

# Shoaling in juvenile guppies: The effects of body size and shoal size

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## Abstract

While factors affecting shoal mate choice have been examined extensively in adult guppies (*Poecilia reticulata*), few studies have focused on the shoaling behavior of juveniles. In this study, juvenile guppies were tested for their ability to shoal as well as their response to shoal mates of different body size and to shoals with different numbers of individuals. In dichotomous choice tests, 10-day-old guppies (mean body length = 8.83 mm), 30-day-old guppies (13.17 mm) and 50-day-old guppies (18.6 mm) were given the opportunity to swim near shoals of five fish or an empty chamber. In most cases, the juvenile fish demonstrated shoaling behavior, swimming near a group of fish rather than an empty chamber, regardless of the age of the stimulus shoal. When presented with two shoals, one of similar age and body size and one of dissimilar age and body size, only the 50-day-old guppies showed a significant preference for the age-matched shoal. Similarly, when choosing between a large shoal and a small shoal, only the 50-day-old guppies spent significantly more time near the larger shoal. Thus, while juveniles at each age shoaled, only 50-day-old fish demonstrated the shoal mate discrimination seen in adult fish.

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## 1. Introduction

Shoaling behavior provides distinct benefits for fish, including enhanced acquisition of food and potential mates, as well as protection from predators (Pitcher, 1983; Krause and Ruxton, 2002). Numerous studies have shown that individuals from many fish species actively discriminate between potential shoal mates in ways that will enhance these benefits. In banded killifish, (*Fundulus diaphnus*), individuals choose to shoal with conspecifics over non-conspecifics (Krause and Godin, 1994), which may increase the potential to find mates. In guppies (*Poecilia reticulata*), individuals choose large shoals over small shoals, which may facilitate food acquisition (Day et al., 2001) and increase predator protection through numerical dilution (Krause and Godin, 1995) and predator inspection (Magurran and Seghers, 1994). Finally, individuals from many species choose shoal mates with similar phenotypic characteristics to themselves, enhancing the predator protection benefits of the “confusion effect,” which is thought to occur when predators have difficulty targeting a single prey fish within a group of phenotypically similar individuals (Godin, 1986; Landeau and

Terborgh, 1986). Inclusion of a phenotypically dissimilar individual in a shoal results in the ‘oddy effect’ causing the individual to stand out, leading to more successful predator attacks (Theodorakis, 1989; Ohguchi, 1981). For this reason, fish typically associate with phenotypically similar shoal mates. Examples include mollies (*Poecilia latipinna*) that choose shoal mates of similar coloration (McRobert and Bradner, 1998), guppies that choose shoal mates of similar body size (Croft et al., 2003), and even banded killifish that choose shoal mates with similar spotting patterns on their bodies caused by parasites (Krause and Godin, 1996).

The shoaling behavior of the common guppy (*Poecilia reticulata*) has been analyzed in numerous studies. It has been shown that female guppies actively assort into shoals according to body size and prefer to shoal with familiar individuals (Griffiths and Magurran, 1997; Croft et al., 2003). In addition, male guppies spend more time near larger shoals than smaller ones (Lindstrom and Ranta, 1993), most likely because larger shoals may provide guppies with added protection from predation and allow them to find food faster (Krause and Godin, 1995; Day et al., 2001).

The majority of shoaling studies in guppies and other species have been performed on adult fish. However, juvenile fish are extremely vulnerable to predation (Fuiman and Magurran, 1994; Sogard, 1997), and it seems reasonable to expect that juveniles may benefit from shoaling as so adult fish. Both juvenile

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roach, *Rutilus rutilus*, and juvenile pollock, *Pollachius virens*, are known to shoal as an anti-predator defence strategy (Persson and Eklov, 1995; Rangeley and Kramer, 1998). Since juvenile guppies suffer from predation by the pike cichlid, *Crenicichla alta*, and the killifish, *Rivulus hartii* (Mattingly and Butler, 1994), they may demonstrate shoaling behavior as a method of group protection.

In this study, we examined the behavior of juvenile guppies to determine whether they demonstrate basic shoaling behavior, whether they choose size-matched individuals as shoal mates and whether they show a preference for large shoals over small shoals.

## 2. Materials and methods

Newborn guppies reared from a domesticated commercial strain were collected within 24 h of birth and housed in isolation until testing date in 250 ml opaque plastic beakers set in a water bath at 26 °C, on a 12:12 LD cycle. All fish were fed Nutrafin flake food.

A test tank was created by dividing a 38-l aquarium into three chambers by adding two glass walls 13 cm from each end of the tank. On the exterior of the central chamber vertical lines were drawn 6 cm from each glass wall indicating preference zones for each end of the tank. Overall tank dimensions were 50 cm length × 32 cm height × 26 cm depth. The tank was maintained at 26 °C and illuminated by a 15-W incandescent light bulb. Brown paper was used to cover the back and sides of the tank.

