

# OVERVIEW OF SOIL XENOBIOTICS AND THEIR BIOLOGICAL REMEDIATION STRATEGIES

## ABSTRACT

Xenobiotic pollution of the environment is a global phenomenon brought on by human activity as a result of increased urbanisation and population expansion. Pesticides, petrol, solvents, alkanes, polycyclic hydrocarbons (PAHs), antibiotics, artificial azo dyes, pollutants (polychlorinated biphenyls and dioxins), and polyaromatic, chlorinated, and nitroaromatic substances are examples of xenobiotics. Xenobiotics are primarily defined as substances that are foreign to a living thing and have an inclination to collect in the **environment, where they can be harmful for existing ecosystem**. The natural environment is impacted in a number of ways, both short- and long-term, when these dangerous contaminants are released. The scientific study of xenobiotic compound biodegradation—which uses microorganisms' catabolic powers to break down poisonous and hazardous xenobiotics—has received a lot of attention in the last few decades. Numerous bacteria possessing remarkable capacity for biodegrading xenobiotic substances have been identified, and their novel degradation routes have been clarified. The field of molecular approach to biodegradation research is fairly young. Therefore, in order to create efficient and environmentally beneficial "green" technology, it is imperative to investigate the microbial biodiversity.

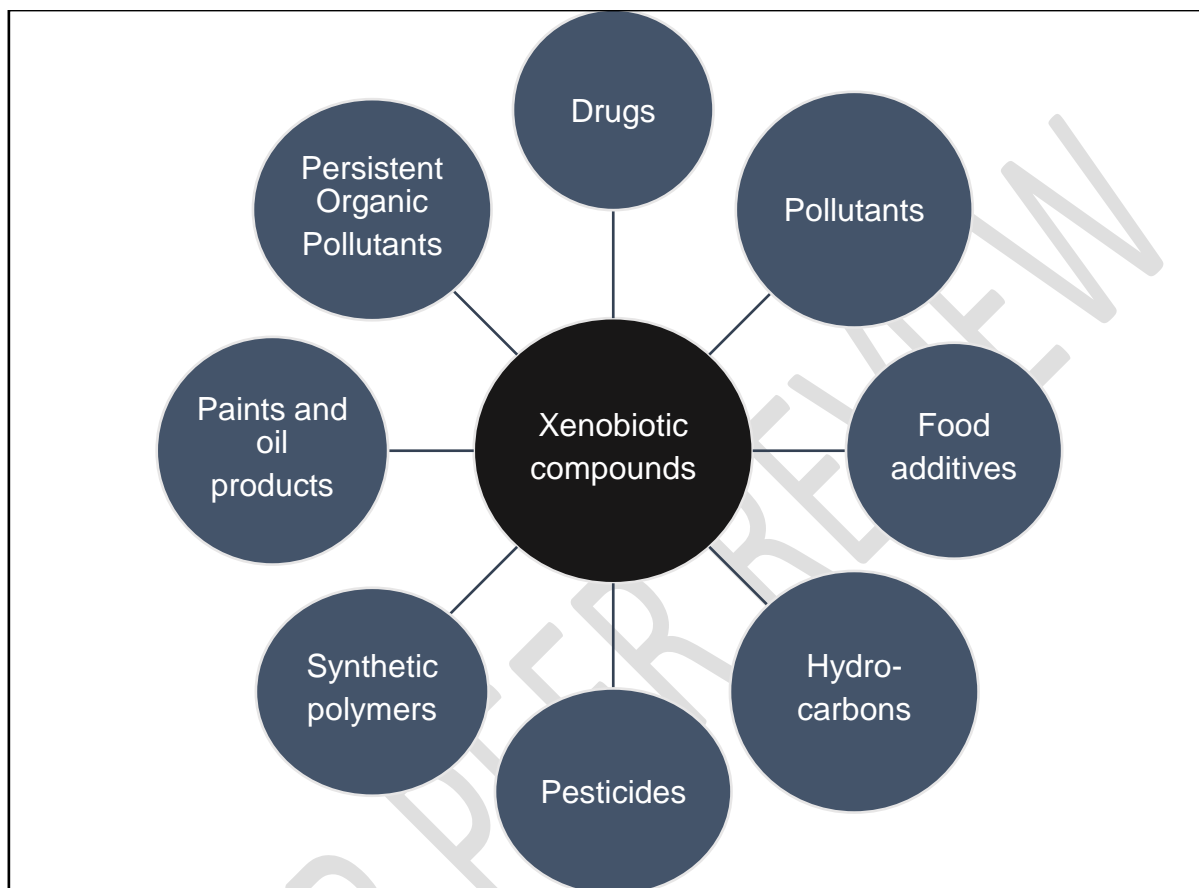
**Keywords:** Xenobiotics, Bioremediation, Phytoremediation and sustainability.

