

Applications of Environmental DNA (eDNA) in Wildlife Monitoring and Biodiversity Conservation

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Abstract

Environmental DNA (eDNA) has emerged as a transformative molecular tool for biodiversity assessment, wildlife monitoring, and ecosystem conservation. Unlike conventional survey methods that rely on direct observation, trapping, or specimen collection, eDNA enables the detection of organisms through genetic material naturally released into environmental matrices such as water, soil, sediments, snow, and air. This non-invasive approach has significantly improved the efficiency, sensitivity, and cost-effectiveness of monitoring rare, elusive, endangered, and invasive species while minimizing disturbance to natural habitats. Recent advances in high-throughput sequencing, metabarcoding, quantitative polymerase chain reaction (qPCR), digital PCR (dPCR), and bioinformatics have substantially expanded the applications of eDNA in ecological research and conservation biology. Environmental DNA has become an indispensable tool for species inventory, habitat assessment, population monitoring, community structure analysis, invasive species surveillance, pathogen detection, and evaluation of ecosystem health across aquatic, terrestrial, and aerial ecosystems. Moreover, integration of eDNA with geographic information systems (GIS), remote sensing, artificial intelligence (AI), machine learning (ML), and ecological modelling has enhanced the accuracy and predictive capability of biodiversity assessments. Despite its considerable advantages, several technical challenges remain, including DNA degradation, environmental inhibition, contamination risks, incomplete reference databases, and limitations in quantitative interpretation. Standardization of sampling protocols, laboratory procedures, and bioinformatics pipelines is therefore essential for improving reproducibility and reliability. This review provides a comprehensive overview of the principles, methodologies, analytical approaches, and diverse applications of environmental DNA in wildlife monitoring and biodiversity conservation. Furthermore, recent technological developments, current limitations, future research priorities, and policy implications are critically discussed to highlight the expanding role of eDNA in supporting evidence-based conservation strategies and sustainable biodiversity management under changing global environmental conditions.

Keywords: Environmental DNA, eDNA, Wildlife monitoring, Biodiversity conservation, Metabarcoding, Bioinformatics, Next-generation sequencing, Conservation genetics, Invasive species, Ecosystem monitoring.

1. Introduction

The rapid decline in global biodiversity has become one of the most pressing environmental challenges of the twenty-first century. Accelerated habitat destruction, climate change, pollution, biological invasions, overexploitation of natural resources, and emerging infectious diseases have collectively contributed to unprecedented losses in species diversity and ecosystem integrity. Accurate assessment of biodiversity is fundamental for effective conservation planning, ecological restoration, and sustainable management of natural resources. However, conventional wildlife survey techniques—including direct observation, live trapping, camera trapping, acoustic monitoring, and morphological identification—often require considerable financial investment, specialized expertise, extensive field effort, and prolonged sampling periods. These methods may also underestimate species richness, particularly for rare, cryptic, nocturnal, migratory, or low-density organisms, thereby limiting their effectiveness in comprehensive biodiversity assessments.

Recent advances in molecular ecology have transformed wildlife monitoring through the development of environmental DNA (eDNA)-based approaches. Environmental DNA refers to genetic material released by organisms into their surroundings through biological processes such as skin cell shedding, hair loss, mucus secretion, saliva, feces, urine, blood, reproductive products, and decomposing tissues. This extracellular or intracellular DNA can be recovered from environmental samples including freshwater, marine water, soil, sediment, snow, ice, and even air, allowing the presence of organisms to be inferred without the need for direct capture or visual observation [1]. Consequently, eDNA has become a powerful non-invasive technique for detecting species across diverse ecosystems while minimizing disturbance to wildlife populations.

The widespread adoption of eDNA technology has been facilitated by significant improvements in molecular biology and DNA sequencing technologies.

