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TEMPORAL EFFECT ON PREDATION OF ASTYANAX LACUSTRIS AND POECILIA RETICULATA FINGERLINGS BY ODONATA NYMPHS IN AN EXPERIMENTAL ENVIRONMENT

EFEITO TEMPORAL NA PREDACÃO DE ALEVINOS DE ASTYANAX LACUSTRIS E POECILIA RETICULATA POR NINFAS DE ODONATA EM AMBIENTE EXPERIMENTAL

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Abstract: The study evaluated the predatory impact of nymphs from the order Odonata on two fish fry species: *Astyanax lacustris* (native) and *Poecilia reticulata* (invasive), both commonly found in aquaculture systems. The experiment was conducted over 72 hours, with fry counted and replaced every 12 hours, testing three treatments: only *A. lacustris*, only *P. reticulata*, and both species together. Each aquarium contained one Odonata nymph that had fasted for 24 hours, under constant temperature (26 °C) and continuous lighting. Statistical analyses (Kruskal-Wallis and Dunn's test) revealed no significant differences in predation between the two species, suggesting a lack of selectivity. However, significant differences were observed over time, with higher predation rates during the first 12 hours, possibly due to predator satiation after initial consumption. These findings indicate that Odonata predation is not species-specific but is influenced by temporal feeding dynamics. Therefore, management strategies in aquaculture should consider the timing of predator feeding behavior to minimize economic losses due to fry mortality, particularly in the initial hours after stocking, when fry is more vulnerable to predatory nymphs.

Keywords: Aeshnidae, fingerlings, Libellulidae, pisciculture, predation rate.

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Resumo: O estudo avaliou o impacto predatório de ninfas da ordem Odonata sobre duas espécies de alevinos: *Astyanax lacustris* (nativa) e *Poecilia reticulata* (invasora), comuns em sistemas de piscicultura. O experimento foi conduzido por 72 horas, com contagens e reposições de alevinos a cada 12 horas, testando três tratamentos: apenas *A. lacustris*, apenas *P. reticulata* e ambas as espécies juntas. Cada aquário continha uma ninfa de Odonata em jejum, com temperatura constante (26 °C) e iluminação contínua. As análises estatísticas (Kruskal-Wallis e Dunn) não indicaram diferenças significativas na predação entre as espécies, sugerindo ausência de seletividade. Contudo, observou-se variação significativa ao longo do tempo, com taxas de predação mais altas nas primeiras 12 horas, possivelmente devido à saciedade das ninfas após o consumo inicial. Esses achados indicam que a predação por Odonata não depende da espécie de alevino, mas é influenciada pela dinâmica temporal. Assim, estratégias de manejo em piscicultura devem considerar o comportamento alimentar desses predadores ao longo do tempo para minimizar perdas econômicas associadas à mortalidade de alevinos, especialmente nas primeiras horas após a introdução em tanques, quando estão mais vulneráveis ao ataque de ninfas predadoras.

Palavras-chave: Aeshnidae, alevinos, Libellulidae, piscicultura, taxa de predação.



INTRODUCTION

The order Odonata represents a group of extreme ecological relevance, acting as key predators in aquatic ecosystems during most of their life cycle, contributing to population control of aquatic and terrestrial organisms (Bock; Padovani, 2000, De Faria et al., 2001). In fish farming systems, Odonata nymphs are important predators of fish larvae, resulting in significant economic losses (Fortunato et al., 2020). To mitigate these losses, fish farmers often resort to strategies such as chemical control and inappropriate management practices. However, these methods can result in negative environmental impacts, including water pollution and ecosystem destabilization (Fortunato et al., 2020).

Odonata has a hemimetabolous development, with aquatic nymphs and aerial-aquatic adults widely distributed depending on climatic conditions and environmental quality (Corbet, 1980, Corbet, 1999; Braune et al., 2008). During the nymph stage, predatory behavior is based on ambush strategies and projection of the modified lip onto the prey to capture it (Corbet 1980, Soares et al., 2001). The predation rate of nymphs is modulated by factors such as substrate type, prey availability, environmental conditions and developmental stage (Corbet, 1980, Corbet 1999, Soares et al. 2001, Lacerda et al., 2011). The suborder Anisoptera is characterized by larger body size, higher predation rate and greater ability to adapt to artificial environments such as fishponds (Santos et al., 1988, Corbet, 1999, Juen et al., 2014). In addition, the larval development time of Anisoptera is faster compared to other suborders, which maximizes their impact on production systems (Moura et al., 2020).

