

# Mucus proteomics in minnows from nearby creeks to determine the impact of aqueous film



## forming (AFFF) usage at three southern Ontario airports

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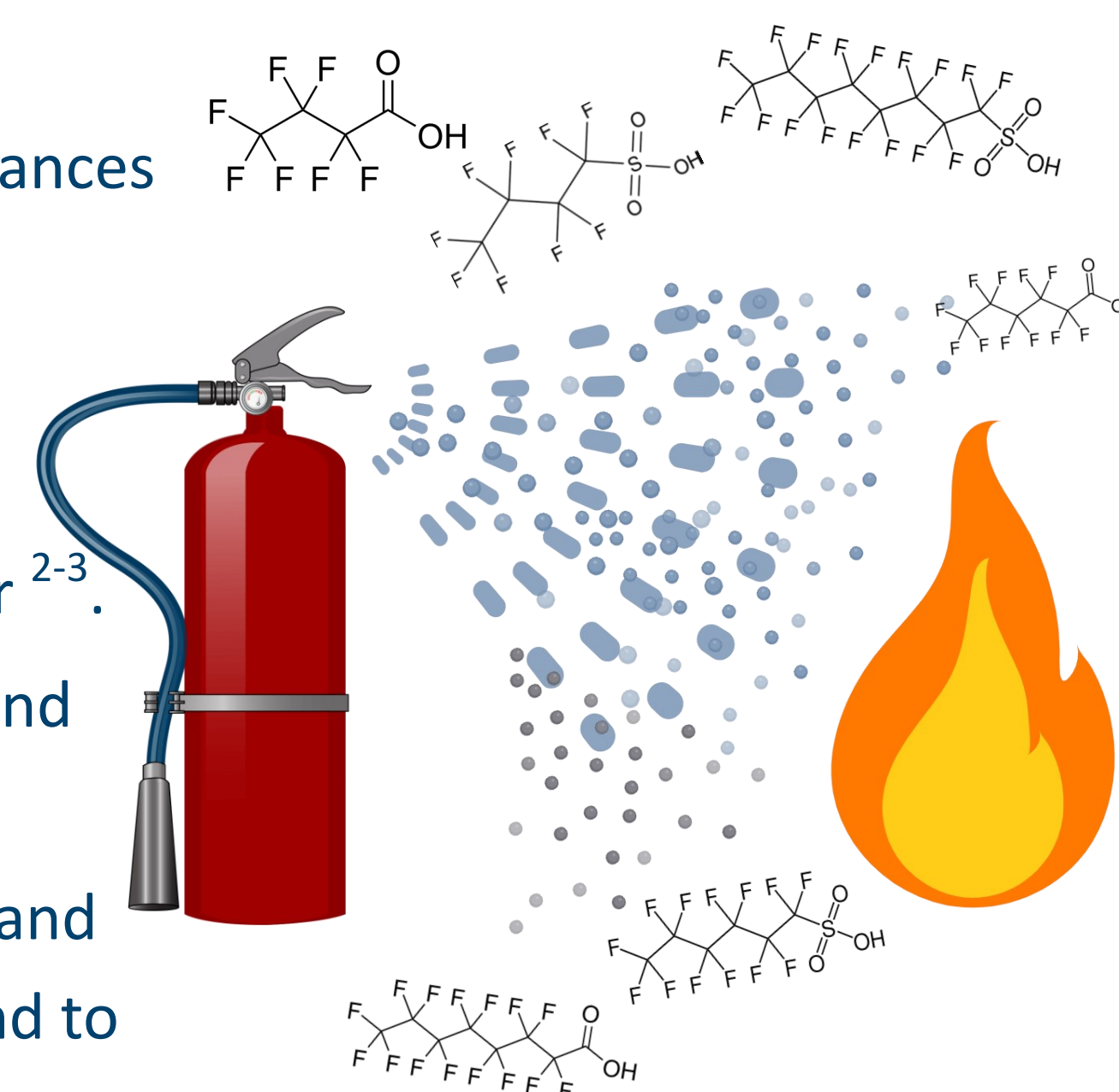
### Introduction

#### Why do we care about AFFF's?

- AFFF's are surfactant containing fire suppressants used to extinguish chemical fires. Perfluoroalkyl Substances (PFAS) are a major component of AFFF's and provide AFFF's their functionality.
- Perfluoroalkyl Substances (PFAS) are synthetic compounds commonly found in water resistant products, aqueous film forming foams (AFFFs) and non-stick cookware<sup>1</sup>.
- PFAS exposure has been shown to impact vital organs such as the liver and brain, as well as cause cancer<sup>2-3</sup>.
- The toxicity of PFAS coupled with its persistence in the environment has made them an environmental and human health concern<sup>4</sup>.
- Airports are a common source of AFFF's as they are commonly used as training grounds for firefighters, and house large volumes of AFFF's in case of chemical fires. Therefore, creeks that run alongside airports tend to contain elevated levels of several PFAS analogues.

