

# Vertical bars on male *Xiphophorus multilineatus*: a signal that deters rival males and attracts females

Molly R. Morris, Michelle Mussel, and Michael J. Ryan

Department of Zoology, University of Texas, Austin, TX 78712, USA

We examined the function of the vertical bar pattern on male swordtails (*Xiphophorus multilineatus*) as a signal in both male–male competition and female choice. This pattern had previously been described as an aggressive signal because males intensified the bars during male–male encounters in the laboratory. Our field observations supported this observation and also showed that bars intensified when males courted females. The intensity of bars was correlated with access to females in the field. Within the size range of males that have bars, however, neither bar number nor male size appeared to influence access to females. We used freeze-branding to remove the bars from males in the laboratory so that we could control for characters correlated with bar intensity, and tested males and females separately so that we could separate the influence of these two components of sexual selection. We compared the responses of males and females to males that had their bars removed and control males freeze-branded between the bars. Test males responded more aggressively to males without bars as compared to control males. In addition, females showed a preference for control males over males that had their bars removed. These results suggest that the bars may function as a signal that deters rival males and attracts females. *Key words*: aggressive signal, courtship, dichromatism, female choice, male–male competition, Poeciliidae, sexual selection, vertical bars, *Xiphophorus multilineatus*. [*Behav Ecol* 6:274–279 (1995)]

The significance of intrapopulation variation in color patterns in fish has been examined extensively (Barlow, 1983; deMartini, 1985; Endler, 1983; Long and Houde, 1989; McLennan and McPhail, 1989; Semler, 1971; Stepien et al., 1988). Many species of teleost fish can rapidly modify dichromatisms and often do so during social interactions (deMartini, 1985). Several studies have examined changes in coloration and pigment patterns as signals of aggression, and it has also been suggested that some of these signals play a role in female choice (Heiligenberg, 1976; Kingston, 1980; Martin and Hengstebeck 1981; Neil, 1984; Thresher, 1984). One of the difficulties in determining whether these color and pigment changes function as signals that deter rival males and attract females is that they are often correlated with other male traits. Few studies have examined the function of these color or pigment patterns through experimental manipulations that control for correlated characters (although see Kodric-Brown 1989; Semler, 1971).

Through field observations and laboratory manipulations, we examined the function of a pigment pattern that can be facultatively expressed on males in the swordtail fish *Xiphophorus multilineatus*. *X. multilineatus* is a member of the northern swordtail clade (Rauchenberger et al., 1991). The vertical bars found on *X. multilineatus* and several other species in the genus *Xiphophorus*, as well as in other genera of poeciliid fishes (e.g., *Heterandria*, *Phallichthys*), are composed of hundreds of closely associated melanophores (Gordon, 1931). The intensity of the bars increases dramatically during male–male interactions in several species of *Xiphophorus* (Franck, 1964; Zimmerer and Kallman, 1988). Male size is highly variable in *X. multilineatus* due to four alleles at the *P*

locus on the Y chromosome, which control the age at which males reach sexual maturity and cease growth (Kallman, 1984, 1989). The three largest size classes of males have vertical bars, while females and males from the smallest size class do not. Zimmerer and Kallman (1988) demonstrated that the bars have a polygenic basis with one sex-linked factor having a major effect. The bar genes appear to influence both bar number and bar intensity. Bar number and male size are positively correlated in the larger males, and the lack of bars in small males is due to a suppressor gene (Zimmerer and Kallman, 1988).

The goals of this study were to determine (1) whether changes in the intensity of the bars in the field occurred in the same situations as changes in the laboratory, (2) whether variation in the intensity of the bars or in the number of bars influenced access to females, and (3) through laboratory manipulations whether the bars functioned as a signal that would deter rival males and attract females.

## METHODS

### Field observations

We made field observations with snorkel and mask in March 1990 in the Rio Coy south of Ciudad Valles, San Luis Potosi, Mexico (18° N, 89° W). All observations were made between 0900 and 1400 h.

