## Assessment of bacterial diversity in polluted and unpolluted surface freshwater bodies and adjacent sediments using environmental DNA and metabarcoding techniques

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## <u>Abstract</u>

In the environment, a wide range of organisms, including plants, animals, and microorganisms, leave behind genetic material termed environmental DNA (eDNA), which can be collected from various local habitats, including air, water, sediment, snow, and soil. Here, aquatic samples were obtained from a variety of freshwater bodies, including previously polluted and traditionally clean sites (estuaries, tributaries, man-made waterways, lacustrine, and lentic environments), using a multifaceted approach that employed two different pore-size filters (0.22 µm and 0.45 µm) for the filtration of water samples. In an exploratory effort to potentially enhance the comprehensiveness of the knowledge of the environmental conditions, sediment samples were collected from the surrounding regions of the previously acquired water samples. Following the collection of samples, DNA extraction, and quantification were conducted. Subsequently, the V4 hypervariable region of the prokaryotic 16S ribosomal RNA (rRNA) gene was amplified by polymerase chain reaction (PCR) using a universal primer set (515f and 806r). After using PCR and gel electrophoresis to determine the size and quantity of amplified DNA fragments, our current research is centered on DNA metabarcoding of approximately thirty candidate PCR amplicons using next-generation sequencing. The primary objective of our study is to assess the bacterial diversity and abundance in local aquatic habitats, as studying the diversity and abundance of freshwater microbiomes is of utmost importance in understanding their role in ecosystem functioning, their impact on the health and survival of larger organisms, and their potential to aid in identifying environmental stressors and promoting conservation efforts.