





# The Pragmatic Sceptic: A Practical Approach for Integrating Environmental DNA Into Marine Stock Assessment and Fisheries Management

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### **ABSTRACT**

The 'omics revolution' has advanced scientific understanding of marine ecosystems and led to a rapid increase in data that can inform species' population structure, distribution, and abundance. Of the 'omics data types, environmental DNA (eDNA) may present the most cost-effective opportunity for developing quantitative estimates of abundance trends, a key input for stock assessment models. However, eDNA has yet to be widely adopted for stock status determinations within regional fisheries management organisations. We review progress towards addressing key challenges that limited the application of eDNA in marine fisheries management, including advances in (1) the quantitative relationship between eDNA observations and species biomass, (2) reducing false-negative and false-positive detections, (3) defining the spatial scale of eDNA, (4) collecting biological data from eDNA surveys, (5) quantifying uncertainty in eDNA surveys, and (6) responding to scepticism of new survey methods. We use a case study with Pacific hake (*Merluccius productus*) to demonstrate the development of an eDNA index and its direct integration into an age-structured stock assessment model. Given the many ways in which the field of eDNA has matured, we propose that eDNA can meaningfully inform a range of fisheries management needs, and outline a roadmap for using eDNA in stock assessment models in data-limited to data-rich species. A primary impediment to operationalising eDNA as stock assessment model inputs is the lack of interdisciplinary research teams, including geneticists, ecological modellers, and stock assessment scientists, which are necessary to interpret methods and results across scientific disciplines and ensure data are used appropriately.

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### 1 | Introduction

The 'omics revolution is touted as a potential sea change for managing marine resources (Rodríguez-Mendoza and Saborido-Rey 2023). Given rapid fluctuations in species distributions and abundance due to changing environmental conditions, methods to aid scientifically informed management are critical (De León et al. 2023). Indeed, many fisheries management organisations have identified investment in 'omics as a high priority to support and supplement stock assessments (ICES 2020), which form the scientific basis for robust fisheries management. Of the many types of 'omics data, environmental DNA (eDNA) may present the most costeffective opportunity for developing quantitative estimates of abundance, a key input for stock assessment models. Over the last decade, the field of eDNA has significantly matured with proliferating empirical studies, development of best practices and substantial improvements to the analytical landscape. However, we know of only one recent example where eDNA has been directly evaluated in a stock assessment model as an abundance index for a marine species (Johnson et al. 2025).

We suspect that the lag between scientific developments in eDNA approaches and realised applications for fisheries management decision-making is due, at least in part, to the scarcity of interdisciplinary research teams, the tendency for research to be siloed, and the lack of a common nomenclature among geneticists and fisheries scientists. The disconnect is evident given the number of eDNA studies that provide general recommendations for use in fisheries management, yet do not provide an approachable template for how data or results should be used by non-geneticists (but see Stein et al. 2024 for a framework for communicating eDNA science). Where interdisciplinary collaborations between eDNA researchers and natural resource managers do exist (e.g., Maine-eDNA and eDNAqua-Plan), there is progress developing standardised practices and moving eDNA towards adoption (Kelly et al. 2024; Lee et al. 2024). And yet, most eDNA studies focus on technical laboratory details and tend to appear in specialised journals, which makes them less accessible to stock assessment scientists and managers (Jo et al. 2022; Ramírez-Amaro et al. 2022; but see Gilbey et al. 2021). Similarly, reviews of hypothetical applications for eDNA tend to stop short of demonstrating how to develop an eDNA abundance index that corresponds to the spatiotemporal scale of inference for an assessment (i.e., an entire 'stock' or population) and lack methods to quantify uncertainty (Rodríguez-Mendoza and Saborido-Rey 2023; Rodríguez-Rodríguez et al. 2022). Integrating eDNA products within fisheries management frameworks will require interdisciplinary research teams and discussions on eDNA data collection, use, and how sampling and measurement errors can be adequately quantified.

Here, we review the current state-of-the-science as it pertains to using eDNA data as a quantitative input for marine fisheries stock assessments (i.e., statistical population models used for determining stock status, see Table 1 for definition). We describe progress addressing key challenges that have been highlighted as potentially limiting factors for integrating eDNA (i.e., Beng and Corlett 2020; Hansen et al. 2018), including (1) the quantitative relationship between eDNA observations and species

abundance or biomass; (2) false-negative and false-positive detections; (3) defining the spatial scale of eDNA; (4) the lack of available biological data from eDNA surveys; (5) quantifying uncertainty in eDNA surveys; and (6) scepticism of new survey methods. Using examples from recent empirical studies, we discuss how these challenges can be overcome. A case study with Pacific hake (Merluccius productus) then summarises the development of an eDNA index and demonstrates the subsequent direct integration into an age-structured stock assessment model (Johnson et al. 2025). Finally, we propose a roadmap for integrating eDNA into management decision-making and associated stock assessment models. For data-moderate and datarich species assessments, the threshold for including an index of abundance from an eDNA survey is necessarily higher than for data-limited species, for which eDNA might add to otherwise limited information. Here, we present explicit real-world examples, described with limited jargon, in an attempt to bridge the communication gap between geneticists and stock assessment scientists.

### 2 | Current Use of eDNA in Stock Assessments

Despite much anticipation about the use of eDNA in marine fisheries stock assessments (Gilbey et al. 2021; Hansen et al. 2018), few studies provide quantitative information across the entire spatial management unit of a stock, as required for developing an eDNA index related to abundance. A prime example of this type of study is Shelton et al. (2022), in which an 86,000 km<sup>2</sup> eDNA survey was designed to parallel a traditional acoustic-trawl survey for evaluating the distribution and abundance of Pacific hake. Multiple years of data collected on this survey then contributed to developing an eDNA index that was evaluated in the stock assessment model (Johnson et al. 2025). We provide an in-depth look at this example in Section 4.1.1. Other eDNA surveys have covered the spatial extent of fisheries stocks, but have not extended their analyses to develop quantitative indices of abundance for stock assessment models. For example, the BIOMAN 2017 survey (Fraija-Fernández et al. 2020) sampled eDNA across 120,000 km<sup>2</sup> in the North Atlantic, but the aim of this study was biodiversity assessment, and a quantitative relationship between eDNA and species biomass was not established, making it of little use for a stock assessment. In freshwater, Pont et al. (2023) surveyed 2850 km of the Danube River and its tributaries and correlated DNA copies with biomass for multiple species based on traditional electrofishing methods. Here, the quantitative relationship between eDNA and biomass could be extended to develop an index of abundance if stock assessment models were an objective. Similarly, many eDNA studies provide evidence of positive correlations between eDNA and species abundance or biomass, and suggest future applications for stock assessments (Hata et al. 2025; Osathanunkul and Madesis 2022; Wang et al. 2021; Yoshitake et al. 2021). Although this positive relationship between eDNA and abundance is a reasonable expectation if eDNA were to be used in a stock assessment, this alone does not make the data usable in an assessment model. Ideally, an eDNA index is based on a carefully designed, multi-year survey (as described in Dias et al. 2025). Empirical studies correlating eDNA concentration with biomass and abundance are globally distributed,

