MAINTENANCE OF THE BUTTERFLYFISH Chaetodon striatus (PERCIFOMES: CHAETODONTIDAE) IN A RECIRCULATION SYSTEM

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ABSTRACT

The high extractivism of marine fish to supply the demand of the aquarium market has worried the researchers and, in this context, the aquaculture consists of an alternative to the problem. The family Chaetodontidae represents one of the main families of ornamental marine fish and has not received enough attention on their cultivation research. Considering the absence of this information about the butterflyfish Chaetodon striatus, the present work had the objective of obtaining information about the species in captivity. The fishes were kept in tanks of 100, 200, 300 and 1,000 L, with 6 fishes in each tank, with 3 repetitions for each volume, totaling 72 individuals and were observed for 30 days, being offered living small invertebrates and frozen and fresh mussels for the feeding. Also, for the attempt of reproduction in captivity, two couples, collected directly from the sea, with the entry of only two adults in the trap at a time, were used. Each couple was acclimated in a 100 L tank and we observed their behaviors for 8 hours a day. There was no mortality in tanks of 300 and 1,000 L and, for feeding, small invertebrates and fresh mussels were well accepted. Some behaviors were observed, most of which were already described for chaetodontids. This is the first report of C. striatus in captivity and this contribution may provide subsidies for a future technological package.

KEYWORDS: Aquarism; behavior; ornamental marine fish; Chaetodon striatus

INTRODUCTION

The production of ornamental fishes is considered a highly profitable activity (Araujo et al., 2012) and even the marine species representing only 10% of the aquarium market, this activity is expanding (Sampaio & Nottingham, 2008). The high values achieved by this market, increase the sector and certainly the name of the luxurious hobby is pertinent, on a basis of unit weight, ornamental fish constitute the most valuable commodity in the world (Hardy, 2003). According to the Food and Agriculture Organization of the United Nations (FAO - UN), the values of the ornamental fish trade have grown significantly over the past few decades, and between 2000 and 2011 global exports of ornamental fishes increased from 181 million to 372 million dollars (Manning et al., 2019). If considered the retail sales values, associated materials, wages and not exported products, industry values may reach 15 billion dollars annually (Bartley, 2000; Whittington & Chong, 2007; Prang, 2008; Moorhead & Zeng, 2010).

The ornamental fish industry is characterized by a set of numerous species from different countries, which became evident throughout the year 2000, when approximately one billion ornamental fishes were exported, coming from more than 100 countries located in tropical regions, such as Brazil, Colombia, Indonesia, Malaysia, Nigeria, Peru, Sri Lanka and Thailand. These countries, sometimes still in development, find a source of income by marketing these organisms to developed countries (Moravec et al., 1999).

Despite all the economic benefits generated by this activity, there is another emerging issue that has aroused the researchers concern - exacerbated extraction. Around 90% of the marine fishes species used for this purpose are captured from the natural environment (Rubec et al., 2001; Calado, 2006; Olivotto et al., 2006), because there is no control over the production of these reef and coastal fishes in captivity, especially in Brazil.
In this context, marine ornamental aquaculture is part of effective management plans, since species produced in captivity can supply market demand, thus reducing extraction for this purpose (Tlusty, 2002; Bellwood et al., 2004).

Among the main families of marine ornamental fishes, the Chaetodontidae family stands out, is highly requested in the aquarium market because it includes species that have attractive coloring and different morphotypes (Nagpure et al., 2006). This family has approximately 130 species, known as butterflyfish, distributed in 11 genera present in tropical and sub-tropical oceans (Pratchett et al., 2013). Butterflyfish generally live in monogamous pairs for years and each pair inhabits a certain area on the reef or on the coast, defending itself from other pairs of the same species (Driscoll & Driscoll, 1988).

The Chaetodon striatus (Linnaeus, 1758) species, popularly known as banded butterflyfish, is found on consolidated bottoms (rocky and coral reefs) in tropical waters, in the Western Atlantic Ocean, from Massachusetts, United States to Santa Catarina, Brazil, including the Gulf of Mexico and the Caribbean Sea (Carvalho-Filho, 1999; Pinheiro et al., 2017). Its body is flattened laterally and has a color that varies from yellowish-white to beige, always with four black vertical bars, and juveniles have an ocellus in the posterior portion of the body (Sampaio & Nottingham, 2008).

