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Review

Microbial degradation of toluene

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Zephirus environment is polluted by the toluene from profuse range industries. In spite of positive potential application, toluene results in many mishaps especially health hazards; hence amputation of toluene is crucial for human welfare as well as environmental issues. This review deals with destruction of toluene using microbial degradation. The overall aerobic biodegradation of toluene into carbon dioxide, water and biomass by bacteria, fungi and mixed culture has been studied comprehensively. Biodegradation technique has a promising prospective for the reason that it results in effective and economical treatment than traditional techniques. The amputation of toluene contributes to an assortment of various techniques utilized at various levels of investigation which are characterized based on quantification techniques. A comparison of the removal capacity based on continuous and batch mode of operations was studied proficiently. This methodology opens up a window for future exploration in field of biodegradation of toluene.

Key words: Toluene, biodegradation, microorganism, health hazards, bioreactor.

INTRODUCTION

Toluene, formerly known as toluol, is a clear, water-insoluble liquid with the typical smell of paint thinners. It is a mono-substituted benzene derivative, that is, one in which a single hydrogen atom from six groups of atoms from the benzene molecule has been replaced by a univalent group, in this case CH_3 (Michael et al., 2011) (Figure 1).

It is an aromatic hydrocarbon that is widely used as an industrial feedstock and as a solvent. Like other solvents, toluene is sometimes also used as an inhalant drug for its intoxicating properties; however, inhaling toluene has potential to cause severe neurological harm. Toluene is an important organic solvent, but is also capable of dissolving a number of inorganic chemicals such as sulfur (Broberg et al., 2008). Toluene (methylbenzene, toluol, phenyl methane) is an aromatic hydrocarbon (C_7H_8) commonly used as an industrial solvent for the manufacturing of paints, chemicals, pharmaceuticals, and rubber. Toluene is found in gasoline, acrylic paints, varnishes, lacquers, paint thinners, adhesives, glues, rubber cement, airplane glue, and shoe polish. At room temperature, toluene is a colorless, sweet smelling, and a volatile liquid. The name toluene was derived from the older name toluol, which refers to tolu balsam, an aromatic extract from the tropical Colombian tree Myroxylon balsamum, from which it was first isolated (Stine, 1943). It was originally named by Jöns Jakob Berzelius. Benzene, also known as C_6H_6, or benzol, is an organic chemical compound which is a colorless and flammable liquid with a pleasant, sweet smell. Benzene is a known carcinogen. It is a component of gasoline. Toluene reacts as a normal aromatic hydrocarbon towards electrophilic aromatic substitution. The methyl group makes it around 25 times more reactive than benzene in such reactions. It undergoes smooth sulfonation to give p-toluenesulfonic acid, and chlorination by Cl_2 in the presence of $FeCl_3$ to give ortho and para isomers of chlorotoluene. Toluene is a type II carcinogenic agent, an aromatic hydrocarbon, which was commonly used as industrial solvent. The repeated exposure of toluene to the pregnant ladies may increase the risk of damage to the fetus. It is illegally abused with spray paint and inhaled through nose and mouth to get euphoria sensation, which lead to death or unconsciousness. The Occupational Safety and Health

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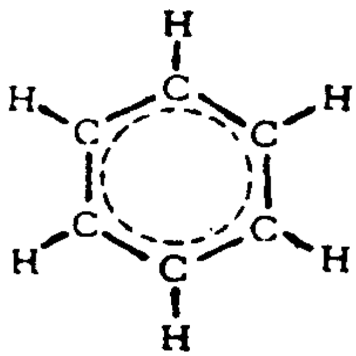

Figure 1. Structure of toluene.

Table 1. Methods available for toluene degradation.

Methods used for degradation	Reference
Ozone	Aydin et al. (2011)
Absorption	Byungjoon et al. (2008)
Adsorbent	Tham et al. (2011)
Biofiltration	Ortiz et al. (2003)
Incineration	Raul et al. (2007)
Catalytic oxidation	Faisal et al. (2000)
Condensation	Raul et al. (2007)
Membrane	Madan et al. (1996)
Photochemical oxidation	Zaishan et al. (2010)
Activated carbon	Farshid et al. (2010)

Administration (OSHA) have determined the acceptable level of occupational exposure to toluene for people in the workplace. The Permissible Exposure Limit (PEL) of 200 ppm is considered as an acceptable level of exposure with a time-weighted average for an 8 h workday. Toluene exposure level of 500 ppm is considered as an immediate dangerous level to life and health.

