Plant Archives Volume 21, No 1, 2021 pp. 1739-1748 *e-ISSN:2581-6063 (online), ISSN:0972-5210*

Plant Archives

Journal home page: www.plantarchives.org

DOI Url: https://doi.org/10.51470/PLANTARCHIVES.2021.v21.no1.237

A REVIEW ON BIOREMEDIATION OF SUGAR MILL EFFLUENT

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(Date of Receiving-23-01-2021; Date of Acceptance-30-03-2021)

ABSTRACT

In the field of bio refinery, zero waste concepts have gained a lot importance in research impetus to boost the environment and bio economy in a sustainable manner. The wastewater from sugar industries contains miscellaneous compounds and need to be treated chemically or biologically before being discharged into water bodies. Efficient utilization of wastewater produced by sugar industries is a key point to improve its economy. Thus, interest in the sugar industry wastes has grown in both fundamental and applied research fields, over the years. Although, traditional methods being used to process such wastewaters are effective yet are tedious, laborious and time intensive. Considering the diverse nature of wastewaters from various sugar-manufacturing processes, the development of robust, cost-competitive, sustainable and clean technologies has become a challenging task. Bioremediation is emerging as an effective and attractive management tool to treat and recover the environment, in an ecofriendly manner. Bioremediation has been used at a number of sites worldwide, with varying degrees of success. Bioremediation, both in situ and ex situ have also enjoyed strong scientific growth, in part due to the increased use of natural attenuation, since most natural attenuation is due to biodegradation. Bioremediation technology, which leads to degradation of pollutants, may be a lucrative as well as environmentally friendly alternative. Biological treatment systems have various applications, such as cleanup of contaminated sites such as water, soil, sludge, and streams. Several methods have been designed and developed but more often, these process again produce secondary pollutants, which again are costing the environment. This review article discusses the role of microbes in sugar mill effluent treatment methods in different field and puts forward thoughts and scope for further research in the field.

Keywords: Bioremediation, Biodegradation, pollution, sugar mill effluent.

INTRODUCTION

Our earth was covered with about 70% of water and the lives cannot survive without water which plays important role in metabolism of living organism. Rapid industrialization, urbanization, intensive farming and other human activities have resulted in land degradation, environmental pollution and a decline in the crop productivity in all sectors of agriculture. Human activities have increased pressure on the natural resources and have become the source of a myriad of pollutants. Because of the anthropological activities of manmade things causes fresh water pollution at alarming pace. World Health Organization (WHO) says that 4% of deaths and 5.7% of disabilities in peoples are due to the water pollution (Peters and Meybeck 2000); (Matta and Kumar 2017). The industrial waste materials mixing with water bodies playing important mess in biological life activity of aquatic life (Ansari *et al*., 2012).

The first and greatly affected ecosystems in any country are the aquatic ecosystems, affected by either point or non-point source of pollution. Point sources of pollution occur when the polluting substance is emitted directly into the waterway. The common point sources of pollution are municipal and industrial wastewater effluents; run-off and leachate from solid disposal sites; run-off from industrial sites; run-off and drainage from industrial sites; discharge from vessels. The non-point source includes flow of water from agricultural fields and orchards, urban run-off from unsewered areas, etc. about 0.7 cubic meter of wastewater is generated per ton of crushed sugarcane (Samuel, 2011) and around 1.5-2 meter cubic water was consumed to process 1 ton of cane and generated approximately 1 meter cube of wastewater (Sahu and Chaudhari, 2015). The effects of water pollution are not only devastating to the aquatic organisms but also to the terrestrial animals and birds. More seriously, contaminated water destroys aquatic life and reduces their reproductive ability. Ultimately, the water becomes unfit for human consumption or domestic usage, at severe cases even hazard to human health. Waste disposal has an environmental cost and a financial cost too, which can be reduced by use of bio remediating agents. To speed up the bioremediation process, seeding of contaminated wastewater with competent microflora that are capable of degrading hazardous waste, is usually practiced in most treatments. The inoculated microorganisms either may be naturally occurring types or genetically engineered to attack the target waste. There are several initiatives being followed by Indian distilleries to minimize their water consumption and recycle the treated wastewater. In addition, research to address existing gaps is also necessary to provide cost effective solution to enable industries to consume lower water and zero discharge units.

