Chapter 16
Bioremediation of Contaminated Paddy Soil

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16.1 Background

A paddy field is a flooded parcel of agricultural land that is used for growing semiaquatic rice. Rice cultivation in the flooded land should not be confused with paddy cultivation. Rice is grown in the flooded land (deep water rice) with water at least 50 cm (20 in.) deep for a minimum period of one month (Molina et al. 2011; Fig. 16.1). Paddy field farming is practiced in Asia; namely, in Cambodia, Bangladesh, Indonesia, Taiwan, China, India, North Korea, Iran, Japan, Myanmar, South Korea, Malaysia, Pakistan, Nepal, Philippines, Sri Lanka, Thailand, Laos, and Vietnam. In Europe, paddy field farming is practiced in northern Italy, in the Camargue in France (Riz et al. 2013), and in Spain, particularly in the Albufera de València wetlands in Valencia, the Ebre Delta in Catalonia, and the Guadalquivir wetlands in Andalusia. This type of farming is also carried out along the eastern coast of Brazil, in the Artibonite Valley in Haiti, and in the Sacramento Valley in California, among other places. Paddy fields are also considered as an origin of atmospheric methane, contributing approximately 50–100 million tons of the gas annually (D Reay GHG 2018).

Studies have shown that draining the paddies, to allow the soil to aerate, interrupts methane production, significantly reducing its emission while also boosting crop yield. Studies have also shown variability in the assessment of methane emission using local, regional, and global factors, and have called for better intervention based on micro-level data (Mishra et al. 2012). Rice is the global staple food for up to 60%

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Fig. 16.1 A rice paddy on the outskirts of Lahore, Pakistan

of the world's population (Kiritani 1979) and China is the top-ranking world rice producer (Kögel-Knabner et al. 2010). Paddy fields are anaerobic at the time of plant development; however, they are aerobic during the period between the harvest of the ripe crop and replanting, and are considered as most reasonable area for the planting of rice sprouts (Liesack et al. 2000).

In Taizhou, crop rotation, which is applied by planting vegetables during the first half of the year and single-crop of rice in the second half of the year, also gives rise to a succession of dry and flooded conditions in paddy fields.

Repeated replacement of reductive and oxidative conditions is a result of water table fluctuations, oxygen diffusion through the water column, and oxygen transport through above-ground plant tissues into the rhizosphere (Armstrong 1971).

Such alterations of redox conditions in paddy fields can lead to the staggered appearance of aerobes, such as facultative and obligate anaerobes, which are expected to exhibit a commutative predominance of quantity and activity in different time-space conditions (Liesack et al. 2000; Brune et al. 2000). Additionally, Walker et al. (2003) stated that the rise in the amount of altered pH and aeration conditions and readily accessible carbons derived from root exudation in the rhizosphere of rice further result in the promotion of microbial activity.

In 1998, Holliger et al. revealed that the more chlorinated biphenyls via halorespiration might be transformed potentially, involving naturally arising micro-organisms (using electron acceptor PCBs, gaining energy). In another report by Abramowicz (1990) and Field and Sierra-Alvarez (2008), they acknowledged that high chlorinated congeners must be completely biodegradable in an arrangement of dechlorination of anaerobic reductiveness followed by less chlorinated items for aerobic mineralization. Many researchers have reported that the combination of these two procedures can effectively demolish PCBs (Evans et al. 1996; Master et al. 2001). However, thus far, studies involving sequential PCB transformation mostly depends on the preliminary enrichment of functional guilds or bioaugmentation, which is not suitable for large-scale application in situ (Payne et al. 2013).

16.1.1 Anoxic Environment

The anoxic environment is obligatory to produce redox condition and is induced by flooding redox state by dechlorination, and the land deprived of such conditions cannot be used for the rice cultivation.

16.1.2 Oxic Environment

Oxic conditions formed during the fallow period provide a favorable environment for the subsequent degradation of dechlorination products. Moreover, the paddy soil represents an improved micro-environment where the transformation of PCB is escalated.

16.2 Types of Bioremediation

There are two different types of bioremediation: in situ and ex situ.

16.2.1 In Situ Bioremediation

In situ bioremediation involves treatment of the contamination on site. Mineral nutrients are added in this type of bioremediation to treat soil contamination. These nutrients in the soil cause microorganisms to enhance their degradation capability.

Sometimes new microorganisms are added to the contaminated area. Microorganisms can sometimes be genetically engineered to degrade specific contaminants (Fig. 16.2).

16.2.1.1 Example of a Genetically Engineered Microorganism

An example of a microorganism that has been genetically engineered is *Pseudomonas fluorescens* HK44. These genetically engineered microorganisms can be designed for conditions at the site. This method relies on the relationship between

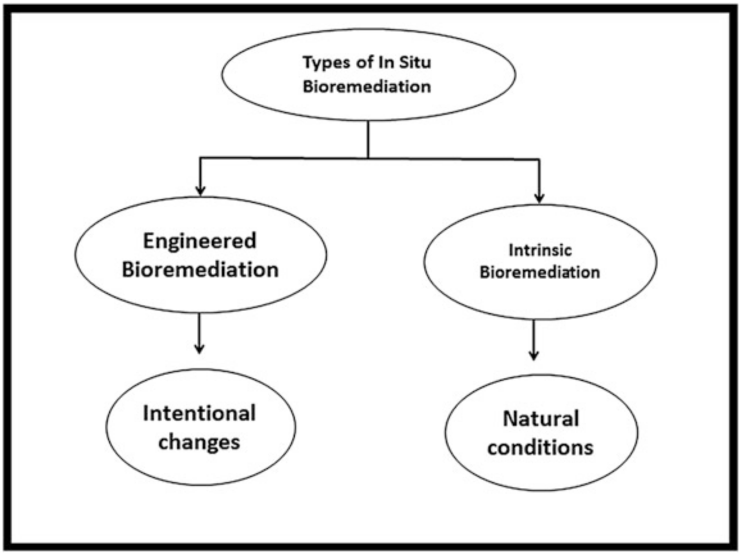

Fig. 16.2 Types of in situ bioremediation

the kind of contamination and the type of microorganisms effectively present at the contamination site. For example, if the microorganisms already present are appropriate to break down the type of contamination, cleanup crews may only need to "feed" these microorganisms by the addition of fertilizers, nutrients, oxygen, phosphorus, etc.

16.2.1.2 Methods of Supplying Oxygen to the Microorganisms

There are two frequently used methods of supplying oxygen to the microorganisms, bioventing and hydrogen peroxide injection.

Bioventing This method consists of blowing air from the atmosphere into the contaminated soil.

16.2.1.2.1 Procedure

1. Injection wells must be drilled into the contaminated soil; the number of wells, how near they are to each other, and how deeply they are drilled all depend upon the factors influencing the rate of degradation (e.g., type of contamination, kind of soil, supplement levels, and the contaminant groups).
2. With an air blower, the air supply that is given to the microorganisms can be controlled after all the injection wells have been drilled.