#### What is "omics"?

- Omics' is a powerful tool that provides a protein, lipid or metabolite profile of an organism in response to a toxicant without the need for severe adverse effects<sup>5</sup>. Through generating a response-based profile, it is possible to understand the mechanism behind an organisms' response to a toxicant and deduce larger scale outcomes.

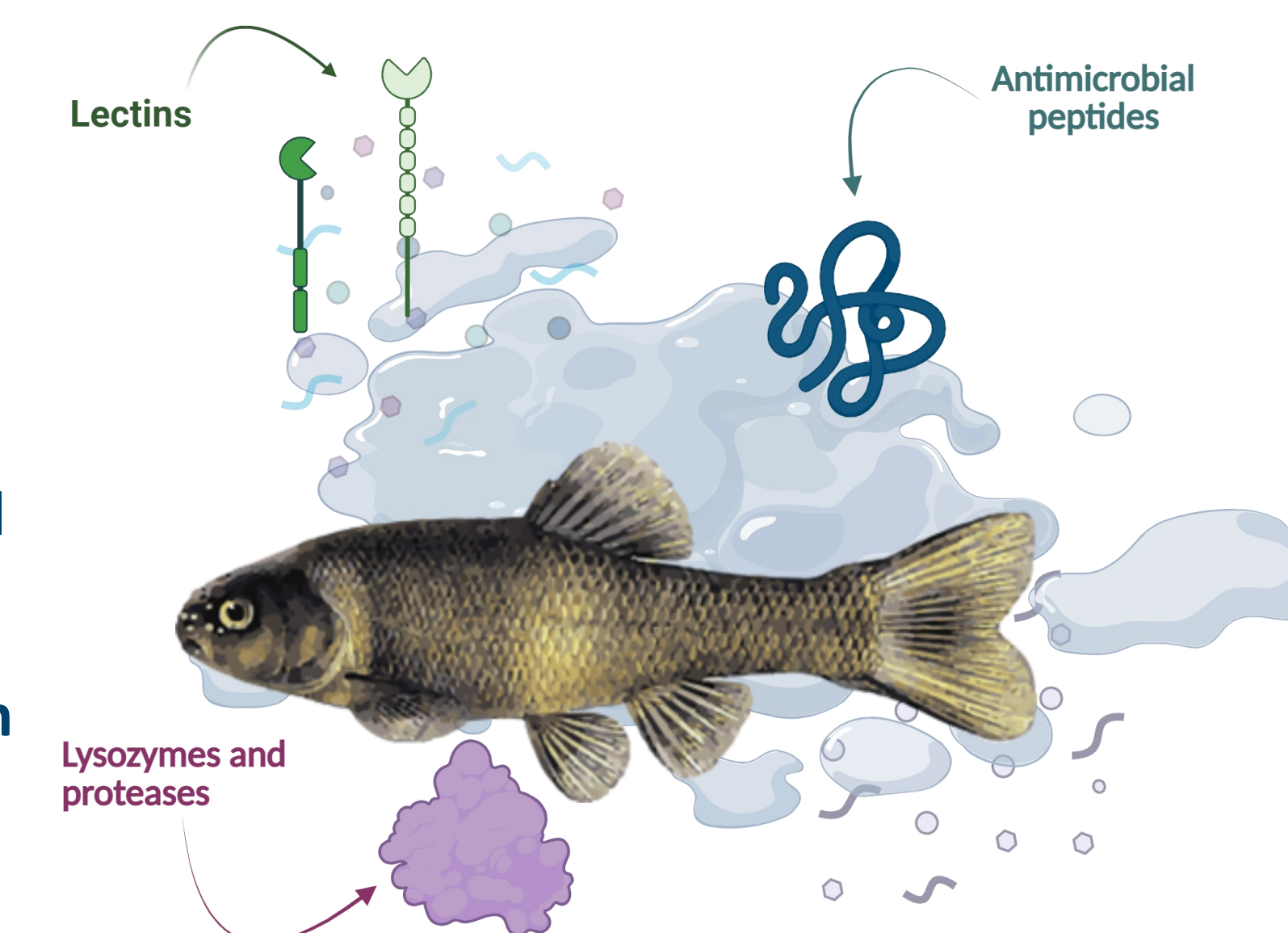


#### Why sample mucus?

In short; its *kind of* like taking a blood test!