We used the same methods as described in Morris et al. (1992) with *X. nigrensis*. Before behavioral observations commenced, we counted the number of females and males within a 1-m<sup>3</sup> area. The river bottom dropped off within 3 m of the bank, and most of the fish stayed in this shallow portion of the river (approximately 1.5 m deep). A rope marked with tape every 1 m was tied parallel to the bank of the river to aid in locating quadrats that had been sampled. We made the assumption that when we moved to a new area we were observing different males. Results from a previous study with *X. nigrensis*, the sister species to *X. multilineatus*, in which

M. R. Morris is now at the Department of Natural Sciences, University of Maryland, Eastern Shore Campus, Princess Anne, MD 21853, USA.

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marked males returned to within 1 m of the same site in subsequent days, support this assumption (Morris et al., in press).

Males used in the focal observations were observed for several minutes prior to the beginning of the observation period to determine if they were likely to remain in the area of the quadrat. We made focal observations on 22 males. Each male observed was classified as large (>32 mm) or intermediate (>25 mm but <32 mm) by holding a small ruler, clearly marked at 25 mm and 32 mm, up close to the fish. We did not catch the fish to measure them, and therefore these estimates are used only as relative measurements of male size and not indicators of genotype. We counted the number of bars on 13 of the 22 focal males. When bar number varied from side to side, an average number of bars for the two sides was recorded.

We made focal observations for 10 min or until the focal male was lost from view. When observers were in position (approximately 1 m from the quadrat), their presence did not appear to disturb the behavior of the fish. Males courted females and chased males within 10 cm of the observers. Not all behaviors recorded occurred within the original 1-m<sup>3</sup> quadrat, but all did occur within a 0.5-m range of the quadrat. Focal periods averaged 7.98 min and ranged from 3 to 10 min. The difference between the duration of the focal observations for the large ( $\bar{x}$  = 8.55 min, SD = 2.22) and intermediate size classes ( $\bar{x}$  = 7.54 min; SD = 2.63) was not statistically significant ( $t$  = 0.98;  $df$  = 21;  $p$  = .34). During focal observations, we recorded the number of times each male displayed to females (glided back and forth either to the side or in front of a female; Ryan and Causey, 1989). We also recorded the number of times a male was chased by another male (flees), the number of times he chased other males (chases), and the relative size of the other male involved in a chase when possible. We noted changes in bar intensity in many cases. Based on observations in the field and the laboratory, these changes were translated into a qualitative index of bar intensity with four categories: completely faded (not visible), faded, intermediate, and dark.

We determined whether male size influenced the rate of interactions with other males (chases/min or flees/min) or interactions with females (courts/min) with one-way analyses of variance. We also examined the relationships between bar number and interactions with males and females with linear regressions to determine whether bar number influenced these interactions in a manner similar to male size.

To determine whether bar number or bar intensity might influence a male's mating success, we examined the relationships between both of these components of the bars and access to females. We measured access to females as the number of females on the 1-m<sup>3</sup> quadrat prior to the focal observations divided by the number of males on the quadrat. The maximum intensity of the bars does not vary among individuals (Zimmerer and Kallman, 1988). However, bar intensity changed depending on whether males were chasing males, courting females (bars intensified), or fleeing (bars faded, see Results below). The duration of a chase was very similar to the duration of a court (male glides back and forth in front of a female). We subtracted the number of times a male fled (bars fade) from the total number of times he chased another male or courted a female (bars intensified) to get an index of the relative time the bars were intensified. We tested relationships for significance with linear regressions.

### Laboratory experiments

We determined the function of the bars in *X. multilineatus* by testing the responses of both males and females to control males with bars as compared to males with bars removed. We

removed bars from males by freeze-branding (Hert, 1986; Raleigh et al., 1973). Twelve pairs of males were matched for size (within 0.5 mm) and anesthetized with MS222. One male from each pair was freeze-branded on the bars (bars removed) and one between the bars (control). Given a difference in size in some cases (within 0.5 mm), the larger of the two males was chosen to be the bars removed male. Pigmentation in the branded areas faded after 2–3 days, and the behavior of the males did not appear to be affected by the manipulation. Freeze-branding in this manner produced no other visible marks. During contests, the intensity of the bars on control males was either intermediate or dark. The bars removed males simulated males with a bar intensity of completely faded. Freeze-branded males (bars removed and controls) are referred to as the experimental males.

