Provisional chapter

Scientific Swift in Bioremediation: An Overview

Ranjith N. Kumavath and Pratap Devarapalli

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1. Introduction

A pure environment gives a quality of life on earth. In ancient times, it was believed that people on earth had an unlimited abundance of land and resources; today, however, the resources in the world show a greater or lesser degree of carelessness and negligence in using them. In many parts of the globe, the problems associated with contaminated sites are now growing up. The actual cause of this scenario resulted from past industrial activities when awareness of the health and environmental effects connected with the production, use, and disposal of hazardous substances were less well recognized than today. It became a global complication when the estimated number of contaminated sites became significant. Several traditional methods have been applied to overcome this inconvenience. From the list of ideas that have been applied, the best ones are to demolish the pollutants, if possible completely, or at least to transform them into innoxious substances. Bioremediation is an option that utilizes microbes to remove many contaminants from the environment by a diversity of enzymatic processes. However, it will not always be suitable, as the range of contaminants on which it is effective is limited, the time scales involved are relatively long, and the residual contaminant levels achievable may not always be appropriate. We attempted to assist by providing information on how bioremediation is linked with cutting-edge sciences such as genomics, transcriptomics, proteomics, interactomics, and bioinformatics.

Some novel molecular biology approaches, such as genetic engineering, transcriptomics, proteomics, and interactomics, hold considerable potential as tools for studying the processes that control mineralization pathways. To comprehend the mineralization process in a meaningful way, strategies that integrate transcriptomics and proteomics data must be refined. These methods have considerable potential in predicting organism metabolism in contaminated environments and predicting microbial-supported attenuation of contaminants to expedite bioremediation. Bioinformatics is a technique for identifying and and protein functions, interactions, metabolic and regulatory pathways, etc. Bioinformatics analysis will make it easier

and faster to comprehend cellular processes to treat and regulate microbial cells such as factories. Understanding molecular mechanisms and cellular manipulation using bioinformatics will focus on the coming decade.

Bioremediation is a method of removing contaminants from the environment using bacteria and a variety of enzymatic processes. The main advantages of bioremediation are its cost-effectiveness and low-tech approaches, which are often well accepted by the public and can usually be carried out on site (Robb et al., 1995). However, because the spectrum of contaminants on which it is effective is restricted, the time scales required are quite long, and the residual contamination levels achieved may not always be sufficient, they will not always be suitable. Bioremediation has been applied in a number of locations across the world with varying degrees of effectiveness (Ajay et al., 2009). Here, we attempted to assist by providing information on how bioremediation is linked with cutting edge sciences such as genomics, transcriptomics, proteomics, interactomics, and bioinformatics (Fleming et al. 1993., Schena et al. 1998., Sikkema et al. 1995., Kuhner et al. 2005., Ellis et al. 2000).

2. Genetic analysis of genes involved

The presence and expression of important genes involved in bioremediation can reveal more information about microbial activities than 16S rRNA sequence analysis (Rogers and McClure. 2003). The relative number of genes involved in bioremediation and the capacity for pollutant degradation have a positive association in general (Rogers and McClure. 2003., Schneegurt and Kulpa. 1998). On the other hand, the genes for bioremediation may be present but not expressed. As a result, quantifying mRNA levels for critical bioremediation genes has become more important. Increased mRNA concentrations are frequently linked to greater contaminant degradation rates, at least qualitatively (Schneegurt and Kulpa. 1998). For instance, the concentrations of mRNA for *nahA,* a gene involved in the aerobic degradation of naphthalene, were positively correlated with rates of naphthalene degradation in hydrocarbon-contaminated soil (Fleming et al. 1993). One method for removing mercury from water is the conversion of soluble ionic mercury, Hg(II), to volatile Hg (0); the content of mRNA for merA, a gene implicated in Hg(II) reduction, was highest in mercury-contaminated waters with the highest rates of Hg(II) reduction (Nazaret et al. 1994). However, the concentration of merA was not necessarily proportional to the rate of Hg(II) reduction (Nazaret et al. 1994, Jeffrey et al. 1996), indicating that factors other than gene transcription can influence bioremediation rates.

There are currently highly sensitive technologies for detecting mRNA for essential bioremediation genes in single cells (Bakermans and Madsen. 2002). When combined with 16S rRNA probing of the same environmental samples, this approach might reveal which organisms' phylogenetic groupings express the genes of interest. Analysis of mRNA concentrations for genes not directly engaged in bioremediation might provide further information about the variables that influence the rate and extent of bioremediation. The development and metabolism of organisms participating in bioremediation in contaminated environments might be

hampered by low nutrition levels, pH, salinity, and other environmental factors. Molecular approaches are used in ecological studies of phytoplankton to assess the stress response of photosynthetic microorganisms in the environment (Palenik and Wood. 1998). Similarly, assessing the metabolic status of bioremediating microorganisms by analyzing mRNA quantities for key genes involved in stress response might aid in identifying changes to polluted settings that could increase bioremediation.