TOLUENE

Toluene is an aromatic hydrocarbon which is used as an industrial feedstock and as a solvent. It is also used as an inhalant drug for its intoxicating properties; however, inhaling toluene has potential to cause severe neurological harm (Sercu et al., 2005). It is an important organic solvent capable of dissolving a number of notable inorganic chemicals. Toluene occurs naturally at low levels in crude oil and is usually produced in the processes of making gasoline or making coke from coal (Aydi et al., 2011). Final separation takes place in a Benzene-Toluene-Xylene plant. It has been found that toluene is illegally combined with petrol in fuel tank outlets for sale as a standard vehicular fuel. It attracts no fuel excise, while other fuels are taxed over 40%, hence the fuel suppliers are able to profit from substituting the cheaper toluene for petrol. This substitution is likely to affect engine performance and result in additional wear and tear. In the recent finding, toluene has been used as one of the component for jet fuel surrogate blends.

Techniques used for toluene amputations

More rigorous requirements for removing toluene from atmospheric air in recent era necessitate the expansion of innovative cost effective treatment alternatives. Numerous physical and chemical gas cleaning methods such as biofiltration, bioremediation, biodegradation, absorption, incineration, wet scrubbing, adsorption, thermal and catalytic oxidation have been developed to treat BTX (Table 1). The costs of chemical energy consumption and further treatment or disposal of secondary waste are the two major issues when compared with conventional techniques. Biofiltration is an inexpensive *modus operandi* and efficient in treating large volumes of soggy air streams with low concentration of toluene (Farshid et al., 2010). Biofiltration is a pollution control technique that uses living material to capture the biologically degraded process pollutants. Common uses include processing waste water, capturing harmful chemicals or silt from surface runoff, and micro biotic oxidation of contaminants in air. The elementary principle of biofiltration of polluted air is that gaseous pollutants are destroyed in the process being converted into carbon dioxide, water and biomass by microbial metabolic reactions (Byoung et al., 2008). During the biofiltration, polluted air is passed through biofilter medium where the pollutant is transferred from gas to liquid solid phase where they are degraded by biofilim containing various microorganisms which favors the suitable environment for toluene removal.

MICRO-ORGANISMS USED FOR THE BIODEGRADATION

Mostafa et al. (2009) examined biofiltration

Aerobic microbial species degrade contaminants in 2 steps: One is oxidation of ketone functional group by molecular oxygen and second one is breakdown of energized molecule to CO_2 and water. The contaminant diffuses through pores on the active biofilm which is formed by the microbial population (Adriana et al., 2001). Selectivity of the microbial population is one of the important factor for the aggrandize of removal efficiency and the performance. Wan et al. (2003) reported that the major groups of microbes are bacteria, virus, fungi, protozoa, actinomycetes, algae, and helminthes (Table 2). They interact with the environment and are used in several industries to acquire the desired products. Bacteria and fungi with its dehydrogenase activity (an indicator) are the two dominant groups used for biofiltration.

Table 2. Removal of toluene by different type of reactor and species with their removal efficiency.

