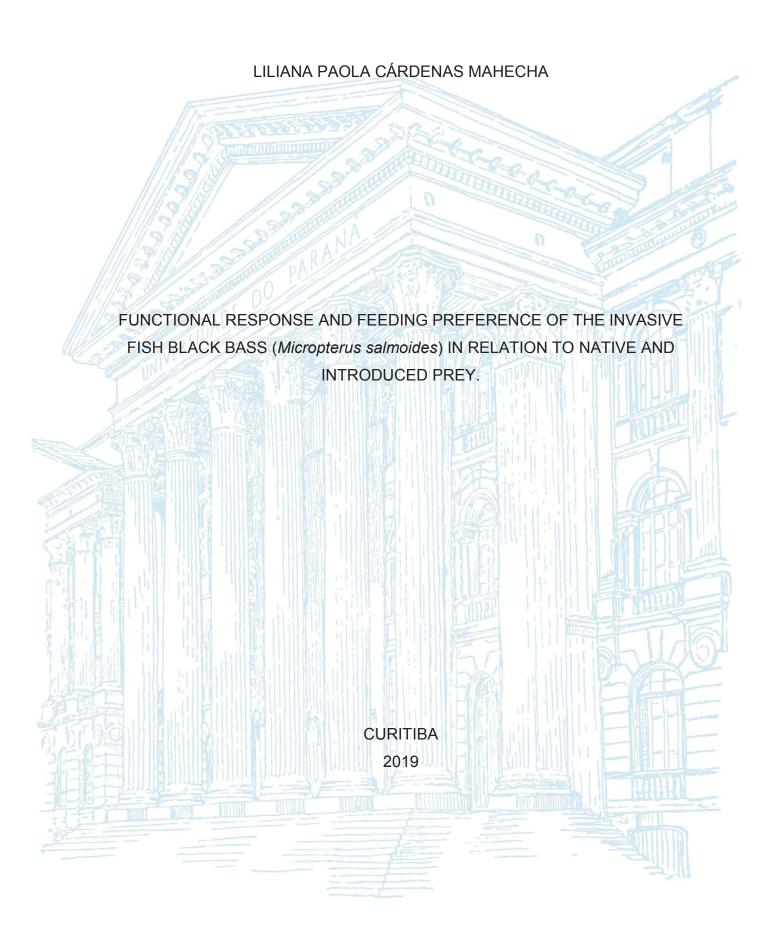
UNIVERSIDADE FEDERAL DO PARANÁ



LILIANA PAOLA CÁRDENAS MAHECHA

FUNCTIONAL RESPONSE AND FEEDING PREFERENCE OF THE INVASIVE FISH BLACK BASS (*Micropterus salmoides*) IN RELATION TO NATIVE AND INTRODUCED PREY.

Dissertação apresentada como requisito parcial à obtenção do grau de Mestre em Ecologia e Conservação, no Curso de Pós-Graduação em Ecologia e Conservação, Setor de Ciências Biológicas, da Universidade Federal do Paraná.

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RESUMO

A invasão biológica é uma das maiores causas de perda de biodiversidade. Entre seus impactos estão a mudança na abundância e riqueza de espécies nativas. O black bass (Micropterus salmoides) é uma das principais espécies invasoras no mundo. É originário da América do Norte e tem sido introduzido no Brasil para atividades pesqueiras. É generalista e tem alta voracidade. Nos reservatórios da região metropolitana de Curitiba, além do M. salmoides também foram introduzidas outras espécies, especialmente as tilápias (Oreochromis niloticus e Coptodon rendalli), que junto com a espécie nativa cará (Geophagus brasiliensis) compõe grande parte da dieta do *M. salmoides*. O objetivo deste estudo é medir a magnitude do impacto da predação por *M. salmoides* sobre a espécie nativa e as introduzidas através de análises da resposta funcional. Para isso executamos um experimento no qual se esperava que o *M. salmoides* se comportasse como 'generalista oportunista', apresentando uma curva de resposta funcional de tipo II segundo o descrito na literatura. O experimento consistiu em colocar um predador e diferentes densidades dos dois tipos de presas durante 48 horas, após esse período a quantidade de presas restante foi contabilizada, e determinamos o tipo de resposta funcional e seus parâmetros. Os resultados mostram que existe preferência do M. salmoides pelas presas introduzidas quando tem alta disponibilidade de alimento. Para os dois tipos de presa, a resposta funcional se ajusta ao tipo II, com uma diferença significativa no tempo de manipulação mais não na taxa de ataque. Nossos resultados são relevantes principalmente considerando a interação entre múltiplas espécies invasoras nos ecossistemas. Quando se tem mais de uma espécie introduzida em um mesmo local é importante o entendimento das relações tanto entre elas com as espécies nativas, quanto entre elas mesmas já que podem ter efeitos sinérgicos ou antagónicos, ou os dois ao mesmo tempo. As relações são complexas e sua compreensão é necessária para tomada de decisões de gestão ambiental.

Palavras-chave: Invasões. Predação. Equação de Rogers. *Largemouth Bass*. Neotropical. Reservatórios.