- Fish epidermal mucus serves a variety of functions as it helps the immune system and aids in wound healing.
- Mucus can be collected non-invasively and non-lethally.

Therefore, the collection and analysis of fish epidermal can provide information on the health of a fish without animal sacrifice.



#### Study Goals:

To use proteomics to determine the toxicological outcomes of environmental PFAS exposure and AFFF runoff on wild minnows.

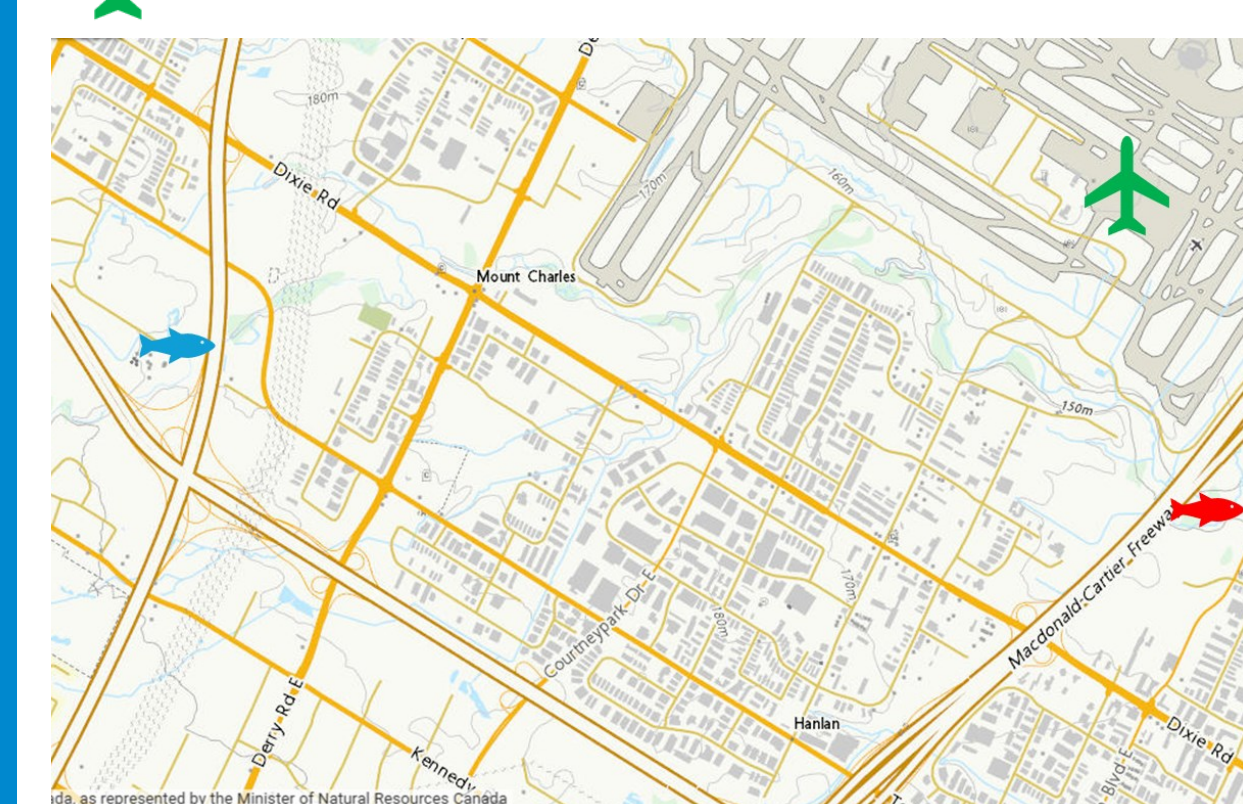
To assess the use of fish epidermal mucus as a reliable biofluid for toxicological assessment *in-situ*.

