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Importance of Proteomics in Bioremediation

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Abstract: ***Bioremediation can be used to help clean up contaminated regions today since environmental contamination is a major issue. The use of microbial mediated bioremediation offers a lot of potential for effectively restoring a polluted environment, but a lack of detail about the parameters that determine whether certain microbial communities can grow and reproduce in a polluted environment makes this kind of bioremediation challenging. Transcriptomics, proteomics, and interactomics are flourishing fields that hold great potential for addressing long-standing issues about the molecular mechanisms that drive the mineralization pathway. With the aid of microarray technology, transcriptomic approaches have been applied to study the structure and expression of transcripts during mineralisation. However, transcripts cannot generally cause any physiological effect; instead, they must be translated into proteins with significant functions. Two-dimensional polyacrylamide gel electrophoresis (2-DE) is a powerful tool for identifying these proteins via proteomic techniques. The current advancements in mass spectrometry (MS) and protein microarrays are playing an important role in functional proteomics. A comprehensive genome-wide analysis of differentially expressed proteins and a screen for proteins that interact with specific mineralization factors would allow us to gain a better understanding of bioremediation.***
Keywords: bioremediation, transcriptomics, proteomics, interactomics, microarray

I. INTRODUCTION

The rapid development of industries has not only improved human lives in many ways, but also had a detrimental effect on the environment.

As a result of industrial waste and other sources, soil, water, and the air are contaminated with toxic pollutants. Reduced availability of arable land, potable water, and clean air has become a global crisis. In spite of the fact that hazardous waste disposal has become more recognized than in the past, there is still a significant amount of land and water that is contaminated. Various physical and chemical methods are used to clean up contaminated soil, but they do not restore the site's biodiversity once the treatment is completed (Zhang, Li, and Nie 2010).

Unlike these methods, bioremediation is a non-toxic, cost-effective, and sustainable method for removing toxic contaminants from our environment. It utilizes a multitude of microorganisms to remove pollutants from our environment. There are, however, some limitations associated with bioremediation, including the fact that it takes time and has a narrow action range (Maphosa et al. 2012).

A culture-independent technique of genomics can now be used on-site for the analysis of uncultivable microorganisms used for bioremediation. As a result of these efforts, several new 'omics' fields have been created: transcriptomics, proteomics, metabolomics, interactomics, etc.

Analyzing proteins can be very challenging. Unlike other cellular macromolecules, proteins are found in all subcellular compartments that are defined by the envelope architecture or membrane invaginations. Typically, soluble proteins are located in the cytoplasm or periplasm (if present).

They are also secreted to the extracellular milieu. In view of the huge structural and functional diversity of proteins, only rational combinations of diverse analytical approaches can provide an overview of the overall state of a cell. Measuring the complexity of microbial proteomes depends on three factors: genome size, localization within cells, and adaptation to environmental changes. The widespread adoption of proteomic methods in the past decade was due primarily to the availability of robust analytical methods (such as two-dimensional gel electrophoresis), and the affordability and accessibility of mass spectrometers. (Wöhlbrand, Trautwein, and Rabus 2013).

A proteomics study analyzes all of the proteins present in a living organism in order to generate a comprehensive picture of how the proteins function within the specific environment (Peter Chovanec 2017). The proteomics approach provides a comprehensive view of the protein complement of biological systems and, working in combination with other omics technologies, it has a significant role to play in helping us discover the mechanisms of these cellular processes and, consequently, advance the development of environmental biotechnologies (Lacerda and Reardon 2009).

Proteomics is a valuable technique that is more complex than genomics, since an organism's genome is more or less constant, but the proteome varies from cell to cell and over time. By studying the sequence of proteins produced by bacterial cultures (proteomics) and in environmental samples (metaproteomics), it may be possible to uncover differences in the composition and production of proteins and find many proteins that play a critical role in the physiological response of microbes to pollutants. (Dangi et al. 2019).

In addition, metaproteomics has also been used to detect protein expression profiles of microbial communities in environmental samples without culturing any bacteria in the samples. This approach reflects the actual functional activities of microbes in a particular ecosystem.

Due to the advent of two-dimensional gel electrophoresis (2-DE) combined with mass spectrometry and protein sequencing, as well as improvements in protein structure databases, this approach is increasingly feasible (Herbst et al. 2016).

II. IMPACT OF PROTEOMICS ON BIOREMEDIATION.

The proteome of membrane proteins plays an important role in bioremediation, particularly in terms of PAH biodegradation, where alterations in any particular bacterium affect cell surface proteins and receptors. With the introduction of MudPIT, a new method of multidimensional protein identification, 2-DE is now more suitable to be used in compartmental proteomics (Singh and Nagaraj 2006).

PAHs, ubiquitous environmental pollutants, must be eliminated from the environment. The use of naturally occurring microorganisms and genetically engineered microorganisms has been partially successful in the *in situ* and *ex situ* bioremediation of PAHs (Dell' Anno et al. 2021).

