eDNA Sampling in Waterways – *an introductory guide*

This resource is to help you understand the advantages/limitations of using eDNA sampling in waterways, and guide how you can best use this new environmental sampling tool.

A brief introduction

The genetic information for every living organism is stored within a molecule called DNA (deoxyribonucleic acid). It acts as a blueprint for how the organism will look and function.

Each species has short DNA sequences unique to that species that can be used for identification. A full copy of an organism's DNA is in nearly every cell, which means a single cell can identify what species the organism belongs to – even if the cell is no longer attached to the organism, i.e., it comes from material that has dropped off or been released by an organism.

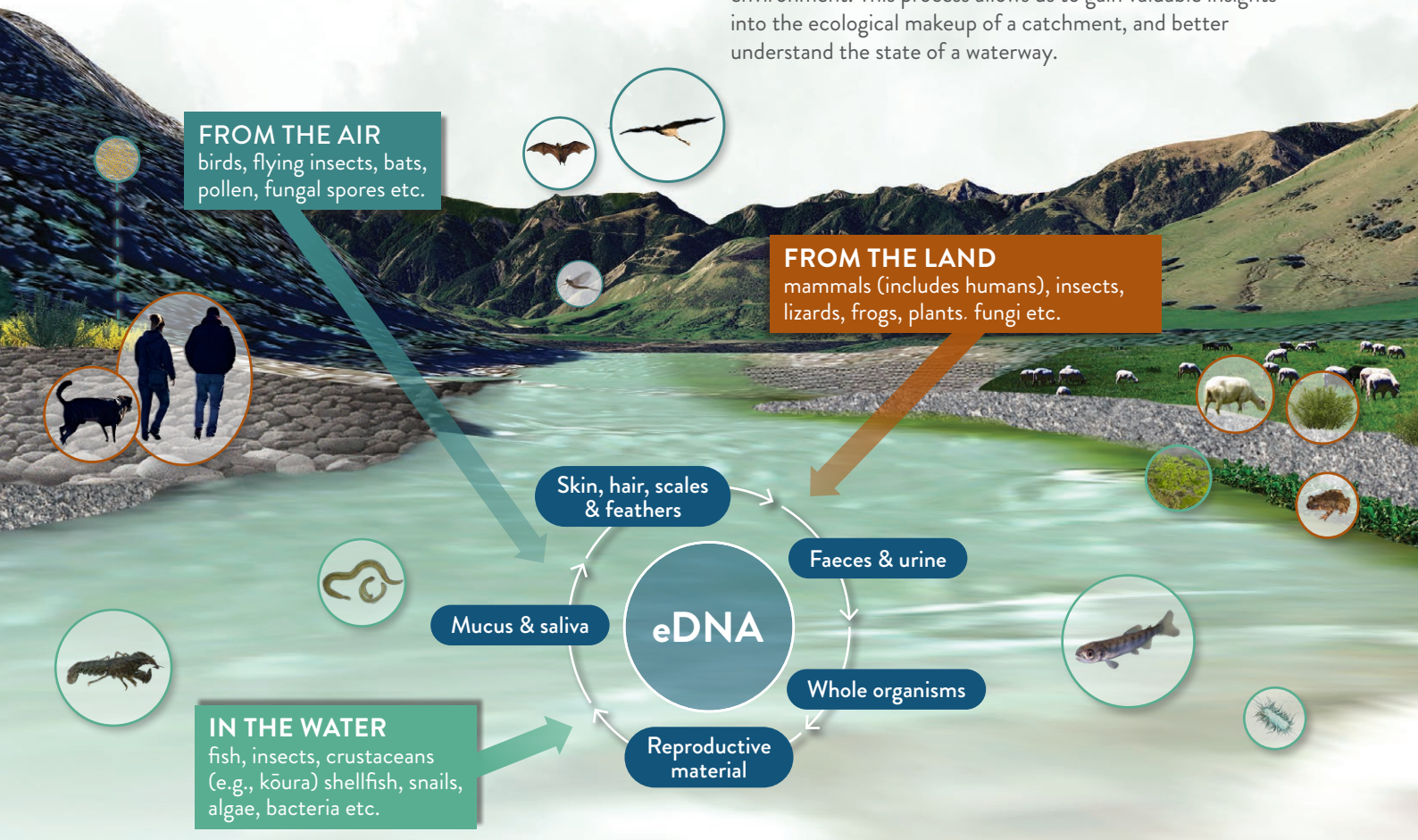
DNA shed by an organism into water, soil or air while going about its day is called environmental DNA, or eDNA.
The infographic below illustrates this concept.

How eDNA helps us understand a catchment

Our waterways are a soup of genetic material – collecting and transporting eDNA released by organisms living in the water, the air, and the land near the water.