Predation by Odonata nymphs is one of the main challenges for fish farming, especially in countries with tropical climate such as Brazil, where favorable climatic conditions and the great availability of water resources have driven the growth of the sector (De Faria et al., 2001, Fortunato et al., 2020). In this scenario, the economic interest associated with fish production often outweighs concerns about environmental risks, leading to the introduction of exotic species as a control measure. However, this practice has been widely criticized for its negative impacts on native biodiversity and aquatic ecosystem integrity (Ziller et al., 2007, Leão et al., 2011).



Despite the economic and ecological relevance of the subject, there is a lack of experimental data relating the predatory behavior of Odonata nymphs to their food preferences and consumption rates at different time intervals, especially in the context of fish farming. Given this gap, the aim of this study was to compare the predation rates of Lambari, *Astyanax lacustris* (Lütken, 1875), and Guppies, *Poecilia reticulata* Peters, 1859, by Odonata nymphs over a 12-hour period, assessing the existence of specific food preferences and possible variations in consumption rates as a function of time. The results obtained should support the development of more effective management strategies capable of minimizing economic losses in fish farming systems, as well as increasing the understanding of the ecological impact of Odonata nymphs in natural and artificial environments. In this way, the study will contribute to the integration of sustainable production practices and the conservation of aquatic biodiversity.

Additionally, understanding the interplay between native and invasive species under pressure contributes to broader ecological insights. Invasive species often exhibit traits such as rapid reproduction or behavioral plasticity, yet these traits may also increase their susceptibility to native predators. Odonata nymphs, being generalist predators with high adaptability, serve as a natural test case for evaluating this hypothesis. This work, therefore, offers an experimental contribution to invasion biology by assessing the potential of native predators to suppress invasive prey populations without human intervention.

MATERIAL AND METHODS

Sampling and Experiments

Odonata immatures were sampled in a temporary lagoon Mayaca (21° 57' 59"S, 47° 53' 00' W) located in a cerrado fragment in the grounds of the Federal University of São Carlos (UFSCar). This lagoon has marked seasonal characteristics, drying up completely during periods of drought. At the time of collection, the lagoon was in a flood. The region's climate is classified tropical with wet summers and dry winters (Cwa), according to the Köppen classifications (Rolim et al., 2007).



The Odonata nymphs were collected using a net, selecting only Anisoptera, which are the most common in fishponds (De Marco Junior; Resende, 2004, Fortunato et al., 2021). Immediately after collection, each immature fish was placed in an individual bottle containing water from the pond itself, to prevent cannibalism and guarantee the integrity of the specimens until the start of the experiment.

The fingerlings used in the experiments were of the species *A. lacustris* and *P. reticulata*, donated by the National Center for Research and Conservation of Continental Fish (CEPTA), located in Pirassununga, SP. These species were chosen because they were available in quantities and sizes suitable for the experiment. All juveniles were less than two centimeters in length and were kept in aquariums with adequate oxygenation under controlled conditions until they were used in the treatments.

The experiments were carried out in the Plankton Laboratory of the Hydrobiology Department of the Federal University of São Carlos. Four replicates were used for each treatment, consisting of aquariums with a volume of two liters of water and an Odonata nymph, which was fasted 24 hours before the start of the experiments. After this period, 10 *A. lacustris* fingerlings were introduced into each of the four aquariums. The same procedure was followed in a second set of aquariums to which 10 *P. reticulata* were added. A third set of aquariums was stocked with five *A. lacustris* and *P. reticulata*. In addition, a control group was maintained that contained only one Odonata nymph, without the presence of juveniles.

The experiment lasted 72 hours in total, and every 12 hours we counted how many and which larvae were preyed upon. During the observations, the behavior of the nymphs and the fingerlings was recorded in order to identify patterns of predation and interactions between the species. The ambient temperature was maintained at 26°C and the lighting was controlled to remain constant throughout the experiment.

At the end of the experiment, Odonata nymphs were sacrificed and preserved in 70% ethanol. Identification was performed to the genus level using the specialized literature (Carvalho; Pessacq, 2018, Neiss et al., 2018).

