

MALE MATE CHOICE IN RELATION TO FEMALE SIZE IN THE SAILFIN

MOLLY, *POECILIA LATIPINNA*

THESIS

Presented to the Graduate Council  
Texas State University-San Marcos  
in Partial Fulfillment  
of the Requirements

for the Degree

Master of SCIENCE

by

Rosie Gonzalez, B.S.

San Marcos, Texas  
August 2007

**COPYRIGHT**

by

Rosalinda Gonzalez

August 2007

## **ACKNOWLEDGEMENTS**

First and foremost, I would like to offer my sincerest gratitude to my advisors, Dr. Caitlin Gabor and Dr. Andrea Aspbury, who convincingly conveyed encouragement and patience in regard to research and scholarship. I would also like to offer my thanks for their support throughout my personal obstacles. Without their guidance and persistent support, this thesis would not be possible. I would also like to thank my committee members, Dr. Clay Green and Dr. Floyd Weckerly for their thoughts and suggestions with this thesis. I thank the GASP lab for being amazing and stimulating lab mates. I am also grateful to my parents and sisters for their support. Special thanks are due to Cynthia Morgan who endured this challenging process with me, continuously offering support and encouragement.

This manuscript was submitted on June 22, 2007.

## TABLE OF CONTENTS

	<b>Page</b>
ACKNOWLEDGEMENTS.....	iv
LIST OF FIGURES.....	vi
CHAPTER	
I. INTRODUCTION.....	1
II. DIFFERENTIAL SPERM EXPENDITURE IN RELATION TO MALE AND FEMALE SIZE IN <i>POECILIA LATIPINNA</i> .....	14
III. EVALUATING MATING PREFERENCES: PREFERENCE FUNCTIONS IN MALE <i>POECILIA LATIPINNA</i> .....	23

## CHAPTER I

### INTRODUCTION

Sexual selection is differential reproductive success resulting from variation in mating success (Darwin 1871). Sexual selection operates via two mechanisms: intrasexual selection, which is contest competition among members of one sex for access to mates, and intersexual selection, which occurs when members of the choosier sex determine which members of the other sex will have a chance to mate. Generally, females tend to be the choosier sex because they are most often the limiting sex in the population. This arises because ova are generally more expensive to produce than sperm (Trivers 1972), females often invest more in offspring, and because the operational sex ratio (OSR) is often male biased (Emlen & Oring 1977).

Several hypotheses have been proposed to explain why females preferentially mate with certain males and not others. A variety of benefits, including both direct and indirect, are obtained by females mating with certain males (Andersson 1994). Direct benefits include mating with males that are more likely to produce enough sperm to fertilize a female's clutch of eggs (Halliday 1978), or mating with males that provide some resource that increases a female's fecundity (Andersson 1994). When males offer no such resources, females may select males based on indirect benefits. Hypotheses for

the evolution of female choice based on indirect benefits fall into two broad groups: runaway sexual selection and good-genes models (Kokko et al. 2003). Runaway selection theory proposes that discriminating females acquire sperm with genes whose primary effect is to influence their daughters to prefer attractive males and to endow their sons with attributes that will be preferred by most females (Andersson 1994). In addition, if a slight majority of females in an ancestral population had a preference for a certain male characteristic, perhaps initially because the preferred trait was indicative of some survival advantage enjoyed by the male, then females that mate with preferred males would have produced offspring that inherited the genes for the mate preference from their mothers and the genes for the attractive male character from their fathers (Andersson 1994). This pattern could generate a runaway process in which more extreme female preferences and male traits spread together as new mutations occur (Andersson 1994).

Good-genes models postulate that the degree of expression of male ornaments reflects the overall viability or quality of the male (Andersson 1994): the displays or traits that males possess are indicators of genetic quality. If males with more attractive traits are genetically superior to males with less attractive traits, then choosy females will secure better genes for their offspring (Zahavi 1975). These genes might advance the survival chances of their offspring by conferring hereditary resistance to parasitic infection or disease or by promoting adaptive foraging skills (Andersson 1994). The “handicap principle” further proposes that females specifically mate with males who possess phenotypes that negatively affect them in daily survival because only the “best males” can bear the cost of elaborate traits (Zahavi 1975).

### Male mate choice and sperm production

The traditional viewpoint of sexual selection theory has been that females are the choosier sex (for reasons described above). Male mate choice is, however, expected when males provide paternal care, when the OSR is female biased, when mating is costly to males, or when there is a limited supply of highly viable females (Andersson 1994). For example, males should mate preferentially with larger females in species where female fecundity increases with body size, because male fitness increases with the number of matings and offspring sired.

Implicit in much of the historical discussion of sexual selection is the assumption that male sperm production has a negligible cost compared to female egg production. However, although per unit egg production is relatively more costly than per unit sperm production, sperm production does entail energetic costs (Dewsbury 1982; Nakatsuru & Kramer 1982; Shapiro et al. 1994; review in Wedell et al. 2002). For this reason, we expect that males may also preferentially select mates. When females differ in reproductive value, males are expected to preferentially mate with females with a higher potential reproductive rate (Farr 1989). Egg production increases with body size in many species of fishes (Perrone 1978). Such an increase is often expected simply because larger females have more internal space for eggs. There is a potential advantage of male choice of mates in species where females differ markedly in fecundity related to body size. Although many studies have demonstrated a female association preference for larger males (reviewed in Andersson 1994), less studies have demonstrated male mate preferences for larger females (for exceptions see: fish: *Poecilia latipinna*: Aspbury & Gabor 2004 a; Gabor 1999; Ptacek & Travis 1997; guppies, *P. reticulata*: Herdman et al.

2004; Dosen & Montgomerie 2004; smooth newt, *Triturus vulgaris*: Verrell 1986; sand lizard, *Lacerta agilis*: Olsson 1992). In many poeciliid fish, females are larger than males, and larger females are more fecund (Farr 1989; guppies; *P. reticulata* see Reznick & Endler 1982; Herdman et al. 2004; *P. latipinna*; Konkin-Garcia et al. unpublished data), suggesting that large female size may be a trait males would prefer. In addition, when female reproduction is based on energy reserves accumulated over a long period, large female size may confer a fecundity advantage because the capacity for storing energy reserves increases more rapidly with body size than do metabolic costs (Millar & Hickling 1992). To the extent that mating takes time or depletes male sperm or other resources, and reduces males' chances of fertilizing other females, males should favor the larger, most fecund females as mates (Parker 1970). Therefore, males may be prepared to invest more of their potentially limited reproductive resources in larger females.

Another proposed hypothesis to explain male preference for larger females may be that these females are more likely to be the focus of greater male-male competition (Gage 1998). Males may be prepared to invest more sperm in mating with larger females as suggested by the higher rates of multiply sired broods (Trexler et al. 1997). Sexual selection by male-male competition occurs when individual males can monopolize access to females, however, the competition between males does not necessarily stop when copulation is over: the primary determinant of a male's mating success is not whether he copulates, but whether his sperm fertilize eggs (Parker 1970).

Male mate choice has been measured in *P. latipinna* by examining association times (Gabor 1999), male mating attempts (Ptacek & Travis 1997), and male sperm production (Aspbury & Gabor 2004 a,b). Because both males and females mate multiply

in *P. latipinna*, sperm competition is a possible factor influencing sperm production and possibly sperm transfer (Aspbury 2007). When males are provided with stimuli from females, physiological changes associated with sperm production occur, and is referred to as the priming response (Olsén & Liley 1993; Bozynski & Liley 2003; Aspbury & Gabor 2004 a,b). Sperm priming has been used to examine male mate choice in *P. latipinna* (Aspbury & Gabor 2004 a). Male *P. latipinna* prime more sperm in the presence of larger females than in the presence of smaller females and smaller males prime less sperm than larger males (Aspbury & Gabor 2004 a). Smaller males primed less sperm when isolated from females, and males primed more sperm in the presence of females (Aspbury & Gabor 2004 a). Sperm may therefore be more energetically costly for small males, thus limiting reproductive success. However, there may be no relationship between sperm primed and sperm transferred. Females prefer larger males; therefore, larger males may mate with numerous females and adjust sperm transfer accordingly (Aspbury & Gabor 2004 a; Aspbury 2007).

