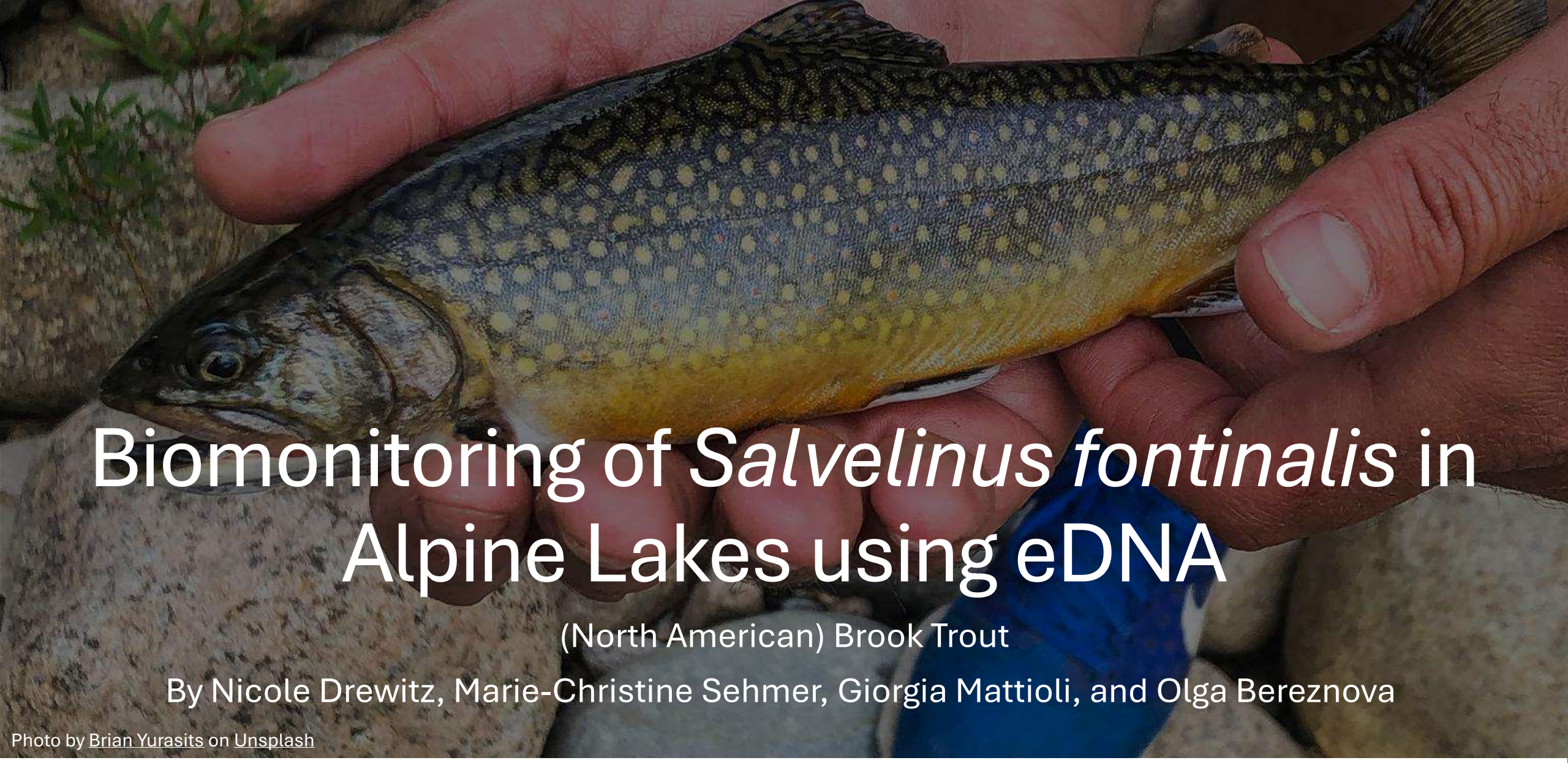


Biomonitoring of *Salvelinus fontinalis* in Alpine Lakes using eDNA

(North American) Brook Trout

By Nicole Drewitz, Marie-Christine Sehmer, Giorgia Mattioli, and Olga Bereznova

Photo by Brian Yurasits on Unsplash

