Brook Trout Environmental DNA Detections Comparable to Two Conventional Methods in **Southern Ontario Creeks**

REMTECH 2022

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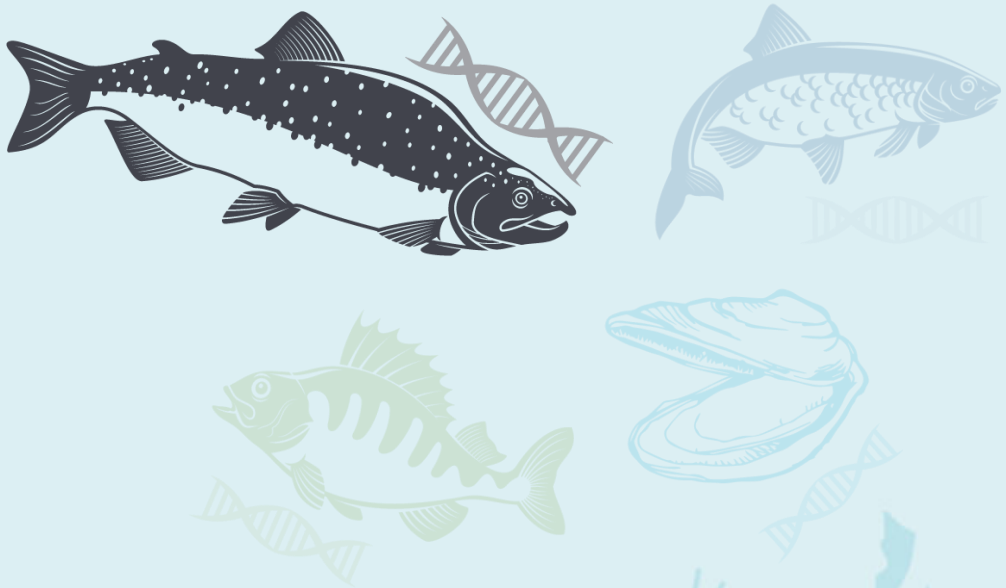
Overview

- Introduction to eDNA
- eDNA for Biomonitoring
- How eDNA Testing is Conducted
- Study Location & Overview
- Survey Techniques
- Results
- Conclusions

What is “eDNA?”

eDNA for Biomonitoring

eDNA can **characterize diversity** on several scales:



Species

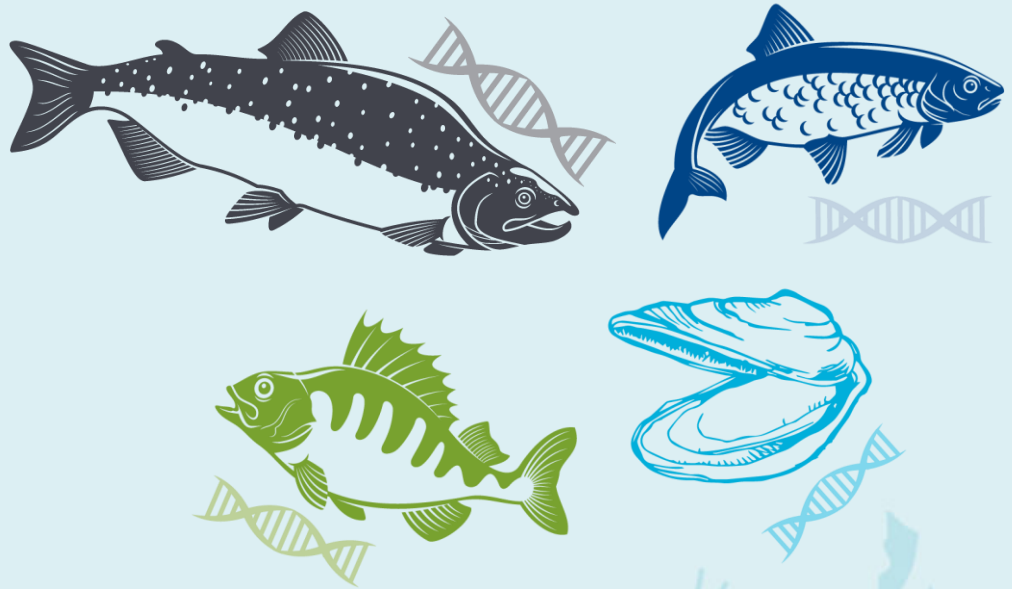
eDNA for Biomonitoring

eDNA can **characterize diversity** on several scales:

Population

eDNA for Biomonitoring

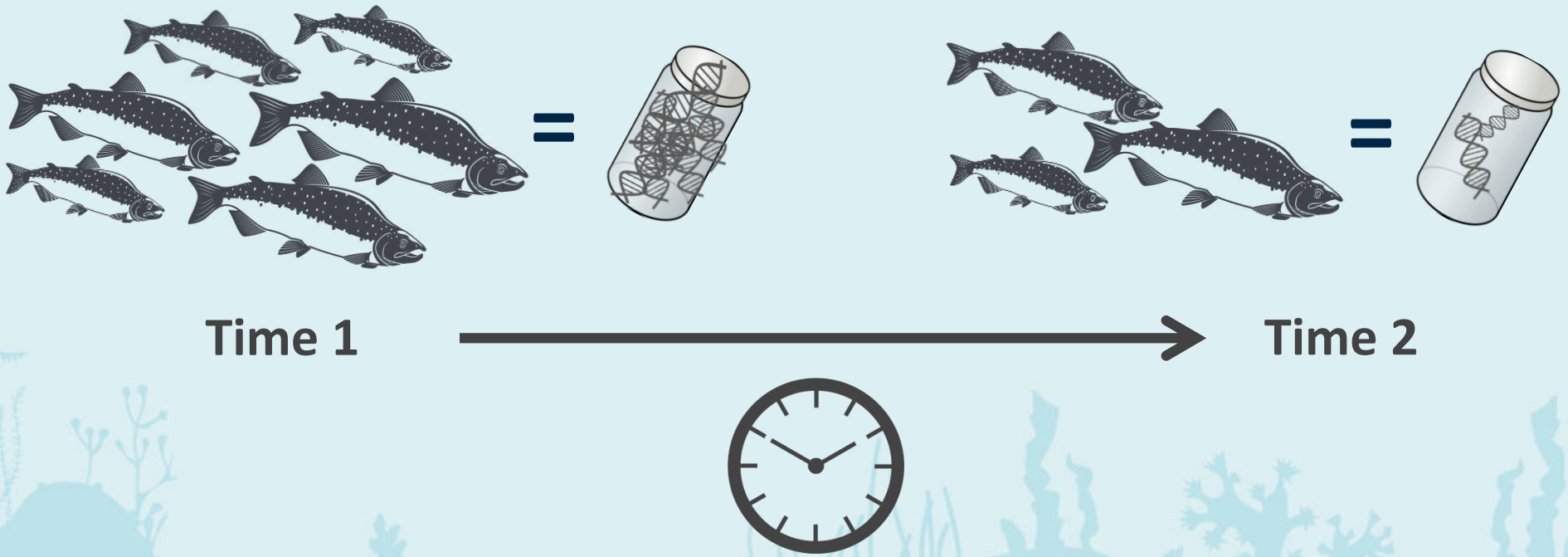
eDNA can **characterize diversity** on several scales:



Community

eDNA for Biomonitoring

eDNA can also reflect **seasonal** or **temporal changes** in relative abundance:



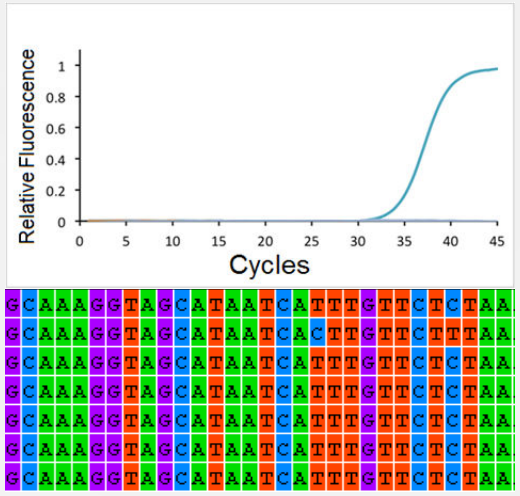
How is eDNA Testing Conducted?