### Methods

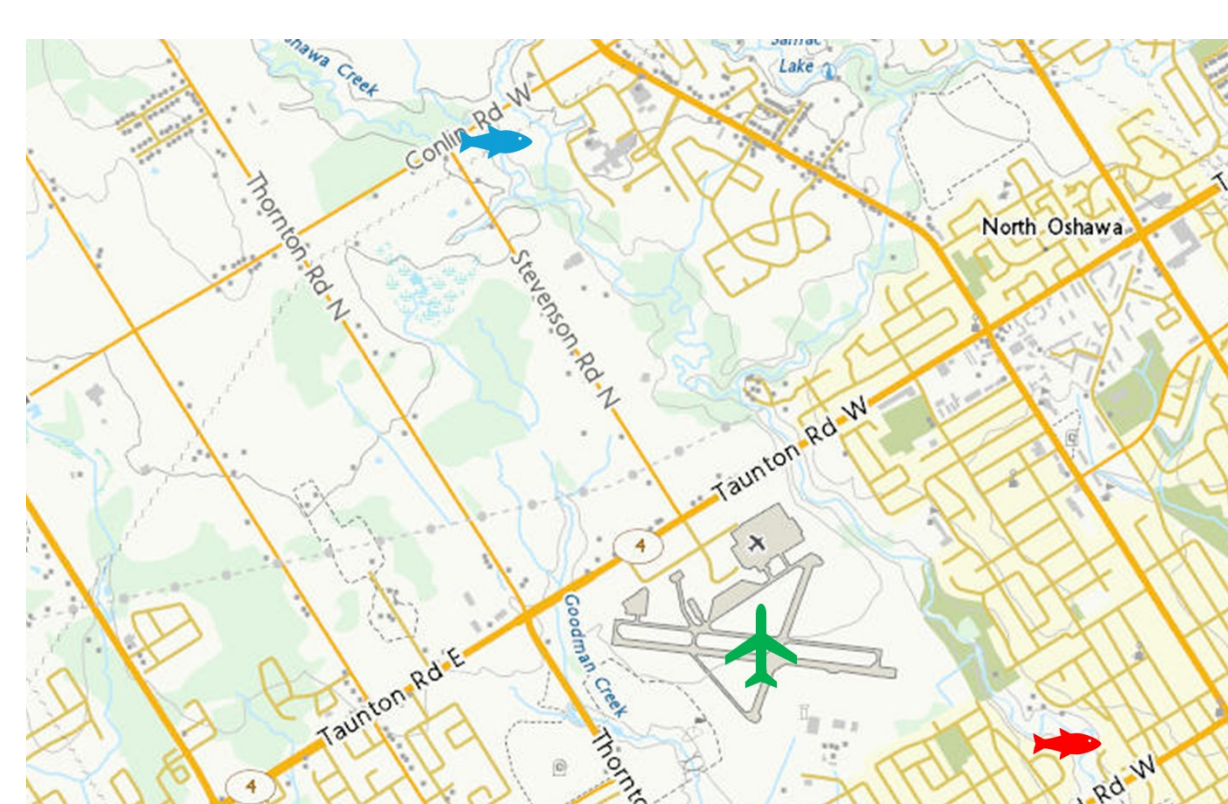
#### Site Selection

Three airports and three adjacent creeks within the greater Toronto area (GTA) were selected:

