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Phytoremediation and Detoxification of Two Organophosphorous Pesticides Residues in Riyadh Area

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Abstract: A study was undertaken to detect and monitor the degradation of two organophosphorous pesticide residues (dimethoate and malathion) by four plants namely Amaranth, *Amaranthus caudate*, Lettuce, *Lactuca sativa*, Water cress, *Nasturtium officinale* and Kidney bean, *Phaseolus vulgaris* and microbial degradation based on the recommended rates for insects control in Kingdom of Saudi Arabia. Pesticides residues were quantified by gas chromatography mass spectrometry (GC-MS) analysis. The ability of these biological agents to detoxify and clean up these pesticide residues in both soil and plants was studied. In soil samples, the highest percentages of malathion-degradation were ≥60 and ≥90% after 28 and 42 days, respectively. The half-life times of malathion were accounted by 24, 23, 25 and 25 days for Amaranth, Kidney bean, Lettuce and Watercress, respectively. For dimethoate, results of time course studies in soil degradation revealed that Amaranth and Kidney bean were the most effective plants in dimethoate degradation, whereas ≥90% was degraded after 42 days. The half-life times of dimethoate were accounted by 28, 30, 25 and 30 days for Amaranth, Kidney bean, Lettuce and Watercress, respectively. In plant tissues, only ≤5 and ≤3% of malathion and dimethoate still in plant tissue, respectively. These results suggest that phytoremediation ability of plants could accelerate the degradation of malathion and dimethoate residues in plant tissues. Random amplified polymorphic DNA-polymerase chain reaction (RAPD-PCR) patterns of plants´ DNA exhibited sever mutagenic effects on plants´ DNA. Malathion caused extreme effects on plants´ DNA. These mutagenic effects supported the plants phtoremediation ability besides keeping the plants healthy. Moreover, efficiency of a *Pseudomonas frederiksbergensis*, isolated from soil sample, for degradation of malathion and dimethoate was also investigated. This isolate could degrade 100% of malathion and dimethoate after 3 days. These rhizosphere-associated bacterial could support plants in the rhizospheric region to increase their ability to degrade these pesticides. The results indicated that these biological agents works together and may be considered as potential candidates for the detoxification and degradation of these organophosphorus compounds in contaminated soil in Riyadh area, Saudi Arabia.

Key words: Degradation . Dimethoate . Malathion . Microorganisms . Organophosphorous . Plants . Phytoremediation . *Pseudomonas frederiksbergensis*

INTRODUCTION

Millions of tons of pesticides applied annually are used in modern agriculture to increase production through controlling harmful effects caused by the targets organisms including insects, fungi, bacteria, viruses as well as grasses grown in between the economical crops [1]. However, less than 5% of these products are estimated to reach the target organisms. One of the most important problems with the use of pesticides is their possible persistence in the environment and therefore, their possible incorporation into the food chain affects ecosystem and all human beings [1]. Therefore, these toxic compounds have been implicated in various disorders and diseases including cancer, adverse reproductive outcomes, peripheral neuropathies, neurobehavioral disorders, impaired immune functions and allergic sensitization reactions, particularly of the skin, cumulative inhibition of cholinesterase activity because of long-term low doses of exposure [2]. Synthetic Organophosphorus Pesticides (OPs) are the most widely used pesticides and unacceptable levels of environmental residues of these compounds have been found in many countries worldwide. Although most OPs compounds are not persistent, they still cause broad area pollution from

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continued use in agriculture and public health [3]. These pesticides are frequently not used as recommended dosage, leading to an accumulation in the environment [4].

Furthermore, many of these compounds are mobile in the environment, which may lead to diffuse contamination, leading to an accidental release or disposal that caused heavy pollution in certain areas. High concentrations of the OPs as malathion and dimethoate are degraded in soil to acceptable levels only after many years [5]. These pesticides are acetylcholinesterase (AChE) inhibitors, mostly are liposoluble and pose a hazard to humans. The loss of AChE results in acetylcholine accumulation, which interferes with muscular responses and produces serious symptoms in vital organs, eventually leading to death [6].