Data Analysis



Statistical analyses were performed using R version 4.4.0. Initially, the *Shapiro-Wilk* test was applied to assess the normality of the predation variable, followed by *Levene's* test for homogeneity of variances. Due to the lack of normality of the data and the lack of homogeneity of the variances, we opted for the non-parametric Kruskal-Wallis's test, complemented by the *Dunn* test for multiple comparisons. A Pearson correlation test was performed to examine the relationship between nymph size and predation rate. The significance level was set at $p < 0.05$.

RESULTS

The Kruskal-Wallis's test showed no significant difference between treatments in terms of the number of larvae consumed ($X^2 = 6.75$; $p > 0.05$). The Kruskal-Wallis's test showed a significant difference between the variables in relation to time and the number of fingerlings consumed ($X^2 = 12.57$; $p < 0.05$).

Dunn's test showed a significant difference in the predation of Odonata on *Poecilia reticulata* (invasive) between the times: 12/48 hours ($p = 0.0447$), 12/60 hours ($p = 0.0149$) and 12/72 hours ($p = 0.0149$). This shows a higher consumption rate in the first hours of measurement (Figure 1).

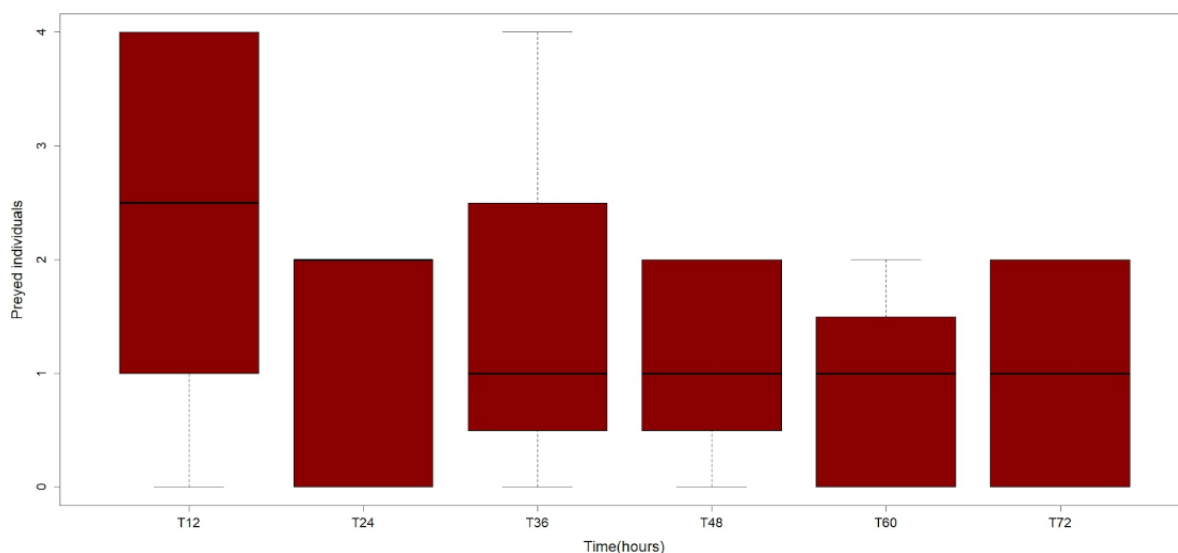


Figure 1. Dunn's test revealed a significant difference in Odonata predation on *Poecilia reticulata* (invasive) across time intervals: 12 vs. 48 hours ($p = 0.0447$), 12 vs. 60 hours ($p = 0.0149$), and 12 vs. 72 hours ($p = 0.0149$).



These results indicate a higher consumption rate during the initial hours of observation. These findings suggest that predation may follow a Type II functional response, where predator consumption increases rapidly at low prey densities and then plateaus as predators become satiated. In aquaculture environments, this could imply that short-term exposure to Odonata nymphs poses a greater risk to newly introduced fingerlings than prolonged exposure. Recognizing this temporal pattern can support better management strategies, such as the timing of fingerling introduction or temporary predator exclusion during the most vulnerable early hours. The nymphs utilized in the study were identified as *Anax* (53.83%) and *Tramea* (46.15%), representing the Aeshnidae and Libellulidae families, respectively. The findings revealed that the size of the nymphs did not affect the predation rate ($r = 0.42$, $p > 0.05$).

During the experiment, the nymphs remained at the bottom of the aquarium, with no movement along the water column, in contrast to *A. lacustris* and *P. reticulata*, which remained in motion.

