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Looking Further and Deeper into Environmental Protection, Regulation and Policy Using Environmental DNA (eDNA)

Abstract

DNA sequencing technologies are transforming how environments are monitored. In this article, we pose the question: is environmental DNA (eDNA) the tool that Aotearoa New Zealand needs, but does not yet realise it does? The step change with eDNA is that genetic 'breadcrumbs' left behind in the environment can identify every living thing, from microbes to mammals, thus providing a more nuanced and holistic lens on ecosystems. Using eDNA, we can explore the biological networks that underpin healthy environments. Here we explore whether changes in policy setting, guidance, or pathways for uptake of eDNA are needed. Can eDNA help us make better decisions, inform policy and protections, track restoration, and act as a deterrent to reduce environmental harm?

Keywords eDNA, biomonitoring, environmental protection, environmental policy, ecosystems

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The field of genetics is developing quickly

The use of real-time genomics has played a central role in Aotearoa New Zealand's ability to track and trace outbreaks of Covid-19 around the country (Jelley et al., 2022). Just a few years ago, this would not have been possible. To put the speed of change into context, the first-ever human genome was announced in the year 2000, having taken about a decade to be completed at a cost of approximately US\$4 billion (International Human Genome Sequencing Consortium, 2001). The same can be done now for about US\$1,000 using a benchtop instrument the size of a microwave. While these DNA sequencing instruments that unravel the A, T, C and Gs¹ are transforming medical genomics and tracking the evolution of viral variants, they are also, using environmental DNA (eDNA), catalysing a change in how environments are monitored, protected and restored.

Pick up any recent New Zealand state of the environment report (e.g., Ministry for the Environment and Statistics New Zealand, 2022) and read it alongside the recent Intergovernmental Panel on Climate Change (IPCC) report (IPCC, 2021) – it's a sobering read. New Zealand's land and ocean ecosystems are increasingly under stress, and we are all to blame, directly or indirectly. We don't contend that we can live without impact, but most of us, Māori and Pākehā alike,

would agree that there are environmental bottom lines that have become, at best, a little fuzzy and, at worst, ignored.

At the core of this problem is the fact that it is difficult to make decisions when you cannot measure or track biotic impacts, especially when relating to ecosystem health. In his 2019 report Simon Upton, the Parliamentary Commissioner for the Environment, lamented the fragmented nature of environmental reporting across the motu and advocated for dedicated research funding and more joined together thinking (Parliamentary Commissioner for the Environment, 2019). How can our team of five million respond to this challenge? While arguably not as immediate

biodiversity at a given site (the basis of ecosystem-based monitoring, EBM), but do not have the expertise to identify everything we might find. Added to this, some organisms can be difficult to identify without sacrificing them. Enter eDNA.

The morphological features of an organism are not its only identifiers; inside the cells of each organism lie its genetic code. Akin to a barcode on any supermarket item, there are DNA regions (known as DNA barcodes) that can definitively distinguish one species from another. From some parts of our genomes we can tell individuals apart (for example, forensic DNA analysis conducted at crime scenes), but DNA barcoding works at a higher level

The use of distinct species as biological indicators has long been established; for example, the often-cited canary in the coalmine idiom. But as we broaden our ability to identify taxa, the question of what combinations of species are the best barometers (across a variety of potential disturbances) comes into play. The easy solution is to measure everything; however, until eDNA came on the scene, this was impractical from both logistical and financial perspectives. While eDNA still can't measure 'everything', it can measure a wide diversity of biota from which many indicators can be selected and then refined. Figure 1 provides a window into what is now possible using eDNA recovered from just a few litres of river water. While this 'tree of life' does not capture all the diversity present in the waterway (the bacteria and viruses are missing, but could be added), it gets far closer to an ecosystem-wide picture, and thus opens the potential for us to be able to measure, monitor and better understand the biological networks that underpin a range of environments.

The 2020 National Policy Statement for Freshwater Management specifically emphasises this need, in stating that we must 'recognise the interconnectedness of the whole environment, from the mountains and lakes down the rivers to hāpua (lagoons), wāhapū (estuaries) and to the sea' (New Zealand Government, 2020, p.13). Environmental DNA has the ability to respond to this challenge, but for it to be utilised to its full potential, an overhaul of existing monitoring approaches and reporting is likely required.

