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Review

Phytoremediation and Bioremediation of Pesticide-Contaminated Soil

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Abstract: Management and destruction of obsolete pesticides and the remediation of pesticide-contaminated soil are significant global issues with importance in agriculture, environmental health and quality of life. Pesticide use and management have a history of problems because of insufficient knowledge of proper planning, storage, and use. This manuscript reviews recent literature with an emphasis on the management of obsolete pesticides and remediation of pesticide-contaminated soil. The rhizosphere of plants is a zone of active remediation. Plants also take up contaminated water and remove pesticides from soil. The beneficial effects of growing plants in pesticide-contaminated soil include pesticide transformation by both plant and microbial enzymes. This review addresses recent advances in the remediation of pesticide-contaminated soil with an emphasis on processes that are simple and can be applied widely in any country.

Keywords: pesticide; phytoremediation; soil; persistent; rhizosphere; obsolete

1. Introduction

The production and application of pesticides has a long history because of the many benefits associated with their use. There is a huge literature related to biodegradation of pesticides, microorganisms that degrade pesticides, bioremediation and phytoremediation of pesticide-contaminated soils, and soil amendments for pesticide remediation [1–305]. This review addresses recent advances in the remediation of pesticide-contaminated soil with an emphasis on processes that are simple and can be applied widely in any country. An attempt is made to include methods, content and literature that will be valuable to professionals in developing countries who are addressing pesticide remediation and management of pesticide-contaminated soils. Bioremediation and phytoremediation are inexpensive and can be implemented in many locations, and this manuscript has a focus on these remediation methods. In many locations, crops like vegetables need pesticides because of significant yield losses without pest control [82,149]. Pesticides have enhanced food production to meet the needs of a growing population [17,18,46,82,105,119,129,220,247].

However, large scale application of pesticides has often had a negative impact on human health and the environment [2,3,5,9,30,33,36,39,51,55,67,109,123,131,132,135,140,143,145,150,153,167,192,194,196,207,212,215,229,230,257,258,263].

The Peoples Republic of China, United States of America, Argentina, Thailand, and Brazil are the top five countries in pesticide use [16]. The use of pesticides in Africa has reportedly increased from 2055 to 4000 tons of active ingredients from 2004 to 2011 on a per country basis [68] but the data is of uncertain quality. Cameroon is a medium consumer of pesticides in Africa compared to higher consumers like Ghana and Mauritius and lower consumers such as Mozambique [194].

2. Types of Pesticides

Pesticides may be grouped according to the pest to be controlled (algicides, bactericides, fungicides, herbicides, insecticides, nematicides, and rodenticides) [34,234]. Chemical classes of organic pesticides include organochlorine, organophosphorus, acetamides, carbamates, triazoles and triazines, neonicotinoids and pyrethroids [179]. This review does not address inorganic pesticides including lead arsenate, chromated copper arsenate, copper acetoarsenite (Paris Green), Bordeaux mix (copper sulfate + calcium oxide), borax and boric acid complexes. The dithiocarbamate compexes Zineb (Dithane) and maneb, contain zinc or manganese, respectively. Removal of inorganic elements from soils requires different technologies than for organics [8,57,104,110,113,136,195,259].

One important group of pesticides are those that have been included on the list of persistent organic pollutants (POPs). Fourteen organochlorine compounds such as DDT (dichlorodiphenyltrichloroethane), chlordane, heptachlor, and toxaphene are included in the POP list [238,239]. These pesticides are persistent in the environment and have a high octanol/water partition coefficient which causes them to bioconcentrate in the lipid-rich tissue of organisms [2,67,90,95,167]. Some may cause cancer [170,197]. Some volatile POPs are transported through the air to locations where they pollute environments where they have not been applied [138,269]. Because of health and environmental issues, many organochlorine pesticides are no longer approved for use in many parts of the world [238,239,245]. There is still some approved use of DDT in some parts of Africa and India because of malaria [51,247,270].

The chemical and physical properties of pesticides are important because they impact the application process, effectiveness, and potential for in situ remediation. These include chemical composition, chemical structure, volatility (melting temperature, boiling point, vapor pressure, Henry's constant), half-life in the application environment, octanol/water partition coefficient (Kow), soil adsorption coefficient (Kd), and diffusivity in both air and water. Many values may be found in the published literature for many pesticides [40,102,132,141,144,277,291].

