

PARASITISM BY *Gussevia asota* IN GILLS OF JUVENILES OF *Astronotus ocellatus* CULTURED IN THE PERUVIAN AMAZON**Patrick Mathews Delgado^{1*}, John Paul Mathews Delgado², Rosa Ismiño Orbe³**¹ Department of Parasitology, Institute of Biomedical Sciences, University of São Paulo, São Paulo, Brazil² Department of Botanic, National Institute of Amazonian Research, Manaus, Amazonas, Brazil³ Department of Aquatic Ecosystems, Research Institute of the Peruvian Amazon, Iquitos, Loreto, Peru

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Abstract: The present study aims to evaluate the monogenean infestation in *Astronotus ocellatus* bred in a fish farm in the Peruvian Amazon. Fifty individuals of the species *A. ocellatus* were collected between July and August 2011 from a semi-intensive fish farm, located in the northeast of the State of Loreto, Peru. This study identified a high infestation in gills by monogenean specie of *Gussevia asota* in a cultivation of *A. ocellatus*. The prevalence was 100%, with mortality of all fish. The mean intensity and mean abundance of the parasite were 189.8 of parasites per individual. This is the first report of infestation high by *G. asota* in juveniles of *A. ocellatus* cultured from the Peruvian Amazon.

Keywords: *Astronotus ocellatus*, *Gussevia asota*, Parasites of fish, Peruvian Amazon

Öz: **Peru Amazonunda Yetiştirilen *Astronotus ocellatus* Balığı Yavrularının Solungaçlarında Karşılaşılan *Gussevia asota* paraziti**

Bu çalışma, peru amazonunda yetiştirilen *Astronotus ocellatus* balığında görülen monojen istilasını değerlendirmektedir. 2011 senesinde temmuz ve ağustos aylarında 50 adet *A. ocellatus*, kuzeydoğu Loreto eyaleti Peru'da bulunan yarı intensif balık üretim çiftliğinden alınmıştır. Çalışmada, örneklerin solungaçlarında yoğun olarak *Gussevia asota* istilasının olduğu görülmüştür. İstila tüm örneklerde (100%) görülmüş ve tamamı ölmüştür. Bireylerde ortalama yoğunluk ve miktar birey başına 189.8 parazit olarak hesaplanmıştır. Bu peru amazonunda yetiştirilen *A. ocellatus* bireylerinde *G. asota* istilasının ilk raporudur.

Anahtar Kelimeler: *Astronotus ocellatus*, *Gussevia asota*, Balık parazitleri, Peruvian Amazon

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Introduction

In fish farming the intensive exploitation allows the handling of high densities of organisms per unit area. Indeed, this type of management frequently leads to break the balance between pathogen and host, consequently resulting in the emergence of infectious and parasitic diseases that cause various problems ranging from slow up growth, reduced fertility rates, until the appearance of severe epidemics resulting in high mortality (Thatcher 1991; Scholz 1999; Cable et al., 2002).

Cichlids have a wide geographical distribution. Currently there are 1,533 known species, with 320 reported for South America (Kullander 1988). These species inhabit a wide variety of aquatic ecosystems. Moreover, the fish represent high economic importance, given that they are marketed for human nutrition with a promising potential for intensive and extensive aquaculture (Kullander and Ferreira 2006; Araujo et al., 2009).

The oscar *Astronotus ocellatus* (Agassiz, 1831) can reach up to 45 cm in length and 1.6 kg of total weight (Fracalossi et al., 1998) and Oscar is a much appreciated species for its meat, which has a firm consistency and lacks intramuscular bones with great acceptance on the Amazonian market being regarded as a food fish of the highest quality. The *A. ocellatus* is normally found in Amazon floodplain areas, and is characterized as a hypoxia tolerant species (Almeida-val et al., 1995; Almeida et al., 2000). Recent experiments carried out in laboratory have shown that the adult animals tolerate 6 h anoxia at 28°C by reducing their standard metabolic rates (Muusze et al., 1998). Due to its zootechnical characteristics, the *A. ocellatus* is considered a species with great potential for management in controlled environments aiming human nutrition and ornamental purposes. However, to allow the breeding to become entirely feasible, it turns out the necessity to solve the problem of diseases and parasites upsurge affecting this species in controlled environments, as a consequence of intensive farming under inadequate management (Varella and Malta 1995).