3. Role of transcriptomics

The transcriptome, a dynamic connection between the genome, the proteome, and the cellular phenotype, is a subset of genes transcribed in each given organism. One of the most important mechanisms for adjusting to changes in environmental circumstances and, consequently, survival is gene expression control. Transcriptomics is a term that defines this process over the whole genome. DNA microarrays are highly sophisticated transcriptomics platforms that allow determination of the mRNA expression level of almost any gene in an organism (Schena et al. 1998., Golyshin et al. 2003., Diaz. 2004). The most difficult aspect of microarray studies is data interpretation (Dharmadi and Gonzalez. 2004). Hundreds of genes may be up- or downregulated in response to a stress condition. Several statistical concerns, such as compensating for random and systematic errors and conducting poor analysis, become extremely complicated in this scenario (Singh and Nagaraj. 2006).

4. Applications of DNA microarray

However, with entire genome sequences of microbes with bioremediation capabilities (Golyshin et al. 2003., Tiedje. 2002., Heidelberg et al. 2002., Seshadri et al. 2005., Rabus et al. 2005), studies are not hastening in a rapid manner. With the finished genome sequences, whole-genome DNA microarrays may be used to examine the expression of all genes in each genome under diverse environmental variables (Gao et al. 2004., Muffler et al. 2002. Schut et al. 2003). This type of genome-wide expression research is helpful in discovering regulatory circuits in these organisms (Lovely. 2003., Rabus et al. 2005., Muffler et al. 2002). DNA microarrays have previously been used to assess the physiology of pure environmental cultures and track the catabolic gene expression profile in mixed microbial communities (Schut et al. 2003). (Dennis et al. 2003). When a DNA microarray had been used to analyze variations in mRNA expression levels in Bacillus subtilis cultured under anaerobic circumstances, more than 100 genes were identified to be influenced by oxygen-limiting conditions (Ye et al. 2000). Only genes from populations contributing more than 5% of the community DNA can be identified in PCR-based cDNA microarrays. Therefore, sensitivity may be a part of the problem. The sensitivity of spotted oligonucleotide DNA microarrays and their usefulness for bacterial functional genomics were tested using a number of factors (Denef et al. 2003). The 50-C6-amino-modified 70 mers printed on CMTGAPS II substrates at a 40 mM concentration and the application of tyramide signal amplification labeling were the best parameters.

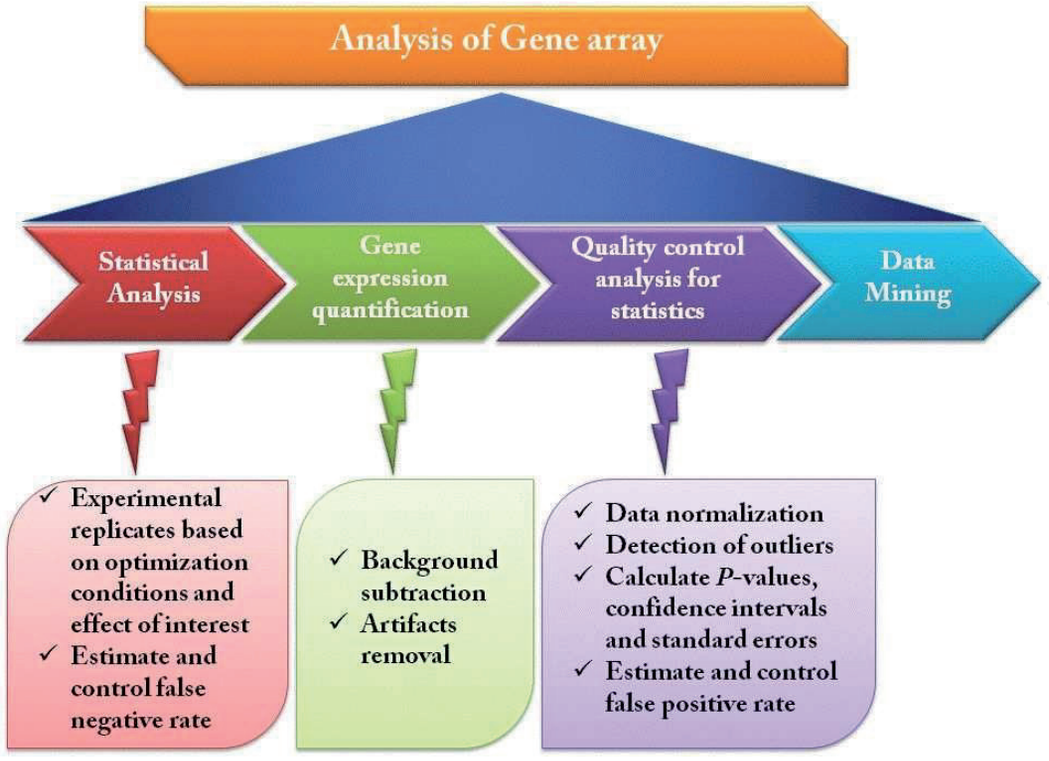

Figure 1. Gene array analysis workflow. A graphical representation depiction of DNA microarray data analysis and relative constraints for each data analysis area is shown during data mining.

