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e DNA: AN EMERGING TOOL FOR BIOMONITORING

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Short Communication

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Environmental DNA (eDNA) sequencing is a rapidly emerging method to study biodiversity and monitoring ecosystem changes. eDNA is DNA that is collected from a variety of environmental samples such as soil, seawater, snow or even air rather than directly sampled from an individual organism. As various organisms interact with the environment, DNA is expelled and accumulates in their surroundings. eDNA analysis utilizes these left-behind genetic signatures to provide information about all the species present in a sample – from microbes to large creatures. eDNA analysis can provide clues about the presence of a species without any disruption to the ecosystem. e DNA sequencing is an emerging tool in conservation and monitoring past and present biodiversity, water testing, soil testing and many more.

Analysis of e DNA

Natural samples contain DNA in a variety of packages, including free DNA, virus particles, and prokaryotic and eukaryotic cells.As species interact with the environment, they continuously expel DNA to their surroundings. For higher organisms, this DNA may come from excreted cells or tissue such as urine, faeces, hairs, skin and also from dead individuals. The content of an eDNA sample is typically analyzed by amplification using polymerase chain reaction (PCR) and subsequent DNA sequencing. The amplification is done either by a single-species approach using specific primers or by multiple-species (multiple-taxon) approach using generic primers for a given focal group of organisms.

Next Generation Sequencing methods such as DNA metabarcoding –utilize mass DNA sequencing for the simultaneous molecular identification of multiple taxa in a complex sample.

Every organism has a unique DNA sequence, or barcode, associated with it. This DNA barcode is a highly variable region interspersed between conserved genomic regions. eDNA metabarcoding involves target-specific amplification and sequencing of these barcodes, often mitochondrial cytochrome oxidase 1 (CO1) or the 18S ribosomal subunit. These are useful approaches for distinguishing between higher-order eukaryotes.

The Barcodes may include These target genes have basically been chosen due to their high resolution at the species level, but high copy number per cell of mitochondria, chloroplasts and rRNA genes also make them useful in eDNA studies, since they are more likely to be picked up than single-copy nuclear DNA.

Environmental metagenomics has typically relied on sequencing the 16S or internal transcribed spacer (ITS) rRNA genes for detecting bacteria or fungi, respectively. Both 16S and ITS rRNA gene sequencing are well-established methods for comparing sample phylogeny and taxonomy from environmental samples.

Long-range PCR can be used to amplify large DNA sequences, such as mitochondrial genomes. These longer DNA sequences can help distinguish between species when smaller DNA barcodes are not available. This approach is favourable for sequencing DNA that has not been degraded by the environment

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Challenges for eDNA Approaches
Despite of useful applications of eDNA, several issues associated with this approach should be considered such as:

Contamination
The most serious pitfall of eDNA is the risk of contamination and thus possibility of false positive results. Contamination of samples can occur from taking the samples in the field to every step of analyses in the laboratory.

Inhibition
Humic acids or humic substances, co-extracted with DNA in environmental samples, strongly inhibit enzymes such as the Taq Polymerase used in PCR reactions to amplify DNA.

Errors
Incorrect DNA sequences may give rise to biased results like that of contaminants. Errors can occur either before sampling in long-term preserved DNA.

Reference DNA databases
Identification of DNA sequences derived from environmental samples depends on reliable reference DNA-sequence databases.

Interpretation of results
Another important issue in eDNA studies concerns about the criticalinterpretation of final results.

Advantages of eDNA Approaches
Despite of crucial challenges associated with eDNA approaches it also offers some great advantages over traditional methods in biodiversity monitoring, such as:

It facilitates improved detection of native species as well as early detection of invasive Species

Scientists studying environmental DNA often analyze trace amounts of DNA per species in a given sample, without knowing the types or abundance of species represented.

eDNA is a truly non-invasive method that inflicts no damage on the species or habitats under study.In contrast, physical surveys of natural environments require manual data collection and can be disruptive.

eDNA is more cost-efficient than traditional methods as this depends on the target species and take shorter handling time and lower cost as compared to traditional monitoring techniques which are not effective for processing large or complex samples.

CONCLUSION
Thus the potential of eDNA metabarcoding in biological research seems limitless, but the technique requires advancement and optimization of procedures. Overcoming its pitfalls metabarcoding will surely emerge as an essential tool for a number of scientific tasks not only in biodiversity monitoring but also in several fields for the assessment of communities for numerous purposes making it extremely versatile and vital to the future of molecular research.

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