Therefore, it is essential to decontaminate OPs in the environment. Although previous studies have focused on persistent of the organic pollutants, little attention has been paid to the remediation of OPs contamination. The metabolic fate of pesticides is dependent on abiotic environmental conditions, microbial community, plant species (or both), pesticide characteristics and biological and chemical reactions. Abiotic degradation is due to chemical and physical transformations of the pesticide by processes such as photolysis, hydrolysis, oxidation, reduction and rearrangements [7]. Furthermore, pesticides may be biologically unavailable because of compartmentalization, which occurs as a result of pesticide adsorption on soil colloids without altering the chemical structure of the original molecule. The simultaneous cleanup of multiple, mixed contaminants using conventional chemical and thermal methods is both technically difficult and expensive; these methods also destroy the biotic component of soils. Alternative approach, phytoremediation, has been considered as a potentially low cost, very effective and an environmentally friendly method to detoxify these compounds. It may provide a better option for dealing with such diffuse contamination [8].

Phytoremediation is an eco friendly approach for remediation of contaminated soil and water using plants. It takes advantage of the unique and selective uptake capabilities of plant root systems, together with the translocation, bioaccumulation and contaminant storage/degradation abilities of the entire plant body. Plant-based soil remediation systems can be viewed as biological, solar-driven, pump-and-treat systems with an extensive, self extending uptake network (the root system) that enhances the below-ground ecosystem for subsequent productive use [9].

This technology is comprised of two components, one by the root colonizing microbes and the other by plants themselves, which degrade the toxic compounds to further non-toxic metabolites [10]. Plants have a natural ability to metabolize OPs, as reported in a previous phytoremediation study of the use of the hydrophyte *Typha latifolia* to remove malathion and dimethoate pollutants from soil. *Pterocarpus indicus* and *Jatropha curcas* were able to remediate hexavalent chromium polluted soil of less than 90 mg/kg [11]. Plants also prevent contamination from spreading to other areas via wind, rain or groundwater an advantage of using phytoremediation to address diffuse OPs pollution at scattered sites [12].

In Saudi Arabia, the rapid development of agriculture necessitated the use of a wide number of pesticides on agricultural field. The presence of pesticides in soils constitutes pervasive problems for public health and therefore, there is a growing concern for reducing the pesticides contamination in the environment. Meanwhile, this study was conducted to evaluate four plants, Amaranth, *Amaranthus caudate*, Lettuce, *Lactuca sativa*, Watercress, *Nasturtium officinale* and Kidney bean, *Phaseolus vulgaris* L. and microbial activity in rhizospheric region for the detoxification and phytoremediation of malathion and dimethoate residues from soil in Riyadh area, Saudi Arabia.

MATERIALS AND METHODS

Soil sample preparation: Laboratory bench-scale soil washing experiments were performed to clean the contaminated soil. The soil samples were collected from 15-30 cm depth of Riyadh agricultural area. Samples were supplied with air dried at 35°C and ground to <2mm. The soil samples were washed by using 0.1 M HCl and 0.05 M sulfuric acid, where 70 L of distilled water was mixed together with the acid and 100 kg soil sample. The whole mixture was shacked for 6-7 hrs continuously. Afterwards, the soil sample was washed again with the ordinary water for 3 hrs. After 24 hrs, the soil sample was washed with 6% H_2O_2 for 6 hrs before washing again with distilled water, then left for 24 hrs. Samples were air dried by exposing to the sunlight directly for 12 hrs. Visible insects and pests were removed from the soil.

Soil characteristics: Soil samples were collected from agricultural area, south of Riyadh city. The soil type and chemical analysis were carried out according to the method [13].