A comprehensive 50-mer-based oligonucleotide microarray for effective monitoring of biodegrading populations was designed based on the most known genes and pathways involved in biodegradation and metal resistance (Rhee et al. 2004). This type of DNA microarray successfully analyzed naphthalene-amended enrichment, and soil microcosms indicated that microflora evolved in diverse ways depending on incubation circumstances (Cho and Tiedje. 2002). During the breakdown of alkylbenzenes, a global gene expression study showed the coregulation of multiple previously discovered genes (Kuhner et al. 2005). In addition, DNA microarrays were utilized to identify bacterial species, quantitative approaches of microbial genome stress gene analysis, and genome-wide transcriptional profiles (**Figure 1**) (Muffler et al. 2002., Greene and Voordouw. 2003).

5. Footprints of proteomics

In 1995, the words 'proteomics' and 'proteome' were used to describe a crucial postgenomic aspect that evolved from expanding massive and complicated genome sequencing data (Wasinger et al. 1995). Because the observed phenotype is a direct outcome of the activity of proteins rather than the genome sequence, proteomic study is very important. This technique has historically relied on extremely effective separation technologies such as two-dimensional polyacrylamide gel electrophoresis (2-DE) and contemporary bioinformatics tools in combination with mass spectrometry (Hochstrasser. 1995). Nevertheless, 2-DE has been regarded as

a limiting method for extremely basic and hydrophobic membrane proteins in compartmental proteomics. The proteome of membrane proteins is of great importance in bioremediation, particularly in PAH biological degradation, where various changes in each site-specific bacteria impact cell-surface proteins and receptors (Sikkema et al. 1995). The introduction of an alternate technique for multidimensional protein identification technology has improved 2-DE for use in compartmental proteomics (Paoletti et al. 2004). Mass spectrometry has revolutionized environmental proteomics by allowing researchers to analyze small molecules, peptides, and proteins. This has increased the sensitivity of protein identification by many orders of magnitude while also reducing the time it takes to identify proteins from hours to minutes (Aebersold and Mann. 2003). Proteomics for protein identification has benefited greatly from advances in mass spectrometry methods combined with database searches (Singh and Nagaraj. 2006).

By generating peptide mass fingerprinting, matrix-associated laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS) is the most often utilized mass spectrometry technique for detecting proteins of interest excised from 2-DE gels (Aebersold and Mann. 2003., Aitken and Learmonth. 2002., Landry et al. 2000). The combination of direct sample fractionation on a chip and MALDI-TOF-MS analysis is known as surface-enhanced laser desorption ionization mass spectrometry (SELDI-TOF-MS) (Merchant and Weinberger. 2000., Seibert et al. 2005). SELDITOF-MS was used to examine a range of differentially expressed signature proteins in blue mussels (Mytilus edulis) subjected to PAHs and heavy metals (Knigge et al. 2004). The liquid chromatography–mass spectrometry (LC–MS) approach has opened a new analytical opportunity for detecting and identifying potential water contaminants (Joo and Kim. 2005). Furthermore, metabolites and degradation products have been considered in determining the fate of organic pollutants such as pesticides, surfactants, algal and cyanobacterial toxins, disinfection byproducts, and medications in the environment during water treatment strategies (Singh and Nagaraj 2006., Joo and Kim. 2005).

6. Interaction of interactomics

Genome-wide mRNA profiling cannot determine the activity, organization, or eventual destination of the gene products, the proteins. On the other hand, many proteomic techniques can successfully deliver direct solutions. When cellular proteins and several other relevant cellular expressions are on crest during the physiological response in the bioremediation process of any contamination, it is extremely unusual for any protein molecule to operate as a unique pillar (Muffler et al. 2002., Eyers et al. 2004., Segura et al. 2005., Kuhner et al. 2005., Singh and Nagaraj. 2006). With numerous proteins participating in multicomponent protein aggregation, cellular life is structured in general by a complicated protein interaction network. At the proteome level, affinity tag/pull-down/MS/MS methods are often used to identify these aggregated proteins, referred to as 'interactomics' (Lee and Lee. 2004., Coulombe et al. 2004., Gingras et al. 2005). One of the primary focuses of functional proteomics and/or second-generation proteomics is studies on protein–protein interactions and supermolecular complex formation (Singh and Nagaraj. 2006).