3. Obsolete Pesticides

There are significant quantities of obsolete (out-dated, or banned) pesticides in many countries. These pesticides need to be processed to reduce their toxicity because they are no longer useful or legal to use [16,24,42,52,61,65,69,73–75,101,138,161,170,173,177,209,231,235,260]. For example, Tarla and coworkers reported finding 210,047 kg and 309,521 L of obsolete pesticides in Cameroon. This included 4146 kg of POPs [231]. The risk assessment identified 195 sites with contamination [231]. In Kazakhstan 354.7 tons of pesticides were reported in 26 stockpiles [169,174]. In Ukraine, more than 3000 pesticide storage sites and 2000 polluted soil sites have been reported [156,157]. There has been some progress in transporting some of the hexachlorobenzene from a factory in Ukraine to Poland and Germany where it has been incinerated [140]. In some African countries obsolete pesticides have been collected and destroyed as part of the Asian Stockpiles Program [14,146,271]. There are reports of obsolete pesticides in Russia, former Soviet Union countries, Central and Eastern Europe, Asia, and the Middle East [6,55,61,70,91,94,117,134,170,246,252,261,271]. The Food and Agriculture Organization (FAO) has provided leadership and financial support for some of the efforts to address obsolete pesticide problems [271]. FAO estimates that the quantity of obsolete and unwanted pesticides is about

500,000 tonnes. About 10% of this is in Africa, and about 40% may be in the 12 former Soviet Union Republics [271]. While there has been significant progress in the incineration of obsolete pesticides, a significant portion of the 500,000 tonnes has not been destroyed as of 2019.

Incineration of pesticides at high temperature has become the method of choice to destroy obsolete pesticides [271–273] although this approach also has some disadvantages such as generating toxic emissions and ash that may contain potentially hazardous substances. Incinerators designed for thermal processing of pesticides and cement kilns that produce cement while burning pesticides as a fuel have been successfully used [271–273]. Sometimes a mixture of fuels is burned in order to achieve the desired results.

Risk assessment and risk management are important in working with pesticides because spills in storage sheds and onto soil may present risks for those who remediate the site [7,58,126,271]. Education of those who work with pesticides as well as the general public must be a high priority in order to implement health and safety programs that are designed to protect workers and the public [271,272].

There are many locations in the world where pesticides have been put into storage sheds that need to be cleaned up because the site is no longer needed for storage and the pesticides that are present at the site can no longer be applied for their intended purpose. There are safety concerns at some of these sites because of leaking containers and spills. In many cases there is soil contamination at storage sites and surrounding areas because pesticides have been spilled and dispersed [169,173,174].

4. Analysis of Pesticides to Monitor Clean-Up

Successful clean-up of obsolete pesticides requires sensitive analytical methods to monitor and verify that the clean-up process is working. There is significant literature on analytical methods to determine concentrations of pesticides in soil, water and foods [11,106,168,213,234,240,241]. This includes the US Environmental Protection Agency (EPA) Internet site under the heading Environmental Chemistry Methods [241], and AOAC International online and printed products [11]. Methods for organic pesticides include gas chromatography (GC), high-performance liquid chromatography (HPLC) [19,127], thin-layer chromatography (TLC) [96], spectrophotometry [120], polarography [208]. With GC, electron capture (ECD), nitrogen phosphorus (NPD), and flame photometric (FPD) detectors and mass spectrometry (GC–MS) have been used [56,77,142,203,213,248]. Liquid chromatography has been used with mass spectrometry (LC–MS) [213,240].

Sample preparation methods are an important part of chemical analysis of pesticides in soil and plants [11,241]. Numerous methods have been developed and used for the separation of pesticides from soil and plant matter. Liquid extraction processes have been used extensively for sample preparation [63,77,250]. Sonication has been used to enhance mixing and extraction effectiveness [77]. Solids are often present when pesticides are associated with plants or soils, and the extraction process may include extraction from a solid phase. Furthermore, many pesticides have much higher concentrations in fat and lipids. Determining the concentration of a pesticide is difficult if part of the pesticide is distributed in lipid-rich regions. In the analysis of pesticides, solid-phase extraction [11], solid-phase microextraction (SPME) [213], microwave-assisted extraction [108], and supercritical fluid extraction [203] have been used. There is a quick, easy, cheap, effective, rugged, and safe (QuEChRS) method for the preparation of pesticide samples [240].

