A CREDIBILITY, RELEVANCE AND LEGITIMACY FRAMEWORK ANALYSIS OF ENVIRONMENTAL DNA FROM THE POINT OF VIEW OF POTENTIAL END-USERS

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ABSTRACT

As threats to natural resources multiply, the need for effective science-policy interfaces (SPIs) that account for the incorporation of ecological and social issues into decision-making is increasing. A feasible solution to successfully accomplish fast and reliable environmental information is to make use of advanced technologies, such as sampling genetic material shed by organisms into the surrounding environment, also known as environmental DNA (eDNA). While there are many studies on the scientific and technical aspects of eDNA, not much attention has been paid to users' and decision-makers' perspectives on eDNA and their implication in aquatic environmental monitoring and assessment. Aiming to verify if the eDNA tool and data generated by its use could make its way into decision- and policy-making, I conducted semi-structured interviews with potential end-users and stakeholders, and reviewed some Acts and Regulations, in order to better understand how eDNA is perceived and accepted. Results were interpreted and discussed using the credibility, relevance and legitimacy (CRELE) framework, as the balance among those criteria is seen as key to effective SPIs. Analysis demonstrated that eDNA is already seen as a relevant tool in inventory-monitoring studies, especially for preliminary assessments and targeted monitoring, but it would be applied as an addition to current techniques and programs, instead of a replacement option. The main challenges for eDNA relate to its resolution, its validity and its users. Although current legislation presents more opportunities than challenges for eDNA incorporation, better reproducibility and repeatability are necessary for strengthening eDNA's credibility and legitimacy. Hence, through a combination of social perceptions, regulatory information and ecological knowledge, this study enhanced scientists and decision-makers knowledge about the tool, facilitating the identification of relevant points to be addressed towards an improved SPI.

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POSITIONALITY STATEMENT

How we view and approach our research is linked to our life experiences, our values and our knowledge of the world (Pitard, 2017; Crouzat et al., 2018), even though this last one is "inevitably incomplete and situated" (Simandan, 2019, pg. 1). Regarding my values, the main examples I can see related to how I practice and see science are equity, integrity, justice, respect and responsibility. I once read that you cannot discuss sustainability without equity and justice. I believe a balance between those values is crucial. When considering my positionality, I feel like my strongest social characteristic that plays a role is my education. Throughout these 15 years in which I have been involved with the academic world, I have found my passion in aquatic environments, especially in the ichthyology realm, navigating from freshwater environments as an undergrad to marine ecosystems during a PhD. During all that time I have mainly been involved in quantitative research, but it felt good to be brought to different 'waters' (qualitative) by this current project. Aware of my standpoint and accounting for reflexivity, interview questions were formulated to have as little bias as possible and were designed to get the most of a participant's insight on the topic in a viable timeframe. Even though English is not my mother language, this didn't seem to affect the interview process and didn't result in inaccurate reporting.

In sum, I find myself most comfortably following a post-positivist paradigm, which holds that reality can be known only probabilistically, meaning that no single method or perspective can provide the answer, nor capture an external reality in its totality (McGregor & Murnane, 2010; Ponterotto, 2005). This paradigm has value for knowledge creation and evidence-based policy development, an essential starting point for interdisciplinary work (Phoenix et al., 2013; Ryan, 2006) - just like this research.

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CHAPTER 1

INTRODUCTION

"Current rates of extinction, habitat degradation and emerging challenges show that freshwater ecosystems already face pressures larger than any other ecosystem, and threats will intensify in future as the exploitation of freshwater resources grows to meet human demand."

(Reid et al., 2019, pg. 864)

Freshwater ecosystems are regarded as precious natural resources for their biodiversity and services provided, but that has not prevented them from suffering increased species extinction and high environmental degradation (Arthington, Dulvy, Gladstone & Winfield, 2016; Reid et al., 2019; Thackeray & Hampton, 2020). Building upon a highly cited review article on the subject (Dudgeon et al., 2006), Reid et al. (2019) classify the global pressures on fre shwater ecosystems as persistent and emergent. Overexploitation, water pollution and habitat degradation fall into the persistent category, while some examples of newly emergent pressures include e-commerce and invasions, harmful algal blooms, freshwater salinization, cumulative stressors and, of course, climate change (Reid et al., 2019). As threats to natural resources multiply, the need for effective science-policy interfaces that account for the incorporation of ecological and social issues into decision-making is increasing, along with a strong claim from practitioners for not only more reliable knowledge, but importantly, decision relevant and actionable information (Görg et al., 2016; Greig & Duinker, 2011; Treweek, 1995). In addition, because climate change can impact and amplify some of the previous mentioned pressures and threats (Arthington et al., 2016; Poesch, Chavarie, Chu, Pandit & Tonn, 2016; Reid et al., 2019) an "improved interjurisdictional integration" is suggested for successful management of freshwater ecosystems (Poesch et al., 2016, pg. 385). For example, Canadian Prairies areas, such as in Alberta and Saskatchewan, are particularly prone to droughts and reduced volumes of snow, while eastern Canadian freshwater ecosystems may experience changes in water temperature and a mismatch of phenology and life cycle, resulting in habitat loss for some species and increased availability for others (Poesch et al., 2016; Schindler, 2001; Singh, Pirani & Najafi, 2020).

Environmental policy demands a constant stream of information that enables it to perform effectively, so that any negative changes stemming from development are addressed sufficiently early and remedial and/or corrective measures may be applied to protect aquatic ecosystems and

the services they provide (Evans & Lamberti, 2018; Kelly, Port, Yamahara, Martone et al., 2014). Crucial to policy, environmental monitoring and assessments can provide the knowledge needed by different stakeholders at different levels of governance (Nichols et al., 2017). Environmental monitoring and assessments are distinct from each other, with environmental monitoring usually considering the "wellbeing of the whole system as its starting point", and environmental assessment focusing "on the effects and consequences of specific human undertakings" (Fleskens, 2017, pg. 1). Yet these processes exist in tandem, with activities sometimes linked or complementary (Kilgour, Dubé, Hedley, Portt & Munkittrick, 2007). Common challenges of environmental assessments and monitoring, that can reduce their efficacy in informing policy, are overlooking their multi-disciplinary nature (Arciszewski et al., 2017); mismatches among approaches and criteria, suggesting insufficient communication between actors (Bonada, Prat, Resh & Statzner, 2006); and the slow implementation of more diagnostic frameworks (Baird & Hajibabaei, 2012).

In order to respond to different information needs at different stages of decision-making and, at the same time, overcome current environmental monitoring and assessment challenges, stakeholders have been looking into emergent technologies, such as molecular techniques and methods (Fleskens, 2017; Nichols et al., 2017). Compared to traditional sampling methods (e.g., seining, bottle traps, core sampling), molecular methods, specifically DNA-based ones, offer several potential benefits to attend to the demands for timely information to support policy and management decisions, such as increased sensitivity, speed of processing, and reduced costs (Hering et al., 2018). One example of a DNA-based tool is environmental DNA (eDNA), a non-invasive procedure that involves collecting genetic material from the environment, such as in samples of water instead of from the organism itself, and then analyzing the samples to infer taxonomic composition and distribution in that environment. Already identified as having remarkable potential in conservation of inland waters in Canada (Pérez-Jvostov et al., 2019), eDNA opportunities extend beyond biological knowledge. For example, it has been demonstrated that eDNA can help in the control of zoonoses by providing different insights into the eco-epidemiology of Leptospira in irrigation water (Gamage, Sato, Kimura, Yamashiro & Toma, 2020); it can offer an indication of groundwater connectivity and areas at risk of contamination (Oberprieler et al., 2021); and it can reveal potentially harmful algae, associated with environmental, health and economic challenges, outside of bloom events and expected ranges (Jacobs-Palmer, Gallego, Cribari, Keller & Kelly, 2021).

There is a growing consensus that to solve current and future environmental problems we need to pay at least as much attention to social aspects, such as decision-making and stakeholders, as we do to natural science (Perrings, Duraiappah, Larigauderie & Mooney, 2011; Shackleton, Larson, Novoa, Richardson & Kull, 2019). A recurring question that afflicts scientists from many areas is "how can we effectively integrate social, economic and biological knowledge into effective decision- and policy-making" (Barclay et al., 2017, pg. 426). Something else to be considered is how to apply the large amounts of detailed genetic and genomic information into monitoring programs in a way that is useful for regulatory assessment, impact management, and decisionmaking (Pawlowski et al., 2018; Schenekar, Schletterer, Lecaudey & Weiss, 2020; Thackeray & Hampton, 2020). In recent years, Canada's government has directed increasing efforts to o vercome the disconnection between science information and policy analysis, including the establishment of a federal chief science advisor and the review of several federal Acts and Regulations. After some criticism that previous changes to federal legislation, such as the Fisheries Act and Canadian Environmental Assessment Act 2012, lacked scientific engagement and were even responsible for making environmental regulation weaker (Hutchings & Post, 2013; Roach & Walker, 2017), revised federal legislation and new commitments to science-policy integration, such as through science-based departments and agencies open data directives (Roche et al., 2020), present a policy window and perhaps an opportunity for the incorporation of new and improved scientific information in policy and regulatory decisions (Rose et al., 2017).

Hence, the purpose of this study was to identify the key barriers and opportunities for implementation of eDNA approaches in aquatic assessment, policy and management in Canada. To do so, this research engaged a range of stakeholders to explore their understanding of and their perceptions about eDNA, supplemented by a review of key aquatic environmental legislations. While its scope is limited to Canadian inland water ecosystems, related legislation and stakeholders, the research design can be replicated to include other environments and regions. The intended audience of this study includes end-users with a direct and indirect interest in monitoring and assessment of inland aquatic environments. The research question guiding this study is: how can the new environmental DNA (eDNA) methodology be incorporated into existing policies and management frameworks? Addressing this question also requires consideration of what type(s) of information is needed by regulators and other decision-makers in management of aquatic systems to

support decisions; and whether eDNA is accepted by end-users. Aiming to suggest improvements in policy and management, the specific research objectives were to:

- A. Build an understanding of the eDNA tool from a social science perspective, by examining end-user monitoring and decision support needs; and
- B. Identify opportunities and challenges for effective incorporation of eDNA in existing monitoring and assessment programs.

CHAPTER 2

LITERATURE REVIEW

"... efforts to mobilize S&T [science and technology] for sustainability are more likely to be effective when they manage boundaries between knowledge and action in ways that simultaneously enhance the salience, credibility, and legitimacy of the information they produce." (Cash et al., 2003, pg. 8086)

A common view among the conservation science community is the existence of a gap between knowledge generation and knowledge use (Choi et al., 2005; Gavine et al., 2018; Lawton & Rudd, 2014). A frequent complaint among decision-makers relates to the inability of scientists to match policy needs and translate research into feasible solutions (Gluckman, 2016; Sepulveda, Nelson, Jerde & Luikart, 2020). On the other hand, many scientists become frustrated with the policy-making process as participating in it has little academic value, requires their time and energy, and their results are often disregarded or not fully utilized (Engels, 2005). In addition to having different goals and different perceptions of time (Gavine et al., 2018), other issues leading to this discrepancy in opinions relate to the way both groups understand evidence, to whom they are accountable, and how the problems are ultimately structured (Choi et al., 2005; Engels, 2005; Fernández, 2016; Lawton & Rudd, 2014).

Furthermore, environmental problems do not respect political boundaries and, in many cases, may need similar evaluations and approaches conducted between different jurisdictions, leading to complex, challenging and sometimes suboptimal management (Lodge et al., 2016). In Canada, this situation can be aggravated due to the constitutional division of powers (Campbell & Thomas, 2002). For example, when looking into regulations for invasive alien species, Smith, Bazely and Yan (2014) concluded that the federal-provincial framework is disconnected and uncoordinated, resulting in ineffective legislation to deal with the issue.

Recognizing the importance of better linking the areas of science and policy, this literature review explores the science-policy interface and some quality criteria associated with it. This review section also focuses on eDNA, a new tool with the potential to complement traditional sampling methods and a viable option for supplementing monitoring and conservation programs

(Evans & Lamberti, 2018), possibly resulting in a more efficient and strategic use of resources and efforts.

2.1 Defining terminology

Recognizing the importance of defining terms to provide clarity and enable more effective communication, I deemed it necessary to start this section by explaining key terms found throughout this work, namely policy, legislation and regulation. While proper definitions are hard to come across, the descriptions provided here reflect my view on the terms after careful literature review.

As stated by Ball (1993), "the meaning of policy is taken for granted" (p. 10), with the term being used in different ways, to describe different things. The author explains that policy can be seen as things, processes, and outcomes, but also recognizes his limitations in providing a clear definition (Ball, 1993). Some other explanations revolve around policy being also "perceived as a pluralist, consensual process mediated by the state" (Blackmore & Lauder, 2005, p. 97), including several different dimensions (Torjman, 2005) and being subjectively defined, "involv[ing] behaviour as well as intentions, and inaction as well as action" (Hill & Hupe, 2002, p. 4). Due to its complexity, insights from several areas are used when theorizing about policy, such as sociology, psychology, law, and economics (John, 2012). In addition, policy is also used as a synonym for public policy, which "can be generally defined as a system of laws, regulatory measures, courses of action, and funding priorities concerning a given topic promulgated by a governmental entity or its representatives" (Dean G. Kilpatrick, electronic resource).

Just like with policy, legislation and regulation are hard to distinguish, with definitions connected to possible perspectives, such as having different sources or one being a subset of the other (Kosti, Levi-Faur & Mor, 2019). The perspective often employed by researchers, and with which I concur, is a distinction between legislation and regulation. Hence, "while legislation sets out the principles of public policy, regulation implements these principles, bringing legislation into effect" (Kosti et al., 2019, p. 171).