**TABLE 1** | Glossary of common terms for eDNA and stock assessment.

Term	Definition		
Environmental DNA (eDNA)	DNA (cells and extracellular material) released into the environment by a variety of physical (e.g., shedding) or biological mechanisms		
DNA primers	Short synthetic DNA sequences that are complementary to the beginning and end of the desired DNA sequence to be amplified using PCR, qPCR, ddPCR or metabarcoding. Primers can be designed to target a single species, group of species or more broadly (i.e., teleost fishes)		
Quantitative PCR (qPCR)	Single-species quantification based on a target sequence. Quantification is relative to known standards that have a predetermined number of DNA copies of the target		
Digital-droplet PCR (ddPCR)	Single-species absolute quantification of DNA copies independent of standard curves. Species-specific primers bind to the target DNA sequence.  Optimal approach for rare and low-concentration targets		
DNA metabarcoding	Multispecies compositional data generated by targeting and amplifying a short genomic DNA sequence. Metabarcoding data are compositional and often reported as relative proportions. Quantitative metabarcoding uses absolute quantification of at least one species to correct for amplification biases across the species present in the compositional data		
Index of abundance	A time series of relative population (or stock) size (informing biomass or numbers) from a fishery-independent or fishery-dependent survey, which is often used within a stock assessment model to infer both population trends and magnitude (based on the scaling coefficient—catchability—parameter estimate)		
Selectivity	The relative probability of capture by size (i.e., by length or age) for a given sampling gear for individuals that encounter the gear (i.e., are available to the gear at the time of sampling), often scaled to a reference size where selectivity is the highest		
Catchability	The relative proportion of a fish population that is sampled by a unit of survey or fishing effort (i.e., the scaling coefficient between relative abundance and absolute abundance)		
Availability	The proportion of the fish population that is within a sampling unit at the time of sampling and, thus, will be able to interact with the sampling gear (i.e., the proportion of the population that is vulnerable to the sampling gear)		
Stock assessment model	A statistical population model that calculates population trends and absolute magnitude through estimation of key population parameters by fitting to observed data (i.e., landings, indices of abundance and age or length composition data), thereby providing estimates of stock health (status, relative to a target biological reference point) and the impacts of fishing on the population; associated catch forecasts project harvest levels based on a pre-defined harvest control rule (HCR) that defines long-term sustainable yields based on current and future stock status		
Uncertainty	Incomplete knowledge regarding the dynamics of a system, including both human error (e.g., measurement error during data collection) and unknown states of nature (e.g., error in model parameterisation or parameter estimates)		
Harvest control rule (HCR)	A decision rule that defines the management action to be undertaken (i.e., quota or TAC level) based on current and future status of the resource; an HCR is typically used within a catch projection model		
Total allowable catch (TAC)	The prescribed catch (quota) to be removed from the resources in a given time period (i.e., a fishing season) as determined by the fishery management body		

cover both marine and freshwater applications, and reinforce the notion that eDNA data are poised to benefit fisheries assessments for a wide range of species, but remain limited in their direct application to stock assessment models.

More common are applications for eDNA in fish community and biodiversity assessments and aquatic monitoring (Blancher et al. 2022; Li et al. 2019). Often, biodiversity applications

apply molecular methods that allow for the identification of many species simultaneously (i.e., metabarcoding, see Table 1) and may use site occupancy or frequency of occurrence as proxies for abundance (Hänfling et al. 2016). More recently, species-specific quantitative information can be extracted from multispecies data using models based on DNA mixtures of known species constructed in a laboratory (i.e., mock communities; Shelton et al. 2023). The details of multispecies and

single-species approaches for eDNA data are described extensively elsewhere (Gilbey et al. 2021; Ramírez-Amaro et al. 2022; Rourke et al. 2022) and should be part of the decision-making process for an interdisciplinary team that includes eDNA researchers and stock assessment scientists.

### 3 | Key Challenges for Integrating eDNA Into Assessments and How to Overcome Them

A stock assessment aims to estimate the absolute biomass. time series trends and population status (i.e., biomass relative to a defined long-term reference point) of a species within a spatial management unit. To be integrated into a stock assessment, an eDNA survey must provide data that informs current biomass or abundance while ideally establishing a time series of these observations to inform population trends. Moreover, inputs to stock assessments must include quantitative estimates of observation uncertainty, which enable the model to balance statistical fit to multiple data inputs (surveys, age or length compositions, and fishery catch) when estimating parameters (Maunder and Punt 2013). The following sections discuss proposed challenges identified as limiting factors for eDNA integration into assessment processes (Hansen et al. 2018) and highlight why these challenges may no longer be significant obstacles.

# 3.1 | Challenge 1: Developing a Quantitative Relationship Between eDNA and Abundance

Recent studies demonstrate strong positive relationships between eDNA concentration and biomass across a range of laboratory and field studies, habitats, and spatial scales (reviewed in Rourke et al. 2022). Positive comparisons between biomass estimates from eDNA and diverse gear types included the following: trawls of Atlantic cod (Salter et al. 2019), North Sea flatfishes (Maes et al. 2023) and commercial fishes in a Norwegian fjord (Guri, Shelton, et al. 2024); and acoustic surveys of reef fishes (Sato et al. 2021) and Pacific hake (Shelton et al. 2022). However, there remain uncertainties associated with eDNA data including understanding the effects of availability (i.e., what portion of the population is being sampled) and demographic structure of individuals in the 'sampling unit' (i.e., the species, length or age composition of the samples; see next section); but these challenges are present in virtually all fishery surveys. As with any operational survey, an eDNA survey should be developed using a study design in which the methodology and sampling design are determined a priori and maintained across years, such that the scaling between the relative index and absolute abundance (i.e., 'catchability') remains consistent. Moreover, eDNA sampling can be combined with existing sampling or survey methods (e.g., trawl surveys), which may help overcome limitations related to size selectivity. For example, the size structure of a population can increase the uncertainty of the eDNA abundance/biomass relationship because the proportionality between eDNA shedding rates and fish biomass may vary with fish size (Ledger et al. 2024). However, the composition of fish contributing to eDNA could be inferred by using concurrent sampling from other methods (e.g., trawl), and then estimating the maximum

difference in shedding rates between different size classes for a given species. Whereas uncertainty in the size composition of the sampled population is a common issue for eDNA as well as other survey types (e.g., acoustic surveys), eDNA can effectively eliminate other sources of uncertainty that are difficult to address for non-genetic methods (e.g., species identification; Fall et al. 2024; see Section 4.3).