Microbes used to degraded	Types of packing material used in reactor	Removal efficiency (%)	Reference
Rhodococcus erythropolis	Peat biofilter	99	Malhautier et al. (2008)
Indigenous to filter media	Palm shells and activated sludge biofilter	100	Pakamas et al. (2005)
Rhodococcus fascians	Peat moss and sugarcane stem (bagasse) biofilter	100	Yi-chen et al. (2011)
Paecilomyces lilacinus	Perlite biofilter	53	Gabriel et al. (2008)
Acinetobacter sp. *Rhodococcus rhodochrous*	Peat and glass beads biofilter	100	Mario et al. (2001)
Consortium	Vermiculate biofilter	90	Pineda et al. (2000)
Mixed culture	Polyurethane foam biofilter	100	Singh et al. (2010)
Consortium	Peat, vermiculite, A mixture of vermiculite and activated carbon, tree bark and, porous glass rashig rings biofilter	65 to 95	Ortiz et al. (2003)
Paecilomyces variotii	Ceramic rings biofilter	70	Aitor et al. (2005)
Sphingobacterium multivorum; Comamonas testosteroni Pseudomonas; putida Pseudomonas fluorescens; Chryseobacterium indologenes	compost and perlite biofilter	70	Eva et al. (2006)
Penicillium sp. *Aspergillus niger Trichoderma viride*	-	75 to 92	Bhuvaneswari et al. (2012)
P. putida	inorganic/polymeric composite chip Biofilter	80	Byoung et al. (2008)
Cladosporium sphaerospermum	Biofilter	-	Frans et al. (1995)
Alcaligenes xylosoxidans	Biofilter	98	Andrew et al. (2003)
Phanerochaete Chrysosporium	powdered compost biofilter	92	Seyed et al. (2011)
Mixed culture	Membrane biofilter	84	Madan et al. (1996)
Alcaligenes (Achromobacter) xylosoxidas	Ceramic enters to activated carbon bed bioactive foam emulsion bioreactor	89.59	Farshid et al. (2010)
Mixed culture	Coal biotrickling filter	100	Anil et al. (2008)

Biodegration of volatile organic compounds (VOC) by using bacteria

Bacteria are the oldest form of cellular life. They are also the most widely dispersed and conceivable microclimate on the planet. They vary in shape, size and in arrangements. General shapes include Cocci, Bacilli, Spirilla and Spirochetes. They are identified by means of microscopic, macroscopic as well as by biochemical, serological, molecular and genetic characteristics. Non odorous sulphide groups are synthesized by bacteria by means of oxidizing ionic sulphide groups in an aerobic condition during biofiltration. Bacterial microorganism such as *Pseudomonas putida, P. flurorescence. Rhodocous faccians, Alcaligens xyloxidans, Burkholderia cepacia, Hypomicrobium, xanthobactor, Acinetobactor* are utilized for the degradation of VOC's. Luis et al. (2006) reported that the performance efficiency of biofilter reaches almost 100% when fed with 0.5 g/m^3 of the pollutant or less while using bacterial population. Particularly, *P. putida*has maximum VOC removal efficiency of 83% with the inlet of 1 g/L pollutant concentration.

Biodegradation of volatile organic compounds (VOC) by using fungi

Babak et al. (2009) described that fungus microbial population such as *Cladosporium sphaerrospermum, F. solani, Paecilomyces variotii, Pacilomyces,* and *Phanerochaete chrysoporium* is used for the VOC's degradation. Due to enzymatic oxidation by lactases and peroxidase, fungi tend to degrade more complex molecules. They are able to colonize and degrade a great member of substrates by using them as the sole source of carbon and energy; for this reason they are largely employed in the biotechnological process. Long duration of operation with high VOC's organic load leads to dryness, acidification, and nutrient depletion which decrease in the microbial activity and VOC's removal efficiency. Fungi is highly preferred than bacteria because of their tolerant capacity in growth during the environmental changes like temperature, pH and humidity; bacteria require more than 0.9 water in the environment for its activity whereas fungi can withstand its growth even in the decrease of moisture content up to 0.6. Hypothesized aerial mycelia of fungi in direct contact with the gas phase can offer faster contaminant mass transfer rate than flat aqueous bacterial biofilter surface.

Biodegradation of volatile organic compounds (VOC's) by mixed culture

Sandeep et al. (2010) isolated microorganism from sewage sludge or strain and immobilized this on the surface of packing material. Generally, biofilm contains between 10^6 and 10^{10} colony-forming unit (CFU) of bacteria and antinomycetes per gram of bed changes of the number of strains in the biofilm relates the removal efficiency of VOC's. Mixed culture produce high RE of organic compounds in VOC than pure culture because it can release several toxic intermediates during biodegradation (Raul et al., 2007).

