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Review Article

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Utilisation of Environmental DNA for Conservation Biology and Ecological Assessment: A Mini Review

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Abstract
Environmental DNA (eDNA) is constantly released DNA into species surroundings as they interact with the environment. eDNA has the potential to revolutionise biodiversity science and conservation efforts by making it possible to count species globally and almost instantly. eDNA investigations can becommunity-based or species-specific and can be done on terrestrial as well as aquatic ecosystems. Understanding the ecology of eDNA is important for conservation based research applications utilising eDNA. Population characteristics like abundance, dispersion, and biomass along with population genetics studies which includes studying haplotype ratios, effective population size and population structure can be performed taking eDNA. The current review summarises the types of eDNA studies and their applications in improving biodiversity and ecological understanding as well as in population genetics along with the challenges faced during eDNA analysis.

Keywords: environmental DNA, ecological, conservation, biodiversity, population

Introduction

Recent rapid developments in non-invasive genetics, which examines genetic material inside traces of creatures rather than full animals, such as hair, faeces, and other shed biological components, have benefited conservation and ecological understanding. Environmental DNA (eDNA) discovery and application have recently attracted a flurry of scholarly attention in the field of molecular ecology, which applies genetic techniques to ecological issues.It specifically refers to the examination of genetic material obtained from bulk environmental samples such as soil, water, or air rather than through targeted techniques like setting up fur traps or gathering fresh scats (Barnes and Turner, 2016).Species constantly release DNA into their surroundings as they interact with the environment. For higher organisms, this DNA can originate from expelled cells or tissue like urine, faeces, hair, and skin, as well as explicitly from deceased individuals releasing genetic material.It may be possible to allocate specific individual's genetic information via direct organismal traces. eDNA can be used to infer information about an organism's abundance, population size, density, nutrition, and sex from these non-invasive samples. Estimates of gene flow, evolutionary relevant units, genetic diversity (such as inbreeding within a population), and mating systems (such as multiple paternity) have also been obtained. Environmental DNA research (eDNA) has the potential to revolutionise biodiversity science and conservation efforts by making it possible to count species globally and almost instantly. Utilisation of eDNA samples from soil, water, or air is advantageous, but, when it is difficult to identify and assess individual traces. Before doing intense, invasive sampling in difficult-to-sample environment, for instance, eDNA can be implemented to pinpoint sites of suspected occupancy. Combining conventional methods with eDNAmetabarcoding of environmental materials may show greater biodiversity than either approach by itself (Adams *et al.,*

2019).Although eDNA applications seem intriguing, finding eDNA in freshwater and marine environments will require well-defined hypotheses and appropriate experimental design, both in the lab and in the field, that takes into account the habitat, life history, and behaviour of the target species for accurate detection of eukaryotes using eDNA in freshwater and marine ecosystems(Diaz-Ferguson and Moyer, 2014).While eDNA has undergone significant experimentation as a tool for monitoring biodiversity and biosecurity with a strong taxonomic focus, its potential as a source of population genetic data has not yet been adequately investigated.

Types of eDNA studies

The two primary categories of eDNA investigations are semi-targeted (community-based) and targeted (species-specific). Although both categories are frequently discussed at the same time, their approaches, interpretations, and accuracy are very different. Studies that focus on individual DNA fragments in an environmental sample employ assays designed for that species. Studies may use a variety of tests on a single environmental sample or concentrate on a particular species (Seymour, 2019).Community-based eDNA investigations aim to link all readily accessible DNA strands in an environment to their species of origin. Since most genomic databases are still insufficient for any specific investigation, accurate identification of sequences to species frequently depends on course taxonomic assignments. In species-specific eDNA studies, standard PCR is still employed to determine the presence or absence of the targeted DNA, as in traditional eDNA investigations, while quantitative PCR or digital PCR chemistry is becoming increasingly common for quantification while for community-based approaches, wide range of high-throughput sequencing technologies, includingmetabarcoding, long-read, shot-gun mitogenomics, genome skimming, etc. are utilised.eDNAmetabarcoding primer sets have been employed to simultaneously identify numerous haplotypes across different species including arthropods (Elbrecht*et al.,* 2018).

Environmental DNA investigations in aquatic ecosystem

Ecosystems in both freshwater and marine environments are significant eDNA reservoirs. The pace at which DNA is released and destroyed by biotic and abiotic processes, as well as the abundance of the target species, are both connected to the detection of eDNA. The distribution of eDNA in freshwater systems is often homogeneous while in marine ecosystems, the high density of marine life makeseDNA more likely to dilute and disperse due to the large amount of sea water compared to biomass. Although less accurate than in freshwater systems, eDNA can nevertheless be detected in marine environments with more difficulty as compared to freshwater systems due to the dynamic nature of marine settings and the higher inhibition of subsequent molecular procedures caused by the high salinity conditions (Diaz-Ferguson and Moyer, 2014).Additionally, tropical marine and freshwater ecosystems have high surface temperatures (often above 30°C) and significant UV radiation at sea level, which may accelerate eDNA breakdown and shorten its stay in the water, lowering the likelihood that it will be detected over time and space. Because of this, prior to the start of any field detection protocol, the appropriate detection method, laboratory and aquarium eDNA assays taking into account species size, vertical distribution, current or water flow, life history of the target taxa, and degradation rates among different environments, are required(Kelly *et al.,* 2013).