The fight intensity of test males in contests with control males compared to their fight intensity in contests with bars removed males the same size as the control males was used as an indicator of male response to bars. Test males were 0.5–4.5 mm smaller than the experimental males, and only one test male was tested with each pair of experimental males. Experimental and test males were kept isolated in individual 2.5-l tanks throughout the testing period. Tests were conducted in a 45 × 60 × 41 cm tank with gravel on the bottom and black plastic covering the ends and back side. We placed one test male and one experimental male on either side of an opaque partition that divided the test tank into two equal parts. After 24 h, we removed the partition and recorded the number of bites delivered by each male, the length of the contest, and the winner of the contest. Contest length was measured as time from when the first male approached to the time when one male became dominant. A male was considered dominant when the other male lowered his dorsal fin and retreated when approached. We used bites/min as a measure of fight intensity. The following day, the same test male was tested against the other experimental male from the pair. We tested half of the test males against the bars removed males first and half against the control males first to control for any influence of prior fighting experience (Franck and Ribowski, 1987). We compared the fight intensity of test males in contests with control males to their fight intensity in contests with bars removed males using a Wilcoxon matched-pairs signed-rank test. We also examined relationships between fight intensity and size difference between males to determine whether the relative size of the opponents influenced how the males responded to the bars.

We tested the responses of females with the same 12 pairs of experimental males used to test the responses of males. Prior to testing, males and females were separated for at least 1 week. The test aquarium measured 45 × 90 × 41 cm and was divided into five equal sections. Plexiglass separated the sections at each end from the three central sections. One male from each pair was placed on either end of the test tank, and a female was placed in an opaque cylinder in the center of the test tank. All three fish were allowed to acclimate for 10 min. After removing the opaque cylinder, we recorded for 20 min the time the female spent in each of the sections near the males. The males were then switched end to end and the entire test repeated to control for side-bias. We compared the total time females spent with control males and with bars removed males using a Wilcoxon matched-pairs signed-rank test.

## RESULTS

### Field observations

The average number of females per m<sup>3</sup> quadrat was 8.0 (SD = 5.29,  $N$  = 22), while the average number of males was 3.18

**Table 1**  
Rates of behaviors in the field for large males compared to intermediate males

Behavior	Large males		Intermediate males		df	ANOVA	
	$\bar{x}$	SD	$\bar{x}$	SD		$F$	$p$
Flees/min	0.13	0.14	0.98	0.58	1,21	20.1	.0002
Chases/min	1.93	0.87	0.79	0.67	1,21	12.5	.002
Courts/min	2.18	1.29	1.97	1.29	1,21	0.14	.71

(SD = 1.53,  $N = 22$ ). Number of females was positively correlated with number of males per quadrat ( $r^2 = .51$ ,  $F = 21.1$ ,  $N = 22$ ,  $p = .002$ ). Most interactions between males consisted of chases, but we observed three parallel swims (Franck, 1964) between males that appeared of equal size. Of the 32 chases in which the relative size of the two males was noted, the larger male chased the smaller male in all interactions except for five interactions involving one particular male. Size explained a significant amount of the variation in the rate of chasing and the rate of fleeing (Table 1). Male size class did not explain a significant amount of the variation in interaction rates with females (Table 1).