BIOFILTRATION

Biofiltration is a novel biological process used to treat contaminated air that takes advantage of the ability of micro-organisms to transform organic and inorganic pollutants into less toxic compounds (Zarook et al., 1997a) than other conventional techniques (Zarook et al., 1997b; Zarook et al., 1998a). The proliferation of fungi in biofilters is frequent due to their ability to tolerate acid and low humidity conditions (Kennes and Veiga (2004). Fungal biofilters have shown greater elimination capacities for some hydrophobic compounds when compared to their bacterial counterparts. The complete aerobic biodegradation of toluene to carbon dioxide, water and biomass by bacteria has been studied extensively (Perry and Green, 1997). It is considered as an alternative to air pollution control technologies, because of low cost. Both fungi and bacteria are known to degrade toluene and other aromatic hydrocarbons. It has been proved that fungi played a vital role of degrading benzene, toluene, ethylbenzene, and xylene (BTEX) than bacteria. Biofiltration has proven to be an applicable method for gas treatment since it is economical compared to other eight techniques. The contaminant removal efficiency is very high and also minimum CO_2 is produced by this technology (Mostafa et al., 2009). Fungi show significant preferences compared to bacteria since they are most resistant to environmental changes such as temperature, pH and humidity. For instance, the water activity, which is defined as the amount of required water that is free in a given environment is 0.6 for fungi where as it is more than 0.9 for bacteria (Seyed et al., 2011). Using different species, the degradation of benzene, toluene, ethyl benzene and xylene are preceded under bioreactors.

Removal of toluene using *Cladosporium sphaerospermum*

Frans et al. (1995) reported that the growth of eukaryotic organism with toluene is the sole source of carbon and energy. The fungus *C. sphaerospermum* was used for the removal of toluene from waste gases. The oxygen consumption and enzyme activities of toluene grown *C. sphaerospermum* indicate that it is degraded by initial attack on the methyl group. They discussed that the fungi involved in biofiltration system results in complete biodegradation of toluene. Due to the interaction of methyl group with fungi, the oxygen consumption and biomass are high in rate.

Removal of toluene by *Paecilomyces variotii*

Elena et al. (2005) reported that the biodegradation of toluene using two fungal strains namely *P. variotii* was isolated on it as the sole carbon and energy source, mineralizing the substrate into carbon dioxide. It is rare that fungal strains isolated degrade the pollutant. The fungus degraded over the range of pH 3.9 to 6.9 and temperature range of 23 to 40°C. The activity of strains decreased gradually at pH<4, so the use of nitrate was tested, which maintains the constant pH instead of ammonium. So far fungal growth on alkyl benzenes has not been reported at 40°C. But this work has proved that nitrate was the suitable nitrogen source for *variotii* strain fungal growths on alkyl benzenes at 40°C; showed its activity on higher temperature which was included.

Removal of toluene by *Paecilomyces lilacinus*

Gabriel et al. (2008) were the first to report the use of *P. lilacinus*, which consumed toluene as the sole carbon source. In a gas-phase bio-filter, along with packing material, the species obtained an average elimination capacity of 50 gm^{-3} h^{-1}. It has the removal efficiency of 53% and a final biomass of 31.6 mg biomass gm^{-1} dry support. The molecular weight of hydrophobin protein, which was produced from mycelium, was identified after its purification and precipitation. It confirmed that hydrophobin protein presented high homology with sequences of class I hydrophobin proteins from other ascomycetous fungi and the same was compared against national center for biotechnology information database. *P. lilacins* yielded about 1.1 mg hydrophobin per gm biomass. These proteins modified the hydrophobicity of Teflon by lowering the contact angle from 130.1° to 57° for supporting sodium dodecyl sulfate (SDS) washing.

Removal of toluene using a fungal biofilter

Seyed et al. (2011) showed that during intermittent loading, the performance of a compost bio-filter treating toluene vapor was investigated by a bio-filtration system set up. The system was inoculated with white rot fungus, *Phanerochaete chrysosporium* and was loaded 10 h per day on different air flow rates (0.096, 0.024, 0.06 m^3/h) and pollutant concentration (173.1 and 52.6 mg m^{-3}) and there was no aeration during the remaining 14 h of the day. The maximum removal efficiency of toluene and its elimination capacity obtained were about 92% and 1913.7 mg m^{-3} h^{-1}. By application of Monod type equation, the kinetic of biological reaction was studied and its constants were evaluated as 3.495 gm^{-3} h^{-1} and 50 gm^{-3} h^{-1}. Under harsh condition, fungal system could remove toluene effectively without adding excess nutrient solution.