For this particular study, it helps to think about policy, legislation and regulations not as one versus the other, but as multiple levels in policy-making following a logical sequence (Brown, 2003).

"While government always retains the ultimate responsibility for the formulation of policy, it is best to delegate nuanced policy decisions, micro policy, to regulators. Doing so makes for less politicization, more predictability, more transparency, and more informed decision-making." (Brown, 2003, p. 1)

2.2 Science-policy interface

Science-policy interfaces (SPIs) are defined as social flexible processes denoting a variety of ways in which knowledge can be used to enrich decision-making (van den Hove, 2007). For example, the literature presents five ways through which knowledge may impact policy decisions, such as by co-producing information, shaping minds, building support, generating action and producing improved outcomes (McKenzie et al., 2014; Posner, Getz & Ricketts, 2016). This knowledge may also be used at different stages of policy planning and through different modes, such as instrumentally to make specific decisions, conceptually to expand knowledge and raise awareness, and *strategically* to support and promote policy options or justify previous beliefs (McKenzie et al., 2014; Posner et al., 2016). Knowledge may also interact with the decisionmaking process through different links, with these links differing mainly by the level of interaction exhibited between the areas of science and policy. When the source of knowledge and policy do not exert direct influence on each other, co-existing but still being independent of each other, it is said to be a static link (Giebels, van Buuren & Edelenbos, 2015). An interactive link, like the name suggests, is characterized by a connection between science and policy, but when the interaction occurs through a long process and there is co-production of knowledge it is said to be an *adaptive* link (Giebels et al., 2015).

A common mistake among environmental scientists is to ignore the complexity of the SPIs and focus on a linear approach, as if it was up only to them to feed policy demands, acting as information suppliers (Boswell & Smith, 2017; Crouzat et al., 2018; Fernández, 2016). An important point often overlooked by researchers is that scientific knowledge is not always the limiting factor in politics and that sometimes different types of knowledge are needed by policy-

and decision-makers (Fernández, 2016; Giebels et al., 2015; van den Hove, 2007). As stated by Lawton & Rudd (2014, pg. 855), "the reality, as seen from the 'other side' of the science-policy gap, is that evidence is one of a set of equally important inputs into societal decisions". A study engaging Canadian scientists and policy-makers identified that, according to the respondents, the top three current strategies to bridge the gap in SPIs are: 1) science-policy forums, 2) focus on policy, and 3) conferences (Choi et al., 2016). When asked about ideal strategies, the main ones were: 1) focus of policy, 2) policy briefs, and 3) science-policy forums. Some activities that rank high as current strategies, such as conferences (#3) and journal publications (#4), are not seen as ideal strategies (#12 and #14, respectively). The study also indicates space for more collaboration in study designs (#4) and in analysis (#5), as compared to the current situation (#9 and #12).

2.2.1 CRELE attributes

It has been widely suggested that the key for knowledge to be useful and usable, resulting in effective SPIs, is to balance three quality criteria: credibility, relevance (or salience) and legitimacy – referred to collectively as CRELE (Cash et al., 2002; van Voorn, Verburg, Kunseler, Vader & Janssen, 2016). Different definitions of these quality criteria are available in the literature, with different combinations of them resulting in three different "modes of scientific authority" that can be applied in sustainable development governance: assessment-oriented, advice-oriented and solution-oriented (van der Hel & Biermann, 2017, pg. 217). These are not mutually exclusive and the same researcher or institution, for example the Scientific and Technological Community Major Group, may combine different aspects of the modes of authority, seeking credibility, relevance and legitimacy through multiple strategies (van der Hel & Biermann, 2017). In this study, I better identify with the advice-oriented mode, where *credibility* is about the perception and trust of the knowledge and technical credentials by involved actors; *relevance* or *salience* refers to knowledge that is timely, appropriate and informative about societal needs and problems; and *legitimacy* relates to the fairness of the process through formal recognition, with it being transparent and considering multiple perspectives (Cash et al., 2002; van der Hel & Biermann, 2017).

Even though the application of CRELE attributes may enhance the acceptance of environmental assessments in policy, it also presents challenges. A key one is that while CRELE attempts a prescriptive application, it has been mostly used as an evaluative and descriptive tool,

with little discussion of how to actually apply the criteria to direct the SPI (Tangney, 2017). Other important challenges are that the pursuit of one criterion may counteract another, that different users may have conflicting views or perceptions about what constitutes relevant, legitimate or credible research and information, and that perception is also subject to change with time (Cash et al., 2002; Cook, Mascia, Schwartz, Possingham & Fuller, 2013; van Voorn et al., 2016). For example, in a study related to soil carbon science and involving multiple stakeholders, the authors recognized the CRELE attributes as emerging pillars structuring the study (Ingram et al., 2016). Once made aware about the difference in time perception related to crop production, strategic decision-making and soil carbon management, the authors were able to provide guidelines considering both short and long-term impacts, hence increasing the relevance of the project. However, it was also stated that the iterative methodology, responsible for increasing legitimacy, brought negative effects on the relevance and credibility criteria, since with a wide range of stakeholders also came different interests, which may make some information irrelevant to part of the audience (Ingram et al., 2016).

2.3 Environmental DNA

The international interest in SPIs and current demands to respond to different information needs at different levels and times of decision-making challenges governments worldwide, calling for new approaches and new tools to gather and consider evidence that fits the above-mentioned criteria (credibility, relevance and legitimacy). Genetic analysis has long been useful in biological studies and legal investigations, but the potential to access policy-relevant ecosystem level information from a glass of water is relatively new (Kelly, Port, Yamahara, Martone et al., 2014). Environmental DNA refers to the DNA obtained from environmental samples such as sediments, ice, water and even air. The first reference to eDNA dates back to 1987 and concerns a method for extracting microbial DNA from sediments (Ogram, Sayler & Barkay, 1987), but it was not until the early 2000s that a clear interest emerged (Evans & Lamberti, 2018; Jiang & Yang, 2017). Jiang and Yang (2017), reviewing papers from 1992 to 2016, demonstrated that publications related to eDNA are widely distributed in many source journals, covering various subject categories and published by authors around the world. Papers and citations have increased steadily with time and eDNA has emerged as a high-resolution tool with potential application for conservation biology, understanding of ecosystems and policy-making decisions (Bohmann et al., 2014). To date, eDNA

analysis in aquatic science has focused primarily on proof-of-concept, followed by application of the tool to the detection and monitoring of invasive and at-risk species, but its capability to improve assessments of rare species, estimate biodiversity and assist in routine sampling have also been highlighted (Evans & Lamberti, 2018; Jiang & Yang, 2017; Kelly, Port, Yamahara & Crowder, 2014).

The application of eDNA in monitoring and assessments studies consists of field, laboratory and bioinformatics work. While studies with more detailed information on each dimension can be widely found in published literature (Deiner et al., 2017; Goldberg et al., 2016; Ruppert, Kline & Rahman, 2019; Tsuji, Takahara, Doi, Shibata & Yamanaka, 2019), as a brief overview, the field part typically comprises obtaining a sample, collecting the material on a filter and immediately preserving it to avoid degradation. The most common steps in the laboratory are processing procedures, such as eDNA concentration, extraction, amplification and detection. The bioinformatics work is done along the way, such as in primer design and transforming sequencing reads into measures to be used in biodiversity analyses. The difference in the method used for DNA amplification and reading is responsible for dividing eDNA detection into two main types: species-specific (sometimes referred as qPCR – the name of the most efficient tool used for detection) and metabarcoding (via high-throughput sequencing – HTS) (Deiner et al., 2017; Goldberg et al., 2016). While the former has higher sensitivity and, for that, has been more widely used, being better suited for targeted species; the latter is more appropriate for monitoring the biota, as it gives a broader taxonomic scope (Tsuji et al., 2019). While it is important to understand each step and what can impact the results, in the end, workflows will be determined primarily by the study questions, and then by a combination of available funding and equipment, as well as by personal choice and expertise (Ruppert et al., 2019).

Regardless of the preferences within the workflow, eDNA monitoring has many potential advantages over conventional methods: increased sensitivity, more rapid results, reduced cost and reduced need for taxonomic expertise. DNA based detection outperforms other traditional capture based biological survey methods in terms of number of species detected and does so with noninvasive sampling (Darling, 2015; Evans & Lamberti, 2018; Kelly, Port, Yamahara, Martone et al., 2014). If fish are used as examples, sometimes species may have been unrecorded due to difficulty in sampling their habitat or their active avoidance of conventional sampling methods.

Also, different habitats and species habits demand different types of sampling gear, resulting in a non-standardized set of data, which can impair comparisons. On top of data, depending on the species and habitat, physically capturing animals may be difficult, expensive, or simply not acceptable in case of endangered or rare species, as it may pose a risk for an already small population (Pimm et al., 2015). Environmental DNA methodology has the potential to create a worldwide standard database and without jeopardizing conservation efforts, by applying the same protocol to assess species in a non-invasive manner in very different types of habitats, maybe even making use of other new developed technologies, such as employing unmanned aerial vehicles for collecting water samples (Doi et al., 2017).

However, a degree of caution is required due to possible biases in detection. Just like any other methodologies, projects using eDNA will need to carefully adapt study designs (i.e., choose appropriate sample analysis methods, prevent contamination, test and validate samples), standardize data storage and analysis, critically consider influences of temporal and spatial processes, and assess influences of abundance and uncertainty on positive and negative results (Goldberg et al., 2016; Yoccoz, 2012). Scientists are just beginning to determine how environmental conditions influence DNA persistence times and transport distances, as well as how methodological choices of sample volume, storage, or laboratory processing can influence eDNA detection and quantification (Souza, Godwin, Renshaw & Larson, 2016). Sequencing depth, differential DNA shedding rates and/or preferential amplification of species may be important considerations when interpreting community composition with eDNA (Kelly, Port, Yamahara & Crowder, 2014). A successful application of eDNA in field surveys strongly depends also on sitespecific conditions and temporal selection, as a combination of single environmental factors may cause both false positives and false negatives (Stoeckle et al., 2017). For example, the transport of eDNA by water over long distances may create a false positive result due to the uncertainty about its actual origin (Clusa, Miralles, Basanta, Escot & García-Vázquez, 2017; Evans & Lamberti, 2018). False negatives may derive from the life cycle of the target species, since many species vary their activity depending on season, or from low concentration if the detection limit is not sufficiently low (Clusa et al., 2017; Souza et al., 2016; Stoeckle et al., 2017). Last, but not least, the implementation of large-scale eDNA-based ecological studies is dependent on the availability of a completed reference library, significant computational capacity, highly advanced facilities, credible laboratories, and well-trained personnel.

In an illustrative experiment, a tank mesocosm was sampled at the Monterey Bay Aguarium to check if eDNA would indeed be a viable tool to indicate composition of the tank community that included green sea turtles, sandbar and hammerhead sharks, one species of pelagic stingray and eight bony fish species from the Pacific Ocean (Kelly, Port, Yamahara & Crowder, 2014). From the nine taxonomic families accounted for, eDNA was able to identify four of them, some even at the genera level, all pertaining to bony fishes. They also obtained positive results from species that were not present in the tank, due to different inputs of eDNA sources, such as an intake seawater pipe and feed sources. When knowing what to look for, for example the turtle species, the authors made use of a different molecular marker and obtained a positive presence match. Because the authors knew the exact composition and species abundance of the tank, they were able to say that the rank abundance of modeled eDNA generation matched the rank abundance of biomass, but could not establish a model or identify if it is a consequence of mass/surface area, number of individuals or species metabolic rates. Other core challenges that must be overcome before informative relative abundance data can be generated through eDNA analysis are related to eDNA persistence in a broad range of habitats and climates, and how environmental factors may affect eDNA concentrations (Bohmann et al., 2014). It has been shown that temperature does influence eDNA concentrations (Buxton, Groombridge, Zakaria, & Griffiths, 2017; Tsuji, Ushio, Sakurai, Minamoto & Yamanaka, 2017) but, because this can also be affected by relative abundance and maybe even other factors not accounted for, more experimental studies need to be done to determine the importance of each variable.

2.3.1 eDNA and science-policy interface

A real-life example of eDNA applicability to monitoring and decision-making is the surveillance for the invasive Asian carp in the U.S. Midwest region, around the Chicago area and the Great Lakes. In an overview of the project, Darling and Mahon (2011) highlight the value of eDNA when assessing for low abundance/difficult to capture species (e.g. invasive ones) and mention that eDNA results were even responsible for triggering management actions, such as intensive monitoring and plans to correct blockage bypasses. The authors also comment on the paradox of the tool: while eDNA grew in recognition due to its increased sensitivity over traditional methods, its results are still expected to be validated by these same limited methods. This is not uncommon, as new tools typically go through stages of acceptance and even 'macrobial'

scientists have been resistant to accept eDNA results (Kelly, 2016). Perhaps in an attempt to put things in perspective, other authors have even compared eDNA technology as to being 'sight-unseen' (Jerde, Mahon, Chadderton & Lodge, 2011) or as to 'tracking ghosts' (Wilson et al., 2014). However, the lack of (expected) visual data confirmation leads to dissatisfaction and intense scrutiny by some stakeholders, and eventually the discussion around the use of eDNA for monitoring of invasive species can become more political than scientific (Darling & Mahon, 2011).