Metaproteomics has given a new window into the functional and phylogenetic processes involved in the biodegradation of hydrocarbons in soil. During PAH biodegradation, dioxygenases convert aromatic hydrocarbons to cis-dihydrodiol. A lower proportion of metaproteomes occurs in compost treated soils, which may be associated with the biodegradation of petroleum-derived alkanes and PAHs. Members of *Sphingomonadaceae* plays a key-role in the biodegradation of aromatic compounds in the compost-amended soil (Bastida et al. 2016).

To date, proteomic analysis of *Pseudomonas putida KT2440* incubated with aromatic compounds has identified 110 proteins involved in hydrocarbon degradation. A few of them are benzoate dioxygenase (BenA, BenD), catechol 1,2-dioxygenase (CatA), protocatechuate 3,4-dixoygenase (PcaGH), β-Ketoadipyl CoA thiolase (PcaF) and 3-oxoadipate enol-lactone hydrolase (PcaD) (Kim et al. 2006)**.** Similarly, the proteomic approach has recently led to the identification of about 250 proteins involved in hydrocarbon degradation pathways in *Pseudomonas sp*.

The predominant drivers of the bioremediation process are F420-dependent oxidoreductase and phthalate 4,5-dioxygenase grown in the presence of pyrene (Swati et al. 2020).

Fungus such as *Pycnoporus sanguineus* shows efficient and promising metabolic mechanism for bioaugmentation and biodegradation of TPhP-polluted water-sediment, which is of great significance to make better use of white rot fungi in TPhP contamination bioremediation.

It has desired ability to degrade TPhP under optimal conditions. Proteomic analysis suggests that TPhP is hydroxylated, oxidatively cleaved, and methylated by cytochrome P450s, aromatic compound dioxygenases, oxidative species-generation enzymes, and methyltransferases (Feng et al. 2021).

Organohalides are highly toxic and persistent in the environment, and bioremediation techniques should be used to remove or neutralize them. (Wohifarth and Diekertt et al.1997.) Anaerobic bacteria that can perform organohalide respiration have therefore been highly sought after as candidates for bioremediation in sites with low oxygen concentrations, such as aquatic sediments, submerged soils, and groundwater (Anon 1998a; Smidt and de Vos 2004).

By analysing how organohalide respiring bacteria respond to various conditions, proteomic approaches can be used to develop concepts that describe interactions between these bacteria and its cellular components with respect to its environment. Species of bacteria that respire organohalide are often found in consortia containing other anaerobes, such as *Desulfovibrio, Eubacterium, Acetobacterium, Citrobacter, Spirochetes,* and *Clostridium,* that produce hydrogen and acetate from organic substrates (Anon 1998b; Duhamel and Edwards 2006; Lee et al. 2006; Richardson et al. 2002).

Table 1. List of Micro-organisms involved in bioremediation.

Sr. No.	Name of Micro-organism	Degraded Pollutant	References
1.	*Pseudomonas putida KT2440*	Aromatic compounds	(Swati et al. 2020)
2.	*Sphingomonadaceae spp.*	Polycyclic aromatic hydrocarbon	(Bastida et al. 2016)
3.	*Pycnoporus sanguineus*	Triphenyl phosphate (TPhP)	(Feng et al. 2021)
4.	*Desulfovibrio*, *Eubacterium*, *Acetobacterium*, *Citrobacter*, *Spirochetes*, *and Clostridium*	Organohalides	(Anon 1998b; Duhamel and Edwards 2006; Lee et al. 2006; Richardson et al. 2002)
5.	Consortia of *Alcanivorax*, *Halomonas*, *Marinobacter*, *Oleispira*, *Thalassolituus*, and *Oleiphilus*.	Petroleum hydrocarbons	(Dell'Anno et al. 2021; Yakimov, Timmis, and Golyshin 2007)
6.	*Halomonas*, *Dietzia*, and *Arthrobacter*	Diesel oil	(Dell'anno et al. 2020)
7.	*Cladosporium*, *Aspergillus*, *Cunninghamella*, *Penicillium*, *Fusarium*, and *Mucor*	Aliphatic hydrocarbon degradation	(Amend et al. 2019)

III. APPROACH TOWARDS PROTEOMICS

The majority of proteomics studies require a pre-fractionation of the samples prior to mass spectrometry (MS) analysis. This can be achieved via gel electrophoresis or by certain gel-free techniques. In terms of resolution and reproducibility, 2D-Gel electrophoresis (2D-GE) is now the best method for separating complex protein mixtures. In spite of this, there are some drawbacks such as limited ability to fractionate hydrophobic proteins and glycoproteins successfully, detection of small peptide molecules, and quantitative uncertainty (Lambert et al. 2005). Generally, 2D-GE is not as reproducible as LC-based separation, which is an important advantage for comparative proteomics. As an example of a statistical package which facilitates semiquantitative proteomics, we can mention Progenesis (Nonlinear Dynamics), ImageMaster 2D Platinum (Ge Healthcare, Amersham Biosciences) and PDQuest (Bio-Rad).