Removal of toluene by fungi using biofiltration contains ceramic rings

Different packing materials were used for the inhibition of mycelia growth in fungal bio-filters. As we are aware that, fungal bio-filters are better than bacterial bio-filters for the removal of toluene and also other aromatic hydrocarbons, here Aitor et al. (2005) used fungus *Paecilomyces variotii* CBS 115145. The organisms were tested with two rigid packing materials that attempts high growth of mycelia. The two bioreactors containing 4.25 L sections was taken and packed with ceramic Raschig rings, provided with water retention capacity and internal porosity. The maximum elimination capacity was found to be 245 gm m^{-3} h^{-1} which was measured after optimization. Under lower water content, higher elimination capacity of about 290 gm m^{-3} h^{-1} was measured in the ceramic ring. Also, he worked with 2 L sections of bio-filters which results in bacterial contamination but the fungal activity responded with 70% removal of toluene.

Biodegradation of toluene by *Exophiala oligosperma*

Elena et al. (2005) stated that fungi are more resistant to extreme environmental conditions than most of the bacteria such as low pH, reduced water content, etc. Such characterizations are also interesting in soil biodegradation in environments hostile to bacterial activity. Indeed, it is recently observed that high toluene removal efficiencies can be obtained in biofilter with filamentous fungi rather than bacteria being dominant. In India there is no autonomous regulation in and of itself to ascertain VOCs emission. Since the haze of photo-chemical gradually increases in the atmosphere due to the oxidation of VOCs, OSHA (Occupational Safety and Health Administration) and title-1 US-EPA (United States-Environmental Protection Agency) takes stringent effect to direct with the citation of standard air degree of ozone 0.12 ppm where is mandatory. The vital role of microbes for biofiltration is enormous and its above factors were investigated in this present study. This review open ups the growing innovation for future research in the field of destruction of VOCs.

Removal of toluene in waste gases using a biological trickling filter

Anne and Erik (1995) reported that many volatile organic compounds are discharged to the atmosphere from human activities and from natural process. Those techniques are applicable for emissions of volatile organics in low concentration which are biologically degradable and to some extent water soluble. Only few mathematical models have been proposed for the removal of volatile organics in different types of biological filters in the literature until now. The model consists of a set of non - linear differential equations, which can be solved by a numerical procedure.

Toluene removal from air using differential biofiltration reactor

Abraham (2008) describes VOC's as known environmental pollutants that are often toxic and/or carcinogenic as well as contributors to odour problems, global warming and ozone depletion. One of the advantages of using biological air pollution control methods is that the pollutants are actually destroyed by biological oxidation. A downside is that some pre-treatment of the air and pollutant may be required, for example pre-filtration for particulates, temperature adjustments and humidification of the gas flow.

Removal of benzene, toluene, ethylbenzene, and xylene (BTEX) by isolated fungal species

Francesc et al. (2001) suggested that in air filters, the fungi grown at low pH and low water activities favored the purification of gas streams containing aromatic hydrocarbons. Some of the aromatic hydrocarbons are benzene, toluene, ethyl benzene and the Xylene isomers, which are collectively known as BTEX. They are considered as most abundant components from the water soluble fraction of crude oil and refined fuels. From enrichment cultures, five fungal strains were isolated. In this study three different techniques were used namely solid state-like batches, air bio-filters and liquid cultures. The pollutant soil near petroleum industries were used for inoculation. The isolates were identified as *deuteronmycete* and the *ascomycete*. The kinetic parameter such as the apparent half saturation (ranging from 5 to 22 M) and the concentrations of toluene inhibiting degradation activity were compared and were reported. The degradation activity was inhibited by 50% at toluene concentrations ranging from 2.4 to 4.7 mM. The results show that fungus grown at low pH and low water activities can be used for the purification of gas streams by the doubling times of about 2 to 3 days.

Removal of AMEETX by fungi using biofilters

Bing and William (2006) suggested that the culture grown under acidic conditions (pH 3) was inoculated with polyurethane foam medium in two bio-filters, supplied with gas stream containing five component mixture (acetone, methyl ethyl ketone, ethyl benzene, toluene and *p*-Xylene). The loading rate of total VOC of about 80.3 gm m^{-3} h^{-1} is used to simulate treatment of air emissions. One bio-filter was operated under continuous loading conditions and another was supplied with contaminants 8 h/day. Nutrient solutions are supplied with pH 3 and the work presented the data which can be used to treat paint solvent mixtures in bio-filters. After startup of three weeks, the bio-filter received continuous loading and provided higher removal efficiency while intermittent loading showed the efficiency after six weeks. The performance of bio-filter with continuous loading is about 97 to 99%. The results show that the degradation of ketone in both filters was more and aromatic hydrocarbons were unstable even after continuous operation for two months.