Solid-phase extraction of samples from soil followed by chemical analysis using GC–MS has been reported for 103 pesticides including organochlorines, carbamates, organophosphates, and pyrethroid compounds [186]. EPA Method 508 has been used for analysis of chlorinated pesticides in water [241]. EPA Methods 508.1, 525.2, 527, 8081 B, and 8151 A have been used for pesticide analysis [241,242]. There are many reports with applications of chemical analysis methods and results at specific sites [19,56,107,125,186,203,213,240,248]. Silva et al. have used liquid chromatography tandem mass spectrometry (LC–MS/MS) and gas chromatography–high-resolution mass spectrometry (GC–HRMS) [282].

5. Soils Contaminated with Pesticides

Good productive soil has a vital role in agriculture, and it is a key component of terrestrial ecosystems. Pesticide-contaminated soil is a global concern because of spills, cleaning of spray equipment and empty containers, and land-use changes from agriculture to residential living. Erosion and flood waters may move pesticides from both treated land and storage sites to soils down-gradient and in the flood plain. Where high concentrations of active ingredients are released, both plants and microorganisms may be killed. There may be health risks when animals and people come to contaminated sites because of toxicity to non-target organisms [41,48,66,67,118,161,170]. Inappropriate pesticides may be taken up into garden vegetables and fruit. Pesticides in soil may contaminate ground water that is used as drinking water. Pesticides in soil from prior use may affect the fate of, or contaminate a different crop that is planted.

Silva et al. have reported results for 76 pesticide residues in topsoil for 11 European countries for 6 cropping systems. Many soils had more than one pesticide present; the maximum total pesticide concentration reported was 2.87 mg/kg [282]. Detection levels were 0.05 mg/kg for glyphosate, 0.01 mg/kg for LC–MS/MS detection, and 0.005 mg/kg for GC–HRMS detection [282]. No pesticide residues were reported in 17% of the fields. A single pesticide was identified in 25% of the fields, while more than one pesticide residue was found in 58% of the sites [282].

Yadav et al. have reviewed the literature on organochlorine pesticides in soil and reported values for 4 cities in Nepal [293]. A range of values is reported for data from China, India, Mexico, Uganda, Kenya, Pakistan, Central Europe, and Nepal for DDT, hexachlorocyclohexane, and endosulfans [293]. The largest value reported is 2.179 mg/kg = 2179 ng/g for DDT and metabolites of DDT [293]. Most values for DDT and its metabolites are greater than 0.1 ng/g and smaller than 100 ng/g [293]. The values reported by Zhang et al. for China are in a similar range [294].

Historically, excess or obsolete pesticides were buried in trenches, placed in dumps, or deposited in unlined landfills in many countries [30,84,246]. Disposing of pesticides in burial sites and landfills is not recommended in most countries, because of the risk of contaminating ground water. Remediation processes have been implemented to remove, contain, or degrade buried pesticides at a number of sites [84]. Monitoring of concentrations in ground water near landfills and pesticide burial sites has enabled communities to be informed of contamination issues and take appropriate action. It is important to collect information on burial sites and locations where ground water is contaminated in order to inform the public and prevent exposure to pesticide contaminants. Information on locations and contents of burial sites can help prevent using these sites for new construction [117,246].

6. Pesticides and Soil Ecology

One aspect of soil ecology is biodiversity of soil fauna. In healthy soils there are bacteria, fungi, protozoa, nematodes, arthropods, and earthworms. Bacteria and fungi are important for biodegradation of pesticides and other organic matter [59,114,191,199,200]. Arbuscular mycorrhizal fungi are associated with plant roots and rhizodegradation, and they improve ecological health [115,218]. Symbiotic fungi are beneficial for plants in contaminated soil, and they have a positive impact on pesticide degradation in the rhizosphere [116,187]. Mycorrhizal colonization of roots results in an increase of root surface area and enhances nutrient acquisition for plants [218,295]. Microbial endophytes protect plants and contribute to plant tolerance to herbicides [232] while rhizosphere-associated bacterial populations enhance pesticide degradation [references cited in 122, 133, 305]. Nematodes, arthropods, and earthworms are beneficial in soil as indicator organisms, organic matter degraders, and soil structure modifiers [49,93]. The populations of these organisms are negatively affected by some pesticides [41,62,93].