## INTRODUCTION:

The Greek terms xeno (foreign) and biotics (of or linked to life) are the source of the phrase "xenobiotic." **Xenobiotics are substances that are not essential to an organism, or that do not normally form part of its usual nutrition.** Common examples of xenobiotics are mixtures of pharmaceuticals, artificial chemicals, food additives, and contaminants found in the environment. "Xenobiotics are foreign chemical substances that are found in organisms but are not typically or naturally anticipated to be there". When natural substances are transported from one organism to another, they can act as xenobiotics. For example, fish living downstream of a sewage treatment facility may absorb natural human hormones, or certain species may create chemicals as a chemical defense against predators (Afify et al. 1997). However, because dioxins and polychlorinated biphenyls are man-made substances that were not present in nature before their synthesis by humans, the term "xenobiotics" and their effects on the biota are rarely used concerning chemical pollutants in the biological system.

**As xenobiotic compounds break down molecules of medications or drugs, microbial enzymes** that metabolize xenobiotics are crucial to the pharmaceutical business. Similarly, the length of time that medications remain in the body is influenced by xenobiotic transporters. Although man has long used xenobiotics, knowledge of the metabolism of foreign compounds did not emerge until the middle of the 1800s when the principles, practices, and knowledge of organic chemistry were first applied to the study of foreign compounds. Biotransformation was commonly associated with "detoxification," or the removal of a compound's biological activity, for over a century after that. Any method that restores the environment to its pre-pollution state by using microorganisms or their enzymes is known as bioremediation. Another way to put it is as "a treatability technology that uses biological activity to lessen or reduce the toxicity or concentration of a pollutant." **Detoxification**

that is removal of harmful synthetic compounds and mineralization i.e. conversion of molecules from organic to inorganic form or complex to simpler form, are the steps in bioremediation process that transform hazardous waste into inorganic substances including carbon dioxide, water, and methane. When xenobiotics are persistent in the environment, their biodegradation frequently happens in a variety of ways using different enzyme systems or different microbial communities.



**Fig 1. Types of Xenobiotic compounds in the environment**

One of the major and crucial methods for getting rid of or detoxifying ecologically hazardous substances is through the microbial biodegradation of xenobiotics. It has been established that using potential microorganisms to break down xenobiotic chemicals is a useful way to remove hazardous and toxic waste. Bioremediation is used in contaminated wastewater treatment plants, ground or surface waters, soils, sediments, and air when there has been an unintentional or deliberate release of chemicals or pollutants. It is, of course, in everyone's best interests to remove pollution and handle garbage sensibly (Muhammad et al., 2017). This paper emphasizes the potential significance of microbes for environmental cleaning, given the multitude of issues in the realm of xenobiotics.