The water soluble recalcitrant colouring compounds, melanoidins are highly resistant to conventional biological processes such as activated sludge treatment process. So, these waste water need pretreatment before its disposal into environment (Mohana *et al*., 2007; kumar and Chandra 2006; Anita *et al.,* 2013). Microorganisms due to their inherent capacity to metabolize a variety of complex compounds have been utilized since long back for biodegradation of complex toxic and recalcitrant compounds present in various industrial wastes for environmental safety (Gupta and Mukarjee, 2001). The aim of the review was to express current trends, application or role of microbes on bioremediation of industrial effluent and to contribute relevant background which is identified gaps in this thematic area and promisingly valuable process to solve.

Biological treatment of sugar mill effluent- a sustainable approach

The sugar industries, backbone for the rural, agricultural and socio-economic development of country. Basically, countries such as Brazil, India, china and Pakistan grows sugarcane as cash crop. The products and by-products of the sugar industry plays a vital role in the states development. In the sugar industry, water consumed in the different units for cleaning purposes such as washing of the milling house floor, clarifiers, vacuum pans, boiling house i.e. evaporators, and centrifugation, periodic cleaning of SO2 producing house and limewater are also the main wastewater contributors (Fito *et al*., 2018; Samuel and Muthukkaruppan, 2011). Notably, process house and mill houses are the two major sectors involved in wastewater generation in sugar industry. The process house wastewater is polluted with high organic matters, whereas the wastewater produced from mill house is contaminated mainly with suspended solids and grease and oil (Memon, Soomro, and Ansari 2006). It has been reported that 30,000-40,000 liters of sugar mill effluents was generated per tons of sugar processed (Bevan, 1971; Hendrickson, 1971; Belliappa 1991).But the disposal of industrial waste was the major cause for environmental pollution. The sugar mill effluent of various places were having different qualities and quantities (Jadhav *et al*., 2013) The wastewater of the sugar industry contains heavy metal (Cu, Pb, Mn and Zn), oil and grease and some other chemicals (Damodharan and Reddy 2012), which cause very adverse effects on the living organisms.

Sugar industry effluent has obnoxious odor and unpleasant color, contaminants such as chloride, sulphate, phosphate, nitrate and magnesium are discharged to agricultural lands (Samuel and Muthukkaruppa 2011; Fito *et al*., 2019). It causes injurious to plants, animals and human beings (Swamy *et al*., 2001; Hampannavar and Shivayogimath 2010; Samuel and Muthukkaruppan 2011). Therefore, direct utilization of sugar industry wastewater without any suitable treatment for crop irrigation substantially impedes seed propagation and seedling development (singh *et al*., 1985; Siddiqui and Waseem 2012). These effluents affect the seed germination, growth and yield of crop plants (Fakayode, 2005; Swamy *et al*., 2001). Effluents from the sugar mills with elevated COD and BOD levels can rapidly deplete the oxygen availability in the water bodies and compromise the life of aquatic flora and fauna (Elayaraj, 2014; Hampannavar *et al*., 2010; Siddique *et al*., 2012). The physico-chemical characters of sugar mill effluent had BOD of 1,000 – 1,500 mg/l (Jesudass and Akila, 1996; Priya and Kaushik, 2003; Adnan Amin *et al*., 2010).

Farmers using these effluents for irrigation to reduce water demand have found that plant growth and crop yield were reduced and soil health was compromised. Because sugar industry effluents are commonly used for irrigation, it is essential to determine how crops respond when exposed to industrial effluents. Many studies have indicated that the effluent discharge from sugar mill consist of a number of organic and heavy metal pollutant in dissolved or suspended form that can bring about changes in the physical, chemical and physiological sphere of the biota (Salequzzaman *et al*., 2008). The effluents of industries has ultimate disposal in agriculture field, which can alter the soil properties and crop yielding (Baskaran *et al*., 2009; Samuel and Muthukkaruppan, 2011; Saifi and Singh, 2011). Seed germination studies have been made on many crops such as green-gram, Sorghum, Black gram, Moong, Raphanus and Sugarcane (Doke *et al*., 2011, Siva and Suja, 2012, Elayaraj, 2014). Vaithiyanathan *et al*., reported the effect of sugar mill effluent on germination and growth of African marigold. Seed germination is a critical stage that ensures reproduction and controls the dynamics of plant populations, so it is a critical test of probable crop productivity.

