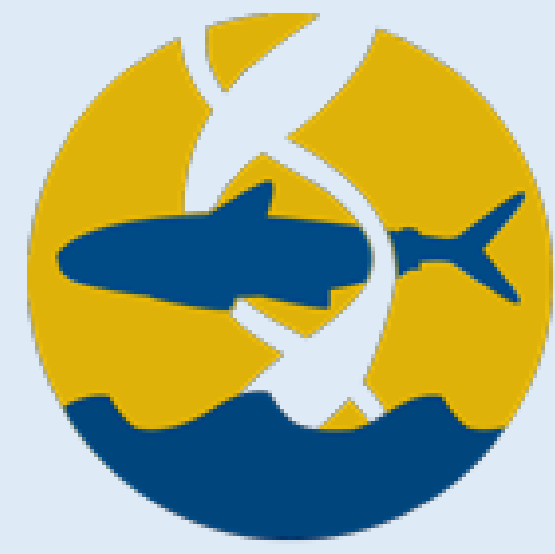


Salinity Tolerance of Delta Smelt Sperm



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Background



Delta smelt (*Hypomesus transpacificus*) are a small anadromous fish found in the Sacramento-San Joaquin Delta. This species inhabits the estuary region for most of the year, but most individuals migrate upstream to spawn in freshwater following the initial winter flow events. Multiple hypotheses have been proposed to explain these movement patterns, including microhabitat requirements for reproduction. One unstudied aspect may include the water chemistry necessary for sperm activation and mobility.

Methods

- Delta smelt were sourced during the breeding season (**March–April 2020**) from a genetically managed refuge population, maintained in freshwater (salinity: 0.2 ppt) at 12 °C at the Fish Conservation and Culture Laboratory (FCCL) in Byron, CA.
- Fish were anaesthetized and their **milt collected** directly from the genital duct using a pipette while squeezing the abdomen.
- Milt was diluted in a non-activating extender solution and kept in a refrigerator.
- **Ten treatment solutions** were prepared: distilled water (**0 ppt**), river water (**0.2 ppt**), and eight saline treatments made by adding Instant Ocean® sea salt to distilled water (**1, 2, 4, 6, 8, 10, 20, and 35 ppt**), reflecting natural variability from the upper river to the estuary (35 ppt represents ocean saltwater).
- **Milt was activated** on a microscope slide using one of the treatment solutions and **a video captured of the sperm movement**.
- The OpenCASA plugin in ImageJ was used to track sperm movement and measure **average sperm velocity and linearity** (i.e. how straight they swim) for each video.
- Milt from 14 fish was analyzed with at least two replicates within each treatment solution per fish.