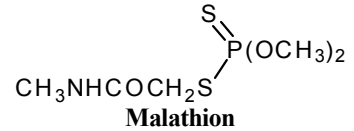
Plant materials: Amaranth, *Amaranthus caudate*, Lettuce, Lactuca sativa, Water cress, *Nasturtium*

Table 1: Standard curves of dimethoate and malathion concentrations

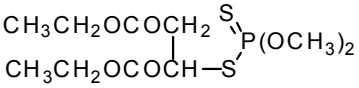
Pesticides	Calibration curve equation	Regression coefficient (r^2)	Concentrations ppm
Dimethoate	y = 0.106x-0.028	0.98	423.109
Malathion	y = 0.267x-0.00146	0.97	259.329

officinale and Kidney bean, *Phaseolus vulgaris* L. were selected to examine their phytoremediation ability. The plants were chosen because of their short life cycle and their phytoremediated background, which makes them suitable for this investigation, besides they are edible. The seeds source was obtained from College of Food and Agricultural Sciences, King Saud University. The seeds were healthy, vigorous, plumy, well matured and free from mixture of other crop seeds and extraneous materials. Seeds were sterilized in 2% hypochlorite solution for 15 min. After being thoroughly washed, they were kept between two pads of moistened cotton, for 3 days. The experiment was conducted by growing the plants in pots under green house conditions at the experimental site in Department of Botany and Microbiology, King Saud University, Saudi Arabia during November, 2008 to January, 2009. All the pots were watered daily during the experiment and were randomized on the greenhouse bench. Untreated checks were non cultivated soil with dimethoate and/or malathion.

Pesticides and application: The organophosphate insecticides used in this study were: dimethoate (Cekuthoate®, 95%) (dimethyl S-(N-methylcarbamoylmethyl) phosphorothiolothionate) and malathion (Carbophos®, 97%) (diethyl dimethoxythiophosphorylthio) succinate. They were purchased El-Raghy Agricultural Company Ltd., Kingdom of Saudi Arabia. Samples were prepared in deionised water using ethylacetate. All pesticides and reagents were high purity and analytical grade. Methanol and acetonitrile HPLC grade were purchased from Merck (Darmstadt, Germany). Calibration curve for each selected pesticide concentrations were prepared and then quantified by GC-MS (HP G1800A system, Capillary column HP5 (30 m x 0.25 mm) Hewlett-Packard, Germany). Regression coefficient (r^2) obtained from the plotted data was calculated (Table 1).



Malathion



Dimethoate

The precision of the method, determined from 3 replicate preparations from the same extracted sample were analyzed. Appropriate volumes were mixed with the soil and bedding materials to give 2000 ppm of dimethoate. The recommended concentration of malathion is 2500 ppm [14]. The concentration of each insecticide was chosen based on the recommended rates for insects control in Kingdom of Saudi Arabia. Dimethoate and malathion were applied by mixing with water. The emulsion within the spray tank was shaken well and sprayed covering the soil. Uniform mixing in soil was achieved by spreading the soil on a plastic sheet and spraying it with pesticide solution followed by thorough mixing. The uniform mixing of pesticide was checked by removing random samples and analyzing for pesticide residues. The analysis of pesticide residues were carried out at the College of Sciences, King Saud University.

Determination of pesticides residues in soil: Three replicates from each treated soil were taken around each plant; afterwards they were mixed homogeneously and subjected to analysis. Extraction of the pesticides and quantified by GC-MS analysis of dimethoate and malarhion was adapted [15] with some modifications. The procedures were based on a wet methanol and acetonitrile extraction of soil extraction. Standard stock solutions of the target pesticides were prepared in methanol at a concentration of 5 mg/ml. GC-MS conditions were: initial temperature, 80°C; temperature increased initially by 15 min^{-1} up to 280°C; injector port temperature, 250°C; detector temperature, 250°C. Retention time for dimethoate was 20.5 min. Hexane was used as an internal standard in this respect.