3. The injection wells may be used to add nitrogen and phosphorus, thus maximizing the rate of degradation.

16.2.1.2.2 Hydrogen Peroxide (H_2O_2) Injection

In cases where the contamination has already reached the groundwater, bioventing will not be very successful. Instead, hydrogen peroxide is used. It functions in much the same way as bioventing, with the hydrogen peroxide, instead of air blowers, delivering oxygen to the microorganisms. If the soil is shallow, the hydrogen peroxide can be administered by spray systems. Injection wells are also used if the groundwater level extends very deep below the surface.

In situ bioremediation involves a direct approach to the microbial degradation of xenobiotics (pollutants) at the sites of pollution (soil, groundwater). The addition of adequate quantities of nutrients at the sites promotes microbial growth. When these microorganisms are exposed to xenobiotics, they develop metabolic ability to degrade them.

The growth of the microorganisms and their ability to carry out biodegradation are dependent on the supply of essential nutrients (such as nitrogen and phosphorus). In situ bioremediation has been successfully applied for the clean-up of oil spillages in the ocean and on beaches. There are two types of in situ bioremediation—intrinsic and engineered.

16.2.1.2.3 Intrinsic Bioremediation

The inherent metabolic ability of microorganisms to degrade certain pollutants is regarded as intrinsic bioremediation. In fact, the microorganisms can be tested in the laboratory for their natural biodegradation capacity and then appropriately utilized (Fig. 16.3).

16.2.1.2.4 Engineered In Situ Bioremediation

The inherent capacity of the microorganisms for bioremediation is generally slow and limited. However, by using suitable physicochemical means (good nutrient and O_2 supply, addition of electron acceptors, optimal temperature), the bioremediation process can be engineered for the more efficient degradation of pollutants.

16.2.1.2.5 Advantages of In Situ Bioremediation

1. Cost-effective, with minimal exposure to the public or people working at the site.
2. Sites of bioremediation remain minimally disrupted.

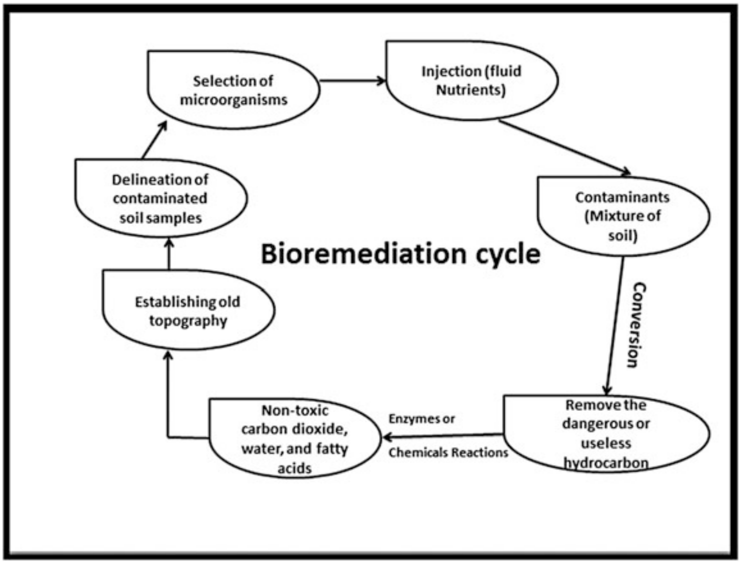

Fig. 16.3 The intrinsic bioremediation cycle

16.2.1.2.6 Disadvantages of In Situ Bioremediation

1. It is a very time-consuming process.
2. Sites are directly exposed to environmental factors (temperature, O_2 supply, etc.).
3. Microbial degrading capacity varies seasonally. http://www.biologydiscussion.com/biotechnology/biodegradation/biodegradation-and-bioremediation-with-diagram/11043.

16.2.2 Ex Situ Bioremediation

Ex situ bioremediation consists of the physical extraction of contaminated media to another location for treatment. If the contaminants are only in the soil, the contaminated soil is excavated and transported for treatment. If the contamination has reached the groundwater, the groundwater must be pumped out and any contaminated soil must also be removed (Fig. 16.4).

16.2.2.1 Stopping the Spread of the Contamination

One major effect of this removal of contaminants is that it stops the spread of the contamination. Provided that the cleanup crews do a good job in the excavation

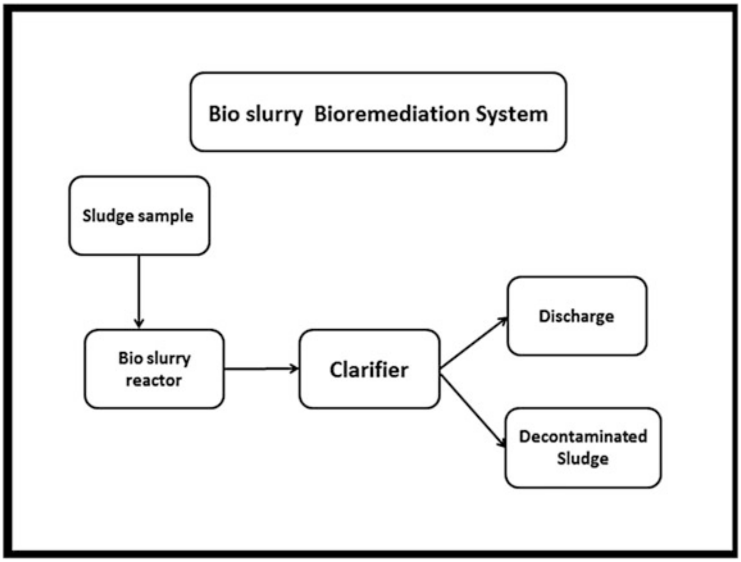

Fig. 16.4 Schematic diagram of bio-slurry bioremediation system

process, there should ideally be no remaining contaminants; however, if minimal amounts of contaminant remain in the soil, they can likely be broken down by the naturally occurring microorganisms already present.

16.2.2.2 Conventional Techniques

Conventional techniques are aimed at remediation that involves either digging up contaminated soil and transporting it to a landfill or capping and containing the contaminated areas of a site. As with every method, this technique also has some drawbacks.

The primary conventional technique involves just moving the contamination somewhere else, and there may be critical dangers in the exhuming, transport, and handling of harmful material. Furthermore, this method is extremely troublesome and it becomes more and more costly to discover new landfill destinations for the final transfer of the material. The cap-and-contain strategy is just an interim solution, since the contamination stays nearby or on site, requiring checking and maintenance of the isolated material long into the future, with all the related expenses and potential obligations.

An approach that is superior to these customary strategies is to completely destroy the toxins if possible, or, if this cannot be done, to transform them into harmless substances. A few technologies that have been utilized are high-temperature incineration and different kinds of chemical decomposition (base-catalyzed dechlorination, ultraviolet oxidation). These methods can reduce the spectrum of contaminants by complete or partial elimination, but have a few disadvantages, including operational complications, cost-effectiveness, and procedural deficiencies such as burning of contaminants after a certain stage, which indirectly can affect the laborers at the site and inhabitants close by.

16.2.2.3 Types of Ex Situ Bioremediation

There are two main types of ex situ bioremediation, referred to as the solid phase and the slurry phase.

Solid Phase In this phase, the technique involves placing the excavated materials into an above-ground enclosure.