# 3.2 | Challenge 2: Reducing False-Positive and False-Negative Detections

Failure to detect an organism that is present (false negatives) or detecting an organism that is not present (false positives) are both potential sources of uncertainty for eDNA (Darling et al. 2021; Jerde 2021). False negatives can arise due to physical dilution (i.e., high water volume-to-biomass ratio in marine environments) or technical issues associated with laboratory or analysis protocols (i.e., inadequate or incorrect reference sequences or lack of species-level resolution in the target DNA sequence; Hansen et al. 2018). Conversely, false positives may arise from collecting eDNA from trawl vessels, where the act of trawling can potentially bias (high) eDNA concentrations in water samples or, perhaps more relevant, ambient fish DNA from trawls can contaminate eDNA samples being filtered onboard. Addressing low-concentration or dilute eDNA requires filtering larger volumes of water, thereby collecting more eDNA (Thomas et al. 2018) and generating quantitative data at very low eDNA concentrations, for example, using digital-droplet PCR (ddPCR; Dias et al. 2025; Guri, Ray, et al. 2024).

Issues encountered during analysis of eDNA data are now better understood and methods have been developed to address potential false negatives. For instance, the availability of reference data has increased, particularly for commercially important fishes, and an eDNA survey must ensure that focal species have reference data, including geographic representation, as well as sequence information for closely related species. Similarly, incorrectly identified sequences can be identified and filtered out based on new software for building reference databases (Jeunen et al. 2023; Keck and Altermatt 2023). New eDNA primer sets have been designed to provide species-level identification for desired taxa (Ledger et al. 2024), and recently developed taxonomic assignment approaches exist for analysing data (Pipes and Nielsen 2024). Moreover, improvements in the use of controls allow for evaluating sources of both false positives and false negatives (Govindarajan et al. 2022; Kelly et al. 2018). False positives (i.e., contamination from sampling aboard fishing vessels) can be prevented through collection of eDNA prior to trawling at a given station, or through the use of specialised sampling equipment that allows water to be sampled, filtered, and preserved in situ, which minimises the likelihood of capturing ambient DNA from a trawling vessel. Also, it is important to note that trawling at one station is unlikely to impact the concentration of eDNA at another station many kilometres away (see next section). Therefore, false detections in eDNA are likely to be increasingly rare for commercially harvested marine species, and possibly much lower than in most other fisheries sampling methods that rely on expert judgement (i.e., species assignment from acoustic survey) or are prone to human error (e.g., species assignment by human observers on mixed-species trawls).

## 3.3 | Challenge 3: Defining the Spatial Scale of eDNA

Studies demonstrate that the strongest concentration of eDNA is in closest proximity to the organism, with a precipitous drop-off in eDNA with distance and time (Andruszkiewicz et al. 2019; Collins et al. 2018; Jensen et al. 2022). Although eDNA can be detected some distance away, experimental and empirical studies suggest that maximum eDNA transport does not exceed ~5 km (Baetscher et al. 2024; Shea et al. 2022). More typically, eDNA is only detected within 10-100s of metres of the organism, depending on the biomass of the target species (Murakami et al. 2019). The vertical behaviour of eDNA emulates other particles that interact with ocean depth based on density: eDNA attached to large or weighted particles (faeces, tissue) will sink quickly, whereas scales and skin cells are more restricted to the ocean depth layer where they were shed, with diffuse sinking behaviour (Jeunen et al. 2020). Additional evidence for the vertical dispersion of eDNA comes from studies of diel vertical migration, where eDNA paired with acoustic sensors identified species undergoing diel migrations (Easson et al. 2020). Thus, water collections for eDNA should be within the same density layer as the species of interest (i.e., sampling for groundfish should be aligned with the depth where trawl surveys traditionally sample for these species). Moreover, eDNA does not persist for long periods of time because of rapid dispersal and dilution of genetic material and biological or physical degradation (Allan et al. 2021; Lamb et al. 2022).

In many commercially exploited marine fisheries, the spatial scale of management is 1000–10,000 s of km<sup>2</sup> and sampling locations for many fishery-independent trawl surveys are separated by at least 1-10s km (Clemons et al. 2024; Stauffer 2004). Thus, eDNA from water samples and detection probabilities at the scale of a regional survey will only reflect organisms that are currently present at the sampling station or were present very recently (i.e., within < 8 h; Ely et al. 2021). Although this allows for the possibility that a highly mobile species could be detected across multiple survey locations—even 10 km apart—the same critique would be true for acoustic or camera surveys, when animals are sampled non-invasively. The implementation and treatment of an eDNA survey should be envisioned similar to other survey types (e.g., acoustics or a trawl net), where a fish will only be observed if it is in the path of the sampling gear at the time of the survey. Therefore, assumptions regarding the availability, relative catchability, and selectivity (see Table 1) of the eDNA survey gear are comparable to other survey types (e.g., acoustic surveys that assume only mature or immature fish aggregate together) and can be aided through combined surveys (e.g., using trawl or camera gear to measure the size or age structure of the population being sampled via eDNA).

Ultimately, operationalising eDNA surveys will require increased investment in large-scale studies that match the unit of inference for assessment and management across population or management units, or large marine ecosystems. Broadscale studies are more applicable to fisheries management, which will improve the likelihood of eDNA surveys being adopted for decision-making. Furthermore, the ability to derive quantitative information from eDNA may be more robust at broader spatiotemporal scales because potential sampling biases (e.g., dispersal

of genetic material and likelihood of detection) and associated uncertainties tend to decline as the unit of inference increases. For example, Shelton et al. (2022) demonstrate the development of an eDNA index of abundance for Pacific hake, which integrated eDNA concentrations across nearly 90,000 km², and the 2025 Pacific hake assessment includes this biomass index as a model sensitivity (see Section 4.1.1. Case Study: Pacific hake; Johnson et al. 2025).