Given the already demonstrated utility of eDNA identification of invasive species by providing early detections and allowing faster conservation responses, Thomas, Hanner & Borisenko (2016) make a strong case in favor of the incorporation of this tool into Canadian legislation dealing with invasive aquatic species. If eDNA assessments and monitoring were officially recognized by the government, it would not only represent a major commitment to Canada's obligations for strategic planning for biodiversity, but also require the creation of a database, promote inter-agency communication, represent a large saving of analytical costs, and allow for data standardization (Thomas et al., 2016); this last factor being extremely critical in the role of environmental assessments to make better predictions (Roach & Walker, 2017). Even so, the idea of incorporating eDNA into regulatory policy has received little attention. The reasons may be related to some of the already identified hurdles that likely prevent genetic information from effectively informing decision-making, such as unfamiliarity with the tool, procedures and results, and insufficient engagement of potential end-users (Darling, 2015).

Studies involving relevant stakeholders and that balance quality criteria (CRELE) are more likely to influence SPIs (Cash et al., 2003; van Voorn et al., 2016). According to a review article on stakeholders and invasive species, social science studies have increased robustness due to better information context, such as by integrating local knowledge with scientific knowledge and providing insights beyond numerical data (Shackleton, Adriaens et al., 2019). The same research also demonstrated that the main reasons for studies to engage with stakeholders are to "assess their knowledge and perception on the topic (67%)", as well as "to inform policy and management planning (41%)"; while the main outcomes and benefits are to "build scientific knowledge and evidence (43%)" and obtain "information for policy and management development and implementation (14%)" (Shackleton, Adriaens et al., 2019, pg. 93). Even though stakeholders from different areas such as in science, policy or society may not share the

same perceptions on some topics, assessing and protecting biodiversity is almost universal and automatic. The grounds behind it can be of ecological, economic and/or social origin, such as moral, cultural and religious importance, and the values attributed to species are mostly translated through ecosystem services and the benefits obtained (Bennett et al., 2015; Sagoff, 1996). While the importance of assessing knowledge and perceptions in policy development and environmental governance is well known (Carlson & Cohen, 2018; Moon, Blackmand, Brewer & Sarre, 2017; Reed, 2008; Shackleton, Richardson et al., 2019), with early stakeholder engagement influencing the use of new sampling tools (e.g., molecular ones) (Darling, 2015, 2019; Moon et al., 2017; Shackleton, Adriaens et al., 2019), I was unaware of studies considering those regarding eDNA and with a focus on Canadian inland aquatic environments.

CHAPTER 3

METHODS

"Drawing on social-science theory and methods to increase scientific understanding (...) can contribute to improved policy and management decisions."

(Bruskotter, Toman, Enzler & Schmidt, 2010, pg. 947)

With the intention of answering the proposed research question and achieving objectives, I sought the perspectives of potential end-users towards eDNA through semi-structured interviews. An outline of the study and relevant materials, such as information forms, consent forms and an interview guide were submitted to the University of Saskatchewan Research Ethics Board and received an "exempt status as per Article 2.1 of the Tri-Council Policy Statement (TCPS): Ethical Conduct for Research Involving Humans, December 2014" (#BehID182).

Purposive random sampling was used to consider and select potential interviewees. In cases where selected interviewees suggested other candidates, snowball sampling was also used. Both sampling techniques are widely employed in qualitative studies, but while the former is normally used to "identify and expand the range of variation or differences", the latter is a way of "narrowing the range of variation and focusing on similarities" (Palinkas et al., 2015, pg. 534). Hence, the rationale for choosing these sampling methods was to add trustworthiness, while attempting to obtain information-rich cases (Palinkas et al., 2015).

As I anticipated different inputs from participants with diverse professional backgrounds, interviewees were grouped into five categories according to their occupations: 1) academia, consisting of those employed primarily in academic environments; 2) community, for those involved in community-based and/or non-governmental organizations; 3) government, comprised of provincial and federal government employees, including those working for regulatory agencies; 4) law, for those practicing law and/or with legal expertise; and 5) private, being those working in the private (for-profit) sector. The rationale for selecting those categories was to establish a diverse pool of potential direct and indirect end-users of the eDNA tool in the science-policy domain, ensuring groups with important or distinctive perspectives were represented (Robinson, 2014). The only mandatory common characteristic among all groups was that participants should have at least

heard about eDNA, as otherwise interviews would be ineffective. When gathering data, no distinction was made between different eDNA types [e.g. species-specific (qPCR) or multi-species (metabarcoding)], as the focus of the study was to obtain the general perceptions of end-users in relation to the tool.

Because I was focusing on obtaining knowledge about eDNA use related primarily to freshwater aquatic habitats and at the same time was questioning if geo-political boundaries could have any influence, participants were selected from different Canadian geographic regions, namely the provinces of Alberta (AB), Saskatchewan (SK) and Ontario (ON). Alberta and Saskatchewan are neighboring provinces and home to the transboundary Saskatchewan River basin. The basin is an area subjected to multiple land use and development pressures (e.g. urban development, agriculture, intensive livestock, heavy industry, hydro power) (Ball, Noble & Dubé, 2013; Strickert et al., 2016), and under much scrutiny for water security because of competing water users (Wheater & Gober, 2013). The province of Ontario shares with the United States the control of the Great Lakes, one of the world's largest freshwater systems and a region of immense social, economic and ecological value to the province, but also an aquatic system that is affected by many cumulative stressors, such as habitat loss/destruction, nutrient run-off from land, overexploitation and invasive species (Allan et al., 2013; Boston, Randall, Hoyle, Mossman & Bowlby, 2016). As I started compiling a list of possible participants from these provinces and considering which perspectives were necessary, some participants did not align with provincial jurisdictions because of the pan-Canadian focus of their work, and so a fourth category was created - nationwide (Nat.).

Participants were first contacted by electronic mail and/or by phone. The interview guide was designed to encompass themes related to both ecological assessment and governance (Appendix A). The majority of the interviews was done by audio-only phone calls, recorded, and transcribed with the help of an online automated transcription service. Interview responses received in writing were added to those previously transcribed. Transcriptions were later coded and analyzed using the software NVivo v12.5.0. Analyses were mainly question-oriented, but common themes emerging throughout the interviews were also considered. One question in the interview guide (#4) involved assigning quantitative scores that ranked eDNA on a scale from 0 to 10 based on several characteristics (accuracy, applicability, bias, detail, legitimacy and sensitivity). These results were examined through jitter graphs plotted using the free software

PAST v3, with values close to zero being worse than those close to ten. The results for 'bias (x)' were converted to y (x+y=10) to be comparable to the other characteristics being ranked and were henceforth referred to as 'unbiased'.

From August 2018 to April 2019, 64 participants were contacted, resulting in 40 coded interview reports (Table I and Appendix B). The number of codes in each question do not total the number of participants, since participants were free to provide more than one answer at each time or abstain from commenting. The final number of interviewees was determined by achieving saturation. One of the first signs of reaching saturation was that recommendations for new potential interviewees were overlapping with the list of participants already considered or interviewed (Bleich & Pekkanen, 2013). A second sign was based on the researcher's impressions of data saturation (Saunders et al., 2018), when the interviewer noted that most of the information learned in the latest interviews evoked similarities to previous conversations and acquired data. At this point, I was confident that the study had reached or closely approximated a satisfactory 'conceptual depth' (Saunders et al., 2018).

Table I: Number of analyzed transcribed interviews by occupation and geographic categories.

| Geo categories/ Occupation | Alberta | Saskatchewan | Ontario | Nationwide | total |
|----------------------------|---------|--------------|---------|------------|-------|
| Academia | 2 | 1 | 1 | | 4 |
| Community | 4 | 2 | 2 | 3 | 11 |
| Government | 3 | 4 | 2 | 4 | 13 |
| Law | 1 | 1 | | 2 | 4 |
| Private | 2 | 2 | 2 | 2 | 8 |
| Total | 12 | 10 | 7 | 11 | 40 |

Secondary information to support the interview data was also obtained through an unstructured review of existing federal and provincial Acts and Regulations in Canada, with a focus on Alberta, Ontario and Saskatchewan (i.e. inland waters). I accessed consolidated Acts and Regulations on the Justice Laws Website (https://laws.justice.gc.ca/eng/) during the years of 2018 and 2019. Alberta environmental legislation was obtained through Alberta Queen's Printer Laws

Online/Catalogue (https://www.qp.alberta.ca/Laws_Online.cfm) during the same period. The selected documents (Table II) were examined to verify potential applications for eDNA-based methods in their context, an approach akin to Kelly, Port, Yamahara, Martone et al. (2014), who identified some contributions of environmental genetic tools to legal or policy goals in the United States and European Union, after reviewing selected statutes and directives.

3.1 − *Study limitations*

In the hope of increasing validity and reliability, I would like to recognize some known and considered weaknesses of this study. The first issue relates to the selection of participants. I believe I was successful in avoiding getting impressions only from people already working in the area and/or trying to develop eDNA as a research method, which would presumably be biased in favour of the tool. However, it is known that snowball sampling tends to select people with similar characteristics (Palinkas et al., 2015), which could lead to similar points of view. Hence, note that I did not snowball potential participants from those few interviewees who had negative perceptions towards eDNA, as all our snowballed interviewees were recommended only by those participants who showed neutral or positive attitudes. Secondly, even though having knowledge about eDNA was an essential pre-requisite for a candidate to be considered, I made the choice to still include in the analysis the transcription of the interview with one participant (P20) who claimed to have never heard about eDNA until our first contact. I see it as justifiable since the participant in question still managed to provide powerful insights within the interview guide on how the theme could be approached/seen in their field of occupation.

On the same note, I have decided to not include in the analysis one interview conducted in person with representatives from an Indigenous community in Saskatchewan (P15). I show profound respect for all I learned regarding their relationship with the water and aquatic environments but, unfortunately, I was not able to acquire insights related to the eDNA theme. With this, I acknowledge that when designing the study and the interview guide, I have failed to consider the effective participation of all groups of interest and, therefore, this study does not carry Indigenous perspectives on the theme. By recognizing that Indigenous peoples understand water as an alive and sacred being, acting as caretakers for aquatic environments (McGregor, 2012), and that the importance of traditional knowledge in watershed management is undeniable (Baldwin et al.,

2018), I recommend that future work should address how the applicability and findings of eDNA as a monitoring and assessment tool would fit, connect and translate into the Indigenous belief system.

CHAPTER 4

RESULTS

Before reporting on the eDNA interview theme itself, and to better understand the participants, I first asked the interviewees what they considered to be the main problems facing aquatic environmental studies in Canada. Popular topics such as climate change and water quality/quantity were only mentioned one and three times, respectively, and only by representatives from the Community and Private groups. Instead, for nearly half of interviewees (n=16), the biggest problems with monitoring and assessments were data related, ranging from data collection and establishing baselines to data access and management.

I also asked participants about their familiarity with eDNA to better understand their subsequent interview responses (Appendix B). Only one participant (P20; Law – Nat.) said they had no knowledge of the tool at all, while the level of expertise of others was variable. Nine participants said they were aware of the tool but demonstrated some level of uncertainty in understanding. Fifteen participants said they were familiar with the topic, with a few even explaining that they gained familiarity by reading about eDNA, attending conferences or workshops where eDNA studies were featured, and/or using the tool directly. The highest number of participants indicating that they were familiar with eDNA were in Alberta and Saskatchewan, while if the distribution by occupation is looked at, those participants be longed mostly to the Community and Private groups. The number of participants who said they were knowledgeable about eDNA was the same of those who said they were familiar with the tool (n=15) but, in this case, they were mainly professionals from the Government group (n=8). From those participants who affirmatively responded to have a comprehensive understanding of the topic, nine declared to be using eDNA at the moment and one mentioned having used it before.

The main interview results are presented in five sections. In the first section, I assess how interviewees perceive some of the technical characteristics of eDNA and investigate possible relations among the level of expertise from participants, their occupations and geographic categories. The second section connects uses of eDNA with possible users, followed by two sections identifying the main general opportunities and general challenges facing eDNA in freshwater aquatic studies, according to the participants. Lastly, I present the results of a line of

questioning that was more focused on a legal perspective and how eDNA may find its space in environmental governance.

4.1 Quantitative scoring of eDNA characteristics

Overall, interviewees scored eDNA as a very applicable tool for studies of aquatic environments (individual scores ≥ 7 , except for one participant in AB and two in ON), with a high degree of sensitivity to detection (individual scores ≥ 7 , except for one participant in ON and one in the Nat. group) (Fig. 1). The legitimacy of eDNA was scored slightly lower than applicability and sensitivity but individual scores were mostly located on the favourable side of the spectrum (n ≥ 5 , except for three participants). Impressions about eDNA for all other characteristics (accuracy, detail, and unbiased) were more divergent, indicating that the tool has room for improvement in these areas. For example, when looking into accuracy, scores provided by participants from the group Academia ranged from 5.5 to 8, while in the Community group they ranged from 1 to 10.

Even though I provided a broad explanation for all characteristics, several interviewees emphasized that how these are assessed is very much tied to the objectives (i.e. what is being asked when eDNA is being used) and to the level of training of the user (human factor). Potential endusers from the Nationwide category, who on average felt like they were more knowledgeable about eDNA overall, had a range of scores that was only slightly lower when compared with respondents from different provinces (e.g. the average values for applicability per geographic category were: Nat. = 8.56; SK = 8.28, AB = 8.13 and ON = 7.25) (Fig. 1). This led me to believe that background experience did not have a considerable influence on the answers. Participants from Ontario ranked eDNA with the lowest averages for all characteristics, while the highest ones were shared by the Alberta (accuracy, legitimacy and sensitivity) and Nationwide categories (applicability, detail and unbiased). I did not identify any substantial pattern and/or difference among groups by occupation.