1. Inside this enclosure, the contaminated soil is spread on a treatment bed. This treatment bed generally has some sort of built-in aeration system.
2. By utilizing this system, cleanup groups can control the moisture, nutrients, heat, pH, and oxygen. This enables them to augment the efficiency of the bioremediation. The soil can be cultivated like farmland, providing oxygen and empowering the extra-aerobic biodegradation of the contaminants. Solid phase treatment is particularly effective if the contaminants are fuel hydrocarbons. However, this treatment requires a lot of space and sometimes it cannot be utilized for that very reason.

   There are three solid-phase bioremediation techniques:

1. Landfarming
2. Biopiling
3. Composting.

Slurry Phase In the slurry phase the contaminated soil is excavated and removed from the site as completely as would be prudent. The contaminants are then put into a large tank, known as a bioreactor, in which the contaminants and microbes are mixed by cleanup crews. The mixing process maintains the microorganisms in continuous contact with the contaminants. Nutrients, water, and oxygen are added to the mix.

When the cleanup groups have controlled the conditions in the bioreactor; they can make changes until they achieve the optimal conditions for the degradation of the contaminants. Subsequently, the degradation can be kept at or close to optimal conditions, and the contaminants can be itemized within a short time.

Slurry phase bioremediation is considerably quicker than numerous other bioremediation techniques. It is exceptionally helpful in cases in which the contaminants should be itemized rapidly.

Also, slurry stage bioremediation can sometimes, but not always, provide a permanent solution. Its success is highly dependent upon the chemical properties of the soil and the contaminants. Slurry phase bioremediation definitely has some disadvantages. For example, the rate of treatment is limited by the size of the bioreactor. That is, if a small bioreactor is being used, the rate of degradation will be very slow. Also, additional treatment of the wastewater is required. After the additional treatment, the wastewater must then somehow be disposed of. These things add quite a bit to the cost. They are part of the reason that slurry phase bioremediation has a high operating cost as well as a fairly high capital cost.

http://matts-bioremediation.tripod.com/id4.html.

16.2.2.4 Advantages of Ex Situ Bioremediation

1. Better controlled and more efficient process than in situ bioremediation.
2. The process can be improved by enrichment with desired microorganisms.
3. The time required is short.

16.2.2.5 Disadvantages of Ex Situ Bioremediation

1. It is a very costly process.
2. Polluted sites are highly disrupted.
3. There may be disposal problems after the process is completed.

16.3 Phytoremediation

Phytoremediation is a rising technology for cleaning up contaminated sites; it is economical and has long-term applicability and it also has esthetic advantages. It is best used at sites with shallow contamination by organometallic pollutants, with a nutrient that is suitable for any one of the following five applications: (a) phytotransformation, (b) rhizosphere bioremediation, (c) phytostabilization, (d) phytoextraction, and (e) rhizo-filtration. The technology includes effective utilization of plants to eliminate or immobilize (detoxify) ecological contaminants in a growth matrix (soil, water, or sediments) through the biological, natural, and chemical or physical characteristics of the plants.

16.3.1 Phytoremediation of Heavy Metals (HMs)

Water and soil pollution by HMs is now an environmental concern. Various inorganic contaminants and metals represent the most widespread types of contamination found at many sites and their remediation in soils and sediments is troublesome. The high cost of existing cleanup technologies has promoted the pursuit of new cleanup procedures that involve minimal effort, have few side effects, are safe to use, and are environmentally sound. Phytoremediation is a cleanup idea that includes the utilization of plants to clean or balance a contaminated environment.

Phytoremediation can be utilized to disinfect soils contaminated with inorganic toxins. In situ, solar-driven technology makes use of vascular plants that aggregate and translocate metals from their roots to their shoots. Harvesting the plant shoots can then permanently eliminate these contaminants from the soil.

Phytoremediation does not have a dangerous effect on soil fertility and structure, such as corrosive extraction and soil washing, that some more vigorous conventional technologies have. Phytoremediation can be applied in situ to remediate groundwater, surface water bodies, and shallow soil. In addition, phytoremediation is green and more environmentally friendly than more dynamic and intrusive remedial techniques; it is a low-tech substitute for such techniques.

16.4 Microbial Remediation and Mycoremediation

Mycoremediation uses the digestive enzymes of fungi to break down contaminants such as pesticides, hydrocarbons, and HMs, while microbial remediation is used for aromatics-contaminated soil, which is of specific environmental concern, as these agents have cancer-causing and mutagenic properties. Bioremediation is a natural/biological strategy for the removal of soil contaminants; it has several advantages over conventional soil remediation procedures, including high efficiency, complete pollutant removal, low cost, and operational efficiency over a large range of contaminants. Bioaugmentation, which is characterized as the use of particular strains or consortia of microbes, is a generally applied bioremediation technology for soil remediation. In this way, it is closed which a few effective investigations of bioaugmentation of aromatics-polluted soil by single strains or blended consortia.

In recent decades, various reports have been published on the metabolic machinery of aromatics degradation by microorganisms and the capacity of these microorganisms to adjust to aromatics-contaminated environments. With these characteristics, microorganisms are the chief players in site remediation. The bioremediation/bioaugmentation process depends on the enormous metabolic limits of organisms for the transformation of aromatic pollutants into, principally, safe, or at any rate, less harmful compounds.

Aromatics-contaminated soils are effectively remediated with single strains of bacteria or fungi, as well as bacterial or fungal consortia. There are also a few novel methodologies in which physical and biological factors or chemicals are used to boost the efficacy of microbes in the remediation of aromatics-contaminated soil. Environmental factors also have a considerable effect on the bioaugmentation procedure. A biostatistics strategy is suggested for examination of the impact of bioaugmentation treatments (Fig. 16.5).