Before pesticides are released on the market, thorough testing of their active ingredients' ecotoxicological effects must be performed to evaluate whether the risk is at an acceptable level [300,304]. However, numerous studies are reporting the toxicity of pesticides on soil ecosystems [41,48,100]. These effects fall into three general categories: inhibition of microbial functions, reduction of microbial group abundance, changes in the composition and diversity of soil microbial communities. Reviews by Lo [298] and Puglisi [301] provide a general overview of the response of soil microorganisms to pesticides. It should be noted that Puglisi concluded that the usage of the recommended dosage and other good agricultural practices would help to minimize their detrimental effects on microorganisms and suggested further promoting policies focused on implementation of these practices. Researchers, as well as organizations such as European Food Safety Authority (EFSA), recently pointed out the need for new tests to better evaluate the impact of pesticides on soil microbial communities as well as overall soil quality [302,303].

Numerous studies focus on the effects of pesticides on specific groups of soil biota [76,93]. For instance, the effects of pesticides on earthworms have been reviewed recently [296]. Damage to DNA, changes in enzyme activity and altered membrane stability have been reported. For earthworms, the most harmful families of pesticides are carbamates, nicotinoids, organophosphates, strobilurins, sulfonylureas, and triazoles [296]. Earthworm, nematode, and protozoa populations may be reduced by application of herbicides [297]. Zaller et al. [299] cite half a dozen examples of major non-target impacts on flora and soil fauna from a range of common pesticides.

Microbial biomass in soils is a good measure of soil quality [297]. Lo has reviewed the literature on the effects of 21 pesticides on soil microorganisms, and concluded that it is difficult to predict organism responses from knowledge of pesticide chemistry [298]. Beneficial microbial communities are important in crop production but their effectiveness can be reduced by pesticide application [297–299]. Mycorrhizal fungi associated with roots are negatively affected by application of fungicides; in situ activity of enzymes such as dehydrogenases which are linked with microbial respiratory processes, may be affected by pesticide applications, and there may be interactive effects on soil biota when more than one pesticide is applied [299].

7. Remediation of Pesticides

The remediation of pesticides in soil and ground water is important because of public health, the need for healthy productive soils, and for drinking water that is safe to consume. There are several reviews of research on the remediation processes for pesticide contaminated soil [4,13,21,34,40,78,111,114,122,147,163,192,200,214,219,222,223,228,236,262,274–281,283,284]. The review on physicochemical remediation processes [275] is a very recent addition to the literature, and the review on bioaugmentation [277] was published in 2017. These two reviews have good content on the specific topics that are reviewed. Cycon et al. [277] have a table of physico-chemical properties of selected pesticides and a table of characteristics of the degradative potential of pesticide-degrading microorganisms for selected pesticides. Remediation using Fenton reagent has been identified as an effective physicochemical process by Baldissarelli et al. [275]. Rodrigo et al. [278] have provided a very good review on electrochemically assisted remediation of pesticides. Rodrigo et al. review the electro-Fenton process, in which the hydrogen peroxide is produced electrochemically in situ. One of the positive results of the oxidation of pesticides with the Fenton process is the completion of the oxidation such that there are few toxic intermediates in the processed product. The Fenton process is very appropriate for treating wastewater containing pesticides. It can be used to treat rinse water from pesticide containers and spraying equipment. Oturan and coworkers have reviewed the degradation of pesticides in water using the electro-Fenton process [281].

The present review will focus on biological remediation processes including phytoremediation. These processes are appropriate for application in all countries because they are cost-effective and relatively simple to implement. Bioremediation can take place under aerobic and anaerobic conditions [25,111,133,221]. The science of bioremediation is important for both in situ bioremediation and in applications where pesticide-contaminated soil is treated using composting, land farming or biopiles [34,111,264,277,279,283,284].

Eevers and coworkers have developed tables that report information on the microorganism, the pesticide that it degrades, the environment where the organism was found, and the published reference for bacteria and fungi [283]. Hussain et al. have developed tables for commonly used pesticides with listings of microorganisms that degrade the pesticide and the corresponding references where further information is available [111]. In addition there is a table with information on the genes and enzymes that are involved in detoxification of each pesticide [111].

Genetic engineering research has been conducted to develop microorganisms that are effective for the biodegradation of pesticides and pesticide residues [47,60,179,205,287]. Hussain and coworkers have reviewed research progress on this topic [111]. Recently, Huang et al. [285] have reviewed microbial degradation of pesticide residues by both natural and recombinant microorganisms. This paper includes a table with names of microorganisms and a listing of the pesticides that they degrade [285]. Liu et al. have reviewed recent progress with genetically engineered organisms that have been prepared to improve biodegradation of pesticides [286]. Table 1 in Reference 236 provides a stepwise process to develop genetic capability, which includes identification of the genetic ability, evaluation of the capability, modification to improve effectiveness, reevaluation of the improved microorganism, development of detection methods to evaluate effectiveness if needed, and application of the newly developed microorganism [286]. The advantages of recombinant methods in developing better microorganisms for biodegradation are reviewed, and a table listing important genes and host microorganisms for pesticide biodegradation is provided [286].

