2008 Study Plan for Dr. David C. Heins
Fish Resource Permit Application

Collaborators and History

The research on threespine stickleback is part of a long-term series of studies being conducted in collaboration with Drs. John A. Baker and Susan A. Foster of Clark University, Worcester, MA; Dr. Bill Cresko at the University of Oregon, Eugene, Oregon; and Dr. Michael A. Bell of the State University of New York at Stony Brook, NY. We are conducting research in the lakes of the Matanuska-Susitna (Mat-Su) Valley and Kenai Peninsula of south central Alaska. The diversity of populations in Alaska and the relative ease of access to most of them have made Alaska a most profitable study region for research on threespine stickleback.

I have been involved with Drs. Baker, Foster, and Bell in studies collectively involving the behavior, morphology, and life history of threespine stickleback. More recently we have been investigating the same aspects of the biology of ninespine stickleback.

Projects and Goals

Four major projects are included in the work in which I am involved: 1) host-parasite relationship between the threespine stickleback (*Gasterosteus aculeatus*) and the cestode macroparasite *Schistocephalus solidus*, 2) host-parasite relationship between the ninespine stickleback (*Pungitius pungitius*) and the cestode macroparasite *Schistocephalus pungitii*, 3) ecological and evolutionary factors in the life history of the threespine stickleback (*Gasterosteus aculeatus*), and 4) ecological and evolutionary factors in the life history of the ninespine stickleback (*Pungitius pungitius*).

The threespine stickleback *Gasterosteus aculeatus* is a widely studied model organism worldwide. The Matanuska-Susitna Valley and Kenai Peninsula of south-central Alaska presently form one of the most important regions for studies of the host-parasite relationship and the life history of *Gasterosteus aculeatus*. The work on *Pungitius pungitius* has already and should continue to provide new scientific information on the life history and the host-parasite relationship of *Pungitius pungitius*, a species about which we know very little worldwide despite its having a widespread distribution similar to the threespine stickleback.

1.
Host-parasite relationship between *Gasterosteus aculeatus* and *Schistocephalus solidus*

The purpose of this research is to study the host-parasite relationship between the threespine stickleback (*Gasterosteus aculeatus*) and the cestode parasite *Schistocephalus solidus*, including outcomes for both the host fish and the parasite. Our research suggests that this work will provide new scientific insights into the different ways in which this relationship can play out (Heins and Baker, 2008). Thus far we have shown that, contrary to earlier reports, reproduction of threespine stickleback occurs when levels of parasitic infection are high and that females are capable of producing clutches of eggs despite substantial infections (Heins et al., 1999; Heins and Baker, 2008). We also have shown that egg sizes of stickleback females are reduced due to side effects of parasitism (Heins and Baker, 2003). Likely these effects are in part the result of the parasite reducing the body condition of host fish (Bagamian et al., 2004; Schultz et al., 2006). And we have demonstrated that size characteristics of host fish affect the sizes of their parasites (Heins et al., 2002). The size of the parasite attained in the threespine stickleback host is important because it influences the ability of the parasite to infect and reproduce in about 40 species of aquatic birds. Questions now being addressed include the following:

1) Do levels of parasitic infection of stickleback vary among lakes? If so, what factors determine the varying levels of infection? Levels of parasitism are measured in terms of the percentage of infected individuals (prevalence), the number of parasites per infected fish (intensity), and the mass of parasites in relation to the host mass (severity).

2) Do levels of parasitism in stickleback vary among seasons within a year or among years within one lake? If so, what factors result in the observed changes?

3) Do levels of parasitic infection differ among sexes or among age/size classes of stickleback? If so, what factors result in the observed differences?

4) Does the parasite delay sexual maturation in male and female stickleback?

5) Does parasitic infection result in effects on clutch mass and clutch size of stickleback? Do these effects vary with time, and if so, what factors are involved?

6) Do any variations in levels of parasitism across years result in annual variation in the reproduction of the stickleback?

7) Do parasitic infections have effects on the population sizes of host fish? If so, what are those effects?