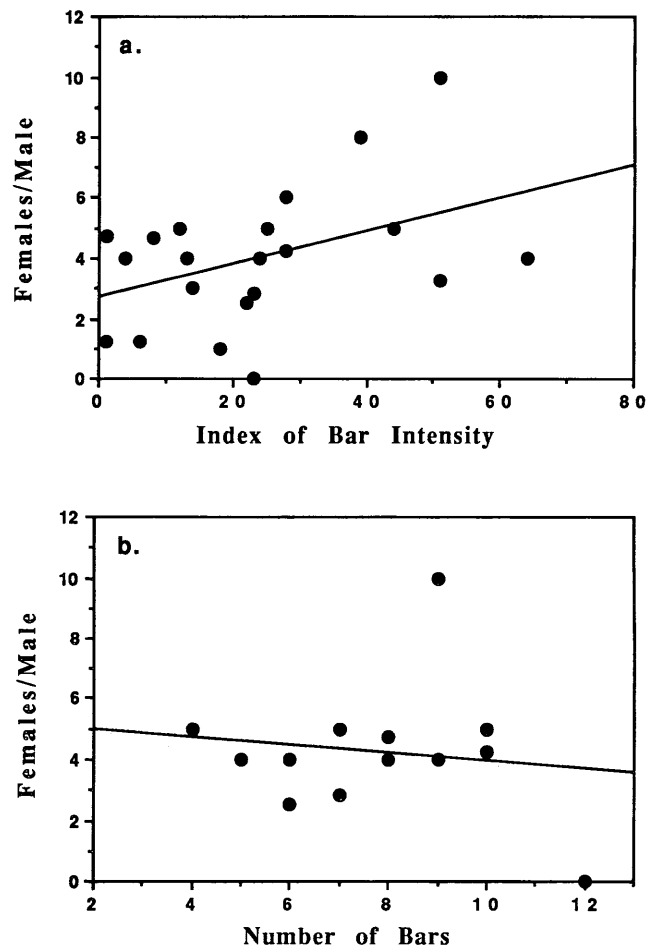
Large males had significantly more bars than intermediates (large  $\bar{x} = 9.8$ , SD = 1.48,  $N = 5$ ; intermediate  $\bar{x} = 5.78$ , SD = 2.64,  $N = 8$ ; Mann-Whitney  $U$  test,  $p = .006$ ), and number of bars showed a pattern similar to male size in relation to male competition and female choice. A significant negative relationship existed between the number of bars and the rate of fleeing (Kendall Correlation Coefficient,  $Z = -2.83$ ,  $N = 13$ ,  $p = .005$ ). The relationship between number of bars and the rate of chasing showed a positive trend, but was not statistically significant (Kendall Correlation Coefficient,  $Z = 1.7$ ,  $N = 13$ ,  $p = .09$ ). The one male that chased males larger than himself had 12 bars, which was the highest number of bars counted by Zimmerer and Kallman (1988) for this species and the highest for the males we observed. No significant relationship, however, existed between bar number and rate of courtship (Kendall Correlation Coefficient,  $Z = -1.27$ ,  $N = 13$ ,  $p = .20$ ).

Males varied the intensity of their bars depending on their interactions; however, the bars never faded completely on males in the field. In all cases where we observed changes in the intensity of the bars, they occurred during an interaction; the bars on males that were fleeing faded ( $N = 11$  males), and the bars on males chasing ( $N = 14$ ) or courting females ( $N = 12$ ) intensified to dark. Males in the vicinity of females but not courting or interacting with other males had intermediate bar intensity.

Our index for the relative time the bars were intensified (number of chases + number of courts - number of flees) was positively correlated with access to females (Figure 1a). Large males did not have significantly greater access to females (5.02 females/male, SD = 2.84) than did intermediate males (3.97 females/male, SD = 2.2; Mann-Whitney  $U$  test,  $df = 21$ ,  $p = .20$ ). Likewise, number of bars was not significantly correlated with access to females (Figure 1b).

### Laboratory experiments

The intensity index for the bars on males prior to staged male-male interactions ranged from completely faded (not visible) to intermediate. The bars on all control and test males intensified to dark once the contest began. After dominance had been established, the bars on the subordinate male faded,