Exploring ecology of eDNA for conservation

The swift progress of eDNA-based conservation applications and potential research contributions are intriguing, however, capturing and identifying eDNA as well as interpreting the results of these efforts will benefit from an improved knowledge of ecology of eDNAwhich includes the origin, state, transport, and fate of eDNA molecules.Finding answer to originofeDNA puts emphasis on the physiological sources of eDNA productionwhich includes both intracellular and extracellular forms.It appears likely that multicellular organisms shed DNA into the environment in two stages, first, as sloughed tissues (extracellular form) and later, when those cells degrade (intracellular form).Increasing our understanding of the connection between organism size, age, and/or biological activity and eDNA generation can help us collect eDNA more effectively. In

order to select sampling windows where genetic materials accumulate, conservation efforts using eDNA may be able to maximise effectiveness by taking into consideration temporal (such as seasonal events like mating or die-offs) and geographical (such as diel or other cyclic migrations) patterns (Barnes and Turner, 2016).Aquatic invasive species (AIS) detection, biodiversity and community assessment, population dynamics, ecosystem wellness, trophic relationships dietary experiments, and species historical trends of distribution are just a few of many uses of eDNA that are available for use in the fields of marine ecology and conservation biology.

Population Genetics and Environmental DNA

The identification and measurement of eDNA can serve as a benchmark or an auxiliary indicator of population characteristics like abundance, dispersion, and biomass. Freshwater systems' biomass and organism distributions have been linked to eDNA concentrations(Takahara*et al.,* 2012, 2013). Thus, in amphibians, fish, and reptile species, the concentration of eDNA has been utilised as a proxy for population dispersal. The link between eDNA concentration and species distribution, abundance, and biomass has not been studied in marine species. However, during blooming events, the concentration of DNA in coastal waters has been associated with the geographical and temporal oscillations of bacterial and phytoplankton groups.It is necessary to collect genetic samples from relevant creatures for population genetic studies. However, tissue sample has detrimental repercussions on the intended organism, such as deadly sampling, discomfort or injury, deformity, or stress (Adams *et al.,* 2019), therefore, in recent years, non-invasive sampling techniques have been introduced to lessen this suffering.

Numerous studies have examined population structure (Taguchi *et al.,* 2015), effective population size (Castro *et al.,* 2007)and haplotype ratios (Elbrecht*et al.,* 2018)using markers found in the mitochondrial DNA. However, because mitochondrial DNA (mtDNA), at least in higher animals, very infrequently recombines, the mitochondrial genome only represents a single evolutionarily separate locus for such investigations. Because mitochondria are typically only passed down through the maternal lineage, their effective population size is typically smaller than that of the nuclear genome in a given population, which can result in genetic differentiation patterns in mtDNA and nuclear genotypes that differ from one another (Sigsgaard*et al.,* 2020).

eDNA has been utilised to discover hitherto unrecognised intraspecific genetic diversity (Parsons *et al.,* 2018) in addition to assignment to known haplotypes.Research using seawater eDNA on killer whales(*Orcinus orca*) in the Northeast Pacific examined whether eDNA-obtained haplotypes could be attributed to known haplotypic variation and how long genetic material can be discovered after known target animal presence (Baker *et al.,* 2018).Assigning sequences to specific people may be one of the largest challenges in the move from species detection to population genetics utilising eDNA. Individuals cannot yet be distinguished from one another using environmental samples. Several individuals may have the same haplotype, depending on the sample, but it can also signify both (Adams *et al.,* 2019). Since population genetic theory is concerned with the existence and evolution of genetic variation within individuals within and between populations, confidence in individually-sorted genetic data, however collected, is crucial for downstream analysis.

Challenges faced during eDNA analysis and their addressal

While eDNA may provide a variety of useful applications in population genetic research, these methods also have associated challenges applicable to both genome-wide and single-marker techniques, as well as to mtDNA and nuDNA including PCR and/or sequencing mistakes resulting in false-positive haplotype detections, allelic dropout (Adams *et al.,* 2019) caused by DNA fragmentation or low DNA abundance (Alberdi*et al.,* 2018), biased PCR amplification, or poor capture efficiency, unknown number of individuals contributing to the eDNA pool and the challenge of linking sequences from various markers to specific individuals(Sigsgaard *et al.,* 2020).

For accurate, repeatable eDNA capture and amplification in eDNA population genetic investigations, testing procedures and the use of numerous markers become crucial (Adams *et*

al., 2019).Understanding changes in allelic frequency between and within populations requires relating allelic abundance to eDNA genetic variation (Vasselon*et al.,* 2018). Answering population genetic problems would also benefit from increasing the length of sequences and diversity of marker types that can be produced from eDNA (Adams *et al.,* 2019). Potential copy number will be maximised by selecting the best substrate for sampling a target organism based on ecological knowledge. Given that method choice affects the genotypes acquired, it is crucial to carefully examine the extraction methods, assay, sequencing, and bioinformatic methodologies suited for the biological issue sought.

Conclusion

Environmental DNA will only serve as a tool for biodiversity monitoring, providing quick and accurate insights on species distribution, estimates of abundance, and perhaps population sizes—all of which serve as the foundation for effective conservation measures. Due to its complexity and continued need for political will, commitment, and action, it will never directly address the biodiversity catastrophe. Future research should prioritise figuring out the minimal level of sample required to provide consistent results and describing the conditions in which eDNA analysis is preferable to other methods. But eDNA-based population genetic approaches present an alluring route for improved monitoring and biological research, at least for secretive, endangered, and commercially significant species.Insights into the population genetics of threatened and difficult-to-sample species worldwide will be rapidly and widely provided by eDNA technology.

Conflict of interest

The authors declare no conflict of interest.

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