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# Shoaling preferences of two common killifish (*Fundulus heteroclitus* and *F. diaphanus*) in the laboratory and in the field: A new analysis of heterospecific shoaling

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#### ABSTRACT

Heterospecific grouping behavior of mummichogs (Fundulus heteroclitus) and banded killifish (Fundulus diaphanus) was analyzed in the laboratory and in a freshwater tidal marsh in Cremona, Maryland. Several parameters of wild, intact shoals were measured, including species composition, body length, parasite load, gender, and any physical abnormalities. Fish collected were used for laboratory analysis of shoaling preferences. When size was equal, banded killifish and mummichogs preferred conspecific shoals to heterospecific shoals, consisting of mummichogs, banded killifish, and sheepshead minnows (Cyprinodon variegates). Shoal collection from the field resulted in mixed species shoals with individuals predominantly unaffected by parasites or other physical abnormalities. Size appeared to be a sorting mechanism. A temporal shift in lengths was evident. Initial shoals caught contained significantly smaller fish compared to the final shoals caught. Results are compared with previous studies on heterospecific shoaling in killifish and new characteristics of heterospecific shoals inhabiting freshwater tidal marshes are discussed.

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#### 1. Introduction

Social groups exist within a wide variety of species such as flocks of geese, herds of cattle, and shoals of fish. In each case, the costs and benefits associated with grouping behavior may impinge upon an individual's decision to join a group (Pitcher and Parrish, 1993). Fish are ideal organisms for studying social grouping due to the propensity of different species to shoal, the ease with which shoaling tests can be developed, and the diversity of morphology, ecology, and behavior seen across different fish species.

Individual fish gain benefits from joining a shoal that may include increased foraging efficiency (Coolen et al., 2003; Ruxton et al., 1995; Yozzo and Smith, 1998), increased mating opportunities (Dosen and Montgomerie, 2004; Nordell and Valone, 1998), and a reduction in the risk of predation (Krause, 1993; Magurran, 1990). One proposed mechanism for reducing the predation risk is through the confusion effect, where a predator's sensory capabilities are impaired when faced with a large, uniform group of prey. It has been proposed that predators have difficulty singling out a specific individual to attack within a large group of phenotypically similar fish (Landeau and Terborgh, 1986; Milinski, 1979). Therefore, by associating with fish of similar appearance, individuals bene-

fit from the confusion effect while simultaneously gaining other shoaling benefits such as access to potential mates and increased foraging efficiency.

Fish increase the phenotypic homogeneity of a shoal by choosing shoal-mates that have a similar shape, size, pattern, or color to themselves (Krause et al., 2000a). Not surprisingly, this often results in shoals being comprised of individuals of the same species. However, when different species of fish appear phenotypically similar to each other they may shoal in multi-species (heterospecific) groups (Krause and Godin, 1994; Krause and Godin, 1996; Landeau and Terborgh, 1986).

Shoaling research on killifish has focused primarily on field and laboratory studies using populations of banded killifish (*Fundulus diaphanus*) collected from Morice Lake, New Brunswick, Canada (Krause and Godin, 1994; Krause and Godin, 1996; Krause et al., 1996a, 1999, 2000b; Hoare et al., 2000). The laboratory studies found that shoal-mate choice in banded killifish was affected by body length, shoal size, and parasite presence. Specifically, banded killifish always preferred size-matched individuals regardless of differences in the size of stimulus shoals (Krause and Godin, 1994) and both parasitized and unparasitized fish preferred unparasitized stimulus shoals to parasitized stimulus shoals (Krause and Godin, 1996).