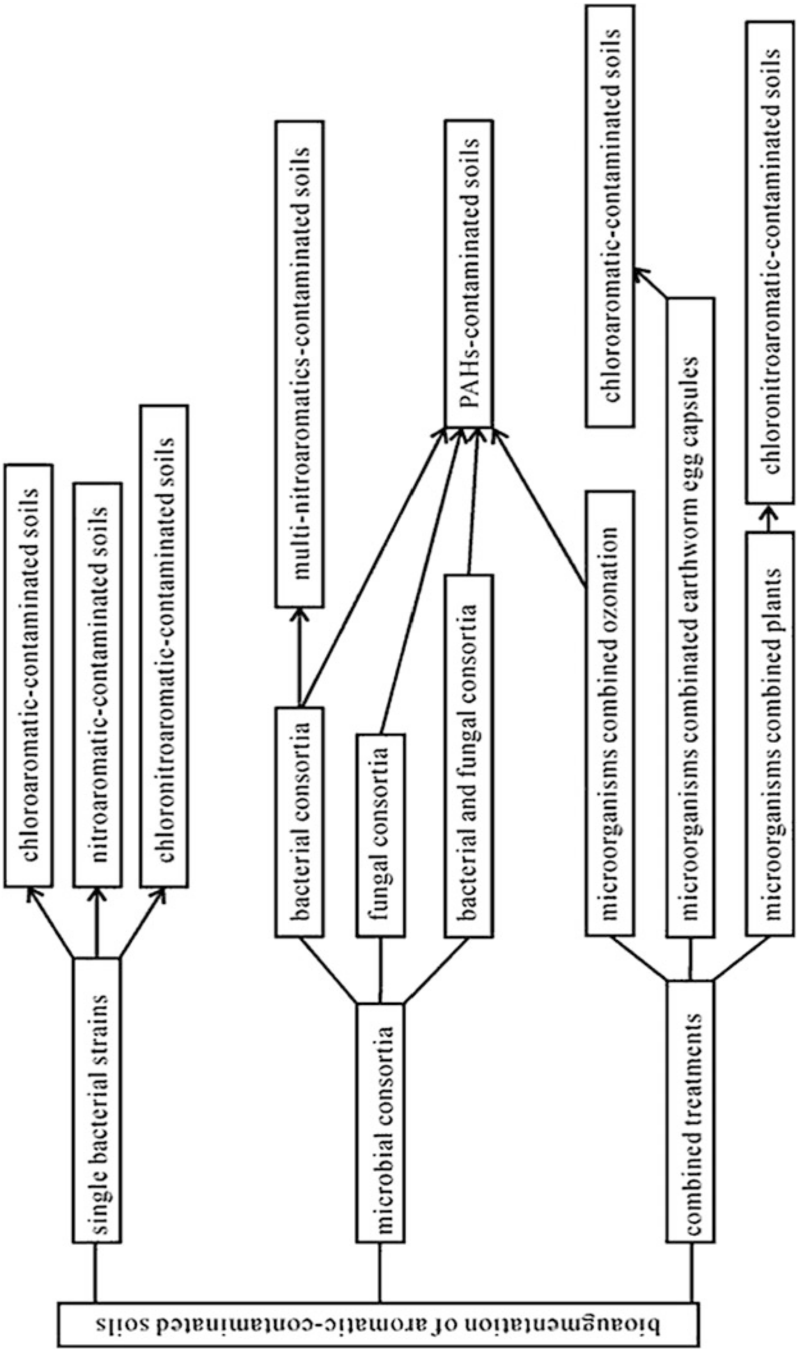

Fig. 16.5 Microbial remediation of aromatics-contaminated soil. Source: (Xu and Zhou 2017)

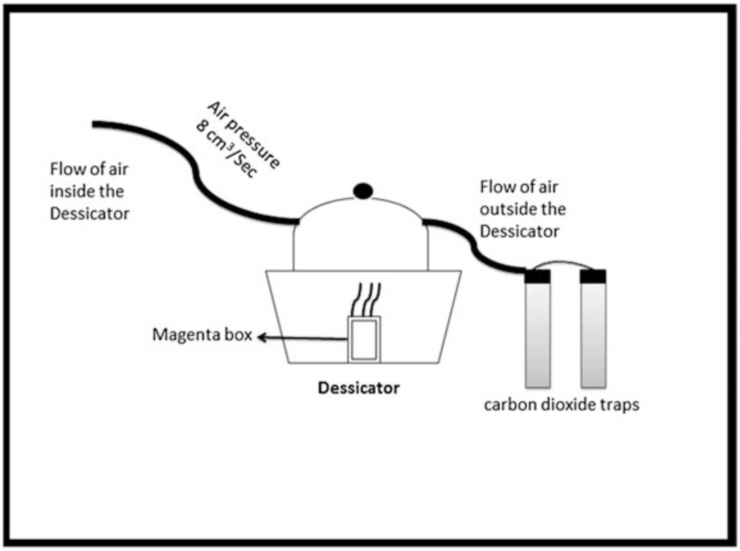

Fig. 16.6 Schematic diagram of microbes used for the examination of mineralization through barley colonized with *Mycobacterium* KMS

16.5 Rhizoremediation

Rhizoremediation is a process whereby microorganisms degrade soil contaminants in the rhizosphere. Soil pollutants that are remediated by this method are generally organic compounds that cannot enter the plant because of their high hydrophobicity. Plants are generally not considered as the main mode of remediation in this technique. Rather, the plant creates a niche that allows rhizosphere microorganisms to degrade the soil contaminants. Rhizosphere microbes are served by the plant functioning as a solar-powered pump that attracts water and the contaminant, while the plant carries out synthesis of substrates that advantage microbial survival and development. Root exudates and root turnover can fill in as substrates for microorganisms that perform toxin degradation (Fig. 16.6).

The selection of organisms that may be useful in rhizoremediation has been successful. Using contaminated soil as the initial media from which to select, bacteria that can survive on the contaminant of interest can be enriched. This enriched fraction can then be inoculated into plants where selection for root colonization can be done. Using this process will result in a host plant that supports a pollutant degrader in its rhizosphere. Wild-type organisms are selected in this process and therefore there is no constraint on their usage, as there is with genetically modified microorganisms. Kuiper et al. (2001) describe the successful use of this

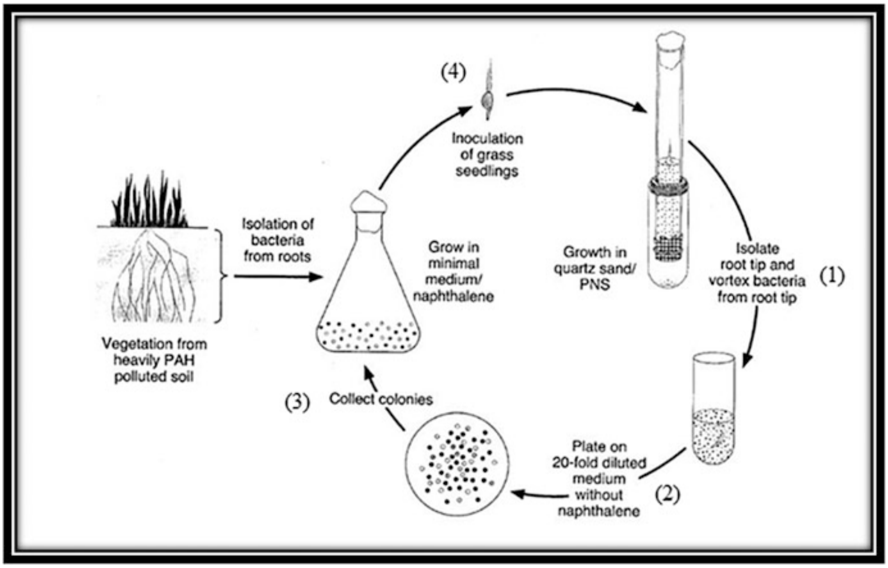

Fig. 16.7 Schematic diagram of Napthalene-degrading media for the screening of root colonizing bacteria (1) isolation of bacteria (2) bacterial isolation on 20-fold on soya agar medium (3) grown in standard naphthalene medium (4) on *Lolium multiflorum*-colonized cultivar Bamultra in a gnotobiotic system. The process is repeated twice

technique to identify rhizosphere polycyclic aromatic hydrocarbon (PAH) degraders.

Another technique used in rhizoremediation research is a method for determining the mineralization of organic pollutants in the rhizosphere. Labeled carbon dioxide that is produced during pollutant degradation is trapped and analyzed (Fig. 16.7).