The popularity of butterflyfish in the aquarium trade makes them obvious candidates for consideration in the development of captive breeding methods (Ogawa & Brown, 2001). Besides, according to Wabnitz et al. (2003) this family ranked the sixth most traded fish in the world in 2003 and, from 2004 to 2005, the USA, alone, imported 97 species of chaetodontids (Rhyne & Tlusty, 2012).

A fact already known by researchers and hobbyists is that many marine ornamental fishes represent a challenge for production, since there is no mastery of technology and there are no reports about maintenance and reproduction in captivity, such as the butterflyfish C. striatus. Although some species of butterflyfish are rustic or even unfeasible in aquariums, as Chaetodon capistratus, which has a highly specialized diet and restricted to certain species of corals (Wabnitz et al., 2003), there are many promising species with good characteristics for the activity (Pratchett et al., 2013). In addition, the development of products and technologies in ornamental aquarium in recent decades has provided the solution to major problems.

Despite their importance in aquarium, butterflyfish have not received the necessary attention, and research in captivity is scarce. Captive breeding has been carried out in some species as Forcipiger flavissimus (Madden & May, 1977), Chaetodon nippon (Suzuki et al., 1980), Chaetodon modestus (Tanaka et al., 2001) and Chaetodon miliaris (Degidio et al., 2017) and the comparison between captive and wild spawning in Chaetodon nippon (Suzuki et al., 1980), Chaetodon aculeatus, Chaetodon capistratus (Colin, 1989), Chaetodon multicintcus (Lobel, 1989), Chaetodon trifascialis (Yabuta & Kawashima, 1997) and Chaetodon modestus (Tanaka et al., 2001). As for C. striatus, the gametogenesis of males and females has recently been described (Bayona-Perez et al., 2020), being fundamental for the future production of the species.

Therefore, in an attempt to acclimatize this species in captivity and to induce the entry of couples in courtship, followed by a possible spawning, the present study aimed to keep individuals in tanks of different volumes (100, 200, 300 and 1,000 L) in a recirculation system, to verify whether there is acceptance of live small vertebrates and frozen and fresh mussels in the diet and to check the behavior of captive couples in the attempt of reproduction.

MATERIAL AND METHODS

Specimen Collection

The collections and maintenance of the fishes were carried out in accordance with the rules of the Ethics Committee on Animal Experimentation of the Biosciences Institute of Botucatu (protocol number 805 - CEEA-IBB/UNESP). The specimens of Chaetodon striatus were collected in Ubatuba, São Paulo, Brazil, from March 2015 to June 2017, using covo type traps implanted in rocky shores close to Itagua beach and through diving with a hand net, in Rapada Island. The traps were made in a trapezoidal and square format, covered with plastic mesh tied with a 0.5 mm nylon line, remaining semi-fixed in the sea through ropes and anchors. The capture depths varied from 3 to 8 m (depending on the variation between low and high tides).

During the collection of specimens, we frequently observed the entry of only two adults in the traps at a time, forming a couple, which were selected for the breeding attempt.

Specimen Maintenance

After collection, the specimens were transported alive in thermal boxes and in 60 L gallons, with sea-
Then, the fish were acclimated, gradually changing the water used for transportation to the water in the tanks where they would stay. That done, the individuals were distributed in tanks with a saltwater recirculation system, equipped with mechanical filtration (100 micron bag), chemical filtration through ozone, skimmer and sterilization of water through ultraviolet lamps. The saltwater recirculation system was installed in a masonry building of 300 m². The circulation rate of the tanks was 200% (the total volume of water in the tanks was renewed twice a day). The tanks were siphoned daily, once a day.

As a maintenance strategy for the recirculation system, the skimmer’s storage “cup” was removed daily and washed with freshwater to eliminate the accumulated residue. The “bag” filter was also removed daily and washed with blast water under pressure. All pumps received a monthly cleaning of the impellers to avoid locking. The water temperature was kept constant at 28 ± 1.0 ºC. The photoperiod was maintained at 12L:12D and salinity at 35 ppt.