The increased need for microarray-based assays is being driven by genomics and proteomics for the investigation of gene and protein function from a global bioremediation perspective. Previously, protein microarray technology has been effectively used in fundamental and applied proteome research for protein identification, quantification, and functional analysis (Labor and Ramachandran. 2005). A broad range of protein microarray-based techniques has already been validated, and this technology is capable of filling the gap between transcriptomics and proteomics and the DNA chip (Liu and Zhu. 2005). Microarray-based protein–protein interaction studies in bioremediation, on the other hand, are still needed to understand the chemotaxis phenomena of any location-specific bacteria toward environmental contaminants (Singh and Nagaraj. 2006).

7. Evolution of genomics

The use of genetics in bioremediation has resulted in significant advancements in the research of pure cultures (Nierman & Nelson, 2002). Next-generation genome sequencing methods are critical for improving our understanding of the physiological and genomic characteristics of microorganisms that are important to bioremediation. For some organisms that are significant in bioremediation, whole or almost complete genome sequences are currently available (**Table 1**). Researchers' perspectives have shifted after applying bioremediation to modern sciences such as genomics, which yielded distinct results. *Geobacter* species, for example, have been shown to be crucial in the bioremediation of organic and metal pollutants in subterranean habitats, according to molecular analysis. The genome sequencing of multiple *Geobacter* species and closely related organisms has profoundly changed our understanding of how *Geobacter* species behave in contaminated subterranean habitats. *Geobacter* species were formerly assumed to be nonmotile until their genomes were sequenced. However, genes producing flagella were later revealed in the Geobacter genomes (Childers et al. 2002). According to further research, *Geobacter metallireducens* develops flagella only when it is growing on insoluble Fe(ra) or Mn(IV) oxides. Chemotaxis genes were found in the *Geobacter* genomes, and experiments demonstrated that *G. metallireducens* had new chemotaxis to Fe(II), which might allow it to navigate to Fe(III) oxides under anaerobic circumstances. Pili genes are present and are expressed especially during insoluble oxide development (Childers et al. 2002). According to genetic research, the pili's function is to help in attachment to Fe(III) oxides and movement along sediment particles in search of Fe(III).

This energy-efficient process in *Geobacter* species for finding and reducing Fe(ra) oxides differs from Fe(III) reduction mechanisms in other well-studied organisms, including *Shewanella* and *Geothrix* species. Other species produce Fe(III) chelators, which solubilize Fe(m) from Fe(m) oxides (Nevin and Lovley, 2002), as well as electron shuttling compounds, which take electrons from the cell surface and then reduce Fe(m) oxides (Newman and Kolter. 2000., Nevin and Lovely. 2002a). *Shewanella* and *Geothrix* species can decrease Fe(III) without directly touching Fe(m) oxide. The synthesis of chelators and electron shuttles, on the other hand, requires a large amount of energy, and the lower metabolic energy requirements of the *Geobacter* approach are likely the reason why *Geobacter* species consistently outcompete other

Microorganism	Relevance to bioremediation	Web site for genome documentation
Dehalococcoides ethanogenes	Chlorinated solvents are reductively dechlorinated to ethylene. *D. ethanogenes'* 16S rRNA gene sequence is closely similar to sequences found in subsurface habitats where chlorinated solvents are degraded.	http://www.tigr.org
Geobacter sulfurreducens Geobacter metallireducens	Reductive uranium precipitation and anaerobic oxidation of aromatic hydrocarbons. During anaerobic in situ bioremediation of aromatic hydrocarbons and uranium, 16S rRNA gene sequences closely linked to recognized *Geobacter* species prevail.	http://www.jgi.doe.gov http://www.tigr.org
Rhodopseudomonas palustris	*Rhodopseudomonas palustris* is the main organism for revealing anaerobic aromatic chemical metabolic pathways and regulating this metabolism.	http://www.jgi.doe.gov
Pseudomonas putida	*Pseudomonas putida* has a wide metabolic range capable of aerobically degrading a wide range of organic contaminants. Beneficial organism for improving bioremediation capabilities through genetic engineering.	http://www.tigr.org
Dechloromonas aromatica	*Dechloromonas aromatica* represents a widely distributed genus of perchlorate-reducing bacteria capable of anaerobic benzene oxidation in conjunction with nitrate reduction.	http://www.jgi.doe.gov
Desulfitobacterium hafniense	*Desulfitobacterium hafniense* enables chlorinated solvents and phenols to undergo reductive dechlorination. Desulfitobacterium species may be found in a wide range of habitats.	http://www.jgi.doe.gov
Desulfovibrio vulgaris	*Desulfovibrio Vulgaris* has been shown to precipitate uranium and chromium reductive. It is yet to be proven that it plays a role in polluted settings.	http://www.tigr.org
Shewanella oneidensis	In culture, a closely related *Shewanella* species reduced U(vi) to U(iv); however, *Shewanella* species have still yet to be relevant in a metal reduction in sedimentary contexts.	http://www.tigr.org
Deinococcus radiodurans	*Deinococcus radiodurans* are highly radiation-resistant. They might be genetically modified for bioremediation of radioactive situations.	http://www.tigr.org

Table 1. Overview of genomes of microorganisms pertinent to bioremediation.