ABSTRACT

Biological invasion is one of the most important causes of biodiversity loss. Among its impacts are the change in abundance and richness of native species. The black bass (Micropterus salmoides) is one of the main invasive species in the world. It is native of North America and has been introduced in Brazil for fishing activities. It is generalist and has a high voracity. In the reservoirs of the metropolitan region of Curitiba, in addition to the *M. salmoides* other species were introduced, especially the tilapia (Oreochromis niloticus and Coptodon rendalli), which together with the native species (Geophagus brasiliensis) are a large part of the diet of the M salmoides. The objective of this study is to measure the magnitude of the impact of predation by M. salmoides on the native and introduced species through a functional response analysis. For that, an experiment was conducted in which it is expected that M. salmoides to behave as an 'opportunist generalist', presenting a type II functional response curve as described in literature. The experiment consisted of put a predator and different densities of two types of prey for 48 hours, after that period the remaining amount of prey was counted, and the type and parameters of functional response were determined. The results show that there is a preference of M. salmoides by the introduced prey, particularly when food availability was high. For both types of prey, the functional response is adjusted to type II, with a significant difference in handling time, but not in the attack rate. Our results are relevant considering the interaction of multiple invaders in an ecosystem. When more than one species is introduced in the same ecosystem, it is important to understand the relationships between them, and with the native species, since they can have effects synergistic, antagonistic or both at the same time. Relationships are complex and their understanding is necessary for environmental management decision-making.

Keywords: Invasions. Predation. Rogers equation. *Largemouth Bass.* Neotropics. Reservoirs.

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1 DISSERTATION PRESENTATION

Human play an important role modifying the original ecosystems (ALBERTI et al., 2003; HOBBS et al., 2006). This occurs mainly due to urbanization, which generates an increase in the demand for resources and services by the inhabitants of an urban nucleus (HAASE et al., 2014). To meet this demand, the land of native vegetation is conditioned for agriculture and livestock, in the same way the aquatic ecosystems are also altered to meet the needs of the human population (DEFRIES et al., 2004; MCKINNEY, 2006).

The metropolitan area of Curitiba city is located in the 'Primeiro Planalto Paranense' it is a region with abundant rainfall and a large water supply. However, the growth of the demand for water for public supply, made necessary to build reservoirs (HASSLER, 2006) altering the water dynamics and therefore the quality of habitat and biota (AGOSTINO et al., 2008). Many times, despite the creation of preservation areas such as the Environmental Protection Area of the Passaúna (Lei Estadual nº 13027/2000), the negative anthropic impacts continue with the expansion of the agricultural frontier, the influence of industrial activities and the contribution of domestic and agricultural effluents (YAMAMOTO, 2011). The dams construction also allowed the implementation of other anthropic activities such as aquaculture, for both human consumption and recreational uses such as sport fishing. This activity generates great impacts including the introduction of non-native species that can potentially become invasive (NAYLOR, et al., 2001) which, is known, is one of the greatest causes of loss of diversity in the world (CIRUNA et al., 2004).

Within this context it is important to understand the magnitude of the impact of introduced species on native species, understanding the complex network of relationships between them. For this, we present below the results of a dissertation that through an experiment aims to understand more clearly the depredation relationship between some of the most representative species of reservoirs in the Metropolitan area of Curitiba. The dissertation is formatted as a scientific article according to the norms of the Hydrobiologia journal.

FUNCTIONAL RESPONSE AND FEEDING PREFERENCE OF THE INVASIVE FISH BLACK BASS (*Micropterus salmoides*) IN RELATION TO NATIVE AND INTRODUCED PREY.

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ABSTRACT

The black bass (*Micropterus salmoides*) is one of the main invasive species in the world. In the reservoirs of the metropolitan region of Curitiba, in addition to the *M. salmoides* other species were introduced, especially (*Oreochromis niloticus* and *Coptodon rendalli*), which together with the native species (*Geophagus brasiliensis*) are a large part of the diet of the *M. salmoides*. The objective of this study is to measure the magnitude of the impact of predation by *M. salmoides*. For that, we performed an experiment of functional response. The results show that, there is a preference of *M. salmoides* by the introduced prey, particularly when food availability was high. For both types of prey, the functional response is adjusted to type II, with a significant difference in handling time, but not in the attack rate. Our results are relevant considering the interaction of multiple invaders in an ecosystem. When more than one species is introduced in the same ecosystem, it is important to understand the relationships between them, and with the native species, since they can have effects synergistic, antagonistic or both. Relationships are complex and their understanding is necessary for environmental management decision-making.

Keywords: Invasions, predation, Rogers equation, *Largemouth Bass*, Neotropics, reservoirs.

2 INTRODUCTION

As usual in areas near urban centers, the metropolitan area of Curitiba, Brazil has been severely altered by anthropic interventions, generating negative impacts on the original ecosystems and the native fauna (Lima, 2001). The rivers were dammed by building reservoirs that altered the dynamics of the ecosystems (Bonetto et al., 1989; Agostino et al., 2004; Reidy et al., 2012; Vitule et al., 2012) and favored the introduction of nonnative species (McAllister et al., 2001; Johnson et al., 2008) including the invasive fish black bass (Micropterus salmoides (Lacepède, 1802)), and the tilapias (Oreochromis niloticus (Linnaeus, 1758) and Coptodon rendalli (Boulenger, 1897)), the first one introduced for sport fishing and the second for food purposes (Santos, et al., 2008; Alvarez, et al., 2013). The introduced species can potentially become invasive that are one of the greatest causes of biodiversity loss in the world and one of the greatest challenges in the protection of ecosystems (Ciruna, et al., 2004), presenting ecological, socioeconomic and human health negative impacts (Ciruna, et al., 2004; Karatayev, et al., 2009). They can act as ecosystem engineers, changing the habitat of other species (Baxter, et al., 2004; Capps & Flecker, 2013) and modifying the networks of interactions within the communities (Benjamin, et al., 2013) spreading these impacts through trophic chains by top-down and bottom-up effects (Gallardo, et al., 2016; Penk et al., 2017) and changing abundance and richness of native species (Abekura, et al., 2004) through predation, hybridization, parasitism or competition.

