Proc Indian Natn Sci Acad **85** No. 4 December 2019 pp. 963-983

DOI: 10.16943/ptinsa/2019/49720

Review Article

Microbe-Assisted Biodegradation, Bioremediation and Metabolic Engineering

HEMANT J PUROHIT[1,*], S DAYANANDA[2] and PHALE PRASHANT[3]
[1]*Environmental Biotechnology & Genomics Division, CSIR-National Environmental Engineering Research Institute, Nagpur*
[2]*School of Life Sciences, University of Hyderabad, Hyderabad, India*
[3]*Department of Biosciences and Bioengineering, IIT-Bombay, Powai, Mumbai, India*

(Received on 03 April 2019; Accepted on 05 November 2019)

We summarize here the recent research and development in the area of microbe assisted biodegradation and remediation processes by different research lab in the country. The various substituted hydrocarbons as pollutants have been studied by different groups. The depth of the study includes the characterization of key bacterial metabolic pathway for target pollutants through both genome and proteome levels. By understanding the pathway, in few cases the metabolic engineering approached have been demonstrated for the efficient utilization of pollutants. The better understanding to key enzyme were explored using protein modeling or by understanding its expression through heterologous host. The review also presents few case studies where pilot scale demonstration of contaminated sites with selected pollutants, have also been carried out effectively.

Keywords: Biodegradation, Bioremediation; Metabolic Engineering

Introduction

A wide range of aromatic compounds including solvents are being used in industries for the production of pesticides, dyes, plasticizer, paints, textiles, etc. The impact of these pollutants is very serious as these are capable of inducing endocrine disruption, genotoxic, mutagenic and carcinogenic effect on human health. The effects are more alarming due to long term persistence of these pollutants in the environment. Therefore, to overcome the effects, it is essential to remediate these compounds. The aromatics in the environment are eliminated by processes like volatilization, photooxidation, chemical oxidation, adsorption to soil matrix or biodegradation by microorganisms (bioremediation). Bioremediation is an environmentally "green" method of using microbes – either naturally-occurring or introduced (in naturally-occurring form or engineered) – to break down pollutants to non- toxic substances. Bioremediation can be *in situ* or *ex situ*. The biological processes for treating toxic pollutants are gaining interest over chemical and physical methods especially in terms of economic and environmental development; where instead of toxic by-products generated under non-biological methods, biological processes produce useful and relatively less toxic intermediates that enter the biogeochemical cycles. Bioremediation not only serves removal of contaminants but also has other positive aspects including enhancement of the soil quality, waste water treatment, carbon sequestration etc. However, bioremediation also encounters some limitations viz. toxicity of the compound, presence of simpler carbon source, salinity and other stressors. There are number of research groups working in the area of bioremediation and environmental engineering, which will help to clean up the environment. This report gives the recent advances in the field of microbe-assisted biodegradation and bioremediation and metabolic engineering in the country.

**Author for Correspondence: E-mail: hj_purohit@neeri.res.in*

Microbe-Assisted Biodegradation and Bioremediation

Some of the reported bioremediation technologies include natural attenuation, bioaugmentation, bioleaching, bioreactor, biostimulation, electro-kinetic mediated bioremediation and phytoremediation (Paliwal *et al.,* 2012, Pinjari *et al.,* 2013, Kundu *et al.,* 2014, Reddy *et al.,* 2016, Chatterjee *et al.,* 2017, Juwarkar 2012). The starting point for any bioremediation activity is the assessment of the inherent microflora at a contaminated niche. Based on their catabolic capacities, remediation processes can be designed. A large number of bacteria including *Pseudomonas*, *Alcaligens*, *Sphingomonas*, *Stenotrophomonas*, *Rhodococcus*, *Bacillus* (Qureshi *et al.,* 2007, Tripathi *et al.,* 2011, Verma *et al.,* 2011, Sreenivasulu *et al.,* 2012, Ghosh *et al.,* 2017) etc. have been reported for bioremediation of various contaminated sites.

One strategy of bioremediation is phyto-remediation that involves plant-microbe interaction and by which sustainable growth of plants in the contaminated sites could be restored. Microbes-soil-plant interactions for *in-situ* treatment of pollutants in contaminated sites have been reported as Microbe-Assisted-Phytoremediation (MAP) technology (Juwarkar, 2012). MAP technology has been reported to be ecofriendly, economic and effective for restoration of degraded land such as mines, spoil dumps (Juwarkar, 2012). The report has also illustrated the application of MAP technologies at sites where shallow contamination or pollution exist. It has been reported that, at shallow pollution sites, processes such as phytotransformation, rhizosphere bioremediation, phytostabilization, phytoextraction and/or rhizofiltration were best possible to be implemented (Juwarkar, 2012). Industries like textile mills, tanning- leather manufacturing industries, fertilizer factories, mining industries were major sources of contamination where phytoremediation technologies have been promising. To evaluate the potential applications of the plant-microbe interactions remedial strategies, studies have been performed by researchers (Niti *et al.,* 2013, Sarkar *et al.,* 2010).

Biodegradation and Bioremediation of Chloro/ Nitro/Amino Substituted Aromatic Compounds

Aromatic compounds namely chlorophenols, p-nitrophenol (PNP), aminophenols, chloronitrophenols and their derivatives are important environmental pollutants because of their toxicity to many living organisms (Arora and Jain 2012, Arora *et al.,* 2014, Kanekar *et al.,* 2014, Sengupta *et al.,* 2015, Kumari *et al.,* 2017).

Both aerobic and anaerobic biodegradation pathways of aromatic compounds and their derivatives have been deciphered by researchers (Arora *et al.,* 2014, Malik *et al.,* 2014, Tiwari *et al.,* 2017, Ghosh *et al.,* 2017); more extensively the aerobic degradation. In bacteria, major pathways reported for catabolism of aromatic compounds have revealed that initial conversion steps were carried out by different enzymes, but the compounds were transformed into a limited number of metabolites, such as protocatechuate and catechols. These intermediate metabolites were channeled into central metabolic routes via; different ring cleavage pathways enzymes. This generalized scheme of catabolic pathways for aromatic compounds suggests that microorganisms have extended their substrate range by developing "peripheral" enzymes, which were able to transform initial substrates into one of the central intermediates like catechol, resorcinol, benzoquinones etc.

Arora *et al.* 2014 have reported about bacterial degradation of the toxic compound 4-chloro-3-nitrophenol (4C3NP) and formation of 4-chlororesorcinol intermediate in the degradation pathway. 2-chloro-4-aminophenol degradation by bacteria has also been reported by the same group (Arora *et al.,* 2014, Malik *et al.,* 2014). 4C3NP-mineralizing bacterium, *Pseudomonas* sp. JHN was isolated, by enrichment, from a wastewater sample collected from a chemically-contaminated area in India. Detoxification of 2-chloro-4-nitrophenol (2C4NP) by *Cupriavidus* strain a3 has been reported by Tiwari *et al.* 2017. These microbes show potential for application in bioremediation. Microbial Biotechnology Research Laboratory (MBRL) reported a bacterium isolated from limestone deposits in Ukhrul, Manipur, India; two strains, (HS4-2 and HS6-1) were isolated as p-nitrophenol (PNP) degraders via hydroquinone and –nitrocatechol pathways (Ningthoujam *et al.,* 2012). In PNP degradative pathway, 4-nitocatechol and benzenetriol intermediates were mainly formed in Gram negative organism (Chauhan *et al.,* 2010, Qureshi *et al.,* 2002,

Qureshi *et al.,* 2001). Enzyme p-nitrophenol 4-hydroxylase has been reported to be active for parathion biotransformation and further degradation of PNP (Khan *et al.,* 2011, Verma *et al.,* 2011, Wasi *et al.,* 2011, Saha *et al.,* 2014, Goyal and Bansiwal, 2017). Polyphenol oxidase (PPO) enzyme for phenolic contaminants from industrial wastewater has been reported (Mukherjee *et al.,* 2013). Other enzymes such as catechol 1,2-dioxygenase (C12O) and catechol 2.3-dioxygenase (C23O) have been reported to enhance bioremediation process under different conditions (Pandeeti and Siddavattam, 2011, Kumari *et al.,* 2017, Ghosh *et al.,* 2017). Nitroaromatic pollutants trinitrotoluene (TNT) and dinitrotoluene (DNT) degradation has mainly been reported under aerobic conditions (Goel *et al.,* 2012). Also, other pollutants such as 4-nitrobenzoate, 2-nitrobenzoate, 2-nitrotoluene, nitrobenzene, p-nitroaniline and p-nitrophenol were reported to be used as growth substrates by bacteria (Mulla *et al.,* 2011, Srivastava *et al.,* 2013, Qureshi *et al.,* 2001, Qureshi *et al.,* 2002, Mishra and Sardar, 2014, Arora and Sharma, 2015). Under anaerobic condition, nitroaromatic pollutant reduction and biotransformation have been reported by bacteria and their nanobiocomposite as catalyst (Srivastava *et al.,* 2013, Chaudhary *et al.,* 2013).

Biodegradation and Bioremediation of Organochlorines and Organophosphates

Organochlorines viz., DDT- type compounds, cyclodienes like aldrin, dialdrin, endrin, heptachlor, chlordane, endosulphan, lindane, polychlorinated biphenyls etc, organophosphates such as methyl parathion and many chlorinated/nitro/amino-substituted compounds employed as insecticides and pesticides have also been categorized as pollutants. Recently it has been assessed in the Vasai Creek water near Mumbai that organochlorine and organophosphorus pesticide residues were major cause of endocrine disruption leading to ecological risk (Singare, 2016).

Over the past 20 years of organochlorine insecticide lindane production the industry has utilized inappropriate storage and disposal practices, resulting in widespread environmental hexachlorocyclohexane (HCH) contamination and a large number of HCH dumpsites (Jit *et al.,* 2011). One of the HCH dumpsites at Ummari village, Lucknow has been studied for characterization of the microbial diversity by culture dependent approach (Singh *et al.,* 2009, Dadhwal *et al.,* 2009, Bala *et al.,* 2010, Sharma *et al.,* 2010, Kumar *et al.,* 2013, Negi *et al.,* 2014, Verma *et al.,* 2014). These studies revealed the presence of potent HCH degraders and tolerant bacterial species that arose under the HCH stress. Specifically, many of these bacteria shared components of a unique catabolic system –named the *lin* system which allowed for the degradation of HCH isomers (Kumari *et al.,* 2002, Sharma *et al.,* 2006, Lal *et al.,* 2010). These *lin* genes seem to have played a major role in shaping the degradation of xenobiotic compounds (Dogra *et al.,* 2004) and are diverging to form several catabolic functions (Lal *et al.,* 2006). *linA* (HCH dehydrochlorinase) and linB (halidohydrolase) act as the primary enzymes in the pathway. The *linA* GC content is lower than that of the linXBCD genes (Suar *et al.,* 2005) linB acts on β-HCH, δ-HCH and pentachlorocyclohexanol, yielding a 2,3,4,5,6-pentachlorocyclohexanol (2,3,4,5,6-PCHL), and tetrachlorocyclo hexanediol, respectively (Sharma *et al.,* 2006). LinB protein has a very broad substrate preference for halogenated compounds whereas the substrate range for LinA protein is very restricted to α-, γ- and δ-HCH and their corresponding PCCH products (Kumari *et al.,* 2002). It is due to HCH pressure that LinA proteins are evolving to a greater extent at the dumpsite. LinB, which is evolutionary more advanced and evolved, is evenly distributed at all the contaminated sites (Kumari *et al.,* 2002, Sharma *et al.,* 2006). It has been found that the upper pathway *lin* genes are constitutively expressed whereas the lower pathway genes are primarily induced in the presence of α- and γ-HCH not β- and δ-HCH (Suar *et al.,* 2004). The high residue level is not the only factor: other biological factors like HGT by IS elements and plasmids may be responsible for the high copy number. Bioremediation of HCH contaminated sites is being attempted by bioaugmentation, biostimulation and enzymatic bioremediation approaches (Lal *et al.,* 2010). Commonly used pesticides namely chloropyrifos have been reported to be utilized by Pseudomonas strain C2A1 as the sole source of carbon and energy (Anwar *et al.,* 2009, Gupta *et al.,*2016). Degradation of chlorpyrifos have been demonstrated in soil suspension by Das and Adhya 2015. Also reports showed that the capability of microorganism for degradation could be enhanced by characterization and transfer of

pesticide-degrading genes, induction of catabolic pathways and display of cell surface enzymes (Hussain *et al.,* 2009). Recently researchers are trying to explore Pseudomonas sp to degrade chlorinated pesticides belonging to different chemical categories (Gupta *et al.,* 2016). Endosulphan degradative pathways and the generated metabolites have also been reported (Bajaj *et al.,* 2010).