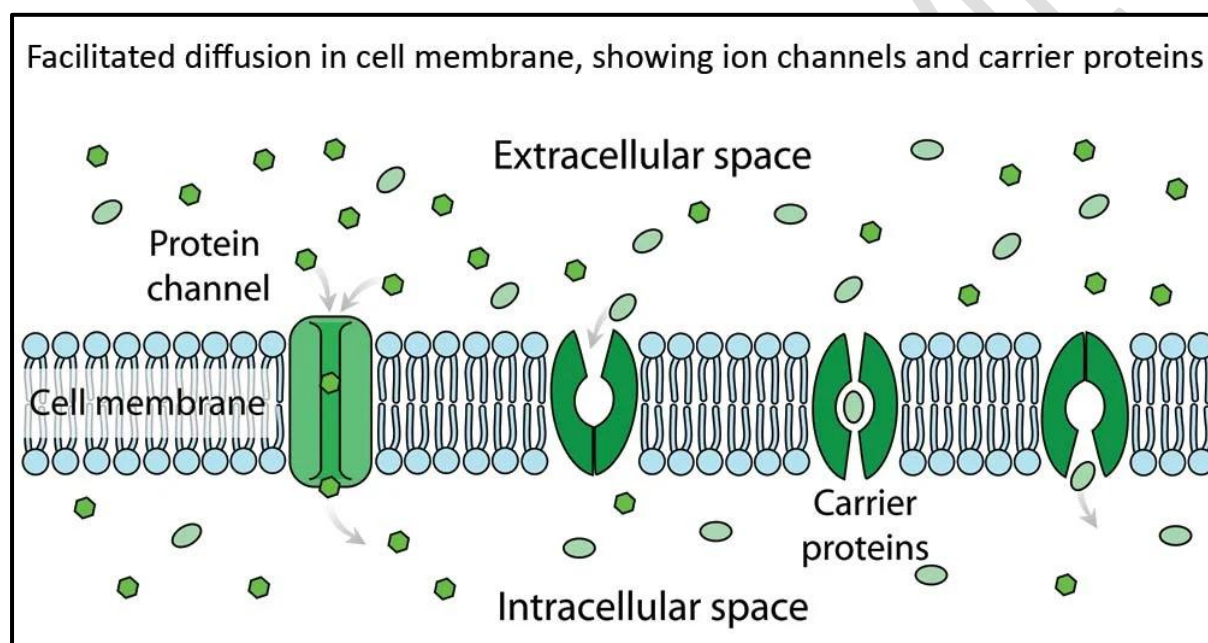
Every day, a huge number of new xenobiotic and natural substances are introduced into the environment, such as dyes, preservatives, cosmetic products etc.. It may be possible to remove harmful substances from the environment as well as convert and produce beneficial end products by investigating and using the unrealized potential of bacteria and their products. For example, Dalapon herbicide containing chlorinated fatty acid is converted into pyruvate by *Arthrobacter* (Smitha and

Singh, 2017). This article highlights basic approaches and practices that will assist meet the expectations of various user/reader levels.

### Xenobiotics-Protein interaction:

Xenobiotic interaction with protein (how xenobiotic and living organisms react to each other) defines the positive and negative impacts of xenobiotic compounds. This interaction facilitates entry of harmful complex compounds in plant or animal cell for which following steps are followed:

- a. Penetration to targeted organism
- b. Transported to the action site
- c. Disrupt or alter vital function.