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Initially, species-specific detection relied primarily on conventional polymerase chain reaction (PCR), which was subsequently enhanced through quantitative PCR (qPCR) for sensitive quantification of target DNA. More recently, digital PCR (dPCR) has enabled highly accurate detection of low-abundance DNA molecules, while high-throughput sequencing (HTS) and next-generation sequencing (NGS) platforms have revolutionized biodiversity assessment through DNA metabarcoding. These technologies permit simultaneous identification of hundreds or thousands of taxa from a single environmental sample, providing unprecedented opportunities for comprehensive ecosystem monitoring. The increasing availability of bioinformatics software and curated genetic reference databases has further improved taxonomic assignment, sequence quality control, and ecological interpretation of complex metabarcoding datasets. Environmental DNA has demonstrated exceptional utility across numerous areas of zoological research and conservation biology. In aquatic ecosystems, eDNA has been successfully employed to detect endangered fish, amphibians, molluscs, crustaceans, marine mammals, and aquatic reptiles. In terrestrial habitats, soil-derived eDNA has facilitated the identification of mammals, birds, reptiles, insects, and soil invertebrates, whereas airborne eDNA has recently emerged as a promising approach for monitoring terrestrial vertebrates and arthropods [2]. Beyond species detection, eDNA is increasingly used to investigate community composition, monitor seasonal population dynamics, evaluate habitat connectivity, detect invasive alien species at early stages of colonization, identify wildlife pathogens, assess ecological restoration success, and support long-term biodiversity surveillance programs.

The integration of environmental DNA with computational biology has greatly expanded its analytical capabilities. Modern bioinformatics pipelines enable automated processing of millions of sequencing reads through quality filtering, chimera removal, sequence clustering, amplicon sequence variant (ASV) inference, taxonomic classification, and biodiversity analysis. Coupling eDNA datasets with geographic information systems (GIS), remote sensing technologies, ecological niche modelling, artificial intelligence, and machine learning has further strengthened predictive conservation science by enabling spatially explicit assessments of species distributions, habitat suitability, and ecosystem change under future climate scenarios. Despite these remarkable advances, several methodological and analytical challenges remain. Environmental conditions such as ultraviolet radiation, temperature, microbial activity, water chemistry, and pH influence DNA degradation and persistence, thereby affecting detection probabilities. Laboratory contamination, PCR inhibition, primer bias, incomplete reference databases, and inconsistencies in sampling protocols may also compromise data reliability. Moreover, translating eDNA concentration into estimates of species abundance or biomass remains an active area of investigation requiring further methodological refinement and ecological validation. As international conservation initiatives increasingly emphasize evidence-based biodiversity management, environmental DNA is becoming an indispensable component of ecological monitoring programs [3]. Its capacity to provide rapid, highly sensitive, and non-invasive biodiversity information has substantial implications for wildlife conservation, protected area management, invasive species control, environmental impact assessment, and ecosystem restoration.

2. Principles of Environmental DNA (eDNA)

Environmental DNA (eDNA) refers to genetic material released by organisms into their surrounding environment through natural biological processes, including the shedding of skin cells, hair, feathers, scales, mucus, saliva, feces, urine, gametes, blood, and decomposing tissues. Unlike traditional molecular studies that require direct collection of tissues or whole organisms, eDNA enables species detection using environmental samples such as water, soil, sediment, snow, ice, and air. Consequently, it has become a non-invasive and highly sensitive approach for biodiversity monitoring [4]. The principle underlying eDNA analysis involves four major steps: sample collection, DNA extraction, molecular detection, and bioinformatic analysis. Environmental samples are first collected under sterile conditions to minimize contamination. DNA is subsequently extracted using commercial kits or optimized laboratory protocols. Target DNA fragments are amplified through polymerase chain reaction (PCR)-based techniques or sequenced using next-generation sequencing (NGS). Finally, the resulting sequences are compared with reference databases to identify the organisms present in the sampled environment.

The persistence of eDNA is influenced by numerous environmental factors including temperature, ultraviolet radiation, microbial activity, salinity, dissolved oxygen, pH, and water flow. Under favorable conditions, DNA may persist for several weeks, whereas high temperatures or intense microbial degradation can rapidly reduce detection efficiency. Understanding these environmental variables is therefore essential for accurate interpretation of eDNA data. Compared with conventional wildlife surveys, eDNA offers several advantages. It facilitates early detection of rare or invasive species, reduces field sampling effort, minimizes habitat disturbance, improves detection sensitivity, and permits large-scale biodiversity assessments with relatively low operational costs [5]. Consequently, eDNA has become an important component of conservation biology, ecological monitoring, fisheries management, and environmental impact assessment.