Many reports are available on organophosphate (OP) pesticides biodegradation by bacteria (Qureshi *et al.,* 2009; Dubey and Fulekar 2012; Sasikala *et al.,* 2012; Iyer *et al.,* 2013; Pinjari *et al.,* 2013; Kundu *et al.,* 2014; Reddy *et al.,* 2016; Chatterjee *et al.,* 2017). Even, several novel bacterial strains have been reported from agricultural field especially located at Narigram in Burdwan district of West Bengal, India. Bacillus aryabhattai, a novel endospore forming strain have been reported to tolerate high concentration (up to 500 $\mu g\ mL^{-1}$) of pesticide parathion (Pailan *et al.,* 2015). Study on bioremediation of methyl parathion using *Pseudomonas* sp have also been reported from Visakhapatnam, India (Mulla *et al.,* 2011, Srivastava *et al.,* 2013, Begum and Arundhati, 2016). OP degrading enzymes have been designated as Phosphotriesterases (PTEs) as they are involved in hydrolysis of triester linkage found in OP compounds. The PTEs are classified into three major groups, i.e. organophosphate hydrolases (OPH), methyl parathion hydrolases (MPH) and organophosphate acid anhydrases (OPAA). The OPAA is later shown to be dipeptidase and its activity on OP compounds is shown due to structural similarities between OP compounds and dipeptides having proline at the C-terminus. However, the physiological substrates for OPH and MPH are unknown. Considering the catalytic efficiency and structural similarities with the structure of quorum quenching lactonases, the OPH, the product of organophosphate degrading (*opd*) gene, is assumed to have evolved from phophotriesterase like lactonases (PTLL). The genetic organization of *opd* gene in both *Flavobacterium* sp. and in *Pseudomonas diminuta* have been elucidated. These two strains have been reclassified and renamed as *Sphingobium fuliginis* ATCC 27551 and *Sphingophyxis wildii* (Parthasarathy *et al.,* 2016a). In these two soil isolates the *opd* gene is found in large indigenous plasmids. The 65 kb plasmid pCMS1, isolated from *Sphingopyxis wildii* has been shown to be a self-transmissible plasmid and is involved in spreading *opd* information among soil microflora (Pandeeti *et al.,* 2011). Similarly, the plasmid pPLD2 is shown to be an Integrated Mobilizable Element (IME) (Pandeeti *et al.,* 2013). In plasmid pPDL2, the *opd* along with an ORF, orf306 is flanked by mobile genetic elements (Pandeeti *et al.,* 2013). The Orf306 is an esterase. When it is expressed it E. coli, the cells have started degrading p-nitrophenol, the recalcitrant degradation product generated during OPH mediated *hydrolysis* of OP insecticides. Transcriptome analysis for the *E. coli* strains grown in presence and absence of Orf306. The Orf306, in an unknown way, is shown to down regulated glycolysis and TCA cycle and upregulated both *hca* and *mhp* operons involved in degradation of phenylpropionate and hydroxylphenyl propionate. The enzymes encoded by hca and mhp operons are shown to degrade p-nitrophenol (Chakka *et al.,* 2015). The IME borne *opd* product (OPH) along with Orf306 is shown to contribute for mineralization of insecticide residues found in soil by stimulating the innate nitrophenol degrading ability of recipient strains. The work done has also shown OPH association with membrane associated transport system involved in phosphate transport. The genetic and biochemical evidences suggest that OPH is involved in phosphate acquisition in soil bacteria (Parthasarathy *et al.,* 2016b).

Biodegradation and Bioremediation of Hydrocarbons and Crude Oil

Crude oil and hydrocarbons comprises of saturates/ paraffins, aromatics, resins and asphaltenes. Scientific communities are being urged to develop strategies to minimize the concentrations polyaromatic hydrocarbons (PAHs) from the Gujrat coastlines and restore sites by investigations (Dudhagara *et al.,* 2016). Reports are available consistently on metabolic diversity and versatility of pure cultures and mixed cultures respectively to utilize hydrocarbon pollutants in petroleum crude as carbon source (Varjani and Upasani, 2016). It has been reported that even co-culture(s) of bacteria and fungi enhance degradation rates of diesel oil and PAHs under laboratory conditions (Varjani and Upasani, 2013).

Bioremediation technologies have been reported to be applied to restore petroleum hydrocarbon polluted environments that makes use of natural microbial

biodegradation activity (Varjani, 2017). Both in-situ and on-site treatment process have been reported to breakdown crude petroleum oil components at contaminated sites. In 2007, Das and Mukherjee have studied and compared the efficiency of thermophilic *Bacillus subtilis* DM04 strain and *Pseudomonas aeruginosa* M bacteria isolated from North East India for biodegradation of crude petroleum oil.

Bacillus spp. and *Pseudomonas* have been found suitable for practical field applications for effective PAH and aromatic compounds bioremediation at contaminated sites (Khanna *et al.,* 2011, Sood *et al.,* 2010, Phale *et al.,* 2013, Phale *et al.,* 2007). Even high-altitude bacteria have been reported to be capable of degrading engine oil (Jain *et al.,* 2010). Recently, *B. licheniformis* CFR1 is reported as an excellent candidate for bioremediation and detoxification of aflatoxin (Rao *et al.,* 2017). Novel, thermally stable, halotolerant enzymes like cellulases have been reported to perform in harsh conditions such as high salt, heat and acidic environments (Chandel *et al.,* 2007, Garg *et al.,* 2016).

Bacterial community found in an Indian coal bed has been reported to be capable of in-situ biotransformation of coal into methane (Singh *et al.,* 2012, Singh and Tripathi 2013, Mohanty *et al.,* 2016). Such findings would assist in bioremediation of such contaminated ecosystems.

Recently it has been reported that oleophilic microorganisms could be used for oil spill bioremediation (Varjani, 2017). Scientific communities are being urged to develop strategies to minimize the concentrations of pollutants for examples PAHs from the Gujrat coastlines and restore sites by investigations (Dudhagara *et al.,* 2016). In the similar lines, there is a big break through achieved, in the area of remediation of crude oil contaminated sites, where the technology "Oilzapper" has been successfully demonstrated at commercial scale at various site and received global acceptance (*Sharma et al*., 2019).

Biodegradation and Bioremediation of Metals and Heavy Metals

Toxicity of metals to ecosystems and their mobility in biological systems needs to be highlighted in Indian scenario. In 2014 Mani and Kumar have reviewed the bioremediation options especially using microorganisms and plants with eco-friendly and sustainable approach in Indian context. Reports have been available for metal binding proteins or peptides for their potential towards tolerance and accumulation in living systems especially with cadmium which is found significantly in foods (Mejare and Bulow, 2001). Molecular level studies of metal tolerance genes in microorganisms and hyper accumulator plants showed involvement of functional genes for detoxification (Dixit *et al.,* 2015). Also, bioabsorbent such as fungi viz; *Phanerochaete chrysosporium*, *Aspegillus awamori*, *Aspergillus flavus*, *Trichoderma viride* are reported for removal of heavy metals from industrial effluent by Joshi *et al.,* 2011. Fungi such as Tirchoderma with plant growth promoting (PGP) abilities have been reported as efficient biocatalyst and potent antimicrobial agents (Mishra *et al.,* 2014, Monaharachary *et al.,* 2014, Singh *et al.,* 2014). Removal of metal(loids) using organic amendments such as compost, MSW as nutrients and soil conditioners have been discussed with practical implications to bioavailability by Park *et al.,* 2011. Role of EPS from various sources with special emphasis on bioremediation of heavy metals have been studied by researchers (Pal and Paul 2008, Ahemad 2012). The applicability of chromium resistant fungal species *viz., Micrococcus* sp and *Aspergillus* sp for removal of chromium and nickel up to 10,0000 mg/L have been reported (Congeevaram *et al.,* 2007). The potential of bacterial genera (*Pseudomonas, Entetobacter, Bacillus, E.coli*), yeast and fungal species for bioremediation of metal toxicity have been discussed and their impact on environment is also illustrated by Ray and Ray 2009. The mechanism of removal of metal by biological means have been reported where it was found that Pi precipitates the heavy metals as phosphate-metal complexes (Chaudhari *et al.,* 2017). Bioremediation of electroplating industrial wastewater with simultaneous nanoparticles synthesis also have been reported (Sathyavathi *et al.,* 2014, Dasgupta *et al.,* 2015). Several biological methods of heavy metal remediation have been reported by both bacteria and fungi (Kundu and Gupta 2007, Singh and Prasad 2015, Chaudhary *et al.,* 2017, Nagvenkar and Ramaiah 2010).

Treatment of Textiles Dyes

Ichalkaranji and nearby places in Maharashtra are

well known as the textile hub of the state. A large number of large, small and even household dye processors are located in this part. Because of the uncontrolled discharge of the toxic textile dye-stuff in environmental sink, water bodies and soils in the said area have become heavily polluted. The Department of Biochemistry, Shivaji University research group lead by Prof. S. P. Govindwar has been actively involved to find out novel and developed ecofriendly bioremediation strategies for the treatment of these noxious dyes.

In the last five years, a number of bacteria such as *Lysinibacillus* sp. RGS, *Providencia rettgeri*strain HSL1, *Pseudomonas* sp. SUK1, *Bacillus thuringiensis*, *Pseudomonas* sp. LBC1, *Pseudomonas monteilii* ANK, *Pseudomonas putida* PgH, *Brevibacillus laterosporus*, *Bacillus cereus* EBT1 and *Kocuria rosea* MTCC 1532 etc. have been explored for the treatment of textile dyes. Fungal species namely, *Galactomyces geotrichum* MTCC 1360 and *Aspergillus ochraceus* NCIM-1146 were also utilized for detoxification of dyes (Saratale *et al.,* 2011). Novel lichen species called *Permeliaper lata* was also found to treat the textile dyes to significant extents (Kulkarni *et al.,* 2014). Combinatorial systems of bacteria and fungi such as *Galactomyces geotrichum* MTCC 1360-*Brevibacillus laterosporus* MTCC 2298 and *Aspergillus ochraceus* NCIM-1146-*Pseudomonas* sp. SUK1 was observed to be more efficient than the individual cultures in treatment and decolorization of dyes. Of late, plant and bacterial hybrid systems of *Zinnia angustifolia-Exiguobacterium aestaurii*, *Portulaca grandiflora-Pseudomonas putida* and *Pogonatherum crinitum-Bacillus pumilus* were also explored in the constructed wetlands systems for efficient removal of textile dyes from real effluents (, Khandare and Govindwar, 2015). A novel bioreactor with *Ipomoea hederifolia* adventitious roots and its endophyte *Cladosporium cladosporioides* was developed for textile dye degradation (Patil *et al.,* 2016). A variety of bioreactors using bacteria were also developed for the treatment of dyes from effluents. The decolorization of real textile industry effluent using immobilized bacterial yeast consortium of *Galactomyces geotrichum* MTCC-*Brevibacillus laterosporus* MTCC in a newly developed triple layered fixed bed bioreactor and the efficiency and performance was tested in continuous operation (Kurade *et al.,* 2017). Degradation and detoxification of methylene blue dye adsorbed on water hyacinth in semi continuous anaerobic-aerobic bioreactors was also carried out in a novel microbial consortium of *Sacharromyces cerevisiae* and *Bacillus* sp. STIS (Bedekar *et al.,* 2015). Solid state fermentation of the bioadsorbed textile dyes stuffs and distillery industry waste-yeast biomass has been successfully carried out using *Bacillus cereus* EBT1 (4, Kadam *et al.,* 2013). In independent studies, influence of diazo dyes on soil bacterial population was monitored. Molecular analysis of shift in community after different time intervals was monitored by using PCR-DGGE targeting V3 region of bacterial 16S rRNA gene. UV irradiation was used to introduce random mutations in *Pseudomonas* sp. LBC1. Genetic alterations induced by UV irradiation in selected mutant bacteria were confirmed by random amplification of polymorphic DNA technique. The mutant bacteria named as *Pseudomonas* sp. The dye treatment efficiency of the mutant was found to be improved after mutagenesis (Joshi *et al.,* 2013). Different oxido-reductive microbial enzymes like lignin peroxidase, manganese peroxidase, aryl alcohol oxidase, laccases, tyrosinase, azo reductase, malachite green reductase, DCIP reductase and riboflavin reductase attacks the complex dyes structure and metabolize them through steps like deamination, desulfonation, dehalogenation, hydroxylation, oxidation, reduction, ring opening and cleavage mechanisms. The direct roles of these enzymes have been proved using standard purification protocols (Telke *et al.,* 2015). The metabolites of dye degradation were analyzed by using UV-Visible and FTI-R spectroscopy, HPLC, HPTLC, GC and/or LC-MS and NMR techniques. Textile effluents generally have very high COD, BOD, TOC, TDS, TSS, alkalinity, conductivity etc., therefore the microbes which can only survive these stressful conditions could only show dye removal efficacies. The toxicity assessment of dyes, effluents and their metabolites after bioremediation have been carried out using plant, microbes and animal models looking for cytotoxicity and genotoxicity. An intensive research on utilization of bacteria at the large industrial scales is underway and an efficient reactor set up is still awaited.