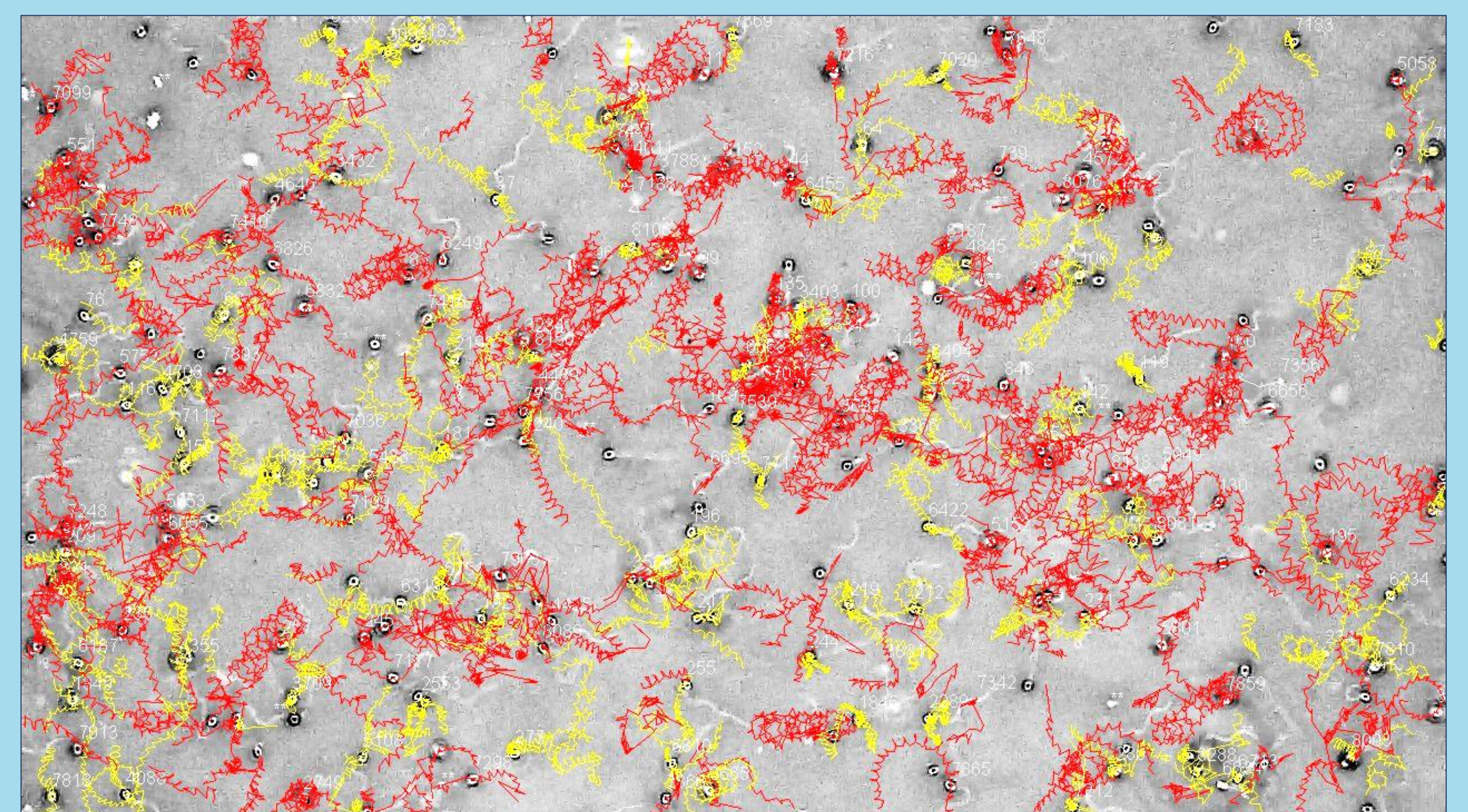