Bioaugmentation is known to be beneficial in many applications; however, there are a number of actions to consider that may be helpful. The moisture content, pH, temperature, organic matter content (nutrients), pesticide concentration, and inoculum concentration are important. The recommended quantity of cells for bioaugmentation is of the order of a million to a billion organisms per g of soil. In some cases repetition of bioaugmentation has been beneficial. Immobilization of cells on supports has been reported to improve the effectiveness of bioaugmentation [277]. The food and nutrition for the organisms is important. It needs to be considered because at low concentration the pesticide may not supply sufficient C or N to support growth. At high concentrations, the pesticide may be toxic. Cycon et al. [277] have reviewed research on bioaugmentation for pesticide-contaminated soil for a number of microorganisms and for a number of pesticides. They have prepared a table that includes the effective organism, the pesticide, inoculum size, and the references.

7.1. Composting

Composting of wastes is a simple process that can be implemented easily in all parts of the world. Yanez-Ocampo et al. have reviewed the composting of pesticides in soils and provide a listing of microorganisms that degrade selected pesticides [264]. One concern with composting of pesticides is the extent of degradation of the pesticide during composting [32,34,158,216,243,264]. The toxicity of the product after composting should be evaluated. When pesticides in soil are composted by adding manure and other wastes to the soil, there is some beneficial dilution. It is important to add microorganisms that can biodegrade the pesticide. Soil from a field where the pesticide has been used regularly may have effective microorganisms. Leaves, manure, yard trimmings, straw, corn stalks, food wastes, and biosolids from wastewater treatment are examples of materials to add to a composting process to provide food for the organisms. Complete mineralization of pesticides should not be expected in composting of pesticides [32,35,216]. Reports on composting in biobeds in Europe [80,81,121] and Africa [92] are available.

7.2. Land Farming

Land farming of soils containing pesticides involves spreading the solids over a land area that is designed for bioremediation of the pesticide [34,103]. Land farming of pesticides has been reported in Mali [98,99] under conditions where plants were established to enhance the remediation. The spreading of the contaminants over a large area dilutes the pesticides. Biodegradation is the desired result, but there may also be loss by volatilization. This method is appropriate for pesticides that are well known to be biodegradable. It should not be used for persistent pesticides.

7.3. In Situ Bioremediation and Phytoremediation

There has been significant progress in developing in situ bioremediation for pesticides [25,86–88,92,111,112,134,151,154,159,162,180–182,185,188,202,204,206,244,277,283,284,286]. These methods fall into three general categories; phytoremediation, bioremediation by indigenous organisms and bioaugmentation. Vegetation has been beneficial in the bioremediation of pesticides in many locations for many different pesticides [1,22,26,38,44,45,50,54,79–81,83,85,111,122,147,148,152,171,175,176,180,221,244,253,283,284]. With vegetation present in the contaminated soil, degradation of the pesticide near the roots is supported by substrates coming from the roots that enhance microbial growth and the magnitude of the active microbial population [22,187,207]. Bacteria and fungi in the root zone provide beneficial genes and play important roles in the biodegradation of pesticides [206,218]. Some pesticides are taken up into the plant and transformed by plant enzymes. The octanol/water partition coefficient of the pesticide is important in phytoremediation because it affects the solubility of the pesticide in water, the uptake into plants, and the distribution of the pesticide in the soil and in the plant [13,124,180,193,256]; see Paschke et al. for values of the octanol/water partition coefficient for pesticides [291]. Biodegradation of pesticides by endophytic bacteria associated with plants is important in many biodegradation processes. Listings of plants that take up pesticides and the pesticides that have been reported to be transformed by plant enzymes are available [111,152,283]. The process of establishing vegetation and nurturing growth is inexpensive compared to many of the other remediation processes. It can be applied at field scale [88,113,160,164,165,184,210]. The vegetation reduces wind-blown dust and the movement of the pesticides from the site [156]. Nurzhanova et al. [169,173,174] have provided a list of 17 pesticide-tolerant plants that grew in soils where high pesticide concentrations were present. The growth of plants in contaminated field soil often improves soil quality by increasing microbial populations and organism diversity, and by adding organic matter to the soil [10,26,53,189,190]. The common results after phytoremediation of contaminated soil are that pesticide concentrations in soil are reduced and pesticide concentrations in roots are less than the concentrations in the soil, but larger than the concentrations in the stems and leaves of the plants [4,156].