Sample Collection

Typically by water filtration

DNA Extraction

Separate DNA from environmental matrices



DNA Analyses

For single-species or multi-species detection

Species Detection

Presence/absence or enumeration

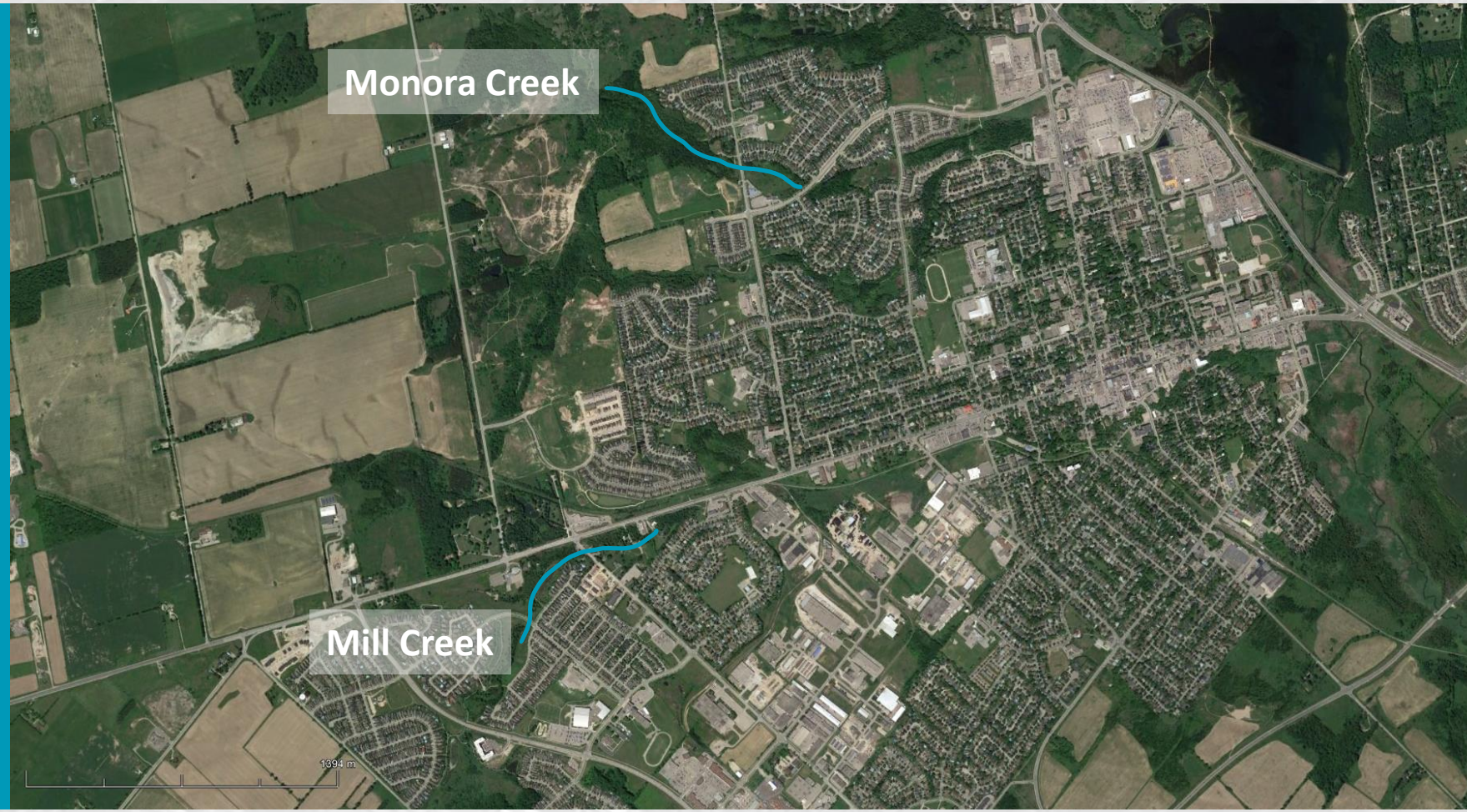
Study Location & Overview

Location: Orangeville, ON
- Mill and Monora Creeks
- Groundwater fed

Target: Brook Trout
- Ecosystem health indicator
- Utilize groundwater for spawning
- Sensitive to human disturbances

3 Monitoring Techniques
- Backpack electrofishing (July)
- Visual spawning surveys (November)
- eDNA Collections (Jul and Nov)

Duration: 2 years* (2020 – 2021)
- Monitoring began in 1997
- SLR since 2014
- Twice a year (July and November)