Fe(III)-reducing microorganisms in a variety of subsurface environments (Nevin and Lovely. 2002b). Understanding this and a slew of other previously unknown physiological properties of *Geobacter* species is critical for directing the adjustment of subsurface conditions to maximize *Geobacter* species' capacity to remove organic and metal pollutants from contaminated groundwater.

Research on the physiology of additional bioremediation-potential bacteria whose genomes have been sequenced is accelerating in a similar way. With the finished genome sequences, whole-genome DNA microarrays may be used to examine the expression of all the genes in each genome under different environmental conditions. It is possible to identify which proteins are expressed using proteomic techniques (Nierman & Nelson, 2002). This genome-wide expression analysis provides important data for identifying regulatory circuits in these

organisms (Baldi and Hatfield, 2002). This would be important because the mechanisms that control the regulation of catabolic and respiratory genes, which are most crucial in bioremediation, are widely undefined. It will be feasible to understand the function of numerous previously unknown genes and interpret bioremediation processes when genetic systems for these environmentally relevant organisms become accessible. The accessibility of *Geobacter* genomes and a genetic framework for these organisms, for example, allows researchers to determine which of the more than 100 c-type cytochromes found in the genome play a significant role in electron transport to metals (Lloyd et al. 2003., Leang et al. 2003).