The fishes were kept in tanks of 100, 200, 300 and 1,000 L, with 6 fishes in each tank, with 3 repetitions for each volume, totaling 72 individuals and were observed for 30 days. In tanks starting from 200 L, 50 cm long polyvinyl chloride (PVC) tubes with 15 or 25 cm of diameter were placed, which functioned as a shelter for the fishes (Figure 1).

To feed the individuals, we used nets of 30 cm, which remained in the sea for 30 days. In these nets there was a process of succession of organisms, becoming replete with live food such as larvae of various invertebrates and polychaetes. These nets were removed from the sea and tied to the tanks where they remained for 7 days. Another food used was frozen and fresh mussels, which had their liquid removed and offered with the shells open or in small pieces. For each fish was offered a unit daily.

Reproduction Attempt

For the attempt of reproduction in captivity, two couples, collected directly from the sea, with the entry of only two adults in the trap at a time, were used. In order to not separate the couples, these couples were not kept with other fishes. Among the specimens collected, the female had a bulging abdomen and both individuals with total length ranging from 12.5 to 15.0 cm, corresponding to adult specimens.

Each couple was acclimated in a 100 L tank, under the conditions mentioned above (Figure 2). The observation of these individuals was carried out for 8 hours a day.

Figure 1. Tanks used for the maintenance of Chaetodon striatus in captivity. A: 100 L tank battery; B: 200 L tank battery; C: 300 L tank battery and D: 1,000 L tank. Figures A to C show all the external area of the tanks and figure D the internal area with the animals.
Data Analysis

All the individuals were measured regarding the weight and length and the mean values were calculated.

RESULTS AND DISCUSSION

Maintenance of Specimens

The repetitions of each volume tank presented very similar results regarding weight and length of fishes. So, we calculate the average values of all specimens, weight and length were 52.0 g and 11.7 cm, respectively.

All specimens kept in captivity, in all repetitions of all volumes tanks were able to feed properly from the first day of maintenance, with great acceptance of live small invertebrates and frozen and fresh muscles, that were offered daily and accepted each time. Despite this, the fishes kept in tanks of 100 and 200 L (Fig. 1A-B) were unable to survive, with mortality of all individuals by the fourth day of captivity in the tanks of 100 L and for the 200 L tanks, by the eighth day of captivity (in all three repetitions of each volume tank). Mortality was due to aggression among individuals that increase in smaller tank volumes.

In the 300 L tanks (Fig. 1C) there were fewer aggressions and mechanical shocks, and there was no mortality, probably because there were more space and volume of water for the fishes. Likewise, there was no mortality of any individual in the 1000 L tanks (Figure 1D).

To be able to compare the feeding behavior of a higher number of individuals, we used three repetitions of each tank volume, totaling 72 fishes, this way we could compare if all the fishes accepted the same food sources in the same amount (every day). The numbers of repetitions provided calculation of the mean values of weight and length of fishes, with a more reliable result. Also this way, we can assure that the water volume of the tank affect the survival of fishes. One or two repetitions could induce error, and there are no necessary of a higher collection of specimens, spearing this way, this population.

Although many species of marine ornamental fish represent a challenge for breeding in captivity, the present study demonstrated that it is possible to acclimatize ornamental species, even those considered difficult to maintain, such as butterflyfish.

One of the greatest difficulties in keeping wild species in captivity concerns food. However, in the present study, it can be proved that live foods were easily accepted by Chaetodon striatus, possibly because it is a generalist zoobentivorous species (Liedke et al., 2016). However, this fact does not rule out the possibility of the animal being gradually conditioned to feed on dry food or even frozen food, as was also observed in this work, which allows its breeding away from the marine environment.
In the present study, the reproduction in captivity of *C. striatus* proved to be appropriate in large spaces, as in other species of butterflyfish (Michael, 2004), avoiding the mortality of individuals, as occurred in the experiments in tanks of 100 and 200 L. Considering the natural aggressiveness of the species, a space of around 300L is required for management in captivity (Michael, 2004), allowing individuals to occupy their own pre-established niches, avoiding attacks and territorial disputes among specimens, as observed in *Chaetodon lunulatus* (Yabuta & Berumen, 2013).