Stickleback will be collected from a number of lakes. They will be trapped using 1/8" or 1/4" mesh minnow traps. Alternatively, some samples may be taken using dip nets or minnow seines. Specimens will be anaesthetized and then fixed in 10% formalin in the field. The preserved specimens will be shipped back to Tulane University. After transfer to the laboratory, the stickleback will be dissected following measurements of standard length and blotted wet mass. Parasites will be removed from infected stickleback, and the parasites will be counted and weighed (blotted wet weight). Gonads of sticklebacks will be removed and staged to determine reproductive condition. Female sticklebacks bearing clutches (mature, ripening and ripe stages of condition) will be used to determine clutch sizes, clutch masses and egg sizes of both infected and uninfected stickleback. Other organs such as the liver or kidney may also be examined to determine their condition. Standard methods will be employed throughout the study. Standard statistical analyses of the data will be used to address the questions posed for these studies.

2.
Host-parasite relationship between *Pungitius pungitius* and *Schistocephalus pungitii*

The purpose and methods of this project are similar to those of the study of the relationship between *Gasterosteus aculeatus* and *Schistocephalus solidus*. There is, however, one significant difference between the two projects. Although the host-parasite relationship involving the threespine stickleback has been widely studied, little is known about the ninespine stickleback and its relationship to its cestode parasite, making any contribution to science likely to be new information. To date, we have published one paper addressing the host-parasite interaction (Heins et al., 2004). The results for the ninespine stickleback contrast rather strongly with those for the threespine stickleback, the ninespine stickleback being much more affected by the parasite.

3.
Ecological and evolutionary factors in the life history of *Gasterosteus aculeatus*

The major thrust of this research, which is largely being coordinated by Dr. Baker, is to determine the ecological and evolutionary causes of variation in the life histories of populations of stickleback in Alaska. We are studying local, geographic, and long-term patterns of variation in clutch size, egg size, size/age at sexual maturity, and population age structure. A recent contribution from this effort (Baker et al., 2005) addresses fish from Benka Lake in the Matanuska-Susitna Valley, and an overview of research in Alaska (Baker et al., 2008) was just published.

Each year collections of gravid females and adult males are made from a set of populations that previous research has indicated are most informative for our purposes. These collections are preserved and shipped back to the laboratory for dissection and analysis in the same way as described above. These continuing annual collections maintain continuity in the research and are yielding an important long-term database on the level of year-to-year

variation in the primary populations. In addition, we also try to make collections from populations that have not yet been sampled to expand the overall data set.

4.

Ecological and evolutionary factors in the life history of *Pungitius pungitius*

The purpose and methods of this project are the same as those of the study of the life history of *Gasterosteus aculeatus*. Again, there is a significant difference between the two projects in that little is now known about the life history ninespine stickleback whereas the threespine stickleback has been the subject of numerous life history studies. Thus, our knowledge base for the ninespine stickleback is quite rudimentary, and almost any contributions to our knowledge of the ninespine stickleback are likely to be new. We have recently published two papers on the life history of this poorly known species (Heins et al., 2003; Heins et. al, 2005) and we have another in preparation at the present time.

Literature Cited

Baker, J. A., W. A. Cresko, S. A. Foster, and D. C. Heins. 2005. Life history differentiation of benthic and limnetic ecotypes in a polytypic population of threespine stickleback (*Gasterosteus aculeatus*). Evolutionary Ecology Research 7: 121-131.

Baker, J. A., D. C. Heins, S. A. Foster, and R. W. King. 2008. An overview of life-history variation in female threespine stickleback. Behaviour 145: 579-602.

Bagamian, K. H., D. C. Heins, and J. A. Baker. 2004. Body condition and reproductive capacity of three-spined stickleback infected with the cestode *Schistocephalus solidus*. Journal of Fish Biology 64: 1568-1576.

Heins, D. C. and J. A. Baker. 2003. Reduction of egg size in natural populations of threespine stickleback infected with a cestode macroparasite. Journal of Parasitology 89: 1-6.