Effluents generated from the textile mills are one of the major sources of point pollution in rivers. In view of the requirement of color free discharge and detoxified waters, several lines of treatment have

been tested. These include conventional physical as well as advanced biological oxidation processes such as treatment with TiO_2, UV, Fenton and photo-Fenton oxidations. Many of these do not work effectively on complex azo and pthalocyanine dyes. Laccases and several other oxidoreductases have been identified as promising candidates for decolouration as well as detoxification. The group of Prof. Saroj Mishra uses laccase from *Cyathusbulleri* (Salony *et al.,* 2006) effectively for decolouration of several category of dyes. The reaction centre of this enzyme is made from closely spaced Cu atoms arranged in the form of three reaction centres, namely T1, T2 and T3. Out of these, the geometry at the T1 Cu centre determines the redox potential of the enzyme and its ability to oxidize dyes of high redox potential (Salony *et al.,* 2008). The property of laccase to act on complex dyes as well as real effluent can be further augmented by addition of small mediators in the reaction mixture, protein engineering, supplementation with other powerful oxidoreductase enzymes such as cellobiose dehydrogenases or by producing laccase on complex lignocellulosics such as wheat bran. These approaches have been successfully used in the lab. Amongst a number of mediators, 2,2'-azino-bis (3-ethyl-benzothiazoline-6-sulphonic acid) or ABTS and natural mediator vanillin were found to be very effective (Chhabra *et al.,* 2008) against a wide range of dyes as well as combined effluent collected from textile mills. Based on these findings, a continuous membrane reactor was designed and successfully operated for decolouration of real effluent (Chhabra *et al.,* 2009) using laccase and a synthetic mediator. The treatment with laccase was extended to complex effluents by combining a chemical step (alum treatment) during the early processing stage (Chhabra *et al.,* 2015a). Operationally, the system was equally effective in handling a variety of Reactive dyes (Chhabra *et al.,* 2015b). In the second approach, random mutagenesis was carried out of the C-terminal half of the coding gene (Garg *et al.,* 2012; Kenzom *et al.,* 2014) and mutant library (Kenzom *et al.,* 2015) screened for catalytically superior enzymes. Many of these were effective on pthalocyanine dyes as well. Through biochemical and model building, amino acid residues were identified that affect catalytic activity. (In another approach, combination of laccase with cellobiose dehydrogenase was found to be highly active on pthalocyanine dyes (Bashir *et al.,* 2015; Gangwar *et al.,* 2016) with extensive degradation noticed of this category of dyes. The treatment on real effluent indicated this combination of enzymes (wild type and the mutant ones) to be effective for both decolouration and reduction in phytotoxicity. Several other mutants available in the author's laboratory are being analysed to identify sites in the enzyme that determine the redox potential as well as the electron transfer property of laccase. The efficacy of laccases produced on complex lignocellulosics was recently demonstrated on complex effluent from denim dying industry (Vats and Mishra, 2017) indicating these to be promising in treatment of textile wastewaters.

Bioremediation of Soil Salinity

Papers published in Nature in 1950s claimed saline soil reclamation using native populations of cyanobacteria, based on which ICAR advocated use of these microbes for such purpose. Dr. Apte's laboratory demonstrated that actually sodium exclusion and its entrapment in extracellular polysaccharides underlie cyanobacterial tolerance to salinity (Apte and Thomas, 1986). While this provides temporary relief to a growing crop by transient sequestration of sodium/chloride ions and by supplementing fixed nitrogen, the technology offers no permanent solution to the problem of soil salinity which is restored subsequent to death and decay of cyanobacteria (Apte and Thomas, 1997). The work also revealed entirely different mechanisms to be involved in salinity and desiccation tolerance (Apte, 2001). Agronomic practices which curtailed sodium influx enhanced salt tolerance (Apte *et al.,* 1987). Desiccation tolerance, in contrast, depended on de novo synthesis of osmotic stress proteins (Fernandes *et al.,* 2000).

Integrating Conventional Bioremediation Technology with Genomics

Over the years it has been learnt that for successful implementation of biological remediation technologies, multidisciplinary approach from experts of various areas are required. Moreover, for effective bioremediation, complete understandings of environment (i.e. physical, chemical and biological properties) to be treated are essential. With the advancement in the technology, our lab has integrated the conventional approach with genomics-driven strategies to make bioremediation technologies more

potent. We perused the hypothesis that microbes are the best answer for counteracting the devastating consequences of our activities on environment. And to demonstrate the viability of our proposition, we have developed and enriched more than 100 bacterial consortia (mixed cultures) and isolated bacterial pure cultures from polluted soil, sediments, water, industrial and CETP effluents exhibiting significant degradation potential for various dyes and dye intermediates, aromatics, polyaromatic hydrocarbons (PAH), distillery spent wash and detoxification of heavy metals under different environmental conditions (anaerobic, microaerophilic, aerobic, sequential microaerophilic-aerobic and anaerobic-microaerophilic) (Patel *et al.,* 2012a and b; Jain *et al.,* 2012; Shah and Madamwar, 2013a; Patel and Madamwar, 2013; Balapure *et al.,* 2014; Chattaraj *et al.,* 2016a; Shah *et al.,* 2016a). The flask scale results were reevaluated during scaling up process in various bioreactors (anaerobic biphasic fixed film, sequential anoxic-oxic batch process, sequential anaerobic-microaerophilic process, periodic discontinuous batch operation) and processes were optimized for simulated and real industrial effluents (Acharya *et al.,* 2011; Shah *et al.,* 2016b; Chattaraj *et al.,* 2016b; Venkata *et al.,* 2012; Balapure *et al.,* 2016). It is imperative to know the degradation mechanism of the xenobiotic compounds to make technology more robust. Our studies show that due to consorted metabolic activities of the consortia complex aromatic compounds such as dyes, PAH are gradually degraded into simpler intermediates through generating different intermediates and finally led to the complete mineralization via TCA cycle. With two different consortia the degradation pathways for degradation of reactive azo dye Remazol Brilliant Violet 5R (i.e. Reactive Violet 5R) was proposed and observed that the dye was metabolized through formation of â-naphthol and benzene sulphoate (Jain *et al.,* 2012; Shah *et al.,* 2016a). In PAH degradation (phenanthrene), the study revealed that the compound was degraded through phthalic acid-protocatechuate acid pathway (Patel and Madamwar, 2013). During azo dye degradation the initial step is the reduction of azo bonds (–N=N–), catalyzed by azoreductase. One of such azoreductases was cloned from consortia V9 of 537 bp containing an ORF of 178 amino acids showing optimum activity in the presence of NADPH. The host harboring azoreductase clone showed 90% azoreduction within 7 min in cell free extracts (Shah and Madamwar, 2013a). On the course of developing feasible bioremediation strategy, whole genome of three bacterial cultures, *Paenibacillus* sp. strain DMB20, *Achromobacter* sp. strain DMS1 and *Paenibacillus* sp. strain DMB5 (showing <97% similarity of phylogenetic marker with available database) was sequenced. Annotation of these genomes revealed the presence of various genes required for aromatic degradation, antibiotic and heavy metal resistance, towards stress response in all three strains, which signifies their active role in xenobiosis (Shah *et al.,* 2015; Amin *et al.,* 2015; Johnson *et al.,* 2016). The complete degradation pathway for benzonitrile, benzamide, benzoate, and catechol degradation was mapped in *Paenibacillus* sp. strain DMB5 (Johnson *et al.,* 2016). Besides whole genome, we have sequenced more than 10 metagenomes from different polluted sites across the 'Golden Industrial Corridor' of Gujarat State, corresponding to about 8 Gb base pairs. Nearly 100,000 reads corresponding to >300 enzymes and enzymatic pathways potentially involved in biodegradation of xenobiotic compounds were mapped. Complete and partial degradation pathways for several xenobiotic compounds were mapped (Shah *et al.,* 2103b). The diversity community shift due to persistent pollution at certain polluted sites was also evidently observed (Patel *et al.,* 2014, 2015). The work describes the potential of bacterial system to utilize the recalcitrant xenobiotics and community organization at polluted environment along with their functional capabilities by knowing their genetic composition. Nonetheless, it is a preliminary study for complete understanding of entire ecology/ ecosystem of polluted environment to improve the natural bioremediation.

Consortium Development in Bioremediation

Remediation of contaminated resources is one of the major thrust areas of the National Environmental Engineering Research Institute (NEERI). Since, bioprocesses play key role in remediation, especially when the contamination is of organic nature, major thrust was taken in the area of optimization of such processes (Purohit *et al.,* 2003a, Purohit *et al.,* 2003b, Moharikar *et al.,* 2005, Kapley and Purohit, 2009). Bioremediation processes are employed either for bio-stimulation or for bio-augmentation (Khardenavis *et al.,* 2008, Khardenavis *et al.,* 2010, Paliwal *et al.,* 2012, Purohit *et al.,* 2016). The strategy involved in

this program is to enrich and select bacteria, which have the potential to utilize target pollutants as carbon source for their growth. The selected pollutants were hydrocarbon, -nitro and -chloro substituted pesticides and intermediates used for synthesis of pesticides and pharma products. The studies over a period of 10 years a bank of isolates was established from different environment with wide catabolic potential (Selvakumaran *et al.,* 2008; Thangraj *et al.,* 2008, Verma *et al.,* 2011). Since, most of these contaminated sites show a multi-substrate challenge, the defined mixtures of bacteria with selected genotype were used to design consortia with flexible degradative capacities (Selvakumaran *et al.,* 2011).

The pollutant targeted in the program included the hydrocarbons from crude oil (aromatics and long chain aliphatic), nitro and -chloro substituted aromatics or heterocyclics. Enrichment techniques were designed to isolate for these difficult to degrade molecules. One of the major achievements was the development of a consortium of four bacteria, which could emulsify and degrade the crude oil efficiently in 12 h. These cultures were used for development of wastewater treatment process (Moharikar *et al.,* 2005, Kapley *et al.,* 2007, Domde *et al.,* 2007). Another area of application was the remediation of contaminated soil. Initially controlled microcosm experiments were planned using designed bacterial consortia, which were further evaluated using pilot scale field trials; and demonstrated to user agencies (Qureshi *et al.,* 2009). Different bioremediation processes were designed either by using bio-stimulation (balancing of nutrients or induction of degradative capacities using inducer molecules) or by bio-augmentation (applying the required bacteria to bring out the degradation). These studies were supported by characterization of the associated genotypes that were predicted by annotation of genome sequence data and revealed the basic mechanism of degradation pathways for hydrocarbons, p-nitrophenol and atrazine (Tikharia *et al.,* 2016).

The biostimulation to enhance the expression of degradative capacities was carried out using the intermediate of the metabolism such as catechol, salicylate and even phenol and analyzed for different aromatic ring cleavage enzymes (Selvakaumaran *et al.,* 2011). Similarly, the metal ions were also used while the sludge was recycled for enhancing the required catabolic potential to microbial community. The other issues is for difficult to degrade wastewaters such as dye containing waste for that consortiums has been prepared (Dafale *et al.,* 2010) and also to solve the issue of generated increase in ammonia level has been dealt with specialized organisms (Khardenavis *et al.,* 2007; Pal *et al.,* 2015).