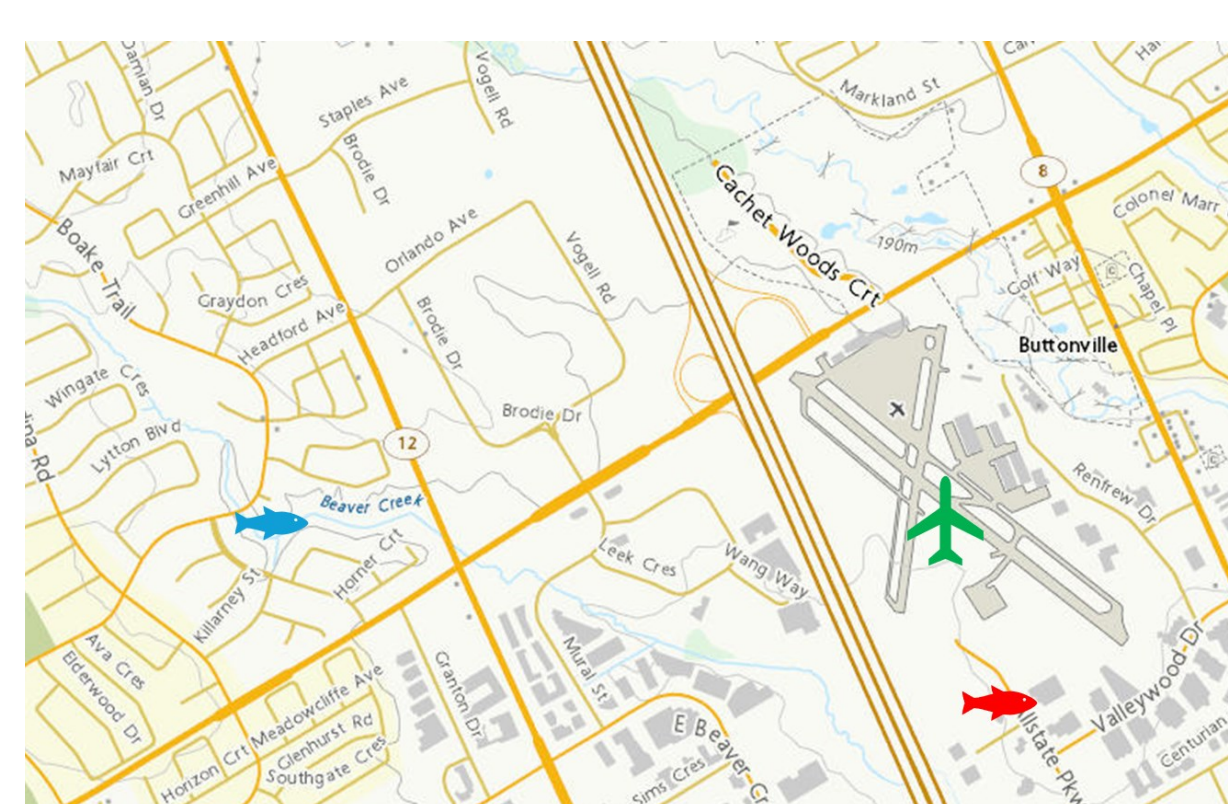
✈️ - Airport (Source)    🐟 - Reference (Control) Site    🔴 - Contaminated (Experimental) Site



Pearson Airport, Etobicoke Creek



Oshawa Executive Airport, Oshawa Creek

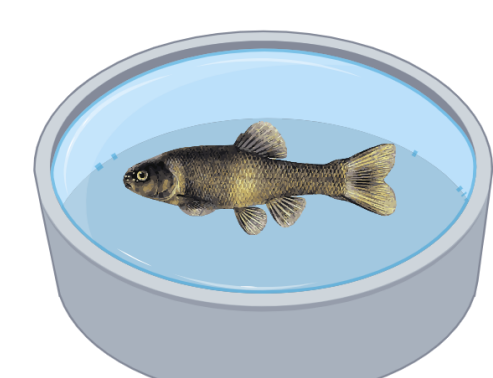
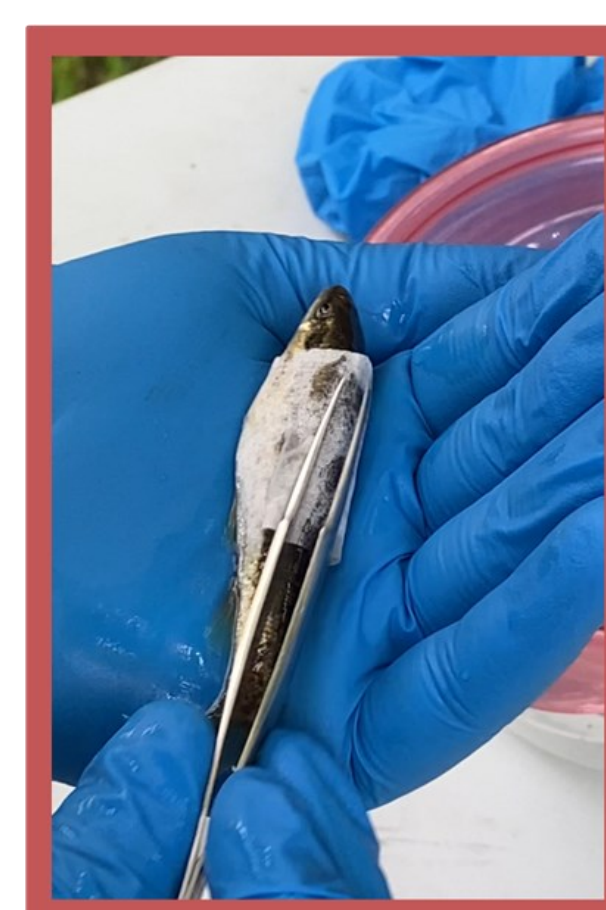


Buttonville Airport, Beaver Creek

#### Minnow Capture and Sampling

Three target species were selected; fathead minnow (*Pimephales promelas*), bluntnose minnow (*Pimephales notatus*), western blacknose dace (*Rhinichthys obtusus*), longnose dace (*Rhinichthys cataractae*).