Therefore, with the gradual increase of intensive and semi-intensive fish farming in the Peruvian Amazon, there is a need for constant monitoring of the fish for the diagnosis and timely control of infestations by monogeneans. In this

sense, the present study aims to evaluate the monogenean infestation in *A. ocellatus* bred in a fish farm in the Peruvian Amazon.

Materials and Methods

Between July and August 2011, which corresponds to the relative dry season, 50 individuals of the species *A. ocellatus* were collected with drag nets, from a semi-intensive fish farm, located in the northeast of Loreto (Peru), between latitudes 3° 48' 48.9" N and 073° 19' 18.2" W, with average annual temperature of 26.3°C and relative humidity of 85% at 328 mean sea level.

Fish were fed twice daily with extruded diet containing 25% crude protein and 2.6 Mcal/kg of digestible energy and feeding rate of 5% of the biomass of the pond. The sampled fish presented length of 12.60 ±0.10 cm and weight of 50.08 ±0.86 g. Having identified the parasite infestation, the fish were transferred to concrete tanks covered with tiles to undergo long-term baths containing 0.5 to 2.0 ppm of potassium permanganate and 1% formalin during one hour. In the absence of improvement, we sacrificed and burned all the fish from the respective pond.

Using a stereoscope we examined the body surface, fins, nostrils, mouth, opercula and gills, looking for possible injuries and excess of mucus production. By means of a scalpel, we also performed scraping of the skin, fins and gills to observe possible attached parasites.

For examination of the gills, the samples were separated and placed in glass containers with a 1:4,000 formalin solution. After one hour, the gills were stirred in the liquid and then removed from the container. Helminths were allowed to settle on the bottom and were subsequently collected with the aid of a small probe and a dissecting microscope (Nikon SM-30). The identification of the parasites was based on the methodology of Kritsky et al. (1989), Thatcher (2006) and Abdallah et al., (2008).

To study the monogeneans, permanent slides were prepared with total parasites assembly according to Thatcher (1991). For the study of sclerotized structures, parasites were fixed in a solution of ammonium picrate glycerine (GAP) and mounted in Canada balsam. Some specimens were mounted unstained in Gray and Wess' medium. To visualize internal structures, parasites

were fixed in hot formaldehyde solution (4%) for staining with Gomori's trichrome. The parasitic indexes calculated for assessing the level of infestation of parasites in the fish were prevalence, mean intensity and mean abundance (Bush et al., 1997).

Results and Discussion

The necropsy of juveniles from *A. ocellatus* bred in controlled environments in the Peruvian Amazon evidenced the infestation by the monogenean *G. asota* in the gill filaments of the fish.

Indeed, the totality of the examined fish showed a high parasitic infestation by *G. asota*. The mean intensity was equal to the mean abundance, provided that the number of parasitized fish was the same as those examined (Table 1).

Several studies report the parasitism of neotropical cichlids by monogeneans belonging to the genus *Gussevia* Kohn and Paperma 1964 (Kritsky et al., 1989; Vidal-Martinez et al., 2001; Mendoza-Franco et al., 2010; Yamada et al., 2011; Mathews et al., 2013a). For south America thirteen species of *Gussevia* have been described for eight species of cichlids (Kritsky et al., 1986), evidencing a high specificity of the genus *Gussevia* in parasitizing cichlids.

In the Central and Peruvian Amazon several species of monogeneans of the genus *Gussevia* have been reported parasitizing cichlid of economic importance for human nutrition and ornamental purposes (Kritsky et al., 1989; Mendoza-Franco et al., 2010; Azevedo et al., 2010). However, little is known about the parasitic infections of farmed *A. ocellatus*, because these studies have been carried out, in general, in wild fish. In our study we report for the first time the parasitism by the *G. asota* in juveniles of *A. ocellatus*

bred in controlled environments in the Peruvian Amazon.