Advances in science and technology and industrial revolution contribute to economical development and as a side natural resources such as water and soil get polluted (Haferburg and Konthe, 2007). The pollution has become global phenomenon, which has demanded attention from all over the countries. Bioremediation, natural and environmental friendly, cost, effective, aesthetically pleasant, soil organism friendly, diversity enhancer, energy derivation from sunlight (Chaney *et al*., 2005) and more important retain fertility factor of soil after removal of heavy metals (Kirkham, 2006).

Process of bioremediation

Bioremediation simply using biological agents used to clean up contaminated sites. It is an biotechnological approach involving microorganisms for solving or removing pollutants through biodegradation (El Fantroussi and Agathos 2005). Bioremediation refers to the optimization of naturally occurring remediation processes carried out by living organisms that degrade, alter or remove toxic organic compounds (Van Hamme *et al*., 2003). This biological strategy depends on the catabolic activities of organisms and on their ability to contribute to the degradation of contaminants of organic origin when using them as a source of food and energy (Pilon-Smits., 2005). Bioremediation is a process in which beneficial microbiological agents, such as yeast, fungi or bacteria are used to clean up contaminated soil and water. It is defined as the elimination, attenuation or transformation of polluting or contaminating substances by the application of biological processes.

Various classical physical and chemical methods such as flocculation, coagulation, filtration, sedimentation, and various combinations of these approaches have been attempted to remediate distillery spent wash and sugar industrial wastewater, but all these were found inefficient and non-effective (Sahu and Chaudhari 2015). Furthermore, the application of these approaches presents some ultimate disadvantages in terms of extreme chemical and energy necessities, generation of a large amount of sludge and toxic by-products (Thanapimmetha *et al.,* 2017). Therefore, increasing interest has been redirected toward biological treatment approaches, which are the potential ways for wastewater remediation to remove nutrients, solids, and organic matter (Valderrama *et al.,* 2002; Fito and Alemu 2019). It is revealed that the bio-based treatment method is highly efficient for intensely contaminated wastewaters discharged from the ethanol distilleries and sugar industries (Pant and Adholeya 2007).

In nature, the bacteria and fungi carry out the decomposition process and perform their function perfectly without having any side effects on the environment. In the biological treatment of industrial wastewater, microbes are used to dissolve the pollutant and convert them into harmless materials because they have the capacity to degrade the toxic material, which has no side effect on the ecosystem (Sirianuntapiboon and Ungkaprasatcha 2007). The principle of bioremediation is the encouraging microbes to work by providing optimum level of nutrients and other essential for the metabolism to degrade or detoxify the hazardous environmental pollutants. Its application often involves manipulation of environmental parameters to allow microbial growth and degradation to be occur in faster rate (Kumar *et al*., 2011; Abatenh *et al*., 2017). There are different types of treatment technologies or techniques come under bioremediation processes shown in figure 1.

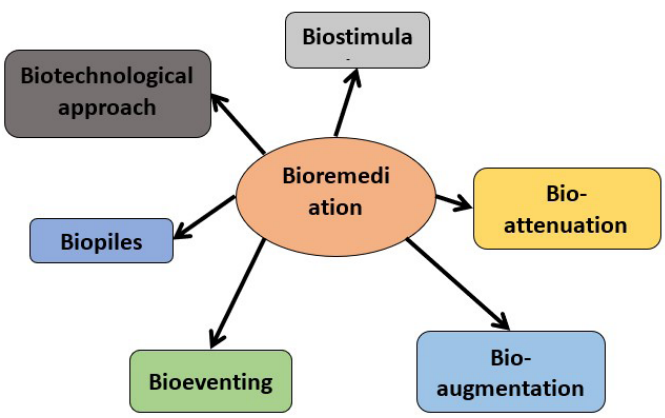

Figure 1: various types of Bioremediation process

It involved degrading, removing, altering, immobilizing, detoxifying the environment pollution. Biotic factors such as mutation, horizontal gene transfer, enzyme activity, interaction, growth, biomass, population size and composition (Boopathy 2000 and Madhavi and Mohini 2012). Abiotic factors such as pH, temperature, moisture, soil structure, redox potential and oxygen content, lack of training, physico-chemical bioavailability of pollutants such as contaminant concentration, type, solubility, chemical structure and toxicity (Madhavi and Mohini 2012). On the other hand, chemical methods sometimes affect the environment by producing secondary pollutants. The biological treatment has a very low cost as compared to physical and chemicals methods for wastewater treatment (Mohana, Desai, and Madamwar 2007).