Speaking specifically of the species introduced in these reservoirs *M. salmoides* is one of the main invasive species in the world and causes negative impacts in diverse regions of the globe (Abekura, et al., 2004), including in Brazil (Daga, et al., 2015; GISD, 2017). The natural distribution of this species occurs from the eastern of Canada to the Rio Grande basin in northwestern of Mexico (Alvarez, et al., 2013). Its presence was reported in Brazil since 1922 (Schulz & Leal, 2005). In this country *M. salmoides* has been widely distributed in artificial systems, as reservoirs, from Rio de Janeiro state to Rio Grande do Sul state (Schulz & Leal, 2005). The fact that this species reproduces and grows rapidly in semi-natural systems can be considered a threat to the conservation of Brazilian ecosystems (Schulz & Leal, 2005; Daga, et al., 2016). *M. salmoides* is a ravenous predator that generates strong pressure in native species (Daga, et al., 2015). Its diet includes small fish, macroinvertebrates, crustaceans,

amphibians and even aquatic plants, depending on their life stage (Hickley, et al., 1994; García-Berthou, 2002; GISD, 2017). On the other hand, *O. niloticus* and *C. rendalli* are also omnivorous species, adapted to an extensive culture, with a rapid growth and high importance in world aquaculture (Santos, et al., 2008). Also, considered a highly invasive species especially in the tropics (GISD, 2018). In several countries where is reported an adverse ecological impact after its introduction (Konings, et al., 2018).

A single invasive species has the capacity to alter the ecosystem and generate the impacts mentioned above, but given the current dynamics of globalization, most ecosystems have suffered the introduction of several invasive species simultaneously, making the understanding of interactions more complex that the generated between native and introduced species (Simberloff & Von-Holle, 1999).

One of the most conspicuous and easily quantifiable interaction between different species of fauna is predation. With respect to *M. salmoides*, previous studies show that their diet in the reservoirs is mainly composed of *O. niloticus* and *C. rendalli* and the native fish cará (*Geophagus brasiliensis* (Quoy & Gaimard, 1824)) (Ribeiro, 2013). These three species belong to the same family (Cichlidae) and have similar functional characteristics, so they are probably energetically equivalent. These three species as prey of *M. salmoides* are a good model to study predation relationships between native and non-native species in these reservoirs, for presenting high abundances and living there together for several years.

In this scenario, we asked if there is a food preference of *M. salmoides* for the introduced or native species, and if the functional response and its magnitude differs with respect to prey type. So, the objective of this study is to verify the magnitude of the impact on both native and introduced small fish.

It is possible to analyze these predation relationships through the functional response. Functional response has the ability to predict the impacts of piscivorous fish by describing the rate of resource capture as a function of their density (Dick, et al., 2013; Guo, et al., 2017). The functional response is the relationship between the number of prey consumed by predator per unit of time as a function of the density of the prey in a given space (Solomon, 1949; Holling, 1959; Fernández & Corley, 2004; Wasserman,

et al., 2016). Predators can present functional responses of type I, II or III (Holling, 1959) (Fig. 1). In type I, the predator increases consumption (Nc) linearly with increasing density (N) (Fig. 1a) with a fixed proportion (Nc/N), that is, it is denseindependent at low densities (Fig. 1b), but with the increase in density the curve reaches an asymptote because the predator is satiated. Thereafter, the number of prey consumed (Nc) remains constant and the consumption rate (Nc/N) decreases with increasing density, then the Nc/N becomes inverse dense-dependent, that is, the relationship between predator and prey it is not regulatory and the prey population can continue to increase (Juliano, 2001). This type of functional response is more common in filtering organisms, which have high food availability and require few capture effort (Holling, 1959; Fernández & Corley, 2004). Type II is where the consumption (Nc) increases with the density (N) of the prey (Fig. 1c), but the proportion (Nc/N) decreases due to the increase of the handling time of the prey with the density (Fig. 1d). This generates an inverse dense-dependent relation, making that in high densities the preys are less likely to be attacked than in low densities. This is the most common type of functional response found for example in vertebrates that has to use a large amount of energy in each prey (Holling, 1959; Fernández & Corley, 2004). Finally, in type III the consumption of prey depending on density results in a sigmoid curve, where the number of prey consumed per unit of time is accelerated with the increase in density until the handling time begins to limit the consumption (Fig. 1e). This type of functional response produces a dense-dependent mortality since prey in high densities are more affected than in low densities, thus exercising control over the populations (Fig. 1f) (Holling, 1959; Juliano, 2001).

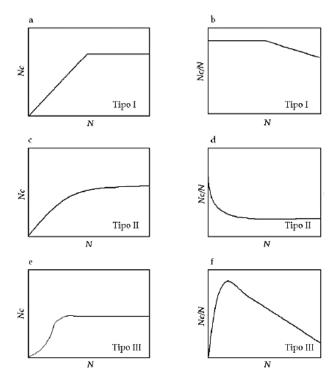


Figure 1. Three main types of functional response. "a", "c" and "e" showing the number of prey consumed (Nc) in relation to the number of prey offered (density (N)); and "b", "d" and "f" the relationship between the proportion of prey consumed (Nc/N) and the number of prey offered (N). Source: Juliano, 2001.