### **Evaluating mating preferences: preference functions**

When examining mate choice, it is also important to describe how the strength of mating preference for a given trait varies with the degree of expression of the trait. Jennions & Petrie (1997) examined empirical and theoretical studies of variation in individual mating preferences and concluded that individual variation in preference is common and can have major consequences for models of sexual selection. Mating preferences are determined by preference functions and degree of choosiness (Jennions & Petrie 1997). Preference functions characterize the form of individual mating preferences and allow for

the comparison of variation within and among individuals of one sex (Wagner et al. 1995). A preference function describes how the strength of an individual's mating response to a trait varies with the degree of expression of the trait and can be measured for individuals as well as populations (Wagner 1998). Preference functions can be measured in relative as well as in absolute terms (Wagner 1998). Relative preference functions describe how the strength of preference varies as a function of the difference in trait value between two mates presented simultaneously (Wagner 1998; MacLaren et al. 2004). Absolute preference functions describe how the strength of response changes with variation in the trait value of stimuli presented individually (Wagner et al. 1995). Prior to deciding whether to test relative or absolute preference functions, it is important to consider how individuals of the species in question encounter potential mates in the wild. Absolute preference functions are the preferred method of assessment for species in which females rarely encounter multiple mates simultaneously (MacLaren & Rowland 2006). Conversely, relative preference functions are the preferred method of assessment for species that frequently encounter more than one mate at a time (MacLaren & Rowland 2006). However, some species may encounter potential mates both singly and simultaneously. When both designs were tested using female sailfin mollies (*P. latipinna*), MacLaren & Rowland (2006) found that female sailfin mollies exhibit a much stronger preference for larger males presented simultaneously than when presented in sequence. While all prior studies on preference functions have been performed on females, preference functions may also describe how the strength of a male's mating response to a female trait varies with the degree of expression of the trait. No known studies have explored male preference functions.

## Natural history

### *Poeciliidae*

The family Poeciliidae contains 200 species in 20 genera of livebearing fishes. They are found in tropical and subtropical latitudes of the New World. Females typically have a 30 d ovarian cycle, and are usually more receptive to males for 1-2 d after dropping a brood (Liley 1966). Insemination takes place after a male inserts his gonopodium, which is a modified anal fin used to transfer sperm packets, into the females' gonopore. Female guppies, *Poecilia reticulata*, store sperm for several months (Baerends et al. 1955) and many females mate multiply.

### *Poecilia latipinna*

*Poecilia latipinna*, the sailfin molly, is a livebearing fish native to brackish waters of southern Mexico near Rio Tuxpan along the Gulf of Mexico through North Carolina. *Poecilia latipinna* has also been introduced to areas outside of its native range, including the San Marcos and Comal rivers in central Texas (Brown 1953). Males are sexually dimorphic with secondary sexual characteristics consisting of a large dorsal fin and enhanced coloration compared to gray females with no enlarged fin. The dorsal fin is erected and presented to the female with a sigmoid curving of the body in courtship displays (Travis & Woodward 1989). This is accompanied by nibbling on the female's gonopore and gonopodial thrusting at the female. Males may also mate without courtship by gonopodial thrusting. This is an attempt at insemination without female cooperation in which the male orients himself behind a female and attempts to insert his

gonopodium into the female's gonopore. While larger males more often court females, smaller males rely primarily on gonopodial thrusting (Travis & Woodward 1989). Males exhibit genetic polymorphism for body size that is associated with differences in secondary sexual characters and behavior patterns (Farr et al. 1986; Travis & Woodward 1989). Large males aggressively display to other smaller males and limit their access to females (Travis & Woodward 1989; Travis et al. 1990). Females gain no known material benefits from males, but females prefer to associate with larger males (Ptacek & Travis 1997; Ptacek 1998; Gabor 1999). Males also prefer to associate with larger females (Travis & Trexler 1987; Ptacek & Travis 1997; Gabor 1999) and prime more sperm for larger females (Aspbury & Gabor 2004a).

### **Research objectives**

Given the prior work performed on *P. latipinna*, I explored further questions about the effects of male and female size on male sperm expenditure. I also examined male preference functions in response to different size female sailfin molly models.

In Chapter II, I examined the proportion of sperm lost while mating with individual females relative to female size. I examined whether sperm availability after mating reflects sperm expenditure. Male *P. latipinna* increase sperm availability in the presence of larger females (Aspbury & Gabor 2004a), thus I expected that males would expend more sperm when mating with larger females.

In Chapter III, I explored male preference functions using males from two different populations, Vicente Guerrero, MX and Alfred Bonfil, MX. Preference functions are useful because they reveal the shape of the preference and hence the

potential for selection on the preferred trait. Because males prefer to mate with larger females (Ptacek & Travis 1997; Gabor 1999) I predicted that males would prefer to associate with the larger female of a given pair. However, male sailfin mollies from one of these two populations (Alfred Bonfil) avoided mating with larger heterospecifics over smaller sailfin mollies (Gumm & Gabor 2005) thus I predicted that these males would only prefer larger females when the difference in size was the least. Whereas, I expected that males from the second population (Vincente Guerrero) would prefer the larger of each of the female model pairs.

The results from these experiments will help shed light on how sexual selection via male mate choice can affect the evolution of female traits, primarily female body size. This differs from the traditional approach of considering how female choice affects the evolution of male traits.

## REFERENCES

- Andersson, M.** 1994. *Sexual Selection*. Princeton, New Jersey: Princeton University Press.
- Aspbury, A.S.** 2007. Sperm competition effects on sperm production and expenditure in sailfin mollies, *Poecilia latipinna*. *Behavioral Ecology*, **18**; 776-780.
- Aspbury, A.S. & Gabor, C.R.** 2004a. Differential sperm priming by male sailfin mollies (*Poecilia latipinna*): Effects of female and male size. *Ethology*, **110**, 193-202.
- Aspbury, A.S. & Gabor, C.R.** 2004b. Discriminating males alter sperm production between species. *Proceedings of the National Academy of Sciences*, **101**, 15970-15973.
- Baerends, G.P., Brouwer, R. & Waterbolk, H.T.J.** 1955. Ethological studies on *Lebistes reticulatus* (Peters). I. An analysis of male courtship pattern. *Behaviour*, **8**, 249-335.
- Bozynski, C.C. & Liley, N.R.** 2003. The effect of female presence on spermiation, and of male sexual activity on 'ready' sperm in the male guppy. *Animal Behaviour*, **65**, 53-58.
- Brown, W.H.** 1953. Introduced fish species of the Guadalupe River basin. *Texas Journal of Science*, **5**, 245-251.
- Darwin, C.** 1871. *The Descent of Man and Selection in Relation to Sex*. Murray, London, United Kingdom.
- Dewsbury, D.A.** 1982. Ejaculate cost and male choice. *American Naturalist*, **119**, 601-610.
- Dosen, L.D. & Montgomerie, R.** 2004. Female size influences mate preferences of male guppies. *Ethology*, **110**, 245-255.
- Emlen, S.T. & Oring, L.W.** 1977. Ecology, sexual selection, and evolution of mating systems. *Science*, **197**, 215-223.
- Farr, J.A.** 1989. Sexual selection and secondary sexual differentiation in poeciliids: determinants of male mating success and the evolution female choice. In: *Ecology and Evolution of Livebearing Fishes* (ed G.K. Meffe & F.F. Snelson), New Jersey: Prentice Hall. pp. 91-123.
- Farr, J.A., Travis, J. & Trexler, J.C.** 1986. Behavioral allometry and interdemic

- variation in sexual behavior of the sailfin molly, *Poecilia latipinna* (Pisces, Poeciliidae). *Animal Behaviour*, **34**, 497-509.
- Gabor, C.** 1999. Association patterns of sailfin mollies (*Poecilia latipinna*): alternative hypotheses. *Behavioral Ecology and Sociobiology*, **46**, 333-340.
- Gage, M.J.G.** 1998. Mammalian sperm morphometry. *Proceedings of the Royal Society of London Series B-Biological Sciences*, **265**, 97-103.
- Gumm, J.M. & Gabor, C.R.** 2005. Asexuals looking for sex: conflict between species and mate-quality recognition in sailfin mollies (*Poecilia latipinna*). *Behavioral Ecology and Sociobiology*, **58**, 558-565.
- Halliday, T.** 1978. Sexual selection and mate choice. In: Krebs JR, Davies NB (eds) *Behavioural Ecology: An Evolutionary Approach*. Sinauer Associates, Sunderland, Massachusetts, pp 180-213.
- Herdman, E.J. E., Kelly, C.D. & Godin, J.G.J.** 2004. Male mate choice in the guppy (*Poecilia reticulata*): Do males prefer larger females as mates? *Ethology*, **110**, 97-111.
- Jennions, M.D. & Petrie, M.** 1997. Variation in mate choice and mating preferences: A review of causes and consequences. *Biological Reviews of the Cambridge Philosophical Society*, **72**, 283-327.
- Kokko, H., Brooks, R., Jennions, M., & Morley, J.** 2003. The evolution of mate choice and mating biases. *Proceedings of the Royal Society of London Series B-Biological Sciences*, **270**, 653-664.
- Liley, N.R.** 1966. Ethological isolating mechanisms in four sympatric species of poeciliid fishes. *Behaviour (Suppl.)*, **13**, 1-197.
- MacLaren, R.D. & Rowland, W.J.** 2006. Differences in female preference for male body size in *Poecilia latipinna* using simultaneous versus sequential stimulus presentation designs. *Behaviour*, **143**, 273-292.
- MacLaren, R.D., Rowland, W.J. & Morgan, N.** 2004. Female preferences for sailfin and body size in the sailfin molly, *Poecilia latipinna*. *Ethology*, **110**, 363-379.
- Millar, J.S. & Hickling, G.J.** 1992. The fasting endurance hypothesis revisited. *Functional Ecology*, **6**, 496-498.
- Nakatsuru, K. & Kramer, D.L.** 1982. Is sperm cheap? Limited male fertility and female choice in the lemon tetra (Pisces, Characidae). *Science*, **216**, 753-755.
- Olsson, M.** 1992. Contest success in relation to size and residency in male sand lizards,