There have been several efforts to develop transgenic plants for phytoremediation of pesticide-contaminated soils [284]. The process that plants use may involve 1) addition of a reactive group such as OH to the pesticide, 2) conjugation of another compound to the pesticide through reaction with the reactive group, and 3) integration of the conjugated pesticide into cell-wall components or another part of the plant where the new substance is less toxic [236,283,284]. Atrazine degradation with transgenic plants has been reviewed by Dhankher et al. [284].

Endophytes with biodegradation ability can be added to the site to assist the plant with phytoremediation [284,288]. Genetic modification of endophytic bacteria is one pathway to develop the successful pesticide degradation by a plant, aided by bacteria associated with the plant. Endophytic bacteria may survive better than soil bacteria in field applications [283,288]. In phytoremediation, biodegradation in the rhizosphere and the plant are both important.

7.3.1. Biodegradation of DDT

The insecticide 1,1,1-trichloro-2,2-bis(p-chlorophenyl) ethane (DDT) is known to be persistent in the environment, and its use is banned in many locations [289]. However, it is still in use in Africa and India. It has very low solubility in water, about 0.001 mg/L and is more soluble in lipids [289,290]. The log of the octanol/water partition coefficient is 7.48 [292]. Eevers et al. have reported that DDT is taken up and transformed by several different plants [283]. Zucchini and pumpkin are two garden plants that take up DDT [254–256]. DDT can be removed from soil and transported into alfalfa, rye grass and tall fescue also [139]. Pan et al. [289] have reported their research to isolate a bacterium that biodegrades DDT and uses DDT as a carbon and energy source. The mineralization pathway is reported [289]. Nano-scale zero valent iron has been found to enhance the biodegradation rate of DDT [290]. Adding these nano particles several times resulted in biodegradation of the DDT [290] and further biodegradation of some of the initial degradation products. Spent mushroom waste (with white-rot fungus), and biosolids have been reported to be effective for degradation of DDT [137,198]. There are many plants that take up DDT and its metabolites and are beneficial in the remediation of DDT-contaminated soil [1,44,79,139,155,254–256,274,283].

Recently, Nurzhanova et al. [172] have reported their work on phytoremediation of soil containing DDT and metabolites of DDT. Table 1 contains data from their research [172]. In the table, DDE refers to 1,1-dichloro-2,2-bis(p-chlorophenyl) ethylene, and DDD refers to 1,1-dichloro-2,2-bis(p-chlorophenyl) ethane. When both chlorines on the benzene rings are in the para positions, 4 is used to show this. The two indicates that one of the chlorines is in the ortho position. MAC refers to the maximum allowable concentration, which is 0.1 mg/kg or 100 µg/kg. In this soil, the average total concentration of DDT and the metabolites was 6.181 mg/kg, which is 62 times the MAC.

Table 1. The concentration of DDT (1,1,1-trichloro-2,2-bis(p-chlorophenyl) ethane) and metabolites in soil and miscanthus after phytoremediation during one growing season *.

Samples	Soil or Vegetation	Mass. kg	Concentration. µg kg^{-1}			Total Extraction µg
			4,4′-DDE	2,4′-DDD	4,4′-DDT	
M. giganteus						
2MAC	Soil	3	146 ± 22	3 ± 1	92 ± 21	723
	Above ground	0.031	0.2 ± 0.1	18.3 ± 6.1	98.4 ± 27.1	3.6
	Root	0.035	35.2 ± 6.5	352.3 ± 32.2	1810.4 ± 126.3	76.9
	Soil	3	105 ± 18	2 ± 1	83 ± 12	572
Dying sprouts of *M. giganteus*						
6 MAC	Above ground	0.015	50.2 ± 14.2	15.3 ± 6.2	36.3 ± 12.5	1.5
	Root	0.026	158.1 ± 87.5	134.3 ± 25.1	139.8 ± 23.6	11.2
13 MAC	Above ground	0.010	41.4 ± 21.2	17.3 ± 11.5	36.7 ± 15.7	0.9
	Root	0.022	246.3 ± 123.3	135.5 ± 27.1	191.1 ± 55.1	8.8
33 MAC	Above ground	0.008	22.2 ± 9.2	36.1 ± 18.2	32.0 ± 16.0	7.2
	Root	0.020	267.4 ± 42.1	60.4 ± 19.1	212.6 ± 27.1	10.8
45 MAC	Above ground	0.007	36.1 ± 27.2	112.3 ± 20.5	27.9 ± 9.9	1.2
	Root	0.021	453.3 ± 30.1	113.4 ± 15.2	52.1 ± 11.1	13.0
62 MAC	Above ground	Died				
	Root					
M. sinensis						
62 MAC	Soil	3	2750 ± 88	933 ± 48	2498 ± 45	18,543
	Above ground	0.014	151.2 ± 45.3	78.4 ± 29.3	12.4 ± 7.0	3.4
	Root	0.009	570.5 ± 53.3	45.1 ± 22.0	247.2 ± 76.4	7.8
	Soil	3	1230 ± 49	888 ± 79	1991 ± 221	12,327