In this way, to respond to the objective we conducted an experiment of feeding preference and functional response (Fig. 2) with juvenile *M. salmoides* as predators, with fingerlings of tilapias (*O. niloticus* and *C. rendalli*) as introduced prey and fingerlings of *G. brasiliensis* as native prey. The hypotheses of this experiment were the following: I) Regarding the food preference, is expected that *M. salmoides* to behave as an opportunistic generalist (Hickley, et al., 1994; García-Berthou, 2002), without differentiating between native and introduced species. II) In relation to the functional response, it is expected that the resulting curve for native and introduced prey will be adjusted to a type II response, because according to literature it is the most common for vertebrates (Juliano, 2001).

The analysis of these functional responses allows us, through the type of response, to analyze if *M. salmoides* is exercising some kind of dense-dependent control over the prey populations and if this depends on the prey type. This giving us an idea of the magnitude of the impact on their populations. This allows us to improve the understanding of the interactions between several introduced and a native species, as well as an advance in the general knowledge of invasive species and provides a basis for environmental management decision-making.

3 MATERIALS AND METHODS

Juvenile *M. salmoides* (predators), and fingerlings of tilapias (O. *niloticus* and *C. rendalli*) as introduced prey and fingerlings of *G. brasiliensis* as native prey were collected at the Passaúna reservoir (Fig. 3) In March 2018. The reservoir is administered by the Paraná Sanitation Company (SANEPAR) located in the municipalities of Almirante Tamandaré, Araucária, Campo Largo, Campo Magro and Curitiba, in the state of Paraná, Brazil. The reservoir was inaugurated in 1979 and has an area of 14 km² with an approximate volume of 48 million m³ (Costa, et al., 2018). The Environmental Protection area of Passaúna was created by Decree 458 of 05/06/1991. The selected species present high densities in this reservoir and in general in the reservoirs of the metropolitan region of Curitiba. The prey collection done under permanent license for collection zoological material SISBIO N° 24779 and was done with trawl (10 mm) and the predators with manual rod, the transport to the laboratory was made in 50 L tanks with aerators to guarantee the oxygen concentration during transport.

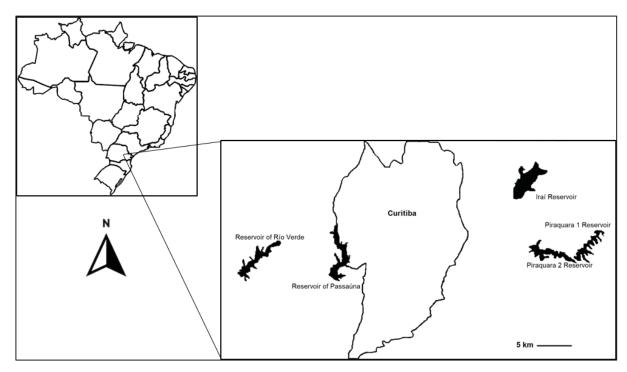


Figure. 2 Reservoirs in metropolitan area of Curitiba, the fish were collected in the Passaúna Reservoir.

The fish were taken to the laboratory where they were placed in 100 L aquariums and 310 L water boxes for acclimation. The process of acclimatization lasted approximately one month. The temperature remained around 23 °C. The preys were fed with commercial ration for fish, while the predators were fed with beetle larvae (*Tenebrio molitor* (Linnaeus, 1758)). During acclimation, both prey and predators were treated with Aqualife and Ictio (Labcon ®) to prevent disease.

After acclimatization in the stock of prey and predators, we made an acclimatization of the individual predators directly in the experimental boxes (35 L), with oxygenators and fed with tenebrios until they observed that they were feeding normally. Later, predators were starved for 72 hours. The experiments consisted in leaving a predator and different densities of prey. The two types of prey (introduced and native) were offered simultaneously in equal amounts to individual predator for this have the opportunity to choose between them. The individual preys were measured before experiment. Prey sizes varied between 2 and 5 cm, and predators varied between 15 and 20 cm. From five to seven replications were made for each treatment level (different prey densities) of the experiment.

Table 1. Levels of treatment with the prey number placed in each of them.

| Treatment Level | Number of introduced prey | Number of native prey | Total prey | Replicates |
|--------------------|---------------------------------|-----------------------|---------------|------------|
| 1 | 1 | 1 | 2 | 6 |
| 2 | 2 | 2 | 4 | 7 |
| 3 | 3 | 3 | 6 | 7 |
| 4 | 4 | 4 | 8 | 6 |
| 5 | 5 | 5 | 10 | 6 |
| 6 | 6 | 6 | 12 | 6 |
| 7 | 7 | 7 | 14 | 6 |
| 8 | 8 | 8 | 16 | 6 |
| 9 | 9 | 9 | 18 | 6 |
| 10 | 10 | 10 | 20 | 5 |
| 11 | 13 | 13 | 26 | 5 |
| 15 | 15 | 15 | 30 | 5 |
| 16 | 16 | 16 | 32 | 5 |

Experiments were conducted from May to December 2018 and lasted for 48 hours. After that, the amount of remaining prey was counted. Each individual predator was used in the different treatment levels. Levels applied for each individual were randomly chosen, and predators stayed in a recess period of at least three days with diet of tenebrios and two of fasting before the new essay. At the end of experiments, predators and remaining prey were euthanized using Benzocaine 80 mg / L or Eugenol 70-90 % following the protocols of CONCEA and the (AVMA, 2013) and discarded according to the procedures of the 'Divisão de Gestão Ambiental' (DGA) of 'Universidade Federal do Paraná'. The use of animals in the experiments and the procedures performed were authorized by the certificates N° 1027 and 1199 of CEUA/BIO-UFPR.