- Lacerta agilis*. *Animal Behaviour*, **44**, 386-388.
- Olsén, K.H. & Liley, N.R.** 1993. The significance of olfaction and social cues in milt availability, sexual hormone status, and spawning behavior of male rainbow trout (*Oncorhynchus mykiss*). *General and Comparative Endocrinology*, **89**, 107-118.
- Parker, G.A.** 1970. Sperm competition and its evolutionary consequences in insects. *Biological Reviews of the Cambridge Philosophical Society*, **45**, 525-567.
- Perrone, M.** 1978. Economy of brood defence by parental cichlid fishes, *Cichlasoma maculicauda*. *Oikos*, **31**, 137-141.
- Ptacek, M.B.** 1998. Interspecific mate choice in sailfin and shortfin species of mollies. *Animal Behaviour*, **56**, 1145-1154.
- Ptacek, M.B. & Travis, J.** 1997. Mate choice in the sailfin molly, *Poecilia latipinna*. *Evolution*, **51**, 1217-1231.
- Reznick, D. & Endler, J.A.** 1982. The impact of predation on life history evolution in Trinidadian guppies (*Poecilia reticulata*). *Evolution*, **36(1)**, 160-177.
- Shapiro, D.Y., Marconato, A. & Yoshikawa, T.** 1994. Sperm economy in a coral-reef fish, *Thalassema bifasciatum*. *Ecology*, **75**, 1334-1344.
- Travis, J. & Trexler, J.C.** 1987. Regional variation in habitat requirements of the sailfin molly, with special reference to the Florida Keys. *Nongame Wildlife Technical Report Number 3, Florida Game and Fresh Water Fish Commission: Tallahassee, Florida*, **3**, 1-47.
- Travis, J., Trexler, J.C. & Mulvey, M.** 1990. Multiple paternity and its correlates in female *Poecilia latipinna* (Poeciliidae). *Copeia*, 722-729.
- Travis, J. & Woodward, B.D.** 1989. Social context and courtship flexibility in male sailfin mollies, *Poecilia latipinna* (Pisces, Poeciliidae). *Animal Behaviour*, **38**, 1001-1011.
- Trivers, R.L.** 1972. Parental investment and sexual selection. In: *Sexual Selection and the Descent of Man* (Ed. By Campbell, B.), pp. 136-172. Chicago: Aldine Atherton.
- Verrell, P.A.** 1986. Male discrimination of larger, more fecund females in the smooth newt, *Triturus vulgaris*. *Journal of Herpetology*, **20**, 416-422.
- Wagner, W.E.** 1998. Measuring female mating preferences. *Animal Behaviour*, **55**, 1029-1042.
- Wagner, W.E., Murray, A.M. & Cade, W.H.** 1995. Phenotypic variation in the mating

preferences of female field crickets, *Gryllus integer*. *Animal Behaviour*, **49**, 1269-1281.

**Wedell, N., Gage, M.J. G. & Parker, G.A.** 2002. Sperm competition, male prudence and sperm-limited females. *Trends in Ecology & Evolution*, **17**, 313-320.

**Zahavi, A.** 1975. Mate selection-a selection for a handicap. *Journal of Theoretical Biology*, **53**, 205-214.

## CHAPTER II

### DIFFERENTIAL SPERM EXPENDITURE IN RELATION TO MALE AND FEMALE

#### SIZE IN *POECILIA LATIPINNA*

#### INTRODUCTION

It is increasingly evident that spermatogenesis can be energetically costly to males, thus limiting reproductive success (Dewsbury 1982; Nakatsuru & Kramer 1982; Shapiro et al. 1994; review in Wedell et al. 2002; Aspbury & Gabor 2004 a). Males may exercise mate choice by differentially allocating sperm to maximize reproductive success. The physiological changes associated with sperm production when males are provided with stimuli from females are referred to as the priming response (Olsén & Liley 1993; Bozynski & Liley 2003, Aspbury & Gabor 2004 a). The priming response may be a mechanism by which males conserve energy associated with sperm production in the absence of females (Liley & Kroon 1995).

In addition to reducing physiological costs associated with spermatogenesis, differential sperm production for desirable females, such as larger, more fecund females, may increase male mating success and can indicate male mate choice (Aspbury & Gabor 2004 a, b). In many species, female fecundity increases with size, and males exhibit preferences for large females (*Poecilia latipinna*: Aspbury & Gabor 2004 a; Gabor 1999; Ptacek & Travis 1997; guppies, *P. reticulata*: Herdman et al. 2004; Dosen & Montgomerie 2004; smooth newt, *Triturus vulgaris*: Verrell 1986; sand lizard, *Lacerta*

*agilis*: Olsson 1992). Therefore, male mating preferences may translate into males expending more sperm for larger females.

It is also possible that there is no relationship between sperm primed and sperm transferred. Females prefer larger males; therefore, larger males may mate with numerous females and adjust sperm transfer accordingly so that no relationship exists between sperm primed and sperm transferred (Aspbury & Gabor 2004 a; Aspbury 2007). The following experiment examines sperm availability and how it relates to male and female size.

In sailfin mollies, *P. latipinna*, males show a greater preference for larger females than smaller females, measured as association times and mating attempts (Ptacek & Travis 1997; Gabor 1999), and males prime more sperm when in the presence of larger females (Aspbury & Gabor 2004a). Additionally, males of different sizes differ in the level of sperm primed. Smaller males primed more in response to female stimuli than larger males (Aspbury & Gabor 2004a). The objective of this study was to determine how the previously demonstrated greater strength of male preferences for larger females over smaller females relates to sperm availability.

It is important to examine sperm availability as opposed to sperm transfer because sperm transferred to females may reflect insemination success while sperm availability may be a more accurate measurement of male investment (Robinson et al. in press). In addition, sperm loss may occur during sperm transfer. In the eastern mosquitofish, higher sperm expenditure does not always lead to higher sperm transfer to the female (Evan et al. 2003). Therefore, sperm recovered from females after sperm transfer does not precisely indicate male sperm expenditure. Also, sperm loss affects sperm availability

for future matings, and therefore, affects a male's future reproductive success. Since males expend sperm during mating, sperm availability after mating will possibly reflect potential sperm allocation. For example, low sperm availability subsequent to mating may indicate that a large amount of sperm was expended in the course of mating.

If sperm expenditure is a variable that contributes to male mating preferences for larger, more fecund females, then I expect that sperm expenditure by male sailfin mollies will exhibit the same pattern as observed previously in this species: males sailfin mollies are expected to expend more sperm when mating with larger females than when mating with smaller females. Therefore, I predict that males will have more sperm available after mating with smaller females than with larger females and that this response will be greatest in smaller males.

### **Methods**

The sailfin molly, *Poecilia latipinna*, is a livebearing fish native to brackish waters of southern Mexico and the southern United States along the Gulf of Mexico through North Carolina. *Poecilia latipinna* used in this experiment were collected from Spring Lake, in San Marcos, Texas. Fishes were maintained on a 14:10 hour light: dark cycle using natural simulated UV florescent lighting that simulates daylight. Fishes were fed a diet of Aquatronic Spirulina Flake mixed with Ocean Star International Inc. Fresh Water Flake food twice a day and supplemented with live and freeze-dried brine shrimp.

Fishes were separated by sex for at least 30 d before testing in 38 l aquaria (54 cm x 29 cm x 33 cm). The isolation of females for at least 30 d increases the chance that all females will be at a similar stage in the brood cycle. As female sailfin mollies have a 30 d

ovarian cycle, most fishes will have dropped any broods they may have held prior to testing (Farr and Travis 1986; Snelson et al. 1986). Male and female test fish were selected randomly and tested only once.

On day 0, I removed a male (N=60) from its stock aquarium and measured and recorded standard length (SL) using digital calipers. After measuring SL, I extracted sperm following the methods of Aspbury & Gabor (2004 a). The males were placed along the edge of a shallow Petri dish lined with wet cotton with their ventral side up. Gentle pressure was applied to the side of the male, going from the eye laterally, to the base of the anal fin. Spermatozeugmata was collected using an aspirator. The spermatozeugmata was placed into a micro-centrifuge tube with 100 ul of 0.9% saline solution (0.9 g NaCl/100 ml water), and was repeatedly drawn up and expelled from a pipette in order to distribute sperm cells evenly. This process was repeated on each male until no more spermatozeugmata were expelled from the base of the gonopodium.

