Reduced reproductive success at **20°C** and the low viability of **Delta Smelt female × Wakasagi male** crosses help explain why viable hybrids arise primarily from Wakasagi females and have implications for conservation management.

TEMPERATURE EFFECTS ON EMBRYOS OF CULTURED DELTA SMELT *Hypomesus transpacificus*, WILD WAKASAGI *Hypomesus nipponensis*, AND THEIR HYBRIDS

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INTRO

- Delta Smelt and non-native Wakasagi hybridize at low frequency in the San Francisco Bay Delta, but the drivers of these interactions remain unclear.
- Temperature influences smelt reproduction and early development, and the two species share overlapping spawning temperatures.
- Hybrids observed in the wild have been exclusively produced by **Wakasagi females** (Fig. 1).
- Examining temperature effects on embryos of both species and their hybrids informs conservation hatchery management.

METHODS

- Both species were strip spawned and crossed in a full-factorial design to produce pure-species crosses and hybrids in both cross directions.
- Eggs from each cross were incubated at three temperatures, 12, 16, and 20°C, under controlled hatchery conditions.
- Fertilization and hatching success and time to 50% hatch were quantified for each cross and temperature.
- Mixed effects models were used to test the effects of cross type, temperature, and their interaction on reproductive and developmental metrics.

RESULTS

- Fertilization and hatching success declined at higher incubation temperatures across **all crosses** (Fig. 2).
- Pure Delta Smelt crosses showed the highest fertilization success, while Delta Smelt egg × Wakasagi milt (DS × WK) crosses showed the lowest hatching success.
- Time to 50% hatch decreased with increasing temperature for all crosses (Fig. 3).
- Developmental timing differed between species at the highest temperature, with Wakasagi eggs hatching earlier than Delta Smelt eggs (20°C, Figure 3).

DISCUSSION

- Higher incubation temperature reduced fertilization and hatching success across species and crosses.
- Differences in developmental timing between species were observed at the **highest incubation temperature**.
- Lower hatching success in Delta Smelt female hybrids suggests a **post-fertilization barrier** and helps explain the observed directionality of hybrids.
- The results provide temperature-based guidance for Delta Smelt conservation and management.

Fig. 1 (a) Delta Smelt, (b) Wakasagi-Delta Smelt hybrid, and (c) Wakasagi. Photograph by René Reyes.