# 3.4 | Challenge 4: Collecting Biological Data for eDNA Surveys

As a non-invasive survey tool, eDNA has limited ability to provide direct biological information from the population of inference (e.g., size, age, weight or fecundity measurements), which are important attributes that are needed for many stock assessments (Hansen et al. 2018). However, data and inputs for assessments come from a variety of sources, and an eDNA survey does not necessarily need to provide all of the biological data currently obtained from fishery-independent surveys. For instance, weight and fecundity data are often collected by observers or port agents based on fishery-dependent sampling, given that a large proportion of the world's fisheries do not have dedicated fishery-independent surveys (e.g., most tropical tunas, Baidai et al. 2024). In data-limited cases (i.e., when no indices are available) or when there is only a fishery-dependent relative abundance index available (i.e., catch-per-unit effort, CPUE), eDNA represents a powerful tool for understanding population trends, despite potential uncertainties in the size or age structure of the sampled population. Conversely, for more datamoderate and data-rich species where fishery-independent indices exist, biological samples informing age- or length-structure of the sampled population may be inferred from paired sampling platforms and stations (Guri, Shelton, et al. 2024). Thus, expert knowledge and complementary survey techniques could inform assessment decisions regarding the population selectivity of the eDNA index, akin to assumptions regarding size composition of acoustic surveys (Fall et al. 2024). Ultimately, we envision that for data-moderate and data-rich species, eDNA will be most suitable as a complement to existing surveys by augmenting survey data and filling in data gaps. For example, the non-invasiveness, quick collection, and short processing time for eDNA sampling would allow increased cost-effectiveness of vessel-based surveys by decreasing the spatiotemporal gear footprints, while simultaneously increasing total geographic coverage and sampling density. eDNA sampling could be especially useful for difficult to survey habitat (e.g., untrawlable habitats or nearshore vegetated habitats like kelp forests), areas closed to fishing (e.g., marine protected areas) or areas that restrict traditional survey gear due to marine spatial planning activities (e.g., offshore energy). These attributes make eDNA a compelling option for sampling species or stocks for which a non-invasive method is important (e.g., for threatened or endangered species).

# 3.5 | Challenge 5: Quantifying eDNA Survey Uncertainty

The previous four challenges outline potential drivers of statistical bias in an eDNA index. Each challenge describes potential

causes of disconnect between fish biomass and what is measured (DNA occurrence or concentration). Another important component of an eDNA-derived abundance index is understanding the processes that affect precision and how they may differ from those derived from other survey methods. In many aspects, an eDNA survey is identical to other traditional surveys where samples are collected across the spatial domain of interest in a given season and combined to provide an estimate of the index (e.g., using design- or geostatistical-based methods). Uncertainty in any index estimate derives from the variability among observations and depends upon the estimator used. In general, eDNA presents the potential for investigating variability at a given sampling site unavailable for other methods. Replicate water samples from a single location and time allow for repeated eDNA concentration estimates that inform the observationassociated variability for eDNA. In trawl surveys, this would be equivalent to the variability if the same location was sampled repeatedly at the same time, but only if the trawl did not deplete the number of fish. Understandably, such replicated sampling designs are uncommon in most fisheries surveys, but are relatively simple to implement for eDNA sampling. The benefit of having replication at fine scales is that it becomes possible to separate fine-scale observation variability from larger-scale spatial or temporal variation. This, in turn, may potentially reduce uncertainty around eDNA indices relative to other survey methods (see Shelton et al. 2022), although this topic deserves further research. Furthermore, if other processes that could bias estimates remain unaccounted for; then, the eDNA index that only accounts for variability among observations will underrepresent the uncertainty in the overall index (e.g., if small fish shed DNA at a much higher rate per kg than large fish and the survey does not account for such differences). Again, though, this is not a feature unique to eDNA surveys as similar concerns exist for nearly all surveys (e.g., backscatter strength related to size composition for acoustic surveys).

### 3.6 | Challenge 6: New Survey Scepticism

The integrated assessment framework encourages incorporating new data types, and consequently, there have been entreaties for wider inclusion of novel data sources (Goethel et al. 2023). However, the bar for inclusion in fisheries assessment models or management decision-making is often exceedingly high and ill-defined (Bradley et al. 2019; Shotwell et al. 2023). New survey techniques are further impeded by an unknowable true population size or trend (i.e., comparisons can only be made to similarly uncertain survey techniques; Nakken and Ulltang 1982). Healthy scepticism of new techniques for monitoring fish populations is important, to a degree, to ensure that observations adequately reflect the population unit of inference, and that the technology is vetted for potential bias, error, and uncertainty. Yet, the history of fisheries science is filled with examples where new data collection techniques have been maligned until begrudgingly accepted. An example is the use of hydroacoustic surveys to monitor fish abundance and biomass, which were initially dismissed for similar justifications as eDNA surveys (e.g., uncertainty in detection, lack of biological samples, and limited understanding of methods by non-experts), but are now widely utilised to survey fish abundance (see Coombs and Cordue 1995; Nakken and Ulltang 1982, for review). Although acoustic

surveys are now widely used in stock assessments globally (e.g., De Robertis et al. 2021; Johnson et al. 2025), the process of refining and improving associated data outputs is continual (Fall et al. 2024; Wanzenböck et al. 2003).

The issues facing eDNA surveys are not new and, instead, represent a historical scepticism for new fish population monitoring methods. We argue that eDNA surveys have progressed to the point that primary uncertainties can be overcome and quantified. Further cross-survey comparisons with broadscale trawl, acoustic, video, and fishery-dependent (i.e., CPUE) indices of abundance (Salter et al. 2019; Sato et al. 2021) should lend further credence to eDNA surveys being a reliable indicator of population trajectories. Although uncertainties will remain and eDNA will have its own biases in enumerating absolute abundance, eDNA indices with similar uncertainty as existing fisheries monitoring approaches (e.g., acoustic or video surveys) are obtainable and can be easily integrated into stock assessment models (Johnson et al. 2025). Much like acoustic survey methods, we expect eDNA approaches to be continually refined and improved in the coming decades as they are more widely adopted for broadscale fisheries monitoring.

### 4 | Using eDNA in a Stock Assessment

Similar to other survey methodologies, there are many potential avenues for including eDNA information in a stock assessment. We outline the general path for adding an eDNA index into an assessment to inform abundance or biomass trends (Figure 1), emphasising how the data available to perform an assessment might influence the design of an eDNA survey. The recent assessment of Pacific hake (Johnson et al. 2025) is summarised as one of the first applied examples of integrating an eDNA index directly into a data-rich stock assessment. As a practical guide for eDNA practitioners, we provide a conceptual flowchart highlighting key eDNA survey design facets to consider depending on the context in which the resulting index will be utilised for assessment (Figure 2). We emphasise that it is necessary to communicate among geneticists, stock assessors, survey technicians and managers to ensure that the eDNA survey will meet the needs and criteria for integration into assessment frameworks, and will be properly funded and maintained for the foreseeable

Use of an eDNA index hinges on the assumption that the DNA concentration in the water is proportional to the stock measure of interest, typically biomass. In the following, we assume that the necessary statistical design work, eDNA sampling, and laboratory processing have been completed, and that the samples have been determined to be sufficiently representative of the population to warrant inclusion in a stock assessment. The main decision point is then to either (a) treat the eDNA survey as an independent data source that informs biomass and use that as an input in the assessment or (b) combine the eDNA information with other sources of data that inform biomass to provide a combined or joint index that is then used as an input to the assessment. Both paths are reasonable depending on the situation, with the decision being driven by existing data availability as well as funding constraints (e.g., is a synoptic eDNA survey that spans an entire management unit feasible?).