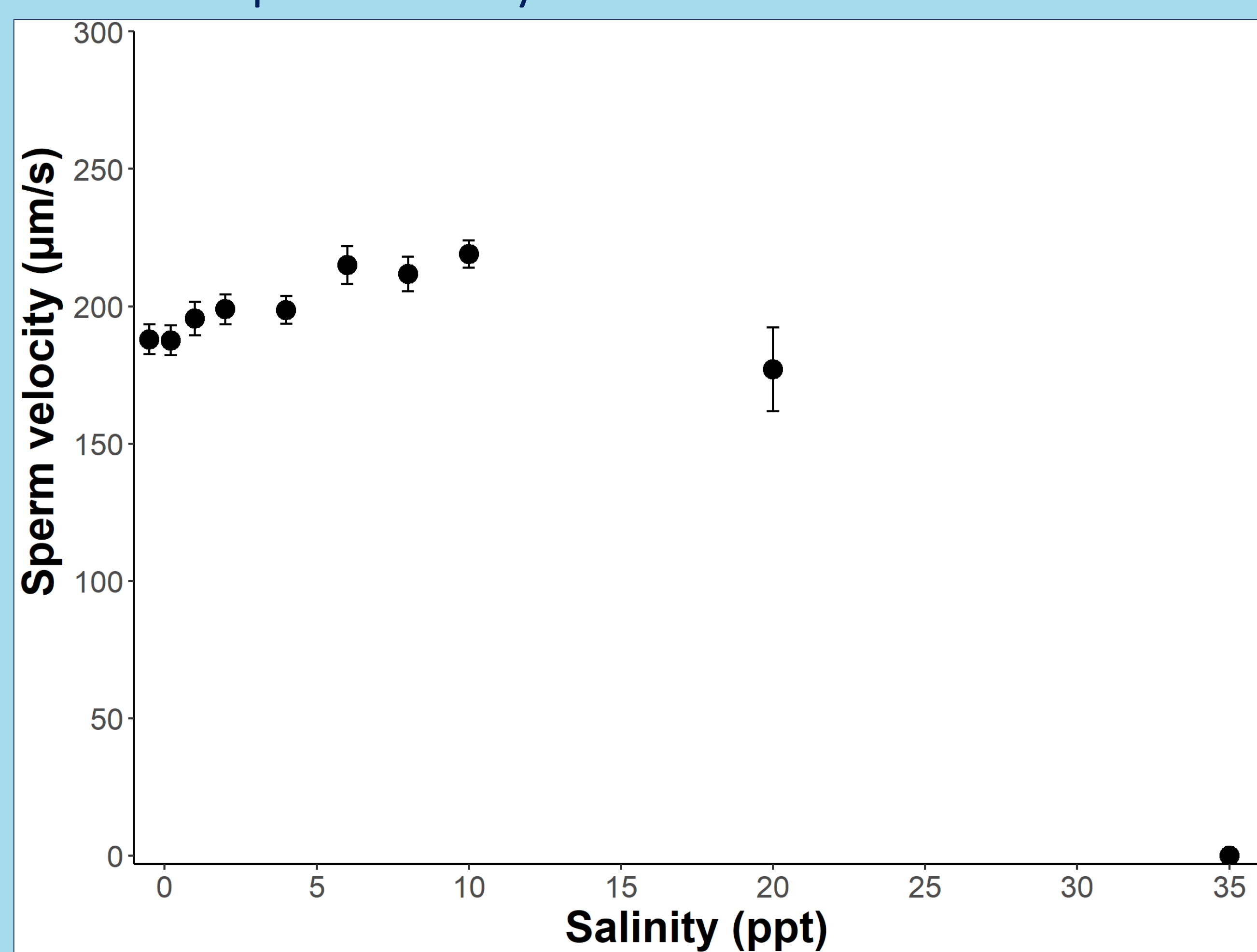