Some examples of the rhizodegradation of PAHs, polychlorinated biphenyls (PCBs), trichloroethylene (TCE), and pesticides are shown in Table 16.1.

16.5.1 Beneficial Plant-Microbe Interaction

Soil and groundwater contamination is an extreme issue worldwide. The negative impacts of pollutants on the surface of the earth and on human wellbeing are various. The search for strategies that can substitute for incineration and excavation in the cleaning of contaminated sites has led to the use of bioremediation methods.

During rhizoremediation, exudates from the plant can stimulate the survival and activity of microbes, bringing about a more effective degradation of poisons. The root arrangement or floral system can spread microorganisms such as bacteria in the soil and help the bacteria to penetrate soil layers that are generally impermeable. The

Table 16.1 Rhizoremediation of various environmental pollutants (from Kuiper et al. 2004)

Plant	Pollutant	Microbes
Rice (cultivar; cv. Supriya)	Parathion	Not identified
Mixture of grass, legume, herb, and pine	TCE	Not identified
Prairie grasses	PAHs	Not identified
Prairie grasses	PAHs	Not identified
Grasses and alfalfa	Pyrene, anthracene, phenanthrene	Not identified
Sugar beet (cv. Rex)	PCBs	*Pseudomonas fluorescens*
Undefined wild plants (*Compositae*) and *Senecus glaucus*	Crude oil	*Arthrobacter*/*Penicillium*
Barley (*Hordeum vulgare*)	2,4-D	*Burkholderia cepacia*
Alfalfa and alpine bluegrass	Hexadecane and PAHs	Not identified
Wheat (*Triticum aestivum*)	2,4-D	*P. putida* strains
Poplar (*Populus deltoides nigra*)	1,4-dioxane	*Actinomycetes*
Wheat	TCE	*P. fluorescens*
Oat, lupin, rape, dill, pepper, radish, pine	Pyrene	Not identified
Reed (*Phragmitis australies*)	Fixed nitrogen	*Nitrospira* sp. and *Nitrosomonas* sp.
Poplar root extract	1,4-dioxane	*Actinomycete amycolatum* sp. CB1190
Corn (*Zea mays*)	3-methylbenzoate	*P. putida*
Astragalus sinicus	Cd^+	*Mesorhizobium huakuii*
Fern (*Azolla pinnata*)	Diesel fuel	Not identified

TCE trichloroethylene, *PAHs* polycyclic aromatic hydrocarbons, *PCBs* polychlorinated biphenyls, *2,4-D* 2,4-dichloro-phenoxyacetic acid

inoculation of toxin-degrading microorganisms on plant seed can be a critical factor for enhancing the efficacy of phytoremediation or bioaugmentation (Kuiper et al. 2004).

16.6 Environmental Remediation

Environmental remediation involves the elimination of contaminants or pollution from environmental media; for example, surface water, soil, groundwater, or sediment. Once requested by a legislature or a land remediation specialist, environmental remediation should be carried out promptly, to reduce adverse effects on human wellbeing and the environment.

To help with environmental remediation, environmental remediation services can be employed. These services help eliminate pollution sources in order to help protect the environment.

16.7 Bioremediation of Contaminated Paddy Soil

Overall, in the e-waste recycling era, paddy fields have significantly lower levels of PCBs than dry land. The nutritional and redox characteristics of planted paddy fields can beneficially affect the fate of PCBs, because these characteristics make enhanced sequential anaerobic and aerobic dechlorination possible. Moreover, it was found that waterlogging benefitted microbial reductive dechlorination, while drying was preferable for the degradation of dechlorination products (Mayer 2001). Also, rice roots accelerated PCB attenuation, showing a faster removal rate than that seen with drying one (Sharma et al. 2018). In view of the low accumulation of PCBs in rice tissues, the use of rice paddy fields, which can act as natural sequential anaerobic-aerobic bioreactors, has proven to be a cost-effective means to accelerate PCB attenuation. Future studies involving plot trials on a large scale to evaluate the effects of natural attenuation of PCBs in situ in paddy fields could provide additional accurate assessments of the effectiveness of natural restoration for reducing the risks associated with PCBs to negligible levels in the agricultural environment.

16.8 Polycyclic Aromatic Hydrocarbons (PAHs)

Phenanthrene, naphthalene, benzo(a) mixes, and pyrene are PAHs; however, toluene, xylene, and benzene are the PAHs that are registered as significant pollutants by the United States Environmental Protection Agency. Because of their low solubility in water and high stability PAHs are difficult to remove from polluted media (Husain 2010).

Fungi transform PAH co-metabolically because they do not use PAHs as a source of carbon. Fungi that are able to degrade lignin (white rot ligninolytic fungi), as well as *Plurotus ostreatus* and *Pityriasis versicolor*, show powerful PAH degradation by means of laccase-mediated transformation, as revealed in numerous studies (Anastasi et al. 2010; Mollea et al. 2005; Bogan and Lamar 1999; Rama et al. 2001). Moreover, PAH degradation and ligninolytic enzymatic production showed a positive correlation (Novotný et al. 1999).

In any case, encouraging results were acquired when free laccases were specifically added to PAH-polluted soil, degrading a blend of 15 PAHs; anthracene degradation accounted for 60% of PAH degradation, this being the highest of the PAHs examined (Wu et al. 2008).

A wide-ranging examination of transmissible ligninolytic fungal strains found that a fungal strain containing the enzyme Mn-peroxidase showed the greatest extent of naphthalene degradation, at 69%, while a strain containing lignin peroxidase and laccase also degraded naphthalene. Likewise, fungi containing Mn-peroxidase and laccase degraded phenanthrene (Clemente et al. 2001).

The non-ligninolytic fungi *Cladosporium sphaerospermum* and *Cunninghamella elegans* degraded PAHs (Potin et al. 2004). Especially, *Cunninghamella elegans* degraded many PAHs (e.g., naphthalene, acenaphthene, anthracene, phenanthrene, benzo[a]pyrene, anthracene, fluoranthene, and pyrene). Many non-ligninolytic fungi degrade PAHs via cytochrome P450 monooxygenase and epoxide hydrolase-catalyzed reactions to form trans dihydrodiols (Reineke 2001).

16.9 Heavy Metal (HM) Mobilization in Contaminated Paddy Soil

For improved phytoextraction, the mobilization of HMs from the soil solid phase to soil pore water is a significant process.

The pot incubation test, which is practical for imitating field conditions, led to research on three substances added to soil for mobilizing HMs from contaminated paddy soil namely, the [S,S] isomer of ethylenediamine disuccinate (EDDS), with application rates of 4.3, 11.8, and 2.3 mmol kg^{-1} of soil; ethylenediamine tetraacetate (EDTA; 7.5, 1.4, and 3.8 mmol kg^{-1}); and sulfur (400, 100, and 200 mmol kg^{-1}).

Subsequent changes occurred in soil pore water HM, and release of carbon fixations along with pH alterations was observed for 119 days. EDDS was the best-added substance to accumulate soil Cu. During the entire experimentation, EDDS was just found compelling amid the initial 24 to 52 days, and was promptly degraded biologically with a partial existence of 4.1 to 8.7 days. The adequacy of EDDS diminished at the most astounding application rate, most presumably because of exhaustion of the promptly prudent Cu pool in the soil. EDTA expanded in the soil pore water while the concentrations of the heavy metals remained powerful during the entire incubation time frame because of their industriousness. The most noteworthy rate of sulfur application prompted a diminishing pH, to nearly 4.

