## QUANTIFYING BIOMASS FROM FISH eDNA: A COMPARISON OF THE SENSITIVITY OF TWO SAMPLING METHODS FOR FLOWING WATER

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liters of water.

samples (<5L).

At the Creston National Fish Hatchery, we compared the sensitivity of novel drift net sampling devices to that of water filtration for the detection of rainbow trout (Oncorhynchus mykiss) environmental DNA. An advantage of drift nets is that they can filter 1000s of liters of water, capturing relatively large particles of size > 10 microns (µm). Here, we used a factorial experimental design to compare the sensitivity of fine mesh drift nets (35-60 µm) using large volume water samples (1000-4000L) vs. small pore filters (1.5 μm) using small volume water samples (5L) to detect rainbow trout eDNA in modified raceways with flowing water. For at least two species of fish, eDNA is most abundant in the 1 – 10 μm particle size fraction and can be as large as 60 μm (Turner et al., 2014, Wilcox et al., 2015). Thus, larger filter pore sizes are still able to capture target eDNA while allowing smaller free-floating particles to pass, but small fragments of targeted eDNA may be missed. This tradeoff is predicted to be offset by sampling larger water volumes (Turner et al., 2014), but this prediction has not been tested.

Questions

- Can we reliably detect environmental DNA (eDNA) from a single rainbow trout fry (very low biomass, ~1g.) in flowing water?
- Can we detect a change in rainbow trout abundance or biomass using eDNA?
- · What effect does volume of water sampled have on the amount of rainbow trout eDNA detected?

**Treatment** 

- Sampling equipment: fine mesh drift nets (35 & 60 microns), small pore filter (1.5 microns).
- · Water volume sampled: 1000L, 2000L, 4000L with drift nets, 5L with filter.
- Rainbow trout abundance: 0, 1 fish, 16 fish, & 32 fish (~1-33g).

Levels



- Consistent detection of 1 fish (~1g biomass) with 2 drift net sampling treatments (2000L with 35 micron mesh, 4000L with 60 micron mesh) and the filter. All three had very similar median amounts of rainbow trout DNA detected (Fig. 1).
- The median amount of rainbow trout DNA was detected 2-3x higher using 4000L samples than with the filter for the same biomass (Fig. 3).

**Abundance** 

- The amount of rainbow trout DNA detected using all sampling techniques was significantly higher for a biomass of 16g (~3-5 fish) than 1g (1 fish) (Fig. 2).
- A consistent increase in the amount of rainbow trout DNA detected for given biomass was observed with higher sample volume. Using the drift net, both mesh sizes, 35 & 60 microns, showed a similar pattern (Fig. 3).

Top: Drift net with fine mesh used to sample 1000s of

Bottom: Typical filter sampler for small volume water

Left: Sloped raceway with drift net sampler positioned for sample collection. Note how flow has extended conical net.

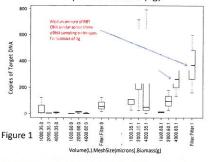
Top Right: Small holding pen below well water tap at top of sloped raceway.

Bottom Right: Bucket showing ~60 rainbow trout used for biomass in experiment. 1 fry weighed ~1-3g.

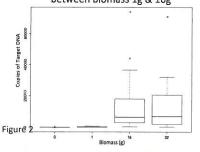
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- 4- USFS, Rocky Mountain Research Station

Experimental methods: All treatment-level combinations were replicated at least three times. All experiments took place in sloped stainless steel troughs (14" W x 8" D x 10.5' L), using a drop of ~3' to replicate flowing water. Troughs and sampling devices were decontaminated with bleach and rinsed with water between each replicate. Fish were acclimated in holding pen for 30' before samples were collected. Well water flowing at 19 gals/min and approximately 5C was run into each trough.

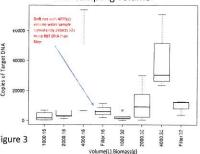
## Consistent detection of RBT DNA even for very small biomass (1g)



Significantly higher median amounts of RBT DNA detected across all sampling methods between biomass 1g & 16g



Median amount of RBT DNA detected increases with sampling volume



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