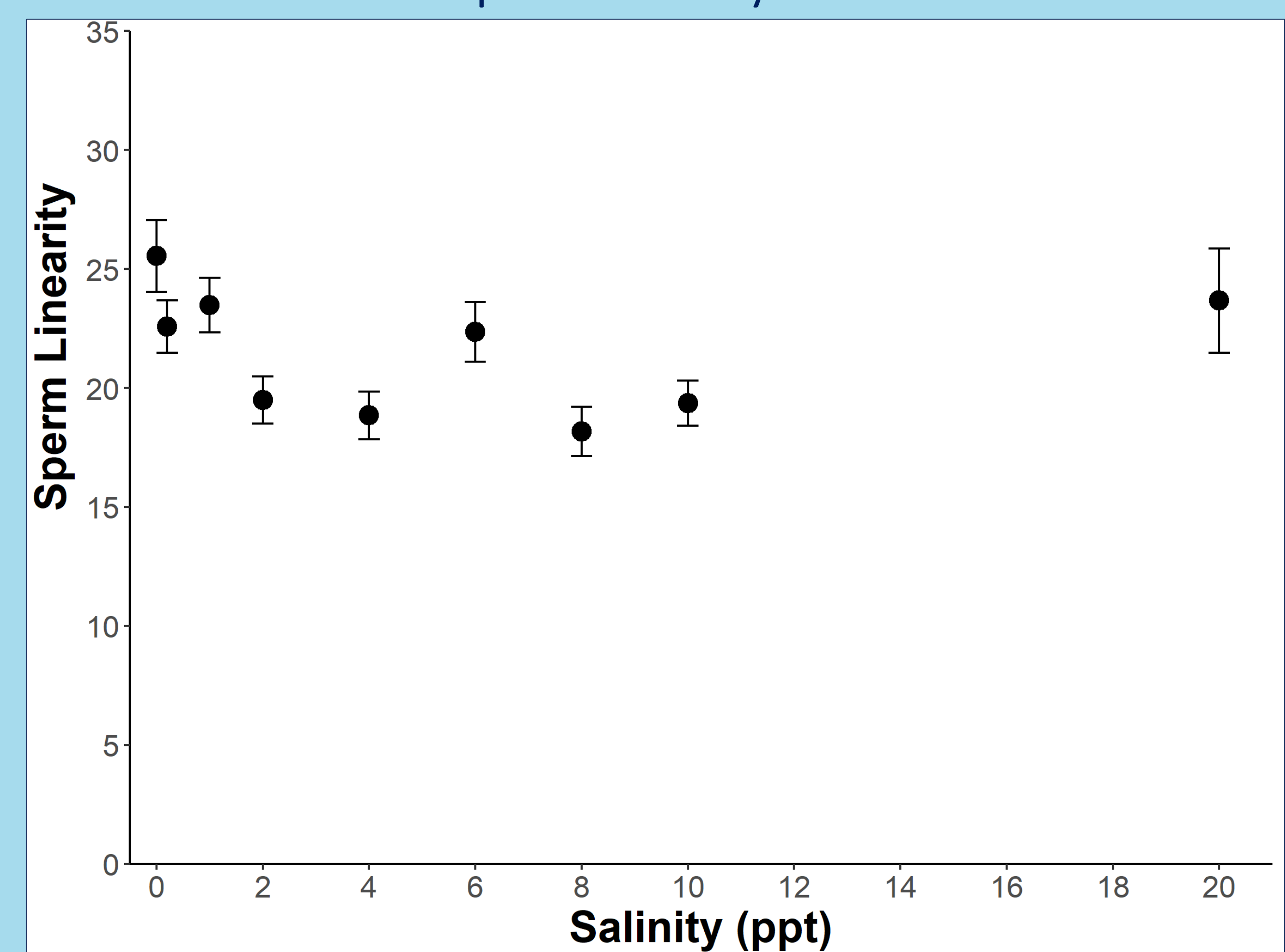
Image displaying sperm tracking from a 2 second video. Red tracks are of sperm moving faster than a set speed threshold.

Results

We found sperm **velocity** was not significantly different when activated in solutions between 0 and 4 ppt salinity, and faster between 6 and 10 ppt. Sperm swam significantly slower at 20 ppt, and were completely inactive at 35 ppt. There were also significant differences in sperm velocity between individual males.



We found sperm **linearity** was higher (sperm swam straighter) when activated in solutions with low salinity, but swam in more circular patterns between 2 and 10 ppt (with the exception of 6 ppt). Sperm movement patterns were highly variable at 20 ppt. There were significant differences in sperm linearity between individual males.



Conclusions



Delta smelt migration upstream for reproduction is unlikely to be purely driven by sperm biology requirements, although sperm motility was decreased at very high salinity levels (20 ppt) and completely inactive at a marine salinity level. Delta smelt sperm are fairly tolerant to a wide range of salinities (<10 ppt). While sperm movement patterns are altered by salinity, it is as yet unknown how these differences may affect fertilization success.

Want to see a video of Delta smelt sperm moving?

<https://ucdavis.box.com/s/x3kqam1i363zol0on19bizmzf3e2rcy>