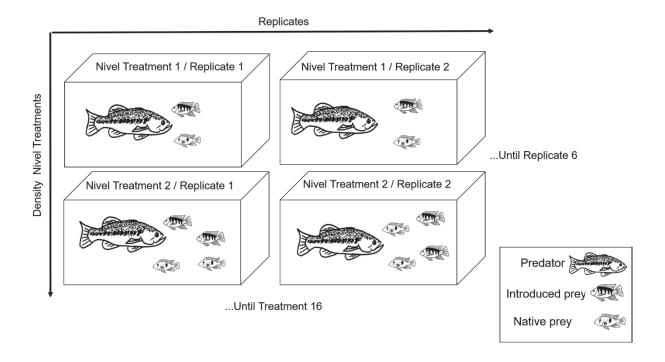


Figure. 3 Experimental design. The two types of prey were presented simultaneously at different densities to a predator for 48 hours.

The statistical analysis was performed in the R software (R Core Team, 2018) Version 3.4.3. Food preference for native or introduced prey was analyzed with a paired t-test, given that both preys were available for predators in the same experimental unit. For that, we did not consider the treatment level, given that the goal was to identify a possible preference across all treatment levels. For the functional response analysis, we use the Frair package (Pritchard, 2017). This package uses the function 'frair test' to determine the type of functional response through a logistic regression of the proportion of prey consumed by the initial density of prey (Trexler, et al., 1988; Pritchard, et al., 2017). This function tests for evidence for type II and III response (Don't test for type I because it only applies for unlimited resources). The result was type II (see results for more details), for that is applicable both the original Hollling's equation, and the equation modified by Rogers. In the original Hollling's equations for type II the prey's density remained constant throughout the experiment so Rogers in 1972 proposed an adjusted model for experiments with prey depletion. This model is more suited to our experimental design. So, we made the fit to Rogers equation using a non-linear regression adjusted for maximum likelihood estimation, of number of prey consumed versus density with the help of the function 'frair fit' of frair package (Juliano, 2001; Dick, et al., 2013; Pritchard, et al., 2017). This procedure also allows to find the parameters of type II functional response which are the following I) a: attack constant also called instantaneous rate of search II) h: Handling time, understood as the time that the predator spends in the search, capture, kill and digest a prey and that therefore it cannot spend in attacking another prey. III) Maximum feeding rate, determined by the handling time and calculated later as (1/hT) where T is the experimental period (Juliano, 2001; Dick, et al., 2013; Pritchard, et al., 2017).

4 RESULTS

In the paired t-test, taking into account each of the treatments levels of density, it was found that the M. salmoides show preference for introduced prey (t = 4.24, df = 75, p < 0.001) As a first approach, in the Figure 4 is visible that preference for the introduced prey occurs mainly in high densities of prey, this is shown more clearly in the functional response results

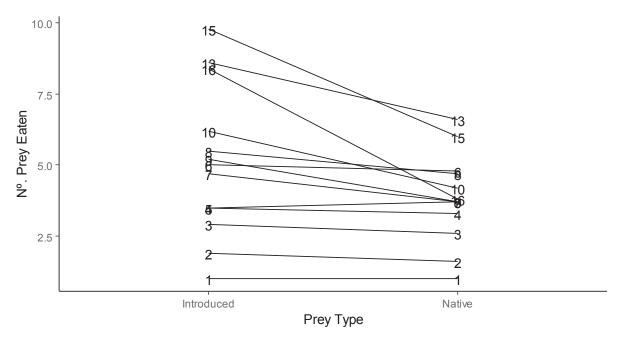


Figure. 4 Graphic of paired t-test. Difference of the average consumption of introduced (*O. niloticus* and *C rendalli*) and native (*G. brasiliensis*) prey taking into account the different levels of density treatment (numbers).

The results of the functional response parameters are found in Table 2, the negative linear coefficient of the logistic regression indicates evidence for type II functional

response. There is no difference in the attack rate for the two types of prey, but if in the handling time, which is greater for the introduced prey.

Table 2. Functional response results

| | Introduced Prey | Native Prey | Difference |
|------|-----------------|------------------------|-------------------|
| Lc | Density = -0.10 | Density = -0.17 | |
| а | 0.047 ± 0.008 | 0.068 ± 0.01 | Da = 0.021 ± 0.01 |
| h | 3.71 ± 0.66 hrs | 8.11 ± 0.79 hrs | Dh = 4.4 ± 1.03 |
| 1/hT | 0.0056 | 0.0026 | |

Lc: Linear coefficient, negative indicates type II functional response; a: Attack rate, h: handling time, 1/hT: Maximum feeding rate, the values with p > 0.001 are in bold.

The functional response equation for *M. salmoides* for native prey is:

$$N_e = N_o (1 - \exp(0.068(8.11N_e - 48)))$$

And for introduced prey is:

$$N_e = N_o (1 - \exp(0.047N_0^q (3.71N_e - 48)))$$

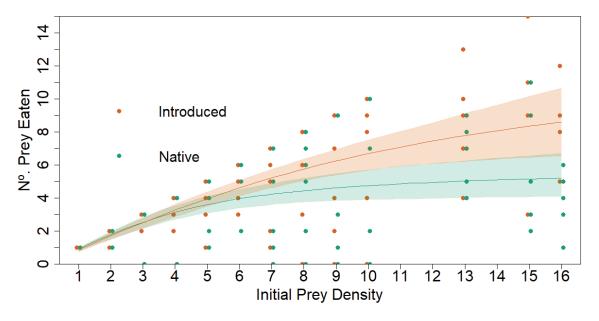


Figure. 5 Functional response of *M. salmoides* for introduced (*O. niloticus* and *C. rendalli*) and native (*G. brasiliensis*) prey. In both cases, the functional response is type II.