Engineering Microbes Towards Bioremediation

Biodegradative potential of degraders can overlap with bacteria from unrelated genera, or alternatively, genes with completely different sequences can catalyze the same reaction. Understanding these complex reactions requires a multi-dimensional approach of whole genome sequencing and bioinformatics analysis of sequence data (Sagarkar *et al.,* 2016). Next generation sequencing technologies allows us to explore the complete catabolic profile of the isolate and this tool has been used to identify many multi-substrate degrading bacteria used in soil microcosm/mesocosm studies. Taking this further into in situ bioremediation of contaminated soil is more complex. Environmental scenarios very often involve more than one contaminant and the microbial community containing different metabolic pathways is governed by diverse soil types and characteristics, climate conditions and anthropogenic activities. These shortcomings are now being addressed via the systems biology approach that analyzes interactions between microbes and their environment and helps predict their function and survival (Chakraborty *et al.,* 2012).

Mapping the microbial populations of a contaminated niche allows for a systematic development of a bioremediation strategy. The metagenomics tool provides insight into the phylogenetic composition of the microbial community as well as its functional genes (Jadega *et al.,* 2014; Yadav *et al.,* 2014). Shifts in microbial community of a wastewater treatment plant have been demonstrated to correlate with performance efficiency (Kapley *et al.,* 2015). Besides analyzing the community function, the new tools generate information on the structure and function of catabolic enzymes, the actual unit carrying out the biodegradation process. Knowledge of their detailed structure offers options for improvement by site directed mutagenesis or cloning

that can improve the efficiency of the reaction or provide the degradative capacity where it did not exist (Bassalo *et al.,* 2016). More recently, researchers have engineered metabolic pathways in micro-organisms to generate engineered biocatalysts. Despite the evolution of tools and increased knowledge database, the field application of bioremediation progresses slowly and more studies need to be directed towards the survival and performance efficiency of target microbes in field conditions.

Engineering Cyanobacteria for Enhanced Nitrogen Biofertilizer Potential in Stressful Agricultural Environments

Discoveries in Dr. Apte's laboratory that stress-responsive gene expression in cyanobacteria has a generic stress response and a stress-specific response (Apte and Bhagwat, 1989; Bhagwat and Apte, 1989; Alahari *et al.,* 2001; Alahari and Apte, 2004), led to discoveries of genes important for survival under major agricultural stresses, such as salinity and heat-shock (Rajaram and Apte, 2003 and 2010; Rajaram *et al.,* 2014), both of which diminish nitrogen fixation. Several of these genes were cloned using the versatile approach of differential RNA hybridization developed by him (Apte and Haselkorn, 1990) or by cloning of stress-specific genes (Rajaram *et al.,* 2001). Mutagenesis and/or overexpression of these genes established their distinct role in general/specific stress tolerance (Chaurasia and Apte, 2009; Rajaram and Apte, 2008). The genes were engineered in to nitrogen-fixing *Anabaena* strains and expressed from strong light-inducible *psbA1* promoter to further enhance the inherent stress tolerance of these strains. An eco-friendly, integrative expression vector was developed for *Anabaena* for the first time and used to construct recombinant strains with enhanced nitrogen biofertilizer potential in stressful environments (Chaurasia and Apte, 2011).

Engineering Cyanobacteria For Oxidative Stress Tolerance

Realization from Dr. Apte's work that oxidative stress is central to all the abiotic stresses, led to the identification of oxidative stress responsive genes and proteins using genomic, transcriptomic and proteomic approaches, in three *Anabaena* strains typical of Indian paddy fields (Panda *et al.,* 2014 and 2015). Biology of several oxidative stress alleviators, such as MnSOD, FeSOD, Mn-Catalases, and 4 peroxiredoxins unique to cyanobacteria was elucidated from his laboratory (Raghavan *et al.,* 2011, 2013 and 2015; Bannerjee *et al.,* 2012a and 2012b). All the corresponding genes and their variants were also over-expressed in *Anabaena* to demonstrate enhanced oxidative stress tolerance as also tolerance to other environmental stresses (Bannerjee *et al.,* 2013).

Engineering *Deinococcus radiodurans* for Enhanced Oxidative Stress Tolerance and Uranium Bioremediation

Dr. Apte's laboratory also took the lead in unravelling the molecular basis of extreme radioresistance in *Deinococcus radiodurans* and contribution of its DNA repair and oxidative stress tolerance abilities to this phenomenon in particular (Misra *et al.,* 2004 and 2006). A novel molecule pyrroloquinoline quinone (PQQ) was characterized from this microbe and shown to play very important role in radioprotection *in vitro* and *in vivo* in *D. radiodurans* (Misra *et al.,* 2004). Transgenosis of PQQ synthase gene was also shown to enhance oxidative stress tolerance of *E. coli* significantly (Khairnar *et al.,* 2003). PQQ was also shown to play a role in radiation-induced signalling and induction of DNA repair through a periplasmic protein kinase (Khairnar *et al.,* 2007). *D. radiodurans* was found to survive very high oxidative stress triggered by exposure to tellurite. Using genomic and proteomic approaches its tolerance to radiation was shown to be superior to tellurite induced oxidative stress, though the two stress responses shared a lot of common genes and proteins (Narasimha *et al.,* 2015).

On account of its radioresistance, *D. radiodurans* is an automatic choice for bioremediation of nuclear waste. Acid and alkaline phosphatase genes (*phoN* and *phoK*, respectively) were engineered into this microbe to construct strains that could bioprecipitate uranium from low concentrations of 0.5 mM up to 10mM over a pH range of 5-9, resulting in impressive uranium loading of up to 7-10 mg/g dry biomass (Appukuttan *et al.,* 2006; Nilgiriwala *et al.,* 2008 and 2009; Kulkarni *et al.,* 2013). The phosphatase activity and uranium bioprecipitation capabilities were retained up to 2 years in lyophiliozed cells (Seetharam *et al.,* 2009;

Appukuttan *et al.,* 2011). At acidic pH precipitated U was cell bound and easy to recover by settling and decantation, while at alkaline pH the extracellular precipitate could be immobilized in to columns and beads for online Uranium remediation in a flow through system and facilitated complete and easy recovery of U (Seetharm-Misra *et al.,* 2012; Kulkarni *et al.,* 2013 and 2016). Methods were also developed for surface display of bioremediation-active proteins in *D. radiodurans* by tagging them to surface layer proteins (Seetharam-Misra *et al.,* 2014). Radiation responsive promoters have been characterized from this microbe (Ujaoney *et al.,* 2010; Narasimha *et al.,* 2016 and 2017) and used to express phosphatases to further enhance U bioremediation in high radiation environments (Seetharam-Misra *et al.,* 2014). The bacterium has also been engineered to express plant *NiCoT* genes for purpose of removal of cobalt from waste solutions (Gogada *et al.,* 2015).

Developing Cyanobacteria for Uranium Bioremediation

Uranium resources on land are getting depleted with time and a novel idea is to recover uranium from extremely low concentrations of 3ppb from the seawater which holds nearly 60% of Earth's uranium. Marine cyanobacteria (*Synechcoccus elongatus* and *Anabaena torulosa*) were developed to sequester U from such low concentration and at an alkaline pH of 7.8, typical of seawater (Acharya *et al.,* 2009 and 2012a). The sequestered U was shown to be immobilized in extracellular polysaccharides or in unique surface associated polyphoshate bodies in marine cyanobacteria (Acharya and Apte, 2013a and 2013b). Proteomic analyses revealed that the ability to prevent availability of U in the cytosol underlies uranium tolerance of certain *Anabaena* species (Panda *et al.,* 2017). Such strains have also been used to remove U from RO water generated from desalination plants (Acharya *et al.,* 2012b).

Developing Microbes for Biodegradation of Organic Solvents and Pesticides

Dr. Apte's laboratory was the first in India to demonstrate the molecular basis of lindane (ã-HCH or ã-hexachlorocyclohexane) biodegradation from *Sphingomonas paucimobilis* (Adhya *et al.,* 1996) that subsequently led to cloning of several of the corresponding *linA* and *linB* genes and their variants from several *Sphingomonas* spp. in India. His laboratory also overexpressed the *linA* gene in (a) *E. coli* to construct a very sensitive and robust biosensor for lindane (Anu-Pratap *et al.,* 2012), and (b) *Anabaena* to develop a lindane biodegrading strain for rice field (Chaurasia *et al.,* 2013). They also isolated and characterized *Sphingobium* sp. strain RSMS, capable of degrading tributyl phosphate (TBP) and utilizing it as carbon and phosphorus source resulting in complete mineralization of this recalcitrant organic solvent of U and Pu in nuclear industry (Rangu *et al.,* 2014). The biochemical pathway of TBP degradation was elucidated and techniques developed to simultaneously degrade TBP and use the phosphate released for precipitation of traces of uranium still present in TBP (Rangu *et al.,* 2016).

Pseudomonas spp*:* Degrader of Many Aromatics

Pseudomonas putida CSV86*: An evader of* CCR. *P. putida* CSV86, a soil isolate from Dr. P Phale lab, utilizes an array of aromatics as the sole source of carbon and energy. This strain has a unique ability to utilize aromatics preferentially over glucose and co-metabolizes aromatics and organic acids (Basu *et al.,* 2006). Aromatics and organic acid mediated repression of glucose metabolism as well as transport was found to be responsible for this ability. In the strain CSV86, glucose to enters only through high affinity ATP dependent ABC transporter. This transport system comprises of a periplasmic glucose binding protein, GBP; outer membrane porin, OprB; two inner membrane ABC transporter proteins, GlcF and GlcG and a membrane bound cytoplasmic ATPase, GlcK (Figure 6C) and present together as a probable transcription unit (Paliwal *et al.,* 2014). The periplasmic glucose binding protein confers specificity to this system and has been studied at the structural level to understand the basis of the specificity (Figure 6D, Modak *et al.,* 2014, Pandey *et al.,* 2016).

Pseudomonas aeruginosa strain PP4. *Phthalate degradation*: *P. aeruginosa* strain PP4 is a soil isolate, capable of degrading phthalate isomers (phthalate, isophthalate and terephthalate) as sole sources of carbon and energy. Studies revealed that strain PP4 utilize phthalates efficiently by overcoming the metabolic inhibition adapting different strategies like modulating different forms of GDH or expressing higher concentration of the same enzyme (Vamsee-

Krishna and Phale, 2010), suggesting it to be a better candidate for the on-site bioremediation.

Pseudomonas sp. strain C5pp*: Evolution in process:* Carbaryl (1-naphthyl *N*-methylcarbamate) is widely used as a broad-spectrum insecticide. *Pseudomonas* sp. strain C5pp isolated from soil metabolizes carbaryl as the sole source of carbon and energy. Carbaryl is metabolized *via* 1-naphthol,1,2-dihydroxynaphthalene, salicylaldehyde, salicylate and gentisate to TCA cycle intermediates. The carbon-source dependent enzyme induction studies suggested that carbaryl degradation pathway is segmented into 'upper', 'middle' and 'lower' (Singh *et al.,* 2013) pathway. The combined approach of genomic library analysis and genome sequencing was used to identify the genes involved in carbaryl metabolic pathway. The sequence analysis suggested genes involved in the pathway have been acquired as a result of horizontal gene transfer.

Strain C5pp appears to be in early stages of evolution and can serve as an interesting model for studying evolution of metabolic pathway for recently introduced pesticide. As strain CSV86 has ability to preferentially utilize aromatics (globally) over simple carbon source like glucose it increases the potential for its use in biodegradation and biorefinery. Moreover, this strain also serves as efficient genetic tool for studying and metabolic engineering, so as to construct a robust strain for efficient removal of aromatic pollutants from the contaminated environment.