The pot incubation test procedure increased the pore water concentrations of HMs, particularly those of Cd and Zn. In the soil pore water, concentrations of HMs can be determined to a high degree by choosing the best possible application rate of sulfur, EDDS, or EDTA. Our pot test, in combination with additional plant test experiments, will, we trust, provide an appropriate apparatus to assess the relevance of various soil additives for achieving upgraded phytoextraction in a particular soil (Wang et al. 2007).

16.9.1 HM Contamination and Remediation in Asian Agricultural Land

It is important to consider the HM contamination status, sources, and remediation in the agricultural land of most Asian countries (in particular, China), which are undergoing rapid economic development. Some farmland soils in the suburbs of most cities and sewage irrigation districts in China are polluted to some extent with HMs such as Cd, As, Zn, Cu, and Hg, resulting in metal contamination of agricultural products, thus posing a potential risk to human health. It has been reported that, in Asian countries, foodstuffs such as vegetables, grains, and domestic animal feed are highly contaminated with HMs. The sources of HMs in arable lands in most Asian countries include natural sources, as well as mining, smelting, agrochemicals, sewage sludge, and livestock manure. There are systematic remediation technologies for contaminated soils, which include physical/chemical remediation, phytoremediation, microbial remediation, and integrated remediation.

16.9.1.1 Soil Contamination with HMs in China and Other Asian Countries

16.9.1.1.1 Accumulation and Impacts of HMs in Agricultural Soils

South and Southeast Asian countries; for instance, Vietnam, peninsular Malaysia, Philippines, India, Indonesia, Pakistan, Bangladesh, and Thailand have paid much attention to the contamination of cultivated crops and soils by HMs, because of the potential effects on the long-term effects on food production and human health in the contaminated areas. It was reported that, in Korea, the average concentrations of copper, zinc, cadmium, and lead (Pb) in the surface layer of rice paddy soils (0–15 cm) were 0.11 mg kg^{-1} (range, 0 to 1.01), 0.47 mg kg^{-1} (0–41.6), 4.84 mg kg^{-1} (0–66.4), and 4.47 mg kg^{-1} (0–96.7), respectively. In orchard fields, the average concentrations of cadmium, copper, Pb, zinc, arsenic (As), and mercury (Hg) in surface soils (0–20 cm) were 0.11 mg kg^{-1} (range, 0 to 0.49), 3.62 mg kg^{-1} (0.03–45.3), 2.30 mg kg^{-1} (0–27.8), 16.60 mg kg^{-1} (0.33–106), 0.44 mg kg^{-1} (0–4.14), and 0.05 mg kg^{-1} (0.01–0.54), respectively. In Japan, the estimated average levels of Cd, Cu, and Zn in rice were 75.9 mg kg^{-1}, 3.71 mg kg^{-1}, and 22.9 mg kg^{-1}, respectively. The average levels of Cd, Cu, and Zn in rice fields were 446 mg kg^{-1}, 19.5 mg kg^{-1}, and 96.4 mg kg^{-1}, respectively (Herawati et al. 2000). China is facing soil contamination problems, especially HM pollution (Luo and Teng 2006; Brus et al. 2009). It was estimated that, in China, nearly 20 million ha of arable soil (approximately one-fifth of the total area of farmland) was contaminated by HM, and this was expected to result in a decrease of more than 10 million tons per annum of food supplies in China (Wei and Chen 2001). The proportion of exchangeable fractions of Cd in the soil of the Zhangshi irrigation area in Shenyang, Liaoning province, with a history of sewage irrigation of more than 45 years, was much higher than these proportions of Cu and Pb. It was suggested that Cd could be the most mobile element in the soil and thus more available to the crops, with a great risk of moving into the food chain. As a consequence, Cd contamination in the arable soils became the most serious problem in this region (Xiong et al. 2003). Other areas, including the Lake Taihu plain and the Pearl River Delta region, which have been under rapid economic development, were all recently found to have moderate to serious contamination by HMs (Huang et al. 2007; Hang et al. 2009). The accumulation of HMs in crops grown in metal-polluted soil may easily damage human health through the food chain. Fu et al. carried out an investigation on HM contents in rice sampled from Taizhou city in Zhejiang province, China, and found that the geometric mean level of Pb in polished rice reached 0.69 mg kg^{-1}, which was 3.5-fold higher than the maximum allowable concentration (MAC) (0.20 mg kg^{-1}) in the safety criteria for milled rice. Cd contents in 31% of the rice samples exceeded the national MAC (0.20 mg kg^{-1}), and the arithmetic mean also slightly exceeded the national MAC. In the Dabaoshan mine area of Guangdong Province, China, the surrounding farmland has been seriously contaminated by Cd and other toxic metals as a result of long-term mining (mainly iron and copper), as well as the discharge of untreated wastewater. The average concentration of cadmium in rice from this farmland exceeded, by150 times, the State Food and Health Standards, which has caused a health risk to local residents.

Norra et al. (2005) reported that the concentration of As in winter wheat grain cultivated in the agricultural area of West Bengal Delta Plain, which is irrigated with As-rich groundwater, could reach 0.7 mg kg^{-1}.

16.9.2 Sources of HMs in Agricultural Soils

It is important to identify the sources and status of soil contaminated by toxic metals so as to undertake appropriate treatments to reduce soil contamination and to maintain sustainable agricultural development.

Natural Sources The initial sources of HMs in soils are closely related materials from which the soils were formed, but the impact of parent materials on the total concentrations and forms of metals in soils is modified to varying degrees by pedogenetic processes (Herawati et al. 2000). In areas affected only slightly by human activities, HMs in the soils, derived from pedogenetic parent materials, and metal accumulation status, are affected by several factors, such as soil moisture and management patterns. Research conducted in Gansu Province, China, by Li concluded that, in three arid agricultural areas, a lithological factor was the main factor responsible for HM accumulation. It was reported that the soil aqua regia-soluble fractions of cobalt (Co), nickel (Ni), lead (Pb), and zinc (Zn) were highly associated with soil aluminum (Al) and iron (Fe). These elements were associated with indigenous clay minerals in soils that were high in Al and Fe.

Mining In mining areas, there are different sources of metal contamination including:

(a) Grinding,
(b) Concentrating ores, and
(c) Tailings disposal (Wang et al. 2004; Adriano 1986).

Inappropriate treatment of tailings and acid mine drainage can pollute agricultural fields surrounding the mining areas (Williams et al. 2009). Taking Tongling copper mine in Anhui Province in China as an example, metal mining has been an important economic activity in this area from ancient times. The major mining areas have been concentrated in a narrow star-shaped basin called Fenghuang Mountain. Long-term mining activities in this area have caused widespread metal pollution. The average soil concentration of total Cu was 618 mg kg^{-1}, with a wide range of 78–2830 mg kg^{-1}. Lead concentration in the soil also showed high variability, with a mean of 161 mg kg^{-1}. The total Zn concentration varied from 78 to 1280 mg kg^{-1}, with an average of 354 mg kg^{-1} (Wang et al. 2004). It was reported that the majority of the agricultural soils in the area were contaminated with As. The high As concentrations in these soils may be attributed to arsenopyrite, which is known to occur in many areas of Southeast Asia, especially in tin mining regions (Patel et al. 2005).