**Entry into Courting**

The couples kept individually in 100 L tanks were fed properly and there was no mortality. After a 2-day acclimatization period, each couple started to exhibit certain behaviors, such as “standing next to each other in opposite directions”, “swimming on a carousel with one fish in front and one behind”, “male touches the abdomen of the female with snout”, “parallel swimming” and “face to face”, which are presented in Figure 3.

To verify the sexual maturation of these couples, two pairs of wild fish were dissected. The gonads of these individuals were sexually mature, occupying a large portion of the abdominal cavity. The testicles were whitish in color and the ovaries were orange in color, with visible oocytes, easily detaching from the gonad (Figure 4).

![Behaviors presented by *Chaetodon striatus*. A: Standing next to each other in opposite directions; B: Carousel swimming with one fish in front and one behind; C: male touches the female’s abdomen with the snout; D: Parallel swimming and E: Face to face.](image-url)
As the couples were captured and kept together, there was a good acceptance by both individuals in captivity. Often, failure to recognize the partner can lead to attacks and territorial disputes (Yabuta & Berumen, 2013) hindering the interaction between the couple. This is due to the fact that the species is monogamous (Liedke et al., 2016), form pairs for spawning (Whiteman & Côté, 2004) and defend itself against other pairs of the same species (Breder & Rosen, 1966). Therefore, it was essential in this study to keep couples already formed, always together, thus increasing the chance of entering into courting in captivity.

In the present study, we observed some characteristic behaviors already described for other species of butterflyfish in a natural environment, including those related to courtship. Many members of the Chaetodontidae family exhibit conspicuous behaviors, such as pairing, proximity and simultaneous swimming (Reese, 1975).

Among the behaviors observed in this study, only “standing side by side in opposite directions” and “face to face” had not yet been described for chaetodontids in the literature. The “Carousel swimming, with one fish in front and one behind” (Neudecker & Lobe, 1982), “male touching the female’s abdomen with the snout” (Neudecker & Lobe, 1982; Fricke, 1986; Yabuta & Kawashima, 1997) and “parallel swimming” (Neudecker & Lobe, 1982) had already been described.

The behavior “male touches the female’s abdomen with the snout” (Neudecker & Lobe, 1982; Fricke, 1986; Yabuta & Kawashima, 1997) is frequently observed in chaetodontids, with the male being able to touch the abdomen, the urogenital papilla as well as the female’s caudal fin. In a study on spawning behavior in a natural environment of C. striatus, the authors described that the male approached a female that had a bulging abdomen and touched the caudal fin with his snout (Colin & Clavijo, 1988). Another behavior related to the spawning of chaetodontids is the “parallel swimming” (Neudecker & Lobe, 1982).

Considering these behavioral aspects, there is no doubt that this is a big step observed in captivity, which can lead to the next stage, which culminates in the reproduction of the specimens. There is still a lot to know about the social structure of butterflyfish, including aggression, territoriality and the mating system (Yabuta, 2000).

Research on the maintenance of ornamental marine fishes in captivity worldwide is scarce (Moorhead & Zeng, 2010). Therefore, a future concentration of efforts for the development of the sector, carried out by more research institutions, could reflect in the mitigation of the exploitation of these resources and in the generation of income, especially for developing countries. In the same sense, the strengthening of the relationship between researchers and the productive sector may favor new perspectives for ornamental aquaculture.

CONCLUSIONS

For the maintenance of Chaetodon striatus, tanks with more and equal to 300 L are suitable and the live foods used here demonstrate viable alternatives for the nutrition of these fish in captivity. Still, elementary information about the possible obtaining of the breeders as well as the behaviors described in this work, can contribute to a future technological package of breeding of the species.

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REFERENCES

Araujo, A.P., Santos, F.W.M., Pincinato, S., & Silva, J.G. (2012). Gestão participativa no comércio de animais aquáticos ornamentais (ensaio). Revista de Educação Continuada Em Medicina Veterinária e Zootecnia Do CRMV-SP, 10(2/3), 6–15.

Bartley, D. (2000). Responsible Ornamental Fisheries. FAO - Fishery Resources Division.