Heins, D. C. and J. A. Baker. 2008. The stickleback–*Schistocephalus* host–parasite system as a model for understanding the effect of a macroparasite on host reproduction. Behaviour 145: 625-645.

Heins, D. C., J. A. Baker, G. De Silva, and E. L. Birden. 2005. Clutch characteristics of two populations of ninespine stickleback from south-central Alaska. Journal of Fish Biology 67: 873-878.

Heins, D. C., J. A. Baker and H. C. Martin. 2002 The "crowding effect" in the cestode *Schistocephalus solidus*: density-dependent effects on plerocercoid size and infectivity. Journal of Parasitology 88: 302-307.

Heins, D. C., J. M. Johnson, and J. A. Baker. 2003. Reproductive ecology of the ninespine stickleback from south-central Alaska. Journal of Fish Biology 63: 1131-1143.

Heins, D. C., S. S. Singer and J. A. Baker. 1999. Virulence of the cestode *Schistocephalus solidus* and reproduction in infected threespine stickleback, *Gasterosteus aculeatus*. Canadian Journal of Zoology 77: 1967-1974.

Heins, D. C., B. Ulinski, J. Johnson and J. A. Baker. 2004. Effect of the cestode macroparasite *Schistocephalus pungitii* on the reproductive success of ninespine stickleback *Pungitius pungitius*. Canadian Journal of Zoology 82: 1731-1737.

Schultz, E., M. Topper, and D. C. Heins. 2006. Decreased reproductive investment of female threespine stickleback *Gasterosteus aculeatus* infected with the cestode *Schistocephalus solidus*: parasite adaptation, host adaptation, or side effect? Oikos 114: 303-310.

Locality List

Lakes to be sampled for purposes of this project will include many of the lakes listed below. Not all lakes listed below will be sampled this year, and some additional lakes may be sampled. The lakes are listed according to present levels of priority.

I am requesting authorization to collect as many as 600 threespine stickleback from all lakes, except for Scout Lake and Walby Lake. At the present time, I am focusing research efforts on Scout Lake and Walby Lake. Thus, I request authorization to collect 1000 threespine stickleback each from Scout Lake and Walby Lake. We request authorization to take as many as 600 ninespine stickleback from each lake, except Dog Bone Lake. Similarly, we are focusing on Dog Bone Lake for ninespine stickleback and request authorization to collect 1000 from that lake.

HIGH PRIORITY
Beverly Lake[Mat-Su]
Big Beaver Lake [Mat-Su]
Boot Lake [Mat-Su]
Cornelius Lake [Mat-Su]
Daniels Lake [Kenai]
Dog Bone Lake [Kenai]
Grant Lake [Kenai]
Horseshoe Lake [Mat-Su]
Loberg (Junction) Lake [Mat-Su]
Matanuska Lake [Mat-Su]
Mud Lake [Mat-Su]
Rocky Lake [Mat-Su]
Scout Lake [Kenai]
Solid Rock Lake [Kenai]
Tern Lake [Kenai]
Walby Lake [Mat-Su]
West Lake [Mat-Su]
Willow Lake [Mat-Su]
Duck Lake [Kenai]
Engineer Lake [Kenai]
Georgina Lake [Kenai]
Lazy Lake [Mat-Su]
Lynda Lake [Mat-Su]
Stepan Lake [Mat-Su]
Suneva [Kenai]
Watson Lake [Kenai]

MODERATE PRIORITY
Blodgett Lake [Mat-Su]
Echo Lake [Kenai]
Faulk Lake [Mat-Su]
Hidden Lake [Kenai]
Lalen Lake [Mat-Su]
Lower Ohmer Lake [Kenai]
Pup Lake [Mat-Su]
Rabbit Slough [Mat-Su]
Seymour Lake [Mat-Su]
Stormy Lake [Kenai]
Trouble Lake [Mat-Su]
Wolf Lake [Mat-Su]

LOW PRIORITY
Bear Paw Lake [Mat-Su]
Big Lake [Mat-Su]
Bruce Lake [Mat-Su]