Smelting and Flying Ash It was reported that atmospheric deposition accounted for 43–85% of the total contents of Hg, Cr, Pb, and Ni in agricultural soils in China. Actually, most of the HM pollutants in the air are derived from flying ash caused by anthropogenic activities (Liu et al. 2006) such as electric power generation, mining, and metal smelting and chemical plants.

Total trace element deposition (wet and dry) to agricultural soils was calculated by Luo from the average deposition fluxes of each element and the total agricultural land area 1.22×10^8 ha in 2005 in China. It was accounted for that the deposition from the climate in China was, by and large, greater than New Zealand with the exception of Zn and equivalent to the region of Tokyo Bay. The most common elements deposited from the atmosphere were Hg, Pb, As, Cd, and Zn, and non-ferrous metal smelting and coal combustion were two of the most important contributants to metal pollutants in the air. Streets et al. (2005) report that, in China, roughly 38% of mercury (Hg) pollution originates from coal burning, with 45% originating from non-ferrous metal refining and 17% originating from different activities, of which battery, cement, and fluorescent light production is of general significance.

In China, Zn was the metal deposited in agricultural soils in the largest amount from the atmosphere, followed by lead (Pb) and copper (Cu).

Fertilizers and Agrochemicals The content of HM in arable soils as a result of fertilizer use is of increasing concern because of the possible risk to environmental health.

Lu et al. (1992) claimed that, among all inorganic fertilizers, phosphate fertilizers are usually the chief source of trace metals, and much attention has been paid to the Cd content in phosphate fertilizers. However, the concentration of Cd in both phosphate rocks and phosphate fertilizers from China is considerably less than that in phosphate rocks and phosphate fertilizers from European nations and the United States. It should be concerned that despite the fact that the fertilizer of lethal metals in the majority of the manures in China was lower than the most extreme restrains, the trace elements input to agricultural land were as yet worth concern, since the yearly utilization of fertilizers accounted for 7.4, 4.7×10^6 and 22.2 tons for K, N and P manures (in unadulterated supplement), separately (NBSC 2006).

In some of the countries mentioned above, phosphate fertilizers have been used for long periods. For instance, the great majority of agricultural soils in Malaysia are heavily treated with this kind of fertilizer, as reported by Zarcinas et al. (2004). Regression analysis showed that log aqua regia-soluble levels of As, Cu, Cd, and Zn in soil in Malaysia were significantly correlated with log aqua regia-soluble phosphate. Soils in these southern Asian countries have phosphate requirments, and the history of the addition of phosphate fertilizer with its related impurities (Cd, Cu, As, and Zn), seems to be longer in these countries than elsewhere (Zarcinas et al. 2004). The agricultural use of pesticides is another non-point source of HM pollution in arable soils. Although the use of pesticides containing Cd, Hg, and Pb was prohibited in 2002, pesticides containing other trace elements, especially copper and zinc, are still in existence. It was estimated that a total input of 5000 tons of Cu and 1200 tons of Zn in agrochemical products was applied annually to agricultural land in China (Wu 2005). Cocoa, groundnut, mustard, and rice had elevated concentrations of HMs (especially Cu and Zn) when compared with findings in other plants (cabbage, oil palm, aubergine, okra). This may be explained by the widespread use of Cu and Zn pesticides on these crops. A survey also showed that HM concentration in surface horizon and in edible parts of vegetables increased over time. Pandey et al. (2000) reported that the metal concentration in soil increased from 8.00 to 12.0 mg kg^{-1} for Cd, and from 278 to 394 mg kg^{-1} for Zn. They also suggested that if the trend of atmospheric deposition continues, it would lead to a destabilizing effect on sustainable agricultural practice and increase the dietary intake of toxic metals. Sinha et al. (2006) concluded that the vegetables and crops growing in such areas in India constituted a risk owing to the accumulation of metals. These researchers also studied the effect of municipal wastewater irrigation on HM accumulation in vegetables and agricultural soils in India. The mean concentrations of Cr, Zn, Ni, Cd, Cu, and Pb in the wastewater-irrigated soil around the Titagarh region were 104, 130, 148, 30.7, 90.0, and 217 mg kg^{-1}, respectively. Also, the concentrations of Pb, Zn, Cd, Cr, and Ni in all vegetables examined (mint, cauliflower, celery, spinach, coriander, parsley, Chinese onion and radish) were over the safe limits. Industrial effluents often contain many HMs. In industrial areas in India, many agricultural fields are inundated by mixed industrial effluent or irrigated with treated industrial wastewater. In one such area, the metal with the highest available content in soil was Fe, with levels of 529 to 2615 mg kg^{-1}, while Ni had the lowest level of, of 3.12 to 10.5 mg kg^{-1}. The results also suggested that the accumulation of Cr in leafy vegetables was greater than that in fruit-bearing vegetables and other crops (Sinha et al. 2006).

Wastewater Irrigation Farmland irrigated by wastewater in China accounted for 36,180,000 ha, occupying approximately 7.3% of the total irrigation area (Bulletin of Environmental Status in China 1998). Sewage irrigation can alleviate water shortages to some extent, but it can also transport some toxic materials, especially HMs, to agricultural soils, and cause serious environmental problems. This is predominantly a problem in overpopulated developing countries where pressure on irrigation water resources is extremely great, such as in northern dry lands in

China. In 2005, the quantity of wastewater used in China had reached 5.25×10^{10} tons, of which industrial wastewater accounted for 2.43×10^{10} tons (SEPAC 2006). The most important wastewater irrigation areas in China are the Zhangshi wastewater irrigation area in Shenyang Liaoning Province, the Xi'an wastewater irrigation area in Shaanxi Province, the Beijing sewage irrigation area, and the Shanghai wastewater irrigation area. In Chhattisgarh, central India, the soil was irrigated with arsenic-polluted groundwater, and people in this region suffered from arsenic-borne diseases. The arsenic concentration in the polluted water ranged from 15 to 825 μg L^{-1}, exceeding the permissible limit of 10 μg L^{-1}. The contaminated soil had a median level of 9.5 mg kg^{-1} of arsenic (Patel et al. 2005). Numerous modern industrial plants in this district work with no or minor wastewater treatment and routinely release their wastewater into drains, which either pollute waterways and streams or add to the contaminant burden of biosolids. Biosolids are progressively being utilized as soil ameliorants, and streams and waterways are the essential sources of water for the water system.