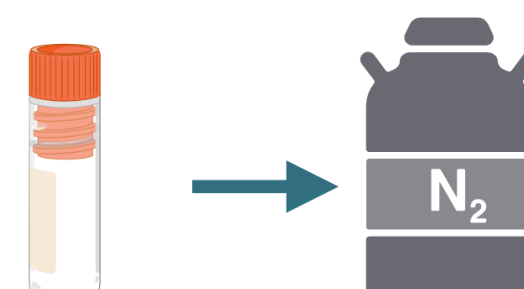
- Fish are baited into minnow traps using Vienna sausage and frozen corn, traps are left to "soak" for ~24hrs.
- Creek water is collected in 500mL amber bottles for non-targeted PFAS analysis.



Anesthetization  
(2ml 1:9 Clove-oil:Ethanol in 2L water)

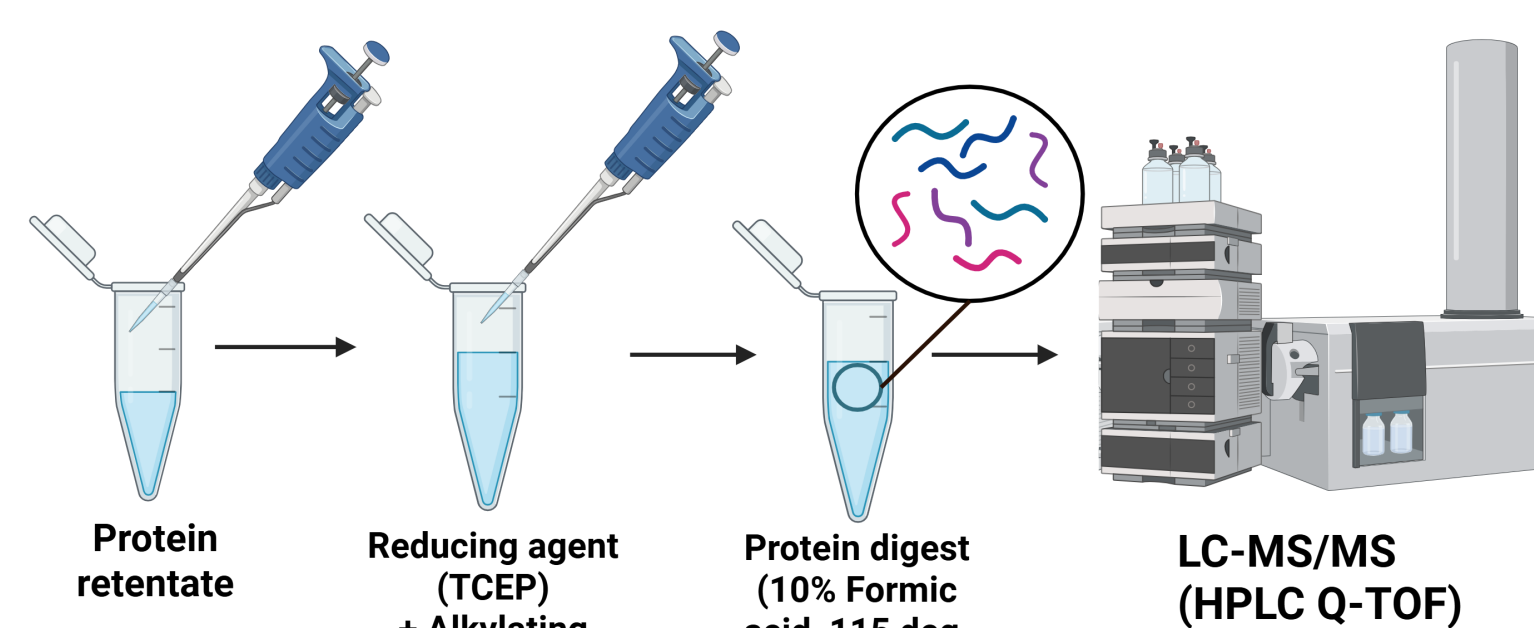


Mucus collection  
(1.5x1.5" kimwipe, 2-3 wipes per fish)