References

Acharya BK, Pathak H, Mohana S, Shouche Y, Singh V and Madamwar D (2011) Kinetic modeling and microbial community assessment of anaerobic biphasic fixed film bioreactor treating distillery spent wash *Water Res* **45** 4248-4259

Acharya C and Apte SK (2013a) Insights into the interactions of cyanobacteria with uranium *Photosynth Res* **118** 83-94

Acharya C and Apte SK (2013b) Novel surface associated polyphosphate bodies sequester uranium in the filamentous, marine cyanobacterium *Anabaena torulosa Metallomics* **5** 1595-1598

Acharya C, Chandwadkar P and Apte SK (2012a) Interaction of uranium with a filamentous, heterocystous, nitrogen-fixing cyanobacterium, *Anabaena torulosa Biores Technol* **116** 290-294

Acharya C, Chandwadkar P, Joseph D and Apte SK (2012b) Uranium (VI) recovery from saline environment by a marine unicellular cyanobacterium, *Synechococcus elongates J Radioanal Nucl Chem* **295** 845-850

Acharya C, Joseph D and Apte SK (2009) Uranium sequestration by a marine cyanobacterium *Synechococcus elongatus* BDU/75042 *Biores Technol* **100** 2176-2181

Adhya TK, Apte S , Raghu K, Sethunathan N and Murthy NBK (1996) Novel polypeptides induced by the insecticide lindane (?-hexachlorocyclohexane) are required for its biodegradation by a *Sphingomonas paucimobilis* strain *Biochem Biophys Res Commun* **221** 755-761

Ahemad M (2012) Implications of bacterial resistance against heavy metals in bioremediation: a review *Journal of Institute of Integrative Omics and Applied Biotechnology* **3**

Alahari A and Apte SK (2004) A novel potassium deficiency-induced stimulon in *Anabaena torulosa*" *J Biosci* **29** 153-161

Alahari A, Ballal A and Apte SK (2001) Regulation of potassium-dependent Kdp-ATPase expression in the nitrogen-fixing cyanobacterium *Anabaena torulosa J Bacteriol* **183** 5778-5781

Amin S, Shah B, Jain K, Patel A, Patel N, Joshi CG and Madamwar D (2015) Draft genome sequence of *Achromobacter* sp strain DMS1, capable of degrading polyaromatic hydrocarbons isolated from the industrially perturbed environment of Amlakhadi canal, India *Genome Announc* **3** e01264-15

Anu Prathap MU, Chaurasia A, Sawant S and Apte SK (2012) Polyaniline based highly sensitive microbial biosensor for selective and rapid detection of lindane" *Anal Chem* (dxdoiorg/101021/ac301077d)

Anwar S, Liaquat F, Khan QM, Khalid ZM and Iqbal S (2009) Biodegradation of chlorpyrifos and its hydrolysis product 3, 5, 6-trichloro-2-pyridinol by *Bacillus pumilus* strain C2A1 *Journal of Hazardous Materials* **168** 400-405

Appukuttan D, Rao AS and Apte SK (2006) Engineering *Deinococcus radiodurans* R1 for bioprecipitation of uranium from dilute nuclear waste *Appl Env Microbiol* **72** 7873-7878

Appukuttan D, Seetharam C, Padma N, Rao AS and Apte SK (2011) PhoN expressing, lyophilized, recombinant *Deinococcus radiodurans* cells for uranium bioprecipitation *J Biotechnol* **154** 285-290

Apte SK (2001) Coping with water stress: cyanobacteria show the way *Proc Indian Natn Sci Acad* (PINSA) **B67** 285-310

Apte SK and Bhagwat AA (1989) Salinity stress induced proteins in two nitrogen-fixing *Anabaena* strains differentially tolerant to salt *J Bacteriol* **171** 909-915

Apte SK and Thomas J (1986) Membrane electrogenesis and sodium transport in filamentous nitrogen-fixing cyanobacteria *Eur J Biochem* **154** 395-401

Apte SK and Thomas J (1997) Possible amelioration of coastal soil salinity using halotolerant nitrogen-fixing cyanobacteria *Plant and Soil* **189** 205-211

Apte S K, Reddy B R and Thomas J (1987) Relationship between sodium influx and salt tolerance of nitrogen-fixing cyanobacteria *Appl Env Microbiol* **53** 1934-1939

Apte SK and Haselkorn R (1990) Cloning of salinity stress-induced genes from the salt-tolerant nitrogen-fixing cyanobacterium *Anabaena torulosaPlant Molec Biol* **15** 723-733

Arora PK, Srivastava A and Singh VP (2014) Bacterial degradation of nitrophenols and their derivatives *Journal of hazardous materials* **266** 42-59

Arora PK, Srivastava A and Singh VP (2014) Degradation of 4-chloro-3-nitrophenol via a novel intermediate, 4-chlororesorcinol by *Pseudomonas* sp JHN *Scientific reports* **4**

Arora PK, Srivastava A and Singh VP (2014) Novel degradation pathway of 4-chloro-2-aminophenol via 4-chlorocatechol in *Burkholderia* sp RKJ 800 *Environmental Science and Pollution Research* **21** 2298-2304

Arora PK and Jain RK (2012) Biotransformation of 4-chloro-2-nitrophenol into 5-chloro-2-methylbenzoxazole by a marine *Bacillus* sp strain MW-1 *Biodegradation* **23** 325-331

Bajaj A, Pathak A, Mudiam MR, Mayilraj S and Manickam N (2010) Isolation and characterization of a Pseudomonas sp strain IITR01 capable of degrading á endosulfan and endosulfan sulfate *Journal of applied microbiology* **109** 2135-2143

Bala K, Sharma P and Lal R (2010) *Sphingobium quisquiliarum* sp nov, $P25^T$ a hexachlorocyclohexane (HCH) degrading bacterium isolated from HCH contaminated soil *Int J Syst Evol\ Microbiol* **60** 429-433

Balapure KH, Jain K, Chattaraj S, Bhatt NS and Madamwar D (2014) Co-metabolic degradation of diazo dye – Reactive blue 160 by enriched mixed cultures BDN *Journal of Hazardous Materials* **279** 85-95

Balapure K, Jain K, Bhatt N and Madamwar D (2016) Exploring bioremediation strategies to enhance the mineralization of textile industrial wastewater through sequential anaerobic-microaerophilic process *Int Biodeterioration* and *Biodegradation* **106** 97-105

Banerjee M, Ballal A and Apte SK (2012a) A novel glutaredoxin domain-containing peroxiredoxin 'All1541' protects the N_2-fixing cyanobacterium *Anabaena* PCC 7120 from oxidative stress *Biochemical J* **442** 671-680

Banerjee M, Ballal A and Apte SK (2012b) Mn-catalase (Alr0998) protects the photosynthetic, nitrogen-fixing cyanobacterium *Anabaena* PCC7120 from oxidative stress *Env Microbiol* **14** 2891-2900

Banerjee M, Raghavan P, Ballal A, Rajaram H and Apte SK (2013) "Oxidative stress management in the filamentous, heterocystous, diazotrophiccyanobacterium, *Anabaena* PCC7120" *Photosynth Res* **118** 59-70

Bashir H, Gangwar R and Mishra S (2015) Differential expression of lignocellulolytic enzymes by *Trametes* sp OE147 on cellulose and lactose *Biochim Biophys Acta- Prot Proteom* **1854** 1290-1299

Bassalo MC, Liu R and Gill RT (2016) Directed evolution and synthetic biology applications to microbial systems *Curr Opin Biotechno l***39** 126-133

Basu A, Apte SK, and Phale PS (2006) Preferential utilization of aromatic compounds over glucose by *Pseudomonas putida* CSV86 *Appl Environ Microbiol* **72** 2226-2230

Bedekar PA, Kshirsagar SD, Gholave AR and Govindwar SP (2015) Degradation and detoxification of methylene blue dye adsorbed on water hyacinth in semi continuous anaerobic–aerobic bioreactors by novel microbial consortium-SB *RSC Adv* **5** 99228-99239

Begum SS and Arundhati A (2016) A study of Bioremediation of Methyl Parathion in vitro using Potential Pseudomonas sp isolated from Agricultural Soil, Visakhapatnam, India *Int J Curr Microbiol App Sci* **5** 464-474

Bhagwat AA and Apte SK (1989) Comparative analysis of proteins induced by heat shock, salinity, and osmotic stress in the nitrogen-fixing cyanobacterium *Anabaena* sp strain L-31 *J Bacteriol* **171** 5187-5189

Chakka D, Gudla R, Madikonda AK, Pandeeti EV, Parthasarathy S and Siddavattam D (2015) The organophosphate degradation (*opd*) island borne esterase induced metabolic diversion in *E coli* and its influence on *p*-nitrophenol degradation *J Biol Chem* **290** 29920-30

Chakraborty R, Wu CH and Hazen TC (2012) Systems biology approach to bioremediation *Curr Opin Biotechnol* **23** 483-490

Chandel AK, Rudravaram R, Rao LV, Ravindra P and Narasu ML (2007) Industrial enzymes in bioindustrial sector development: an Indian perspective *Journal of Commercial Biotechnology* **13** 283-291

Chattaraj S, Johnson J and Madamwar D (2016a) Biotransformation of mixture of dyes by enriched bacterial consortium ASD *Desalination and Water treatment* **572** 1585-21597

Chattaraj S, Purohit H, Sharma A, Jadeja N and Madamwar D (2016b) Treatment of Common Effluent Treatment Plant Wastewater in a Sequential Anoxic–Oxic Batch Reactor by Developed Bacterial Consortium VN11 *Applied Biochemistry Biotechnology* **179** 514-529

Chatterjee S, Deb U, Datta S, Walther C and Gupta DK (2017) Common explosives (TNT, RDX, HMX) and their fate in the environment: Emphasizing bioremediation *Chemosphere* **184** 438-451

Chauhan A, Pandey G, Sharma NK, Paul D, Pandey J and Jain RK (2010) p-Nitrophenol degradation via 4-nitrocatechol in Burkholderia sp SJ98 and cloning of some of the lower pathway genes *Environmental* science and *technology* **44** 3435-3441

Chaurasia AK and Apte SK (2009) Overexpression of the *groESL* operon enhances the heat and salinity stress tolerance of the nitrogen-fixing cyanobacterium *Anabaena* sp strain PCC7120 *Appl Env Microbiol* **75** 6008-6012

Chaurasia AK, Adhya TK And Apte SK (2013) Engineering bacteria for bioremediation of persistent organochlorine pesticide lindane (gamma-hexachlorocyclohexane) *Biores Technol* **149** 439-445

Chaurasia AK and Apte SK (2011) Improved eco-friendly recombinant *Anabaena* sp strain PCC7120 with enhanced nitrogen biofertilizer potential *Appl Env Microbiol* **77** 395-399

Chhabra M, Mishra S and Sreekrishnan TR (2008) Mediator assisted decolorization/detoxification of reactive/acid dyes and dye mixtures using *Cyathus bulleri* laccase *Appl Biochem Biotechnol* **151** 587-598

Chhabra M, Mishra S and Sreekrishnan TR (2009) Degradation and detoxification of triarylmethane dyes by laccase/mediator-assisted laccase of *Cyathus bulleriJ Biotechnol* **143** 69-78

Chhabra M, Mishra S and Sreekrishnan TR (2015a) Combination of chemical and enzymatic treatment for efficient decolorization/degradation of textile effluent: low sludge and high operational stability *Biochem Engg J* **93** 17-24

Chhabra M, Mishra S and Sreekrishnan TR (2015b) Immobilized laccase mediated dye decolorization and transformation pathway of Azo Dye Acid Red 27 *J Environ Health Sci Engg* **13** 38-44

Choudhary M, Kumar R, Datta A, Nehra V and Garg N (2017) Bioremediation of Heavy Metals by Microbes In*Bioremediation of Salt Affected Soils: An Indian Perspective* 233-255 Springer International Publishing

Congeevaram S, Dhanarani S, Park J, Dexilin M and Thamaraiselvi K (2007) Biosorption of chromium and nickel by heavy metal resistant fungal and bacterial isolates *Journal of hazardous materials* **146** 270-277

Dadhwal M, Jit S, Kumari H and Lal R (2009) *Sphingobium chinhatense* sp nov, a hexachlorocyclohexane (HCH) degrading bacterium isolated from an HCH dump site *Int J Syst Evol Microbiol* **593** 140-3144

Dafale N, Agrawal L , Kapley A, Meshram S, Purohit HJ and Wate S (2010) Selection of indicator bacteria based on screening of 16S rDNA metagenomic library from a two-stage anoxic–oxic bioreactor system degrading azo dyes *Bioresource Technology* **101** 476-84

Das S and Adhya TK (2015) Degradation of chlorpyrifos in tropical rice soils *Journal of environmental management* **152** 36-42

Dasgupta N, Ranjan S, Mundekkad D, Ramalingam C, Shanker R and Kumar A (2015) Nanotechnology in agro-food: from field to plate *Food Research International* **69** 381-400

Dixit R, Malaviya D, Pandiyan K, Singh U B, Sahu A, Shukla R, Singh B P, Rai J P, Sharma P K, Lade H and Paul D (2015) Bioremediation of heavy metals from soil and aquatic environment: an overview of principles and criteria of fundamental processes*Sustainability* **7** 2189-2212

Dogra C, Raina V, Pal R, Suar M, Lal S, Gartemann K H, Holliger C, van der Meer JR and Lal R (2004) Organization of *lin* genes and IS*6100* among different strains of hexachlorocyclohexane-degrading *Sphingomonas paucimobilis*: Evidence for horizontal gene transfer *Journal of Bacteriology* **186** 2225-2235