Scientists extract eDNA from water samples using specialised filters. Specialist lab equipment amplifies and sequences the eDNA strands found in the samples, and compares them to a huge reference database of known 'barcodes' (short, unique DNA sequences) for species in New Zealand.

If a section of an eDNA sequence matches a known species barcode, we can confirm that species is/was present in that environment. This process allows us to gain valuable insights into the ecological makeup of a catchment, and better understand the state of a waterway.



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Advantages & limitations of eDNA sampling

Biodiversity is a measurement of the variety of species that live in an area.

Field surveys have traditionally been used to measure biodiversity, which requires manually catching, identifying, and counting all the organisms found in that environment.

While there will always be a place for these traditional methods, eDNA is a newly developed tool that can complement field surveys. eDNA sampling has many advantages, but it won't tell you everything you may want to know about what's living in your catchment. It's essential to understand the limitations of eDNA and what the data will and won't tell you.

ADVANTAGES:

Cheaper, more efficient & more accessibility

Less time and expertise are required for eDNA sampling than traditional methods, and lab processing costs are decreasing, making it more affordable for community groups and citizen scientists.

Comprehensive biodiversity measurement

eDNA samples genetic material from all types of organisms, providing a more comprehensive measure of biodiversity. It is also better than traditional surveys at detecting elusive and small species or those that recently left the area.

Less invasive data collection

eDNA collection causes minimal habitat disturbance, making it suitable for fragile/rare habitats and species.

Versatile in challenging conditions

eDNA collection is minimally invasive and can be used in more challenging conditions (e.g., difficult-to-reach areas, small streams, thick vegetation or conditions with time constraints), providing an alternative where traditional surveys may be impractical.

Wider river-reach sampling

eDNA has demonstrated the ability to detect the genetic material of freshwater fish approx. 4 km downstream of where the organism shed the genetic material, providing a broader assessment area relative to traditional surveys.

Potential future data refinement

Once collected and sequenced, eDNA data can be stored indefinitely – meaning the expansion of reference databases allows for retroactive ID of currently unidentified sequences, improving data outputs in the future.

LIMITATIONS:

Presence/absence indicated only

eDNA can only confirm the presence/absence of species and provides no quantitative information (i.e., it can't tell you how many individuals of each species there are).

Limited duration of detection

eDNA is only detectable for a short period, and the time frame is influenced by environmental conditions (e.g., detection rates may decline after heavy rainfall or due to other environmental factors).

Underestimation of biodiversity

The eDNA reference database may not include barcodes for all NZ species and the short time DNA is detectable in the environment may result in some species going unrecorded.

Overestimation of biodiversity

Human influence (e.g., sewage discharges) can introduce foreign DNA, potentially leading to overestimating biodiversity. Fluctuations in water flow can transport eDNA from upstream, which may also influence perceived species distribution.

Risk of sample contamination

eDNA samples are vulnerable to contamination during collection. Proper sampling practices (i.e., wearing disposable gloves and using clean equipment) are crucial to minimising contamination errors.

Sample processing errors

Despite recent advancements, lab processing errors can occasionally occur. Ongoing efforts to expand species databases aim to enhance accuracy.

Limited personal experience

eDNA sampling offers a minimal personal experience with catchment organisms. Supplementary traditional surveys and community engagement activities can provide a more holistic understanding of catchment ecology.

Collecting eDNA in your catchment

WHY – Purpose & goal setting:

Clearly define why you're sampling and determine the questions you want to answer with eDNA data. This will guide your site selection, timing, and method choices.

Example questions – do you want to...

- do an overall biodiversity assessment?
- find out species presence in areas inaccessible to traditional surveys?
- target areas for detailed traditional surveys?
- look for the presence of threatened, taonga, or pest species?
- compare biodiversity between catchments?
- identify fish migration barriers?
- find out the effects of contamination on local biodiversity?

WHERE – Select sampling sites based on defined purpose:

- Preferably, choose fast-flowing, well-mixed water and avoid stagnant pools for more accurate results.
- Be mindful of potential contamination sources and select
- sites several kilometers downstream from, or upstream of, the contamination source to avoid interference (unless you're studying the impact of contamination).
- Consider spacing sites several kilometres apart to effectively sample larger river reaches.

WHEN:

- You can collect eDNA samples at any time of year, although summer is best for comprehensive biodiversity sampling due to increased aquatic species activity and DNA shedding.
- 1 December–30 April is ideal for detecting freshwater fish species using traditional survey methods, and the same period would work well for eDNA sampling.
- Avoid sampling within 2–3 days of rainfall (more than 10 mm) to prevent DNA dilution or washout.

HOW – 2 methods commonly used:

Which method is right for you depends on the questions you want to answer, the environmental conditions, and how easily you can access your site. Each method requires specialised equipment, which must be sourced before sampling. It is recommended you collect 6 samples (replicates) per site for both methods to ensure the most comprehensive results.