Háskólasetur Vestfjarða
University Centre of the Westfjords

LIFE RESQUE ALPYR

EU CONEXUS
European University for Smart Urban Coastal Sustainability

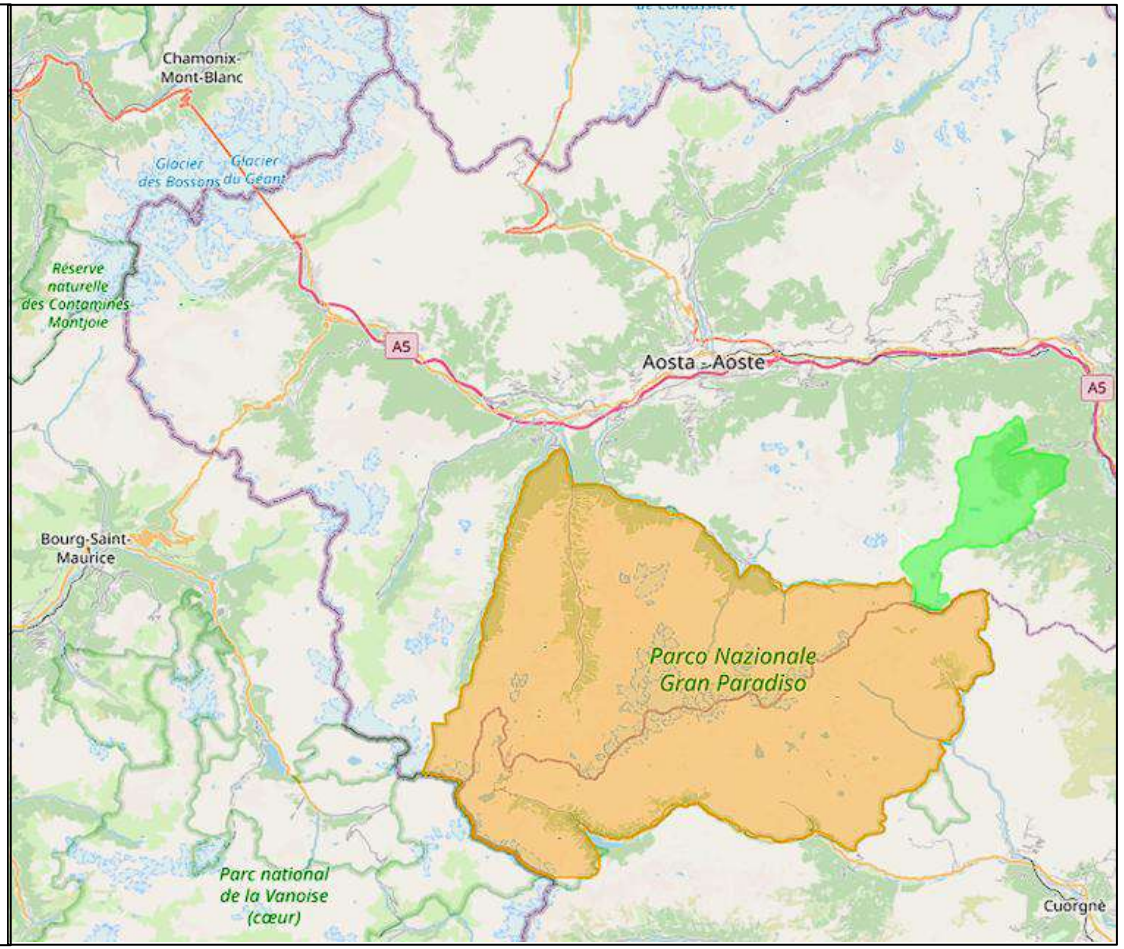
GuardIAS
GUARDING EUROPEAN WATERS FROM INVASIVE ALIEN SPECIES