**Fig 2. Transportation of xenobiotics by facilitated diffusion protein (Afify, 2010)**

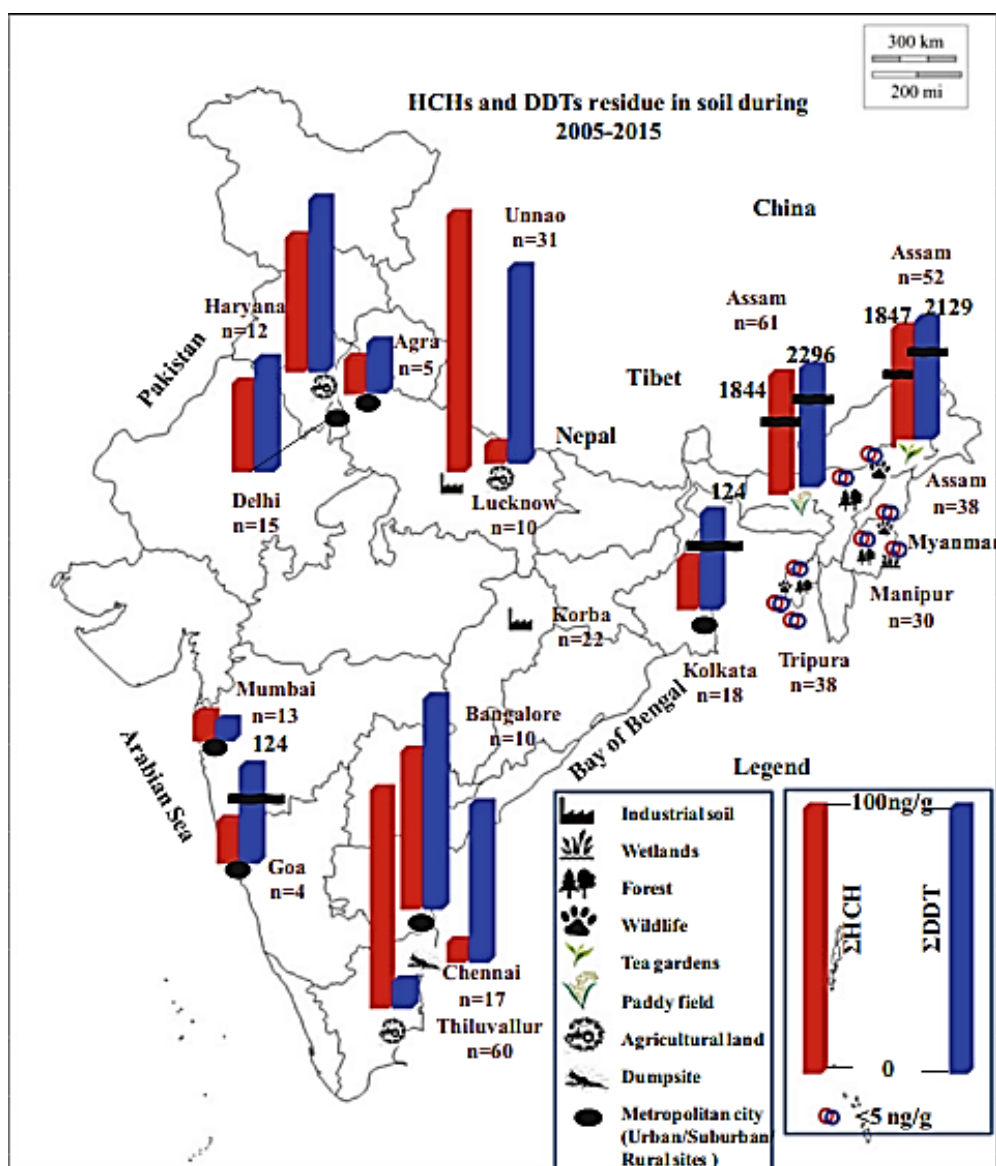
Membrane transport proteins, sometimes referred to as transporters, are proteins that let tiny molecules, ions, and macromolecules like other proteins pass across biological membranes. These proteins are crucial components of membrane proteins and have the power to facilitate the active transport and diffusion of proteins, as well as the movement and transportation of compounds; this process is known as carrier-mediated transport. There are two types of carrier-mediated transport: (a) assisted diffusion and (b) active transport (Crompton 1999). In the absence of energy input, facilitated diffusion protein increases the movement of molecules across membranes; eventually, the associated chemical can only travel down a concentration gradient. The high-specificity pore/channel development that continues to the membrane is most likely the cause of this. Certain amino acid residues plug these "polar holes" in the membrane, lowering the energy barrier to polar molecules' motion (Fig. 2).

### Soil xenobiotics in Indian agriculture:

“India is a nation of farmers with a variety of climate zones.. For more than 50 years, India has been a major user of organochlorine pesticides (OCPs), such as dichlorodiphenyltrichloroethane (DDT) and hexachlorocyclohexane (HCH), for both agricultural and public health objectives. Seventy percent of the yearly pesticide usage (85,000 t) is made up of DDT, HCH, and malathion (an organophosphoruschemical)” (Gupta 2004). “Even though OCPs were outlawed for use in agriculture in the late 1990s, a significant portion of these pesticides are still in use to eradicate insects that carry illnesses like malaria and kala-azar (black fever), among others. These pesticides are widely used, and because of their semi-volatile nature and lengthy environmental lives in soil and water, they continue to pollute many environmental compartments” (Kurt-Karakus *et al.* 2005).

“DDT and HCH build up in the organic matter of soil for a longer amount of time because of their hydrophobic, lipophilic nature, and attraction for particles” (Ockendon *et al.* 2003). “Therefore, by re-emitting these chemicals into the atmosphere, soil serves as both a secondary source and a sink for these pollutants” (Harner *et al.* 2001). “HCHs and other OCPs with comparable physicochemical properties dissipate from soil under tropical and subtropical conditions, resulting in their widespread distribution”. (Babu *et al.* 2003). An overview of HCH and DDT residues in soil found in the northern, eastern, northeastern, western, central, and southern regions of India is given in Fig 3.