5 DISCUSSION

We found that *M. salmoides* showed a feeding preference especially when prey populations are found in high densities. With a low food availability, their behavior is clearly opportunistic. Therefore, we partially supported our hypothesis that behavior of predator fish would be generalist opportunist. The shape of the functional response curve has effects on the predator-prey dynamics (Essington, et al., 2000). The type II functional responses for both types of prey, agrees with that found by Alexander, et al. (2014) in a study with *M. salmoides* as predator of tadpoles (*Hyperolius marmoratus* (Rapp, 1842)). In addition, type II is the most common type of functional response in vertebrates (Juliano, 2001). This type of functional response indicates that the consumption increases with density, but the proportion is decreased due to the manipulation time, generating an inverse dense-dependent relationship, making that in high densities the preys are less likely to be attacked than at low densities (Holling, 1959; Fernández & Corley, 2004). Allowing the increase of the size of the population, since does not exercise controls over it.

The feeding preference evaluated by paired t-test, describes the final result of the consumption, that is to say, the introduced preys are more consumed, however, the fit of the functional response allows to elucidate with clarity why this occurs (Fig. 5). The interpretation can be described as follow. First, we generated evidence that the preference exists mostly at high prey densities. Secondly, the results of the estimation of the parameters allow us to see why this happens, since the attack rates for the two types of prey do not differ. However, the time of manipulation is greater in the native prey, which means that at the end of the experimental time, the predator has consumed more tilapias, because it can consume them more quickly. The handling time for native prey is double that for introduced prey, however, these parameters are not independent, since the experiments were carried out with the two types of prey simultaneously. Theoretically, while one predator consumes one type of prey, it cannot consume another type of prey (Martin, et al., 2010). For this reason, as a complement to this study it would be interesting to carry out another one independently with each one of the types of prey.

The preference for introduced prey is reasonable considering the longer handling time for native prey. As this increases the cost of the prey making it less profitable in the cost/benefit ratio plated by the optimal foraging theory (Pyke,1984). This could suggest that the presence of *O. niloticus* and *C. rendalli* in the reservoirs of the Metropolitan Region of Curitiba has a positive impact on the populations of *M. salmoides*, probably allowing it to reach population sizes that would not be achieved only with the native species, due to the time required for the consumption of these, and added to the large number of tilapia present in the reservoirs (Frehse, 2018) The benefit obtained by *M. salmoides* from these other introduced species could be evidence of an invasional meltdown defined as process by which a non-indigenous species or group of species facilitates the invasion of others, increasing the likelihood of survival and/or of ecological impact (Simberloff & Von-Holle, 1999). Considering our results, we can speculate that *M. salmoides* fitness may be favored by the non-native prey, in relation to the most abundant native prey.

As in any experiment, we must also note some limitations. We did isolate a set of factors, justified by the fact that the prey and the predators are the most abundant (Frehse, 2018) in the reservoir studied. However, 'real life' is surely more complex. We

listed below some issues that in real life may be affecting the dynamics between these three introduced species and the native. I) Life stage of each species: Here we evaluate only one stage of life for each species (juvenile predators, and fry prey). However, O. niloticus and C rendalli are also omnivorous fish that in adult stage can act as egg and small fish predators (Abdelghany, 1993), being able to act as predators of *M. salmoides* and *G. brasiliensis*. Thus, a better understanding of the relationships that arose from the introduction of these species would be reached considering different life stages of prey and predators. II) Existence of other interactions between these species: predation may not be the only interaction considering the generalist behavior of species. For example, they may be competing for food, shelter and other resources (Martin, et al., 2010). III) Presence of other species, both native and introduced, even with lower densities, implies the existence of another set of relationships that can be both negative and positive for the species studied here. In this case, can other species exert pressure for competition or predation, even with relatively lower abundances (Smout, et al., 2010). IV) As is well-known, the control of populations within a predator-prey relationship does not occur unidirectionally, variation in prey populations also affect the predator population through the numerical response. V) presence of other environmental and anthropogenic factors not evaluated that can affect biotic interactions.

The present study contributed to the understanding of the trophic relationships between native and introduced species. In summary, we found that invasive predator benefit from invasive prey given that predation efficiency (mainly due to handling time) is greater than considering the most common native prey. Also, the invasive prey used is known to reach high densities in reservoirs, probably higher than the native counterpart. Therefore, tilapias may likely enhance invasive success of the *M. salmoides* compared to the native prey *G. brasiliensis* even though both prey are cichlids with similar trait values (Yamamoto, 2011).

6 CONCLUSIONS

We conclude that *M. salmoides*, due to presenting a type II functional response, generates in the populations an inverse dense-dependent mortality that does not exert

control over the populations, it was also shown that the greater consumption of introduced prey is probably the result of a greater handling time required to consume native prey.