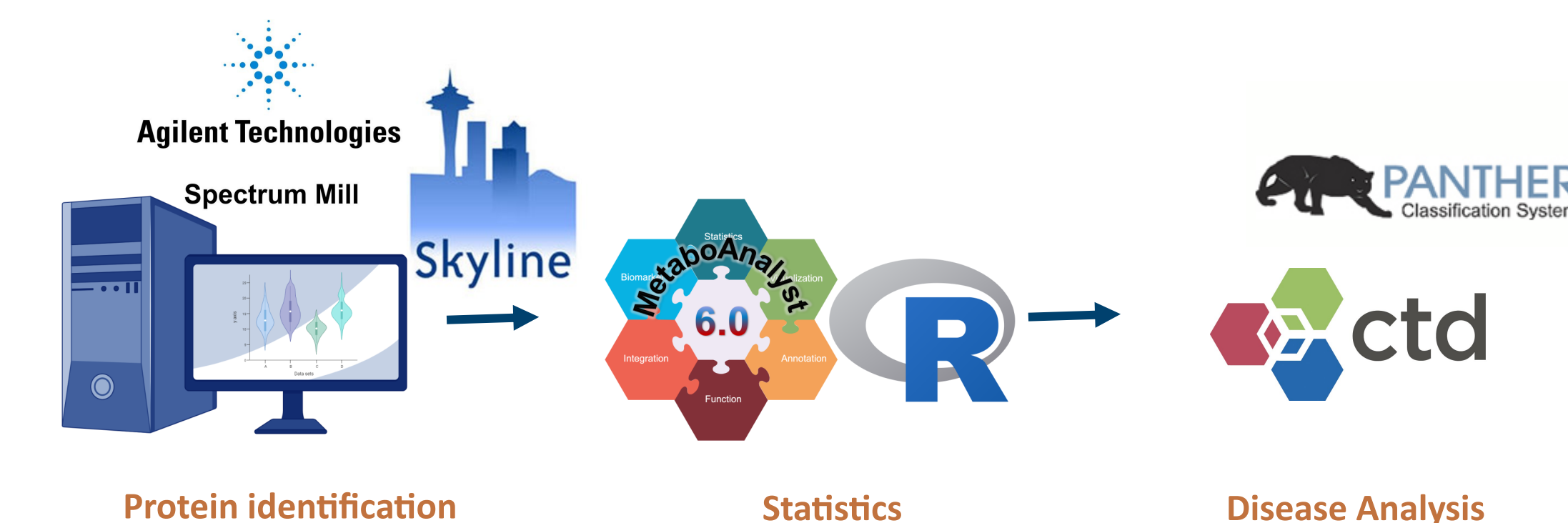


Flash frozen on site until  
storage in -80°C

#### Sample processing



#### Proteomics workflow



### Previous Results

The first component of this study involved a controlled exposure in a laboratory setting. From this study we were able to determine that mucus samples provide plenty of information on the health status of a fish!