Domde P, Kapley A and Purohit HJ (2007) Impact of Bioaugmentation with consortium of bacteria on the remediation of wastewater containing hydrocarbons *Environ Sci Pollut R* **14** 7-11

Dubey KK and Fulekar MH (2012) Chlorpyrifos bioremediation in Pennisetum rhizosphere by a novel potential degrader Stenotrophomonas maltophilia MHF ENV20 *World Journal of Microbiology and Biotechnology* **28** 1715-1725

Dudhagara D R, Rajpara RK, Bhatt JK, Gosai HB, Sachaniya BK and Dave BP (2016) Distribution, sources and ecological risk assessment of PAHs in historically

contaminated surface sediments at Bhavnagar coast, Gujarat, India *Environmental Pollution* **213** 338-346

Fernandes TA and Apte SK (2000) Differential regulation of nitrogenase activity by ionic and osmotic stresses and permeable sugars in the cyanobacterium *Anabaena* sp strain L-31 *Plant Sci* **150** 181-189

Gangwar R, Rasool S and Mishra S (2016) Evaluation of cellobiose dehydrogenase and laccase rich culture fluids of *Termitomyces* sp OE147 for degradation of Reactive blue 21*Biotechnol Rep* **12** 52-61

Garg N, Bieler N, Kenzom T, Chhabra M, Schumacher MA and Mishra S (2012) Cloning, sequence analysis, expression of *Cyathus bulleri* laccase in *Pichia pastoris* and characterization of recombinant laccase *BMC Biotechnol* **12** 75-87

Garg R, Srivastava R, Brahma V, Verma L, Karthikeyan S and Sahni G (2016) Biochemical and structural characterization of a novel halotolerant cellulase from soil metagenome *Scientific reports***6** 39634

Goel A, Babu SG and Celin MG (2012) Bioremediation of nitroaromatic compounds

Gogada R, Singh SS, Lunavat SK, Pamarthi MM, Rodrigues A, Vadivelu B, Phanithi PB, Gopala V And Apte SK (2015) Engineered *Deinococcus radiodurans* R1 with NiCoT genes for bioremoval of trace cobalt from spent decontamination solutions of nuclear power reactors *Appl Microbiol Biotechnol* **99** 9203-9213

Goyal P and Bansiwal RK (2017) Environmental Bioremediation: Biodegradation of Xenobiotic Compounds In*Xenobiotics in the Soil Environment* 347-371 Springer International Publishing

Gupta M, Mathur S, Sharma TK, Rana M, Gairola A, Navani NK and Pathania R (2016) A study on metabolic prowess of Pseudomonas sp RPT 52 to degrade imidacloprid, endosulfan and coragen *Journal of hazardous materials* **301** 250-258

Hussain S, Siddique T, Arshad M and Saleem M (2009) Bioremediation and phytoremediation of pesticides: recent advances *Critical Reviews in Environmental Science and Technology* **39** (843-907)

Iyer R, Iken B and Damania A (2013) A comparison of organophosphate degradation genes and bioremediation applications *Environmental microbiology reports* **5** 787-798

Jadeja NB, More RP, Purohit HJ and Kapley A (2014) Metagenomic analysis of oxygenases from activated sludge *Bioresour Technol* **165** 250-256

Jain K, Shah V, Chapla D and Madamwar D (2012) Decolorization and degradation of azo dye – Reactive Violet 5R by an acclimatized indigenous bacterial mixed cultures-SB4 isolated from anthropogenic dye contaminated soil *Journal of Hazardous Materials* **213-214** 378-386

Jain PK, Gupta VK, Pathak H, Lowry M and Jaroli DP (2010) Characterization of 2T engine oil degrading indigenous bacteria, isolated from high altitude (Mussoorie), India *World Journal of Microbiology and Biotechnology* **26** 1419-1426

Jit S, Dadhwal M, Kumari H, Jindal S, Kaur J, Lata P, Niharika N, Lal D, Garg N, Gupta SK, Sharma P, Bala K, Singh A, Vijgen J, Weber R and Lal R (2011) Evaluation of hexachlorocyclohexane contamination from the last lindane production plant operating in India *Environ Sci Pollut Res* **18** 586-597

Johnson J, Shah B, Jain K, Parmar N, Hinsu A, Patel N, Joshi C and Madamwar D (2016) Draft Genome Sequence of *Paenibacillus* sp strain DMB5, Acclimatized and Enriched for Catabolizing Anthropogenic Compounds *Genome Announc* **4** e00211-16

Joshi PK, Swarup A, Maheshwari S, Kumar R and Singh N (2011) Bioremediation of heavy metals in liquid media through fungi isolated from contaminated sources *Indian journal of microbiology* **51** 482-487

Joshi SM, Inamdar SA, Patil SM and Govindwar SP (2013) Molecular assessment of shift in bacterial community in response to Congo Red *Int Biodeter Biodegrad* **77** 18-21

Juwarkar AA (2012) Microbe-assisted phytoremediation for restoration of biodiversity of degraded lands: a sustainable solution *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences* **82** 313-318

Kadam AA, Kamatkar JD, Khandare RV, Jadhav JP and Govindwar SP (2013) Solid state fermentation: Tool for bioremediation of adsorbed textile dyestuff on distillery industry waste-yeast biomass using isolated *Bacillus cereus* strain EBT1 *Environ Sci Pollut Res* **20** 1009-1020

Kanekar PP, Sarnaik SS, Dautpure PS, Patil VP and Kanekar SP (2014) Bioremediation of nitroexplosive waste waters In *Biological Remediation of Explosive Residues* Springer International Publishing

Kapley A and Purohit HJ (2009) Diagnosis of treatment efficiency in industrial wastewater treatment plants: A case study at a refinery ETP *Environ Sci Technol* **43** 3789-3795

Kapley A, Liu R, Jadeja NB, Zhang Y, Yang M and Purohit HJ (2015) Shifts in microbial community and its correlation with degradative efficiency in a wastewater treatment plant *Appl Biochem Biotechnol* **176** 2131-2143

Kapley A, Thierry B and Purohit HJ (2007) Eubacterial diversity of activated biomass from a CETP *Research in Microbiology* **158** 494-500

Kenzom T, Srivastava P and Mishra S (2014) Structural insights into degradation of Reactive Blue 21 by engineered *Cyathus bulleri* laccase and characterization of the degradation products *Appl Environ Microbiol* **80** 7484-7495

Kenzom T, Srivastava P and Mishra S (2015) Simplified high-throughput screening of *AOX1*-expressed laccase enzyme in *Pichia pastoris Anal Biochem* **489** 49-51

Kenzom T, Srivastava P and Mishra S (2015) Simplified high-throughput screening of *AOX1*-expressed laccase enzyme in *Pichia pastoris Anal Biochem* **489** 49-51

Khairnar NP, Kamble VA, Mangoli SH, Apte SK And Misra HS (2007) Involvement of a periplasmic protein kinase in DNA strand break repair and homologous recombination in *Escherichia coli Mol Microbiol* **65** 294-304

Khairnar NP, Misra HS and Apte SK (2003) Pyrroloquinoline-quinine synthesized in *Escherichia coli* by pyrroloquinoline-quinone synthase of *Deinococcus radiodurans* plays a role beyond mineral phosphate solubilization" *Biochem Biophys Res Commun* **312** 303-308

Khan NU, Varma B, Imrana N and Shetty PK (2011) Phytoremediation using an indigenous crop plant (wheat): the uptake of methyl parathion and metabolism of p-nitrophenol *Indian Journal of Science and Technology* **4** 1-7

Khandare RV and Govindwar SP (2015) Phytoremediation of textile dyes and effluents: Current scenario and future prospects *Biotechnol Adv* **33** 1697-1714

Khanna GP, Goyal D and Khanna S (2011) Pyrene biodegradation by Bacillus spp isolated from coal tar–contaminated soil *Bioremediation journal* **15** 12-25

Khardenavis AA, Kapley A and Purohit HJ (2007) Simultaneous nitrification and denitrification by diverse *Diaphorobacter* sp *Appl Microbiol Biotechnol* **77** 403-409

Khardenavis AA, Kapley A and Purohit HJ (2008) Phenol mediated improved performance of active biomass for treatment of distillery wastewater *International Journal of Biodeteroration and Biodegradation* **62** 38-45

Khardenavis AA, Kapley A and Purohit HJ (2010) Salicylic acid mediated enhanced biological treatment of wastewater *Applied Biochemistry and Biotechnology* **160** 704-18

Kulkarni A , Kadam AA, Kachole MS and Govindwar SP (2014) Lichen *Permeliaper lata*: A novel system for biodegradation and detoxification of disperse dye Solvent Red 24 *J Hazard Mater* **214** 461-468

Kulkarni S, Ballal A and Apte SK (2013) Bioprecipitation of uranium from alkaline waste solutions using recombinant *Deinococcus radiodurans J Haz Metals* **262** 853-861

Kulkarni S, Chitra SM, Gupta A, Ballal AA and Apte SK (2016) Interaction of uranium with bacterial cell surface: Inferences from phosphatase mediated uranium precipitation *Appl Env Microbiol* **82** 4965

Kumar S, Dagar VK, Khasa YP and Kuhad RC (2013) Genetically modified microorganisms (GMOs) for bioremediation In *Biotechnology for environmental management and resource recovery* 191-218 Springer India

Kumari A, Singh D, Ramaswamy S and Ramanathan G (2017) Structural and functional studies of ferredoxin and oxygenase components of 3-nitrotoluene dioxygenase from Diaphorobacter sp strain DS2 *PloS one* **12** pe0176398

Kumari R, Subudhi S, Suar M, Dhingra G, Raina V, Dogra C, Lal S, van der Meer JR, Holliger C and Lal R (2002) Cloning and characterization of *lin* genes responsible for the degradation of hexachlorocyclohexane isomers by *Sphingomonas paucimobilis* strain B90 *Applied and Environmental Microbiology* **68** 6021-6028

Kundu D, Hazra C and Chaudhari A (2014) Bioremediation of Nitroaromatics (NACs)-Based Explosives: Integrating '-Omics' and Unmined Microbiome Richness In *Biological Remediation of Explosive Residues* 179-199 Springer International Publishing

Kundu S and Gupta AK (2007) Adsorption characteristics of As (III) from aqueous solution on iron oxide coated cement (IOCC) *Journal of Hazardous Materials* **142** 97-104

Kurade MB, Waghmode TR, Patil SM, Jeon BH and Govindwar SP (2017) Monitoring the gradual biodegradation of dyes in a simulated textile effluent and development of a novel triple layered fixed bed reactor using a bacterium-yeast consortium *Chem Eng J* **307** 1026-1036

Lal R, Dogra C, Malhotra S, Sharma P and Pal R (2006) Diversity, distribution and divergence of *lin* genes in hexachlorocyclohexane-degrading sphingomonads *Trends in Biotechnology* **24** 121-130

Lal R, Pandey G, Sharma P, Kumari K, Malhotra S, Pandey R, Raina V, Kohler HPE, Holliger C, Jackson C and Oakeshott JG (2010) Biochemistry of Microbial Degradation of Hexachlorocyclohexane and Prospects for Bioremediation *Microbiol Mol Biol Rev* **74** 58-80

Malik DK, Bhatia D and Rathi M (2014) Bacterial Degradation of Some Organophosphate Compounds In *Microbial Diversity and Biotechnology in Food Security* 531-541

Springer India

Manoharachary C, Kunwar IK and Rajithasri AB (2014) Advances in applied mycology and fungal biotechnology

Mishra A and Sardar M (2014) Alpha amylase mediated synthesis of gold nanoparticles and their application in the reduction of nitroaromatic pollutants *Energy and Environment Focus* **3** 179-184

Mishra A, Kumari M, Pandey S, Chaudhry V, Gupta KC and Nautiyal CS (2014) Biocatalytic and antimicrobial activities of gold nanoparticles synthesized by Trichoderma sp *Bioresource technology* **166** 235-242

Misra H, Khairnar NP, Kota S, Shrivastava S, Joshi VP and Apte SK (2006) An exonuclease I – sensitive DNA repair pathway in *Deinococcus radiodurans*: a major determinant of radiation resistance *Mol Microbiol* **59** 1308-1316

Misra HS, Khairnar NP, Barik A, Priyadarshini I, Mohan H and Apte SK (2004) Pyrroloquinoline-quinone : A reactive oxygen species scavenger in bacteria" *FEBS Lett* **578** 26-30

