Into the Belly of the Beast: Traditional and Genetic Prey Detection in Liberty Island's Mississippi Silversides

Bippus[1], T., D. Jordan[1], A. Furler[1], S. Slater[1] (Steve.Slater@wildlife.ca.gov), G. Schumer[2], B. Schreier[3], L. Conrad[3], and M. Baerwald[4]

[1]California Department of Fish and Wildlife, [2]Cramer Fish Sciences, [3]California Department of Water Resources, [4]University of California, Davis

Introduction:

The Mississippi Silverside (*Menidia beryllina*) (Figure 1) is an introduced pelagic fish that preys on Delta Smelt (*Hypomesus transpacificus*) larvae (Baerwald et al. 2012); however, the effects of predation on a population level are poorly understood. Identification of fish larvae in the stomach of predators is difficult given their small size and rapid digestion rate. The purpose of this study was to utilize a novel approach of traditional visual processing followed by genetic testing of Mississippi Silverside stomach contents, with the goal of detecting and quantifying consumption of larval Delta Smelt.

Figure 1. Mississippi Silverside (73 mm FL).

Methods:

Fish collected from Liberty Island by US Fish and Wildlife Service (USFWS) Delta Juvenile Fish Monitoring Program

- Mississippi Silverside were collected by beach seine from March through May 2013 (Figures 2 and 3).
- Specimens were stored in 95% ethanol at -20°C.
- Larval Delta Smelt were collected by trawl in open water.

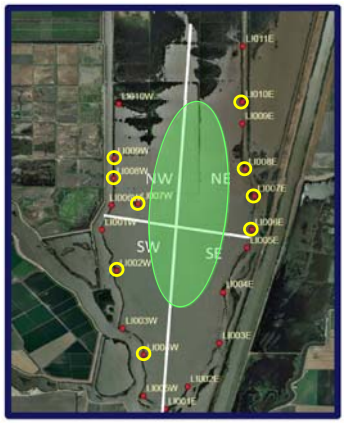
Fish transported to CA Department of Fish and Wildlife (CDFW) Diet Study Lab

- Fish measured and processed with sterile handling protocol to avoid DNA contamination between samples.
  - All tools and glassware bleached and gloves disposed of after manipulation of each carcass and replaced with clean items.
  - All items replaced again and work stations sterilized after complete processing of each stomach sample.

- Digestive tracts removed, stomachs dissected, and contents examined in lysis buffer without Proteinase K within approximately a 60 minute time constraint due to rapid degradation of the samples.
- Stomach contents identified using a dissecting scope and items enumerated to lowest possible taxonomic level (Figure 4).
- All prey items, stomach tissue, and lysis buffer placed in Eppendorf tubes.

Samples sent to Cramer Fish Sciences for genetic analysis

- Delta Smelt TaqMan assay design from Baerwald et al. (2011) employed.
- Proteinase K added to tubes containing stomachs and samples allotted time to dissolve. Complete homogenization accomplished by vortexing tissues with metal pellets under heated conditions.
- DNA extracted, diluted, and processed using quantitative PCR (qPCR) TaqMan assays.



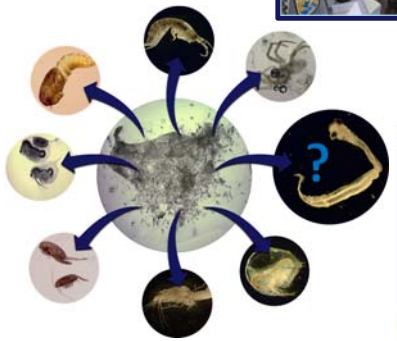


Figure 2. (upper left) USFWS staff collecting samples via beach seine and larval fish net.

Figure 3. (upper right) Map of Liberty Island beach seine sites. Sites that Mississippi Silverside were collected highlighted in yellow. Larval fish trawl area highlighted in green.