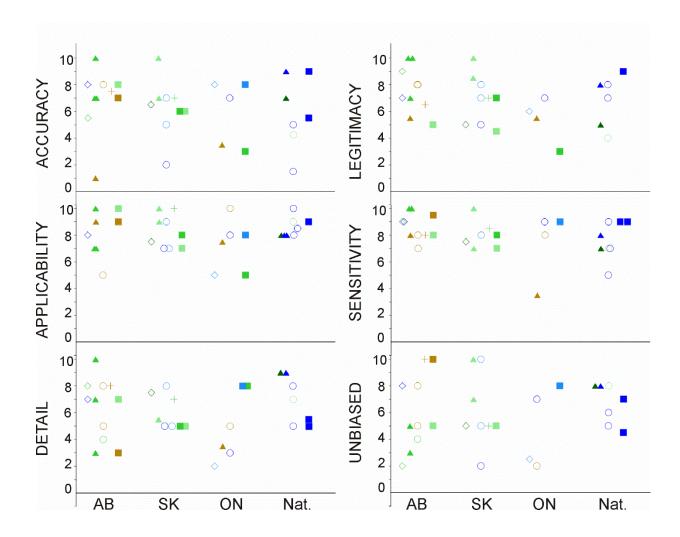


Figure 1: Jitter scatter plot of eDNA characteristics as perceived by interviewees from different geographic categories. Participants were categorized by occupation (Diamond = Academia, Triangle = Community, Circle = Government, Cross = Law and Square = Private) and their knowledge about the tool (Golden = Aware; Green = Familiar with; Blue = Knowledgeable). Darker shades inside a knowledge category indicate a stronger experience with eDNA.

4.2 Users and uses

When asked if and how eDNA results would be valuable to different sectors of society, a similar spectrum of usefulness was identified for all groups, but with different degrees. For example, most interviewees (n=28) agreed that "scientists would be one of the user groups that would find that [eDNA] most valuable" (P04; Community – Saskatchewan), but still expressed that the tool could be limited or even not valuable at all. For example, one interviewee commented

that "...it depends on the questions these scientists are interested in" (P17; Community – AB), with another similarly explaining "going back to researchers, I guess it depends on what you're interested in" (P12; Government – SK). One participant in academia elaborated further, providing examples for ways in which they believed eDNA can be useful, but also ways where they do not see any value in its use.

So, it's valuable for scientists in a bunch of different ways. For looking at species distributions. For being able to sample endangered species information. For providing distribution data. Maybe did I say that twice? For helping to look at invasive species (probably one of the big ones). For looking at escapes from aquaculture facilities to provide a rationale and stimulation for characterizing DNA of new species, rare species. You want ways that it's not valuable? So, in terms of the key areas of concern or main challenges that I defined in the environmental monitoring and assessment, eDNA doesn't help with absence of a framework; it doesn't help with transparency or linking of information; it doesn't help with database; it doesn't help with accessibility and it doesn't help, especially, with decision-making. (P07; Academia – ON)

Interviewees also seemed to agree that the value of eDNA to industry consultants would be similar to that for scientists. For example, a participant from academia commented that "industry consultants usually are scientists. I mean, it can be extremely valuable." (P10; Academia – AB). On the other hand, although some interviewees recognized that eDNA could be potentially valuable for community members, especially to answer specific questions, here is where we saw the highest number of end-users affirming that the tool would not be very important or would require educational efforts in order to make it acceptable. For example, as expressed by interviewees across different occupation groups and geographic categories:

- ...I don't necessarily see a direct use by the community. (P13; Government Nat.);
- You could present the results to them [community]. But generally, in the way you'd have to present the results to them you oversimplify the matter and they lose any appreciation for what the assumptions are behind the technology. (P25; Private Nat.);

- I think it would be valuable if accompanied with the right amount of education as to how it works and what it actually is. (P40; Private AB);
- I think for the information to be valuable it would have to be boiled down into more of like a fact sheet type information. (P11; Community SK).

There was no consensus on who should be leading the development and the application of eDNA. A few interviewees believed the government or the private sector should be the ones responsible, emphasizing whether the roles should be specifically related to the development, application or just support of the tool. However, most participants could be split into two groups: one that attributes solely to academia the responsibility of leading and developing eDNA (n=20), and another that believes this should be the result of collaboration among sectors (n=19). For example, amongst those who believe that academia should assume responsibility for leading and developing eDNA, participants expressed that "the academy ... has to basically get the technique evolved to a point that it provides useful information and it has to articulate some potential uses for that, and then I think after that, the private sector and government can probably have a role in promoting its use", emphasizing that "the first step is for the technique to get to a position that it can be used effectively to answer something practical questions" (P22; Government – ON). A participant from Alberta expressed a similar view, noting that "development of this methodology is coming in academia, but then the application of it is in government and regulatory environment" (P08; Government – AB). For those participants who suggested joint responsibility, some expressed it should be shared among all, "because if one group does it all on their own, it will not fulfill the needs of the other groups and vice versa" (P34; Government – Nat.), while others believed it should be aligned to each group's capabilities, since "the private sector's got the money and the academic sector's got the expertise [and] really the role of government is more in the audit *kind of capacity*" (P35; Government – SK).

The majority of potential end-users agreed that eDNA could be embraced and recommended by regulatory agencies, even if only hypothetically, but some highlighted the need for improvements and rigorous control. For example, a participant from the private sector emphasized that eDNA "definitely could be embraced, absolutely, and recommended" (P40; Private – AB) by regulatory agencies. Others were optimistic but expressed some caution in its uptake by regulators,

noting "there's obviously some issues to resolve in terms of accuracy or at least perception of accuracy, but yes, I would answer positively to this question" (P17; Community – AB). One Community participant from Ontario explained that it could be useful for regulators but "they would need to have confidence in how we collect the information, how accurate it is and under what circumstances", emphasizing that eDNA is a quick screening tool that "agencies would be interested in ... once we worked out the kinks" (P28; Community – ON). Some disagreed with the way the question was formulated, as according to them "...it's unlikely that a government would push or endorse a specific methodology or test" (P20; Law – Nat.). One participant with better understanding of legal procedures explained that "in most cases, it's more likely that it arises either through standard of practice in some sector or be developed in university settings and then promoted for whatever attributes the developers are noticing it has, and then seeking acceptance within sectoral audiences, like water managers and others "(P20; Law-Nat.). Another participant confirmed that governments "never embrace anything", but according to them what happens is that government employees just "typically have it rammed down our throats" (P35; Government -SK). Three interviewees felt like they could not opine on that question and four did not agree with the use of eDNA in this regulatory context.

4.3 General opportunities

Throughout the study I was able to identify some clear areas where eDNA would be better accepted among users due to better chances of favourable outcomes. According to interviewees, eDNA "would be a preliminary assessment tool to give you some sense of whether or not you should be doing more detailed assessment on a particular situation" (P28; Community – ON). One participant who works as a government regulator provided the following statement when asked how eDNA could be used in a specific situation.

In a context of a mine development I see eDNA being used to support baseline data collection and understand the existing environment that has the potential to become impacted through mine development, providing a screening tool to understand distribution species within the potential footprint or surrounding area. (P38; Government – Nat.)

Hence, it is clear that eDNA would be very useful if primarily employed as a first screening tool to provide snapshots of aquatic environments and to establish baselines. However, some

participants still had reservations about its use for assessing whole communities, advising caution "about saying that we can use eDNA all the time for all species" (P02; Community – Nat.) and reminding that "even with the metaBarcoding, you're still best combining eDNA and other assessment methods, [as] there is no assessment method we know that finds all species" (P22; Government – ON). On the other hand, they were extremely confident in the information the tool could deliver when targeting single species, as according to one participant "identifying the presence of an individual species is probably even more defensible using eDNA approaches than using morphological characteristics" (P01; Government – SK).

Participant's comments on singular species helped me to identify another important opportunity for eDNA, which is to be used in the monitoring and assessment of "indicator species" that are important to decision-makers in some way" (P08; Government – AB), such as invasive and endangered species (Appendix C, #6 and 11). Even one participant who seemed, in general, to distrust the tool recognized that eDNA "can play a useful role on a surveillance tier for things like invasive species" (P07; Academia – ON), while another mentioned that "if other industrial activities ongoing on that land base and they are bringing in invasive species, then eDNA can sort of validate that as well" (P08; Government - AB). Specifically for endangered or rare species, participants highlighted that eDNA "would be a great value for presence/absence, where the probability of detection through conventional methods is low "(P12; Government – SK), and could "potentially, alter the plans for that [development] or, you know, induce new mitigation measures" (P08; Government - AB). While the tool's sensitivity and ability to deliver faster results was one of the main reasons for its association with monitoring for invasive species, as eDNA "could be a rapid tool ... to inform on what type of regulatory requirements would be needed in order to protect those [local] fish populations and those [local] fish species" (P28; Community – ON), the non-invasive property of eDNA was especially important in its potential use for assessments involving species at risk. One interviewee importantly reminded me that "scientific assessment in some cases is a threat to the species, [as] things like electrofishing can have very high mortality or gillnetting, so to have something that is noninvasive and has no impact, it would be a huge benefit for monitoring species at risk" (P22; Government – ON). The fact that eDNA presents itself as a tool "that is less invasive, less time consuming, less expensive, ... [with the potential to give a whole other avenue of information" (P19; Private – AB) when compared with other traditional methods can also be seen as an opportunity for this tool to find a place in monitoring studies. One interviewee greatly summarized these attributes, as seen in the quote below:

So, as we continue as to evolve as a society and recognize the importance of protecting a species and treating animals differently, what eDNA provides is a less intrusive method to go out and survey for species, be an invasive species or more so species at risk. So, it is less invasive than going up and trapping, electrofishing or such, which even tromping through a habitat, which also does provide more opportunity to negatively harm the habitat through possible spread of pathogens. ... eDNA provides a tool that is ultimately less costly overall then traditional conventional surveymethods for aquatic species. So they can get to the finish line quicker with more empirical analytical data at hand as opposed to sending out teams. (P29; Private – Nat.)

However, a few interviewees did mention that eDNA is 'not there yet', as it "was presented to us [here referring to general users] as well as a panacea in terms of being able to solve problems and make it cheaper and make it more accessible, and it hasn't necessarily proven to be the case thus far" (P18; Community – Nat.), confirming the need and wish for improvements, since "at present it takes quite a bit of time to both process the sample and preserve it to be sent in to the lab [but] I can see the future where it could be looked at in situ" (P41; Government – SK).

4.4 General barriers

It was already mentioned that eDNA costs are seen both as an opportunity as well as a momentary barrier, as the tool still need improvements. But, according to interviewees and as summarized in the quotation below, the top three challenges that eDNA for aquatic environments are most likely to face relate to the resolution of the tool, its validity and its users.

I think from a cost perspective it can be very cost effective as well. As long as you have, you know, establish markers already done and, you know, again, your methodology is consistent and your actual people on the ground doing the collection are, you know, capable of doing this consistently. (P40; Private - AB)

Regarding eDNA's resolution, the main concerns were related to biodiversity reference information and genetic sequences (primers) development for species identification. Two different

participants from the Government group in Saskatchewan expressed uneasiness since, up to this moment, they "...don't know how ... to evaluate who should be there and who shouldn't, there's very little established understanding of what reference condition is "(P01; Government – SK) and, on top of that, it is known "that you're not going to type all the species that are present in my lifetime, and species are changing "(P35; Government – SK). Those worries align with comments from other groups, who say "some of the barriers would definitely be the depth of the genetic markers that are present currently, the number of species that we have developed markers for" (P40; Private - AB). Another popular claim regarding eDNA resolution was for the tool to deliver quantitative results and provide abundance data, as "on the aquatic side, we often always need to pair that abundance question for any regulatory stuff, like Fisheries Act, all that, you need to understand numbers, not just presence/absence in most cases "(P26; Private – ON). A participant from academia even mentioned that in terms of eDNA "being specific and sensitive, it already does do that quite well and a lot of it keeps getting better, but it needs to become quantitative" (P10; Academia - AB). Even though for some of the participants eDNA already delivers good results in terms of being a specific and sensitive tool, other interviewees mentioned the need for improvement in some of its characteristics, mainly accuracy, supporting data gathered on question #4 (Fig.1). As an example, "... I think in and of itself or just as an individual tool, eDNA has very strong limitations on it. Because there are questions about its applicability, its accuracy, it's bias." (P21; Government - AB)

Accuracy also played a major role in eDNA's validity, with a participant with legal expertise mentioning that "any new tool being used in court cases by lawyers or by the experts of lawyers will face a challenge about its accuracy, and scientific validity, the state of development of the tool, how long it's been in use, how reliable it is" (P20; Law – Nat.). Potential end-users expressed the need for eDNA to be further examined against other tools, in order to be deemed robust. This could be done either through experiments, such as "side by side comparison studies using traditional methodologies versus eDNA approaches in closed systems" (P25; Private – Nat.) and/or "published peer reviewed evaluations of what the differences are between traditional morphological character collections of ... communities and then eDNA collected communities and a reconciliation of how you evaluate those differences" (P01; Government – SK). Over half of interviewees considered that the reliability of eDNA is linked to different levels of certainty (Appendix C, #9). Some suggestions to improve it were related to the development of

"appropriate controls and setting values and more rigorous application under QA/QC protocols" (P27; Academia – AB); and to an increase in the rigor and transparency of methods, which could perhaps make easier for studies to be "published in really prestigious peer reviewed journals" (P04; Community – SK).