Immediately after sperm extraction, males were randomly assigned a female (N=60). Both individuals were placed in a 18 l aquarium separated into two sections by a clear Plexiglas divider. On day 3, I removed the divider, and the male was able to physically interact with the female for 60 min after the first mating attempt (gonopodial thrust). For the first 10 min of this mating trial, the number of gonopodial thrusts were recorded. Immediately following the mating trial, sperm was extracted from the male. Sperm collected from males after mating may be a more accurate measure of male investment as opposed to sperm transfer (the sperm recovered from a female's reproductive tract after mating) because sperm loss may occur during sperm transfer (Evans et al. 2003; Robinson et al. in press). Sperm recovered from a male after mating

affects sperm availability for future matings, and thus affects a male's potential reproductive success.

Sperm cells were placed on an improved Hy-Lite Neubauer chamber hemocytometer under 400x magnification. A digital image of the sperm cells was taken, and sperm cells were counted.

### **Results**

Male sailfin mollies did not differ in sperm expenditure relative to female size (multiple regression:  $P = 0.1121$ ,  $r^2 = 0.046$ ,  $N = 56$ ; Figure 2.1). Larger male sailfin mollies expended less sperm relative to their baseline than did smaller males (multiple regression:  $P < 0.001$ ,  $r^2 = 0.314$ ,  $N = 56$ ; Figure 2.1).

### **Discussion**

In this experiment, neither large nor small males differentially expend sperm relative to female size. Although larger females elicit a greater priming response (Aspbury & Gabor 2004 a), males do not expend more sperm to larger females. Sperm priming does not directly relate to sperm expenditure relative to female size. Males may only exhibit mate choice in terms of sperm priming, but not sperm expenditure. In a similar study, Evans & Magurran (1999) found that sperm stripped in guppies was not a good predictor of fertilization success. In addition, higher sperm expenditure does not always lead to higher sperm transfer to the female (Evan et al. 2003).

While it appears that males may not be able to control or alter sperm usage from these results, we know from a prior study (Robinson et al. in press) that male sailfin

mollies can control sperm usage and preferentially expend more sperm in the presence of heterospecific females over conspecific females. While these results seem surprising, it is possible that males are retaining more sperm for future matings with conspecifics. An increase in sperm availability after mating with a conspecific female could indicate that males expend less sperm when mating with conspecifics, or alternatively, it could indicate that males produce more sperm while mating with conspecifics (Robinson et al. in press).

There was a significant difference in the proportion of sperm loss between large males and small males. Larger males had significantly more sperm available after mating than did smaller males. Therefore, larger males expend less sperm per female than do smaller males. One explanation is that energetic costs associated with sperm expenditure may be higher in smaller males than in larger males (Aspbury & Gabor 2004 a).

Although small males do not have more sperm available than large males, they exhibit a large increase relative to their 'baseline' sperm counts when presented with a female, and large decreases relative to their 'baseline' sperm counts when not presented with females (Aspbury & Gabor 2004 a). This result suggests that sperm is costly and may be more costly for smaller males. In addition, smaller males have less energy available (Pitnick & Markow 1994), and therefore are more likely to show the strongest fluctuations in sperm availability. Sperm competition is a likely factor influencing sperm production (Aspbury 2007) and sperm expenditure in *P. latipinna* because both males and females mate multiply (Travis et al. 1990; Trexler et al. 1997). Smaller males may expend more sperm as a way to offset the same fertilization gains of larger males.

Alternatively, larger males, which are preferred by females, may expend less sperm because they may partition their sperm across several females. Smaller males rely more on sneaky mating, therefore, when given the opportunity to mate, the proportion of sperm loss could be greater. Warner et al. (1995) found that male bluehead wrasse (*Thalassoma bifasciatum*) that mated most frequently distributed less sperm per mating than those, which mated less often.

In conclusion, while male sailfin mollies prime more sperm for larger females and more so in smaller males, male sperm expenditure does not follow this pattern. Sperm competition may be the main explanation for the lack of correlation between sperm priming and expenditure in this species but needs to be further examined.

## REFERENCES

- Aspbury, A.S.** 2007. Sperm competition effects on sperm production and expenditure in sailfin mollies, *Poecilia latipinna*. *Behavioral Ecology*, 18:776-780.
- Aspbury, A.S. & Gabor, C.R.** 2004a. Differential sperm priming by male sailfin mollies (*Poecilia latipinna*): Effects of female and male size. *Ethology*, 110, 193-202.
- Aspbury, A.S. & Gabor, C.R.** 2004b. Discriminating males alter sperm production between species. *Proceedings of the National Academy of Sciences*, 101, 15970-15973.
- Bozynski, C.C. & Liley, N.R.** 2003. The effect of female presence on spermiation, and of male sexual activity on 'ready' sperm in the male guppy. *Animal Behaviour*, 65, 53-58.
- Dewsbury, D.A.** 1982. Ejaculate cost and male choice. *American Naturalist*, 119, 601-610.
- Dosen, L. D. & Montgomerie, R.** 2004. Female size influences mate preferences of male guppies. *Ethology*, 110, 245-255.
- Evans, J.P., Pierotti, M. & Pilastro, A.** 2003. Male mating behavior and ejaculate expenditure under sperm competition risk in the eastern mosquitofish. *Behavioral Ecology*, 14, 268-273.
- Evans, J.P. & Magurran, A.E.** 1999. Male mating behaviour and sperm production characteristics under varying sperm competition risk in guppies. *Animal Behaviour*, 58, 1001-1006.
- Farr, J.A. & Travis, J.** 1986. Fertility advertisement by female sailfin mollies, *Poecilia latipinna* (Pisces, Poeciliidae). *Copeia*, 467-472.
- Gabor, C.** 1999. Association patterns of sailfin mollies (*Poecilia latipinna*): alternative hypotheses. *Behavioral Ecology and Sociobiology*, 46, 333-340.
- Herdman, E.J.E., Kelly, C.D. & Godin, J.G.J.** 2004. Male mate choice in the guppy (*Poecilia reticulata*): Do males prefer larger females as mates? *Ethology*, 110, 97-111.
- Liley, N.R. & Kroon, F.J.** 1995. Male dominance, plasma hormone concentrations, and the availability of milt in male rainbow trout (*Oncorhynchus mykiss*). *Canadian Journal of Zoology*, 73, 826—836.
- Nakatsuru, K. & Kramer, D.L.** 1982. Is sperm cheap? Limited male fertility and female

choice in the lemon tetra (Pisces, Characidae). *Science*, **216**, 753-755.

**Olsén, K.H. & Liley, N.R.** 1993. The significance of olfaction and social cues in milt availability, sexual hormone status, and spawning behavior of male rainbow trout (*Oncorhynchus mykiss*). *General and Comparative Endocrinology*, **89**, 107-118.

**Olsson, M.** 1992. Contest success in relation to size and residency in male sand lizards, *Lacerta agilis*. *Animal Behaviour*, **44**, 386-388.

**Pitnick, S. & Markow, T.A.** 1994. Large-male advantages associated with costs of sperm production in *Drosophila hydei*, a species with giant sperm. *Proceedings of the National Academy of Sciences*, **91**, 9277-9281.

**Ptacek, M.B. & Travis, J.** 1997. Mate choice in the sailfin molly, *Poecilia latipinna*. *Evolution*, **51**, 1217-1231.

Robinson, D. M. A. S. Aspbury, & C. R. Gabor. In press. Differential sperm expenditure by male sailfin mollies, *Poecilia latipinna*, in a unisexual-bisexual species complex and the influence of spermiation during mating. *Behavioral Ecology and Sociobiology*

**Shapiro, D.Y., Marconato, A. & Yoshikawa, T.** 1994. Sperm economy in a coral-reef fish, *Thalassemia bifasciatum*. *Ecology*, **75**, 1334-1344.

**Snelson, F.F., Wetherington, J.D. & Large, H.L.** 1986. The relationship between interbrood interval and yolk loading in a generalized Poeciliid fish, *Poecilia latipinna*. *Copeia*, 295-304.

**Travis, J., Trexler, J.C. & Mulvey, M.** 1990. Multiple paternity and its correlates in female *Poecilia latipinna* (Poeciliidae). *Copeia*, 1990, 722-729.

**Trexler, J.C., Travis, J. & Dinep, A.** 1997. Variation among populations of the sailfin molly in the rate of concurrent multiple paternity and its implications for mating-system evolution. *Behavioral Ecology and Sociobiology*, **40**, 297-305.

**Verrell, P.A.** 1986. Male discrimination of larger, more fecund females in the smooth newt, *Triturus vulgaris*. *Journal of Herpetology*, **20**, 416-422.

**Warner, R.R., Shapiro, D.Y., Marcanato, A. & Petersen, C.W.** 1995. Sexual conflict-males with highest mating success convey the lowest fertilization benefits to females. *Proceedings of the Royal Society of London Series B-Biological Sciences*, **262**, 135-139.