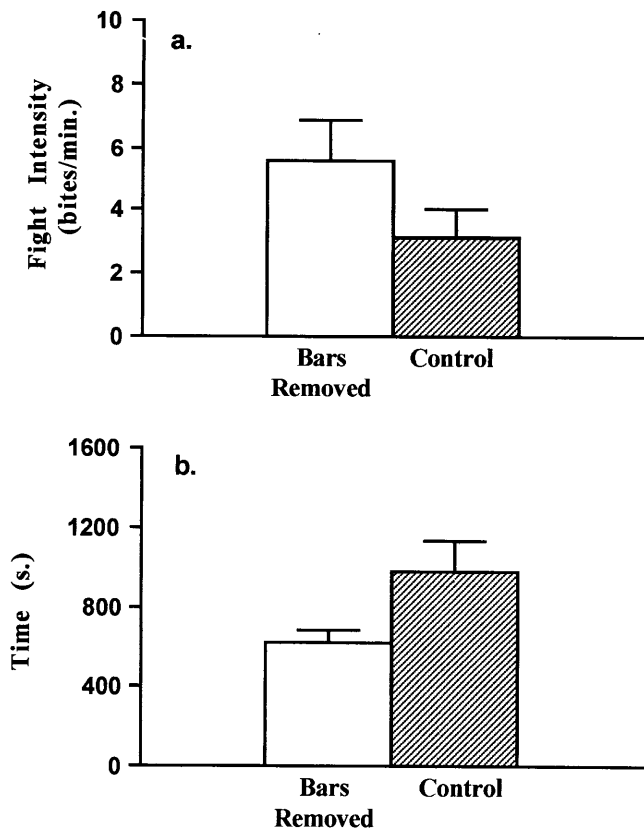


**Figure 1**

Relationships between two components of the vertical body bars on male *X. multilineatus* and access to females (females/male/1 m<sup>3</sup>) in the field. (a) Index of relative time the bars were intensified (number of chases + number of courts - number of flees) was significantly correlated with access to females ( $r^2 = .19$ ,  $N = 22$ ,  $p = .04$ ). (b) Number of bars was not significantly correlated with access to females ( $r^2 = .01$ ,  $N = 13$ ,  $p = .78$ ).

often completely, while the bars on the dominate male remained at an intensity of dark to intermediate. Test males responded more aggressively to the bars removed males compared to the control males (Figure 2a). The intensity of the bars on the control males in the female choice tests was intermediate to dark. Females spent significantly more time with the control males with bars than with the bars removed males without bars (Figure 2b).

In interactions between males, the relationship between fight intensity and difference in size was negative in contests where the larger male was the control male, but this relationship was not statistically significant (Figure 3a). In contests where the larger male had his bars removed, however, a significant positive relationship existed between fight intensity and difference in size (Figure 3a). By subtracting the fight intensity of contests with control males from fight intensity of contests with bars removed males for each test male, we can effectively remove some of the variation due to differences between test males. Differences in fight intensities between contests with bars removed males and contests with control males were significantly positively correlated with difference in size (Figure 3b).