Why we need eDNA

The reason eDNA is gaining traction around the globe (Compson et al., 2020) is because it places a 'Swiss army knife' within our environmental monitoring tool kit. Like all tools eDNA has limitations, but it also has multifaceted applications. Take, for example, the scenario where the Environmental Protection Authority (EPA) might want to explore the impact of a given chemical, 'X', on the environment. While it might be straightforward to measure the concentration of chemical X in, for example, a river, the more nuanced (and biologically meaningful) approach might be to explore how the biota of

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as a pandemic scenario, the ongoing decline in ecosystem health is also in need of a 'surveillance strategy' and science-informed interventions to limit, and perhaps reverse, impacts.

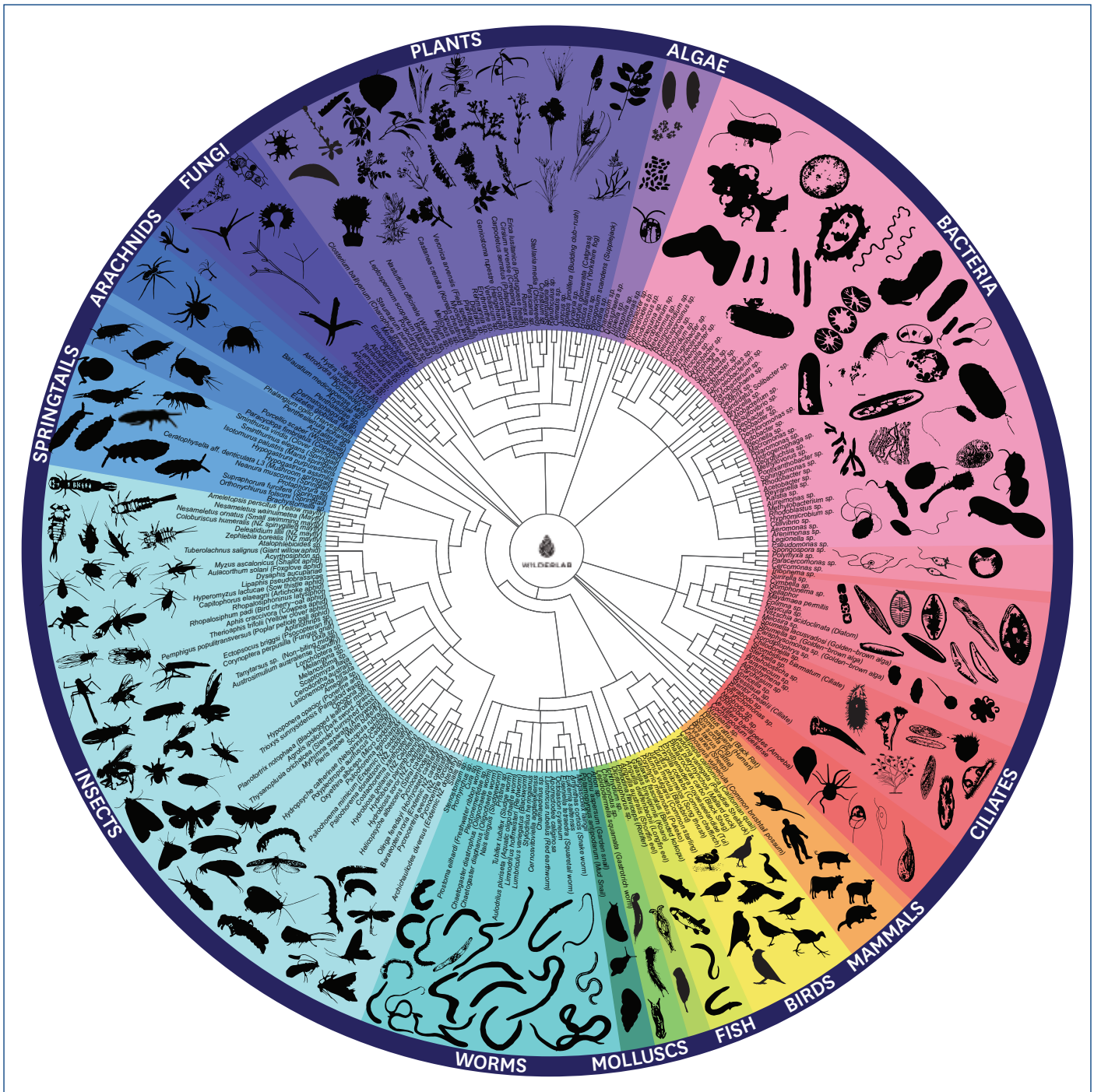
From morphology to molecules

For hundreds of years, the way we have monitored the animals and plants around us has followed, through necessity, a 'catch, look and (sometimes) kill' approach. We literally catch our target and look at it via field surveys (or, more recently, using cameras). This approach has served us well for centuries as we have attempted to catalogue the huge diversity of life on our planet. However, such an approach has limitations – among them, the need to become an expert across a wide range of taxa. While the 'twitchers' out there might be able to identify any New Zealand bird from a hundred paces, such a skill is beyond most people. However, those same expert twitchers would struggle to identify insects from a nearby stream. Increasingly, we want to look at a wide spectrum of