In the study described herein, the juveniles of *A. ocellatus* presented high levels of parasitism by the monogenean *G. asota*. Parasites that have a direct life cycle, such as monogeneans, are more frequently found in lentic environments. Moreover, this type of environment favors the transmission of these parasites (Flores-Crespo et al., 2003; Azevedo et al., 2007), which justifies the fact that the fish had elevated parasite infection, since the same are confined to their culture in earthen ponds where water circulation is almost negligible or nonexistent.

According to Buschmann (2001) and Mariano et al. (2010), intensive fish farming generates a large accumulation of organic matter on the pond bottom produced from the excreta, dead matter and the fraction of uneaten food. This organic matter produces hypoxia and anoxia that creates an unbalance in the homeostasis of the fish, eventually leading to the increase of the oxidative stress of biomolecules, promoting thus various physiological and biochemical alterations, causing cell impairment and death (Sherry 2003; Van der Oost et al., 2003). Therefore, these adverse effects of poor water quality reduce the self resistance of the fish, which turns out as a favorable condition to the parasite proliferation. The *A. ocellatus* is normally found in Amazon floodplain and is characterized as a hypoxia tolerant species (Almeida-Val et al., 1995). However, while adult animals are found in hypoxic waters, juveniles of this species are active and may be easily found in superficial water body layers, where oxygen availability is higher, suggesting a reduced capacity to tolerate hypoxia among juveniles. This fact may justify the high parasitic infestation by *G. asota* in *A. ocellatus* juveniles from fish farming.

Table 1. Parasitic indexes of *Gussevia asota* in juveniles of oscar *Astronotus ocellatus* cultured in the Peruvian Amazon.

Parasitic indexes	<i>Gussevia asota</i>
Prevalence (%)	100
Abundance (Count)	9490
Mean abundance \pm S.D	189.8 \pm 1.7
Mean intensity \pm S.D	189.8 \pm 1.7
Range of intensity	60-175

The results described herein are in accordance with Kritsky et al. (1989), Azevedo et al., (2010), and Kim et al. (2002), who found the monogenean *G. asota* parasitizing gills of *A. ocellatus* and, being common the setting of this kind of monogenean parasite in this organ (Kritsky et al., 1989). Indeed, several studies report the parasitism of *G. disparoides* in gills of *H. severus*, *C. amazonarum* and *C. ocellaris* and all these fish species are currently being raised in confined environments in the Peruvian Amazon.

Furthermore, in a study with *A. ocellatus* captured from the wild, Abdallah et al. (2008) found 62.8% of prevalence and mean intensity of 17.6 monogeneans of *G. asota*. However, the results differ from our study where we found a prevalence of 100% and mean intensity of 189.8 parasites of *G. asota*. A possible reason for the low levels of infestation reported by Abdallah et al. (2008) may be fact that the fish originate from nature. Nevertheless, Kritsky et al. (1989) found 100% prevalence of *G. asota* in *A. ocellatus*, although without informing other parasitic indexes.

Parasites of the genus *Gussevia* are considered specific for cichlids and therefore may show low susceptibility when present in favorable breeding conditions. Kritsky et al. (1989) mentioned that *G. asota* apparently can cause the death of its host, citing the case of an aquarium in Idaho, USA, as an example. This is the first report of *G. asota* parasitizing *A. ocellatus* in fish farming in the Peruvian Amazon. The results of this study and studies addressing various aspects of parasite in other species bred in the same region (Mathews et al., 2007; Dinis et al., 2007; Mathews et al., 2011; Mathews et al., 2013b; Mathews et al., 2013c) confirm the necessity of constant monitoring of fish, seeking the diagnosis and timely control of infestations by monogeneans, in order to reduce fish mortality.

Conclusion

This is the first report of infestation high by *G. asota* in *A. ocellatus* cultured from the Peruvian Amazon. *G. asota* infection probably contributed to the mortality of the captive cichlids.

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