Study area: Alpine Lakes

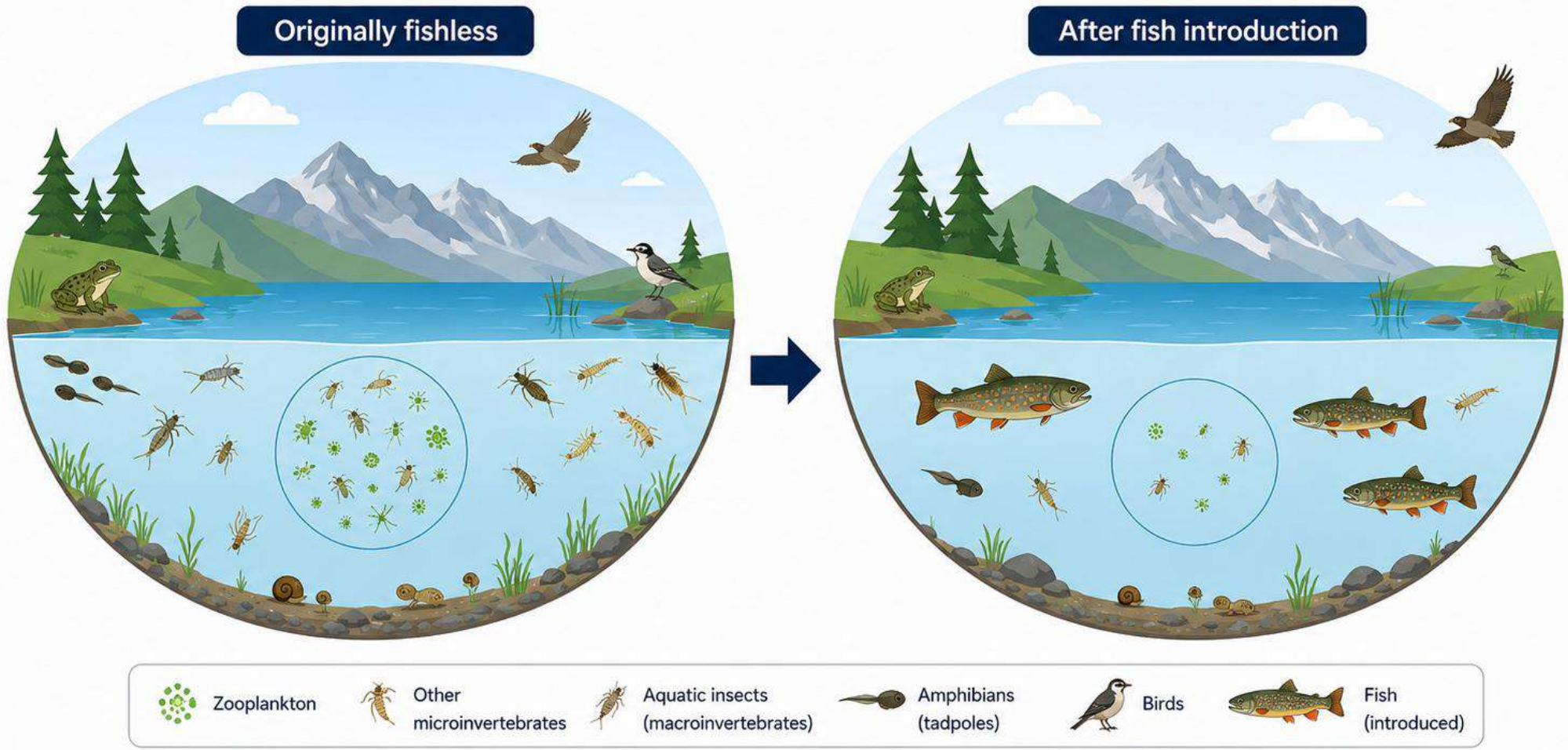




Gran Paradiso National Park
Mont Avic Park

The problem of fish introductions

Stocking fish into originally fishless high-mountain lakes
can trigger cascading effects through the food web.



Eradication methods



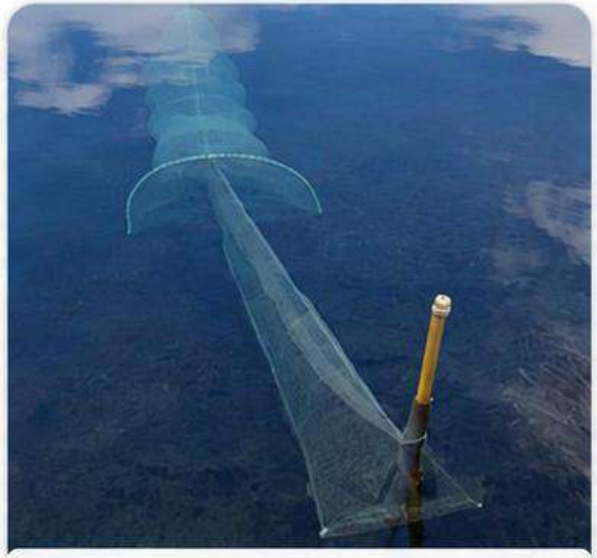
Rod angling

Targeted capture using rod and line.