than this: it is about telling species apart. In most cases, a small segment of DNA just a few hundred A, T, C and Gs in length can, for example, distinguish all the mammals in New Zealand, from native bats to invasive stoats. As an example, here is a short, but unique, DNA barcode for the long-tailed bat (pekapeka-tou-roa): TTTAATTAACCTAACTTACATGACCATA TACACTCTCTATAAGAAATAACAC AAACATGATTAAGTTAGCAATTTAG – which is very different from that of any bird, despite the bat controversially winning the 2021 Bird of the Year contest (Forest & Bird, 2021).

By combining the power of morphology, which sets up the taxonomic playing field, with insights from DNA, we have developed a pathway to building more complete inventories of biota. The importance of this is paramount: put simply, we cannot confidently protect what we cannot measure. Moreover, if we measure the wrong things and make decisions on the basis of these data, we might not be doing the environment any favours.

Figure 1: An eDNA ‘tree of life’ recovered from 6 litres of water from Pāutahanui stream by the Mountains to Sea Wellington educational community group (sampled on 15 April 2022 at the coordinates –41.098943, 174.990792)



Source: Wilderlab

that river is changing in response to the rising amount of chemical X. For example, maybe chemical X is ecotoxic to one type of insect that is a core food for a native fish. Alternatively, it would be possible to use eDNA to rapidly detect the point at which a given ecosystem reaches a chemical tipping point that might be detrimental to the biota and/or the underpinning food webs.

To cite a real-world example, researchers at the Cawthron Institute have developed

an eDNA index of when an aquaculture facility (depositing nutrients into the sea) might be approaching nutrient levels that are detrimental to the surrounding environment (Pochon et al., 2020). These same researchers are also developing a better eDNA biosecurity safety net to quickly detect invasive marine species at our ports (Bowers et al., 2021), and using eDNA to assess the health of lakes across 10% of the lakes within Aotearoa (see the eDNA section of Cawthron’s Lakes380

project: Cawthron Institute and GNS Science, n.d.). In the medical space, the Institute of Environmental Science and Research (ESR) has also used eDNA (actually eRNA) to detect SARS-CoV-2 in our waste water to track not just the amount of viral RNA, but also the variants.

As with the science of anthropogenic climate change, the science of eDNA is settled. It is a powerful tool that has the potential to change how we monitor environments around the globe. Why, then,

is there a lack of urgency to deploy this new technology? To extend the climate change analogy (a bit) further, we would suggest that it is largely because it requires some changes in how we do things, and such changes never come easily. Arguably, it involves dialling back some things (for example, morphological-based surveys for benthos or invertebrate surveys for routine monitoring) and learning new ways. It may also involve deploying our environmental monitoring toolkit in a different order.

Environmental DNA, as an environmental monitoring or compliance tool, is fundamentally simpler than having to undertake physical counts or sampling, but requires a technological laboratory

significant shake-up in the way we collect samples and generate and store data. New Zealand could be leading the way in this area, but we need to address the fragmented nature of environmental funding and reporting to do so (Parliamentary Commissioner for the Environment, 2019).

Getting the eDNA ball rolling: how best to communicate the eDNA revolution

In the last few years, in the shadow of a pandemic, science has been very much in the public eye; thinking back, perhaps not since the Apollo missions have we witnessed such widespread interest in science. Throughout the Covid-19 pandemic, science has again come into

embarked on a new eDNA-based mode of environmental engagement.

Our approach has been surprisingly simple: we let people use eDNA at a place that means something to them, namely their own backyard. In 2020, the EPA partnered with Wilderlab (a commercial eDNA provider) to launch Wai Tuwhera o te Taiao – Open Waters Aotearoa. It was our attempt to get the eDNA ball rolling. We figured that if eDNA could capture the interest and imagination of the communities, iwi and hapū around New Zealand, it could be the catalyst needed to trigger a wider shift in how we monitor our precious waterways, taonga species and wider ecosystems.