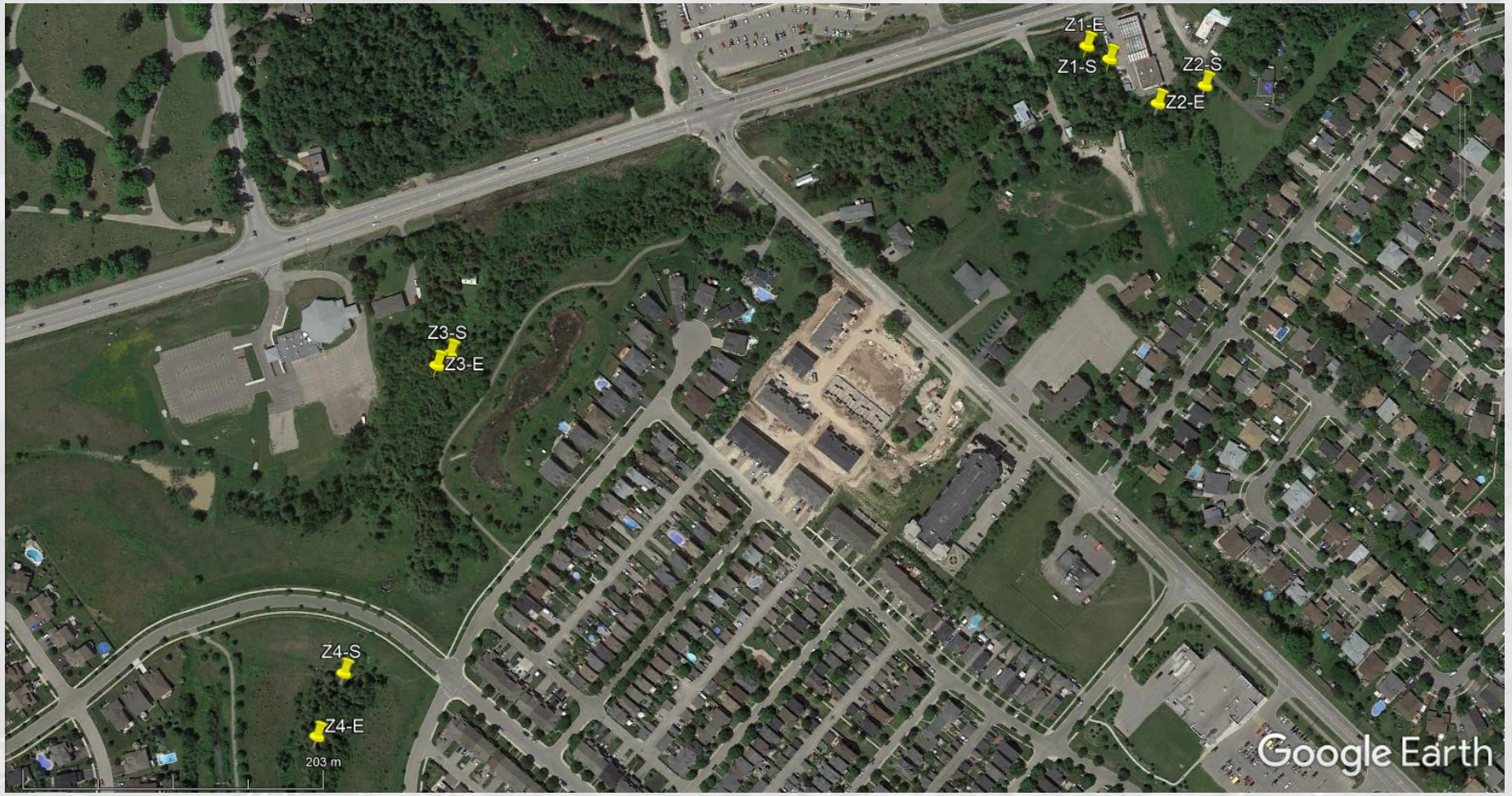
Survey Techniques

Survey Techniques: eFishing

- Conducted in July 2020 and 2021 **in tandem with eDNA**
- ~ 30 m zones
- x3 passes per zone
- Weight, total length, abundance recorded

Monora Creek: 6 Zones

Mill Creek: 4 Zones



Survey Techniques: Visual Spawning Surveys

- Conducted in November/during the spawning window **in tandem with eDNA**
- **CVC protocols**
  (scoring redd quality 1-3; High – Medium - Low)
- **Walk length of each creek**
  (in and between efishing zones)

Survey Techniques: eDNA

Sample Collection/ Filtration
(OSMOS or ANDe)

→

DNA Extraction
(QIAGEN DNEasy w. modifications for filters)

→

Multiplex qPCR
BRK2 assay (Wilcox et al. 2013)
+
Commercial IPC (Applied Biosystems)

↓

Magnetic Bead Inhibition Cleanup
(Based on IPC performance)

↓

Species Presence/ Absence

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Survey Techniques: Pros & Cons

eFishing

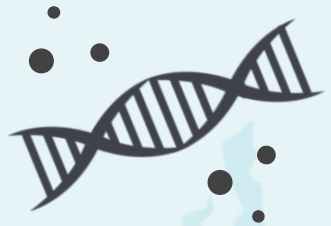
- ✓ Biomass, abundance, life stage, community data
- ✘ Stressful for fish
- ✘ Time-and labor-intensive for staff