Figure 4. (left) Prey species within Mississippi Silverside stomachs (Clockwise from top): Amphipod, terrestrial invertebrate, larval fish (not found), cladoceran, mysid, copepods, cladocerans, terrestrial invertebrate larva.

Results:

- Mississippi Silverside (n = 49) fork lengths (FL) ranged from 44.5 to 89.0 mm FL (mean = 70.0 mm FL).
- All of the Mississippi Silverside stomachs contained prey items, with cladocerans and copepods accounting for 95% of the diet composition by number (Figure 5). Other organisms identified include insects, amphipods, and mysids.
- No larval Delta Smelt were found during the visual examination of Mississippi Silverside stomach contents. This was confirmed by subsequent DNA analysis.
- Delta Smelt larvae were present in Liberty Island during the period Mississippi Silverside were collected for this study (Figure 6), and Delta Smelt fork lengths ranged from 4.7 to 19.2 mm FL (mean = 9.4 mm FL).

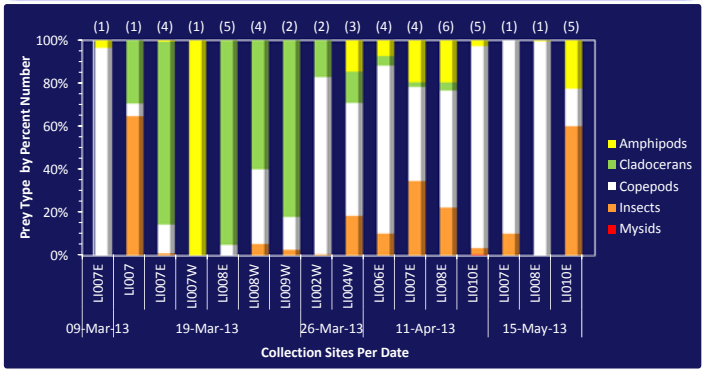


Figure 5: Prey type by percent number found in stomachs of Mississippi Silverside collected by site and date in Liberty Island. Number of stomachs reported in parenthesis above each bar.

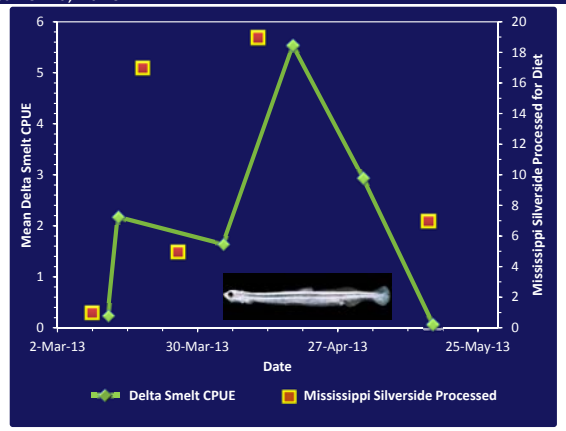


Figure 6: Mean catch-per-unit-effort (CPUE) of larval Delta Smelt per 100 cubic meters of water and number of Mississippi Silverside stomachs examined by date of collection at Liberty Island. Inset photo of a larval Delta Smelt (11.8 mm FL).

Conclusions:

- Mississippi Silverside preyed on organisms of various type and size, some of which were comparable in size to larval Delta Smelt. We found no evidence of predation on Delta Smelt.
- Delta Smelt larvae inhabited the sampling area and were of size to be prey for Mississippi Silverside.
- Mississippi Silverside near-shore may not have interacted with larval Delta Smelt in the open water. In a previous study (Baerwald 2012), Delta Smelt DNA was detected in the digestive tracts of Mississippi Silverside caught offshore by CDFW's Spring Kodiak Trawl, but not in individuals collected by beach seine.
- A lack of Delta Smelt DNA found during genetic analysis indicated no DNA contamination occurred during gut content identification. However, use of sterile techniques proved difficult and time consuming.
- A larger Mississippi Silverside sample size is required for a more thorough assessment of diet and potential impact on Delta Smelt larvae.

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