Removal of isopropyl alcohol from air stream using biofiltration

Smita and Babu (2010) described volatile organic compounds (VOCs) as the large group of organic compounds emitted into the atmosphere by a wide range of industries and are one of the major pollutants released by the industries which contaminate the atmospheric air and the fresh water resources. Due to its adverse effects, it should to be removed from the waste gaseous streams. Out of various techniques such as incineration, ozonation, combustion, and adsorption, biofiltration has emerged to be a cost effective technology for eliminating odorous and toxic VOCs from waste gas streams in recent years. Biofiltration takes place by combination of basic processes such as absorption, adsorption and degradation. Morales et al. (1998) reported that the quality of aquatic environments is deteriorating due to raw and toxic effluent discharges. Detergents are an important source of pollution and often transferred to waterways by industrial and domestic effluents.

Removal of n -hexane by *F. solani* with a gas-phase biofilter

Sonia and Sergio (2005a) exploited biofiltration as a multi- step process involving the partition of the gaseous Raul VOS and oxygen to the liquid phase containing the microorganisms followed by transport through the biofilm to the cells. A filamentous fungus has shown promising results for the degradation of hydrophobic pollutant. Sabrina et al. (2007) suggested the gradual increase in degradation rates measured in time suggested the adaptation of microbial communities towards this class of pollutants. Kyung et al. (2010) reports that the toluene was selected as a representative compound because it is widely used as a solvent and a primary raw material in industries but biodegradation rate is relatively low. However, their release into the environment is strictly controlled and they are classified as prioritized environmental pollutants by the U.S. Environmental Pollutant Agency because they are suspected to be carcinogens and can produce offensive odours. Biofiltration seems to be a proven attractive technology for the treatment of waste gases containing relatively low concentration of VOCs, because of its simplicity, low cost, and non-generating hazardous residues.

Removal of isopropyl alcohol from air stream using biofiltration

William and Randall (1996) suggested that the biological treatment of VOCs has been attracting more attention these days than physical and chemical methods as they are cost effective and create almost negligible secondary pollutant. The need for alternate cost effective treatment technologies led to similar biological treatment processes for waste gas streams. In biofiltration, a humid, contaminated air is passed through a porous support material on which pollutants degrading cultures are immobilized.

Removal of VOCs from air by zeolite based adsorption-catalytic combustion system

Wang et al. (2007) stated that emissions of VOCs do not necessarily give rise to health or environmental concern. Ground-level ozone, the primary component of smog is formed when oxides of nitrogen (NOx) and VOCs react in the presence of sunlight. Ozone is not usually emitted directly into the air, but at ground level, it is created by a chemical reaction between NOx and VOC in the presence of sunlight. Motor vehicle exhaust and industrial emissions, gasoline vapors, chemical solvents and the natural sources emit NOx and VOC that helps to form ozone. Sunlight and hot weather cause ground-level ozone to form in harmful concentrations in the air.

Biodegradation of mono-chlorobenzene by using a trickle bed air biofilter (TBAB)

Anil et al. (2008) illustrated that although, many researchers have reviewed the principles of biological waste air treatment and the advantages over chemical and physiccal techniques, research is still going on to the biological treatment of waste air for the use of effective new packing media, designs, microbial structure analysis and modeling of VOC removal. The use of surfactants has the potential to increase the biodegradation rate of hydrophobic organic compounds in contaminated environments by increasing the total aqueous solubility of these compounds. Although the contaminants encountered indoors may have a substantial impact on occupant health, their concentrations in indoor air are low when compared to those encountered under industrial conditions.