* MAC—maximum acceptable concentrations of metabolites of DDT in the soil 100 µg kg^{-1} from reference [172].

The soil sample is from a contaminated field site in Kazakhstan that also had metal contamination [172]. The toxicity of the pesticide and the metabolites had a greater impact on *Miscanthus x giganteus*, which is a triploid hybrid, compared to the diploid *Miscanthus sinensis*. For *M.x giganteus* the concentrations of DDT and DDE in the roots are greater than those in the aboveground biomass for all cases; however the concentration of DDD is larger in the aboveground biomass than in the root for the *M. sinensis*. The average height of the plants was 137.6 cm for the *M.x giganteus* for 2MAC and 75.3 cm for *M. sinensis* for 62MAC after one growing season [172]. The values for mass include any water that was present. The mass of DDT taken up by the plant was largest for the 2MAC experiment with *M.x giganteus*. The concentration of DDT in the roots is higher than the average value in the blended soil for 2MAC. When one blends clean soil and contaminated soil, one would expect to have some soil particles with much higher concentrations of DDT than the average value of 0.2 mg/kg. In Table 1, for each experiment, the top soil sample data has values before the vegetation experiment and the lower soil sample data has values after the growing season. The notation 2MAC refers to two times the MAC or 2×0.1 mg/kg = 0.2 mg/kg.

7.3.2. Plant Selection

Plants that are used for phytoremediation should be appropriate for the environmental conditions where they are planted. They should be plants that grow well locally under normal rainfall and temperature for the area. Plants that have a history of successful application in pesticide-contaminated soil should be considered for phytoremediation projects; see the listings of plants used previously for phytoremediation [111,152,256,283]. The toxicity of the pesticides in the soil to the plants may be important [54].

The cost of the plants and their establishment as well as the value of any products associated with the growth of the plants may affect the economics of the project [54,57,104]. Pidlisnyuk and coworkers have established miscanthus plots for phytoremediation applications with the goal of having an annual crop to harvest and use [83,172,189].

7.3.3. Soil Amendments

Many beneficial amendments have been added to pesticide-contaminated soil that have increased biodegradation rates and extent while also reducing toxicity. Manure, biosolids, and compost provide nutrients and organisms with enzymes that may be beneficial for biodegradation in vegetated soil [189]. Nutrients and carbon and energy sources for microbial populations should be included in the plan. Bioaugmentation with microbial populations that have enzymes to degrade the pesticide of interest is helpful [277]. Arbuscular mycorrhizal fungi have been reported to be beneficial in phytoremediation because they have beneficial enzymes, and they stimulate microbial activity in the rhizosphere [12,85]. Substances that are not toxic and enhance solubility in the aqueous phase are beneficial because of the low water solubility of many pesticides. Since one goal of phytoremediation is to have land that can be used for its intended purposes, amendments that improve the land should be given a higher priority [29,59,100]. The pH that is desired for the phytoremediation and future use of the land should be included in developing the amendment plan.

The addition of enzymes with the capability to serve as catalysts for biodegradation of the pesticide of interest may be beneficial [87,111,201,227]. It is important for the pesticide and enzyme to find each other in the multiphase mix that includes soil, plant matter, and amendments. The water phase is a good place for this to happen if the pesticide has significant water solubility.