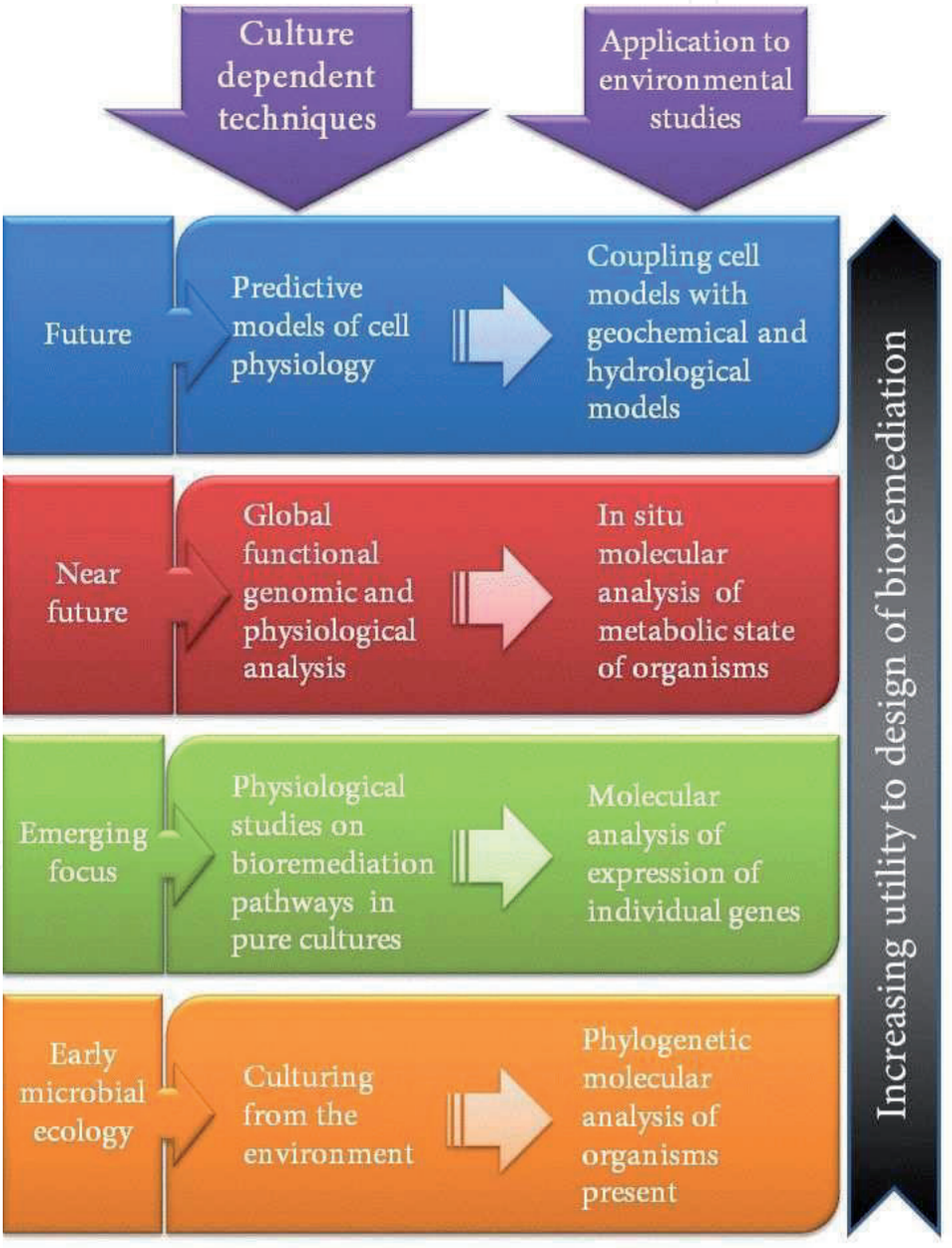

Figure 2. A visual representation of how pure culture studies have become increasingly sophisticated and their application to the study of microbial communities has become more common.

A treatability study is a procedure that involves incubating samples of the contaminated environment in the laboratory and documenting the rates of contaminant degradation or immobilization (Rogers and McClure. 2003). Such research offers an estimate of the potential metabolic activity of the microbial population while providing little information into the microorganisms responsible for bioremediation. When studying bioremediation processes in more depth, most people try to separate the involved species (Rogers. et al. 2003). In microbial ecology, the isolation and characterization of pure cultures have been and will continue to be critical for the development and interpretation of molecular studies (**Figure 2**). The restoration of isolated strains that are representative of the bacteria responsible for bioremediation can be extremely valuable because, as explained below, studying these isolates allows researchers to look into not only their biodegradation reactions but also other aspects of their physiology that may control their growth and activity in contaminated environments. Nevertheless, before applying molecular methods to bioremediation, it was unclear whether the isolated microbes were important in in situ bioremediation or if they were laboratory weeds that grew quickly but were not the primary organisms responsible for the environmental reaction of interest.

8. The 16S rRNA approach

The discovery that the sequences of highly conserved genes present in all microorganisms, most notably the 16S rRNA genes, may offer a phylogenetic characterization of the microorganisms that make up microbial communities was a significant advancement in the area of microbial ecology (Pace et al. 1986., Amann et al. 1995). This was a benefit to the study of bioremediation since it meant that by analyzing 16S rRNA sequences in polluted settings, the phylogenetic position of microorganisms involved in bioremediation processes could be identified unequivocally (Rogers and McClure. 2003., Watanabe and Baker. 2000).

The finding that bacteria that prevail during bioremediation are closely linked to organisms that can be cultivated from subsurface environments has been one of the discoveries of using the 16S rRNA method for bioremediation (Lovely. 2001). This contradicts the general quandary in environmental microbiology, which is that recovering the most ecologically important organisms in culture can be challenging (Amann et al. 1995). For instance, there was a considerable enrichment in microorganisms with 16S rRNA sequences that were closely associated with those of previously grown *Geobacter* species in contaminated aquifers where microorganisms were oxidizing pollutants with the reduction of Fe(m) oxides (Rooney-varga et al. 1999., Snoeyenbos-West et al. 2000., Roling et al. 2001). This, together with the fact that *Geobacter* species can oxidize organic pollutants in pure culture by reducing Fe(III) oxide (Lovley et al. 1989), suggests that *Geobacter* species are significant in in situ contamination degradation. *Geobacter* species may also reduce soluble U(vi) to insoluble U(iv) and therefore remove uranium from polluted water (Lovley et al. 1991). When acetate was added to uranium-contaminated groundwater to promote the microbial reduction of U(vi), the number of *Geobacter* species increased by many magnitudes, contributing to up to 85% of the microbial population in the groundwater, according to 16S rRNA sequence analysis (Anderson et al. In Press, Holmes et al. 2002). 16S rRNA sequences that are 99% similar to the 16S rRNA sequence of a pure culture of the TCE degrader *Dehalococcoides*

methanogens were found in aquifers where the indigenous microbial population was degrading the solvent trichloroethene (TCE) (Fennell et al. 2001., Richardson et al. 2002., Hendrickson et al. 2002). Microorganisms having 16S rRNA sequences closely linked to NaphS2, an anaerobic naphthalene degrader accessible in pure culture, were especially enriched in marine sediments with high rates of anaerobic naphthalene breakdown (Hayes and Lovely. 2002). The number of organisms with 16S rRNA sequences that were more than 99 percent similar to the MTBE-degrading organism, strain PM-1, which is available in pure culture, had a close relationship with the potential for aerobic degradation of the fuel oxygenate methyl tert-butyl ether (MTBE) in groundwater (Hristova et al. 2003).