The response to the programme has been overwhelming (you might want to explore an eDNA sample from a waterway close to you – at www.wilderlab.co.nz/explore). It turns out that New Zealanders have the eDNA exploration gene, and you only need to put a syringe filter in their hand for their eDNA journey to begin. Without exception, the eDNA data prompts the next set of questions: Can we get more tests? Can eDNA tell abundance? How long does the DNA last? Can we use eDNA to track changes over time? And can we use it to monitor the impact of 'X'? Anecdotal reports from councils confirm that they are being asked by their communities to adopt these eDNA approaches after gaining a glimpse of the power of eDNA to reveal the huge amount of biological diversity hidden in their own backyards – from taonga species to bacteria that even Google will struggle to provide information on.

Through Wai Tuwhera o te Taiao, the narrative we are hearing is that, when it comes to environmental monitoring, we need to change what we are doing. Every year our report card seems to get worse, yet we think that the status quo will suffice. We advocate that it is time for environmental practices (and policy settings) to catch up with the technologies, including eDNA (and remote sensing), and that these data types need to start informing better predictive models.

In turn, these models should underpin our decision making and rapidly shine a spotlight on the trajectories of the environments we are all charged with protecting. The universality of the genetic

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'back end'. Some practitioners, especially those in more traditional environmental consulting, may resist this new technique as a threat because they don't yet have the know-how or connections to the right laboratories to enable processing and interpretation of their samples. The arguments that eDNA technologies are 'unproven' or 'experimental' or that it is 'too early to implement' are ever present. This is the gauntlet that the new techniques often have to run; the international literature (reviewed in Compson et al., 2020) is now full of exemplars that demonstrate the utility of eDNA across a wide variety of applications. There are even moves afoot to make the data much more accessible (Berry et al., 2020). Some good reading on this topic is by the US Environmental Protection Agency's John Darling, who wrote the paper 'How to learn to stop worrying and love environmental DNA monitoring' (Darling, 2020).

There is an urgent need to monitor our environments more efficiently and holistically across many biological domains, including drinking water, waste water, rivers, oceans, soils and air. This requires a

the spotlight, with commentators like Siouxsie Wiles, Michael Baker and Ashley Bloomfield becoming household names. Readers may also remember University of Otago professor Neil Gemmill's mission in 2019 to use eDNA in the hunt for the Loch Ness monster. The aim of Gemmill's project was not really to find monsters; it was to promote eDNA as a technique for exploring and recording biodiversity, using as an example a story that might engage people and excite their imagination.

Within central and local government an all-too-common response to our explanations about eDNA technology (and its potential) is that it is 'magic' and 'too good to be true' and 'too experimental'. Rather than undertake further academic research (the literature on eDNA is growing exponentially) or write position papers, we decided that the first step should be to generate a groundswell of understanding, curiosity and support, with a focus on iwi and hapū. After wānanga on eDNA (including sharing of data) within the EPA's national Māori network, Te Herenga, and with Ngā Kaihautū Tikanga Taiao (the EPA's statutory Māori advisory body), we

code (A, T, C and G) might also serve to 'defragment' the environmental monitoring system (the challenge set by the Parliamentary Commissioner for the Environment) and get New Zealand to generate datasets that are truly comparable across time and space.

To flesh this out a little more, while there might not be policies or practices that prevent or block the uptake of eDNA, neither is there a clear pathway to promote their uptake. The small footprint of eDNA within the National Science Challenges is a case in point. We advocate that local and central government, including the EPA, signal more clearly a shift towards the uptake of this new generation of biomonitoring tools. The eDNA ball is starting to roll, albeit slowly: recent eDNA pilots led by Waikato and Hawke's Bay regional councils to explore the utility of eDNA as a fish monitoring tool (compared with electrofishing) have been successful (David et al., 2021) and prompted a nationwide pilot at around 45 sites across New Zealand. Likewise, an eDNA 'barometer' has been approved for use in aquaculture environmental monitoring after years of benchmarking by Cawthron (Pochon et al., 2020).

Start with a few drops of water

The poet and philosopher Kahlil Gibran once wrote: 'In one drop of water are found all the secrets of all the oceans.' With eDNA, this vision is coming to life (although experimental design dictates that we need a few more replicates than a single drop). The power of eDNA to profile the biota from a few litres of water is astounding (again, see Figure 1). However, this is nothing compared with the insights that can be obtained from time-stamped data. Put simply, time is often the missing data from our environmental decision making. Without good baseline data, how is it possible to observe change? And how can we attribute a given activity to the change in biota as opposed to natural variation?