Advantages and disadvantages of biological remediation methods

Various advantages and disadvantages of biological and physical/chemical treatment methods are summarized. That, Mohana, Acharya, and Madamwar (2009) reported that Better stability, low operating costs, and quick recovery after the starvation process. There was no need for recycling and solids separation and also provides resistance to shock loads and inhibitions. Even though, there must be some risk of clog, low reduction of pathogens and nutrients and needs long startup time.

Good performance and high efficiency at low cost, simplicity, efficient quality control in the effluent and flexibility of use and high biogas yield. The disadvantages are It is very slow process require several weeks, concentration gradients appear inside the systems, granulation process is difficult to control, and it requires proper monitoring to maintain the alkalinity to balance the excessive acidity (Suresh, Tripathi, and Gernal 2011; El- Kamah and Mahmoud (2012); Singh and Srivastava (2011); Fito, Tefera, and Van Hulle (2019b); Van Lier *et al.,* (2001); Ghangrekar, Joshi, and Asolekar (2003); Lettinga and Hulshoff Pol (1991))

Role of microbes in Bioremediation process

Microorganisms are the original bioremidiator, which restoring the original natural environment and preventing further pollution (Abatenh, 2017). Microbial bioremediation can cost-effectively and expeditiously destroy or immobilize contaminants in a manner that protect human health and the environment (Heitzer and Sayler, 1993; Gheewala and Annachatre, 1997; Gadd, 2000). A successful, cost effective, microbial bioremediation program is dependent on hydro geologic conditions, the contaminant, microbial ecology and other spatial and temporal factors that vary widely. In any bioremediation process the introduced microorganisms use the contaminants as nutrients or energy sources (Tang *et al*., 2007) . Bioremediation activity through microbes are stimulated by supplementing nutrients (Nitrogen and phosphorus), electron acceptor (oxygen), and substrates (methane, Phenol and toluene) or by introducing Microorganisms with desired catalytic capabilities (Ma *et al.,* 2007, Baldwin *et al*., 2008). A successful, cost

Table : 1 Microorganisms involved in bioremediation of sugar industrial effluents

Bacterial species	Process of remediation	References
Phaseolus aureus	Treated effluent will increase the seedling growth, chlorophyll and amylase content in green gram	Chandra *et al*., 2004
Azotobacter sp, Beijerinckia sp, Azospirullum sp	Microbes treated effluent increases sodium, nitrate and potassium concentration and decreases calcium, magnesium, heavy metals too	Sompony meunchang *et al*., 2006
Bacillus thuringiensis, Bacillus brevis and Bacillus sp	Decolorized synthetic melenoidins	Kumar & Chandra 2006
Phosphobacterium	Increases seed germination and seedling growth in wheat and black.	Kamlesh nath *et al*., 2007
Pseudomonas aeruginosa, Stenotrophomonas maltophila, Proteus mirabilis	Capable to decolorize anaerobically	Mohana *et al*., 2007
Pseudomonad fluorescence	Decolorize melanoidin from sugar effluents	Mohana, Desai, and Madamwar, 2007
Lactobacillus plantarum	Decolorize melanoidins under aerobic condition	Tondee & Sirianun-tapiboon 2008
Bacillus licheniformis, Bacillus sp and Alcaligenes sp.	Consortium decolorised the melanoidin pigment very effectively	Bharagava *et al*., 2009
Alcaligenes faecalis	Decolorization and treated effluent was environmentally safe	Santal *et al*., 2011
Bacillus subtilis, serratia marcescens & enterobacter asburiae	Remediate about 73% of sugar mill effluent	Saranraj & Stella, 2012
Proteus mirabilis, Bacillus subtilis, Serratia marcescens, Enterobacter asburiae, Pseudomonas fluorescens, Bacillus weihenstephanensis, Pseudomonas aeruginosa, Staphylococcus aureus, Escherichia coli and Brevibacterium halotolerance	Bioremediation using bacteria shows reduction in physic-chemical characteristics of sugar mill effluent and Bacillus showed maximum effect	Saranraj & Stella, 2012
Lactobacillus plantarum,	Decolorize melanoidins in the cane sugar wastewater	Kryzwonos 2012
Bacillus consortia of C1 and C2	Decolorize melanoidins in the cane sugar wastewater	Kryzwonos 2012
Proteus mirabilis, Bacillus sp, Raoultella planticola & Enterobacter sakazakii	Consortia ratio of 4:3:2:1 decolorize 75% of melanoidins within 10 days	Yadav & Chandra, 2012
Denitrifying bacteria	Biological denitrification of waste water using sugar industrial waste molasses as corbon source and bagasse charcoal pellets as supporting media	Kumar & sahu *et al*., 2013
Bacillus subtilis and pseudomonas fluorescens	Bacterial consortium immobilized in sodium alginate gel reduced higher percentage of physic-chemical characters in sugar mill effluent	Jagannathan *et al*., 2014
Trichoderma harzianum, pseudomonas fluorescens	Degradation and reduction of pollutants, decolorization of effluent was very effective. Germination of chick pea was high in treated effluent sample	Kashif & Monowar, 2017
Enterococcus faecalis	Lactic acid production in fluidized bed biofilm reactor	Narayanan *et al*., 2017
Fungal species		
Trametes versicolor	Molasses wastes	Benito *et al*., 1997
Rhizopus sp	Sugar Industrial effluent remediation	Dursun *et al*., 2003
Azospirillium brasilense	Sugar mollase degradation	Dursun *et al*., 2003
Penicillium pinophilum, Alternaria gaisen, Aspergillus flavus, Fusarium monolifome, Aspergillus niger	Remediation of sugar industrial effluent	Pant and Adholeya, 2007
Phanetochaete chrysosporium	Treatment of sugar mill waste water	Prabakar *et al*., 2010