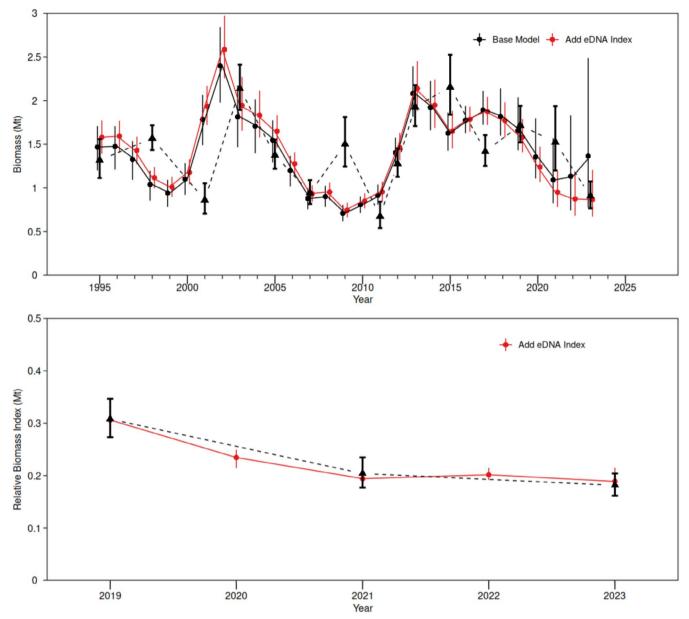
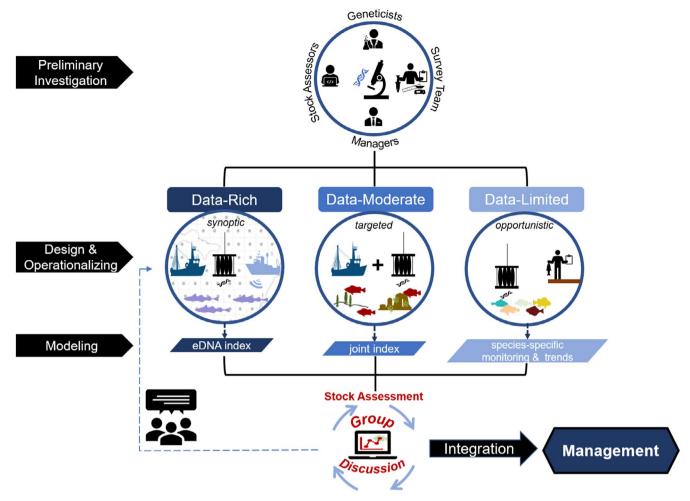


FIGURE 1 | Comparison of alternative Pacific hake stock assessment model fits to the acoustic-trawl fishery-independent survey index (top panel) and the eDNA survey index (bottom panel). In the top panel, the median predicted estimates of the acoustic-trawl survey biomass from the 2025 model used for management ('Base Model') are given by the black solid line and points, whereas the same predictions for the eDNA sensitivity model ('Add eDNA Index') that simultaneously fit the eDNA index are provided by the red solid lines and points. Model fits are relative to the observed acoustic-trawl survey data points, which are given by the black dashed lines and triangles. In the bottom panel, the sensitivity model fit (red solid lines and points) to the observed eDNA index (black dashed lines and triangles) spans the three sampling years (2019, 2021 and 2023). The vertical bars represent the 95% credible intervals for predictions and 95% confidence intervals for observations.

# **4.1** | Data-Rich: Integrating eDNA Indices as Standalone Surveys

For most high-value marine species, there are typically multiple data inputs for an assessment, where various indices of relative abundance (e.g., from survey or fishery CPUE) are often integrated simultaneously as independent data sources (Maunder and Punt 2013). To be considered for inclusion in an assessment, an index of abundance should represent the dynamics of the entire unit being assessed (i.e., the stock), have a relatively consistent methodology over time, provide a reasonably long time series (> 4 years) to assess trends in population

dynamics, and include a measure of interannual variability or uncertainty in the index. Within an assessment, indices of abundance are paired with auxiliary data or expert knowledge to inform stock assessment modelling assumptions related to what portion of the stock interacts with the gear (i.e., availability), relative scaling of the index to the entire population (i.e., catchability), and the size or age composition of the fish being observed by the gear (i.e., selectivity; Table 1). As with all survey methods, eDNA will have imperfect information to inform these parameters. Careful consideration of the experimental sampling design can alleviate many of these concerns and provide guidance to stock assessors on realistic model



**FIGURE 2** | Roadmap highlighting how eDNA can be more widely integrated into fisheries assessment and management frameworks. See Table 2 for text description of the steps depicted here, the participants at each stage of the process, and the different requirements for data-rich, data-moderate, and data-limited species assessments. Data-rich assessments could also use eDNA data in a similar way to either of the more data-sparse approaches.

assumptions. Empirical studies pairing eDNA sampling with other methods as well as studies about dispersion, transport, and decay, will help identify appropriate scaling coefficients (e.g., catchability) for parametrising within an assessment model. Similarly, paired sampling, for both validation studies and ongoing data collection, will provide information about the size composition of the fish being sampled and associated eDNA 'selectivity'. For eDNA surveys, selectivity represents the general size or age classes assumed to be observed by eDNA samples, which, as eDNA is a nearly ubiquitous sampling method, will likely be primarily influenced by the interplay of sample locations and spatiotemporal ontogenetic species distributions (see the next section for selectivity assumptions used in the Pacific hake case study). As with many hydroacoustic surveys, it is likely that expert judgement will be necessary to make case-specific assumptions about the selectivity of eDNA (e.g., sampling of fish greater than a certain age).

When a synoptic eDNA survey with a rigorous experimental design is available, then direct integration into an assessment as a standalone index of abundance is the most likely path. Aside from an assumption about survey selectivity as noted above, the assessment will need to estimate the catchability to

scale the eDNA index to absolute abundance. Calibration experiments with other gear types along with expert judgement will likely be necessary to place priors on the catchability term to ensure model tractability and consistency with existing survey population scaling. In more data-limited situations or when no other surveys are available to help anchor total population abundance or biomass, expert judgement and relative scaling with total catch will be particularly important to determine eDNA index scaling factors (i.e., catchability). Ultimately, stock assessors will need to make assumptions regarding relative data weights in the model, which will influence the amount of emphasis the model places on fitting to each data source where a higher weight ensures the model more closely reflects the scale and trend of a given data source. Depending on model structure, final data weights will be a mix of data uncertainty, expert judgement and iterative tuning of the input weights (Punt 2017). Although parametrising the observation model for an eDNA survey and determining appropriate data weights is complex, the general approach is common across survey types and should not be seen as a barrier for inclusion of eDNA in stock assessments. The following case study with Pacific hake demonstrates the process of how a synoptic, three-year eDNA survey was considered for and integrated into the stock assessment.