Determination of pesticides residues in plants: Twenty-five grams of the whole plant were chopped in small pieces and homogenized with acetonitrile in a blender. Hundred ml of acetonitrile was added and blended for 2-3 min at moderate to high speed. The homogenate was filtered by passing through glass wool and transferred the filtrate to a jar and extracted with an additional 100 ml acetonitrile. The mixture was filtered as before and combined the filtrates. The pesticide residues were quantified and analyzed as previously mentioned.

Plant DNA isolation and random amplified polymorphic DNA-polymerase chain reaction

(RAPD-PCR): Plant genomic DNA was isolated from leaves according to the method [16]. The two primers used in this study were, primer 1 (5'-GGTGCGGGAA) and primer 2 (5'-CCCGTCAGCA) were purchased from Amershambioscience. Amplification reaction solutions were prepared in a final volume of 50 µl containing 10 mM Tris-HCl, pH 8.3, 1.5 mM $MgCl_2$, 50 mM KCl and 100 M each of dATP, dGTP, dCTP and dTTP (Boehringer Mannheim), 2.5 M primer, 1.25 units of Taq DNA Polymerase (Boehringer Mannheim) and approximately 50 ng of DNA. The amplification was performed in an MJ Research programmable thermal cycler (PCR Thermocycle: Elmar Cetus 420, Elmar Cetus USA) where the program was as follows: universal denaturation cycle (5 min at 94°C), 45 cycles of annealing/extension reactions (1 min at 94°C, 1 min at an optimum annealing temperature 36°C for each used universal primer and 2 min at 72°C) and cycle of final extension step (5 min at 72°C) was followed by soaking at 4°C.

Isolation and identification of malathion and dimethoate degrading bacterium: The soil extraction and enrichment of the microorganisms in mineral salt medium (MSM) was prepared [17]. For isolation of the pure culture, the soil extract was serially diluted and by using streak method technique, 250 µl was taken from the soil extract with appropriate dilutions, then were spread on standard agar plates (Nutrient Agar, Merck, Germany) and incubated for 24 hr at 37°C. After overnight incubation, the single colonies were picked and cultivated again on malathion and\or dimethoate MSM. These procedures were repeated until identical colonies in shape and morphological characteristics were observed. The isolation and identification of the isolate were carried out in accordance with Bergey's Manual of determinative Bacteriology [18, 19 and 20].

Sorption: Adsorption of each pesticide on soil particles was determined by the method [20].

RESULTS AND DISCUSSION

Degradation of dimethoate and malathion in soil and plants: In soil sample, malathion degradation increased with increasing of the cultivated time. The highest percentage of malathion-degradation in soil samples were ≥60% and ≥90% after 28 and 42 days, respectively. Except for Kidney bean, all evaluated plants could degrade ≥80% of malathion. The half-life times of malathion were estimated by 24, 23, 25 days and 25 for Amaranth, Kidney bean, Lettuce and Water cress, respectively. For dimethoate, results of time course studies in soil degradation revealed that Amaranth and Kidney bean were the most effective plants in dimethoate degradation, where ≥90% was degraded after 35 days. The half-life times of dimethoate were estimated by 28, 30, 25 and 30 days for Amaranth, kidney bean, Lettuce and Watercress, respectively (Fig. 1).

In plant tissues, for malathion, 10, 125, 110, 68 ppm were found in Amaranth, Kidney bean, Watercress and Lettuce, after 42 days, respectively. For dimethoate, 10, 59, 60, 65 ppm were found in Amaranth, Kidney bean, Watercress and Lettuce, after 42 days, respectively. Therefore, only ≤5 and ≤3% of malathion and dimethoate still in plant tissue, respectively (Fig. 2). In fact, Organophosphorus compounds are a group of highly toxic agricultural chemicals widely used for plant protection. Up to the present these pesticides such as dimethoate and malathion, are still extensively used world wide despite their high toxicity [22]. Due to environmental concerns associated with the accumulation of these pesticides in food products and water supplies, there is a great need to develop safe, convenient and economically feasible methods for pesticide detoxification in environment. Many studies have focused on the phytoremediation of persistent pesticides, but few have been conducted for OPs compounds [23].