Sewage Sludge Application Although the contents of toxic metals in sewage sludge had also been markedly reduced, and most of them were below the national discharge standard of pollutants for municipal wastewater treatment plants, due to the huge increase in the amount of wastewater treated, the sewage sludge produced increased rapidly. As indicated by information from State Environmental Protection Administration of China (SEPAC) (2006), roughly 4.6×10^6 tons (dry weight) of city sewage sludge was created in China in 2005. It was estimated that in China the direct application of sewage sludge to agricultural land accounted for 10% of fertilizer use. Cu is strongly attached to organic material and may be added as a contaminant with organic soil improvements. There is now also a considerable body of evidence documenting long-term exposure to high concentrations of HM (e.g., Cu) as a result of past applications of sewage sludge (McGrath 1994); as a result of past applications of Cu and Zn from animal manure (Christie and Beattie 1989); and as a result of past applications of Cu-containing fungicides (Zelles et al. 1994). In the agricultural areas of Hyderabad, Pakistan, researchers studying the effect of the long-term application of wastewater sludge on the concentrations of HMs in soil irrigated with fresh canal water (SIFW) and soil irrigated with wastewater (SIDWS) reported the following findings: the total mean concentrations of Cd, Pb, Cu, and Zn were 11.2, 105, 21.1, and 1.6 mg kg^{-1}, respectively, in SIFW and 32.2, 209, 67.4, and 4.3 mg kg^{-1}, respectively, in SIDWS. The concentrations of metals in the SIDWS were generally higher than those in the SIFW. The high percentage of Cd and Cr in SIDWS was attributed by the authors to waste effluent from small industries (tanneries and batteries) situated near domestic areas (Jamali et al. 2007).

Livestock Manure In China and in other Asian developing countries, people's demand for meat, eggs, and dairy products has risen greatly over the past decades, owing to the continuous rise in living standards. Heavy metals are present in livestock fodder as additives for animal health and for other purposes. For example, arsenic had been used as a feed additive for growth improvement and for the control of diseases in pigs and poultry. Unfortunately, it was still in use in certain countries;

for instance, in China and the United States, although its use as an animal feed preservative had been prohibited in Europe. According to many reports, the concentrations of HMs in poultry manure have increased with the usage of feed additives. Livestock fertilizers accounted for approximately 69%, 51%, and 55%, of total Zn, Cd, and Cu concentrations, respectively. Among the HMs investigated by Luo et al. in agricultural soils in China, Cd was a top problem, with an average level of 0.004 mg kg^{-1} $year^{-1}$ in the plough layer (top 0–20 cm of soil).

Soil Pollution Control and Remediation Conventional techniques utilized for the remediation of soils polluted by metals and the control of contaminated areas include:

1. ***Land filling:*** the removal of contaminated soils and their transport to an area in which it is permitted to deposit dangerous waste.
2. ***Fixation:*** the chemical processing of soil to immobilize the metals, usually followed by treatment of the soil surface to eliminate penetration by water, and
3. ***Leaching***: using acid solutions as proprietary leaching agents to leach metals from soil, followed by the return of the clean soil residue to the original site (Krishnamurti 2000).

Conventional methods used for metal detoxification are cost effective, but produce large quantities of toxic products. The advent of bioremediation technology has provided an alternative to conventional methods for remediating metal-polluted soils (Khan et al. 2009). Systematic remediation technologies have been developed for contaminated soil; these include bioremediation, physical/chemical remediation, and integrated remediation. Various development trends in soil remediation are summarized as follows: green and environmentally friendly bioremediation, combined and hybrid remediation, in situ remediation, environmentally functional material-based remediation, equipment-based site remediation, remediation decision-supporting systems, and post-remediation assessment. Phytoremediation is another emerging low-cost in situ technology that is employed to remove pollutants from contaminated soils. Much work in metal phytoremediation, based on laboratory, glasshouse, and field experiments, has been carried out in China during the past decade. The effectiveness of phytoremediation can be improved by the careful and cautious application of applicable heavy-metal-tolerant, plant growth-promoting rhizobacteria, e.g., symbiotic nitrogen-fixing organisms (Khan et al. 2009). Leafy vegetables, especially mint, from SIDWS contained higher levels of Zn, Cd, and Pb than other vegetables grown at the same site, suggesting that the cultivation of leafy vegetables should be avoided in SIDWS (Jamali et al. 2007). Mani et al. (2007) investigated the interaction between Cd, Ca, and Zn and organic matter for Cd-phytoremediation in sunflowers. The results suggested that phytoremediation of Cd-contaminated soil could be performed through soil-plant-rhizospheric processes. *Bacillus sphaericus* was shown to be tolerant to 800 mg L^{-1} Cr (VI) and was reduced by >80% during growth (Pal and Paul 2004). A study revealed the relationship between Cd adsorption by soil and the properties of the soil, and the influence on Cd uptake by plant roots. The results indicated that the adsorption capacity of the soils for Cd increased with increases in the pH or alkalinity of the soil. However, the adsorption rate of Cd decreased with increased in pH. The results also indicated that the Cd adsorption capacity of tropical vertisols was higher than that of temperate vertisols (Ramachandran and Dsouza 1999). Adhikari and Singh (2008) studied the effect of city compost, lime, gypsum, and phosphate on Cd mobility by columns. Of all the treatments, lime application reduced the movement of Cd from the surface soil to lower depths of the soil to a large extent. And the combined application of lime and city compost reduced the movement of Cd in the soil profile. These results showed that high soil pH may reduce the mobility of Cd, and organic matter may control the sorption of Cd in the soil. It is imperative to develop safe and cost-effective in situ bioremediation and physical/chemical stabilization technologies that can be used broadly for moderately or slightly contaminated farmland; to develop safe, land-reusable, site-specific physical/chemical and engineering remediation technologies for heavily polluted industrial sites; and to develop phytostabilization and eco-engineering remediation technologies for the control of soil erosion and pollutant diffusion in mined areas. Besides, it is also necessary to develop guidelines, standards, and policies for the management of remediation of contaminated soil. Asian countries should exert more effort in promoting international exchanges and regional cooperation for soil environmental protection and in enhancing the capacity of management and technology innovation.

16.10 New Plans

In regard to bioremediation (crime scene cleanup), the goal is to rid a site of potential biohazards such as blood, body fluids, and items that cause communicable diseases. Rather than clean up a crime or trauma scene with bleach or ammonia, which can have negative effects on the environment, bioremediation companies often sanitize using enzyme cleaners that do not have such effects.

16.11 How Does a Crime Scene Cleanup Work?

At the request of the victim's family, crime scene cleaners usually enter a trauma site after law enforcement officials have finalized their work at the scene. The job of a crime scene cleaner is not only to clean up blood and other potential biohazards that are left behind, but to also deodorize and completely sanitize the scene.

The Aftermath organization has been an industry leader in crime scene cleanups for almost 20 years; the company restores trauma scenes to hospital sanitization levels through the use of adenosine triphosphate (ATP) testing. The goal of using ATP is to identify the presence of organic material onsite by measuring cellular energy molecules. It is important to note that this branch of bioremediation varies greatly from the classic definition of the word, and that Aftermath does not handle the remediation of environmental pollutants.

http://www.aftermath.com/content/types-of-bioremediation

16.12 Conclusion

Paddy soil contamination has always been a challenging task for agricultural scientists and enormous efforts have been made to eliminate such contamination. Although scientific efforts have reduced the prevailing contamination and ameliorated the quality of paddy soil, there is still a long way to go to standardize soil conditions for the maximum production of rice and to meet the growing demand for food.

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