**TABLE 2** | Roadmap for developing and integrating an eDNA index into stock assessments with specific recommendations on critical considerations during each stage of eDNA collection and index development.

Stage	Participants	Primary issue to address	Recommendations
Identify Need	Geneticists, Stock Assessors	Communicate across disciplines to determine if and how an eDNA index could be used for assessment	Discuss assessment data tier (rich, moderate, limited) and needs to determine how an eDNA survey might be best utilised Convey uncertainties and potential eDNA survey limitations to identify whether an eDNA survey is likely to fill data needs Identify modelling skill sets among participants and whether geneticists or stock assessors have bandwidth for syntheses of eDNA data
Scoping	Geneticists	Ensure DNA reference sequences available for the study species of interest	Identify all species (if multispecies survey) for which eDNA indices will be developed Check reference databases for species encountered by the survey, geographic representation and sequence specificity Develop species-specific primer sets or qPCR assays and reference sequences, if necessary
Validation	Geneticists, Survey Technicians	Implement pilot study and perform associated lab analyses to understand detection probability and help identify potential uncertainties	Identify clear survey protocols to avoid false positives (e.g., sample before trawling or use in situ sampling technology)  Identify data analysis approaches to avoid false negatives (e.g., what concentration thresholds are needed, how much water to filter per station)  Consider pairing with another survey method to develop baseline demonstrating eDNA concentrations adequately reflect estimates from other approaches
Resource Acquisition	Geneticists, Managers, Survey Technicians	Identify costs for expanding survey to meet assessment needs (spatiotemporal resolution) and for making it production level (i.e., providing yearly data outputs and maintaining a long-term time series)	Use pilot study as a baseline for identifying costs for implementing broadscale survey Convey need to decision-makers to aid resource acquisition and permanent funding for eDNA surveys Identify synergies with existing surveys, if any, to reduce costs (paired surveys from single vessel) and link eDNA with biological sampling Determine division of labour (survey collection vs. data analysis) and associated resources (personnel) needed to ensure feasibility of production-level outputs
Survey Design	Geneticists, Survey Technicians, Stock Assessors	Based on the intended assessment use, identify appropriate spatiotemporal coverage of eDNA sampling and potential synergies among existing sampling methods	Determine the spatial extent and temporal frequency of sampling along with the distance between sampling stations (i.e., develop a survey grid that covers the entire population of inference, where feasible and as needed)  Implement a specific survey experimental design (e.g., fixed or random stratified) to choose sampling locations, noting that opportunistic or localised surveys are of little value to region-wide assessments  Identify how the eDNA samples will be obtained (i.e., dedicated vessel vs. collected as part of existing survey)  Pair with other survey gear, when feasible, to allow collection of size and age composition (e.g., trawl or video gear) or to inform other survey methods (e.g., species identification for acoustic surveys)  Consider calibration studies for initial survey years to understand relative abundance scaling differences across survey methods and to inform catchability parameter assessments in stock assessments

(Continues)

Stage	Participants	Primary issue to address	Recommendations
Sampling Design	Geneticists, Survey Technicians, Stock Assessors	Develop a stringent and consistent experimental sampling design	Ensure that consistent gear is used and sampling methods are well-defined (e.g., volume of water collected, type of filtration, etc.)  Determine an appropriate sampling depth (e.g., with groundfish species water sampling should be within 10-20 m off bottom) and potentially sample multiple depths, if possible  Identify appropriate technical analyses of the data to ensure consistent year-to-year estimates of relative abundance or biomass from eDNA concentrations, while eliminating potential biases (false negatives, false positives, detection issues) to the extent practicable  Calculate appropriate estimates of uncertainty in the resulting index  Identify methods to ensure appropriate assumptions about the size or age structure (e.g., all life history stages or only mature fish if aggregated by maturity-status) of the population being surveyed can be made in the assessment
Operationalising	Geneticists	Plan for the future and ensure appropriate personnel to provide consistent eDNA survey data products at the time intervals needed for assessment	Account for resource needs to staff survey vessels and analyse data, while also recognising potential time crunches associated with annual assessments (i.e., annual data collection must also account for subsequent analysis, QA/QC, index development and uncertainty calculations, which must be accomplished within <1 year time lines to support assessments)  Allow for periodic technical advancements and improved analyses to be integrated to improve uncertainty Recognise that small scale, one time surveys (e.g., pilot studies) are of limited value in an assessment or management context, so any survey must be maintained (and funded) for the long-term to be effective
Modelling	Stock Assessors, Geneticists	Develop the eDNA index and then carefully test the integration of the eDNA index within the stock assessment model Develop a framework for updating and maintaining the time series, if deemed suitable for integration	Determine appropriate assumptions regarding the size/age structure of the population being surveyed by eDNA (i.e., population selectivity), likely informed by paired survey methods Identify priors for catchability, potentially informed by calibration experiments  Explore uncertainty assumptions (i.e., appropriate probability distributions) and data weighting (i.e., relative uncertainty among data sets) when integrating the new eDNA survey Determine model sensitivity to inclusion of the new survey through leave one out analysis with existing surveys or other data inputs Implement sensitivity runs to each of the above assumptions using plausible alternatives to identify influence of the eDNA survey on model outputs When joint indices are developed (data-moderate cases), explore specific sensitivity in the final index to how the data are combined  For data-limited cases, compare various data-limited models with/without the use of the eDNA index to ensure management advice is robust