Modak A, Bhaumik P and Phale PS (2014) Periplasmic glucose-binding protein from *Pseudomonas putida* CSV86-identification of the glucose-binding pocket by homology-model-guided site-specific mutagenesis *FEBS J* **281** 365-375

Mohanty B, Muduli PR, Behera AT, Mahapatro D, Barik SK, Nag SK, Samal RN and Pattnaik A K (2016) Assessment of petroleum hydrocarbon in a tropical brackish water lagoon: Chilika, India *Chemistry and Ecology* **32** 653-668

Moharikar A, Purohit HJ and Kumar R (2005) Microbial population dynamics at effluent treatment plants *J Environ Monit* **7** 552-558

Mulla SI, Manjunatha TP, Hoskeri RS, Tallur PN and Ninnekar HZ (2011) Biodegradation of 3-Nitrobenzoate by Bacillus flexus strain XJU-4 *World Journal of Microbiology and Biotechnology* **27** 1587-1592

Nagvenkar GS and Ramaiah N (2010) Arsenite tolerance and biotransformation potential in estuarine bacteria *Ecotoxicology* **19** 604-613

Narasimha A, Basu B, Gupta A, Joseph D and Apte SK (2015) Depletion of reduction potential and key energy generation metabolic enzymes underlies tellurite toxicity in *Deinococcus radiodurans Proteomics* **15** 89-97

Narasimha A, Basu B and Apte SK (2016) *In situ* real time evaluation of radiation-responsive promoters in the extremely radioresistant microbe *Deinococcus radiodurans J Biosci* **41**193-203

Narasimha A, Basu B and Apte SK (2017) Proximity of Radiation Desiccation Response Motif to the core promoter is essential for basal repression as well as gamma radiation-induced *gyrB* gene expression in *Deinococcus radiodurans Gene* **615** 8-17

Negi V, Lata P, Sangwan N, Das S, Gupta SK, Rao DLN and Lal R (2014) Draft genome sequence of hexachlorohexane (HCH) degrading *Sphingobium lucknowense* strain $F2^T$ isolated from the HCH dumpsite *Genome Announcement* **2** e00788-14

Nilgiriwala K, Alahari A, Rao AS and Apte SK (2008) Cloning and over-expression of an alkaline phosphatase PhoK from *Sphingomonas* sp BSAR-1 for uranium bioprecipitation from alkaline solutions *Appl Env Microbiol* **1784** 1256-1264

Nilgiriwala K, Bihani SC, Das A, Prashar V, Kumar M, Ferrer J-L, Apte SK and Hosur MV (2009) Crystallization and preliminary X-ray crystallographic analysis of PhoK, an extracellular alkaline phosphatase from *Sphingomonas* sp BSAR-1 *Acta Cryst* **F65** 917-919

Ningthoujam DS, Sanasam S and Mutum A (2012) Characterization of p-nitrophenol degrading actinomycetes from Hundung limestone deposits in Manipur, India *African Journal of Biotechnology* **11** 10210-10220

Niti C, Sunita S, Kamlesh K and Rakesh K (2013) Bioremediation: an emerging technology for remediation of pesticides *Res J Chem Environ* **17** 88-105

Pailan S and Saha P (2015) Chemotaxis and degradation of organophosphate compound by a novel moderately thermo-halo tolerant Pseudomonas sp strain BUR11: evidence for possible existence of two pathways for degradation *Peer J* **3** pe1378

Pal A and Paul AK (2008) Microbial extracellular polymeric substances: central elements in heavy metal bioremediation *Indian Journal of Microbiology* **48** 49

Pal RR, Khardenavis AA and Purohit HJ (2015) Identification and monitoring of nitrification and denitrification genes in *Klebsiella pneumoniae* EGD-HP19-C for its ability to perform heterotrophic nitrification and aerobic denitrification *Func* and *Integ Genomics* **15** 63-76

Paliwal V, Puranik S and Purohit HJ (2012) Integrated perspective for effective bioremediation *Appl Biochem and Biotechnol* **166** 903-924

Paliwal V, Raju SC, Modak A, Phale PS and Purohit HJ (2014) *Pseudomonas putida* CSV86: A candidate genome for genetic bioaugmentation *PLoS One* **9** e84000

Panda B, Basu B, Rajaram H and Apte SK (2014) Methyl viologen responsive proteome dynamics of *Anabaena* sp strain 7120 *Proteomics* **14** 1895-1904

Panda B, Basu B, Rajaram H and Apte SK (2015) Comparative proteomics of oxidative stress response in three cyanobacterial strains native to Indian paddy fields *J Proteomics* **127** 152-160

Panda B, Basu B, Acharya C, Rajaram H and Apte SK (2017) Proteomic analysis reveals contrasting response to uranium in two nitrogen-fixing *Anabaena* strains, differentially tolerant to uranium *Aquatic Toxicol* **182** 205-213

Pandeeti EVP and Siddavattam D (2011) Purification and characterization of catechol 1, 2-dioxygenase from Acinetobacter sp DS002 and cloning, sequencing of partial catA gene *Indian journal of microbiology* **51** 312-318

Pandeeti EVP, Chakka D, Pandey JP and Siddavattam D (2011) Indigenous organophosphate-degrading (*opd*) plasmid pCMS1 of *Brevundimonas diminuta* is self-transmissible and plays a key role in horizontal mobility of the *opd* gene *Plasmid* **65** 226-31

Pandeeti EV, Longkumer T, Chakka D, Muthyala VR, Parthasarathy S, Madugundu AK, Ghanta S, Medipally SR, Pantula SC, Yekkala H and Siddavattam D (2013) Multiple mechanisms contribute to lateral transfer of an organophosphate degradation (*opd*) island in *Sphingobium fuliginis* ATCC 27551 G3: *Genes Genom Genet* **2** 1541-1554

Pandey S, Modak A, Phale PS and Bhaumik P (2016) High resolution structures of periplasmic glucose-binding protein of *Pseudomonas putida* CSV86 reveal structural basis of its substrate specificity *J Biol Chem* **291** 7844-7857

Park JH, Lamb D, Paneerselvam P, Choppala G, Bolan N and Chung JW (2011) Role of organic amendments on enhanced bioremediation of heavy metal (loid) contaminated soils *Journal of hazardous materials* **185** 549-574

Parthasarathy S, Paripatla H, Palmer T and Siddavattam D (2016) Organophosphate Hydrolase is a lipoprotein and interacts with P_i-specific transport system to facilitate growth of *Brevudimonas diminuta* using OP insecticide as source of phosphate *J Biol Chem* **291** 7774-7785

Patel V, Cheturvedula S and Madamwar D (2012a) Phenanthrene degradation by *Pseudoxanthomonas* sp DMVP2 isolated from hydrocarbon contaminated sediment of Amlakhadi canal, Gujarat, *India Journal of Hazardous Materials* **201-202** 43-51

Patel V and Madamwar D (2013) Biodegradation of phenanthrene in bioaugmented microcosm by consortium ASP developed using coastal sediment of Alang-Sosiya ship breaking yard, Gujarat *Marine Pollution Bulletin* **74** 199-207

Patel V, Munot H, Shouche YS and Madamwar D (2014) Response of bacterial community structure to seasonal fluctuation and anthropogenic pollution on costal water of Alang-Sosiys ship nreaking yard, Bhavnagar, India *Bioresource Technology* **161** 362-370

Patel V, Munot H, Shah V, Shouche Y and Madamwar D (2015) Taxonomic profiling of bacterial community structure from coastal sediment of Alang-Sosiya shipbreaking yard near Bhavnagar, India *Marine Pollution Bulletin* **101** 736-745

Patil SM, Chandanshive VV, Rane NR, Khandare RV, Watharkar AD and Govindwar SP (2016) Bioreactor with *Ipomoea hererifolia* adventitious roots and its endophyte *Cladosporium cladosporioids* for textile dye degradation *Environ Res* **146** 340-349

Phale PS, Basu A, Majhi PD, Deveryshetty J, Vamsee-Krishna C and Shrivastava R (2007) Metabolic diversity in bacterial degradation of aromatic compounds *Omics: a journal of integrative biology* **11** 252-279

Phale PS, Paliwal V, Raju SC, Modak A and Purohit HJ (2013) Genome sequence of naphthalene-degrading soil bacterium Pseudomonas putida CSV86 *Genome announcements* **1** e00234-12

Pinjari AB, Pandey JP, Kamireddy S and Siddavattam D (2013) Expression and subcellular localization of organophosphate hydrolase in acephate degrading Pseudomonas sp strain Ind01 and its use as a potential biocatalyst for elimination of organophosphate insecticides *Letters in applied microbiology* **57** 63-68

Purohit HJ, Raje DV, Kapley A, Padmanabhan P and Singh RN (2003a) Genomics tools in environmental impact assessment *Env Sci Technol* 357A-363A

Purohit HJ, Kapley A, Moharikar A and Narde G (2003b) A novel approach for extraction of PCR-compatible DNA from activated sludge samples collected from different biological effluent treatment plants *Journal of Microbiological Methods* **52** 315-323

Purohit HJ, Kapley A, Khardenavis A, Qureshi A and Dafale NA (2016) Insights in waste management bioprocesses using genomic tools *Adv in Appl Microbiol* **97** 121-170

Qureshi A, Prabu SK and Purohit HJ (2001) Isolation and Characterisation of *Pseudomonas* strain for degradation of 4-nitrophenol *Microbes and Environment* **16** 49-52

Qureshi A and Purohit HJ (2002) Isolation of bacterial consortia for degradation of p-nitrophenol from agricultural soil *Annals in Applied Biology* **140** 159-162

Qureshi A, Verma V, Kapley A and Purohit HJ (2007) Degradation of 4-nitroaniline by *Stenotrophomonas strain HPC 135*' *International Biodeterioration and Biodegradation* **60** 215-218

Qureshi A, Mohan M, Kanade GS, Kapley A and Purohit HJ (2009) In situ bioremediation of organochlorine pesticide contaminated microcosm soil and evaluation by gene probe *Pest management science* **65** 798-804

Raghavan PS, Rajaram H and Apte SK (2011) Nitrogen status dependent oxidative stress tolerance conferred by overexpression of MnSOD and FeSOD proteins in *Anabaena* sp strain PCC7120 *Plant Mol Biol* **77** 407-417

Raghavan P, Rajaram H and Apte SK (2013) N-terminal processing of membrane-targeted MnSOD and formation of multiple active superoxide dismutase dimmers in the nitrogen-fixing cyanobacterium *Anabaena* sp strain PCC7120" *FEBS J* **280** 4827-4838

Raghavan PS, Rajaram H and Apte SK (2015) Membrane targeting of MnSOD is essential for oxidative stress tolerance of nitrogen fixing cultures of *Anabaena* sp strain PCC7120 *Plant Mol Biol* **88** 503-514

Rajaram H, Ballal A, Apte SK, Wiegert T and Schumann W (2001) Cloning and characterisation of the major *groESL* operon from a nitrogen-fixing cyanobacterium *Anabaena* sp strain L-31 *Biochim Biophys Acta : Gene Struct Expr* **1519** 143-146

Rajaram H and Apte SK (2003) Heat-shock response and its contribution to thermotolerance of the nitrogen-fixing cyanobacterium *Anabaena* sp strain L-31 *Arch Microbiol* **179** 423-429

Rajaram H and Apte SK (2008) Nitrogen status and heat-stress-dependent differential expression of *cpn60* chaperonin gene influences thermotolerance in the cyanobacetrium *Anabaena Microbiology* **154** 317-325

Rajaram H, Chaurasia AK and Apte SK (2014) Cyanobacterial heat-shock response: role and regulation of molecular chaperones *Microbiol* **160** 647-658

Rangu SS, Muralidharan B, Tripathi SC and Apte SK (2014) Tributyl phosphate biodegradation to butanol and phosphate and utilization by a novel bacterial isolate, *Sphingomonas* sp strain RSMS *Appl Microbiol Biotechnol* **98** 2289-2296

Rangu SS, Basu B, Muralidharan B, Tripathi SC and Apte SK (2016) Involvement of phosphoesterases in tributyl phosphate degradation in *Sphingobium* sp RSMS *Appl Microbiol Biotechnol* **100** 461-468

Rao KR, Vipin AV, Hariprasad P, Appaiah KA and Venkateswaran G (2017) Biological detoxification of Aflatoxin B 1 by Bacillus licheniformis CFR1 *Food Control* **71** 234-241

Ray SA and Ray MK (2009) Bioremediation of heavy metal toxicity-with special reference to chromium *Al Ameen J Med Sci* **2** 57-63