**Wedell, N., Gage, M.J.G. & Parker, G.A.** 2002. Sperm competition, male prudence and sperm-limited females. *Trends in Ecology and Evolution*, **17**, 313-320.

## CHAPTER III

### EVALUATING MATING PREFERENCES: PREFERENCE FUNCTIONS IN MALE

#### *POECILIA LATIPINNA*

### INTRODUCTION

Preference functions describing how the strength of a mating response varies with the degree of expression of the preferred trait are common features in genetic models of the evolution of preferences (Jennions & Petrie 1997; Wagner 1998). A preference function describes how the strength of an individual's mating response to a trait varies with the degree of expression of the trait and can be measured for individuals as well as populations (Wagner 1998). Preference functions provide more useful information than that from traditional dichotomous choice tests, because they reveal the shape of the preference and hence the potential for selection on the preferred trait (Widemo & Sæther 1999).

There are two different forms of preference functions: absolute and relative. Absolute preference functions describe how the strength of response changes with variation in the trait value of stimuli presented individually (Wagner et al. 1995). Relative preference functions are determined by presenting two stimuli simultaneously to individuals and relating the difference in response to the difference in stimuli (Wagner

1998). There are two general types of relative preference functions: standard and variable (Wagner 1998). These two types of relative preference functions differ in the types of stimuli individuals are asked to choose between. The difference in response can be measured with one of the stimuli held constant between tests (standard relative preference function), or both stimuli can vary between tests (variable relative preference functions) (Wagner 1998).

Prior to deciding whether to test relative or absolute preference functions, it is important to consider how individuals of the species in question encounter potential mates in the wild. Absolute preference functions are the preferred method of assessment for species in which females rarely encounter multiple mates simultaneously (MacLaren & Rowland 2006). Conversely, relative preference functions are the preferred method of assessment for species that frequently encounter more than one male at a time (MacLaren & Rowland 2006). However, some species may encounter potential mates both singly and simultaneously. When both designs were tested using female sailfin mollies (*P. latipinna*), MacLaren & Rowland (2006) found that female sailfin mollies exhibit a much stronger preference for larger males presented simultaneously than when presented in sequence.

It is also important to recognize that mating preferences will differ between populations. Geographical variation in mating preferences is reasonably common, and is well documented; examples are known from fish, amphibians, birds and insects (Endler & Houde 1995; Evans & Magurran 1999; Gabor & Ryan 2001; reviewed in Jennions & Petrie 1997). Variation in behavior is expected when there are ecological differences between populations (Endler & Houde 1995; Schwartz & Hendry 2007). Additionally,

geographical variation in mating preferences is expected when two closely related species are sympatric in some regions and not in others (Gerhardt 1994; Sætre et al. 1997; Pfennig 2000; Gabor & Ryan 2001; Higgins & Waughman 2004; Prohl et al. 2006). A strong preference for traits that differentiate between species would be expected as a result of avoidance of mating with heterospecifics or hybrids (Pfennig 1998). Individuals in sympatric populations are expected to emphasize species recognition traits over mate quality traits (Pfennig 1998). Large body size is a mate quality trait that could cause a conflict. When larger size individuals are more likely to be heterospecifics, males and females are expected to evolve in order to emphasize species recognition cues. For example, in the unisexual-bisexual mating system of mollies male *P. latipinna* are sexually parasitized by the gynogenetic species, *P. formosa*. Since *P. formosa* are commonly larger than female *P. latipinna*, male *P. latipinna* encounter a conflict in mate quality recognition and species recognition and more frequently mismatch when *P. formosa* are larger than female *P. latipinna* (Gumm & Gabor 2005).

Although all prior preference function studies have focused on females, preference functions may also describe how the strength of a male's mating response to a female trait varies with the degree of expression of the trait. No known studies have explored male preference functions.

I examined male preference functions in relation to female size in *P. latipinna*. Examining the preference functions of male *P. latipinna* provides more useful information than that from traditional two choice tests, because they reveal the shape of the preference and hence the potential for selection on the preferred trait (Widemo & Sæther 1999). As observed previously in this species, males prefer to mate with and

associate with larger females (Ptacek & Travis 1997; Gabor 1999). Additionally, it is known that male sailfin mollies respond to model fish stimuli and preferentially associate with a conspecific model fish over no model and significantly prefer to associate with a conspecific model over a heterospecific model fish (Gumm et al. 2005). Thus, I presented males with paired model fish of two different sizes. I predicted that as the difference between the female model fish increases in the series of paired stimulus presentations, male association preference for the larger female in the pair would increase. I tested this for males from two populations that were previously tested by Gumm & Gabor (2005) because males from these two populations differed in their preference for larger females. Males from one population, Vincente Guerrero, MX, showed no significant preference but tended to prefer larger heterospecific females, *P. formosa*, than smaller conspecific females while males in the other population, Alfred Bonfil, MX, preferred smaller conspecific females over larger *P. formosa*. Thus I predicted similar results for each population.

### Methods

Trials were run in a 38 l aquarium (54 x 29 x 33 cm) that contained tan gravel and 24 cm of aerated and filtered water maintained at 26 C. A 15 W Sun-Glo full spectrum UV light (General Electric) was placed directly on top of the aquarium and two standard fluorescent lights were placed at an approximately 45 degree angle 5 cm from the back of each side of the aquarium. The lights on the sides lighted up the model fish on either side of the aquarium. The aquarium was placed in a darkroom to block out all other light sources. The back of the tank was covered with black plastic to provide a uniform

background. The sidewalls of the testing chamber (35 cm from the right and left sides of the aquarium) were lined with white poster board in order to provide a uniform background for the transparency models. Tan gravel was placed on the outside left and right of the aquarium (35 cm) to create a similar environment outside the aquarium to the inside. The front of the aquarium was covered with one-way film in order to minimize disturbance of the fishes by outside activity. All fishes were fed 15 min prior to testing.

Markings on the front of the aquarium divided the aquarium into three visual sections. The outer sections (9 cm of each end of the aquarium) were the choice sections while the inner section (36 cm) was the no choice zone and the habituation area. The aquarium was filled with 15.2 cm of water.

I created model test fish using photographs previously created (Gumm et al. 2005). Digital photos using a Nikon Digital CoolPix 950 digital camera were taken of 18 female sailfin mollies from the Vincente Guerrero, MX population. Each female was isolated in a 19 l aquarium (40 x 20.5 x 26.5 cm). To enhance definition of the images, a white paper was placed in the back portion of the aquarium, which provided a background for the photos. Also, the photos were taken in a dark room in order to exclude any outside light, and to provide consistent lighting for all eighteen photos. All photos were taken when fishes had all fins extended. The photos were then prepared for construction of models using Adobe Photoshop 5.5. Each fish image and its corresponding flipped image were printed onto transparencies using a Hewlett Packard 7350 printer. White paper cut into the shape of the body of each fish transparency was placed between the two transparencies in order to enhance a realistic model fish. The resulting two-dimensional models incorporate a much detailed and realistic opaque body

along with transparent fins. For the preference function experiment, three replicates of the 18 model fish were constructed in three sizes: 32 mm, 38 mm, 50 mm. 38 mm is the average size of both male and female fish from these populations.

Male fish were isolated from females for at least 3 d before testing. Male test fish were selected randomly and tested only once in each of the three treatments. I tested males from two populations, Vincente Guerrero, MX (N = 30) and Alfred Bonfil, MX (N = 26). Each male was tested in three different treatments; (1) 32 mm vs. 38 mm (6 mm difference in size), (2) 38 mm vs. 50 mm (12 mm difference in size), (3) 32 mm vs. 50 mm (18 mm difference in size). In this way I was able to evaluate the male preference function for differences in female size in units of 6 mm (Fig. 3.1). The order of the treatments were randomized for each male. Males were tested in all three treatments on the same day with 5 min between treatments. Male test fish were placed in the center of the aquarium under a clear plastic cylinder (12 cm diameter x 15 cm) and allowed to acclimate for 10 min. Model female fish were attached on the right and left of the aquarium and turned on a pulley system rotated with an electric motor. After 10 min, the male was released and time spent by the male in each of the choice sections of the aquarium was recorded for 10 min. At the end of the 10 min, the sides of the models were reversed in order to account for potential side biases. The male was re-acclimated for 10 min with the models rotating. After 10 min, the male was released and time spent by the male in each of the choice sections of the aquarium was recorded for 10 min. The initial left-right position of the transparencies was randomized between tests and each transparency was randomly selected from the 18 models of each class size with no two pairs being re-used. Therefore,

each pair of models was a novel set of stimuli. Once the male had been tested in the three different treatments, I measured male SL.