1. Active sampling

- Uses a large syringe to hand-filter water through an eDNA filter. Use a caulking gun to push water through the filter, as it can be difficult and tiring to do by hand.
- Takes approx. 5–10 minutes per sample.
- Most commonly used in NZ for its effectiveness in most water conditions.
- Use in clear water with moderate flow, avoiding stagnant pools.

2. Passive sampling

- eDNA filters are submerged into the waterway current and secured there. All 6 replicates can be set up at the same time.
- Leave the filter in place for 24 hours.
- Good option for waterways with excess fine sediment or for sampling immediately after a rainfall event.
- Has a high detection rate for terrestrial species. Commonly used by predator-free groups around NZ to identify areas where trapping is needed and investigate the effectiveness of existing pest eradication efforts.
- May not be ideal if access to your site is problematic as it requires two visits (one to set up the filters, one to retrieve them).

TIPS for...

maximising detection:

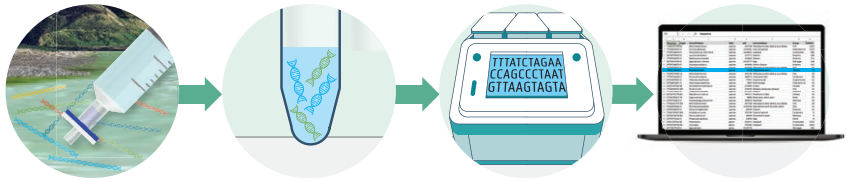
- *Collect multiple replicates per site to enhance species detection. Six replicates are recommended based on a national study completed by Wilderlab in collaboration with New Zealand regional councils.*
- *Preserve samples quickly after collection to prevent DNA degradation.*
- *Avoid sampling after heavy rainfall to maintain sample integrity.*
- *Consider multiple samplings throughout the year and across various sites for migratory or seasonal species.*

minimising contamination:

- *Wear disposable gloves to minimise foreign DNA contamination.*
- *Use new or sterilised equipment to prevent contamination. To sterilise, use a 10% bleach solution or soak equipment in hot, soapy water and thoroughly rinse with clean water.*
- *Avoid entering the waterway if you can. But if you must, enter downstream of your site and walk up (to avoid disturbing the bottom). Face upstream to sample.*
- *Start sampling the furthest downstream site first, then work your way upstream. This means you don't contaminate downstream sites before you've sampled them.*

Processing eDNA samples

Sending your eDNA samples to a specialised laboratory is crucial for proper processing. The labs listed to the right are the ones currently known to provide eDNA services to the public. They have the equipment and expertise needed to extract, amplify, and sequence the genetic material filtered from your water sample. They compare the sequenced DNA to a reference database to identify the species or taxonomic group, providing valuable information about your catchment's biodiversity.



eDNA sampling through filter | DNA extraction & amplification | PCR analysis & sequencing | Species identification

eDNA data ownership

Consideration of eDNA data ownership is essential.

Ownership and access depend on factors such as group affiliation, site location, and funding sources. If your project is publicly funded, data sharing may be expected. For private or culturally significant sites, discussions with landowners or mana whenua are necessary. Te ao Māori considers DNA from native species as taonga, emphasising the importance of consultation and respecting tikanga.

CURRENT PROVIDERS OF eDNA SERVICES IN NEW ZEALAND

Wilderlab
www.wilderlab.co.nz

Wilderlab specialises in eDNA testing, offering several types of user-friendly sampling kits and is the primary supplier for the general public in New Zealand.

Cawthron
www.edna.cawthron.org.nz

Cawthron Institute provides eDNA single-species testing services for various applications, including fish species detection, fish diseases, harmful algae, and marine biosecurity.

Ampersand Technologies
www.ampersandtech.co.nz

Ampersand Technologies is a start-up led by Massey University staff, focusing on agricultural, horticultural, and environmental applications of eDNA.

eDNA – *One tool among many!*

Remember, eDNA is just one tool among many for studying catchments. While it offers insights into biodiversity, combining it with traditional survey methods like visual assessments, kicknet sampling, eletrofishing, trapping/netting, or spotlighting enhances our understanding of freshwater catchments.

Further information:

- Find a more detailed report – 'eDNA Sampling in New Zealand Waterways – an Introduction' – in the Resources section at www.waiconnection.nz.
- NIWA has also produced a good eDNA guide – www.niwa.co.nz/sites/niwa.co.nz/files/eDNA%20guidelines%20report_12122023%20FINAL%20%283%29.pdf
- The EPA has also produced an eDNA guide – www.epa.govt.nz/assets/Uploads/Documents/Wai-Tuwhera-o-te-Taiao/Designing-your-eDNA-project_final.pdf

URLs are current as at April 2024.

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