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Field-ready environmental DNA (eDNA) protocols and tools for sea lamprey assessment

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ABSTRACT:

The Sea Lamprey Control Program uses standardized electrofishing protocols that estimate larval sea lamprey density (especially those >100 mm in length) to prioritize streams for lampricide treatment. However, EF surveys are labor-intensive, can be difficult or dangerous under some conditions (e.g., high water), and are limited seasonally (i.e., possible only between ~April and October). Resources allocated to assessment will reduce those available for the implementation of control. Environmental DNA (eDNA) monitoring programs are becoming routine for other aquatic invasive species, and prior GLFC support fostered development and initial testing of species-specific PCR-based eDNA assays and protocols for sea lamprey. Building off this research, we conducted a series of experiments to optimize a field sampling protocol for eDNA sampling of larval sea lamprey (e.g., determining that eDNA sampling at least 1 month after the end of the spawning period should detect only larval DNA in the system and testing different filter types and two filtration methods) and created standardized eDNA sampling protocols and training materials that sea lamprey larval assessment teams at Fisheries and Oceans Canada and US Fish and Wildlife Service could implement. Over 2 years and with three larval assessment crews, we then conducted a large-scale study comparing the ability of eDNA sampling versus Rapid Assessment (RA) electrofishing surveys to detect sea lamprey at 108 sampling stations on 24 Great Lakes tributaries in Canada and the United States. Each sampling station was sampled for eDNA three times (in the summer, fall, and winter), with the summer and fall sampling activities being paired with an electrofishing survey. When amalgamating the detections at a given station across time periods, we observed that eDNA sampling and electrofishing results agreed at 79 stations (73%). There were only 2 sampling stations (2%) where eDNA failed to detect sea lamprey presence when the species was detected by electrofishing, but credible detections of sea lamprey eDNA were made at 27 sampling stations (25%) where electrofishing failed to find sea lamprey during either survey. Detections were considered credible if there was no evidence of contamination in any of the controls. A weak relationship was observed between eDNA signal strength (proportion of replicates testing positive for sea lamprey) and relative larval abundance (number of larvae collected during electrofishing) at each station; the proportion of detections was consistently high at the few sites with high larval densities, but it was much less consistent at medium and low densities, likely due to the confounding effects of environmental variables that could dilute or inhibit the eDNA signal. Using hierarchical occupancy models to estimate detection probability and error rates, we found that 6 eDNA samples with 3 qPCR replicates each provides a greater than 95% probability of detecting sea lamprey eDNA present at the sampling station, which is equivalent to conducting 1–3 RA electrofishing surveys. The quantitative analysis also revealed an extremely low false positive error rate at both the field sample and laboratory analysis stages. Using parameter estimates from the first quantitative analysis as informed priors for a

hierarchical Bayesian occupancy model, we were also able to estimate the effect of various environmental covariates on the detection probability of sea lamprey eDNA. This analysis revealed that eDNA detectability decreases with decreasing water temperature and increasing distance downstream, while eDNA detectability increases with increasing electrical conductivity (mS) and total dissolved solids (ppm). In addition to optimizing and testing eDNA protocols for sea lamprey, we also developed and tested the ability of eDNA to detect the presence of native lampreys in the Great Lakes, that is, American brook lamprey (*Lethenteron appendix*) and the *Ichthyomyzon* species (northern brook, silver, and chestnut lampreys). We confirmed that all assays were highly genus-specific (i.e., reliably differentiating among sea lamprey, American brook lamprey, and *Ichthyomyzon* spp.), and compared the ability of eDNA and electrofishing to detect American brook lamprey and *Ichthyomyzon* spp. at 54 stations across 12 Great Lakes tributaries. As with sea lamprey, we found that eDNA sampling was more sensitive than traditional electrofishing methods. Our research shows that eDNA can be used effectively to complement existing larval assessment protocols, particularly given its sensitivity at low densities. It could be used to identify upstream infestation limits of sea lamprey prior to treatment, monitor re-infestation of streams above barriers or following treatment, and identify new sea lamprey infestations (e.g., previously unknown spawning populations or range expansions that might be occurring due to climate warming or pollution abatement). It could also be used to better understand the distribution of the native lampreys in the Great Lakes to enhance the ability of the Sea Lamprey Control Program to minimize non-target effects during control.