Numerous studies have indicated that the primary local sources of POPs (**Persistent Organic Pollutants**) are agricultural activities. POPs are found all over the world because of their moderate vapor pressure properties. (Gong *et al.* 2014) and are dependent upon air transportation processes as well as historical local and regional factors (Wang *et al.* 2007). According to UNEP (2012), “polychlorinated biphenyls (PCBs) and organochlorine pesticides (OCPs) are dangerous and very risky for both human health and ecosystems”. In contrast to developed countries, Nasir *et al.* (2014) noted from various literature that “high concentrations of persistent pollutants (POPs) typically observed in developing countries were suggestive of extensive past usage for agricultural and vector-borne diseases as well as the inability to enforce regulation to restrict indiscriminate use and disposal of industrial/agrochemicals”. According to Khan *et al.* (2002), “83% of pesticides were applied to rice, sugarcane, and cotton crops overall”. When it comes to the overuse or deposition of DDT in soil and leaves from agricultural operations, Nepal is likewise not far behind (e.g., Gong *et al.* 2014).



**Fig 3: Maximum HCH and DDT residues in surface soil from different states of India. Concentrations presented in this figure have been obtained from various studies in India (Chakraborty et al. 2015)**

### Antibiotics and Antibiotics Resistance Genes (ARGs) in soil:

Following Alexander Fleming's 1928 discovery of penicillin, the age of manmade antibiotics started (Diggins 1999). Autochthonous soil bacteria that biosynthesize secondary compounds with antimicrobial properties are found in natural soil (Martinez-Fleites et al. 2006;). One of the natural chemical regulating mechanisms found in most organisms, particularly bacteria, is antibiosis. Both naturally occurring and manufactured antibiotics are now widely used in veterinary and human medicine to treat and cure a wide range of bacterial diseases; nevertheless, improper use or abuse of antibiotics leads to the appearance and dissemination of antibiotic resistance genes (ARGs) in the environmental compartments (Tang et al. 2015). Antibiotics are often delivered into the environment via the following routes: (1) unintentional release during manufacture and usage; (2) application against plant diseases; (3) sewage sludge containing antibiotic leftovers from human medication; and

(4) medicated animal manure treated with injectable solutions, ergotropics, or therapeutic feed/water additions. Antibiotics have therefore been found in a variety of environmental settings, including soil, aquatic habitats, and agricultural groundwater (Martinez 2009).