Removal of xylene using a bioreactor containing bacteria and fungi

Li and Liu (2006) proposed that Xylene from off-gas using a bioreactor containing bacteria and fungi can be removed and that work has been attempted by Research Centre of Eco-environmental Sciences and Chinese Academy of Sciences. The work is all about volatile organic compounds, which is considered as constituents of petroleum and fine chemical industries and as carcinogens that can cause offensive odors. A bench-scale combined-bioreactor consists of two zones that is, one zone containing a bacterial suspension and another zone with packed material for growth of fungi. Under steady state, Xylene along with its three isomers is eliminated with capacity of 62 $gm^{-3}h^{-1}$. The removal rate of Xylene was >90 to 24% in the first zone and 67.6% in the second zone. The liquid and gas flow ratios influenced the removal efficiencies of the isomers in both the zones. When the flow ratio of liquid and gas is >0.15, the removal efficiency was increased in the first zone while it decreesed in the second zone. When the zones of the reactor differed, the Xylene was degraded by different kinds of microorganisms.

Removal of n-hexane by *F. solani* with a gas-phase biofilter

Sonia and Sergio (2005a) have attempted the removal of hexane with a gas phase bio-filters which was inoculated by fungus *F. solani*. *Fusarium* was isolated from a mixture of culture grown on hexane vapors. With the packing material, the bio-filter operated with an empty bed residence for a period of 60 s with hexane concentrations and the performances of bio-filter was evaluated for about 100 days. For the growth of fungus and its activity, several nutrients was supplied. This system has the elimination capacity of up to 130 gm^{-3} reactor h^{-1}. Due to biomass development, the mass transfer limitations were confirmed in batch experiments, followed by the inhibitors and pH confirmed the predominant role of the fungus. Finally, the results show that for treating hydro-phobic compounds, fungal bio-filters act effectively.

Removal of hexane by enhanced fungal species in a biofilter

A great work was done by Sonia and Sergio (2005b) in removal of hexane by enhancing the development of fungus in a microbial consortium bio-filter. Her study was limited by gas liquid mass transfer to the biotic aqueous phase where biodegradations occurs. When the use of fungi increased in bio-filters, the removal of hexane improved and it was proved. She allowed the inoculated mixed culture (containing bacteria and fungi) to uptake hexane vapors with 2.6 L per liter-packed bio-filters and that was operated under neutral or acid conditions. In acid bio-filter, increase in the inlet hexane load showed the maximum elimination capacities (150 gm^{-3} h^{-1}). The bacterial inhibitors had no significant effect on elimination capacities in the acid system. The biomass in the acid bio-filter had direct contact with the gas and absorbed the hydrophobic compounds. In this study, two fungal strains were identified namely *Fusarium* and *Cladosporium* species and elimination capacity of hexane was about 40 gm^{-3} h^{-1}. For hexane biofiltration of strain, the biomass content showed the potential of about 30 mg g^{-1} dry L^{-1}.

Biofiltration methods for the removal of phenolic residues

Luis et al. (2006) reported that the ability of isolated microorganism able to degrade pollutants is usually done from natural microbial pollutants by batch cultivation using enriched media containing high pollutant levels. The most recent studies on these technologies, dealing with bacteria isolation, their classification, physiological characterization and molecular analysis of the enzymes responsible for pollutants degradation led to the construction of so-called “super microorganism”.

Performance of biotrickling filters for VOC removal

Chunping et al. (2011) anticipated the control of volatile organic compounds (VOCs) from waste gas streams of low concentration and have attracted more and more public concern. Biofiltration, initially applied to control odor emissions in soil beds, has become an established technology for VOC emission control. Many investigations and applications of biofiltration technology on VOC removal were reported. More recently, several novel biofilters capable of reaching higher removal efficiencies were developed. Experiments were conducted using polyurethane foam plugs and shredded polyurethane foam mixed peat while choosing perlite and vermiculite as support media, concluding that the polyurethane foam did not have properties that were as favorable for biofiltration as other materials tested. Bacteria and fungi are the two dominant microbial groups in bioreactors, but bacteria are most likely to dominate under favorable conditions. Fungi are more tolerant to dry and acidic conditions than bacteria, hence fungi can degrade certain hydrophobic compounds faster than bacteria and have a higher elimination capacity.

