

Using eDNA to Detect The Natural Re-Introduction of Two Fish Species (Atlantic Salmon- *Salmo salar*, Striped Bass- *Morone saxatilis*) in Rivers Where They Were Once Present

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One of the sample sites on the Sissiboo river

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Two of the authors enjoying their work- Annick Blinn (left) & Jessica Dulong (right)

Using scientific technology and local Indigenous knowledge to advance the protection of our marine habitat for future generations

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INTRODUCTION

Atlantic salmon (*Salmo salar)* and Striped Bass (*Morone saxatilis)* were once plentiful throughout the rivers and ocean waters of the Maritime region of Canada (COSEWIC 2018, COSEWIC 2019). Today, however, Salmon populations along the Bay of Fundy and Atlantic coast of Nova Scotia have been assessed as Endangered by the Committee on the Status of Endangered Wildlife (Atlantic salmon (Salmo salar) (COSEWIC- 2018). As well, in the case of Striped Bass its various populations throughout the Maritime region of Canada are considered to be of Special Concern or Endangered (COSEWIC- 2004). With climate change it is believed that the changing habitat of these two species will create an even more uncertain future for their survival (Fisheries & Oceans Canada, 2025).

Therefore, it is critical to see if rivers in above mentioned areas of Nova Scotia, that once held healthy populations of these two species, might contain some remnant populations. One method that has become more scientifically accepted in recent years is the measuring of eDNA found within the habitat of various species (Wang, 2021). In the case of fish, it has certain advantages and disadvantages over traditional methods of population evaluations or taken samplings. When using the eDNA method, water samples (which contain eDNA) are taken directly from the watershed, passed through a filter, and this filter is then sent to a laboratory for analysis. This is usually simple and quick to achieve and do not disturb the fish or the habitat. However, the results only reveal that there are fish present without giving any specific numbers,

or health status, gender, or physiological parameters (ie size, age, etc) of the species being studied. Also at the present time, eDNA is relatively easier to conduct than the usual biological in the field surveys which are time consuming, require high amounts of human effort, and are often harming to the environment. More traditional methods, such as netting, electro-fishing, or barbless hooking are more invasive and disturbing but reveal more precise numbers or health status as to the population studied (Ontario Fisheries, 2026). This project chose eDNA as a way to determine if the presence of the two species were found in the 3 rivers (Grosses Coques River (44° 21' 13" north, Longitude: -66° 05' 35" west), Salmon River (Latitude- 44° 3' 6" north, Longitude- 66° 9' 40" west), and Sissiboo River (Latitude- 44° 24' 26.75" north, Longitude: -66° 00' 12.72" west) located in southwest Nova Scotia and if some population was discovered, a more comprehensive study could be undertaken in the future to evaluate the status of the populations.

In consultation with and assistance from a local Indigenous group (Association des Acadiens-Métis Souriquois (AAMS)- Comeauville, Nova Scotia), the above 3 rivers were chosen as there were reports from the local fishers that possibly both Salmon and Striped Bass had been caught despite the scientific evidence that there were no populations of these 2 fish species thriving in those river habitats. As well, Indigenous history/knowledge spoke of teeming populations of these 2 fish species within these 3 river systems especially with respect to salmon which was once plentiful in the Salmon river watershed. The local Mi'kmaq people history speaks of teeming numbers and how critical salmon was to their diet and society. "The Mi'kmaq people continue to rely on Atlantic salmon for food. Over time, the Mi'kmaq developed an intimate and sacred relationship with the salmon and sought ways to manage populations while sustaining themselves. This relationship continues today and the species remains an important

part of the Mi'kmaq way of life." (2024-2025 Atlantic Salmon Plamu). With this Indigenous knowledge, and fishing history in mind, it was decided that the 3 above rivers should be chosen to begin a long term study to see if there were any natural populations within the watersheds.

Several factors had to be considered when it came to obtaining the water samples for the analysis of the fish eDNA. The species of fish and their life cycle needs (ie water temperature, their spawning cycle, fish migration to and from the river headwaters, hatching of the eggs, bottom sediment, etc) had to be considered, which led to deciding what season (Spring) had the most chance of obtaining the eDNA from the river water. As well, the effect of climate change with respect to the changes in water temperatures or water depths were unknown factors to decide when or how to collect eDNA (COSEWIC, 2018, COSEWIC, 2019, Fisheries & Oceans Canada, 2013).

The areas for collection were chosen based on the 2 species of fish and their life cycles. Fortunately both species have similar life cycles or breeding preferences. Both species are anadromous meaning they require both fresh and marine water to complete their life cycles, they both require clean fresh running water to breed/growth of young, require the same water temperatures, suitable habitat, etc. (COSEWIC, 2018, COSEWIC, 2019, Fisheries & Oceans Canada, 2013).

