COMPARISON OF URBANIZATION EFFECTS ON LIFE HISTORY TRAITS IN *GAMBUSIA AFFINIS*

by

Emily Mae Edgar

HONORS CAPSTONE

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Supervisor:

Caitlin R. Gabor

Second Reader:

Kyndal Irwin

ABSTRACT

Impervious surfaces associated with urban development increase the flow of rainwater run-off entering streams that drain urban catchments and drive urban stream syndrome. Urban stream syndrome is the physical and ecological degradation of urban streams. Streams suffering from urban stream syndrome are generally more homogenous with lower levels of habitat structure due to scouring. We define environmental complexity as the degree of variation in stream habitat structure with low variability equating to low environmental complexity. More scoured streams likely have less resources and this could affect life history tradeoffs in fish. We investigated the relationship between environmental complexity and life history traits of the western mosquitofish, *Gambusia affinis.* Western mosquitofish are urban-tolerant and live-bearing species of fish that persist in a wide range of environmental conditions. I examined the fecundity (eggs and embryo), reproductive allotment (egg and embryo mass), and reproductive tradeoff in adult female G. *affinis* from six different stream populations that vary from high, medium, and low environmental complexity (Pyke GH, 2005). I measured the number of unfertilized gametes and the embryos (and their varying stage of development) in relation to female size and mass. I also measured dried brood mass to explore potential energy tradeoffs in fecundity, and reproductive allotment. I predicted that there would be greater reproductive tradeoff in areas with low environmental complexity due to more predicted limited resources. Specifically, I predicted that fecundity and reproductive allotment would be lower in areas with low environmental complexity. In contrast to our predictions, the fish had higher fecundity in low complexity habitats than in medium and high. Also, the fish had the greatest reproductive allocation in low complexity habitats than in medium and high complexity habitats. I found no significant trade-off across complexity, but high environmental complexity appears to have the highest fecundity and individual propagule mass when compared to low and medium complexity. This suggests that female mosquitofish in these populations are tolerant of a variety of conditions and that might explain their widespread dispersal. But it may account for the loss of intolerant species. With a greater understanding of anthropogenic effects on aquatic vertebrates, we can better understand how to conserve the natural habitat of these ecosystems.

DEDICATION

I dedicate my thesis work to the efforts my parents have made to put me into the position I find myself in. Without their sacrifices I would not have been able to complete the work I have accomplished. I would like to extend my gratitude to my partner who has kept me afloat in my efforts to pursue my education with honors. All of you have been my best cheerleaders.

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Introduction

Urbanization alters natural habitat by replacing previously green spaces with impervious surfaces that can lead to secondary effects such as runoff, therefore increasing the water pollution. (Marques, Manna, Frauendorf, 2020). The level at which an ecosystem has been urbanized can be determined by the percent of impervious surface cover in the sub watershed surrounding the sampling site (Paul and Meyer 2001; Walsh et al. 2005). Urban stream syndrome is the physical and ecological degradation of streams draining urban areas. Symptoms of urban stream syndrome include altered hydrology, increase in temperatures, and elevated changing concentrations of nutrition or pollutants (Walsh et al. 2005). These changes can further result in changes to environmental complexity. Environmental complexity describes the level of heterogeneity in the physical structure and biological interactions (Cannon & John, 2007). We define environmental complexity as the degree of variation in habitat structure with higher levels of habitat structure being more complex. Urban stream syndrome generally reduces levels of habitat structure which limits resources such as food availability, and refuge vegetation that may influence life history traits and overall fitness. Life history tradeoffs are an outcome of the energy used to benefit one trait of an organism that is linked to the detrimental change of another trait due to resource limitations (Stearns, 1989). The most prevalent example of this relationship within an organism is the cost of reproduction with there being two main components: the cost of survival, and the cost to future reproduction (S.C. Stearns, 1989). Life history traits of individuals in a population include: body condition/mass, fecundity, age (Dantzer, Westrick, Kesteren, 2016). Urban stream syndrome results in more homogenous habitat structure and lower levels of biodiversity (Walsh et al., 2005). This homogenization pushes aquatic environments to limit organisms and can be studied via changes in life history characteristics such as reproduction and survival. With altered nutrients in the aquatic environment, some species can capitalize on this availability with increased reproductive output such as in the live-bearing mosquitofish *Gambusia affinis* (Kolonin et al. 2022). Kolonin et al (2022) found that as urbanization (percent impervious cover) increased reproductive output increased. To understand this relationship, we examined the life history traits in *Gambusia affinis* sampled from six sites that varied in environmental complexity.

We hypothesized there will be an overall greater reproductive tradeoff in areas with low environmental complexity. We measured female mass, fecundity, and reproductive allocation in G. *affinis* in relation to environmental complexity to explore these tradeoffs. We predict that there will be a greater tradeoff in areas with low environmental complexity. We predicted that the fecundity, the measure of eggs per reproductive cycle, will be lower in areas with low environmental complexity. We also predict that the reproductive allotment (total brood mass) will be lower in areas with lower environmental complexity. Lastly, we predict the size of individual eggs will be smaller in areas with low environmental complexity.