3. Sources and Types of Environmental DNA

Environmental DNA originates from virtually all living organisms inhabiting an ecosystem. The source of DNA largely determines its persistence, concentration, and suitability for biodiversity assessment. Aquatic ecosystems constitute the most widely studied source of eDNA. Fish, amphibians, aquatic reptiles, crustaceans, mollusks, and marine mammals continuously release DNA into surrounding water through mucus, feces, urine, gametes, epidermal cells, and decomposing tissues. Water samples collected from rivers, lakes, ponds, wetlands, and oceans therefore provide valuable information regarding aquatic biodiversity. Soil represents another important environmental matrix. Soil eDNA contains genetic material from mammals, birds, reptiles, insects, fungi, nematodes, earthworms, bacteria, and plants. Because DNA binds strongly to soil particles, it may persist longer than in aquatic environments, making soil particularly useful for terrestrial biodiversity assessments. Sediment samples often preserve DNA for extended periods owing to reduced microbial degradation. Ancient sedimentary DNA has been successfully employed to reconstruct historical ecosystems, evaluate long-term biodiversity changes, and investigate species distributions during past climatic periods. Airborne eDNA represents one of the most recent developments in molecular ecology.

Air contains microscopic particles carrying DNA released from skin, feathers, pollen, fungal spores, insects, and microorganisms. Recent studies have demonstrated that airborne eDNA can detect terrestrial mammals, birds, amphibians, and insects within natural habitats and zoological parks. Additional environmental matrices include snow, glacier ice, cave sediments, honey, animal nests, feeding sites, fecal deposits, and drinking water sources [6]. These specialized sample types have expanded the applicability of eDNA to extreme environments and difficult-to-access ecosystems.

Table 1: Major Sources of Environmental DNA

Environmental Sample	Target Organisms	Major Applications
Water	Fish, amphibians, aquatic mammals, crustaceans	Aquatic biodiversity monitoring
Soil	Mammals, reptiles, insects, microbes	Terrestrial biodiversity assessment
Sediment	Ancient and extant fauna	Paleoecology and conservation
Air	Birds, mammals, insects	Wildlife surveillance
Snow/Ice	Arctic fauna	Climate change studies
Honey/Nests	Pollinators and insects	Pollinator diversity monitoring

4. eDNA Sampling Strategies

Appropriate sampling strategies are fundamental for obtaining reliable environmental DNA data. The choice of sampling method depends on the study objectives, habitat characteristics, target species, and environmental conditions. Water sampling generally involves collecting between 250 mL and 5 L of water, followed by filtration using membrane filters with pore sizes ranging from 0.22 to 1.2 µm. Filters are preserved at low temperatures or in specialized preservation buffers until DNA extraction. Soil sampling requires standardized collection depths and sterile equipment to minimize contamination. Multiple subsamples are often combined into composite samples to improve representation of local biodiversity. Field contamination represents one of the greatest challenges in eDNA studies. Researchers therefore employ disposable gloves, sterilized instruments, field blanks, extraction controls, and PCR negative controls throughout sample collection and laboratory analysis [7]. Temporal replication is equally important because DNA concentrations fluctuate according to season, reproductive activity, rainfall, water discharge, and species abundance.

Table 2: Comparison of Major Molecular Techniques Used in eDNA Studies

Technique	Sensitivity	Quantification	Multiple Species Detection	Typical Application
Conventional PCR	Moderate	No	No	Species confirmation
qPCR	High	Yes	No	Endangered species monitoring
Digital PCR	Very High	Absolute	No	Rare species detection
DNA Metabarcoding	High	Semi-quantitative	Yes	Community analysis
Next-Generation Sequencing	Very High	Yes	Yes	Large-scale biodiversity assessment

The remarkable progress achieved in environmental DNA methodologies has transformed biodiversity research by providing accurate, rapid, and cost-effective alternatives to traditional ecological surveys. Coupled with advances in high-throughput sequencing and computational biology, eDNA continues to expand its applications across wildlife conservation, ecosystem monitoring, and environmental management. Subsequent sections discuss bioinformatics pipelines, wildlife monitoring applications, biodiversity conservation, invasive species surveillance, and future directions of this rapidly evolving discipline.