## Results

The male behaviors most commonly observed during the transparency presentations from both populations included erected large dorsal fin, sigmoid curving of the body (Farr et al. 1986), and flexing of the gonopodium (personal observation). These actions are attributed to male mating behavior. For males from the Vincente Guerrero, MX population, there was no significant difference between male mating responses between different female size treatments (Repeated Measures ANOVA:  $df = 2$ ,  $F = 1.325125$ ,  $p = 0.2737$ ; Fig. 3.2). But males significantly preferred larger females in all three treatments (Table 3.1).

For males from the Alfred Bonfil, MX population, there were significant differences between male mating responses between different female size treatments (Repeated Measures ANOVA:  $df = 2$ ,  $F = 21.951010$ ,  $p < 0.0001$ ; Fig. 3.3). Based on fisher PLSD the 6 mm treatment was significantly different than the 12mm treatment ( $p < 0.0001$ ) and significantly different than the 18 mm treatment ( $p < 0.0001$ ). Males significantly preferred larger females in the 6 mm treatment but showed no significant preference for larger or smaller females in the 12 mm and 18 mm treatment (Table 3.1).

## Discussion

Males from Vicente Guerrero, when presented simultaneously with a series of female model pairs that differed in size significantly preferred females of larger body size. However, there was no significant difference between different female size treatments (i.e. 6 mm, 12 mm, 18 mm difference). Whereas, males from Alfred Bonfil only preferred larger females when the difference in size was the smallest, but showed no preference between large and smaller females when the size difference between the models was larger than 6mm. While both populations are sympatric with the closely related heterospecific females, only males from Alfred Bonfil avoided larger heterospecific females, as in a prior experiment (Gumm & Gabor 2005). Since *P. formosa* are commonly larger than female *P. latipinna*, male *P. latipinna* encounter a conflict in mate quality recognition and species recognition and more frequently mismatch when *P. formosa* are larger than female *P. latipinna* (Gumm & Gabor 2005).

Males from the Vicente Guerrero population showed a general preference for larger females but the strength of preference did not differ as the difference in female size increased. The general preference, of males in this population, for larger females is consistent with previous studies of male preference for large female body size (Travis & Trexler 1987; Ptacek & Travis 1997; Gabor 1999) when presented simultaneously to males. They also are consistent with the results of Gumm & Gabor (2005) who also tested males from the Vicente Guerrero population. They found that males from this population no longer preferred conspecific females and showed a trend to prefer larger heterospecific females (although this was not significant). Thus, males from this population have yet to resolve the conflict between species recognition and mate quality recognition. This can be seen both when they are presented with larger heterospecific

females than conspecific females and when they have a choice between larger and smaller conspecific females as in this experiment.

Males from Alfred Bonfil, when presented simultaneously with a series of female model pairs showed a difference in response between different female size treatments. Males preferred larger females when presented a 6 mm difference between two female models. However, when presented with a 12 mm or 18 mm difference, males showed no significant preference between larger and smaller females. These results are consistent with the results of Gumm & Gabor (2005), where they found that males from this population, that is sympatric with *P. formosa*, did not prefer to mate with larger heterospecifics than smaller sailfin mollies. They suggested that males are avoiding larger females as size may be more an indicator of species identity than mate quality. In this experiment, males only preferred the larger female when she was close to the average size conspecific female (38 mm) in this population. Whereas males lost the preference for larger females when the larger female was greater than the average size conspecific. As such, it is possible that males from this sympatric population avoid mating with heterospecific females by preferentially mating with average size females that are more likely to be conspecifics.

Another explanation for the Alfred Bonfil results is that males may have exhibited an elevated response to females in general and therefore spent relatively less time with the larger female when there was no other male competing for females. The operational sex ratio (OSR; Emlen & Oring 1977) and density may have an influence on the mating tactics of males. In anurans, OSR and density influence alternative mating tactics of males (Lucas & Howard 1995). The distribution and local density of prospective mates

may influence mate choice, and thus, sexual selection in general. For example, when the density of females is lower, there are increased distances between prospective mates. Therefore, increased energetic and time costs are relevant to sampling (Real 1990). This may lead to a reduction in the amount of effort a male is prepared to invest in finding and assessing females (Jennions & Petrie 1997; Widemo & Sæther 1999). Male sailfin mollies may not have encountered large costs in this choice experiment and thus did not encounter a sampling cost and thus did not differentiate between females of only slightly different sizes. It is important to consider distribution and local density of potential mates as they may influence mate choice in future studies.

Because preferences in a simultaneous experiment are inferred by observing an individual's choice between paired stimuli, an overestimation of the strength of preference for a larger mate may occur (Wagner 1998). On the other hand, because male sailfin mollies frequently encounter multiple females at the same time in nature, simultaneous presentation experiments may accurately reflect mate choice in the wild. A simultaneous choice design can also compromise preference comparisons among individuals, populations, and species because of variation in sampling behavior (Wagner 1998). For example, two males with identical preferences for female body size but different sampling strategies would likely produce different preference functions obtained from simultaneous presentation tests. Moreover, one male may sample repeatedly before making a choice, and another male may sample a female only once before making choice. Therefore, their behavior will likely differ in a simultaneous test even if their preference were identical (Wagner 1998). However, female sailfin mollies, exhibit a much stronger preference for larger males presented simultaneously than when presented in sequence

(MacLaren & Rowland 2006). Thus, it is not likely that overstimulation explained the results found in this study.

In conclusion, males from both populations showed some preference for larger females but each population differed in how this was manifest. These results might be related to the fact that males from both of these populations are sympatric with *P. formosa*, a sexual parasite on male sailfin mollies. The outcome of this relationship is that sailfin mollies may be evolving to avoid mating with larger females that are more likely to be these heterospecific sexual parasites. Perhaps the males from the Alfred Bonfil population have progressed further than males from the Vincente Guerrero population. These results are concurrent with those of Gumm & Gabor (2005). It would be interesting to explore preference functions in a population allopatric to *P. formosa* and compare the shapes of the preference function to these sympatric populations. Gumm & Gabor (2005) found that males from allopatric populations preferred to mate with larger heterospecifics over smaller conspecifics, thus these males may have shown an even greater preference for larger females. To my knowledge, this study is the first to explore male preference functions in multiple populations.