The main drawback of the 16S rRNA technique is that understanding the phylogeny of organisms involved in bioremediation does not always predict crucial elements of their physiology (Pace. 1997., Achenbach and Coates. 2000). Microorganisms with 16S rRNA sequences closely related to the TCE degrader *Dehalococcoides methanogens*, for example, can differ in the chlorinated compounds they can degrade (He et al. 2003., Bunge et al. 2003), and predicting which of these compounds an uncultured organism will degrade may not be possible based on 16S rRNA sequence analysis alone (Hendrickson et al. 2002). When no closely related species are accessible in pure culture, predicting physiology from phylogeny becomes considerably more challenging (Singh and Nagaraj. 2006).

9. Comparative analysis of omics in bioremediation

The complete study of whole-genome sequencing, based on an overarching analysis of transcriptomics and proteomics, is especially beneficial in understanding bioremediation-relevant microorganisms whose physiology has not been examined in detail. The degree of coverage of cellular mRNA and cellular proteins determines global gene expression using DNA microarray technology. In contrast, the full genome coverage reflects all of an organism's genes by definition. The dynamic range of cellular mRNA levels is not as great as that of encoded proteins (Gygi et al. 1999). Whole-genome arrays, rather just proteomic investigations, are thought to provide a considerably more thorough assessment of the real gene expression pattern (Singh and Nagaraj. 2006).

In light of global gene expression studies, both transcriptomics and proteomics endorse the idea that DNA array technologies capture gene expression variations more thoroughly than proteomics (Muffler et al. 2002., Kuhner et al. 2005., Eymann et al. 2002). As a result, genome data are required to complement the proteomics technique (Hegde et al. 2003). On the other hand, proteinomics would continue to play a key role in functional transcriptomics and/or genomics. The primary participants in an on-site microbial mineralization reaction are protein molecules rather than mRNAs; the latter is one of the very unstable transmitters on the way from the genes to the ribosome, while each protein molecule represents the final result of the gene expression (Kuhner et al. 2005). Comprehensive protein profiling offers information about the particular organism and the destiny and destination of protein molecules both within and outside the cell, which can only be identified using combined transcriptomics, proteomics, and interactomics methods (**Figure 3**).

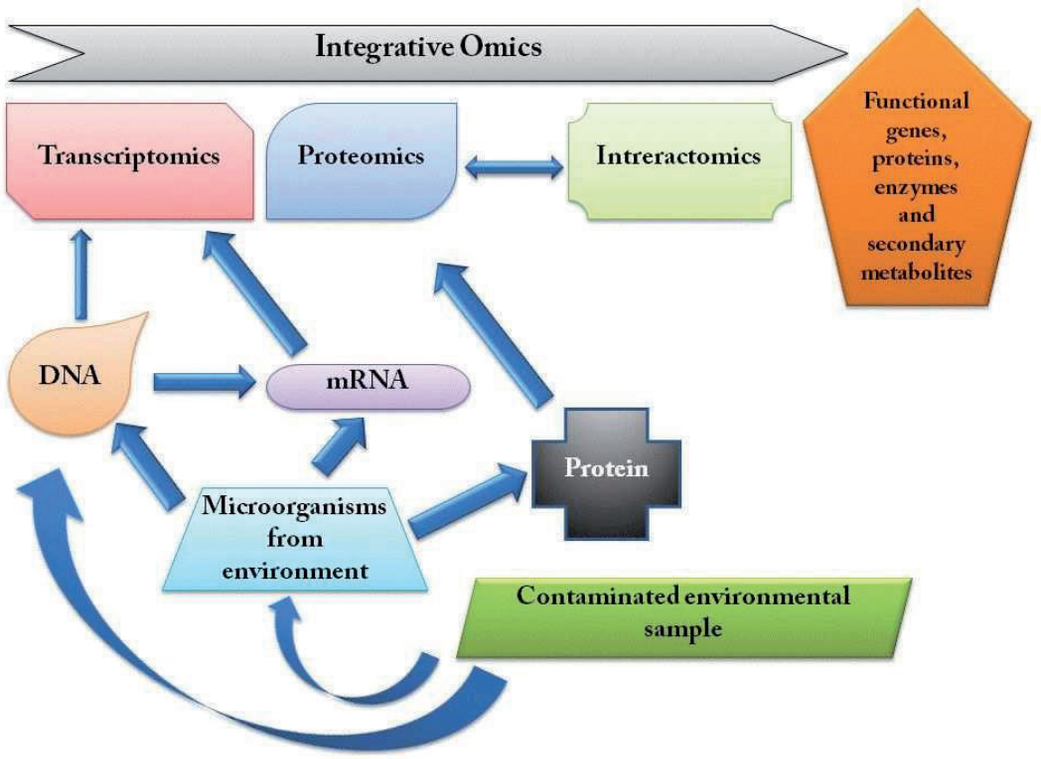

Figure 3. Insights related to bioremediation are tracked utilizing omics technologies and systematic biology approaches. Transcriptomics will use DNA taken directly from contaminated environmental locations and species (DNA microarrays). Transcriptomics will give way to proteomics, which will be followed by interactomics. We will be able to discover novel compounds of interest during the mineralization process by extracting protein from pure culture utilizing 2-DE and protein microarray systems.