Bayona-Perez, I.L., Mazzoni, T.S., & Quagio-Grassiotto, I. (2020). Cellular development of the germinal epithelium during the female and male gametogenesis of Chaetodon striatus (Perciformes: Chaetodontidae). Zygot 28(4), 291-299. https://doi.org/10.1017/S0967199420000118.

Bellwood, D.R., Hughes, T.P., Folke, C., & Nystrom, M. (2004). Confronting the coral reef crisis. Nature, 429, 827-833.

Breder, C.M. & Rosen, D.E. (1966). Modes of reproduction in fishes. T. F. H. Publications.

Calado, R. (2006). Marine ornamental species from European waters: a valuable overlooked resource or a future threat for the conservation of marine ecosystems/Scientia Marina, 70(3), 389–398.

Carvalho-Filho, A. (1999). Peixes: costa brasileira. Editora Melro.

Colin, P.L. & Clavijo, I.E. (1988). Spawning activity of fishes producing pelagic eggs on a shelf edge coral reef, Southwestern Puerto Rico. Bulletin of Marine Science, 43(2), 249–279.

Colin, P.L. (1989). Aspects of the spawning of western Atlantic butterflyfishes (Pisces: Chaetodontidae). Environmental Biology of Fishes, 25(1–3), 131–141.

Degidio, J.M.L.A., Yanong, R.P.E., Watson, C.A., Ohs, C.L., Cassiano, E.J., & Barden, K. (2017). Spawning, embryology, and larval development of the Milletseed Butterflyfish Chaetodon miliaris in the laboratory. North American Journal of Aquaculture, 79(3), 205–215.

Driscoll, J.W. & Driscoll, J.L. (1988). Pair behavior and spacing in butterflyfishes (Chaetodontidae). Environmental Biology of Fishes, 22(1), 29–37.

Fricke, W. (1986). Pair Swimming and Mutual Partner Guarding in Monogamous Butterflyfish (Pisces , Chaetodontidae): a Joint Advertisement for Territory. Ethology, 73, 307–333.

Hardy, R. (2003). Introduction to the special issue on “ornamental fish.” Aquaculture Research, 34(11), 903.

Liedke, A.M.R., Barneche, D.R., Ferreira, C.E.L., Segal, B., Nunes, L.T., Burigo, A.P., Carvalho, J.A., Buck, S., Bonaldo, R.M., & Floeter, S.R. (2016). Abundance, diet, foraging and nutritional condition of the banded butterflyfish (Chaetodon striatus) along the western Atlantic. Marine Biology, 163(1), 1–13.

Lobel, P.S. (1989). Spawning behavior of Chaetodon multicolor (Chaetodontidae); pairs and intruders. In The butterflyfishes: success on the coral reef (pp. 125–130). Springer, Dordrecht.

Madden, W.D.; & May, R.C. (1977). Ornamental fish culture project. Final Report. MAC Task Order The Oceanic Institute and the Hawaii Institute of Marine Biology, 3, 23–23.

Manning, C.G., Foster, S.J., & Vincent, A.C.J. (2019). A review of the diets and feeding behaviours of a family of biologically diverse marine fishes (Family Syngnathidae). In Reviews in Fish Biology and Fisheries (Vol. 29, Issue 2).

Michael, S.W. (2004). Angelfishes & Butterflyfishes: Plus Ten More Aquarium Fish Families with Expert Captive Care Advice for the Marine Aquarist. Microcosm Limited.

Moorhead, J.A., & Zeng, C. (2010). Development of Captive Breeding Techniques for Marine Ornamental Fish: A Review. Reviews in Fisheries Science, 18(4), 315–343.

Moravec, F., Wolter, J., & Körtig, W. (1999). Some nematodes and acanthocephalans from exotic ornamental freshwater fishes imported into Germany. Folia Parasitologica, 46(4), 296–310.

Nagpure, N.S., Kumar, R., Satish K. Srivastava, Kushwaha, B., Gopalakrishnan, A., & Basheer, V.S. (2006). Cytogenetic characterization of two marine ornamental fishes, Chaetodon collare and Stegastes insularis. Journal of the Marine Biological Association of India, 48(2), 267–269.