continue...

Aspergillus niger, Penicillium sp and Fusarium sp	*Aspergillus niger* shows maximum degradation pH(-38.63%), EC(18.76%), TDS(18.74%), BOD(47.62%) and COD(44.68%)	Buvaneswari *et al*., 2013
Clostridium sp & Thiobacillus ferrooxidans	Bioelectricity generation from the sugar mill waste matter	Ravinder kumar *et al*., 2016
Aspergillus, Rhizopus, Alternaria sps, Bacillus sp and Staphylococcus sps	The sugar mill effluent contains both the fungal and bacterial flora	Sangeeta *et al*., 2017
Aspergillus niger, A. flavus, A. terreus, Penicillium verruculosum, Fusarium oxysporum, and Curvularia lunata	Sugar industry effluent stimulated the growth of all these fungal strains of Rhizosphere mycoflora	Shaila Sakhala, 2020
Microalgae strains		
Oscillatoria boryana	Distillery melanoidin pigment	Kalavathi *et al*., 2001
Chlorella vulgaris	Recalcitrant wastewater	Valderama *et al*., 2002; Travieso *et al*., 2008
Chlorella sorokiniana	Alcohol distillery wastewater	Solovchenko *et al*., 2014
Spirogyra sp	Maximum removal of TDS, EC, pH, Mn after 60 days	Vinod kumar *et al*., 2016
phytoremediation		
Polyalthia longifolia, Moringa olefera, Tamarindus indicus, Samanea saman, Azadirachta indica and Acacia nilotica	All tree species showed remarkable reduction of physico-chemical properties of sugar mill effluent	Baskaran *et al*., 2009
Other methods		
Biphasic high rate reactor	sugar industry and ethanol distillery wastewater treated the blended wastewater through a two-stage anaerobic reactor	Fito *et al*., 2018
Adsorption of bagasse fly ash (BFA).	Sugar industry and ethanol distillery wastewater were blended and treated using anaerobic digestion	Fito *et al*., 2019

effective, microbial bioremediation program is dependent on hydro geologic conditions, the contaminant, microbial ecology and other spatial and temporal factors that vary widely. In any bioremediation process the introduced microorganisms use the contaminants as nutrients or energy sources (Tang *et al*., 2007) . Bioremediation activity through microbes are stimulated by supplementing nutrients (Nitrogen and phosphorus), electron acceptor (oxygen), and substrates (methane, Phenol and toluene) or by introducing microorganisms with desired catalytic capabilities (Ma *et al.,* 2007, Baldwin *et al*., 2008). Some common microorganism used in the process of remediation are *Acromobacter, Alcaligenes, Arthrobacter, Bacillus, Cinetobacter, Corneybacterium, Flavobacterium, Micrococcus, Mycobacterium, Nocardia, Pseudomonas, Vibrio, Rhodococcus* and *Sphingomonas* species (Gupta *et al.,* 2001, Kim *et al,* 2007, Jayashree, 2012). The main species involved in effective sugar waste water treatment include lactic acid bacteria-*Lactobacillus plantarum, L. casei and Streptococcus lacti* and Photosynthetic bacteria-*Rhodopseudomonas palustrus, Rhodobacter spaeroide*, etc. (Narmatha and Kavitha, 2012). Microbes possessing such novel properties can be either isolated from natural contaminated sources or obtained through engineering process (Pradeepa *et al*., 2017). Table 1 present as