The absence of baselines has, without a doubt, clouded many a debate on environmental impact, or lack thereof. The 2021 *Policy Quarterly* article by Mike Joy is a case in point (Joy, 2021). What are the natural levels of nitrate in each of our rivers? How are these numbers changing? Looking through an eDNA lens, we might also ask the question: at what level of nitrate are the underpinning biotic

networks beginning to shift, and are these shifts temporary or permanent? Without a time machine, it's impossible to know.

We contend that a set of environmental samples systematically taken through time, where changes in biological communities can be observed, would likely have achieved a more complete picture of the impact of nitrate levels on ecosystem composition/health. We simply do not have water, sediment or air samples, let alone the eDNA profiles, going back in time, but perhaps we could start now? Indeed, we advocate that the archiving or 'biobanking' of environmental samples (for example,

As a field it is maturing, with an increased understanding of sample collection, storage, workflows, false positives/negatives, contamination and data accessibility. This maturation is needed if eDNA is going to withstand the scrutiny of (often contentious) environmental decision making. The legal scrutiny might even be stepped up a few notches if eDNA were used in legal proceedings in the areas of environmental compliance, monitoring and enforcement.

There is nothing in the current New Zealand environmental legislative framework that we believe will prevent the application of eDNA as a regulatory

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filtered water or soil) or the DNA extracts is a key part of any eDNA solution and should perhaps be front and centre of environmental policy reform.

In a move that might surprise some, a number of global resource companies are taking and storing environmental samples for their own baselines so that, in the event of an incident (for example, an oil spill), they can assemble an enhanced picture of the 'before and after' biota. Whether these biological snapshots are for insurance purposes or to truly do the 'right thing' for the environment, there are increasingly compelling arguments for bioarchiving facilities. Should New Zealand be exploring environmental sample archives? Is it part of our journey towards better environmental stewardship and kaitiakitanga?

From decisions to deterrents

On the global stage eDNA is already informing environmental decision making,

control or monitoring technique. While there will be a need for policy work around some aspects (e.g., sample archiving), the EPA is seeking participants with environmental footprints to 'sign up' to the use of eDNA for baseline and ongoing monitoring of the impacts of their activities.

There are a lot of parallels between eDNA analysis and its genetic cousin, forensic DNA analysis. In much the same way as forensic DNA analysis has transformed modern criminal investigations, eDNA will, if given sufficient support, funding, and stature as a biomonitoring tool, be a catalyst in transforming the environmental sector.

As a technique, forensic DNA analysis started off as a research tool, but was quickly adopted by forensic labs across the globe. Hard lessons were learnt about controls and contamination and the need for standard operating procedures. Over about a decade around the turn of the century, forensic DNA cemented itself as a

cornerstone of the crime-fighting toolkit. Forensic DNA analysis continues to be innovated on, refined, and adapted to the social context in which it is applied.

One final parallel between eDNA and forensic DNA is in the area of deterrents. Research suggests that increasing the likelihood of getting caught for a crime has a bigger impact on future behaviour than changing the severity of a sentence. Once offenders know their DNA profile is on a central database, they are less likely to commit a crime. What we find surprising is that, for each convicted felon profile added to a US DNA database, a cost saving of between US\$1,500 and \$20,000 was realised (Doleac, 2017). Such is the impact

not occur in the first instance, or can at least be detected more rapidly. Indeed, in the future, environmental approvals might stipulate that environmental samples be collected and stored in a bid to create the necessary deterrent. Such an approach, especially if environmental data is shared, may have the added benefit of reassuring the public that environmental footprints are being monitored and that robust data underpins a decision to start, stop or control activities with a footprint on the receiving environment. Rather than be seen solely as a 'big stick' approach, this might also provide companies with the social licence they need to continue or modify their operations to minimise environmental harm.

anecdotally, some of our scientists, consultants and policymakers see the move into a DNA 'world' as being a radical departure from the status quo, and one in which a degree of retraining, time and investment is required.

Is the uptake of eDNA an issue of policy setting, slow implementation, or both? The 2020 National Policy Statement for Freshwater Management specifically mentions techniques, of which eDNA is not currently one. Likewise, it is difficult to envisage a shift into archiving of environmental samples occurring without a change in policy that enables the samples to be collected and stored. The Lakes380 project is showing the power of this approach, using eDNA in sediment cores to travel back in time to understand how lakes have changes over the past millennia.