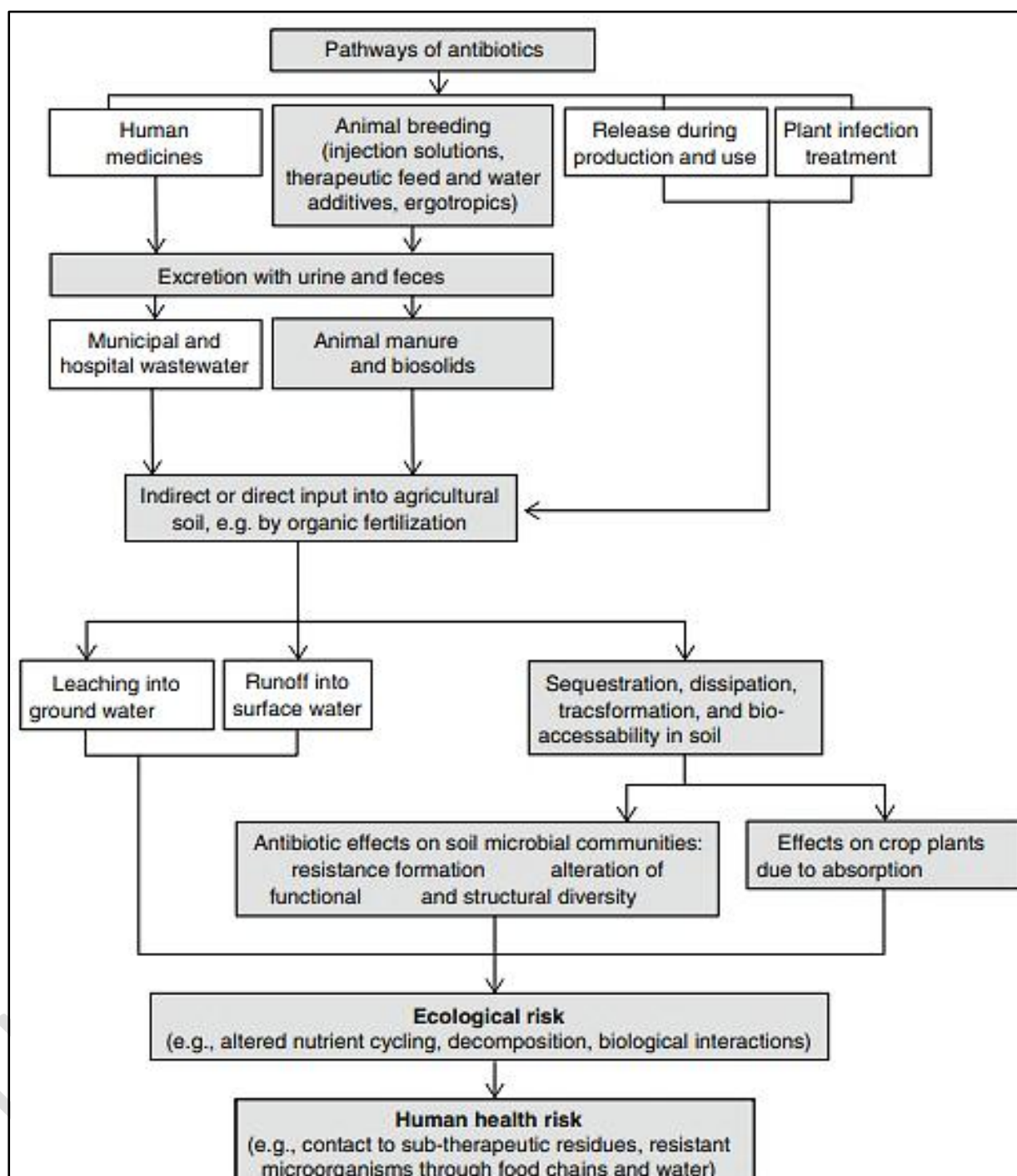


Fig. 4: Pathways of antibiotics (Adapted from Du and Liu 2012)

**Table 1: Biological Remediation Strategies**

Technique	Advantages	Disadvantages	References
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<p><b>Phytoremediation</b> Uses plants in combination with microorganisms to remediate the contaminated area.</p>	<ul style="list-style-type: none"> <li>• Minimal impact on the environment.</li> <li>• Technology powered by solar energy; applicable to a wide variety of pollutants.</li> <li>• Economical for sizable, polluted locations</li> </ul>	<p>It takes two growth seasons.</p> <ul style="list-style-type: none"> <li>• Restricted to groundwater below three meters and soils less than one meter below the surface. Animals that consume the plants utilized in these experiments might introduce contaminants into the food chain.</li> </ul>	-
<p>A) <b>Phytovolatilization:</b> Contaminant is taken in by the plant tissue and then vitalized in the environment.</p>			Danika and Norman (2005)
<p>B) <b>Phytoextraction:</b> Plants remove dangerous elements or compounds from soil or water, most usually heavy metals, metals that have a high density and may be toxic to organisms even at relatively low concentrations.</p>			Marchiol et al., (2007)
<p>C) <b>Phytostimulation:</b> Involves the stimulation of microbial degradation through the activities of plants in the root zone</p>			Rupassara et al., (2002)
<p>D) <b>Phytotransformation:</b> It is the breakdown of organic contaminants sequestered by plants via: (1) metabolic processes within the plant; or (2) the effect of compounds, such as enzymes, produced by the plant.</p>			Kvesitadze et al., (2006)
<p>E) <b>Rhizofiltration:</b> Involves filtering water through a mass of roots to remove toxic substances or excess nutrients.</p>			Miller (1996)
<p>F) <b>Phytoscreening:</b> plants are used as biosensors of subsurface contamination and is a simple, fast, noninvasive, and inexpensive method.</p>			Burken et al., (2011)
<p>G) <b>Phytostabilisation:</b> Root released compounds enhance microbial activity in the rhizosphere</p>			Mendez and Maier (2008)
<p><b>Bioreactors/ Bioslurry:</b> Use of bio based reactors and selected bacteria to biodegrade the contaminants.</p>	<ul style="list-style-type: none"> <li>• Fast degradation</li> <li>• Effective use of inoculants and surfactant</li> </ul>	<p>Soil transport required.</p> <ul style="list-style-type: none"> <li>• Expensive</li> </ul>	Zhang et al., (2001)
<p><b>Biosparging:</b> Air and nutrients are injected into the saturated zone to increase the biological activity of the</p>	<ul style="list-style-type: none"> <li>• Readily available equipment.</li> <li>• Cost competitive.</li> <li>• In situ technology</li> </ul>	<ul style="list-style-type: none"> <li>• Biochemical and physiological interactions are very complex and needs to</li> </ul>	Raag (2000)