Neudecker, S., & Lobe, P.S. (1982). Mating Systems of Chaetodontid and Pomacanthid Fishes at St . Croix. Ethology, 59, 299–318.

Ogawa, T. & Brown, C.L. (2001). Ornamental reef fish aquaculture and collection in Hawai. Aquarium Sciences and Conservation, 3(1–3), 151–169.

Olivotto, I., Holt, S.A., Carnevali, O., & Holt, G.J. (2006). Spawning, early development, and first feeding in the lemonpeel angelfish Centropyge flavissimus. Aquaculture, 253, 270–278.
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Pinheiro, H.T., Bernardi, G., Simon, T., Joyeux, J.C., Macieira, R.M., Gasparini, J.L., Rocha, C., & Rocha, L.A. (2017). Island biogeography of marine organisms. Nature, 549, 82–85.

Prang, G. (2008). An Industry Analysis of the Freshwater Ornamental Fishery With Particular Reference To the Supply of Brazilian Freshwater Ornamentals To the Uk Market. Scientific Magazine UAKARI, 3(1), 7–52.

Pratchett, M.S., Chong-Seng, K.M., Feary, D.A., Hoey, A.S., Fulton, C.J., Nowicki, J.P., Dewan, A.K., Walker, S.P.W., & Berumen, M.L. (2013). Butterflyfishes as a model group for reef fish ecology: Important and emerging research topics. In M.S. Pratchett, M.L. Berumen, & B.G. Kapoor (Eds.), Biology of Butterflyfishes (Issue January, pp. 310–334). CRC Press.

Reese, E. S. (1975). A Comparative Field Study of the Social Behavior and Related Ecology of Reef Fishes of the Family Chaetodontidae. Ethology, 37(1), 37–61.

Rhyne, A.L. & Tlusty, M.F. (2012). Trends in the marine aquarium trade: the influence of global economics and technology. International Journal of the Bioflux Society, 5(2), 99–102.

Rubec, P.J., Pratt, V.R., & Cruz, F. (2001). Territorial use rights in fisheries to manage areas for farming coral reef fish and invertebrates for the aquarium trade. Aquarium Sciences and Conservation, 3(1–3), 119–134.

Sampaio, C.L.S., & Nottingham, M.C. (2008). Guia de Identificação de Peixes ornamentais brasileiros (Vol. 1). Ibama.

Suzuki, K., Tanaka, Y., & Hioki, S. (1980). Spawning Behavior, Eggs and Larvae of the Butterflyfish *Chaetodon nippon* in an Aquarium. Japanese Journal of Ichtyology, 26(4), 334–341.

Tanaka, Y., Hioki, S., & Suzuki, K. (2001). Spawning behavior, eggs, and larvae of the butterflyfish, *Chaetodon modestus*, in an aquarium. Journal of the Faculty of Marine Science and Technology-Tokai University, Japan, 334–341.

Tlusty, M. (2002). The benefits and risks of aquacultural production for the aquarium trade. 205, 203–219.

Wabnitz, C., Taylor, M., Green, E., & Razak, T. (2003). From Ocean to Aquarium. The global trade in marine ornamental species. In UNEP - WCM C. UNEP World Conservation Monitoring Centre.

Whiteman, E.A. & Côté, M.I. (2004). Monogamy in marine fishes. Biology Reviews, 79, 351–375.

Whittington, R.J., & Chong, R. (2007). Global trade in ornamental fish from an Australian perspective: The case for revised import risk analysis and management strategies. Preventive Veterinary Medicine, 81, 92–116.

Yabuta, S. (2000). Behaviors in agonistic interaction of the butterflyfish (Chaetodon lunulatus). Journal of Ethology, 18(1), 11–15.

Yabuta, S. & Berumen, M.L. (2013). Social structure and spawning behavior of Chaetodon butterflyfishes. In M. S. Pratchett, M. L. Berumen, & B.G. Kapoor (Eds.), Biology of Butterflyfishes (pp. 200–225). CRC Press.

Yabuta, S. & Kawashima, M. (1997). Spawning behavior and harem mating system in the corallivorous butterflyfish, *Chaetodon trifascialis*, at Kuroshima Island, Okinawa. Ichthyological Research, 44(2), 183–188.