Electrofishing

Stun and capture using electric current.



Fyke netting

Passive trap that guides fish into a one-way chamber.

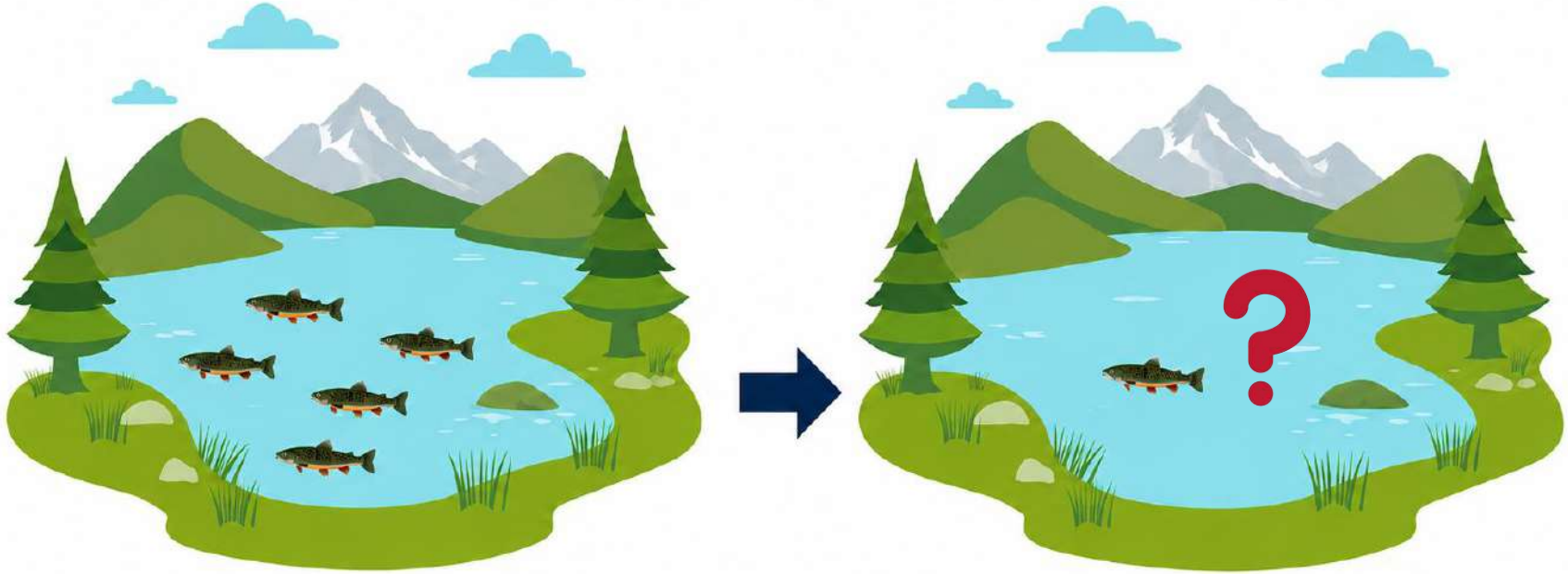


Gillnetting

Entangling net to capture fish by the gills.