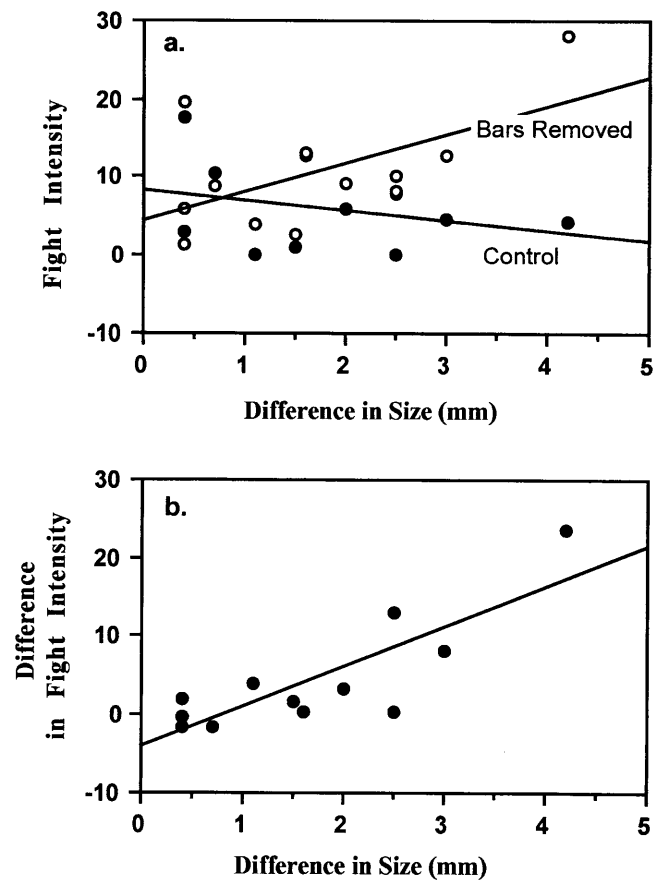


**Figure 2**  
Mean responses of males (a) and females (b) to the same pairs of experimental males. One of the experimental males from each pair had bars removed (white bars), and one male was the control with bars (hatched bars). Standard errors are shown. (a) In controlled laboratory contests, test males responded significantly more aggressively (bites/min) to males without bars as compared to males with bars (Wilcoxon matched-pairs sign-ranked test,  $Z = 2.4$ ,  $N = 12$ ,  $p < .05$ ). (b) Females responded by spending significantly more time with control males as compared to bars removed males (Wilcoxon matched-pairs sign-ranked test,  $Z = 2.1$ ,  $N = 12$ ,  $p < .05$ ).

## DISCUSSION

Field observations supported previous laboratory observations (Franck, 1964; Zimmerman and Kallman, 1988) that the bars on *X. multilineatus* males intensified in aggressive interactions and faded when a male was submissive. The range of male-male interactions and intensity of the bars, however, was greater in the laboratory than in the field. Aggressive interactions in the field did not include biting but consisted almost exclusively of chasing and fleeing, and the bars never faded completely on males in the field as they did in the laboratory. In addition, we observed that the bars on males intensified during courtship, suggesting that the bars may also function in female choice. We used the observation that the intensity of the bars faded when a male was fleeing and intensified when males were courting and chasing to arrive at a relative index of the time a male's bars were intensified. Since this index of bar intensity was positively correlated with access to females and bar number was not correlated with access to females, we suggest that among the males that have bars, bar intensity may play a more important role than bar number in gaining access to females.

Correlations between aspects of the bars and access to females could result from factors correlated with the bars (e.g.,



**Figure 3**  
Relationships between the fight intensity (bites/min) and size difference between opponents in staged laboratory contests. (a) A negative relationship existed in contests in which the larger male was a control male with bars (filled circles,  $r^2 = .10$ ,  $N = 12$ ,  $p = .20$ ) and a positive relationship in contests in which the larger male was a bars removed male (open circles,  $r^2 = .36$ ,  $N = 12$ ,  $p = .04$ ). (b) The difference in the responses of smaller test males to males that had their bars removed compared to control males with bars was positively correlated with the difference in size between opponents ( $r^2 = .69$ ,  $N = 12$ ,  $p = .0008$ ).

male size, condition, behavior) rather than the bars themselves. In addition, since access to females in the field was measured as the number of females per male per quadrat, access could result from females preferring to associate with certain males or from certain males being better at excluding rival males. We controlled for male size and other characters that might be correlated with the bars by manipulating the bars in the laboratory. We then tested males and females separately so that we could distinguish the influences of female choice and male-male competition. Our laboratory results demonstrated that control males with bars elicited less aggressive behavior from the smaller test males than did males of the same size as control males with bars removed. In addition, females preferred to associate with the control males with bars compared to those with no bars. Therefore, when the bars are expressed at some level of intensity (or not completely faded), they may function to deter rival males and attract females in *X. multilineatus*.

Our results suggest that the presence of the bars should reduce the aggressive behavior of rival males and attract females and that increasing the intensity of the bars should increase access to females. Therefore, why don't all of the males have bars expressed at their highest intensity all of the time?

The facultative nature of the bars in the larger males, and the complete suppression of the bars in the small males, strongly suggest a cost to the bars in *X. multilineatus*. We have shown that having dark bars should enhance rather than reduce a male's mating success. Costs could result from predation pressures or social interactions. If vertical bars increase conspicuousness, they could increase risk of predation. In addition, there may be a cost to the bars in aggressive interactions due to injury or unnecessary loss of energy for individuals that continue and/or escalate a contest in which they cannot win (Maynard Smith and Harper, 1988; Rohwer and Ewald, 1981). The presence of the suppressor gene that reduces bar expression in small *X. multilineatus* males strongly suggests that the cost/benefit ratio for the bars is size dependent.