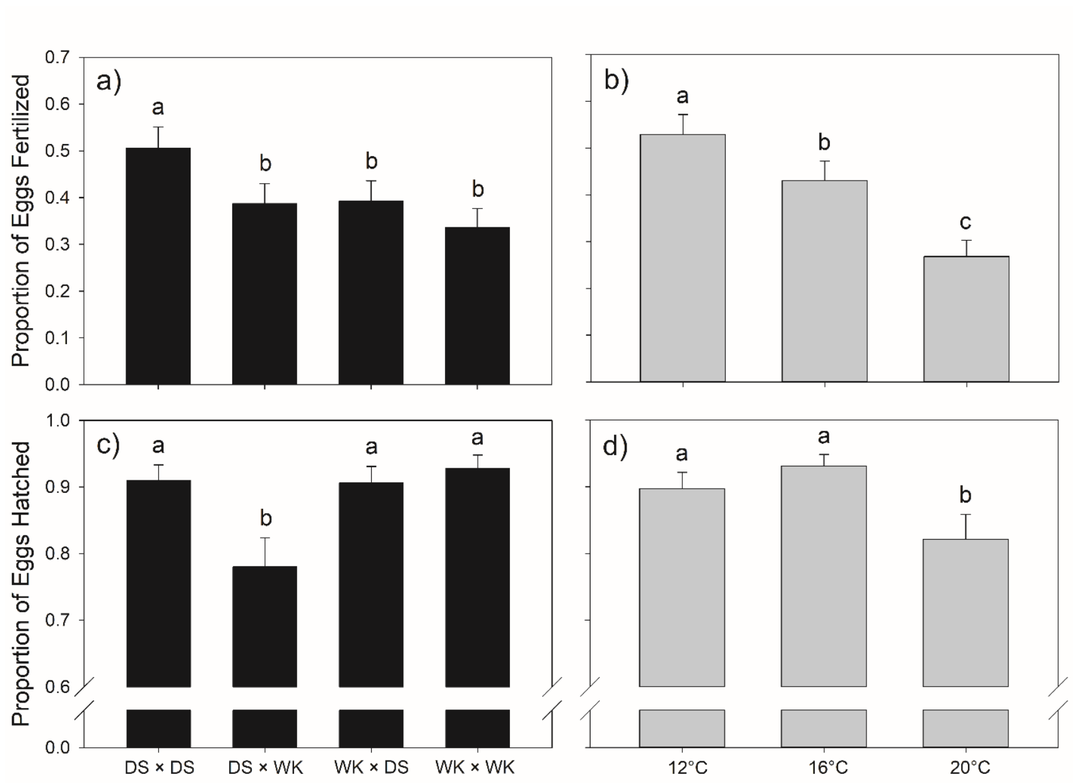

Fig. 2 Back-transformed estimated marginal mean proportion of eggs fertilized (a, b) or hatched (c, d) by cross and temperature. Different letters denote significant differences between groups ($P < 0.05$).

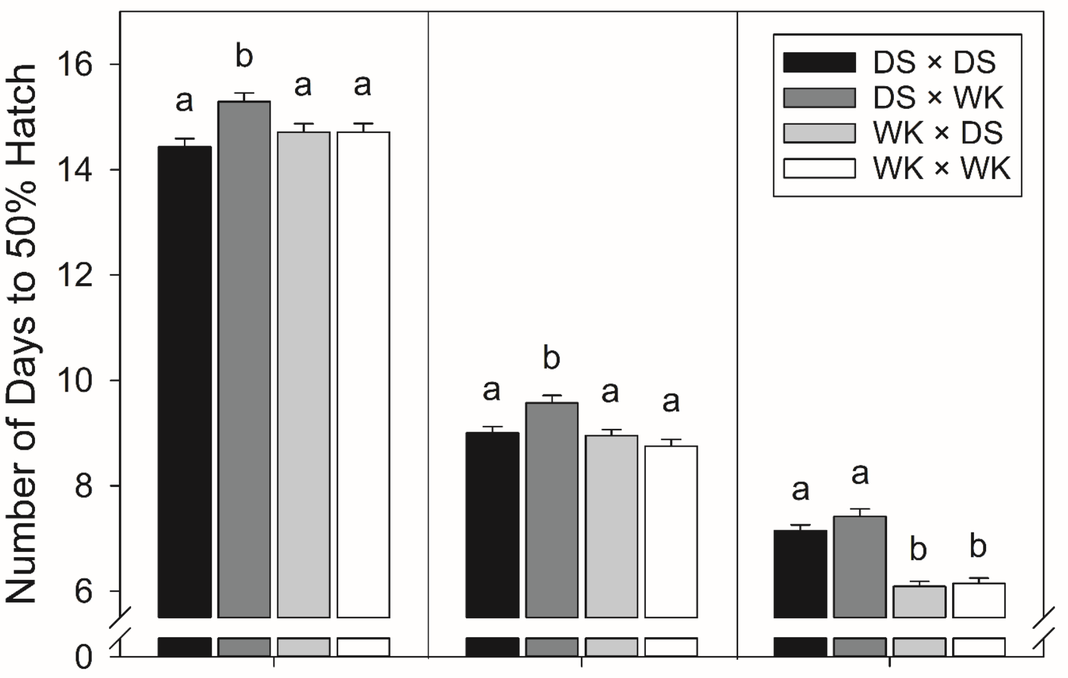

Fig. 3 Back-transformed estimated marginal mean number of days to 50% hatch by cross and incubation temperature. Within each temperature or panel, different letters denote significant differences between cross types ($P < 0.05$).

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