSD test was used for multiple comparisons, and differences were considered statistically significant at $\alpha \leq 0.05$. The above analyses were based on Zar (1999).

2.5. Ethical note

This study was approved by the Ethics Committee on Animal Use of the Federal University of Amazonas, Manaus, AM (protocol n° 035/2011).

3. Results

The lower light intensity treatment reduced the frequencies of approach, attack, chase, flight, threat, and total contest for the group (Kruskal-Wallis, $X^2 > 6.38$; $P < .04$; Fig. 1). A lower frequency of total locomotion was observed for animals subjected to the lowest light intensity (ANOVA, $F > 3.79$; $P < .04$; Fig. 2). There was no difference in the locomotor activity of the animals exposed to intermediate and high light intensities (LSD, $P > .05$).

4. Discussion

In general, light intensity interferes with the display of aggressive behavior in fish and disrupts their social context (Castro and Caballero, 2004; Carvalho et al., 2012; Mukai et al., 2013; Gaffney et al., 2016). The lower aggression can be related to the high melatonin production at low light intensities because the release of this hormone in fish is related to light exposure (Ekström and Meissl, 1997; Falcón et al., 2010). In contrast, the decline in this hormone with increased light intensity increases aggression in tilapia (*Oreochromis niloticus*), which influences the social stability of the school (Falcón et al., 2007; Carvalho et al., 2013).

The effect of melatonin on the motivational behavior system in vertebrates may result in less aggression, which is mediated by the release of neurotransmitters (e.g., Oliveira and Gonçalves, 2008). In addition, tryptophan and serotonin, which are melatonin precursors, also reduce aggression and cannibalism, thus increasing survival in fish (Hseu et al., 2003; Höglund et al., 2005; Lepage et al., 2005; Wolkers

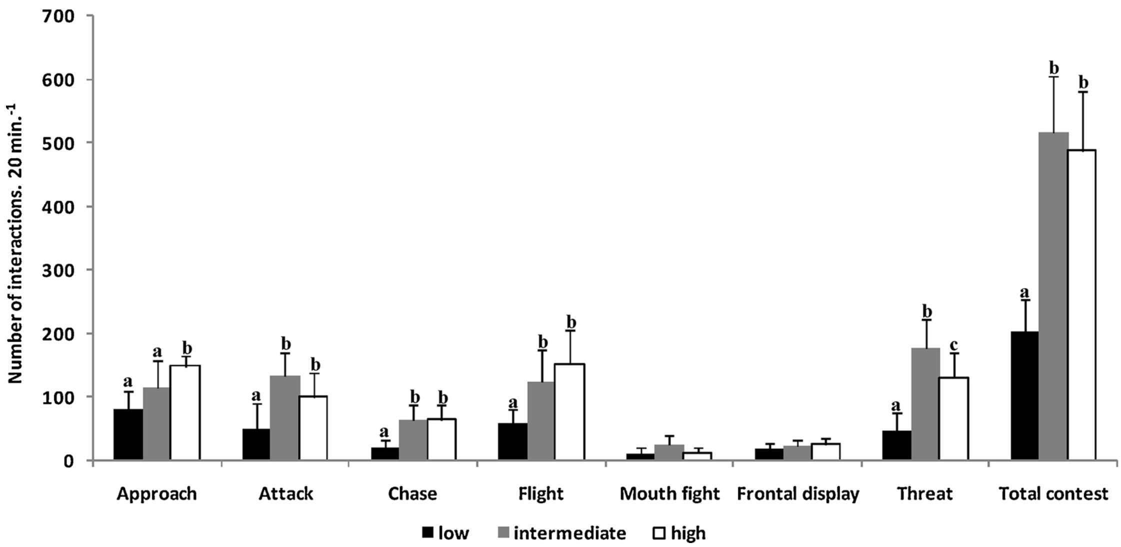

Fig. 1. Mean (± S.D.) frequency of aggressive interactions for the groups at low (17 ± 3 lx), intermediate (204 ± 12.7 lx) and high (1,613.33 ± 499.03 lx) light intensity. Different letters indicate significant statistical differences between treatments (LSD, $P < 0.05$).

et al., 2014). Therefore, it is likely that the low light intensity treatment increased melatonin production and consequently reduced the display of aggressive behavior in matrinxã.