Lastly, several interviewees from different geographic categories and occupation groups expressed concerns about how users' lack of specific training and education on the subject could lead to eDNA being improperly used or oversold, ultimately affecting its reliability and acceptability. For them, "the main challenge is going to be understanding what conclusions you can draw from the data" (P38; Government – Nat.) and for that "the scientific community would have to provide tools or explanations on to help support some of the results from an eDNA to something that's more consumable by practitioners" (P06; Community – ON). According to one participant from Academia, "the potential for abuse exists a little bit more with the consulting companies and potentially a lot more with the general public who don't have that scientific training to be able to accurately interpret it" (P27; Academia – AB). The testimonial below, from a real situation, given by a participant from the private sector, illustrates the dangers of having someone "overstate the application of the tool and not properly acknowledge the potential limitations" (P16; Private – SK), with some of many possible outcomes being the undesirable consequences of confusion and lack of interest by potential end-users.

I know when people get a taste of it, when you see a little presentation at a conference, people get excited about the applicability, the possibility of it solving a problem for them that they've been having for five years. But the other thing that happened at the last conference I was at when people talked about it is you had two groups of people. And I believe it was, as it was explained to me, the gene jockeys, are perceived as overselling its potential. There are those people that are saying, 'it's really cool, it's perfect, it's going to solve all your problems'. And then there's the other end of the spectrum of the same people who do this stuff on a day-to-day basis saying it's not ready yet. ... So, I kind of walked away going 'this is really neat, but I'm not sure where it's there yet'. And I don't know who to believe and I didn't spend a lot of time digging in. (P19; Private – AB)

As stated by another participant, the best way to present eDNA is to "proceed with cautious optimism to ensure that the technology is used in a responsible manner; otherwise, the credibility of the tech will be tarnished" (P42; Private – ON).

4.5 eDNA into policy

A review of legislation revealed far more opportunities than challenges for eDNA to be considered a viable tool in environmental monitoring and assessments (Table II). Even though for most of the identified biological opportunities eDNA would be just another option in the toolbox, for others its non-invasive characteristic would represent an advantage over traditional sampling methods. Political and/or legal opportunities are closely related to the power of regulators to modify permits, create regulations and implement programs, corroborating information obtained from the interviews which indicated that eDNA acceptance by users would represent a greater challenge than legislation itself. One participant from the government group commented that "there's nothing about the documentation that I work from that says that I can't accept a given methodology, it's my discretion to ensure that the information I'm getting from a proponent is adequate" (P38; Government – Nat.), while another mentioned a "reluctance right now ... to accept eDNA findings from industry consultants and the various environmental consulting companies, because the technology is so new and because people are not sure what is the quality of the findings" (P22; Government – ON).

Biological challenges were only identified in two of the twelve analyzed pieces of legislation, namely the Metal and Diamond Mining Effluent Regulations SOR/2002-222 and the Pulp and Paper Effluent Regulations SOR/92-269. The possible biological challenges were all related to the fact that specimens would have to be captured and sacrificed in order to provide all requested information (Table II). Some interviewees also pointed out this mismatch between indicators demanded by legislation and the fact that eDNA "doesn't provide information at the level to which [is] need[ed] to manage a system" (P07; Academia – ON). One participant made a parallel with some of the challenges described in the previous section and mentioned that for them "...questions about accuracy and sensitivity and all that ... are secondary; the first thing the regulator needs to know is what line of evidence can this technique provide that's relevant in some way to the decisions we have to make in our organization, ... what's the basic line of evidence that

can be provided by the technique and how is it relevant to the government "(P37; Government – ON).

However, it is important to highlight that this requested information does not necessarily represent an impediment, as eDNA can find its space even under rigid regulations, as exemplified in this quote below:

I think the MMER [Metal Metal and Diamond Mining Effluent Regulations SOR/2002-222] that you are referencing is super prescriptive. There is an option to suggest a new approach. So eDNA probably will not become part of the pure EEM [environmental effects monitoring] program under the MMER. But where I see it being implemented the soonest and in the most beneficial capacity would be prior to an EEM, so like in that EA [environmental assessment] phase and a baseline in the permitting phase where they're studying the environment and confirming their impacts and predicting the effects of the operation and the effluent discharge and so on. So that is not prescribed in any documentation anywhere. (...) But also under the MMER, if you confirm effects in two years in a row, that's when you step into an investigation of cause phase. And then you get to design your own study and determine how best to answer the question of what's going on. So even under the MMER, if you move into an investigation of cause phase and they talk about an IOC [investigation of cause] in the regulations and they definitely talk about it in the guidance document that supports the regulations, you could use eDNA and likely would want to under a whole host of scenarios under the MMER as well. So the MMER doesn't explicitly bar you from ever using it. It only kind of boxes it out during a regular monitoring program because they tell you exactly what you have to do and you can't really change it. But if you get into an IOC phase, you sure *could use eDNA. (P19; Private – AB).*

Only two minor political and/or legal challenges were identified in the document review: the possible need for a certificate of analysis and for documentation and validation of methods. Without further clarification, both have the potential to be contested in court, for example. Who would be the analyst in eDNA analyses? Does each sample need to be saved and stored for possible counterproofs? What is considered a validated method? Those questions associated with

challenges mentioned in the previous section regarding eDNA validity contribute negatively to the tool and a possible way to minimize it would be by developing standards.

4.5.1 Standards

One of the most common suggestions to increase the levels of certainty and eDNA's validity, especially in a regulatory context, was the need for adoption of some sort of standards, guidelines and/or protocols. Participants mentioned that "for regulatory, for the other groups, the value [of eDNA] is the ability to repeat tests for the presence of something ..., especially because it's not invasive" (P36; Community – AB), "however, standards need to be in place so that sampling could essentially be replicated to ensure consistency and reliability." (P42; Private – ON). A participant from the private sector is of the opinion that "not until there is a certified standard that has an accreditation that [eDNA] will be recognized in a court of law" (P25; Private – Nat.).

Nevertheless, it was not until one potential end-user mentioned their uncertainty of which type of standards (criteria or tests) that I started getting deeper into this matter with other interviewees. According to participant 20 (Law – Nat.), both types are part of "a constant debate" in environmental regulation" on "whether to use prescriptive approaches or whether to use broader definitions of results and objectives. And so, the standard tests go with fairly prescriptive approaches and the criteria are more applicable to broader decision-making processes. And both of these things happen in environmental law in different contexts." Not many interviewees took a stance on choosing which type of standards should be in place, criteria (n=4), tests (n=4) or both (n=1), but some even risked stating these should be more linked to laboratorial procedures (n=2). A participant in favor of criteria stated: "I would say from a regulatory perspective, it would have to be the criteria route because a regulator is not going to endorse a particular test because that *limits the, it basically, it could create a monopoly" (P38; Government – Nat.).* Another individual in favor of tests opined: "That would be my point of view, because then you want repeatability and you want it to be the same for everybody, particularly if it was in a regulation, it has to be the same for everybody. And so, just say 'well you just have to meet, the test has to meet certain criteria' that leaves doubts about what the test might be or how it might be conducted." (P23; Law -Nat.). There was no mention of the different types of standards throughout the reviewed Acts and Regulations, corroborating that the "decision-making framework [is] fairly broad and allows for different types of evidence that will then be weighed by the decision-maker" (P20; Law-Nat.).

Table II: Biological and political/legal opportunities and challenges for environmental DNA in selected provincial and federal current acts and regulations.

| Acts and Regulations | Legislation goals | eDNA opportunities | | eDNA challenges | |
|--|--|--|---|-----------------|---|
| | | Biological | Political and/or Legal | Biological | Political and/or Legal |
| Alberta Environmental Assessment Regulation 112/1993 | To regulate specifics to be followed by proponents and authorities in environmental assessments processes | | | | -1 |
| Alberta Environmental Protection and Enhancement Act e12 | To balance support and wise use of environment with requirements from other areas | Need for continuous monitoring, evaluation and report of Alberta environment's condition [§15.1, §49(i)] Environmental protection orders may fix and/or regulate the methods or procedures to be used for measuring and sampling [§85(1)(k), §122(1)(i)*, §241(1)(b)] | Establishment of policies and programs, and regulations for those [§12(a), §36.1(g)] Carry out and participate in research projects [§12(d)] Existence of a Chief Scientist role [§15.1] Possibility of adoption by reference [§38(1)] Regulations about standards and criteria to be used to verify conservation [§146(b)] | | • Possible need for a certificate of analyst/documentary evidence [§254(1)(a), §255(2)] |
| Alberta Fishery Regulation SOR/98-246 | To regulate many aspects related to fisheries, such as close times, catch limits, baits, gears, etc | Not a clear one. {There is a concern a bout fishing adversely affecting "the conservation or protection of the fish in the fishery to which the permit applies". So, if we consider fishing for scientific purposes, eDNA has an advantage due to its non-invasive characteristic. [§10(b)]} | | | |

| Saskatchewan Fishery Regulation SOR/95-233 | To regulate licenses to fish and close times per area in the province | | | |
|---|---|---|---|--|
| Ontario Fishery Regulations SOR/2007-237 | To regulate many aspects related to general, sport and commercial fisheries, such as close times, catch limits, baits, gears, etc | Not a clear one. {If fish escapes and prevention of spread are related to the introduction of fish in areas other than their natural habitat, then eDNA could be useful. [§4(1)(n)(o)]} | | |
| Canada National Parks Act S.C. 2000, c. 32 | To establish the limits and regulates the protection of federal parks and reserves areas | Contribute to the evaluation, maintenance or restoration of ecological integrity [§8(2), §11(1)] "Governor in Council may make regulations respecting" "the taking of specimens of fauna for scientific purposes" [§16(1)(c)] Not clear: if fish are considered renewable resource (no definition provided in the act), traditional scientific sampling could be impacted by section 17(3)(c) and the non-invasive characteristic of eDNA could be an advantage. | Park's superintendents may "issue, a mend, suspend and revoke permits, licenses and other authorizations" [§16(3b)] Convicted persons may have to carry out, implement or pay for environmental monitoring or research [§30(1)(f)(g)(n)] | |
| Canada Water Act C-11 | To "provide for the management of the water resources, including research" | • Aquatic research and inventory [§5(a)(c), §7] | "Establishment of intergovernmental committees" to advise on research, policies and programs [§4] Potential to diminish costs in a greements [§8(1)(b)(c)] | • Need for a certificate of analyst [§37(1)] |

| Federal Sustainable Development Act S.C 2008, c. 33 | To "provide the legal framework for developing and implementing a Federal Sustainable Development Strategy that will make environmental decision-making more transparent and accountable to Parliament." | + | Existence of a council with representatives from environmental nongovernmental organizations [§8(1)(b)] Creation of regulations to achieve the legislation goals [§13] | | + |
|---|--|---|---|--|---|
| Fisheries Act F-14 | Regulate fisheries and its implications in Canadian waters | • Establishment of standards and codes of practice to better conserve, protect and avoid death of fish [§34.2(1)(a)(b)] | Creation of regulations [§43(1)(b)(n)(o)] Possibility of incorporation by reference [§89] | | • Need for a certificate of analyst [§56.1(2)(3)] |
| Metal and Diamond Mining Effluent Regulations SOR/2002-222 | To establish guidelines for dealing with mining effluents | • Description of the exposure and reference areas where the biological monitoring studies would be conducted [Schedule 5(10)(a)(ii), (10)(c)(iA)] | | Environmental effects and monitoring studies must be conducted in accordance to Schedule 5 [§7(1), §32(1)(c)] In order to attend all requested effects indicators and to provide all information requested in the First Interpretative Report, it would be needed to capture the specimen [Schedule5(1), 5(12)(e)] | • "The studies shall be conducted using documented and validated methods" [§7(2)] |

| Pulp and Paper Effluent Regulations SOR/92-269 | To regulate the management and input in the environment of substances originated from pulp and paper mill activities | • Need for environmental effects (biological) monitoring studies of benthic invertebrates communities, with richness as one of the required information [§28(1), Schedule IV.1 (3)(c), Schedule IV.1 (11)(a)(ii)] | | • Need for environmental effects (biological) monitoring studies of fish population with information that could only be provided by capturing the specimen [§ 28(1), Schedule IV.1 (3)(a), Schedule IV.1 (11)(a)(i)] | |
|---|--|--|---|--|--|
| Species at Risk Act | To protect wildlife species at risk in Canada | May request monitoring of the status of the species and determine when wildlife species are to be assessed [§11(1), §11(2)(a), §12(1), §12(2)(a), §15(1b)] Requires the use of best available knowledge/information and scientific support on the status of a wildlife species [§15(2), §40] Need for a status report before any assessment of the status of a wildlife species [§21(1)] Preparation of a recovery strategy and action plans, which must include the identification of critical habitats, and a statement of the population and distribution objectives [§41(1)(c.1)(d), §49(1)(a)] Proper identification of habitat necessary for recovery or survival of an aquatic species [§80(4)(a)(i)] | Necessity of an annual report [§8(3)] Stewardship action plan containing incentives and recognition to programs [§10.2] Possibility of entering into conservation a greements which can include monitoring programs [§11(1), §11(2)(a), §12(1), §12(2)(a)] The contents of a status report may be established through regulations [§21(2)] Issuing of a greements or permits if made sure that the survival or recovery of the species will not be jeopardized [§73(1)(2)(3)] | | |

CHAPTER 5

DISCUSSION

"Human perceptions of nature and the environment are increasingly being recognised as important for environmental management and conservation. Understanding people's perceptions is crucial for understanding behaviour and developing effective management strategies to maintain, preserve and improve biodiversity, ecosystem services and human well-being."