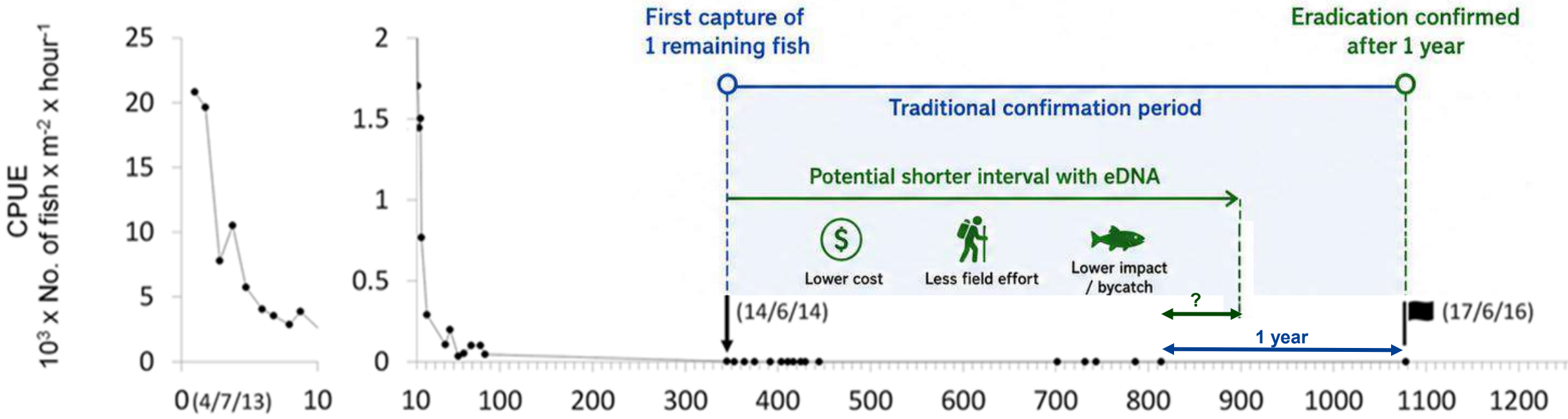
Challenge now: few remaining *Salvelinus fontinalis*

Can eDNA confirm eradication earlier than traditional surveys, reducing costs, field effort, and environmental impact?



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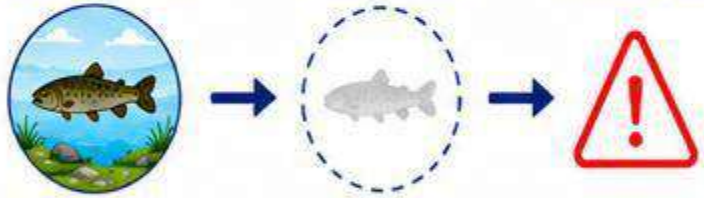
Occupancy models: what are they and how do they work?

They estimate the true presence of a species while accounting for imperfect detection.

1. Why are they useful?



Species present → Not detected → False negative



A species can be present but not observed during monitoring.

Occupancy models account for imperfect detection.

2. How are they applied?

A Repeated visits

Each lake is surveyed several times.



B Detection history

Lake	Visit 1	Visit 2	Visit 3
Lake 1	0	1	0
Lake 2	1	1	0
Lake 3	0	0	0

1 = detected, 0 = not detected

C Covariates



Altitude · Lake area · Maximum depth · Temperature

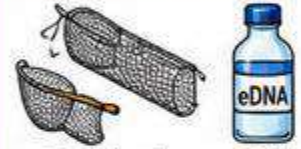
Occupancy model

3. What do we estimate?



a) Occupancy probability (ψ)

Probability that the species is truly present.



Traditional methods · eDNA

b) Detection probability (p)

Probability of detecting the species if it is present.



Traditional methods > eDNA

c) Method comparison

Compare detection probability of traditional methods and eDNA to identify the more effective method.

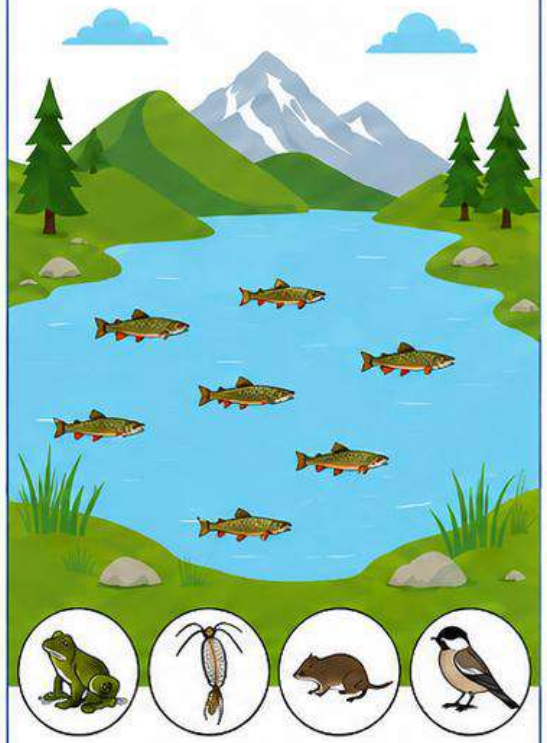


Result: distinguish true absence from non-detection and support monitoring decisions.



Key idea: repeated surveys help estimate true presence and compare which method is more effective.