Multiple sampling events increase detection probability and reduce false-negative results. Standardization of sampling protocols has become a major research priority to facilitate comparisons among studies conducted in different geographical regions and ecosystems.

5. Molecular Techniques for eDNA Analysis

Several molecular techniques have been developed for detecting environmental DNA, each possessing distinct advantages depending on study objectives. Conventional polymerase chain reaction (PCR) remains one of the simplest approaches for species-specific detection. Although inexpensive and easy to perform, PCR is less sensitive than more advanced molecular methods. Quantitative PCR (qPCR) enables simultaneous amplification and quantification of DNA molecules through fluorescence-based detection. This method is widely employed for monitoring endangered species, invasive organisms, and wildlife pathogens because of its high sensitivity and specificity. Digital PCR (dPCR) partitions DNA samples into thousands of independent micro-reactions, allowing absolute quantification without requiring standard curves. The technique provides exceptional sensitivity for detecting extremely low DNA concentrations and has become increasingly valuable for early invasive species detection. DNA metabarcoding combines universal genetic markers with next-generation sequencing to identify multiple species simultaneously from a single environmental sample. Commonly used barcode regions include mitochondrial 12S rRNA, 16S rRNA, cytochrome oxidase I (COI), and cytochrome *b*. Metabarcoding has revolutionized biodiversity assessment by enabling comprehensive analysis of entire biological communities [8]. Next-generation sequencing (NGS) platforms, including Illumina, Oxford Nanopore, and PacBio technologies, generate millions of DNA sequences in a single sequencing run. These technologies provide unprecedented taxonomic resolution and permit large-scale ecological studies involving complex environmental samples. Selection of molecular markers is critical for successful species identification. Mitochondrial genes are generally preferred because of their high copy number, maternal inheritance, and relatively rapid evolutionary rate, making them particularly suitable for animal biodiversity studies.

6. Conclusion

Environmental DNA (eDNA) has transformed wildlife monitoring and biodiversity conservation by providing a highly sensitive, non-invasive, and cost-effective alternative to conventional ecological survey methods. The ability to detect organisms from trace amounts of genetic material present in environmental samples has significantly improved species detection, particularly for rare, elusive, endangered, and invasive taxa that are often difficult to monitor using traditional approaches. Continuous advancements in molecular biology, high-throughput sequencing, and bioinformatics have further enhanced the accuracy, efficiency, and scalability of eDNA-based biodiversity assessments across aquatic, terrestrial, and aerial ecosystems.

The integration of eDNA with next-generation sequencing, metabarcoding, quantitative PCR, digital PCR, geographic information systems (GIS), remote sensing, artificial intelligence, and ecological modelling has expanded its applications beyond simple species detection to include community composition analysis, habitat assessment, population monitoring, pathogen surveillance, ecosystem restoration, and long-term biodiversity monitoring. These multidisciplinary approaches provide valuable scientific evidence for conservation planning, environmental impact assessment, invasive species management, and policy development aimed at protecting biodiversity under rapidly changing environmental conditions, several methodological challenges remain, including DNA degradation, environmental variability, sample contamination, PCR inhibition, primer bias, incomplete reference databases, and limitations in estimating species abundance from eDNA concentrations. The incorporation of artificial intelligence and machine learning into eDNA data analysis is also expected to improve species identification, ecological prediction, and conservation decision-making. An expanding global reference databases and establishing internationally accepted methodological standards will enhance the widespread adoption of eDNA technologies in ecological research and biodiversity management, environmental DNA represents one of the most significant advances in modern conservation science and molecular ecology. Its exceptional sensitivity, versatility, and broad applicability make it an indispensable tool for monitoring biodiversity, evaluating ecosystem health, detecting invasive species, and supporting evidence-based wildlife conservation. As sequencing technologies, computational methods, and ecological applications continue to evolve, eDNA is expected to play an increasingly central role in global biodiversity assessment and the sustainable management of natural ecosystems in the coming decades.

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