Materials and Methods

Environmental complexity

The female *G. affinis* were collected from six different locations, ranging in environmental complexity. We quantified the environmental complexity of *G. affinis* habitat using an optical analysis technique from (Shumway et al. 2007) In a site where we collected *G. affinis* we measured 10M range along the bank and 2M out into the stream. We recorded six videos at each site to capture habitat structure. We used iMovie to randomly extract three frames from each video, the frames were imported to ImageJ to convert to 8-bit greyscale to measure the standard deviation of pixel density. Environmental complexity was quantified using the standard deviation of pixel intensity of each frame from the videos. Then we averaged the standard deviations of pixel intensity for all frames of each habitat. Higher average standard deviation reflected greater variation and therefore higher complexity. The average standard deviation of pixel intensity for each habitat was standardized based on the total mean and standard deviation pixel intensity for all habitats and was used to categorize each habitat structure as less complex (give range), moderately complex(range), or highly complex(range).

Life history traits

Gambusia affinis from each population (n=40) were euthanized using an ice bath and stored individually in 70% ethanol in 15mL falconer tubes. To dissect the fish, each specimen was laid individually under a dissection microscope on a rubber mat. Dissecting scissors were used to make an initial cut at the gonopore while the fish laid on dorsal side supported by forceps holding the anterior end. The cut from the gonopore was made to the operculum, and then the fish was laid on the left lateral side. Two additional cuts were made, one starting at the gonopore and another at the operculum going up to the dorsal side of the fish. These cuts then made a flap in the right lateral side of the fish allowing them to open and remove the clustered brood gently using forceps. The embryos/eggs were separated, individually counted, and staged using the Haynes (1995) classification method and a dissection microscope. We then separated the female and the offspring by placing them in separate microcentrifuge tubes and label with the number of specimen and the location it was collected. The microcentrifuge tubes were placed in drying oven set to 55 degrees Celsius for 24 hours. Once dried, the weight of the dry eviscerated female and dried brood were recorded.

From the recorded data, we calculated reproductive allotment as the total dry mass of all combined eggs or embryos per female (Kolonin et al, 2022). Additionally, we calculated individual offspring dry mass as the total combined number of eggs and embryos by the number of eggs or embryos per female (Kolonin et al, 2022). We used a general linear mixed model to examine the role of complexity on fecundity, reproductive allocation, and tradeoffs. We set population as the random variable. We controlled for dry mass (sqrt) of the females in each model in JMP. Our research was in accordance with IUCAC permit number 83.

Results

The fish had higher fecundity in low complexity habitats than in medium and high, as they get bigger their fecundity also increases more rapidly at low complexity. Overall high complexity had bigger fish, but all had the lowest fecundity (GLMM: F (2,205) = 11.387, p =<0 .0001; Fig. 1). The fish had greater dry brood mass in low complexity habitats than in medium and high (GLMM; F (2,-183) =7.04, p = 0.0011; Fig. 2). There was no tradeoff between individual propagule size and fecundity across complexity (GLMM: F (2,183) = 1.04, p = 0.356; Fig. 3)

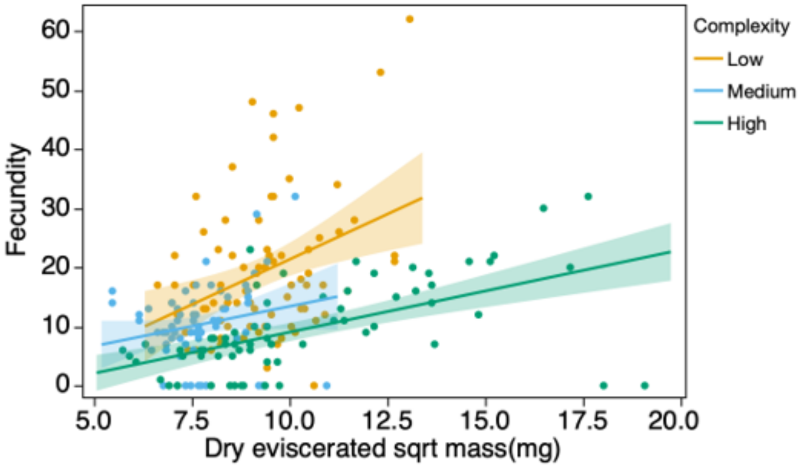

Fig 1. The relationship between fecundity and dry eviscerated square root mass across levels of environmental complexity