Spawning Survey

- ✓ Direct observations possible
- ✘ Relies on accurate prediction of the spawning window and redd ID skills

eDNA

- ✓ Sight-unseen detection
- ✓ Non-invasive
- ✘ Specialized equipment and facilities required to process samples



Results: July 2020 Brook Trout Presence/Absence

Monora Creek		
Zone	eFishing	eDNA
1	✓	✓
2	✓	✓
3	✓	✓
4	✓	✓
5	✓	✓
6	✓	✓

Mill Creek		
Zone	eFishing	eDNA
1	✓	✓
2	✗	✓
3	✗	✓
4	✗	✗

Mill Z4:
Intermittent watercourse

No fish seen, or eDNA detections

Results: November 2020 Spawning Surveys (Monora)

Monora Creek Visit 1			
eDNA Sample ID	eDNA	Redds (Count x Qual.)	Fish Seen
MONS01	✓	N/A	✗
MONS02	✓	N/A	✓
MONS03	✓	N/A	✗
MONS04	✓	4 x 2	✗
MONS05	✓	1 x 2	✓
MONS06	✓	1 x 3	✓
MONS07	✗	N/A	✓
MONS08	✓	4 X 2	✓
MONS09	✓	N/A	✓
MONS10	✓	N/A	✗

Multiple instances of eDNA detections when no fish or redds seen nearby

- Transport of eDNA likely

Monora Creek Visit 2			
eDNA Sample ID	eDNA	Redds (Count x Qual.)	Fish Seen
MONSP1	✓	N/A	✓
MONSP2	✓	2 X 1	✓
MONSP3	✓	1 X 3	✓
MONSP4	✓	N/A	✓
MONSP5	✓	1 x 2	✓
MONSP6	✓	N/A	✓
MONSP7	✓	3 X 2	✗

Results: November 2020 Spawning Surveys (Mill)

Mill Creek Visit 1			
eDNA Sample ID	eDNA	Redds (Count x Qual.)	Fish Seen
MILLS01	✓	N/A	✗
MILLS02	✓	N/A	✓
MILLS03	✓	1 X 2	✓
MILLS04	✓	1 X 2	✓
MILLS05	✓	1 X 2	✓
MILLS06	✓	N/A	✓
MILLS07A	✗	N/A	✓
MILLS07B	✓	N/A	✗
MILLS08	✓	N/A	✓
MILLS09	✓	N/A	✓
MILLS10	✓	N/A	✓
MILLS11	✓	N/A	✓
MILLS12	✓	N/A	✗

Mill Creek Visit 2			
eDNA Sample ID	eDNA	Redds (Count x Qual.)	Fish Seen
MISP1	✓	2 x 3	✓
MISP2	✓	1 X 2	✓
MISP3	✓	1 X 2	✓
MISP4	✗	N/A	✗

Mill Z4 also not supporting Brook Trout during spawning season

Results: July 2021 Brook Trout Presence/Absence

Monora Creek		
Zone	eFishing	eDNA
1	✓	✓
2	✓	✓
3	✓	✓
4	✓	✓
5	✓	✓
6	✓	✓

Mill Creek		
Zone	eFishing	eDNA
1	✓	✓
2	✓	✓
3	✓	✓
4	✗	✗

Results: November 2021 Spawning Surveys

Monora Creek			
eDNA Sample ID	eDNA	Redds (Count x Qual.)	Fish Seen
21MONSP_01	✓	2 x 2	✓
21MONSP_02	✓	N/A	✗
21MONSP_03	✓	3 x 3	✗
21MONSP_04	✓	1 x 2	✓
21MONSP_05	✓	N/A	✗
21MONSP_06	✓	N/A	✓
21MONSP_07	✓	N/A	✓

Mill Creek			
eDNA Sample ID	eDNA	Redds (Count x Qual.)	Fish Seen
21MILLSP2_01	✓	2 X 3	✗
21MILLSP2_02	✓	2 X 2	✗
21MILLSP2_03	✓	1 X 1	✓

Suspected to have missed the spawning window, but Brook Trout eDNA detected from all tested sites

Conclusions

- Brook Trout eDNA is readily detectable in both creeks
- The OSMOS and ANDe instruments are both capable of collecting BKT eDNA
- eDNA consistently provided an additional line of evidence of BKT absence in intermittent Mill Z4
- eDNA presence/absence is not strongly linked to redd count or quality, likely due to eDNA transport by water and fish activity
- An IPC + cleanup protocol is essential for addressing environmental inhibition
  - 9% of our samples **showed negative BKT eDNA detection until a Magnetic Bead Cleanup was performed**

Thank You!

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