### 4.1.1 | Case Study: Pacific Hake

Pacific hake support the largest fishery by volume on the US West Coast (excluding Alaska) and are jointly managed by the US and Canada under the Pacific Hake Treaty (Agreement Between The Government of The United States of America and The Government of Canada on Pacific Hake/Whiting 2003). Annual stock assessments include long time series for catch from multiple fleets, age structure, weight-at-age, and a fisheries-independent index of relative abundance. The index estimates relative biomass from a biennial acoustic-trawl survey that began in 1995 and is currently conducted in oddnumbered years (Clemons et al. 2024; Johnson et al. 2025). Since 2019, a large-scale eDNA survey has been conducted alongside the acoustic-trawl survey, where samples were taken from the same vessel and at each station (see Shelton et al. 2022). As part of the 2025 hake assessment, an eDNA biomass index derived from the three survey years (2019, 2021 and 2023) was developed and then used within an assessment model sensitivity evaluation to determine the impact of including the eDNA index as an independent biomass index (identified as the 'eDNA index' sensitivity; Johnson et al. 2025). To fit the eDNA index in the assessment, it was assumed hake ages 1 and older were fully selected by eDNA sampling 'gear'. This assumption was based on expert judgement regarding the influence of ontogenetic hake distribution patterns on the presence of younger fish in the core survey area, uncertainty regarding the location of spawning grounds and the influence of variable oceanographic conditions on hake larvae drift and survival (Vestfals et al. 2023). Broadly, the eDNA index suggested a similar trend in biomass from 2019 to 2023 as the acoustic-trawl survey (Figure 1). The addition of the eDNA index in the assessment sensitivity run did not substantially affect biological parameter estimates, as would be expected for a survey with only three data points, but it did affect estimates of stock biomass in recent years. Overall, biomass estimates decreased when the eDNA index was included, which reflects agreement in recent time trends among the two surveys. Thus, in the sensitivity run, the consistent signals across survey data streams led to survey data being implicitly upweighted over other types of data (e.g., age compositions) compared to when fitting only the acoustic-trawl index in the operational assessment model used for management. The low levels of uncertainty in both the eDNA and acoustic survey indices also led to further upweighting of these data sources (see Johnson et al. 2025 for more details). Although the eDNA index was not integrated into the operational assessment model, it was straightforward to include the index as a sensitivity evaluation. Overall, the eDNA index will continue to be considered for inclusion in upcoming stock assessments, particularly as the index time series lengthens and estimates of uncertainty are further evaluated.

## **4.2** | Data-Moderate: Developing and Integrating Joint Indices

Data-moderate assessments typically depend on fishery-independent surveys similar to data-rich assessments. However, a common pitfall for species of moderate economic value is that dedicated, single-species surveys cannot be implemented due to

funding or other constraints. Oftentimes, multispecies surveys are used to provide indices of abundance for an array of species simultaneously, but these cannot be optimised for sampling of each species. Thus, survey design becomes a trade-off between sampling the primary habitat and distributional range of the species deemed most 'important', with the consequence of increasing the uncertainty and variability for species that are not within this range and habitat. For example, in the Gulf of Alaska a fishery-independent trawl survey provides the primary index of abundance used in assessment for an array of species with varying life histories and habitat preferences, including many rockfish species that are found in both survey trawlable and 'untrawlable' habitat (Conrath et al. 2019). Thus, estimated biomass from the trawl survey may be biased or inaccurate due the survey not being synoptic for certain species. In such instances, eDNA surveys could provide a cost-effective platform to sample at a broader spatial scale and across more diverse habitats, especially those which are not conducive to traditional sampling methods.

Data from both eDNA and traditional surveys can be integrated in a single modelling framework to provide a more comprehensive understanding of a species' distribution and produce a joint index. Joint integrated indices are typically generated by using a spatially explicit model that can link data from multiple sources into location-specific estimated densities summarised across a larger spatial scale (Grüss and Thorson 2019, 2024; Guri, Shelton, et al. 2024). Synthesising data from multiple sources helps smooth and predict density in locations that would otherwise have little-to-no data. Disparate input data can be integrated as different data types (e.g., biomass, abundance, encounter or eDNA concentration). More data sources increase the sample size and spatial coverage, which theoretically decreases the estimated uncertainty across large spatial scales. Using this joint-model approach, Guri, Shelton, et al. (2024) demonstrated that coupled trawl and eDNA survey data can produce robust, estimated fish densities and has the potential to allow for greater flexibility when data are missing. Thus, for data-moderate assessments, eDNA could be used to improve survey cost-effectiveness by reducing the number of trawl survey stations or increasing the survey footprint (e.g., expanding eDNA-only sampling stations). eDNA could then be integrated into a joint index that is used in the same way as a standalone eDNA index (as outlined for the data-rich assessment case).

# 4.3 | Data-Limited: eDNA Surveys as the Basis of Assessment

For many low-value or bycatch only species, minimal data are often available, which necessitates applying data-limited methods (DLMs), such as simplified, typically trend-based assessment approaches (Cope et al. 2023). Data-limited species may depend exclusively on fishery-dependent data or surveys that only partially or ineffectively sample the population. Likewise, data for these species may be collected at a higher taxonomic level (i.e., aggregating species either by intention due to limited resources or due to high rates of misidentification). An eDNA index could provide a primary or sole data source to assess and monitor population trends for data-limited species. Given

resource constraints for most data-limited species, eDNA indices for these species would likely need to be developed from multispecies eDNA surveys or opportunistically from eDNA sampling initially targeting other focal species (e.g., in water samples from other directed eDNA surveys). Resulting indices are likely to incorporate more uncertainty for informing population trends because the entire spatial distribution for the non-target, opportunistically sampled species (i.e., the datalimited species for which the eDNA index is being developed) is unlikely to be synoptically sampled. In cases where eDNA is the first survey for a species, resultant indices can be used directly as the basis of an index-based DLM approach. Although long-term eDNA sampling would greatly benefit monitoring of data-limited species, even short-term and opportunistic eDNA surveys for these species can fill data gaps. For instance, snapshots from eDNA sampling can indicate range shifts, expansion, or contraction, which can be used as a population status indicator (Dowling et al. 2015). As with DLMs using other data sources (e.g., index-based or catch only), eDNA surveys, regardless of time series length, could form the basis of an empirical harvest control rule (HCR) for calculating appropriate catch levels (Goethel et al. 2023).

For extremely data-limited species that are often managed as a species 'complex', species identification is perhaps the greatest benefit of eDNA data. For instance, many species are often misidentified, cryptic (i.e., morphologically indistinguishable), or assigned to mixed-species groups due to lack of resources (Omori et al. 2021). With the ability to identify taxonomic groups to species, scientists and managers can improve monitoring of individual data-limited species as well as track shifts in species composition. Moreover, eDNA could improve species identification for acoustic surveys, a major source of uncertainty for interpreting acoustic backscatter, which further demonstrates the benefits of synergistically implementing multiple survey types. Thus, eDNA sampling provides a unique opportunity to develop scientifically informed, species-specific management advice based on DLMs for species that do not typically have the data necessary to monitor trends or provide any catch advice.