1. Before eradication



- Brook trout present
- Ecological impacts on native communities
- Low-density survivors may remain difficult to detect

2. Eradication phase



Rod angling



Gillnetting



Electrofishing



Fyke netting

- Mechanical removal
- Repeated efforts across lakes
- Population strongly reduced

3. After eradication monitoring



eDNA sampling



Traditional surveys



- Post-eradication surveys
- eDNA used as a complementary method
- Sampling after reproductive period
- Compare eDNA and traditional methods

4. Main outputs

A Validate complete eradication

B Detect residual brook trout

C Estimate detection probability (occupancy modeling)

D Reduce time, effort, and invasiveness of monitoring

Goal: confirm whether eradication succeeded and improve monitoring of low-density trout populations.

Field work and sampling protocol

- **6 alpine lakes** in Valle d'Aosta
- **Monthly sampling** between June and October for 2 subsequent years
- **5 L water** filtered per sample from selected locations around the lake and middle of the lake (merged subsamples)
- **3 samples per lake:** 2 independent replicates, 1 field blank



Field work and sampling protocol

Filter kit and peristaltic pump





Sampler for deep water



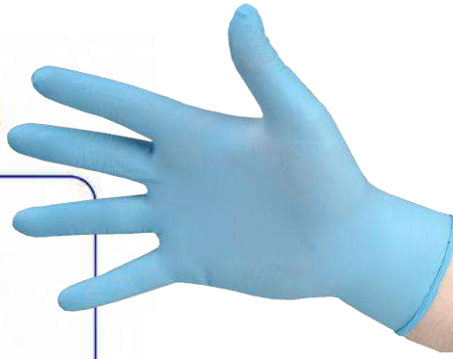


https://www.naturemetrics.com/kit-instructions/maxi-aquatic-edna-kit-pump-filtration

Laboratory workflow for detecting *Salvelinus fontinalis* eDNA

Qiagen DNeasy Blood & Tissue Kit





1 Filters from fieldwork

Keep cold / freeze

2 DNA extraction

Extract DNA from filter

3 Endpoint PCR

Species-specific primers for *S. fontinalis*

4 Electrophoresis + result

Detected

Not detected

Check expected fragment

F: Salfon-S715, 5'-CCTCCCGCCCTCCTTTCTA-3'
R: Salfon-A715, 5'-TGCCAGCTAAATGTAGGGAAAAA-3'
(Thalinger et al., 2021)

Optional alternative: qPCR for more sensitive detection

PCR master mix + F + R + H_2O

DNA extracted from *S. fontinalis* fin tissue

Controls

Field blank

PCR negative control

Positive control

Bioinformatics

1

Generate FASTQ files



Illumina sequencer

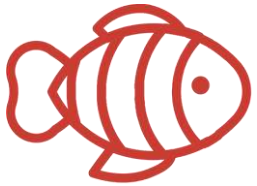
2

Group into ASV with R

a. check read quality
a. filter and trim
b. dada2 package for ASV (Amplicon Sequence Variant) grouping

3

Assess results with MitoFISH BLAST



Web database is simpler for non-bioinformatics researchers to also understand (e.g. existing managers)

4

GBIF and GenBank

GBIF
Global Biodiversity Information Facility

Adherence to FAIR data principles via GBIF and GenBank uploads following their eDNA protocols.

Convention on Biological Diversity

Italian **regulatory agencies** identified on the **Convention on Biological Diversity** website will be **contacted** whenever possible **through prior relationships** with the **existing management program.**



Results will be further interpreted via known Brook trout ecology, occupancy models, and comparisons with the existing monitoring program.

References

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- Thalinger, B., Pütz, Y., & Traugott, M. (2021). Endpoint PCR coupled with capillary electrophoresis (celPCR) provides sensitive and quantitative measures of environmental DNA in singleplex and multiplex reactions. *PLoS One*, *16*(7), e0254356.
- Tiberti, R., Bogliani, G., Brighenti, S., Iacobuzio, R., Liautaud, K., Rolla, M., ... & Bassano, B. (2018). Recovery of high mountain Alpine lakes after the eradication of introduced brook trout Salvelinus fontinalis using non-chemical methods.

Guide to using eDNA to eradicate fish in streams

Environmental DNA Sampling Informs Fish Eradication Efforts: Case Studies and Lessons Learned

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