Isolation of *Pseudomonas frdedriksbergensis* from cultivated soil: *Pseudomonas frdedriksbergensis* was isolated on mineral salts medium (MSM) with dimethoate as a carbon and energy source. On the basis of morphological, cultural and biochemical characteristics, the culture was found to belong to the genus Pseudomonas, according to the Bergey's Manual of determinative Bacteriology [20]. The bacteria are highly oxidative, aerobic and metabolically versatile and have been reported to degrade aromatic hydrocarbons, oil, petroleum products and pesticides. This Gram negative, rod shaped bacterium was 0.5-0.8 µm in diameter and 1.5-3.0 µm long (Fig. 3).

Degradation of dimethoate and malathion *Pseudomonas frdedriksbergensis*: *Pseudomonas frederiksbergensis* showed high efficiency and rapid rate of mineralization to assimilate malathion and dimethoate. When the medium pH was 7.0, the compounds were totally degraded after 6 days (Fig. 4). The half lives of malathion and dimethoate accounted by 3 and 2.3 days, respectively. When the medium pH was 8.0, results of GC-MS revealed that biodegradation by *Pseudomonas frederiksbergensis* began on the first day and degraded totally after 3 days. The half-lives of malathion and dimethoate were 10 and 38 hrs, respectively.

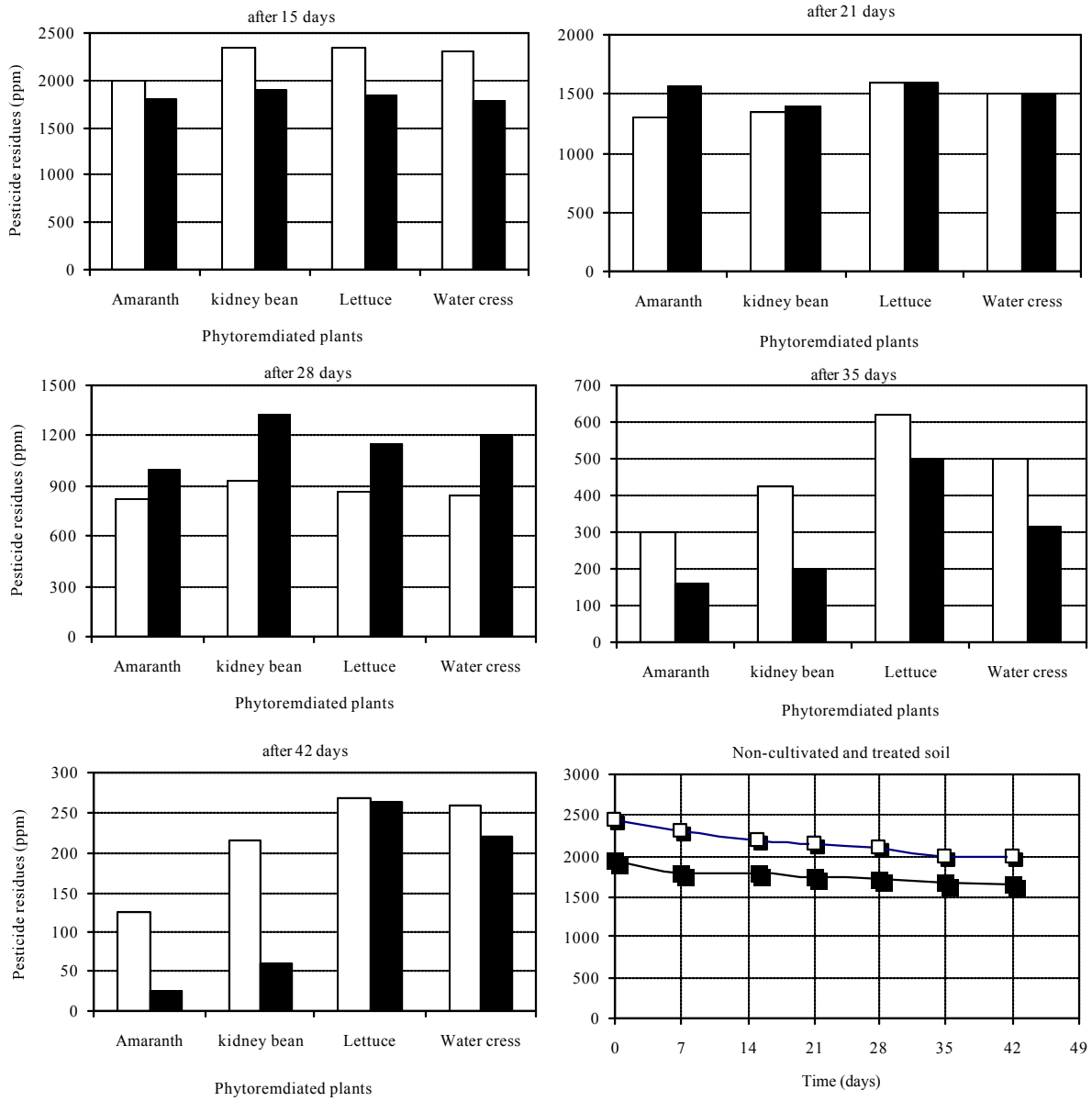


Fig. 1: Degradation of dimethoate and malathion in soil

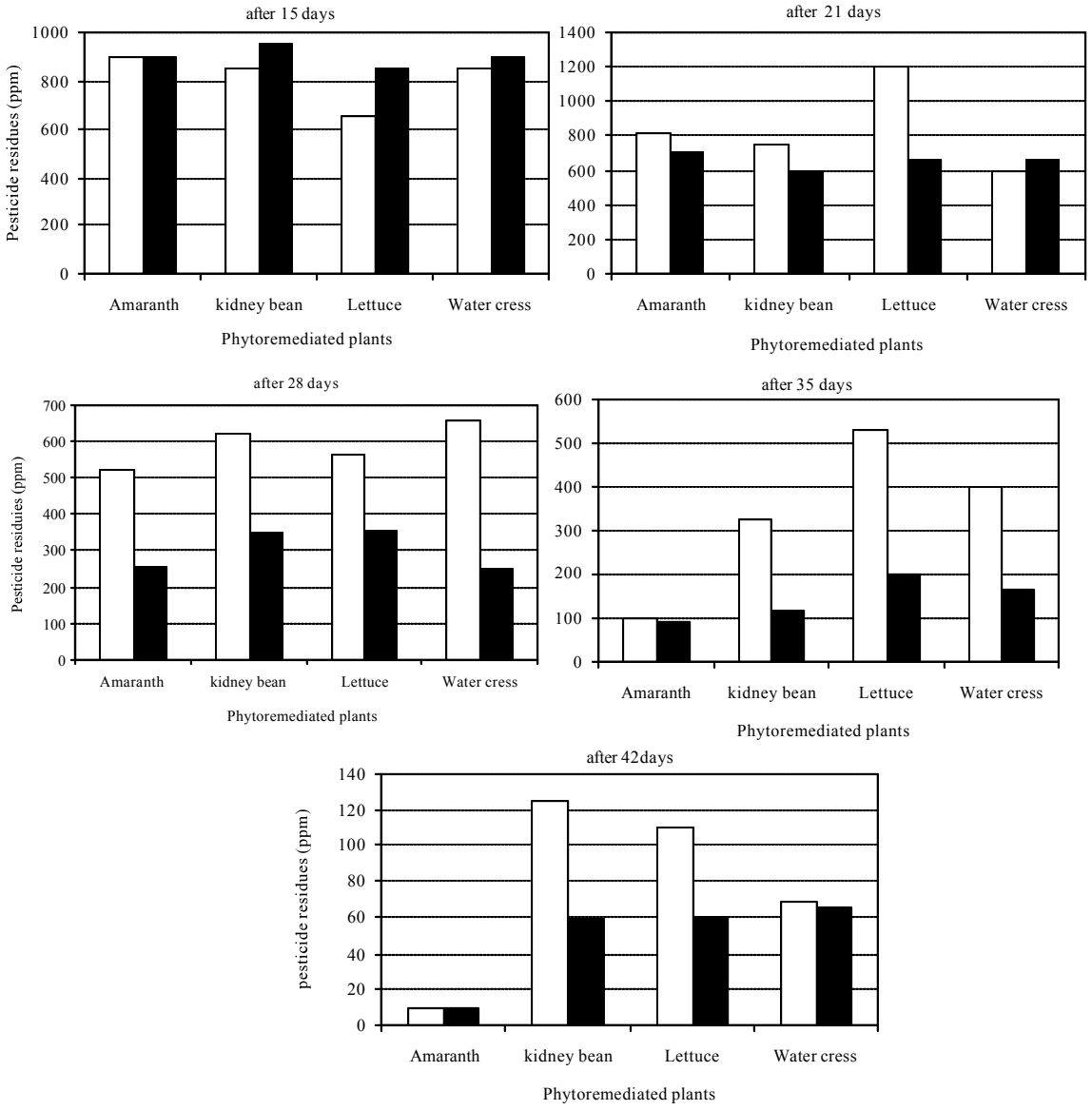
On the other hand, microscopic examination exhibited the ability of these bacteria in assimilating Malathion and dimethoate (Fig. 5 and 6). Formation malaoxon was detected in an incubated sample by GC-MS (Fig. 7&8).