(Shackleton, Richardson et al., 2019, pg. 10)

A unique aspect of this study, in relation to several others comprising the eDNA topic, is the shift of focus from the scientific elements of the tool to one that also takes into consideration the social dimension involved with its application. For example, a recent review study on freshwater eDNA categorized 238 peer-reviewed primary publications on this topic according to ecosystem types; geographic regions; target species and main objectives (invasive species - 32%, endangered/rare species - 25%, methods development - 23%, biodiversity assessment - 11% and others, such as effect of environmental variables or economics – 9%) (Belle, Stoeckle & Geist, 2019). While it is within reason to assume that some of these publications may have commented on the social dimensions of eDNA, it is important to highlight that, if so, these were not considered relevant or significant enough to mention in the review study, despite the recognized importance of stakeholder participation, community learning and policy-making in conservation science and environmental management (Reed, 2008; Shackleton, Adriaens et al., 2019; Shackleton, Larson et al., 2019).

By exploring how potential end-users perceive eDNA and how that may affect its application in existing environmental management and assessment frameworks, my results confirmed that eDNA is best understood as a complementary tool to other traditional sampling techniques than as a stand-alone tool, except for targeted species, indicating higher confidence in results from species-specific methods of quantification despite not specifically making that distinction when gathering data. Results also show that the tool itself or current legislation are not the prime impediments for eDNA's broad acceptance, indicating that social elements may be more decisive in that matter.

In the next sections I analyze and discuss my findings in relation to the CRELE model (Cash et al., 2003), an approach that has been deemed to improve the chances of science being implemented in actionable form, as it gives diverse and useful insights on the investigated topic, hence providing decision-makers with more relevant knowledge (Shackleton, Adriaens et al., 2019).

5.1 − eDNA credibility

Credibility is related to the perception and trust of knowledge by involved actors, and according to my interviewees, one of the best ways to manage the uncertainty and lack of confidence associated with eDNA is by comparing its results with those provided by other traditional sampling techniques. This request for more comparisons is not new (Roussel, Paillisson, Tréguier & Petit, 2015) and a few studies have already provided some insights into it (Aylagas, Borja, Muxika & Rodríguez-Ezpeleta, 2018; Fujiji et al., 2019; Hinlo, Furlan, Suitor & Gleeson, 2017). A meta-analysis article, focusing on the comparison between eDNA and other traditional sampling techniques (e.g., capture-based, visual and/or acoustic methods, such as seine hauls, camera traps and telemetry), selected 194 papers and concluded that eDNA performs worse than traditional methods for reptiles and annelids, but shows higher or equal detection probability for most other groups, making it possible to draw similar and comparable conclusions among techniques (Fediajevaite, Priestley, Arnold & Savolainen, 2021).

Even though it has been suggested that the eDNA approach could replace morphology-based methods, with the workflow remaining largely unchanged or even being improved, authors also note that this is unlikely to happen (Leese et al., 2018; Petruniak, Bradley, Kelly & Hanner, 2020) as it could be complicated and result in both gains and losses, such as identifying changes in richness and at the same time biasing indexes that use those values (Leese et al., 2018; DiBattista et al., 2020). Hence, despite the interviewees not sharing a solution on how to achieve compatibility if eDNA were to replace traditional methods, and published authors not having a uniform wording when talking about it (e.g. complementary, supplementary, integrate, unite, not substitute, not interchangeable, in conjunction), the literature is unanimous on the notion that eDNA is better suited as an addition to current techniques and programs, instead of a replacement option (Belle et al., 2019; Evans & Lamberti, 2018; Harper et al., 2019; Hinlo et al., 2017; Holdaway et al., 2017; Ruppert et al., 2019; Shu, Ludwig & Peng, 2020). That said, traditional sampling methods and

molecular ones each have their own limitations, as their efficiencies vary according to the species of interest and their characteristics, such as size and growth rate, and the surrounding environment (Hinlo et al., 2017). By recognizing that each method is imperfect, we can combine conventional and novel techniques to obtain more complete data, hence overcoming potential weaknesses (Holdaway et al., 2017; Ruppert et al., 2019). For example, Fujiji et al. (2019) compared fish community data from lakes in Japan, using eDNA and seven conventional capture methods, such as minnow traps, gill nets and electrofishing. Despite the high detection rate similarity between the molecular and capture methods, it is of note that at least three species were captured but not present in eDNA analysis, and four sites did not exhibit any eDNA data, possibly due to the presence of PCR inhibitors in the environment. On the other hand, eDNA indicated that most individuals classified only as of the genus *Tribolodon*, due to small body length and consequent difficulties in morphological identification, could be assigned as the species *Tribolodon brandtii*.

Other interviewees' suggestions for enhancing eDNA's credibility are better rigor and transparency, which can be referred to as reproducibility (Marcus, 2015), and repeatability. Considered central tenets of science, reproducibility and repeatability are ways of confirming the validity of results, while protecting scientists and reassuring end-users of the quality of research (Marcus, 2015; Powers & Hampton, 2019; Prager et al., 2019). Reproducibility studies have been under scrutiny in recent years, forcing the proposal and adoption of new steps for research (Marcus, 2015; Munafò et al., 2017), such as pre-registration. Another way to improve credibility is through more peer-reviewed studies that focus on addressing certain gaps in knowledge. While end-users seem already satisfied by applicability and sensitivity of eDNA, with the latter being widely used as a main criterion for analysis (Fediajevaite et al., 2021), their opinions on its accuracy, the level of detail it can provide and its possible biases, are still diverse, as shown by my results. Considering that the credibility of a technique is increased when end-users feel like they have all the information and, therefore, can adjust their expectations, I join the call for the publication of more negative results as well (Belle et al., 2019). With academic success being dependent on publications, and with that process favouring novel and positive results (Fanelli, 2012; Nosek, Spies & Motyl, 2012), scientists are discouraged to reproduce published studies and/or write about negative outcomes (Fanelli, 2012; Nosek et al., 2012; Powers & Hampton, 2019). This practice, which misrepresents reality and possibly skews the scientific literature (Fanelli, 2012), possibly emphasizes eDNA's successes over its weaknesses (Beng & Corlett, 2020) and cautions us to take a step back, tone

down some conclusions and acknowledge exactly what type of information eDNA can deliver (Holdaway et al., 2017; Roussel et al., 2015; Thalinger et al., 2021). Otherwise, by failing to be transparent with methodology and making overly broad claims with insufficient evidence (Fediajevaite et al., 2021; Lortie & Owen, 2020), we risk "underm[ining] legitimate links between evidence and implementation" (Lortie & Owen, 2020, pg. 644).

<u>5.2 – eDNA relevance</u>

Considering relevance as timely, appropriate and informative knowledge, interview results and the literature both suggest that relevance of eDNA in ecological studies is well established and growing, revamping ways to obtain biodiversity data, despite being more likely to complement traditional detection and monitoring methods than to entirely replace them (Blackman et al., 2021; Deiner, Fronhofer, Mächler, Walser & Altermatt, 2016). While results identified scientists as the group with the clearest benefits from its use, eDNA is also relevant and useful to other stakeholders (Harper et al., 2019). When combined with citizen-science and community-based monitoring programs (Biggs et al., 2015; Robinson et al., 2021), eDNA can be used to inform government decisions, hence actively contributing to an enhanced science-policy interface. A classic example would be the monitoring of great crested newts (Triturus cristatus) in the United Kingdom, a species protected under law and that, through some studies with eDNA and citizen-science, saw this technique have an approved protocol for monitoring by Natural England (WC1067) (Biggs et al., 2014; Biggs et al., 2015; Harper et al., 2019). Although promising, some identified caveats to be considered are the translation of eDNA to the general public; how much citizen-derived data is really used to inform water policy (Carlson & Cohen, 2018); and the importance of directing eDNA use towards specific questions instead of just suggesting its use due to its convenience in sampling (Carlson & Cohen, 2018; Robinson et al., 2021).

Interviewees also emphasized that the greatest opportunities for eDNA rely in the inventory-monitoring realm, as a screening and/or a preliminary assessment tool, and that eDNA would be even more relevant if focused on set targets, such as endangered, rare and invasive species. Their impressions reflect the most known, recognized and accepted characteristics of eDNA, its non-invasive properties and sensitivity, and translate the ways eDNA directly benefits conservation and why it has been most used in those areas (Beng & Corlett, 2020; Ruppert et al., 2019). However, this also indicates that the way eDNA is currently regarded and put into practice seems to be under-

utilized (Baillie et al., 2019), since scientific studies have been showing its potential in several other areas. For example, in addition to detecting presence or absence, it has been suggested that eDNA is also able to differentiate between similar (cryptic) species even when they may appear identical during certain life stages and, with that, increase biodiversity data acquisition (Deiner et al., 2017; Evans, Shirey, Wieringa, Mahon & Lamberti, 2017; Reid et al., 2019). Environmental DNA can also ultimately identify trends and changes to community composition, providing a comprehensive overview of the biota (Berry et al., 2019; DiBattista et al., 2020; Ruppert et al., 2019). For example, when investigating marine communities in western Australia, Berry et al. (2019) not only found consistent seasonal assemblages for zooplankton, but they were also able to identify deviations from the regular pattern during environmental perturbations (e.g., heatwaves) and to demonstrate that eDNA is able to deliver a practical whole-sea system approach for investigation. Another study examining biodiversity in coral reefs through different levels of anthropogenic disturbance found that eDNA can "act as a barometer of disturbance" (pg. 1), being able to show changes in richness, fragmentation and indicator taxa (DiBattista et al., 2020).

Furthermore, in times when research and ecological monitoring rely on unstable, long-term financial support (Han et al., 2014; Lortie & Owen, 2020), and an ongoing complaint is the time lag for when results are needed by policy-makers and stakeholders (fixed time frame) versus when they are delivered by scientists (long-term focus) (Shackleton, Richardson et al., 2019), another appealing attribute of eDNA is its cost- and time-effectiveness compared with traditional techniques (Fujiji et al., 2019). For example, Evans et al. (2017) demonstrated that a fish distribution study was at least 33% less expensive with eDNA than presence-absence electrofishing. This was driven by eDNA requiring far less screening effort (6.8 vs. 30 person-hours), suggesting that most of the cost associated with sampling methods result from work remuneration (Evans et al., 2017). Another study, this time on benthic macroinvertebrates from an estuarine and coastal area, showed that analysis cost 55% less and took 72% less time to complete with eDNA than with traditional techniques (van-Veen grab and spade sampling) (Aylagas et al., 2018). In addition, eDNA allows ecological studies to take place over greater temporal and spatial scales (Harper et al., 2019), a characteristic of special value for Canadian studies, where seasonality and accessibility to remote areas may present a challenge.

It is also important to highlight that in my study, through a review of legislation (see Table II) and interviews, I could not find any concrete impediment in legislation that would prevent the

use of eDNA in ecological studies, corroborating previous findings (Sepulveda et al., 2020). By acknowledging that ecological information can be obtained qualitatively and quantitatively (Holdaway et al., 2017), it becomes clear that eDNA is relevant for ecological studies and its use becomes more a case of individual discretion by regulators of what is acceptable, as opposed to any legal obstructions found in legislation.

<u>5.3 – eDNA legitimacy</u>

As legitimacy relates to the transparency and fairness of the process through formal recognition, a recent study reviewing the legal admissibility of eDNA concluded that "eDNA as a method would not be the problem or the concern when validated protocols are used" (Sepulveda et al., 2020, pg. 676). This analysis was done in regard to meeting the *Daubert v. Merrell* (1993) factors, a set of four rules regarding the admissibility of scientific expert evidence:

"...the theory of the new science must be tested; the theory must have been subject to peer review; there must be known error rates and standards; and the theory must have general acceptance." (Booth, Watts & Dufour, 2019, pg. 280)

Even though *Daubert* refers to a U.S. judicial decision, "Canadian law now recognizes those principles for acceptance of novel science" (see *R. v. J.-L.J.*, 2000) (Booth et al., 2019; Glancy & Bradford, 2007, pg. 354); and, as eDNA is a relatively new technique, it is expected that this type of scrutiny will be applied to it.

While standardization of eDNA procedures is seen as a solution to ensure meta-data quality and consistency, a common suggestion by end-users in this study and by other authors (Aylagas et al., 2018; Belle et al., 2019; Harper et al., 2019; Leese et al., 2018; Petruniak et al., 2020; Robinson et al., 2021; Shu et al., 2020), its development is complex and not so easy to achieve. For example, while eDNA meets the Daubert requirement, Sepulveda et al. (2020) also questioned "what are appropriate standards for validating molecular tools prior to implementation" (pg. 676). Linked to this matter is the dualism of standard tests and standard criteria or, as previously mentioned, prescriptive versus broader approaches, for which I could not obtain clear indications from interviewees of what type is better, as they are context dependent. While it is undeniable that the adoption of standards would allow for increased transparency and repeatability (Araújo et al., 2019), thus enhancing not only legitimacy but also credibility, it has been recognized that the development of

eDNA standards that could fit all situations and environments is unlikely (Baillie et al., 2019; Harper et al., 2019; Ruppert et al., 2019), given that existing ones are typically species and site specific (Sepulveda et al., 2020). As standard tests or criteria might also limit the uptake of locally-driven monitoring and Indigenous knowledge (Carlson & Cohen, 2018), I believe the main focus should be in achieving a better balance between prescriptiveness and flexibility, considering availability of methods and quality of those in different risk scenarios (Brown, 2003; Huq et al., 2008; Penas Lado, 2020). Thereby, some current alternatives are the application of decision-trees (Sepulveda et al., 2020; Welsh, Jerde, Wilson, Docker & Locke, 2019), comprehensive workflows (Morisette et al., 2021) and validation scales (Thalinger et al., 2021), which facilitates the science-policy interface between eDNA and stakeholders by offering a simplified, yet not less informative, and possibly more comprehensible, view of the subject. Future initiatives should prioritize quality assurance/quality control measures, such as intercalibration of eDNA with current assessment methods (Hering et al., 2018; Leese et al., 2018), and the adoption of accessible, and clear and consistent terminology across frameworks (Baillie et al., 2019; Lortie & Owen, 2020; Smith et al., 2014).