Field studies on killifish in Morice Lake have shown that banded killifish form heterospecific shoals with golden shiners (*Notemigonus crysoleucas*), white suckers (*Catostomus commersoni*),

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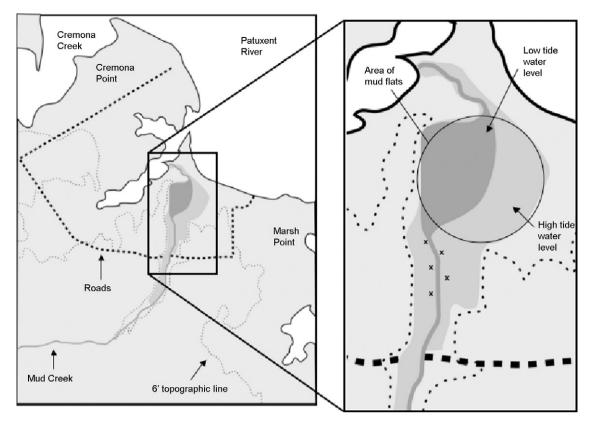


Fig. 1. Mud Creek study location, Cremona, MD. x's denote the position of umbrella nets on the peripheral of the main channel.

threespine sticklebacks (*Gasterosteus aculeatus*), and fourspine sticklebacks (*Apeltes quadracus*). Analysis of shoal composition revealed that the shoals were assorted by species composition and body length. Researchers state that the assessment of shoal suitability in terms of body length and species composition probably takes place within a very short time span, probably less than a few seconds (Krause et al., 1996a, 2000b). When parasites were present, heterospecific shoals were also assorted by parasite load and prevalence (Krause et al., 1999; Hoare et al., 2000).

Banded killifish have also been observed to shoal heterospecifically in a freshwater tidal marsh in Maryland (Wagner, 1999). Freshwater tidal marshes are distributed worldwide along the margins of estuaries as they transition into creeks or marshes. These sites are characterized by salinities less than 0.5 ppt, mudflats with high organic matter, and numerous shallow channels (<1 m depth) created by bi-directional water flow into, and out of, the marsh driven by the lunar tides (see Odum, 1988 for a comprehensive review). Compared to salt marshes, freshwater tidal marshes have both freshwater and estuarine fish, particularly young-of-year, with mixed communities of Cypronodonts (Fundulus sp.) often being numerically dominant. Freshwater tidal marsh fish have increased food resources in the form of insects, amphipods, and detritus from highly diverse plant communities (McIvor and Odum, 1988; Allen et al., 1994). These fish also experience lower predation rates from piscivores in the freshwater tidal marshes, but are subject to higher densities of avian predators (Rozas et al., 1998; Wagner, 1999; Yozzo and Smith, 1998).

Here we present a study on the shoaling behavior of banded killifish (*F. diaphanus*) and mummichogs (*Fundulus heteroclitus*) inhabiting a freshwater tidal marsh (Mud Creek, Cremona, Maryland). Both a field and laboratory study was conducted to analyze the grouping behavior of banded killifish and mummichogs. Results from the study are compared with previous studies on the heterospecific shoaling in killifish. Characteristics of heterospecific

grouping behavior of these fish in a freshwater tidal marsh are also discussed

#### 2. Materials and methods

#### 2.1. Field study

The field study was conducted in June 2007 at a remote site in Mud Creek, Cremona, MD, USA (Fig. 1). Mud Creek flows into a freshwater tidal marsh south of Cremona Point and North of Marsh Point before entering the Patuxent River. Marsh grasses and other vegetation covered the area surrounding the study site. Beyond this vegetation, forested areas could be found on both sides of the creek. The marsh varied in depth, temperature and salinity with the tide.

Shoals were captured using un-baited umbrella traps (Promar 'umbrella net',  $91 \, \text{cm} \times 91 \, \text{cm}$ ,  $5 \, \text{mm}$  mesh-size) that were positioned in the main creek channel through the marsh and attached to poles embedded into the substrate such that the nets could be remotely raised from a boat (Fig. 2). The umbrella traps were modified from their original condition by folding down the sides to allow fish to actively swim over the net surface. In this way, fish did not accumulate in the net (as with a flume net) and samples represent only the fish that were above the net when pulled from the water.

Collections were timed to coincide with the incoming tide as this was the time at which fish were observed to be at their highest abundance and also to facilitate ease of movement of the boat in the marsh. A total of five nets were used and checked sequentially in a manner that minimized disturbance and 'herding' of fish by the boat in the narrow channel. Nets were placed no closer than 5 m from one another and were checked as the tide came in until no shoals were caught in two full rotations of raising the nets (10 nets checked). Although the typical definition of a shoal is any group of two or more fish, for this experiment, a shoal was considered to be any group containing more than four fish. Shoals were considered

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