Effect of substrate henry's constant on biofilter performance

Xueqing et al. (2004) reported that as the waste gas moves through the biofilter, organic compounds diffuse into the attached biofilm surrounding the support medium and are oxidized into mineral end products (for example, water and carbon dioxide) and incorporated into new biomass. A key process involved in a gas-phase biofilter is the transport of VOCs from the gas phase to the aqueous phase. This process frequently is assumed to be related to vapor/liquid partition coefficients or Henry's constants. One of the most useful methods for determining the structure or diversity of bacterial communities is denaturing gradient gel electrophoresis (DGGE). In a study by Byungjoon et al. (2008), DGGE technology was used to examine microbial communities with the emphasis on determining the type and presence of denitrifying organisms in the biofilters. Some of the most impressive advances in waste treatment biotechnologies have taken place in the field of hazardous waste bioremediation. This has resulted in an enormous increase in our understandding and ability to utilize the cost-effective capabilities of microorganisms successfully and efficiently in absorbing and degrading a wide variety of toxic waste materials. However, effective as of December 29, 2004, the definition relies solely on any compounds of carbon, excluding carbon monoxide, carbon dioxide, carbonic acid, metallic carbides or carbonates and ammonium carbonate, which participates in atmospheric photochemical reactions (The United State Environmental Protection Agency, U.S. EPA, Definition of VOC: Code of Federal Regulations: Title 40, Part 51 Section 51.100).

Comparative studies on toluene removal and pressure drop in biofilters using different packing materials

Hee et al. (2009) study was to select the best available packing material for VOCs control; it compared the removal efficiency of toluene in biofilters packed with different packing materials, including porous ceramic (celite), Jeju scoria (lava), a mixture of granular activated carbon (GAC) and celite (GAC/celite), and cubic polyurethane (PU) foam. We also monitored pressure drop and clogging during toluene degradation. Biofiltration technology has been exceedingly developed for the control of low concentration VOCs emitted from various industries due to its simple operation, relatively lower capital and operating cost than conventional technology such as physical and chemical treatment as well as the harmless final product such as water and carbon dioxide and environment friendly.

Comparative study on removal of toluene by batch reactor using different fungal culture isolated from municipal sewage water

Bhuvaneswari et al. (2012) carried out the comparative study on the removal of toluene by batch reactor using different fungal culture isolated from municipal sewage water investigated with various parameters such as effect of different fungal culture and effect of pH and temperature. The microbial degradation of toluene was carried out in a shake and flask or batch reactor kept in a shaker with 150 rpm for seven days with the synthetic effluent prepared with toluene concentration of about 1% and was used for the entire study. The treated effluent was analyzed by GC – FID for toluene degradation and was found to be 75 to 92% by *Penicillium sp*; greater than the *Aspergillus niger* and *Trichoderma viride* with growth conditions pH 7 and 30°C.

CONTINUOUS MICROBIAL DEGRADATION OF TOLUENE

Large scale production of toluene by various reactor setups has been found in extensive usage for the last few decades. Depending on the design of the bioreactors, elimination capacity of the specific volatile molecule such as toluene varies. Prevalently used bioreactors are biofilters, biotrickling filter (Duangfang et al., 2011), Bioscrubber, hollow fiber bioreactor (Madan et al., 1996), hybrid bioreactor (Sonia and Sergio, 2005b), fibrous bed bioreactor (Arief and Peter, 2004), capillary bioreactor (Yieng et al., 2005), biological activated carbon biotrickling filter (Amit et al., 2008), tubular bioreactor (Yieng et al., 2005), flat sheet bioreactor (Yieng et al., 2005), two face partitioning bioreactor (Colin et al., 2003), composite bead bioreactor (Wu-Chung and Hung-Yuan, 2010), fixed film bioreactor (Dolloff and Richard, 1996), GAC-fluid bed bioreactor (Dolloff and Richard, 1996), bioactive foam emulsion reactor (Farshid et al., 2010), and adsorption bioreactors. Hollow fiber bioreactors have been used extensively in immobilizing enzymes and microbial cells because they are able to exhibit large surface area per unit volume.

CONCLUSION

Amputation of toluene is one of the vital needs in the present scenario because it leads to health problems and several environmental hazards. It contributes to various techniques utilized at the industrial level mainly various bioreactors that are useful in the toluene removal. A study on biodegradation of toluene to carbon dioxide, water and biomass was conducted using various micro-organisms such as fungi, bacteria, mixed culture and actenomycetes, etc that points to the elaborate significance of fungi among the others. A comparison of the removal capacity under continuous and batch mode of operations was studied proficiently. This review can be considered as a pioneer exertion in amputation of toluene and hence can be referred for the future exploration in the field of biodegradation.

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