By introducing MS technology to proteomics, we have greatly enhanced the throughput of proteomic studies as compared to electrophoretic and chromatographic approaches. MS allows us to identify post-translational modifications such as phosphorylation and acetylation, both essential for cell signalling and epigenetics (Bantscheff et al. 2012).

IV. CONCLUSION

Developing a successful bioremediation strategy requires an in-depth understanding of degradative microbial communities, a challenging task for microbiologists. Genomic analysis, metabolomics, and proteomics have thus become major tools for the identification of all the unexplored microbial communities capable of degrading heavy metals and the identification of the diverse metabolites produced by organisms to cope with stress.

Compared to conventional remediation methods, bioremediation techniques have been found to be far more efficient and effective and it also maintains the ecological balance of the environment. To conclude, it will be necessary to evaluate *in situ* sediment bioremediation on a large scale, by scaling up laboratory or small-scale studies, as well as estimating economic costs and environmental impacts. As a result, they are important elements for the development of sustainable and eco-compatible bioremediation interventions on contaminated sites.

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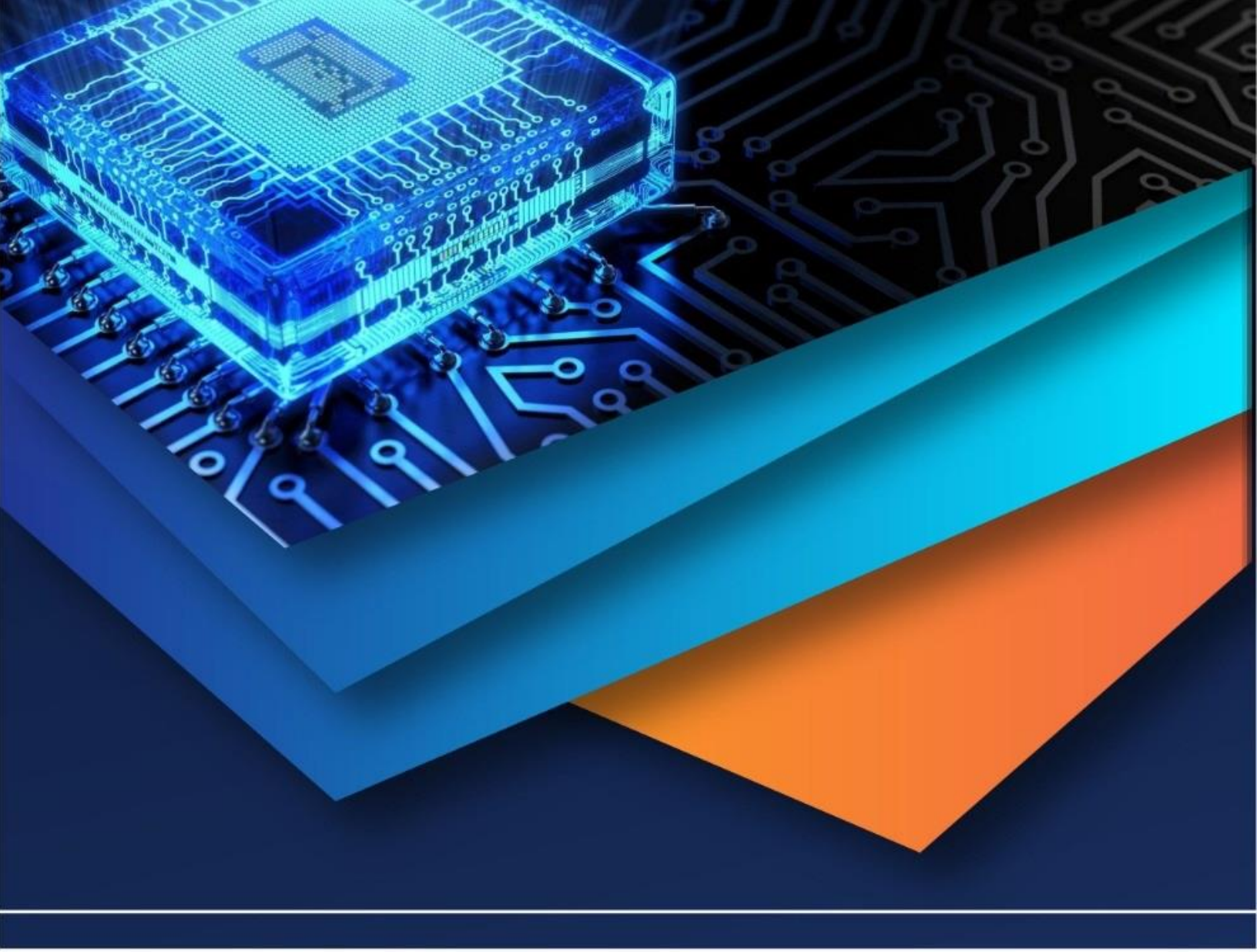

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