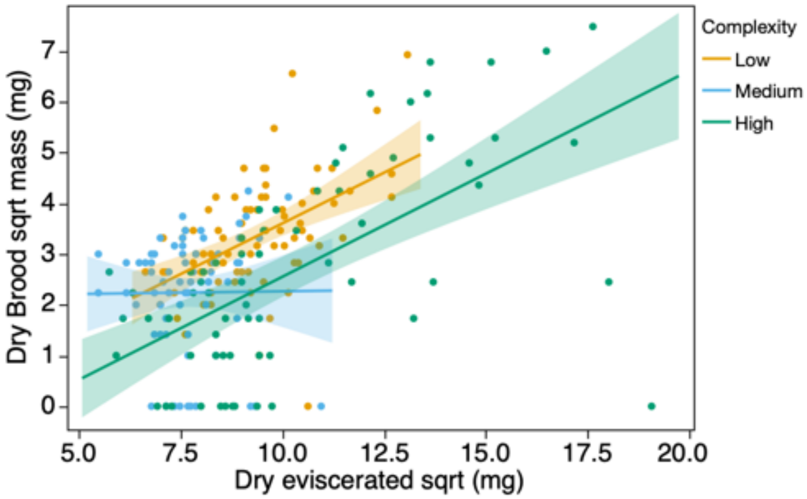

Fig 2. The relationship between dry brood square root mass and dry eviscerated square root mass across levels of environmental complexity

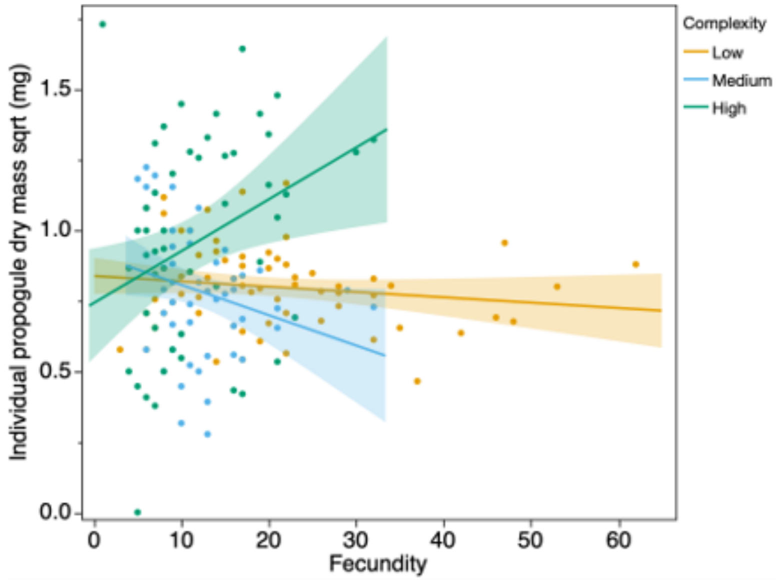

Fig 3. The tradeoff between individual propagule dry mass square root and fecundity across levels of environmental complexity

Discussion

In this study we looked at certain life history traits in *Gambusia affinis* to determine if there was a relationship between these traits and environmental complexity. We expected to see higher tradeoffs in environments with low environmental complexity. Specifically, we expected that low resource availability within environments that had lower environmental complexity would result in a lower individual egg size per female when compared to high environmental complexity. Following this, we predicted lower environmental complexity habitats to have lower reproductive allotment and fecundity. Our results indicate that fish in low environmental complexity have the greatest fecundity. In low environmental complexity, we found that fecundity rapidly increases with fish size, and high environmental complexity had larger fish, but lowest fecundity when controlled by dry eviscerated female mass. Fish in low environmental complexity have the greatest reproductive allotment when controlling for dry eviscerated female mass. Strangely, fish in medium complexity did not show an increase in brood mass with size of female. Finally, we found no significant trade-off across complexity, however there is some indication that females in high complexity environments are having a lower tradeoff due to the increase in fecundity with individual propagule mass.

These results are the opposite of what we predicted. Our prediction was based on the prediction that urbanization of streams led to a reduced quantity of resources in effected environments, which we categorized as low complexity. However, Kolonin et al. (2022) found that females in more urbanized streams had higher fecundity. This then supports our findings too. It may be that resources are not limiting in these environments or that urbanized environments favor individuals that invest more in reproduction in this species (Araya- Ajoy et al 2018). With more developed offspring, females may have more access to sexually mature males, allowing for more females to have a clutch of embryos. Having more access to sexually mature males may be a result of low predation pressure in the environment due to lack of resources available to predatory species because of urbanization (Ghalambor et al, 2004)

Western mosquitofish are native to Texas but have invaded over 40 countries, making them one of the most abundant species of freshwater fish in the world. *G. affinis* is a generalist species that is highly tolerant to environmental changes that may be outside the normal parameters that native species can tolerate (Marques, Manna, Frauendorf, 2020). One of the factors that allows for this population to invade these areas is the ability to allocate more energy to reproduction, growing the density of that species population in the non-native stream (Marques, Manna, Frauendorf, 2020). Additionally, some secondary effects from urbanization such as pollution of nitrogenous compounds runoff into aquatic environments and increase the nutrient availability and consumption for these species (Marques, Manna, Frauendorf, 2020; Kolonin, et al. 2022). Further research looking at intergenerational effects on *G. affinis* offspring would be useful to understand how the presence of higher population density of these invasive species affect native stream ecosystems in the future. With increasing urbanization raises serious questions for how the delicate stream ecosystem will be affected. A major goal of urban ecologists is to understand why certain species decline in urban environments while others thrive. Results from this study provide insight into why *G. affinis* is an urban tolerant species.

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