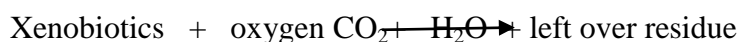
indigenous microorganisms		be understood • Migration of constituents can lead to toxicity elsewhere	
<b>Biopiling:</b> Involves the piling of petroleum contaminated soils into piles or heaps and then simulating aerobic microbial activity by aeration and the addition of minerals, nutrients, and moisture	• Insitu technology therefore no transportation cost.	Need to control abiotic loss • Mass transfer problem • Bioavailability limitation	Filler et al., (2001)
<b>Bioventing:</b> Process of injecting air into the contaminated media at a rate designed to maximize in situ biodegradation and minimize or eliminate the off-gassing of volatilized contaminants to the atmosphere	• Very economic and easy to install • can be combined with other technologies	High concentrations can be toxic for microorganisms. • Low soil permeability doesn't allow proper implication. • Good for unsaturated zones of soils.	Diele et al., (2002)
<b>Bioslurping:</b> Combines elements of bioventing and vacuum enhanced pumping to remediate the contaminated site.	• Applied at shallow as well as deep sites. • Recovers free product, thus speeding remediation	• Low soil permeability hampers remediation. • Soil moisture and oxygen content limits the microbial activities. • Low temperatures slow remediation.	Yen et al., (2003)
<b>Land Farming:</b> Bioremediation treatment process that is performed in the upper soil zone or in biotreatment cells.	• Relatively simple design and implementation • Short treatment times (six months to two years under optimal conditions).	• Required area is high. • Dust and vapor generation may cause some air pollution.	Hejazi (2002)
<b>Natural attenuation:</b> Uses natural processes to limit the flow of contaminants from chemical spills and also reduces their concentration at contaminated sites.	• Remediation waste is least which has less impact on the environment. • Can be easily combined with other technologies.	• Ethical issues remain which needs to be correctly perceived by the people. • Costly and complex site characterization.	Khan and Husain (2002)
<b>Composting:</b> Uses cow manure and mixed vegetable waste to remove the toxicants	• Cheap with rapid reaction rate.	• Treatment time more than other techniques • Requires nitrogen	Atagana (2004)

upto 90% from the contaminated soil		supplementation.	
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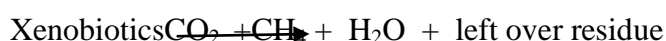
### Role of microbes in xenobiotic degradation:

Normally, the microbes use two pathways for biodegradation of xenobiotics, aerobic and anaerobic conditions.

In aerobic bioremediation, the basic equation will be



In the case of anaerobic bioremediation, it is



“There are vast numbers of potential bacteria, which carry out the bioremediation of xenobiotics such as, *Acidovorax* spp., *Bordetella* spp., *Pseudomonas* spp., *Sphingomonas* spp., *Vario* pp., *Veillonellaalkalescens*, *Desulfovibrio* spp., *Desulfuromonasmichiganensis*, *Desulfitobacteriumdehalogenans*, *Pseudomonasoleovorans* and *Geobacte.metallireducens*. Anaerobic sulfate-reducing bacteria and methanogenic bacterial conditions can be useful to isolate pure culture of anaerobic bacteria to carry out xenobiotic degradation research work” (Jiang and Fan 2008).

**Table 2: Microbes utilized to degrade specific xenobiotic compounds**

Microbial strain	Target xenobiotic compound	Place	Reference
<i>Pseudomonas</i> sp. Ph6	Phenanthrene	China	Sun et al. (2014)
<i>Kocuria</i> sp. CL2	Pentachlorophenol	India	Karn et al. (2011)
<i>Microbacterium</i> sp. strain SL10	Anthracene	Nigeria	Salam et al. (2014)
<i>Streptomyces</i> spp.	Naphthalene	Algeria	Ferradji et al. (2014)
<i>Klebsiella oxytoca</i> PYR-1	Pyrene	China	Zhang and Zhu (2012)
<i>Herbaspirillumchlorophenolicum</i>	Fluoranthene	China	Xu et al. (2011)
<i>Pseudomonas</i> sp., <i>Enterobacter</i> sp., <i>Acinetobacter</i> sp., and <i>Corynebacterium</i> sp.	2-Chlorobenzoic acid	Iran	Kafilzadeh et al. (2012)
<i>Sphingobiumczechense</i> LL01	HCH/lindane (1,2,3,4,5,6-hexachlorocyclohexane)	India	Niharika et al. (2013)
<i>Pseudoxanthobacterliyangensis</i> sp. nov.	DDT (dichlorodiphenyltrichloroethane)	China	Liu et al. (2014)
<i>Novosphingobiumarabidopsis</i> sp. nov.		Taiwan	Lin et al. (2014)
<i>Stenotrophomonas maltophilia</i> and <i>Rhodococcuseryt</i>	Endosulfan compounds	India	Kumar et al. (2007)