Abroad, large initiatives, such as the European Union-funded DNAqua-Net project, have turbocharged the eDNA field and provided researchers with the legal, regulatory, policy and quality assurance/control frameworks that an eDNA toolkit needs to comply with. There are discussions around forming a southern eDNA society across Australasia to build both capacity and cohesion. The EPA's Wai Tuwhera o te Taiao eDNA programme in the community is about building bridges between people and the environment; the Lakes380 project has similar aspirations by connecting people to the wellbeing of lakes across the motu. We hope that these programmes, and others, are the catalyst needed to build further bridges into the policy and environmental management space. The benefits of a concerted shift into eDNA are many – cost, speed, data transferability and resolution among them. But there are also issues to discuss. Who has sovereignty over the data? (a topic debated by the Waitangi Tribunal (Waitangi Tribunal, 2011)). In many respects the same question could be asked of existing environmental survey data; should eDNA data be any different? What can data be reused for? And do guidelines (Hudson et al., 2021) formulated around sequencing entire genomes of native taonga apply to short barcodes recovered from environmental samples?

ESR's ability to sequence a whole SARS-CoV-2 genome in a few hours (contributing to pandemic contract tracing) is a prelude

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of a good deterrent. Might this hold true for environmental crime as well?

In much the same way as a drug tester can turn up at the house of an elite athlete to take a sample, the same system might be used for an eDNA test at, for example, a discharge point on a river. Unless caught in the act or via whistleblowers, environmental crime has been difficult to prove, and even more difficult to determine are the short- and long-term impact(s) on the receiving environment. Environmental DNA-based surveys, coupled with spot inspections, might provide some much-needed evidence to prosecute those who chose not to follow the rules. In some applications, the source of discharge may be difficult to pinpoint (for example, nitrates in agriculture), but in other applications (for example, aquaculture fish farms) the link will be clear.

Environmental DNA might also provide a way of tracking the progress of remediation efforts. Better still, samples held in a bioarchive could be enough to act as a deterrent so that environmental impacts do

The challenges and potential

With pending environmental policy reforms (for example, of the Resource Management Act), coupled with the recent National Policy Statement for Freshwater Management and the advice of the Parliamentary Commissioner for the Environment (on the fragmented nature of environmental reporting), we argue that the time is right for a shake-up of the types of environmental data we gather and how it is reported and shared. Further, we advocate that the power of eDNA is such that it needs a far greater presence in the environmental management landscape across New Zealand than it currently has.

The so-called 'catch, look and (sometimes) kill' approach to biomonitoring will always be present in the biomonitoring and decision-making toolkit, but there is overwhelming evidence that it is time for some of these functions to be complemented and/or replaced by eDNA. This position, we believe, is not controversial to the public, who likely see that a step change is needed. In contrast,

to the near real-time capability of these technologies to enable more rapid decision making. As DNA sequencing technologies get faster, cheaper, more portable and automated, the utility will improve further. While we are still some way off an environmental Star Trek tricorder device,² it is not the pipe dream it once seemed.

Ongoing efforts to sequence the barcodes of more biota from around New Zealand and the globe mean that we are rapidly developing the ability to assign every species' DNA barcode (noting that sequencing a barcode is not the same as compiling an entire genome). In other words, the data we generate today may become even more useful in the future. There are still eDNA challenges, to be sure, principle among them being the question

of how the abundance of DNA barcodes correlates to actual abundance in the sampled environment (in some applications, there are strong correlations; in others, the correlation is not so great).

In sum, we have written this article to highlight the potential applications of eDNA and to help shift thinking around environmental monitoring, policy settings and regulation. Rather than simply highlight the scope of the problem(s) across New Zealand, it is vital that we explore technological solutions that can address our poor environmental report card. Ideally, these solutions will provide pathways to better measure the impacts we are having on the receiving environment. We advocate that eDNA become part of this pathway.

Finally, whether, as a biomonitoring technique, you look at eDNA through a glass half full or a glass half empty lens, the ability of eDNA to sequence the microbes in your half glass might one day save you from drinking something you shouldn't.

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- 1 A, T, C and G are the 'building blocks' of DNA: adenine (A), cytosine (C), guanine (G) and thymine (T).
 - 2 The tricorder is a science fiction creation from Star Trek. It is a handheld prop that is used to scan environments to sense, record and compute multiple features of that environment.

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