10. Bioinformatics in bioremediation

MetaRouter is a system for storing and managing heterogeneous biodegradation data in a framework that allows for its administration and mining (application of methods for extracting new data). It is a program for laboratories that operate in this field and need to keep track of public and private data connected internally and externally and extract fresh information from it. The system is built on an open and modular design that can be customized to meet the needs of different clients. This client/server application, written in Postgre SQL (a relational database standard) and utilizing SRS as an indexing system (to link and query Molecular Biology databases), uses a client/server design to operate on the user station or the business server, allowing it to be securely accessed using a web browser from anywhere around the globe.

A database related to biocatalysts/biodegradation developed by The University of Minnesota (http://www.labmed.umn.edu/umbbd) is in its fifth year and has accomplished all of its objectives. It comprises 250 microbe entries and approximately 100 pathways for microbial catabolic metabolism of predominantly xenobiotic organic chemicals, with information on approximately 650 reactions, 600 substances, and 400 enzymes. It contains details on the most common forms of microbial catabolic responses as well as the organic functional groups that they change. It is poised to proceed beyond its initial objectives now that it has achieved

them. Mirror sites, fold prediction for its sequenced enzymes, closer linkages to genome and microbial strain databases, and the prediction of biodegradation routes for chemicals it does not include are all on the horizon (Ellis et al. 2000).

11. Approaches of systems biology

The emergence of genomic innovations and systems biology offers new ways to biological processes that are now uncontrollable and at the basis of major environmental issues. The biological destiny of the almost 8 operons implicated in this process is a complex problem in this regard. The University of Minnesota's biodegradation database identified thousands of novel chemical compounds widespread in current organic and industrial chemistry. A huge range of microbial strains can thrive on contaminants in the environment (approximately 800 today). Bioremediation has been investigated from a molecular biology perspective, identifying chemical processes and genes; the University of Minnesota has made a groundbreaking effort in compiling practically all of our existing information on biodegradation routes and designing systems that deal with specific data. However, the majority of information on microbial biodegradation of xenobiotics and refractory compounds in the literature focuses on duos consisting of one pollutant vs. one strain, leaving out important components of realistic situations such as gene exchange and metabolic cooperation. This community-based approach to genomes and 'functionomes' (as opposed to organism-based approaches) is leading to the sequencing of 'genomes' of communities and ecosystems rather than single species. These conditions highlight the necessity to classify and represent information in biodegradation databases to allow the total known biodegradative capacity of the microbial world to be matched with the entire collection of compounds known to be partially or completely biodegradable (Kitano 2002).

12. Conclusion

The application of omic sciences to bioremediation research is definitely in its early stages. Many technological challenges must be answered before some of the newer technologies, such as environmental genome sequencing and arrays, can be implemented. It will be essential to recover relevant organisms and research gene function in pure culture to elucidate the function of most genes retrieved from the environment. Microorganisms that are closely related to those found in some polluted environments are currently accessible in culture, and careful reproduction of environmental conditions during isolation will almost certainly yield more. Microorganisms that make up approximately a quarter of the marine microbial community but whose presence has previously been discovered only using 16S rRNA sequences and high-throughput culturing and screening technologies can substantially speed up the search for previously uncultured organisms.

Some novel molecular biology approaches, such as genetic engineering, transcriptomics, proteomics, and interactomics, hold a ton of potential as tools for studying the processes that

control mineralization pathways. Although the applications of these approaches are still in their blooming stage, the massive amounts of data created by today's genomics and proteomics technocrats require gradual organization among relevant databases. To comprehend the mineralization process in a significant way, techniques that integrate transcriptomics and proteomics data must be refined. These methods have considerable potential in predicting organism metabolism in polluted settings and predicting microbial-aided attenuation of pollutants to expedite bioremediation. Bioinformatics and computational biology innovations have been designed to detect and analyze numerous aspects of cells, including gene and protein activities, interactions, metabolic and regulatory processes, etc. IIn particular, analysis using bioinformatics tools will make it easier and faster to comprehend cellular processes to treat and regulate microbial cells as factories. Understanding molecular mechanisms and cellular manipulation using bioinformatics will focus on the coming decade.

Author details

Ranjith N. Kumavath* and Pratap Devarapalli

*Address all correspondence to: rnkumavath@gmail.com
and rnkumavath@cukerala.edu.in

Department of Genomic Sciences, School of Biological Sciences, Central University of Kerala, P.O. Central University, Kasaragod, India

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