METHODOLOGY

eDNA/Water Sampling

To collect the eDNA, a commercially available unit called the *eDNA Citizen Scientist* was used to filter the water samples for eDNA (SMITH-ROOT Inc,



Set-up of the eDNA Citizen Scientist unit

Vancouver, WA, USA 98686). The system is composed of a portable pumping system that pushes the water through a “self-preserving eDNA Filter” to capture the eDNA. At the same time, 1liter of water is also collected to analyse for potential environmental contaminants, water pH, sediments, or any other parameters. These filters are free from contamination if used properly, easy to handle, and are easily shipped for analysis (eDNA Citizen Scientist, 2026).



Removal of the filter after sample is taken

The samples for eDNA were sent to a certified Canadian government laboratory (Agriculture and Food Laboratory/University of Guelph- Guelph, Ontario N1G 2Z4). The methodology for measuring the eDNA is complex but it basically involves separating the eDNA from the filter using an extraction technique. Multiple copies of the extracted eDNA are created using PCR amplification or making millions of copies of the targeted eDNA to be used for analysis which is critical when looking at low concentrations or the identification of rare species. These multiplied eDNA or DNA sequences are used to compare to available online databases to identify the species present in the environment (Tournayre, 2025).

RESULTS

A total of 11 samples were analysed for both fish species (Salmon n= 11, Striped Bass fish=11) from each watershed and resulting eDNA levels as seen in Table 1 below. There were no eDNA found of the 2 fish species within the three river watersheds studies.

Table 1: Samples Analysed From the Various Watersheds and Fish Species*

River Watershed	Atlantic Salmon *(Salmo salar)*	Striped Bass *(Morone saxatilis)*	eDNA Levels
Grosses Coques (n= 4)	4	4	0
Salmon River (n=3)	3	3	0
Sissiboo (n= 4)	4	4	0

* Each individual sample taken was analysed for each fish species, ie n=11 samples taken which were analysed for each fish species. There were no eDNA levels detected in any of the samples taken.

As part of the eDNA sampling procedure, a water sample was taken from each river watershed and the results are shown in the table below (Table 2). Twelve parameters were assessed at each river site during the eDNA sampling event between May/June 2026. Of the three rivers studied, the Salmon river had higher levels of calcium, magnesium, hardness, alkalinity, and total dissolved solids than the other 2 river watersheds. The levels of arsenic, copper, and lead were below detectable limits (0.00) in all 3 rivers (not shown).

Table 2: Results of water samples From the Three Watersheds

Parameter	**Sissiboo River**	**Grosse Coques River**	**Salmon River**
Calcium (mg/L)	0.889	2.905	50.367
Iron (mg/L)	0.256	0.360	0.375
Magnesium (mg/L)	0.615	1.279	158.321
Manganese (mg/L)	0.021	0.089	0.027
Uranium (mg/L)	0.00	0.015	0.036
Hardness (mg/L)	4.8	12.5	777.7
Alkalinity (mg/L)	0.00	5.98	16.78
Total Dissolved Solids (mg/L)	127	56.0	5150
pH (SU)	5.1	6.5	6.4

DISCUSSION

Simply put, the absence of Salmon/Striped Bass eDNA indicates that there were no fish present to shed its DNA for filtration at the time of the study. It is also possible that with a very low population of spawning fish in the watershed there was not sufficient amounts of eDNA present in the water to be filtered out and analysed. As well, it may be that the sampling was not done at the peak time of the spawning season leading to insufficient presence of eDNA. So what could be the reasons for the lack fish eDNA? In order to assess this question, more monitoring data such as proper water temperatures for spawning or rearing of the young, the amount of turbidity in the water system, the pH and heavy metal levels, water levels in the watershed, habitat destruction from human intervention or climate change, etc. would assist in the detection or presence of these fish species.

With respect to water quality, very little recent data can be found of the three studied river sheds that were examined within this study. However, a study from 2010 of streams leading into nearby local lakes looked at the same 9 parameters that were measured in this study which might be used for comparison (Taylor, 2010). The Grosse Coques and Sissiboo rivers had parameters that were close to each other and comparable to the 2010 study. However, when compared to the other two rivers, the Salmon river had some very high values when comparing calcium (almost 25 times more), hardness (over 350 times more), and total dissolved solids (over 94 times more). These values may explain the lack of the two fish species within the Salmon river but not the other two rivers that were looked at (Grosse Coques, Sissiboo).

Much work has been done already but studies that combine scientific procedures along side with Indigenous knowledge at a more local level would create current data bases that would lead to significant protection of these and other species should a population return to these 3

watershed in the future (Weitzman, 2025).

Even though the results did not show the presence of the 2 fish species, it allowed an important collaboration to be formed and valuable baseline data to be established for future work. This future work could reveal when some individuals would be re-introduced in the watershed, a rough estimate of the time of year they returned, and reveal that conditions were favourable for the population to establish themselves. This would be critical for future re-introduction of these species as it would indicate that the habitat is suitable for population growth and to establish recommendations that would protect these populations and their habitat for future generations and continue to play a role in local Indigenous life.



One of the Salmon River sample sites

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