These results stand in contrast to those of Sakakura and Tsukamoto (1997) and Castro and Caballero (2004), who observed lower aggression under high light intensity in *Seriola quinqueradiata* and *Diplodus sargus*, respectively. However, an increase in aggression was observed up to a certain light intensity limit, and beyond this threshold, agonistic interactions declined. Our results indicate that fish maintained at lower light intensity are less aggressive than fish exposed to high light intensity, but we observed no difference between the intermediate and high levels. Thus, the light intensity threshold that reduces the interactions may not have been reached for our matrinxã larvae, as the aggression was higher in treatments of greater light intensity.

The lower light intensity reduced the total amount of locomotion. The lower locomotor activity observed under low light could modulate aggression in matrinxã larvae. As observed by Souza et al. (2014), lower locomotor activity reduces the probability of encounters between animals and, consequently, the social interactions between group members. These results are similar to those of Villamizar et al. (2011) and Carazo et al. (2013), who found low locomotor activity in dark periods and high activity in light periods for fish larvae. In addition, increased locomotion under high light intensity is associated with reduced melatonin in teleosts (Ekström and Meissl, 1997), and low melatonin has been shown to modulate the behavioral responses of juvenile *Melanogrammus aeglefinus* and *Astyanax bimaculatus* in the presence of light (Trippel and Neil, 2003; Navarro et al., 2014).

5. Conclusions

The evaluation of factors that modulate aggressive behavior, such as light intensity, is extremely relevant to improving the performance, profitability and sustainability of *B. amazonicus* cultivation. Lower light intensity (17 ± 3 lx) reduced the display of aggressive interactions between matrinxã larvae at 12 HAH, indicating that this environmental factor can be manipulated to improve conditions for this species during larviculture and other growth phases.

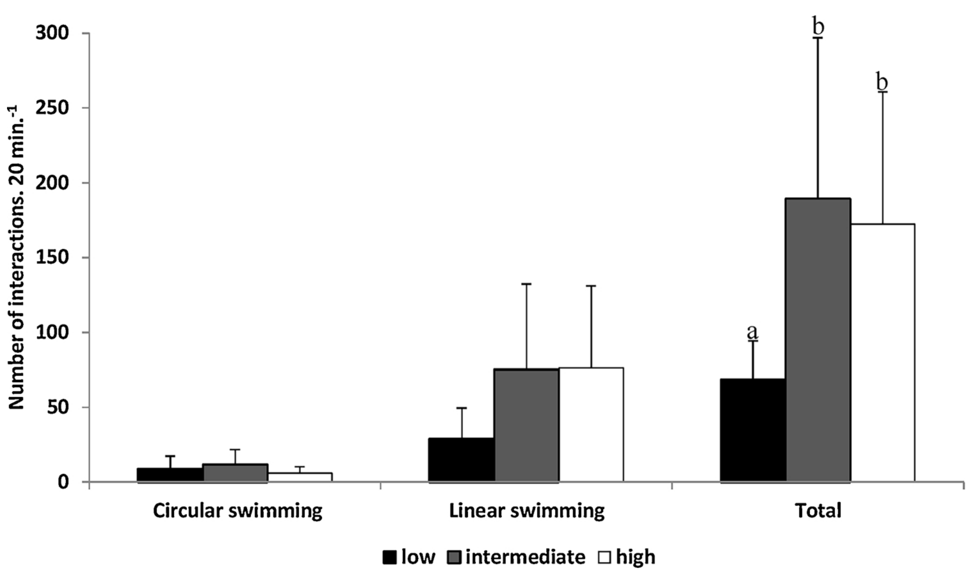

Fig. 2. Mean (± S.D.) frequency of locomotor activity for the groups at low (17 ± 3 lx), intermediate (204 ± 12.7 lx) and high (1,613.33 ± 499.03 lx) light intensity. Different letters indicate significant statistical differences between treatments (LSD, $P < 0.04$).

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