For each behavioral assay, a ‘stimulus’ shoal was placed in an end chamber of the test tank 60 min prior to testing. Each test fish was taken directly from isolation housing beakers and placed in the center chamber and allowed to acclimate for 15 min prior to testing. Each assay required 20 test fish, and a total of five assays were performed. During an assay, the time the test fish spent in the preference zone near either end chamber was recorded over a 10 min time period. Stimulus fish and test fish were not from the same brood and test fish were not used more than once for testing. For each experiment equal numbers of assays were run with stimulus shoals in each end chamber to correct for ‘side biases’.

To examine the ability of juvenile fish to discriminate between potential shoal mates on the basis of age and body size, fish were studied at 10-, 30- and 50-days of age. In the first set of studies, 10- or 30-day-old test fish were given the choice of swimming near: (i) a shoal of five 10-day-old fish or an empty chamber, (ii) a shoal of five 30-day-old fish or an empty chamber, or (iii) a shoal of five 10-day-old fish or a shoal of five 30-day-old fish.

In the second series of tests, 10- or 50-day-old test fish were given the choice of swimming near (i) a shoal of five 10-day-old fish or an empty chamber, (ii) a shoal of five 50-day-old fish or an empty chamber, or (iii) a shoal of five 10-day-old fish or a shoal of five 50-day-old fish.

To study the ability of juvenile fish to distinguish between shoals of different size, 10-, 30- and 50-day-old test fish were given the choice of swimming near a large shoal (fifteen fish) or a small shoal (five fish) consisting of fish of similar age.

The order in which the tests within each assay were performed was randomised. For each experiment, time spent by test fish on either side of the central chamber was compared using a paired *t*-test. The body lengths of 10-, 30- and 50-day-old fish were compared using an ANOVA with Tukey post hoc comparisons.

## 3. Results

### 3.1. Body size

The mean total body lengths of fish from each age group (10-day-old fish =  $8.83 \pm 0.65$  mm, 30-day-old fish =  $13.17 \pm 0.94$  mm, and 50-day-old fish =  $18.60 \pm 0.99$  mm) were significantly different ( $F(2) = 627.46$ ,  $p = 0.001$ , ANOVA).

### 3.2. Assays comparing 10- and 30-day-old fish

Fish from each age group preferred to be near a shoal of fish rather than an empty chamber, with one exception. The 10-day-old fish showed no preference when given a choice between a shoal of 30-day-old fish and an empty chamber ( $t(38) = 1.583$ ,  $p = 0.13$ , paired *t*-test). The 10-day-old fish did, however, prefer to be near other 10-day-old fish rather than an empty chamber ( $t(38) = 5.778$ ,  $p = 0.001$ ), while 30-day-old fish preferred to be near other 30-day-old fish ( $t(38) = 3.038$ ,  $p = 0.007$ ) or 10-day-old fish ( $t(38) = 4.925$ ,  $p = 0.001$ ) rather than an empty chamber. Neither 10-day-old nor 30-day-old fish demonstrated a preference for age-matched (and therefore, size matched) individuals when given the choice between shoals of similar and dissimilar aged fish ( $t(38) = 1.577$ ,  $p = 0.131$ , and  $t(38) = -1.225$ ,  $p = 0.235$  respectively; see Fig. 1).

### 3.3. Assays comparing 10- and 50-day-old fish

Both 10- and 50-day-old fish preferred to be near fish rather than an empty chamber. Ten-day-old fish significantly preferred to be near 50-day-old fish ( $t(38) = 3.861$ ,  $p = 0.001$ ) rather than an empty chamber. Fifty-day-old fish also preferred to be near 10-day-old fish ( $t(38) = 3.472$ ,  $p = 0.003$ ) or 50-day-old fish ( $t(38) = 2.888$ ,  $p = 0.009$ ) rather than an empty chamber. When presented with similar and dissimilar aged fish, only 50-day-old fish demonstrated a preference for size-matched individuals ( $t(38) = -4.31$ ,  $p = 0.001$ ), while 10-day-old fish did not make that distinction ( $t(38) = -1.486$ ,  $p = 0.154$ ; see Fig. 2).

### 3.4. Assays on shoal size

When fish from each age group were presented with a small shoal and a large shoal, only 50-day-old fish preferred to be near the larger shoal ( $t(38) = -1.761$ ,  $p = 0.094$  for 10-day-old fish;  $t(38) = 0.108$ ,  $p = 0.915$  for 30-day-old fish;  $t(38) = -5.06$ ,  $p = 0.001$  for 50-day-old fish, see Fig. 3).

## 4. Discussion

In this study we examined shoaling behavior in juvenile guppies at three ages, 10-, 30-, and 50-days of age to determine

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