Extending storage time for Rainbow Trout (Oncorhynchus mykiss) milt: The effect of temperature
Alex Elizabeth Howell, and Dr. Scott Newbold
Department of Life Sciences, Sheridan College

QUESTION
Can rainbow trout milt (sperm) be stored for longer periods of time and what is the effect of temperature on storage time?

METHODS
- Milt collection took place on 20 March 2017; observations began the same day (Fig. 1).
- Milt samples from 15 male trout were divided and stored at two temperatures.
- Motility was checked every 5 days, for a total of 21 days (Fig. 3).
- Egg fertilization was conducted on 30 Mar and 10 Apr, days 10 & 21 (Fig. 6 & 7A).
- Note: Two different females were used for each temperature on both fertilization days, four females total.
- Egg fertilization rate (% eyed eggs) was determined on 15 Apr and 26 Apr (Fig. 2A&B and Fig. 7B).

RESULTS
- Motility dropped below 50% for most samples by day 10 (Fig. 4).
- Motility slightly higher for samples held at warmer temperature (Fig. 3).
- Despite poor motility, fertilization data from days 10 and 21 showed that the colder temperature had higher fecundity rates (Fig. 5A & B).
- Motility was not a good indicator for viability (Fig. 4 & 5).

HYPOTHESES
Trout milt stored at 1.7°C will remain viable longer and have a higher fertilization rate than milt stored at 4.2°C, and despite motility, milt stored for longer periods (> 15 days) can still fertilize eggs.

CONCLUSION
No strong effect of storage temp on milt storage time based on motility; dropped below 50% (WGFD cutoff) by day 10; interestingly, high fertilization despite poor motility, and higher rate for samples stored at colder temps.

Literature Cited

Acknowledgements
This project would not have been possible without the support and guidance of the WGFD Story Fish Hatchery and Dr. Newbold. We especially thank Jennifer Melenev for insights into the techniques and strategies used to conduct our research. We also thank Brad Hughes, Steve Diekema, and Jen Meineke for helping with field work and data collection. The Story Fish Hatchery generously allowed us access to milt, eggs and other materials needed. We are also grateful to the Sheridan College Life Science department for use of their lab space, equipment, and supplies.

Figure 1. Alex and Brad collecting milt while Steve took photos.
Figure 2. (A) Bad eggs separated from eyed-eggs; (B) fertilized eggs with visible eye spots; (C) checking milt motility with light microscope.
Figure 3. Mean percentage of motile sperm at two temperatures over 21 days.
Figure 4. Percentage of motile sperm from 15 individuals at two temperatures over 21 days.
Figure 5. Mean (±SE) fertilization rate (% eyed-eggs) using milt from 10 males at two temperatures after: (A) 10 days, and (B) 21 days.
Figure 6. Alex, Brad and Jen collecting and fertilizing eggs.
Figure 7. (A) Fertilized eggs in the drip trays; (B) Alex and Jen separating bad eggs from eyed-eggs.
Figure 8. Alex and Brad waiting for a male to become temporarily immobilized for milt collection.

Department of Life Sciences, Sheridan College