Considering our results, we emphasize that the relationships of each introduced species with the native species, as well as between them must be evaluated in considering its likely nature: synergistic, antagonistic or both. We conclude by reinforcing that monitoring invasive species plus basic research on the relationships between species is central for environmental management decisions, to avoid further alteration of trophic dynamics since native species generally turn out to be most affected.

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SUPPLEMENT 1- AUTHORIZATIONS

The authorizations for the collection of zoological material and experimentation with animals are presented below. The specimen's collection was done under permanent license for collection zoological material SISBIO N° 24779 and the experiments were carried out with the organisms authorized by the certificate N° 1199 issued by the *Comissão de ética no uso de animais do Setor de Ciencias biológicas, Universidade Federal do Paraná (CEUA/BIO-UFPR)* and through the procedures authorized in the certificate N° 1027 of the same commission.



Ministério do Meio Ambiente - MMA Instituto Chico Mendes de Conservação da Biodiversidade - ICMBio Sistema de Autorização e Informação em Biodiversidade - SISBIO

Licença permanente para coleta de material zoológico

| Número: 24779-1 | Data da Emissão: 04/11/2010 14:48 | | |
|--|-----------------------------------|--|--|
| Dados do titular | | | |
| Nome: Jean Ricardo Simões Vitule | CPF: 186.514.948-90 | | |
| Nome da Instituição : UNIVERSIDADE FEDERAL DO PARANÁ | CNPJ: 75.095.679/0001-49 | | |

Observações e ressalvas

- As atividades de campo exercidas por pessoa natural ou jurídica estrangeira, em todo o território nacional, que impliquem o deslocamento de recursos humanos e materiais, tendo por objeto coletar dados, materiais, espécimes biológicos e minerais, peças integrantes da cultura nativa e cultura popular, presente e passa da, obtidos por meio de recursos e técnicas que se destinem ao estudo, à difusão ou à pesquisa, estão sujeitas a autorização do Ministério de Ciência e Tecnologia.

 A licença permanente não é válida para: a) coleta ou transporte de espécies que constem nas listas oficiais de espécies ameaçadas de extinção; b) manutenção de
- espécimes de fauna silvestre em cativeiro; c) recebimento ou envio de material biológico ao exterior; e d) realização de pesquisa em unidade de conservação federal ou em caverna. A restrição prevista no item d não se aplica às categorias Reserva Particular do Patrimônio Natural, Área de Relevante Interesse Ecológico e Área de Proteção Ambiental constituídas por terras privadas.
- O pesquisador titular da licença permanente, quando acompanhado, deverá registrar a expedição de campo no Sisbio e informar o nome e CPF dos membros da sua equipe, bem como dados da expedição, que constarão no comprovante de registro de expedição para eventual apresentação à fiscalização;
- Esta licença permanente NÃO exime o pesquisador titular da necessidade de obter as anuências previstas em outros instrumentos legais, bem como do consentimento do responsável pela área, pública ou privada, onde será realizada a atividade.
- Esta licença permanente não poderá ser utilizada para fins comerciais, industriais ou esportivos ou para realização de atividades integrantes do processo de licenciamento ambiental de empreendimentos.
- Este documento NÃO exime o pesquisador titular da necessidade de atender ao disposto na Instrução Normativa Ibama nº 27/2002, que regulamenta o Sistema Nacional de Anilhamento de Aves Silvestres.
- 7 O pesquisador titular da licença permanente será responsável pelos atos dos membros da equipe (quando for o caso)
- O órgão gestor de unidade de conservação estadual, distrital ou municipal poderá, a despeito da licença permanente e das autorizações concedidas pelo ICMBio, estabelecer outras condições para a realização de pesquisa nessas unidades de conservação.
- O titular de licença ou autorização e os membros da sua equipe deverão optar por métodos de coleta e instrumentos de captura direcionados, sempre que possível, ao grupo taxonômico de interesse, evitando a morte ou dano significativo a outros grupos; e empregar esforço de coleta ou captura que não comprometa a viabilidade de populações do grupo taxonômico de interesse em condição in situ.
- O titular da licença permanente deverá apresentar, anualmente, relatório de atividades a ser enviado por meio do Sisbio no prazo de até 30 dias após o aniversário de emissão da licença permanente.
- O titular de autorização ou de licença permanente, assim como os membros de sua equipe, quando da violação da legislação vigente, ou quando da inadequação, omissão ou falsa descrição de informações relevantes que subsidiaram a expedição do ato, poderá, mediante decisão motivada, ter a autorização ou licença suspensa ou revogada pelo ICMBio e o material biológico coletado apreendido nos termos da legislação brasileira em vigor.
- A licença permanente será válida enquanto durar o vínculo empregatício do pesquisador com a instituição científica a qual ele estava vinculado por ocasião da solicitação.
- Este documento não dispensa o cumprimento da legislação que dispõe sobre acesso a componente do patrimônio genético existente no território nacional, na plataforma continental e na zona econômica exclusiva, ou ao conhecimento tradicional associado ao patrimônio genético, para fins de pesquisa científica, bioprospecção e desenvolvimento tecnológico.
- As atividades contempladas nesta autorização NÃO abrangem espécies brasileiras constante de listas oficiais (de abrangência nacional, estadual ou municipal) de espécies ameaçadas de extinção, sobreexplotadas ou ameaçadas de sobreexplotação.