5.4 – Other considerations

According to interviewees, one of the top three challenges eDNA faces, along with technique resolution and its validity, is users having proper understanding of the technique. At several moments during interviews, participants emphasized that eDNA application must suit the objectives in question or there is a potential for misuse and abuse, just like with any other technique (Greenland et al., 2016; Seegert, 2000; Serinaldi, Chebana & Kilsby, 2020). A good example here is the relation between eDNA and quantitative measures, such as the estimation of abundance. While some studies show statistically significant associations between the number of molecular sequencing reads and species biomass or catch per unit effort (Aylagas et al., 2018; Di Muri et al., 2020), others claim these to be weak correlations or not very concrete estimates, as variability among species, such as eDNA shedding and decay rates, and environmental conditions (e.g. freshwater vs. marine habitats, closed or open systems) may influence the measures (Ruppert et al., 2019; Schadewell & Adams, 2021; Yates, Fraser & Derry, 2019). However, in both scenarios, it appears to be accepted that variability in abundance may be explained by eDNA readings (Yates et al., 2019), with the technique thus acting as an "indirect indicator of abundance" (Hinlo et al.,

2017, pg. 97). But, are those indirect measures enough to comply with current needs or will legislation have to adapt its requirements, as quantitative measures are seen as bottlenecks to eDNA contribution (Leese et al., 2018; Ruppert et al., 2019)? Are stakeholders, regulators and decision-makers willing to accept the trade-offs between more detailed information obtained through the capture of specimens for non-traditional abundance measures derived from a cheaper and non-invasive methodology? As suggested by Sepulveda et al. (2020), at the moment, eDNA limitations in uptake would mostly result from a lack of decision-support frameworks and a misaligned interface between eDNA results and management needs. Thus, it is imperative that users must understand those nuances before proposing to use eDNA and analyzing its results, or they risk affecting the credibility and legitimacy of the technique, as mentioned in previous sections.

A new cause of concern, not only for eDNA but for science in general, is that we now live in a post-truth era, defined as "relating to or denoting circumstances in which objective facts are less influential in shaping public opinion than appeals to emotion and personal belief" (Oxford Dictionaries). During these times in which empirical information and experts are contested and mistrusted, many implications for the CRELE criteria and the science-policy interface are still unknown. Are the current norms still enough, and will CRELE hold up as a robust model (Cash & Belloy, 2020)? It is encouraging to know that even in the post-truth era being transparent regarding numerical uncertainty and limits around process only has a small effect on trust instead of backfiring or inviting criticism (van der Bles, van der Linden, Freeman & Spiegelhalter, 2020) and that there are judicial gatekeeping mechanisms responsible for ensuring that the court is presented "with real knowledge, as different from mere opinion" (Booth et al., 2019; Giocoli, 2020, pg. 207). However, it is also imperative we consider how the post-truth movement could affect the integration of science, including methods like eDNA, into decision- and policy-making (Cash & Belloy, 2020; Lin, 2019).

CHAPTER 6

CONCLUSIONS

"Understanding perceptions will highlight where there might be potential conflicts surrounding the management..."

(Shackleton, Richardson et al., 2019).

In a recent paper, Darling (2019) was able to transform science conversations in what could almost be called a therapy session. Known for previous works discussing the connection of molecular tools and management (Darling, 2015; Darling & Mahon, 2011), by changing the way information is presented it feels like the researcher is trying to communicate with new audiences or even old audiences that were somewhat oblivious. Starting with a catchy title (can you really learn to love a molecular tool?), passing by an abstract with attempts to address 'anxiety' experienced by managers and moving to the main body, where we are presented with four 'virtues' one should focus to facilitate engagement with the eDNA, the focus is always on the actor. For example, all the virtues, namely knowledge, self-awareness, preparedness, and patience, are related to the stakeholders and, even though they are discussed in relation to eDNA, they could be applied to any other sampling tool (Darling, 2019). Similar to Darling's work, one of my main research results is that in the matter of eDNA finding its way into policy and management, it is not so much about the tool itself but ultimately about a 'human factor', demonstrating that social elements are actually decisive and should be paid more attention. For example, interview results demonstrated that the detection of single species by eDNA is already broadly accepted, even between those still skeptical of the tool's application for other uses; and, according to reviewed Canadian legislation, the opportunities for eDNA in environmental monitoring and assessments outnumber the challenges, in the end becoming a case of proponent-regulator discretion.

During this study it became clear that communication is essential to bridge science and policy. With stakeholders 'seeking a voice' in social-ecological governance, which could result in policies with higher levels of trust and more effective and achievable decision-making (Moon et al., 2017), we need to stop seeing scientists, regulators and end-users on different sides and aim for stronger links among them (Leese et al., 2018). One way to help with the misaligned interface between eDNA results and management needs is through engagement of stakeholders from the

beginning. Despite the passive nature of this study, it represents an important first step, by informing on current perceptions, so that active engagement can be sought (Shackleton, Adriaens et al., 2019) and a substantial component of eDNA operational implementation can be checked (Morisette et al., 2021). Some examples are the co-development of projects by eDNA practitioners and decision-makers and better use of scientist-manager partnerships (Moon et al., 2017; Morisette et al., 2021), application of joint fact-finding (Cash & Belloy, 2020), and hands-on evidence-informed scenarios (M. Hecker, personal communication, Nov. 2021). Other ways to help eDNA in this transition are by 1) increasing rigor throughout the whole process, in particular adopting controls and values under QA/QC protocols appropriate for each setting; and by 2) increasing transparency, such as reporting negative results and being honest about limitations, as it has been demonstrated that overselling the technique can potentially be more harmful, by negatively affecting credibility. Hence, more investment in education about eDNA for regulators, so that they can manage expectations; in the development of a common way of translating and reporting results, so it is understandable to all; and in trained personnel performing data collection and analysis is imperative for eDNA's path forward.

In conclusion, this study tried to better understand the issues and implications of the perceptions of end-users and legal frameworks on eDNA acceptance and implementation, ultimately achieving notable progress regarding the role of eDNA in a science-policy framework. As eDNA is a relatively new technique, it is natural that there are still more questions surrounding its application than concrete answers. More important though, is that these questions do not translate into doubts of the effectiveness of eDNA when applied in the right contexts, as demonstrated by the majority of my interviews. Hence, the findings of this thesis allow me to conclude that eDNA is ready for application in management and assessment studies, and there is broad support for its increased uptake amongst academics, community members, regulators, practitioners and legal experts. Despite possible different motivations, it is important that future responsibilities are shared among stakeholders and end-users from distinct sectors, with each group contributing with their expertise in order to fulfill everyone's needs. Due to the dynamic and contextual dimensions of perceptions, of the fact that new techniques are constantly being improved, and on how these social and scientific sides interact with the CRELE criteria, studies like this should be conducted more often to better inform the SPI.

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APPENDICES

A.1: Interview guide

- 1. What is your current professional affiliation/position and for how long have you been engaged with it?
- 2. For you, what are the key areas of concern or main challenges in aquatic environment monitoring and assessment, in Canada?
- 3. A lot has been said in the literature about the need for new tools to improve environmental monitoring of aquatic environments. One of these new tools people have been talking about is environmental DNA. Are you familiar with this technique?
- 4. Thinking about some characteristics of the eDNA tool in relation to other existing and currently in use techniques, please attribute a number from 0 to 10, with 0 being very low and 10 being very high, regarding to how you perceive its: *(Allow the interviewee to say s/he isn't sure)
- Accuracy (the ability to identify, measure and translate the nearby environment as closest to the reality as possible | ex: how accurate it could give a portrait of the sampled environment in different times)
- Bias (a particular tendency or inclination, especially one that prevents impartial consideration of a question | ex: tending towards a particular species due to its specimen's size or quantity, and disregarding others)
- Detail (to report or relate explicitly, thoroughly or meticulously | ex: can it capture abundance or location of species or only presence/absence)
- Sensitivity (the capacity to respond to changes in the environment | ex: how fast it could demonstrate a drop in a species abundance or the arrival of a new invasive species)
- Applicability (ex: would you say it is relevant to environmental studies of aquatic ecosystems)

- Accountability and legitimacy (can be defined by clear roles and responsibilities; penalties for performance; responsiveness; transparency; and a free flow of information between actors Moon et al., 2017 | ex: do you think eDNA is capable of explaining the surroundings and sustaining the results, with undisputed credibility?)
 - -> Following up, ask the interviewee why did they chose that number, what was their line of thought for each characteristic.
- 5. My next question is about uses of eDNA information in different sectors of society.
 - How do you think eDNA results would be valuable to 1) Scientists; 2) Regulatory decision-makers;
 - 3) Community members (e.g. members of the public who are concerned about their local environment); 4) Industry consultants and 5) Review panels.
- 6. I would like to have you reflect on a hypothetical scenario where eDNA might be applied. Imagine a new mine development is being considered for approval in northern Saskatchewan. For you, what might be the utility of the eDNA tool in this particular case?
- 7. What do you believe to be the main scientific barriers and/or challenges for the eDNA implementation in aquatic environments assessments and monitoring?
 - -> (Probe A: Do you believe pre-existing perceptions and values of people towards molecular tools may jeopardize the acceptance of this technique?)
 - -> (Probe B: How to achieve compatibility in ecological assessments when replacing conventional by novel methods?)
- 8. We know that sometimes questions related to environmental assessments and monitoring may be involved in legal disputes. Do you believe eDNA would hold up on court?
- 9. How strong does the evidence need to be in order for the tool to be reliable? How to manage uncertainty in your view?
- 10. In face of all discussed here today, do you believe this new methodology could be embraced and recommended by regulatory agencies? Would you care to explain further your answer?

- -> (Probe C: In your opinion what could be a regulatory challenge to implement this methodology?)
- 11. Where do you think the greatest opportunities to implement this methodology are? What would be an immediate gain of implementing it as soon as possible?
 - -> (Probe D: What existing governance arrangements, stakeholder groups and programs could support immediate action; and how could immediate action affect, and be affected by, existing arrangements, structures and institutions?)
- 12. Who should be leading the development and application of the eDNA methodology? Is it up to the government to guarantee its applicability, to the private sector or should this be restricted to the academy at least for now?
- 13. Is there anything else that you would like to comment or recommend regarding your perceptions around the applicability of this new methodology for environmental studies and its implementation in environmental programs? *(Ask if they recommend someone else I should be talking to.)

A.2: Contacted participants by occupation and geographic categories

Those who accepted are represented by Arabic numbers and those who declined or didn't replied are represented by Roman numerals. Snowballed participants are indicated with an asterisk (*) after their number. The information on the column further to the right (level of knowledge about environmental DNA) was provided by the interviewees themselves.

| Occupation | Province | Participant | Status | Interview mode | Duration | Level of knowledge about eDNA |
|------------|-----------|-------------|-------------|---------------------------------|----------|---|
| ACADEMIA | \ | | | | | |
| | Alberta | | | | | |
| | Alberta | 10 | Accepted | Audio-only phone call recording | 35'20" | Knowledgeable (is using it) |
| | | 27 * | Accepted | Audio-only phone call recording | 32'50" | Familiar |
| | | XXII | No Response | Audio-only phone can recording | 32 30 | Tallillai |
| Sac | katchewan | AAII | No Response | | | |
| Sas | Katchewan | 39 | Accepted | Audio-only phone call recording | 29'04" | Familiar (has used before) |
| | | IX | Declined | rudio only phone can recording | 2704 | Tammar (nas asea before) |
| | Ontario | IA | Decimed | | | |
| | Ontario | 7 | Accepted | Audio-only phone call recording | 53'20" | Knowledgeable |
| | | , | Accepted | Audio-only phone can recording | 33 20 | Knowledgeable |
| COMMUNI | TY | | | | | |
| | Alberta | | | | | |
| | | 14 | Accepted | Audio-only phone call recording | 33'34" | Familiar (has read about it) |
| | | 17 * | Accepted | Audio-only phone call recording | 45'46" | Aware |
| | | 32 * | Accepted | Audio-only phone call recording | 30'08" | Familiar (has read about it/attended workshops) |
| | | 36 | Accepted | Audio-only phone call recording | 22'01" | Familiar (has read about it) |
| | | XIV | No Response | <i>y</i> 1 | | , |
| Sas | katchewan | | 1 | | | |
| | | 4 | Accepted | Audio-only phone call recording | 33'37" | Familiar |
| | | 11 | Accepted | Audio-only phone call recording | 19'02" | Familiar |
| | | 15 | Accepted | In Person (but not used) | | |
| | Ontario | | 1 | , | | |
| | | 2 | Accepted | Audio-only phone call recording | 39'36" | Familiar (has used before) |
| | | 6 | Accepted | Audio-only phone call recording | 18'41" | Aware |
| | | 28 | Accepted | Audio-only phone call recording | 37'32" | Aware |
| | | | • | 7.1 | | |