Reddy GS, Rafi MM, Kumar SR, Khayalethu N, Rao DM, Manjunatha B, Philip GH and Reddy BR (2016) Optimization study of 2-hydroxyquinoxaline (2-HQ) biodegradation by Ochrobactrum sp HQ1 *Biotech* **6** 51

Sagarkar S, Bhardwaj P, Storck V, Devers-Lamrani M, Martin-Laurent F and Kapley A (2016) s-triazine degrading bacterial isolate *Arthrobacter* sp AK-YN10, a candidate for bioaugmentation of atrazine contaminated soil *Appl Microbiol Biotechnol* **100** 903-913

Saha SP, Banik SP, Majumder A, Noor A, Biswas K, Hasan N, Banerjee N, Saha P, Das R, Halder S and Parveen S (2014) Bioremediation of Methyl Parathion by Bacterial Strains Isolated from Fresh Vegetables *Journal of Environment and Sociobiology* **11** 43-56

Salony, Mishra S and Bisaria VS (2006) Production and characterization of laccase from *Cyathus bulleri* and its use in decolourization of recalcitrant textile dyes *Appl Microbiol Biotechnol* **71** 646-653

Salony, Garg N, Baranwal R, Chhabra M, Mishra S, Chaudhuri TK and Bisaria VS (2008) Laccase of *Cyathus bulleri*: structural, catalytic characterization and expression in *Escherichia coli Biochim Biophys Acta- Prot and Proteom* **1784** 259-268

Saratale RG, Saratale GD, Chang JS and Govindwar SP (2011) Outlook of bacterial decolorization and degradation of azo dyes: a review *J Taiwan Inst Chem Eng* **42** 138-157

Sarkar S, Seenivasan S and Asir RPS (2010) Biodegradation of propargite by Pseudomonas putida, isolated from tea rhizosphere *Journal of hazardous materials* **174** 295-298

Sasikala C, Jiwal S, Rout P and Ramya M (2012) Biodegradation of chlorpyrifos by bacterial consortium isolated from agriculture soil *World Journal of Microbiology and Biotechnology* **28** 1301-1308

Sathyavathi S, Manjula A, Rajendhran J and Gunasekaran P (2014) Extracellular synthesis and characterization of nickel oxide nanoparticles from Microbacterium sp MRS-1 towards bioremediation of nickel electroplating industrial effluent *Bioresource technology* **165** 270-273

Seetharam C, Soundarajan S, Udas AC, Rao AS and Apte SK (2009) Lyophilised, non-viable, recombinant *E coli* cells for cadmium bioprecipitation and recovery *Proc Biochem* **44** 246-250

Seetharam-Misra C, Appukuttan D, Kantamreddi VSS, Rao AS and Apte SK (2012) Recombinant *D. radiodurans* cells for bioremediation of heavy metals from acidic/neutral aqueous wastes *Bioengineered Bugs* **3** 44-48

Seetharam-Misra C, Mukhopadhyay R and Apte SK (2014) Harnessing a radiation inducible promoter of *Deinococcus radiodurans* for enhanced precipitation of uranium *J Biotechnol* **189** 88-93

Selvakumaran S, Kapley A, Vipin C Kalia and Purohit HJ (2008) Phenotypic and Phylogenic Groups to Evaluate The Diversity of Citrobacter Isolates From Activated Biomass of Effluent Treatment Plants *Bioresource Technology* **99** 1189-1195

Selvakumaran S, Kapley A, Kashyap SM, Daginawala HF, Kalia VC and Purohit HJ (2011) Diversity of aromatic ring-hydroxylating dioxygenase gene in *Citrobacter Bioresource Technology* **102** 4600-4609

Sengupta K, Maiti TK and Saha P (2015) Degradation of 4-nitrophenol in presence of heavy metals by a halotolerant Bacillus sp strain BUPNP2, having plant growth promoting traits *Symbiosis* **65** 157-163

Shah B, Jain K, Patel N, Pandit R, Patel A, Joshi CG and Madamwar D (2015) Draft genome sequence of *Paenibacillus* sp Isolated from Alang ship breaking yard which harbors genes for xenobiotic degradation *Genome Announcements* **3** e00554-15

Shah B, Mohan V, Jain K and Madamwar D (2016a) Microaerophillic symmetric reductive cleavage of reactive azo dye – Ramazole Brilliant Violet 5R by developed consortium VIE6: community synergism *Applied Biochemistry and Biotechnology* **180** 1029-1042

Shah B, Patel A and Madamwar D (2016b) Kinetic modeling and community dynamics of microaerophillic treatment of textile dyes containing effluent by consortium VIE6 *Environmental Process* Doi: 101007/s40710-016-0156-0

Shah V and Madamwar D (2013a) Community genomics: Isolation, characterization and expression of gene coding for azoreductase International *Biodeterioration* and *Biodegradation* **79** 1-8

Shah V, Zakrzewski M, Wibberg D, Eikmeyer F, Schlüter A and Madamwar D (2013b) Taxonomic Profiling and Metagenome Analysis of a Microbial Community from a Habitat Contaminated with Industrial Discharges *Microbial Ecology* **66** 533-550

Sharma N, Lavania M and Lal B (2019) microbes and their secondary metabolites agents in bioremediation of hydrocarbon contaminated site. *Arch Pet Environ Biotechnol*, DOI: 10.29011/2574-7614.100051, 2019

Sharma P, Raina V, Kumari R, Malhotra S, Dogra C, Kumari H, Kohler HPE, Buser HR, Holliger C and Lal R (2006) Haloalkane dehalogenase LinB is responsible for beta- and delta-hexachlorocyclohexane transformation in *Sphingobium indicum* B90A *Applied and Environmental Microbiology* **72** 5720-5727

Sharma P, Bala K, Singh A, Vijgen J, Weber R and Lal R (2010) Evaluation of hexachlorocyclohexane contamination from the last lindane production plant operating in India *Environmental science and pollution research international* **18** 586-597

Singare PU (2016) Distribution and risk assessment of suspected endocrine-disrupting pesticides in creek water of Mumbai, India *Marine pollution bulletin* **102** 72-83

Singh A, Shahid M, Srivastava M, Pandey S, Sharma A and Kumar V (2014) Optimal physical parameters for growth of Trichoderma species at varying pH, temperature and agitation *Virol Mycol* **3** 1-7

Singh A and Prasad SM (2015) Remediation of heavy metal contaminated ecosystem: an overview on technology advancement *International Journal of Environmental Science and Technology* **12** 353-366

Singh B, Kaur J and Singh K (2012) Biodegradation of malathion by Brevibacillus sp strain KB2 and Bacillus cereus strain PU *World Journal of Microbiology and Biotechnology* **28** 1133-1141

Singh DN and Tripathi AK (2013) Coal induced production of a rhamnolipid biosurfactant by Pseudomonas stutzeri, isolated from the formation water of Jharia coalbed *Bioresource technology* **128** 215-221

Singh R, Trivedi VD and Phale PS (2013) Metabolic regulation and chromosomal localization of carbaryl degradation pathway in *Pseudomonas* sp strains C4, C5 and C6 *Arch Microbiol* **195** 521-535

Sood N, Patle S and Lal B (2010) Bioremediation of acidic oily sludge-contaminated soil by the novel yeast strain Candida digboiensis TERI ASN6 *Environmental Science and Pollution Research* **17** 603-610

Sreenivasulu C, Megharaj M, Venkateswarlu K and Naidu R (2012) Degradation of p-nitrophenol by immobilized cells of Bacillus spp isolated from soil *International biodeterioration* and *biodegradation* **68** 24-27

Srivastava SK, Yamada R, Ogino C and Kondo A (2013) Biogenic synthesis and characterization of gold nanoparticles by Escherichia coli K12 and its heterogeneous catalysis in degradation of 4-nitrophenol *Nanoscale research letters* **8** 70

Suar M, van der Meer JR, Lawlor K, Holliger C and Lal R (2004) Dynamics of multiple *lin* gene expression in *Sphingomonas paucimobilis* B90A in response to different hexachlorocyclohexane isomers *Appl Environ Microbiol* **70** 6650-6656

Suar M, Hauser A, Poiger T, Buser HR, Mu¨ller MD, Dogra C, Raina V, Holliger C, van der Meer JR, Lal R and Kohler HP (2005) Enantioselective transformation of alpha-hexachlorocyclohexane by the dehydrochlorinases LinA1 and LinA2 from the soil bacterium *Sphingomonas paucimobilis* B90A *Appl Environ Microbiol* **7** 8514-8518

Telke AA, Kadam AA and Govindwar SP (2015) Bacterial enzymes and their role in decolorization of Azo dyes In: Microbial degradation of synthetic dyes in wastewaters (Ed: Singh) pp 149-168 Springer International Publishing, Switzerland

Thangaraj K, Kapley A and Purohit HJ (2008) Characterization of diverse *Acinetobacter* isolates for utilization of multiple aromatic compounds *Bioresource Technology* **99** 2488-2494

Tikariha H, Pal RR, Qureshi A, Kapley A and Purohit H J (2016) In silico analysis for prediction of degradative capacity of *Pseudomonas putida* SF1 *Gene* **1119** 30491-30497

Tiwari J, Naoghare P, Sivanesan S and Bafana A (2017) Biodegradation and detoxification of chloronitroaromatic pollutant by Cupriavidus *Bioresource technology* **223** 184-191

Tripathi A, Upadhyay RC and Singh S (2011) Mineralization of mono-nitrophenols by Bjerkandera adusta and Lentinus squarrosulus and their extracellular ligninolytic enzymes *Journal of basic microbiology* **51** 635-649

Trivedi VD, Jangir PK, Sharma R and Phale PS (2016) Insights into functional and evolutionary analysis of carbaryl metabolic pathway from *Pseudomonas* sp strain C5pp *Sci Rep* **7** 38430

Ujaoney AK, Potnis A, Mukhopadhyay R and Apte SK (2010) Radiation desiccation response motif (RDRM) like sequences are involved in transcriptional activation of deinococcal *ssb* gene by ionizing radiation but not by desiccation *J Bacteriol* **192** 5637-5644

Vamsee-Krishna C and Phale PS (2010) Bypassing isophthalate inhibition by modulating glutamate dehydrogenase (GDH): purification and kinetic characterization of NADP-GDHs from isophthalate-degrading *Pseudomonas aeruginosa* strain PP4 and *Acinetobacter lwoffii* strain ISP4 *J Bacteriol* **192** 801-806

Varjani SJ and Upasani VN (2013) Comparative studies on bacterial consortia for hydrocarbon degradation *Int J Innov Res Sci Eng Technol* **2** 5377-5383

Varjani SJ and Upasani VN (2016) Biodegradation of petroleum hydrocarbons by oleophilic strain of Pseudomonas aeruginosa NCIM 5514 *Bioresource technology* **222** 195-201

Varjani SJ (2017) Microbial degradation of petroleum hydrocarbons *Bioresource technology* **223** 277-286

Vats A and Mishra S (2017) Decolorization of complex dyes and textile effluent by extracellular enzymes of *Cyathus bulleri* cultivated on agro-residues/domestic wastes and proposed pathway of degradation of Kiton blue A and reactive orange 16 *Environ Sci Pollut Res* **24** 11650-11662

Venkata S, Mohan P, Suresh B, Naresh K, Velvizhi G and Madamwar D (2012) Acid azo dye remediation in anoxic-aerobic-anoxic microenvironment under period discontinous batch operation: Bio-electro kinetics and microbial inventory *Bioresour Technol* **119** 362-372

Verma H, Kumar R, Oldach P, Sangwan N, Khurana JP, Gilbert JA and Lal R (2014) Comparative genomic analysis of nine *Sphingobium* strains: insights into their evolution and hexachlorocyclohexane (HCH) degradation pathways *BMC Genomics* **15** 1014

Verma V, Raju SC, Kapley A, Kalia VC, Kanade GS, Daginawala HF and Purohit HJ (2011) Degradative potential of Stenotrophomonas strain HPC383 having genes homologous to dmp operon *Bioresource technology* **102** 3227-3233

Wasi S, Tabrez S and Ahmad M (2011) Suitability of immobilized Pseudomonas fluorescens SM1 strain for remediation of phenols, heavy metals, and pesticides from water *Water Air* and *Soil Pollution* **220** 89-99

Yadav TC, Khardenavis AA and Kapley A (2014) Shifts in microbial community in response to dissolved oxygen levels in activated sludge *Bioresour Technol* **165** 257-264