In a previous study of unmanipulated *X. multilineatus* males, size was a significant determinant of fight outcome, and fight intensity decreased as size difference between contestants increased, indicating that males assessed the size of their opponents (Morris et al., in press). As expected, the relationship between fight intensity and difference in size in contests with control males was also negative, although not statistically significant. The lack of significance could result from the smaller range of size differences examined in the present study compared to the study of unmanipulated males. What was unexpected, however, was the significant positive relationship between fight intensity and difference in size in contests where the larger male had his bars removed. When we remove variation in fight intensity due to differences between test males by examining the difference in fight intensity between contests with bars removed males and contests with control males, we get an even clearer indication that fight intensity in relation to the difference in size between opponents was changed by removing the bars. Further tests are needed to determine whether this change results from the behavior of the smaller test males, the larger experimental males, or both. One could compare the behavior of test males in contests with males without bars of various sizes, or the behavior of one opponent could be held constant in some manner. While it is difficult to imagine why a smaller test male would fight more intensely with a larger male without bars than with a smaller male without bars, it seems likely that larger males might increase their fight intensity as their opponent's size decreases in relation to their own.

Because the number of vertical body bars on *X. multilineatus* males is correlated with size (Zimmerer and Kallman, 1988) and size determines fight outcome, male *X. multilineatus* could use the number of bars to assess male size. In our field study, number of bars was significantly correlated with the number of times a male fled. However, this relationship could result from assessment of body size and not assessment of bar number. The male we observed with the greatest number of bars is the only male observed chasing males larger than himself, suggesting that males may sometimes rely on bar number over actual size in their assessment of an opponent. Interactions with females, on the other hand, were not greater for males with more bars or larger size. Females may not differentiate between males as long as they use courtship behavior (only the smallest size class of males do not court). This is apparently the case in the closely related species *X. nigrensis*. In *X. nigrensis*, male-male competition was sensitive to differences in intermediate and large males (Morris et al., 1992), while female preference tests indicated that females preferred large courting males over small noncourting males, but showed no preference for large courting males over intermediate courting males (Ryan et al., 1990). Another possibility is that females use bar number to assess males, but only within the context of another variable that we could not control in the field (e.g., male size, bar intensity, or bar symme-

try). Experiments currently underway examine the relationships between number of bars, male size, and female preference by manipulating the number of bars on males in the laboratory.

In summary, within the size classes of males that have bars in *X. multilineatus*, the component of the bars an individual can rapidly modify (bar intensity) appeared to influence access to females in the field to a greater extent than the component correlated with body size (bar number). Laboratory results only pertain to extreme differences in bar intensity, but they also suggest that expressing the bars at some intensity (not completely faded) should increase a male's access to females due to both deterring rival males and attracting females. This system provides an interesting opportunity to examine the evolution of a signal that can be rapidly modified and yet functions as a signal in both components of sexual selection.