Water is important for phytoremediation and for agricultural productivity. In climates where low rainfall may be limiting productivity, soil amendments that improve the water-holding capacity for phytoremediation and future land use are expected to be beneficial. Adding organic carbon and/or biochar may improve water holding capacity, microbial numbers and microbial diversity [10,15,20,23,27–29,37,59,97,100,128,130,191,225,226,251]. Adsorption to the carbon should be expected, which makes the pesticides less available [35,217]. The population numbers and diversity of organisms present may be used to evaluate the quality of the soil [59,100]. Activated carbon additions to soil enhance adsorption and increase the fraction of a pesticide that is adsorbed. This reduces pesticide bioavailability [27,35,37,43,64,76,89,166,178,183,211,217,224,233,237,249,265–268], which is beneficial when there are pesticide residues from past applications. The addition of biochar and/or activated carbon may reduce nutrient and trace metal availability [166,266].

7.3.4. Advantages and Disadvantages of Bioremediation and Phytoremediation

In situ bioremediation and phytoremediation make use of natural processes and are appropriate for application in many locations where soil is contaminated with pesticides. One important issue is the time required for remediation because biological processes are slow compared to excavation and hauling to a landfill for hazardous substances or to an incinerator. It is well known that bioremediation and phytoremediation are less expensive than many alternative remediation processes such as soil removal followed by incineration or chemical oxidation [72,74,110,136,214]. The concentration of the pesticide in the soil may be an issue because biological processes are suitable for conditions where the pesticide is not toxic to the plants and microorganisms that are to be used for remediation. Yet, if it is needed as a C or N source for the micro-organism the concentration must be high enough to fulfill that need. Otherwise, co-metabolism with an additional substrate, perhaps supplied by the plant, will be more effective. Pesticide contamination that is at or near the soil surface is more appropriate for biological processes. Phytoremediation is generally most effective in the root zone of the vegetation [136,214]. There is research progress in developing new genetic capability for biodegradation and phytoremediation [40,136,148,277,289].

8. Implementing and Monitoring Phytoremediation

There are some inexpensive approaches to implementing and monitoring phytoremediation. The initial starting point may be pesticide-contaminated soil where there is knowledge of a spill or where plant growth is poor or plants do not survive because of pesticides. Soil amendments such as compost, manure, and plant residues (such as leaves) can be added to the contaminated soil to add microorganisms, nutrients, and organic carbon to obtain an amended soil with about 5% to 10% amendments. One can blend this amended soil into clean productive soil to obtain a 50/50 soil and a 10/90 soil with 10% of the contaminated soil to reduce toxicity. The different soils can be placed in pots and plants can be transplanted so there is one plant in each pot. The experiment may include (1) The original polluted soil, (2) The original soil with amendments, (3) The 50/50 soil with amendments, and (4) The 10/90 soil with 10% polluted soil with amendments. With three replicates of each soil mixture, there would be 12 pots with one plant transplanted into each pot. These pots can be watered regularly and managed in an environment with good natural light and appropriate temperature.

The fate of the vegetation can be monitored with respect to plant survival and plant growth. The height and mass of the plants can be measured and recorded. There is value in measuring the concentrations of nitrogen, phosphorus, organic carbon, and microbial numbers in the original polluted soil, the clean soil, and the soils with amendments. The concentration of the pesticide should be measured in the original contaminated soil and after the plant growth experiments. Since nematodes, arthropods, and earthworms are measures of soil health, experiments with soil fauna may be beneficial to understanding the fate of these organisms in pesticide contaminated soil.

Once the initial experiments are finished, one may wish to begin the field experiment; however, the results may also suggest that further experimentation is needed. For example, experiments could be carried out with several different plants to try to find a plant that grows well in the polluted soil with amendments and without soil dilution.

9. Conclusions

In many countries significant land area is contaminated with pesticides and/or contains pesticide residues that are of concern. Soil quality is very important for agricultural production, and improving productivity is a high priority because of the need for food, feed, and other products. The biodegradation of pesticides in soil is advancing at many locations. New genetic developments will facilitate this through use of engineered organisms where they are permitted.

Earlier reviews have been cited in this work and recent developments noted. Research on biological remediation methods for pesticides in soil and water continues to be an important research thrust in many countries. Soil amendments that provide genetic diversity and food sources for microbial populations are beneficial. Endophytes with the ability to degrade pesticides are being developed and applied with plants that take up and transform pesticides.

Phytoremediation technologies are very appropriate for application to soils containing pesticides in many countries. New initiatives to develop plants for phytoremediation that can be harvested and used beneficially are receiving research attention. The knowledge and equipment that are needed to implement phytoremediation at field sites are available in many parts of the world.

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