### 5 | Looking Forward: A Roadmap for Developing eDNA Surveys for Use in Marine Fisheries Assessments

Given the high standards for integrating new data into an assessment, an eDNA survey must be carefully designed and implemented to optimise long-term data collection and usability. Here, we outline an idealised roadmap for how eDNA studies should progress when the objective is to develop an index of relative biomass to be used within a stock assessment (Table 2; Figure 2). The approach should be iterative and can be opportunistic. Thus, the roadmap should be used as a loose guide to help avert common pitfalls (e.g., misaligning the scale of a survey to the needs of an assessment), while understanding that steps may not progress linearly, and feedback within and across stages is imperative to refining and improving any potential eDNA survey. Moreover, before any eDNA survey is considered, we recommend that interdisciplinary research teams, including geneticists, ecological modellers, stock assessors, survey technicians, and fisheries managers, be convened to ensure the survey

fills a research or data need while also being collected in a way that can be explicitly used for management. Input and feedback from all groups will be vital to ensure the survey is both logistically feasible and scientifically useful.

Early stages of development for an eDNA survey should begin with identification of the need or interest from all partners, as well as scoping and validation to ensure a study is feasible for the species. Given the hesitancy toward adopting new survey methods, we recommend that eDNA pilot studies be paired with other survey types (e.g., trawl surveys) to allow direct comparison of relative biomass estimates among methods, enable calibration (e.g., identifying appropriate assessment scaling and catchability parameters) and inform size structure of the surveyed population. Once the eDNA survey method has been demonstrated to be feasible, careful communication across disciplines should be undertaken to verify the need for the survey and how it might be used in assessment (e.g., depending on data needs for the study species; see Figure 2). Early in the process of scaling the pilot study up to the spatiotemporal resolution needed to inform an assessment, consideration should be given to how the survey will be funded in the long term. Assessments require adequate time series for all data sources because current population sizes as well as trends over time must be estimated, so short-term and small-scale studies are of limited value and unlikely to be considered for assessment inclusion.

Similarly, the survey and sampling design need to be developed a priori to implementation to ensure adequate resolution and technical analyses, while also maintaining consistency in methods and sampling across years. During the design phases, emphasis should be placed on sampling frequency and design (i.e., consistent stations vs. a random stratified approach), how to address uncertainties (e.g., false positives or negatives) and how uncertainty will be estimated. Moreover, the survey logistics should be considered in terms of whether samples will be taken asynchronously, autonomously (Preston et al. 2024), or paired with existing platforms (e.g., trawl surveys), and whether sampling can work synergistically with other survey methods to expand knowledge of the surveyed population (i.e., using paired trawls or video surveys to collect size or age structure information). Once a stringent experimental design has been implemented and the feasibility has been demonstrated, the final stages involve the intertwined steps of operationalising the survey and integrating the resulting index into the assessment model. From experience (see Section 4.1.1. 'Case Study: Pacific Hake'), there will be considerable feedback and iteration among these steps. For instance, funding to operationalise any survey typically requires demonstrated usefulness for a stock assessment application, yet to prove utility an assessment needs to have an established time series of data. As such, it may be necessary to perform a series of eDNA pilot studies to develop a relative biomass time series, and then geneticists should work with stock assessors to integrate the index into the assessment as a sensitivity run to illustrate the validity and usefulness of the eDNA data. Support from stock assessors and commitment to using the eDNA survey will help aid geneticists in lobbying for funds to operationalise the survey. Likewise, geneticists must be prepared to provide production-level eDNA survey products consistently on the typically annual cycle of assessments, including collection, analysis, QA/QC and calculating the index

with associated uncertainty. Although the process of getting an eDNA index integrated into an assessment is unlikely to be as straightforward as described here, we believe this roadmap can provide useful guidance for navigating the process.

### 6 | Conclusions

Based on our review, we suggest that the major technical and knowledge challenges limiting greater adoption of eDNA in marine fisheries stock assessments have been overcome by improvements in describing the quantitative and spatial characteristics of eDNA, accepted best practices for experimental design and data analysis, rigorous evaluations of sources of uncertainty in eDNA surveys, and because eDNA surveys can be readily paired with complementary data sources (trawl, acoustics, and video). eDNA methods are subject to the same new survey scepticism experienced by acoustic surveys and may follow a similar trajectory to acceptance. Perhaps most persuasively, the case study with Pacific hake shows that an index of biomass based on eDNA can be used within existing model frameworks and evaluated for inclusion in a data-rich stock assessment.

As with any developing scientific discipline, eDNA techniques continue to improve, which will likely lead to more robust eDNA methodologies. Areas of eDNA research that could be of great value to future stock assessments include epigenetic ageing to determine biological age from genetic samples (Balard et al. 2024) and eRNA to determine life stage (Parsley and Goldberg 2024), thereby reducing the need for simultaneous biological collections. Additionally, eDNA studies are beginning to apply population genetic methods that could be used to derive stock structure information (Parsons et al. 2018), presenting an opportunity to bypass traditional tissue-based population genetic methods. Although technical challenges currently limit the application of these novel approaches, rapid progress in the eDNA field suggests that integration of additional 'omics data should be part of future discussions between fisheries managers, stock assessors, and geneticists.

One of the primary limitations to using eDNA in fisheries assessments is the lack of coupling between interdisciplinary research teams, dedicated funding for large-scale eDNA surveys at the relevant scales, and mature analysis methods and pipelines for rapidly producing abundance indices for annual assessment timelines. Existing interdisciplinary collaborations provide some lessons for teams with the goal of data production for stock assessment models, but may be limited in access to funding, analytic capacity or sampling at large spatial scales. Once these hurdles are overcome, we suggest that the best applications for eDNA in assessments are in tandem with other survey methods, which, when paired, could lead to improved geostatistical models and indices. eDNA is particularly suitable for integration and synergy with other emerging technologies, including autonomous platforms. If paired with video, cameras could inform the size distribution of the population sampled by eDNA, while paired with acoustic surveys, eDNA could provide species identity for schooling species. Synergistic paired surveys can demonstrate the combined utility of data from eDNA in tandem with other technologies, and then could eventually be used to decrease survey costs while also increasing the geographic

and habitat coverage. With fewer resources dedicated to monitoring marine fish populations, eDNA surveys present a cost-effective sampling tool that can provide quality data across the array of data availability scenarios encountered when implementing fisheries stock assessments. We envision that eDNA will gain traction in the coming years, especially given increasing resource constraints for monitoring and assessing fish populations, yet increasing demands to provide quantitative, scientifically informed advice for an increasing number of marine species. Thus, increased collaboration among geneticists and stock assessors will be imperative to ensure cost-effective data on trends in fish populations.

### **Author Contributions**

D.S.B., K.L.O. and D.R.G. conceived of the paper. D.S.B., K.L.O., D.R.G., A.O.S. and A.M.B. wrote the original draft. All authors reviewed and edited the manuscript.

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### **Conflicts of Interest**

The authors declare no conflicts of interest.

### **Data Availability Statement**

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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