| Nationwide | V X | Declined Declined | | | |
|--------------|--------|-----------------------------|---------------------------------|----------|---------------------------------|
| | 9 | Accepted | Audio-only phone call recording | 24'07" | Knowledgeable (is using it) |
| | 18 | Accepted | Audio-only phone call recording | 43'31" | Knowledgeable (is using it) |
| GOVERNMENT | | | | | |
| Alberta | | | | | |
| | 8 | Accepted | Audio-only phone call recording | 29'20" | Aware |
| | 21 * | Accepted | Audio-only phone call recording | 1°02'43" | Aware |
| | 30 * | Accepted | Audio-only phone call recording | 25'14" | Familiar (has read about it) |
| | XV | No Response | | | |
| | XX | No Response | | | |
| Saskatchewan | | - | | | |
| | 1 | Accepted | Audio-only phone call recording | 31'42" | Knowledgeable (is using it) |
| | 12 | Accepted | Audio-only phone call recording | 23'14" | Knowledgeable |
| | 35 | Accepted | Audio-only phone call recording | 28'43" | Knowledgeable |
| | 41 | Accepted | In writing | | Knowledgeable |
| | 33 | Accepted but later declined | | | |
| | IV | Declined | | | |
| Ontario | | | | | |
| | 22 | Accepted | Audio-only phone call recording | 1°18'29" | Knowledgeable (is using it) |
| | 37 | Accepted | Audio-only phone call recording | 47'08" | Aware |
| | II | Declined | | | |
| | XIII | No Response | | | |
| Nationwide | | | | | |
| | 13 | Accepted | Audio-only phone call recording | 36'39" | Knowledgeable (is using it) |
| | 31 * | Accepted | Audio-only phone call recording | 36'31" | Knowledgeable (has used before) |
| | 34 | Accepted | Audio-only phone call recording | 48'43" | Knowledgeable (is using it) |
| | 38 | Accepted | Audio-only phone call recording | 48'43" | Familiar |
| | XVIII | No Response | | | |
| | XIX | No Response | | | |

| Alberta | | | | | |
|--------------|-------------|-------------|---------------------------------|--------|---|
| | 24 | Accepted | Audio-only phone call recording | 40'51" | Aware |
| | XXI | No Response | | | |
| Saskatchewan | | | | | |
| | 3 | Accepted | Audio-only phone call recording | 50'51" | Familiar (has read about it) |
| Nationwide | | | | | |
| | 20 | Accepted | Audio-only phone call recording | 24'25" | No |
| | 23 | Accepted | Audio-only phone call recording | 55'44" | Aware |
| PRIVATE | | | | | |
| Alberta | | | | | |
| | 19 | Accepted | Audio-only phone call recording | 34'53" | Aware |
| | 40 | Accepted | Audio-only phone call recording | 28'05" | Familiar |
| | I | Declined | | | |
| Saskatchewan | | | | | |
| | 5 | Accepted | Audio-only phone call recording | 26'31" | Familiar |
| | 16 | Accepted | Audio-only phone call recording | 32'48" | Familiar (has read about it/attended workshops) |
| | III | Declined | | | |
| | VIII | Declined | | | |
| | XI * | Declined | | | |
| Ontario | | | | | |
| | 26 * | Accepted | Audio-only phone call recording | 28'18" | Familiar (has read about it/attended workshops) |
| | 42 * | Accepted | In writing | | Knowledgeable |
| | VI | Declined | | | |
| | VII | Declined | | | |
| | XII | Declined | | | |
| NT 11 | XVI | No Response | | | |
| Nationwide | 2.5 | | | 20125" | |
| | 25 20.** | Accepted | Audio-only phone call recording | 29'35" | Knowledgeable (is using it) |
| | 29 * | Accepted | Audio-only phone call recording | 30'37" | Knowledgeable (is using it) |
| | XVII | No Response | | | |

A.3: Coding table

| | CODES | FILES |
|-------|---------------------------|-------|
| | Climate change impacts | 1 |
| | Cumulative effects - | 10 |
| | Multiple stressors | 10 |
| | Data | 16 |
| | Baselines and trends | |
| | Data management | |
| | Fragmented data | |
| | Monitoring design | |
| | Decision-making | 2 |
| | Depends | 1 |
| | Inconsistency | 5 |
| | Knowledge transfer | 3 |
| QUES- | Lack of | |
| TION | Resources | 9 |
| | Scientific methodol- | |
| 2 | ogy | 6 |
| | Standards | 2 |
| | Support | 3 |
| | Management | 6 |
| | Adaptive management | |
| | On fisheries | |
| | Results | 4 |
| | Endgame | |
| | Imprecision | |
| | Species (Invasive and At | 7 |
| | Risk) | 7 |
| | Unifying political regime | 6 |
| | Water quality and quan- | 3 |
| | tity | 3 |

| | CODES | FILES |
|----------|-------------------------|--------------|
| | Depends on the question | 4 |
| | Migration patterns | 2 |
| QUESTION | Notuseful | 11 |
| QUESTION | Quality monitoring pa- | 1 |
| 6 | rameter | 4 |
| · · | Screening-Snapshot- | 27 |
| | Baseline | 21 |
| | Target work | 12 |
| | Trends - Monitoring | 16 |

| Community Members 3 If they are interested 3 If translated into plain language 5 Limited 4 No direct use 10 Not much different from other programs 3 Potential 11 Valuable for specific questions 5 Invasive Species 2 Decision-Makers Depends on Purpose 7 Weight on evidences 3 Good tool but not there yet 5 Helpful 11 Incredibly valuable 7 Limited 1 Has to be interested in what eDNA can say Not (very) useful 5 Industry Consultants Already using it 2 Depends 2 Helpful/Valuable 18 |
|--|
| If they are interested If translated into plain language Limited No direct use Not much different from other programs Potential Valuable for specific questions Invasive Species Decision-Makers Depends on Purpose Weight on evidences Good tool but not there yet Helpful Incredibly valuable Limited Has to be interested in what eDNA can say Not (very) useful Industry Consultants Already using it Depends 2 Intimited Industry Consultants Already using it Depends |
| If translated into plain language Limited No direct use Not much different from other programs Potential Valuable for specific questions Invasive Species Decision-Makers Depends on Purpose Weight on evidences Good tool but not there yet Helpful Incredibly valuable Thas to be interested in what eDNA can say Not (very) useful Industry Consultants Already using it Depends 10 4 Not much different from other 3 3 4 Potential 11 Valuable for specific questions 5 Invasive Species 2 Decision-Makers 2 Depends 7 Weight on evidences 3 Good tool but not there yet 5 Helpful Incredibly valuable 7 Limited 1 Has to be interested in what eDNA can say Not (very) useful 5 Industry Consultants Already using it 2 Depends |
| Limited No direct use 10 |
| No direct use |
| Not much different from other programs 3 |
| Potential |
| Valuable for specific questions 5 |
| Invasive Species 2 Decision-Makers Depends on Purpose 7 Weight on evidences 3 Good tool but not there yet 5 Helpful 11 Incredibly valuable 7 Limited 1 Has to be interested in what eDNA can say Not (very) useful 5 Industry Consultants Already using it 2 Depends 2 |
| Invasive Species 2 Decision-Makers Depends on Purpose 7 Weight on evidences 3 Good tool but not there yet 5 Helpful 11 Incredibly valuable 7 Limited 1 Has to be interested in what eDNA can say Not (very) useful 5 Industry Consultants Already using it 2 Depends 2 |
| Decision-Makers Depends on 7 Purpose 7 Weight on evidences 3 Good tool but not there yet 5 Helpful 11 Incredibly valuable 7 Limited 1 Has to be interested in what eDNA can say 3 Not (very) useful 5 Industry Consultants 5 Already using it 2 Depends 2 |
| Purpose 7 |
| Purpose 7 |
| Weight on evidences 3 |
| Good tool but not there yet 5 |
| Helpful |
| Incredibly valuable 7 |
| Limited 1 |
| QUESTION eDNA can say 3 Not (very) useful 5 Industry Consultants 2 Already using it 2 Depends 2 |
| QUESTION eDNA can say Not (very) useful 5 Industry Consultants 2 Already using it 2 Depends 2 |
| Not (very) useful 5 Industry Consultants Already using it 2 Depends 2 |
| 5 Industry Consultants Already using it 2 Depends 2 |
| Already using it 2 Depends 2 |
| Depends 2 |
| - |
| |
| Not valuable 1 |
| Potential for abuse 1 |
| Should be open to it 3 |
| Tied with the decision-makers 7 |
| Review Panels |
| All about weight of evidence 8 |
| Depend on the type of panel 4 |
| Has potential but not there yet 3 |
| Informally 1 |
| Not valuable 2 |
| Similar to Decision-Makers 11 |
| Valuable 8 |
| Scientists |
| Limited 8 |
| Depends on their interest |
| Not valuable 1 |
| Valuable 28 |

| | CODES | FILES |
|-------|--------------------------------------|-------|
| | Bioinformatics | 1 |
| | Contamination - False positives | 5 |
| | Cost | 3 |
| | Delivery methods - Capacity | 9 |
| | Knowledge mobilization | 1 |
| QUES- | Politics | 1 |
| TION | Technique resolution | |
| 7 | Biodiversity and Primer availability | 10 |
| | Low Detection | 2 |
| | Quantitative | 6 |
| | Time for results | 1 |
| | Users | |
| | Proper Understanding | 10 |
| | Qualified personnel | 5 |
| | Validity | 12 |
| | Lack of standards | 5 |

| | CODES | FILES |
|-------|--------------------------|-------|
| | Can't comment | 4 |
| | Depends. IF | 17 |
| QUES- | Favored compared to tra- | |
| TION | ditional tech. | |
| | No | 1 |
| 8 | Only way | 1 |
| | Potential for it | 1 |
| | No | 11 |
| | Yes | 12 |

| | CODES | FILES |
|---------------|----------------|-------|
| OHEG | Already has | 3 |
| QUES- TION | Don't know | 3 |
| TION | Hypothetically | 2 |
| 10 | No | 4 |
| 10 | Promising | 3 |
| | Yes | 27 |

| | CODES | FILES |
|----------|----------------------------------|-------|
| | Acceptance | 3 |
| | Already reliable | 2 |
| | Depends on the type of survey | 1 |
| | Can't answer | 2 |
| | Comparison with other tools | 11 |
| | Levels of certainty | |
| | Degrees of probability-Statis- | 5 |
| QUESTION | tics | 3 |
| | Published literature | 5 |
| 9 | QA-QC | 2 |
| | Repeatability | 5 |
| | Rigor and transparency | 7 |
| | Standard of proof-Types of | 3 |
| | dispute | 3 |
| | Low detection limits-High accu- | 3 |
| | racy | 3 |
| | Not an uncertainty issue | 2 |
| | Up to scientists to determine it | 1 |

| | CODES | FILES |
|-----------------|---------------------------------|--------------|
| | Ecosystem recovery | 2 |
| | Inventory-Monitoring | 17 |
| | Less harmful | 1 |
| | Microbial | 1 |
| | No gain yet | 3 |
| | Need for improved library | 1 |
| | Need for numbers | 1 |
| QUESTION | Need for standard protocols 1st | 1 |
| | Partnerships for monitoring | 4 |
| 11 | Collab with communities | 3 |
| | Savings | 8 |
| | Scientific application | 2 |
| | Target tool | |
| | Endangered and rare species | 10 |
| | monitoring | 10 |
| | Invasive species monitoring | 13 |
| | Sampling areas | 1 |
| | Water quality indicator | 2 |

| | CODES | FILES |
|----------|-----------------------------------|-------|
| | Academy | 20 |
| | All together | 19 |
| | Government | 1 |
| | Government - applica- tion | 4 |
| QUESTION | Government - develop- ment | 1 |
| 12 | Government - support | 3 |
| | Private sector | 1 |
| | Private sector - application | 6 |
| | Private sector - develop- ment | 1 |

| | CODES | FILES |
|-------------|----------------------------|-------|
| QUESTION 13 | Accreditation | 2 |
| | Bias view from users | 5 |
| | Broader applications | 2 |
| | Collaboration | 2 |
| | Community involvement | 1 |
| | Limitations | 5 |
| | Obstacles (Political will) | 1 |
| | Short and long term con- | 1 |
| | servation monitoring | 1 |

| STANDARDS | CODES | FILES |
|-----------|------------------|-------|
| | Both | 1 |
| | Criteria | 4 |
| | Don't know / De- | 1 |
| | pends | 4 |
| | For labs | 2 |
| | Protocols | 2 |
| | Tests | 2 |

| | CODES | FILES |
|--------------|--------|-------|
| PROBE | No | 21 |
| \mathbf{A} | Unsure | 8 |
| | Yes | 8 |

| | CODES | FILES |
|------------|-----------------------------|-------|
| PROBE B | Can't answer | 11 |
| | Depends on the evidence out | |
| | there | |
| | Depends on the question | |
| | Inappropriate question | |
| | Potential to stand alone | 10 |
| | Supportive | 16 |

| | CODES | FILES |
|-------|--|-------|
| | Association between eDNA results and projects impacts | 1 |
| | Budget constraint | 1 |
| | Depends | 1 |
| | Gap between science methods and legislation | 1 |
| | Industry role | 2 |
| PROBE | Mismatch between eDNA answers and regulatory questions | 5 |
| C | Need for guidelines and accreditation | 1 |
| | No challenges | 11 |
| | Political will | 2 |
| | Regulatory agencies characteristics | 4 |
| | Uncertainty-Confidence | 9 |
| | Need for robust and peer reviewed scientific evidence | 3 |

| | CODES | FILES |
|------------|--|-------|
| PROBE D | Academic and Government Associations | 3 |
| | CABIN | 1 |
| | Community based programs | 3 |
| | Conservation authorities / Stew- ardship groups | 3 |
| | Industrial funding arrangements | 1 |
| | Invasive species detection programs | 3 |
| | Not able to disclose | 1 |
| | Provincial EIA or ESA | 2 |
| | Sensitive or at risk species detection | 1 |