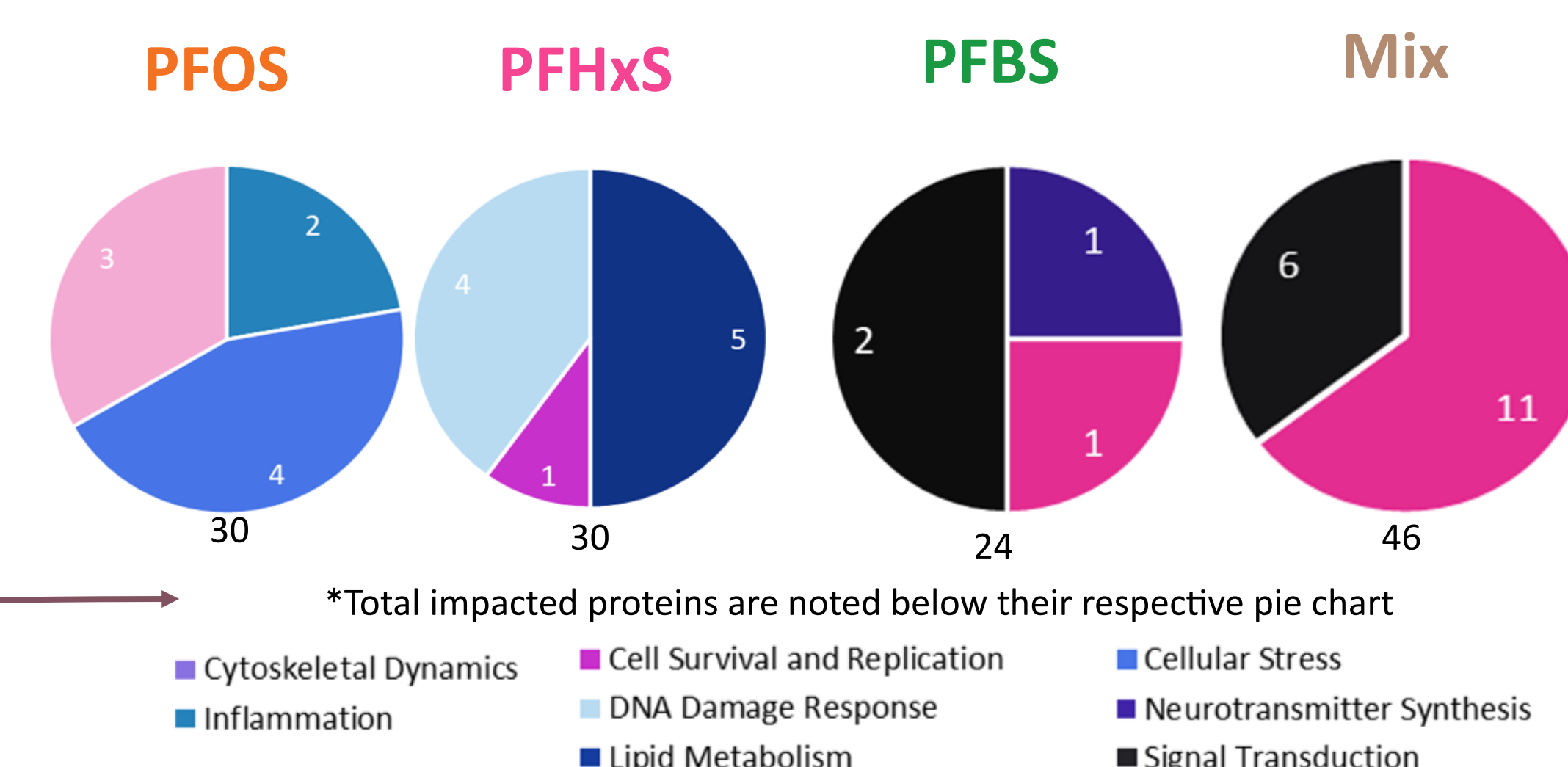
Mucus samples had:

- More impacted proteins in comparison to plasma and brain
- More impacted physiological pathways and disease outcomes in comparison to plasma and brain.

Disease outcomes detected in mucus samples. Each Pie chart represents a different chemical that was tested.

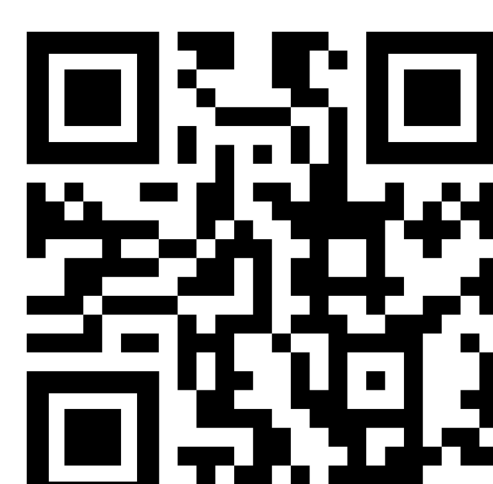
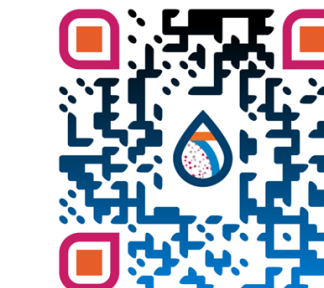
#### We hope to...

- Determine the toxicological/health impacts of AFFF containing run-off on wild fish.
- Compare laboratory results to field results to determine potential biomarkers of PFAS/AFFF exposure.
- Aid in the understanding and optimization of using mucus as a non-lethal assessment tool for biomonitoring and toxicity testing.



#### References

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