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## REFERENCES

- Barlow GW, 1983. Do gold midas cichlid fish win fights because of their color, or because they lack normal coloration? *Behav Ecol Sociobiol* 13:197-204.
- deMartini EE, 1985. Social behavior and coloration changes in painted greenling, *Oxylebius pictus* (Pisces: Hexagrammidae). *Copeia* 1985:966-975.
- Endler JA, 1983. Natural and sexual selection on color patterns in poeciliid fishes. *Environ Biol Fishes* 9:173-364.
- Franck D, 1964. Vergleichende Verhaltensstudien an lebendgebärenden Zahnkarpfen der Gattung *Xiphophorus*. *Zool J Physiol Bd* 71: 117-170.
- Franck D, Ribowski A, 1987. Influence of prior agonistic experience on aggression measures in the swordtail (*Xiphophorus helleri*). *Behaviour* 103:217-240.
- Gordon M, 1931. Morphology of the heritable color patterns in Mexican killifish, *Platypoecilus*. *Am J Cancer* 15:732-787.
- Heiligenberg W, 1976. A probabilistic approach to the study of motivation. In: *Simpler networks and behavior* (Fentress J, ed). Sunderland, Massachusetts: Sinauer; 301-313.
- Hert E, 1986. Freeze branding in fish, a method for eliminating color patterns at the skin surface. *Ethology* 72:165-167.
- Kallman KD, 1984. A new look at sex determination in poeciliid fishes. In: *Evolutionary genetics of fishes* (Turner BJ, ed). New York: Plenum; 95-171.
- Kallman KD, 1989. Genetic control of size at maturity in *Xiphophorus*. In: *Ecology and evolution of live bearing fishes (Poeciliidae)* (Snelson FF, Meffe GK, eds). Englewood Cliffs, New Jersey: Prentice Hall; 163-184.
- Kingston DI, 1980. Eye color changes during aggressive displays in the godeid fish. *Copeia* 1980:169-171.
- Kodric-Brown A, 1989. Dietary carotenoids and male mating success in the guppy: an environmental component to female choice. *Behav Ecol Sociobiol* 25:393-401.
- Long KD, Houde AE, 1989. Orange spots as a visual cue for female mate choice in the guppy (*Poecilia reticulata*). *Ethology* 82:316-324.
- Martin FD, Hengstebeck MF, 1981. Eye color and aggression in juvenile guppies *Poecilia reticulata*. *Anim Behav* 29:325-331.
- Maynard Smith J, Harper DGC, 1988. The evolution of aggression—can selection generate variability? *Phil Trans R Soc B* 319:557-570.
- McLennan DA, McPhail JD, 1989. Experimental investigations of the evolutionary significance of sexually dimorphic nuptial coloration in *Gasterosteus aculeatus* (L.): temporal changes in the structure of the male mosaic signal. *Can J Zool* 67:1767-1777.
- Morris MR, Batra P, Ryan MJ, 1992. Male-male competition and access to females in the swordtail *Xiphophorus*. *Copeia* 1992:980-986.

- Morris MR, Gass L, Ryan MJ, in press. Assessment and individual recognition of opponents in the swordtails *Xiphophorus nigrensis* and *X. multilineatus*. Behav Ecol Sociobiol.
- Neil SJ, 1984. Field studies of the behavioral ecology and agonistic behavior of *Cichlasoma meeki* (Pisces: Cichlidae). Environ Biol Fishes 10:59–68.
- Raleigh RF, McLearen JB, Groff DC, 1973. Effects of topical location, branding techniques, and changes in hue on recognition of cold brands in Centrarchid and Salmonid fish. Trans Am Fish Soc 102: 637–641.
- Rauchenberger M, Kallman KD, Morizot DC, 1991. Monophyly and geography of the Rio Panuco basin swordtails (Genus *Xiphophorus*) with descriptions of four new species. Am Mus Novit 2975:1–41.
- Rohwer S, Ewald PW, 1981. The cost of dominance and advantage of subordination in a badge signalling system. Evolution 35:441–454.
- Ryan MJ, Causey BA, 1989. “Alternative” mating behavior in the swordtails *Xiphophorus nigrensis* and *Xiphophorus pygmaeus* (Pisces: Poeciliidae). Behav Ecol Sociobiol 24:341–348.
- Ryan MJ, Hews DK, Wagner WE Jr, 1990. Sexual selection on alleles that determine body size in the swordtail *Xiphophorus nigrensis*. Behav Ecol Sociobiol 26:231–237.
- Semler DE, 1971. Some aspects of adaptation in a polymorphism for breeding colors in the threespine stickleback (*Gasterosteus aculeatus*). J Zool Lond 165:291–302.
- Stepien CA, Glatke M, Fink MK, 1988. Regulation and significance of color patterns of the spotted kelpfish, *Gibbonsia elegans* Cooper, 1864 (Blennioidei: Clinidae). Copeia 1988:7–15.
- Thresher RE, 1984. Reproduction in reef fishes. Neptune City, New Jersey: TFH.
- Zimmerer EJ, Kallman KD, 1988. The inheritance of vertical barring (aggression and appeasement signals) in pygmy swordtail *Xiphophorus nigrensis* (Poeciliidae, Teleostei). Copeia 1988:299–307.