<i>hropolis</i> <i>Klebsiella pneumonia</i>		South Korea	Kwon et al. (2002)
<i>Arthrobacter</i> sp.C21 <i>Achromobacterdenitrificans</i> strain SP1	Phthalate	China India	Wen et al. (2014) Pradeep et al. (2015)
<i>Micrococcus</i> species <i>Sphingopyxis</i> sp. PVA3	Vinyl chloride	India Japan	Patil and Bagde 2012) Yamatsu et al. (2006)
<i>Raoultellaplanticola</i>	Atrazine	Israel	Swissa et al. (2014)
<i>Pseudomonas</i> sp. and <i>Stenotrophomonas</i> sp. <i>Arthrobacter</i> sp. BS2 and <i>Achromobacter</i> sp. SP1	Diuron(3-(3,4- dichlorophenyl)-1,1- dimethylurea)	France	Batissou et al. (2007) Devers Lamrani et al. (2014)
<i>Xanthomonassp</i>	Propanil	Mexico	Herrera-Gonzalez et al. (2013)
<i>Acinetobacter baylyi</i> strain GFJ2	Chloroaniline	Thailand	Hongsawat and Vangnai (2011)

### Genetic approaches of soil xenobiotic remediation:

Recent developments in sequencing technology, "omics" platforms, and molecular biology provide a molecular toolbox that facilitates the discovery, isolation, and characterisation of structural genes implicated in xenobiotic remediation as well as their transfer between kingdoms.

**Table 3: Plants and their effects in xenobiotic remediation**

Class of pollutant	Plant	Endophytic bacterium	Effect
Heavy metal	Lupinus luteus and Lolium perenne	<i>Burkholderiacepacia</i> and <i>Herbaspirillumseropedicae</i>	Phytoremediation of heavy metals
Solvents	Yellow lupin	<i>Burkholderiacepacia</i> VM1330	Toluene degradation
	Poplar	<i>B. cepacia</i> VM1468	Toluene degradation
	Poplar	<i>Pseudomonas putida</i> W619- TCE	TCE degradation
Herbicide	Pea ( <i>Pisum sativum</i> )	<i>Pseudomonas putida</i> VM1450	2,4 Dichlorophenoxyacetic acid degradation
Polyaromatic hydrocarbons	<i>Pisum sativum</i> var. early onward	<i>Pseudomonas putida</i> strain VM1441 (pNAH7)	Naphthalene phytoprotection and phytoremediation

(Source: Jagtap and Bapat, 2017)

These rehabilitation techniques are still challenging, and in many cases, impossible, to transfer from the lab to the real world. Given this, a thorough research programme is required to translate laboratory findings into environmental practice in a way that optimises contaminated site cleanup and reduces environmental pollution. Our expertise of designing artificial plant systems for the removal of xenobiotic pollutants will grow with a deeper comprehension of plant system biology and the availability of standardised genetics. In order to accomplish this goal, it will be necessary to integrate the expertise and coordinated efforts of soil scientists, plant physiologists, chemists, environmental engineers, government regulators, molecular biologists, microbiologists, systems biologists, and bioinformatics specialists.

### **Conclusion:**

There is a strong chance that our food and water are contaminated with xenobiotics due to urbanisation and population growth. Hazardous xenobiotics have been found in anything from everyday hygiene goods to agricultural applications. To eliminate these newly developing pollutants, choosing an effective treatment plan requires careful consideration of both technical and financial factors. The richness of organisms found in environmental locations, or microbial diversity, offers a vast pool of resources that can be used for restoration. Developing efficient and environmentally friendly "green" technology requires an exploration of the breadth of microbial biodiversity. One such method that uses microorganisms' catabolic properties to break down dangerous and poisonous xenobiotics is bioremediation. In several instances, we have been able to recover locations that were previously irrevocably contaminated, demonstrating the value of our cleanup procedure. However, a thorough knowledge of a microbe's metabolic processes, molecular biology, and capacity for degradation under different conditions is essential to maximising the potential advantages of the microbial community in tackling pollution concerns.

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