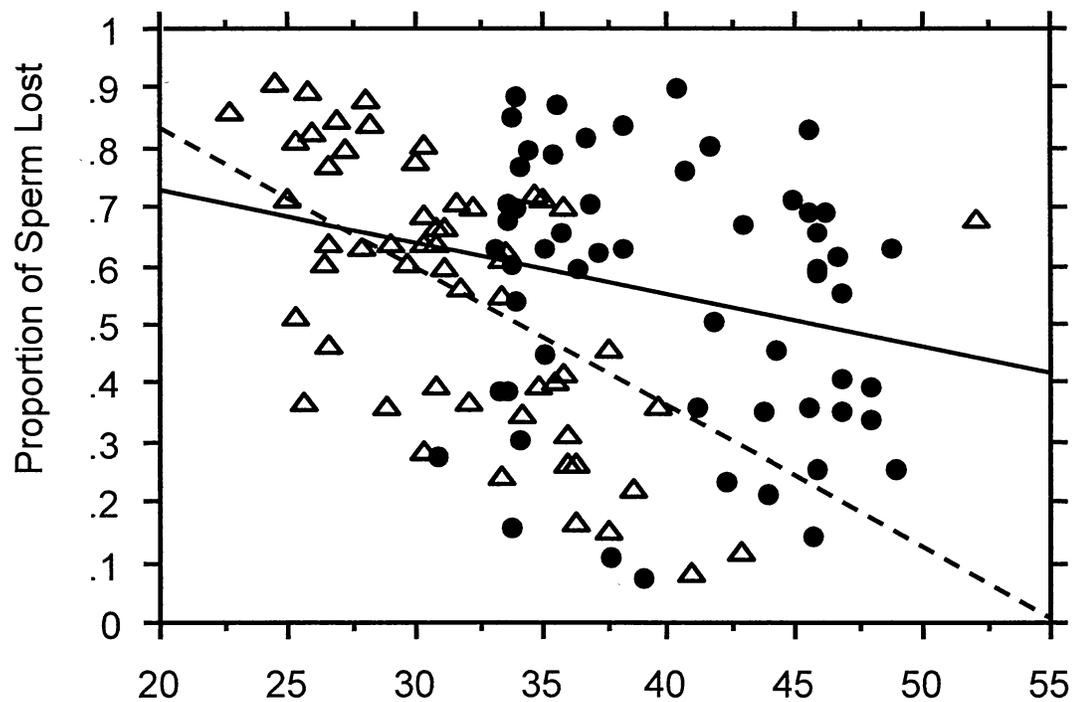
## REFERENCES

- Emlen, S.T. & Oring, L.W.** 1977. Ecology, sexual selection, and evolution of mating systems. *Science*, **197**, 215-223.
- Endler, J.A. & Houde, A.E.** 1995. Geographic variation in female preferences for male traits in *Poecilia reticulata*. *Evolution*, **49**(3), 456-468.
- Evans, J.P. & Magurran, A.E.** 1999. Male mating behaviour and sperm production characteristics under varying sperm competition risk in guppies. *Animal Behaviour*, **58**, 1001-1006.
- Farr, J.A., Travis, J. & Trexler, J.C.** 1986. Behavioral allometry and interdemographic variation in sexual behavior of the sailfin molly, *Poecilia latipinna* (Pisces, Poeciliidae). *Animal Behaviour*, **34**, 497-509.
- Gabor, C.** 1999. Association patterns of sailfin mollies (*Poecilia latipinna*): alternative hypotheses. *Behavioral Ecology and Sociobiology*, **46**, 333-340.
- Gabor, C.R. & Ryan, M.J.** 2001. Geographical variation in reproductive character displacement in mate choice by male sailfin mollies. *Proceedings of the Royal Society of London Series B-Biological Sciences*, **268**, 1063-1070.
- Gerhardt, H.C.** 1994. Reproductive character displacement of female mate choice in the grey treefrog *Hyla chrysoscelis*. *Animal Behaviour*, **47**, 959-969.
- Gumm, J.M. & Gabor, C.R.** 2005. Asexuals looking for sex: conflict between species and mate-quality recognition in sailfin mollies (*Poecilia latipinna*). *Behavioral Ecology and Sociobiology*, **58**, 558-565
- Gumm, J.M., Gonzalez, R., Aspbury, A.S. & Gabor, C.R.** 2005. Do I know you? Species recognition operates within and between sexes in a unisexual-bisexual species complex of mollies. *Ethology*, **112**, 1-10.
- Higgins, L.A. & Waugaman, R.D.** 2004. Sexual selection and variation: a multivariate approach to species-specific calls and preferences. *Animal Behaviour*, **68**, 1139-1153.
- Jennions, M. D. & Petrie, M.** 1997. Variation in mate choice and mating preferences: A review of causes and consequences. *Biological Reviews of the Cambridge Philosophical Society*, **72**, 283-327.
- Lucas, J.R. & Howard, R.D.** 1995. On alternative reproductive tactics in anurans: dynamic games with density and frequency dependence. *The American Naturalist*, **146**, 365-397.

- MacLaren, R.D. & Rowland, W.J.** 2006. Differences in female preference for male body size in *Poecilia latipinna* using simultaneous versus sequential stimulus presentation designs. *Behaviour*, **143**, 273-292.
- Pfennig, K. S.** 1998. The evolution of mate choice and the potential for conflict between species and mate-quality recognition. *Proceedings of the Royal Society of London Series B-Biological Sciences*, **265**, 1743-1748.
- Pfennig, K.S.** 2000. Female spadefoot toads compromise on mate quality to ensure conspecific matings. *Behavioral Ecology*, **11**, 220-227.
- Prohl, H., Koshy, R.A., Mueller, U., Rand, A.S., & Ryan, M.J.** 2006. Geographic variation of genetic and behavioral traits in northern and southern Túngara frogs. *Evolution*, **60(8)**, 1669-1679.
- Ptacek, M.B. & Travis, J.** 1997. Mate choice in the sailfin molly, *Poecilia latipinna*. *Evolution*, **51**, 1217-1231.
- Real, L.** 1990. Search theory and mate choice. I. Models of single-sex discrimination. *The American Naturalist*, **136**, 376-405.
- Sætre, G.-P., Moum, T., Bures, S., Kral, M., Adamjan, M. & Moreno, J.** 1997. Asexually selected character displacement in flycatchers reinforces pre-mating isolation. *Nature*, **387**, 589-592.
- Schwartz, A.K., & Hendry, A.P.** 2007. A test for the parallel co-evolution of male colour and female preference in Trinidadian guppies. *Evolutionary Ecology Research*, **9**, 71-90.
- Travis, J. & Trexler, J.C.** 1987. Regional variation in habitat requirements of the sailfin molly, with special reference to the Florida Keys. *Nongame Wildlife Technical Report Number 3, Florida Game and Fresh Water Fish Commission: Tallahassee, Florida*, **3**, 1-47.
- Wagner, W.E.** 1998. Measuring female mating preferences. *Animal Behaviour*, **55**, 1029-1042.
- Wagner, W.E., Murray, A.M. & Cade, W.H.** 1995. Phenotypic variation in the mating preferences of female field crickets, *Gryllus integer*. *Animal Behaviour*, **49**, 1269-1281.
- Widemo, F. & Sæther, S.A.** 1999. Beauty is in the eye of the beholder: causes and consequences of variation in mating preferences. *Trends in Ecology & Evolution*, **14**, 26-31

TABLE 3.1. Mean  $\pm$  SE time (sec) male sailfin mollies spent with larger and smaller female sailfin mollies across three treatments. AB = Alfred Bonfil; VG = Vicente Guerrero.

Treatment	Population	<i>N</i>	Mean $\pm$ SE (s)	Mean $\pm$ SE (s)	<i>t</i>	<i>P</i>
			larger female	smaller female		
6 mm size difference	VG	30	228.1 $\pm$ 21.40	167.47 $\pm$ 13.73	2.09	0.045
	AB	26	273.62 $\pm$ 22.62	63.81 $\pm$ 6.47	8.77	<0.0001
12 mm size difference	VG	30	278.50 $\pm$ 17.25	171.30 $\pm$ 22.52	3.23	0.003
	AB	26	131.89 $\pm$ 26.76	201.77 $\pm$ 16.00	-1.87	0.0733
18 mm size difference	VG	30	274.97 $\pm$ 18.91	142.10 $\pm$ 19.07	4.02	0.0004
	AB	26	133.15 $\pm$ 29.48	196.54 $\pm$ 25.73	1.288	0.21



## Standard Length (mm)

FIG 2.1 Relationship of the proportion of sperm lost for male *Poecilia latipinna* from Spring Lake, Texas. Proportion of sperm lost was calculated by subtracting 1 from the quotient of Day 3/Day 0. (N=56). The triangles represent male, and the solid points represent each female. The solid line represents the relationship of sperm lost based on female size. The dashed line is the relationship of sperm lost based on the size of females.

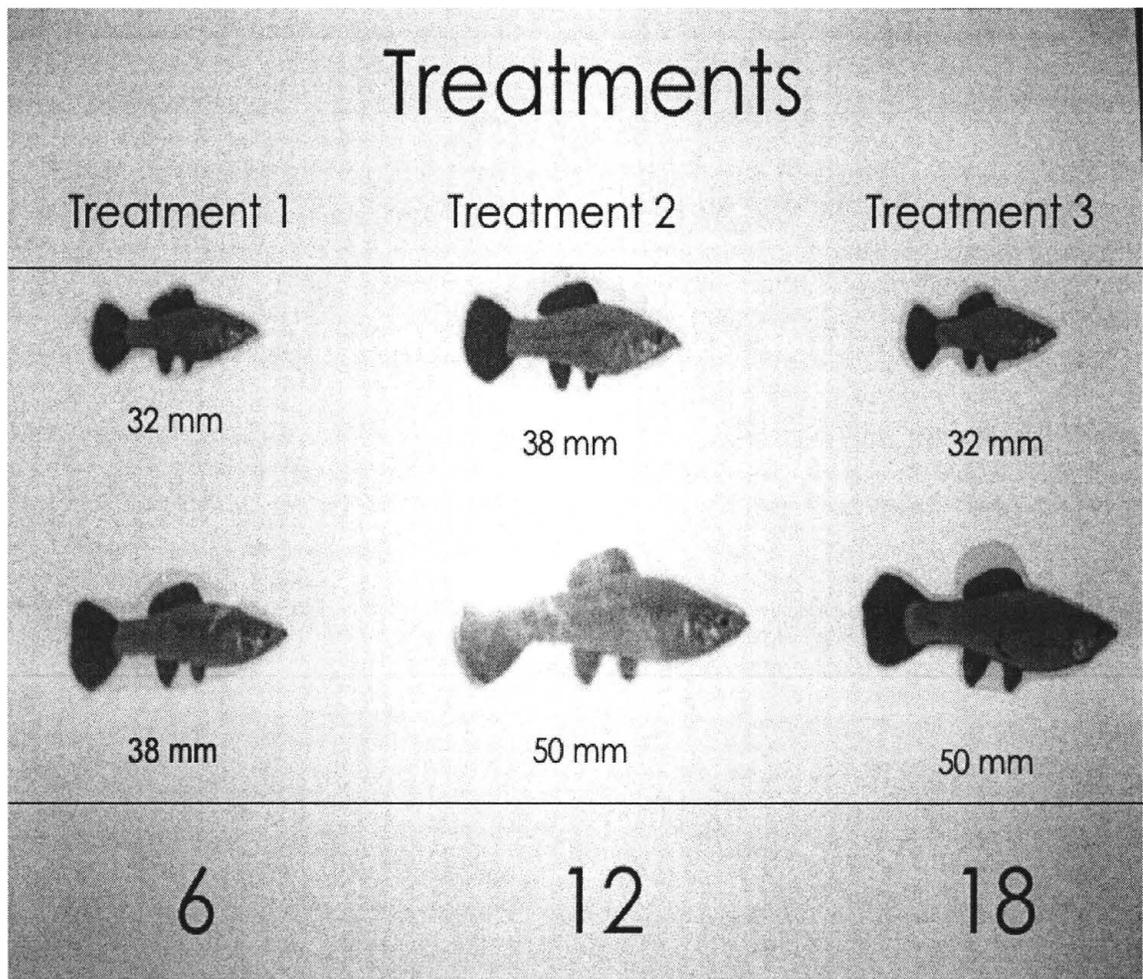


FIG 3.1 Size treatments (6 mm, 12 mm, 18 mm) illustrating female *Poecilia latipinna* models for testing male *Poecilia latipinna* preference functions.