Bacteria

A wide diversity of bacterial strains such as *Pseudomonas aeruginosa, Pseudomonas putida, Lactobacillus planatarum, Bacillus circulans, bacillus megaterium, bacillus firms, bacillus thuringiensis, bacilluscereus, lactobacillus hilgaridii, lactobacillus coryniformis, xanthomonas fragariae* were reported for its property in degradation and decolorization of sugar industrial effluents (Bezuneh 2016). Both aerobic and anaerobic bacterial strains are not economical due to higher energy consumption for aeration. Nowadays aerobic bacterial strains are advantageous due to low energy consumption and minimize cost of wastewater treatment (Ohmono *et al*., 2011).

Fungi

Fungi, their biology, economic value and pathogenic capabilities are not new to human society. They have been used from fermentation of foods to production of pharmaceuticals. Fungi thrive well in inhospitable habitats with environmental extremes because of their enzyme system (Cooke, 1979). Fungi are involved in the biodegradation of undesirable materials or compounds and convert them into harmless, tolerable or useful products.

Many organisms are involved in the biodegradation of organic waste, which has resulted in the production of novel substances of biotechnological importance.

Fungi are recognized for their superior aptitudes to produce a large variety of extracellular proteins, organic acids and other metabolites, and for their capacities to adapt to severe environmental constraints (Lilly and Barnett, 1951; Cochrane, 1958). Fungal systems appear to be most appropriate in the treatment of colored and metallic effluents (Ezeronye and Okerentugba, 1999).

Fungi not only produce various metabolites like citric acid, homogeneous proteins, heterogeneous proteins, peroxidases but have shown their effectiveness for removal, reduction and detoxification of industrial effluents ingredients. Therefore in this review paper an attempt has been made to bring out the capabilities of fungi for bioremediation of industrial effluents. Bioremediation refers to the productive use of microorganisms to remove or detoxify pollutants, usually as contaminants of soil, water or sediments that otherwise threaten public health. Microorganisms have been used to remove organic matter and toxic chemicals from domestic and industrial waste discharged for many years (Gupta and Mukerji, 2001).

Algae

Microalgae are unicellular phototropic microorganisms known for their capacity in biological adsorption and degradation of toxic chemical pollutants as phenols, heavy metals, pesticides, polycyclic aromatic hydrocarbons, Xenobiotics and melanoidins from the industrial effluents which directly mixed to the water bodies (Maynard *et al*., 1999; Shashitekha *et al*., 1997). Compared to the bacterial and fungal system, the algal remediation is more advantageous due to its high potential to utilize contaminants such as ammonium, nitrate and phosphate as nutrient hence minimizes nutrients; it can grow rapidly and adapt very harsh conditions. The main advantage was microalgae produce valuable products such as ethanol, methane, live-stock feed, used as organic fertilizer (Mata *et al*., 2010). Various species of microalgae such as *Chlorella vulgaris, Oscillatoria boryana, Chlorella pyrenoidosa, Chlorella sorokiniana, coenochloris pyrenoidosa, Nostoc muscorum, Neochloris oleoabundas, Phormidium valderianum, Chlorella zofingiensis and Chlorella ellipsoidea, etc* have been used in the process of bioremediation of waste

CONCLUSION

Bioremediation is the cleaning up process of environmental pollution by technically enhancing the natural biodegradation process. By understanding the microbial communities and their response to remediate the environment from pollution. The exploring knowledge of microbial diversity will improve our capabilities to degrade pollutants. As natural resources are the major assets to humans, bioremediation of contaminated sites and ecosystems will be the best ecofriendly approach to presence and also ensure efficient recycling of wastes. It is the peak research field, which producing many products to improve nutrient release in agricultural land, improving composting of industrial waste, removal of toxic chemicals, heavy metals and so on. However, large number of reports on fungal, bacterial and algal treatment has been limited to laboratory level experiments. The large scale process was still inconvenient due to lack of stability, nutrient, supplement, growth cycle, spore formation, loss of extracellular enzyme, reactor system, etc. In future research has to be carried out in contaminate based by isolate, characterize and genetically improve microbes for better bioremediation yield.

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