DISCUSSION

The impact of Odonata nymphs on fingerlings in fish farming systems is well documented, yet there is a scarcity of research investigating the predatory behavior of nymphs, their food preferences, consumption rates, and temporal patterns (Delgado et al., 1995). The present study sought to address this knowledge gap by assessing the predation rates of nymphs on lambari (*A. lacustris*) and guppies (*P. reticulata*) fingerlings in a controlled environment, highlighting on predation over time. The results demonstrated that exposure time significantly influenced predation, with a higher consumption of fingerlings occurring during the initial hours of the experiment. These findings provide novel insights into the predation dynamics of Odonata in fishponds, providing knowledge for the development of more effective management strategies to mitigate the economic impacts caused by nymphs.

The lack of significant differences between treatments in the number of juveniles predated suggests that Odonata nymphs exhibited consistent predatory behavior regardless of juvenile species. This result may be related to the similarity of experimental



conditions such as prey availability, juvenile size, and the absence of complex environmental variables such as the presence of substrate such as vegetation.

In addition, it is well known that Odonata nymphs are generalists (Corbet, 1999), but this study showed that there was a greater preference for predation over time by *P. reticulata*, a non-native species that requires further study for reasons that have not yet been identified. Odonata nymphs are known to exploit different types of prey by adapting to variations in their availability and behavior (Pritchard, 1965, Londoño; Sánchez, 2022). However, the lack of behavioral traits of prey, such as coloration, size or agitation, may have maintained the difficulty of capture, resulting in the lack of significant differences between treatments (Soares et al., 2001).

The results of the experiment demonstrated a higher predation rate during the initial 12 hours, followed by a gradual reduction as the experiment progressed to 24 hours and an equalization after 36 hours. A notable difference emerged in the number of fingerlings preyed upon, with a marked increase observed during the initial 12-hour period, followed by a subsequent decline in the last three intervals (48, 60, and 72 hours). This finding suggests that the predatory activity of the nymphs is more pronounced at the onset of interaction with the prey. This phenomenon may be attributed to larval satiety, where after an initial period of heightened activity, consumption gradually decreases as energy demands are met. Another possible explanation is related to predator learning and environmental acclimatization. As time progresses, nymphs may either adapt to the simplified environment, reduce exploratory behavior, or experience a decline in prey encounter rates due to subtle behavioral changes in the fingerlings. Such feedback mechanisms between predator and prey dynamics are well recognized in behavioral ecology and warrant future experimental exploration under varied structural complexities.

The temporal dynamics of predation may also be influenced by factors such as digestion time, low oxygenation, and the type of prey (fingerlings) (Pritchard, 1965). Furthermore, the fingerlings behavioral response to the predator over time may involve an initial less efficient avoidance strategy, which could be adapted over time to facilitate more efficient escape.



The substantial variations observed in Dunn's test for the predation of *P. reticulata* (an invasive species) during the initial hours of the experiment can be attributed to two primary factors: Firstly, the prey's initial vulnerability to predation is often marked by its inability to promptly detect the presence of a predator, leaving it exposed to potential attacks. Secondly, Odonata nymphs display a distinct behavioral pattern: they exhibit a surge in activity and aggressive feeding behavior upon initial contact with prey, driven by the immediate need to meet their energy requirements. However, as their energy demands are satisfied, their activity levels gradually diminish (Paradis et al., 1996). These dynamic highlights the interplay between prey susceptibility and predator feeding strategies in shaping ecological interactions. Furthermore, *P. reticulata* may exhibit reduced evasive behavior or ineffective defense strategies compared to native species, rendering it more vulnerable to predation during the initial hours (Jeffries 1988).

These findings highlight the potential of Odonata nymphs as biological control agents for invasive species, such as *P. reticulata*, offering a sustainable alternative for their management. By regulating invasive species populations in a natural manner, Odonata nymphs can contribute to the restoration and maintenance of ecological balance in aquatic ecosystems (May, 2019). However, the implementation of this strategy necessitates further research to assess the effectiveness of nymphs on a large scale, as well as their potential impacts on non-target species and the dynamics of aquatic communities.

During the course of the experiment, it was observed that the Odonata nymphs remained predominantly at the bottom of the aquarium, while the fingerlings of both species studied were distributed more widely in the water column. This behavior may have influenced predation efficiency, given that the spatial distribution of prey and predator directly affects the interactions between them (Fulan; Anjos, 2015). The aggregation of nymphs at the base of the aquarium may have constrained their access to the fingerlings that were moving at higher levels, thereby reducing the opportunities for capture.