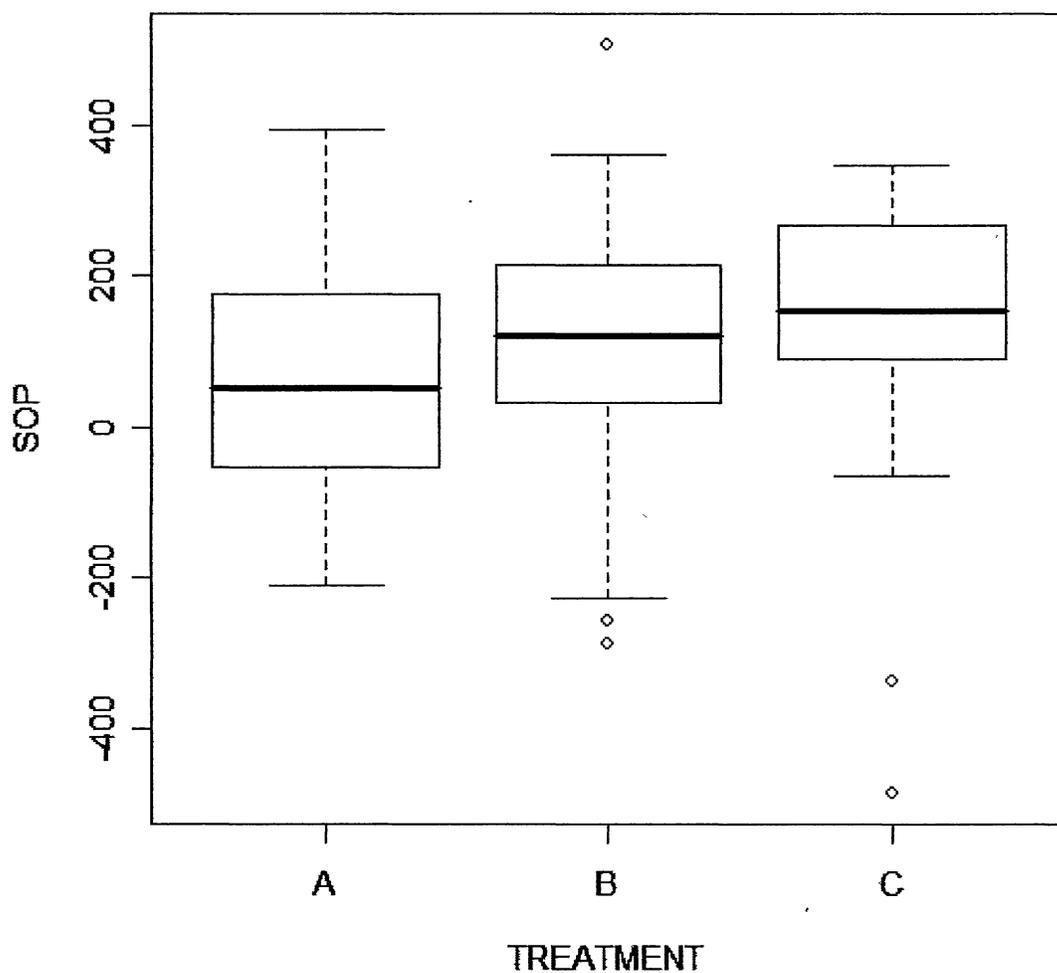


FIG 3.2 Box plots illustrating the strength of preference exhibited by male *Poecilia latipinna* from Vicente Guerrero, MX (N=30). SOP refers to strength of preference. Treatment A represents a 6 mm difference between female models. Treatment B represents a 12 mm difference between female models. Treatment C represents an 18 mm difference between female models. The horizontal line inside the box indicates the median.

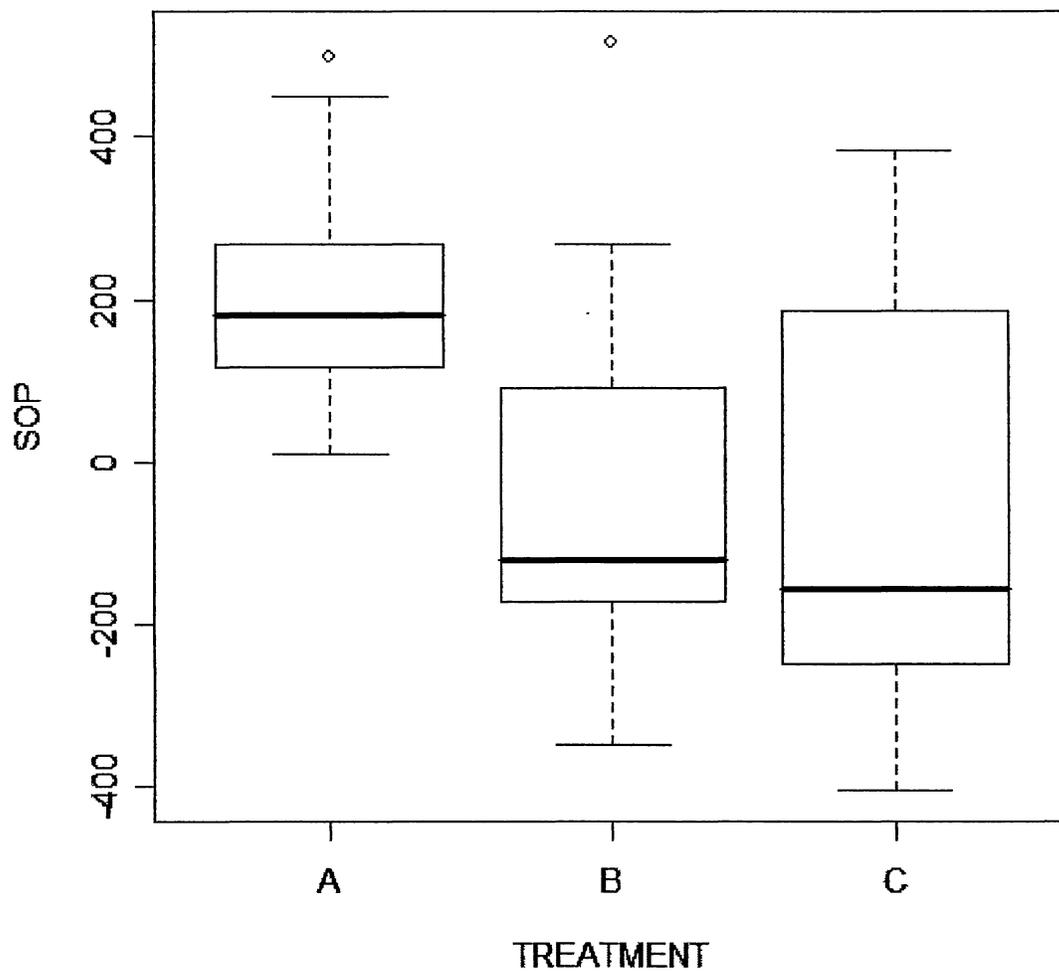


FIG 3.3 Box plots illustrating the strength of preference exhibited by male *Poecilia latipinna* from Alfred Bonfil, MX (N=26). SOP refers to strength of preference. Treatment A represents a 6 mm difference between female models. Treatment B represents a 12 mm difference between female models. Treatment C represents an 18 mm difference between female models. The horizontal line inside the box indicates the median.

## VITA

Rosie Gonzalez was born in McAllen, Texas, on September 24, 1980, the daughter of Jose Antonio Gonzalez and Maria Elena Gonzalez. After completing her work at Nikki Rowe High School, McAllen, Texas, in 1999, she entered the University of Texas Pan American, Edinburg, Texas. After completing basic coursework, she transferred to Texas State University-San Marcos. At Texas State, she received the degree of Bachelor of Science in Microbiology in 2004. She began the Master of Science program in Biology at Texas State in August 2004. While at Texas State, Rosie was employed as a research assistant and taught labs in Organismal Biology.

Permanent address: 3000 Upas

McAllen, Texas 78501

This thesis was typed by Rosie Gonzalez.