Táxons autorizados

| # | Nível taxonômico | Táxon(s) |
|---|------------------|--------------------------------|
| 1 | CLASSE | Elasmobranchii, Actinopterygii |
| 2 | | |

Destino do material biológico coletado

| # | Nome local destino | Tipo Destino |
|---|--|--------------|
| 1 | Zoológico de Curitiba e Museu de História Natural - SMMA/PMC | coleção |

Este documento (Licença permanente para coleta de material zoológico) foi expedido com base na Instrução Normativa nº154/2007. Através do código de autenticação abaixo, qualquer cidadão poderá verificar a autenticidade ou regularidade deste documento, por meio da página do Sisbio/ICMBio na Internet (www.icmbio.gov.br/sisbio).

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Sistema de Autorização e Informação em Biodiversidade - SISBIO

Licença permanente para coleta de material zoológico

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| Nome da Instituição : UNIVERSIDADE FEDERAL DO PARANÁ | | | CNPJ: 75.095.679/0001-49 | |
| | | | | |

Registro de coleta imprevista de material biológico

De acordo com a Instrução Normativa nº154/2007, a coleta imprevista de material biológico ou de substrato não contemplado na autorização ou na licença permanente deverá ser anotada na mesma, em campo específico, por ocasião da coleta, devendo esta coleta imprevista ser comunicada por meio do relatório de atividades. O transporte do material biológico ou do substrato deverá ser acompanhado da autorização ou da licença permanente com a devida anotação. O material biológico coletado de forma imprevista, deverá ser destinado à instituição científica e, depositado, preferencialmente, em coleção biológica científica registrada no Cadastro Nacional de Coleções Biológicas (CCBIO).

| Táxon* | Qtde. | Tipo de amostra | Qtde. | Data |
|--------|-------|-----------------|-------|------|
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^{*} Identificar o espécime no nível taxonômico possível.



Ministério da Educação UNIVERSIDADE FEDERAL DO PARANÁ Setor de Ciências Biológicas Comissão de Ética no Uso de Animais (CEUA)



N° 1199

CERTIFICADO

A Comissão de Ética no Uso de Animais do Setor de Ciências Biológicas da Universidade Federal do Paraná (CEUA/BIO – UFPR), instituída pela Resolução Nº 86/11 do Conselho de Ensino Pesquisa e Extensão (CEPE), de 22 de dezembro de 2011, **CERTIFICA** que os procedimentos utilizando animais no projeto de pesquisa abaixo especificado estão de acordo com a Diretriz Brasileira para o Cuidado e a Utilização de Animais para fins Científicos e Didáticos (DBCA) estabelecidas pelo Conselho Nacional de Controle de Experimentação Animal (CONCEA) e com as normas internacionais para a experimentação animal.

STATEMENT

The Ethics Committee for Animal Use from the Biological Sciences Section of the Federal University of Paraná (CEUA/BIO – UFPR), established by the Resolution Nº 86/11 of the Teaching Research and Extension Council (CEPE) on December 22nd 2011, **CERTIFIES** that the procedures using animals in the research project specified below are in agreement with the Brazilian Guidelines for Care and Use of Animals for Scientific and Teaching purposes established by the National Council for Control of Animal Experimentation (CONCEA) and with the international guidelines for animal experimentation.

PROCESSO/PROCESS: 23075.034420/2018-59

APROVADO/APPROVAL: 31/07/2018 – R.O. 06/2018

TÍTULO: Olhando através dos olhos do predador: O outro lado da "Ingenuidade Ecológica".

TITLE: Looking through the predators' eyes: The other side of the "predator naïveté" theory.

AUTORES/AUTHORS: Jean Ricardo Simões Vitule, Larissa Strictar Pereira.

DEPARTAMENTO/DEPARTMENT: Engenharia Ambiental

Profa. Dra. Katya Naliwaiko Coordenadora da CEUA

ANEXO



Ministério da Educação UNIVERSIDADE FEDERAL DO PARANÁ Setor de Ciências Biológicas Comissão de Ética no Uso de Animais (CEUA)



N° 1027

CERTIFICADO

A Comissão de Ética no Uso de Animais do Setor de Ciências Biológicas da Universidade Federal do Paraná (CEUA/BIO – UFPR), instituída pela Resolução № 86/11 do Conselho de Ensino Pesquisa e Extensão (CEPE), de 22 de dezembro de 2011, CERTIFICA que os procedimentos utilizando animais no projeto de pesquisa abaixo especificado estão de acordo com a Diretriz Brasileira para o Cuidado e a Utilização de Animais para fins Científicos e Didáticos (DBCA) estabelecidas pelo Conselho Nacional de Controle de Experimentação Animal (CONCEA) e com as normas internacionais para a experimentação animal.

STATEMENT

The Ethics Committee for Animal Use from the Biological Sciences Section of the Federal University of Paraná (CEUA/BIO - UFPR), established by the Resolution № 86/11 of the Teaching Research and Extension Council (CEPE) on December 22nd 2011, CERTIFIES that the procedures using animals in the research project specified below are in agreement with the Brazilian Guidelines for Care and Use of Animals for Scientific and Teaching purposes established by the National Council for Control of Animal Experimentation (CONCEA) and with the international guidelines for animal experimentation.

PROCESSO/PROCESS: 23075.160195/2016-43

APROVADO/APPROVAL: 18/10/2016 - R.O. 09/2016

TÍTULO: Resposta funcional de peixes nativos do rio Guaraguaçu - PR considerando-se a estruturação de habitat causada por uma gramínea invasora

TITLE: Functional response of native and nonnative fishes of Guaraguaçu river – PR considering the habitat structuration promoted by an invasive grass

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DEPARTAMENTO/DEPARTMENT: Engenharia Ambiental

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