In natural habitats, the presence of aquatic vegetation, varied substrates, and light gradients can significantly alter predation dynamics (Corbet, 1999, Lacerda et al., 2010).



The absence of these elements in the experimental environment may have simplified interactions, resulting in more predictable and less variable behavior than that observed in natural ecosystems. Furthermore, the inclusion of different families or genera of fingerlings could reveal significant variations in predation rates, thereby broadening our understanding of the ecological dynamics between predators and prey (De Faria et al., 2001, Soares et al. 2001). Finally, reducing the intervals between observations and increasing the total duration of the experiment are necessary measures to improve the precision and comprehensiveness of the results.

The interaction between physical factors such as temperature, humidity, luminosity, and wind has been demonstrated to interfere with both the availability of prey and the predatory behavior of Odonata (Corbet, 1999, Woodward; Hildrew, 2002). The predatory behavior of Odonata is closely related to their highly developed vision, which allows them to locate and capture prey by detecting movement, light, and specific wavelengths (Pritchard, 1965, Corbet, 1999, Soares et al., 2001, Londoño; Sánchez, 2022).

In the context of fish farming, it is imperative to investigate behavioral changes that can reduce predation by Odonata nymphs, particularly given the paucity of data on the subject. The quality of the environment, for instance, can influence the release of chemical substances by the fingerlings, which can inhibit or stimulate predatory behavior (Carreau; Pyle, 2005). Consequently, the implementation of effective monitoring techniques to assess the population dynamics of Odonata nymphs within fishponds, coupled with the adoption of sustainable management strategies to mitigate their ecological impacts, is imperative to ensure the economic viability of these aquaculture enterprises (Fortunato et al., 2020).

Moreover, integrated pest management (IPM) approaches in aquaculture should consider Odonata as part of a broader trophic network rather than isolated threats. While the short-term impact on fingerlings can be significant, these predators also play a crucial role in regulating other nuisance invertebrates and contributing to water quality by limiting decomposer overpopulation. Future studies might explore how Odonata predation interacts with nutrient cycling and overall pond health.



It can be concluded that Odonata nymphs exhibited a higher predation rate during the initial 12 hours of contact with fingerlings, followed by a gradual reduction and stabilization after 36 hours. This pattern suggests that the predatory activity of the nymphs is more intense at the onset of contact with the prey, possibly due to the nymph's attaining satiation after an initial period of high activity. Furthermore, the results indicated that Odonata nymphs exhibited no significant preference for any specific species of fingerlings (*A. lacustris* or *P. reticulata*), suggesting a well-documented generalist predatory behavior (Corbet, 1999). A notable finding was the heightened vulnerability of the invasive species *P. reticulata* during the initial hours of the experiment, suggesting the potential for Odonata nymphs to serve as biological control agents in aquatic ecosystems. However, the efficacy of this strategy on a broader scale and its potential impact on non-target species necessitates further investigation under natural conditions.

The study's findings underscore the necessity of incorporating environmental and behavioral factors into future research endeavors. These factors encompass the complexity of the habitat, including aquatic vegetation and varied substrates, including non-natural ones, as well as the influence of physical variables such as temperature and luminosity. These variables have the capacity to substantially alter predation dynamics under natural conditions. The results of this study provide a foundational understanding that is crucial for the development of more effective management strategies within fish farming systems. The primary objective of these strategies is to mitigate the economic losses incurred due to predation on Odonata nymphs.

This study highlights the importance of time as a critical factor in predation dynamics involving Odonata nymphs and fish fingerlings in aquaculture systems. The findings indicate a peak in predation within the first 12 hours of contact, with no significant selectivity between native and invasive species. However, the elevated vulnerability of *P. reticulata* suggests potential for using native predators as biological control agents against invasive populations.

The implications for aquaculture are multifold. Managers should prioritize the protection of fingerlings during their initial introduction to tanks and consider adjusting stocking times or incorporating structural refuges. Additionally, the generalist behavior



of Odonata nymphs reinforces the need for ecosystem-based approaches that incorporate predator-prey dynamics into planning decisions.

Finally, this study contributes to a growing body of literature that seeks to reconcile production efficiency with ecological sustainability. By understanding natural predator-prey relationships, we may develop aquaculture strategies that reduce reliance on chemical interventions and align more closely with biodiversity conservation goals.

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