In fact, the degradation rate of dimethoate and malathion is strongly related to medium pH together with the abiotic hydrolysis. In our results, the dimethoate and malathion-degrading bacterium exhibited a complete degradation in 3 days. Soil pH has also been shown to be important for maintaining the enhanced degrading capability of soil for prolonged periods (Table 2). These results are similar to the findings [24] that found that the fluctuation of pH had a big influence on chlorpyrifos degradation in soil. The degradation of malathion and dimethoate are due to broad substrate spectrum of *Pseudomonas frederiksbergensis* specificity. Our results demonstrate that the microorganism is olated based on its ability to utilize and mineralize these organophosphorous compounds was quantitatively most efficient in utilizing these compounds as a sole carbon source. The results support the presence of carboxylesterases. This shows the importance of microbial action in the degradation of these pesticides [25].

Table 2: Soil type, chemical and mechanical analysis

Particles size (%)						**meq^{-1}					
Sand	Silt	Clay	Texture class	pH	*EC (ds m^{-1})	$CaCo_{3\ (\%)}$	Ca^{+2}	Mg^{+2}	Cl^{-}	K^{+}	Na^{+}
71.65	16.05	12.3	Sandy Loam	8.2	63	46.9	188.0	325.8	985.36	33.56	35.58

*Electric conductivity, **meq^{-1} Millie equivalents per liter



■ Dimethoate concentrations (ppm), □ Malathion concentrations (ppm)

Fig. 2: Degradation of dimethoate and malathion in plants

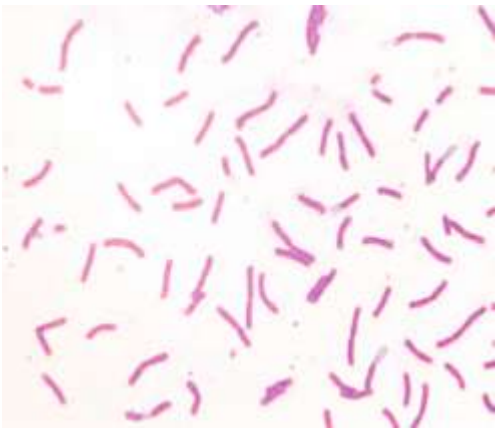


Fig. 3: Photomicrograph of *Pesudomonas fredersriksbergensis* grown in malathion MSM after 3 days

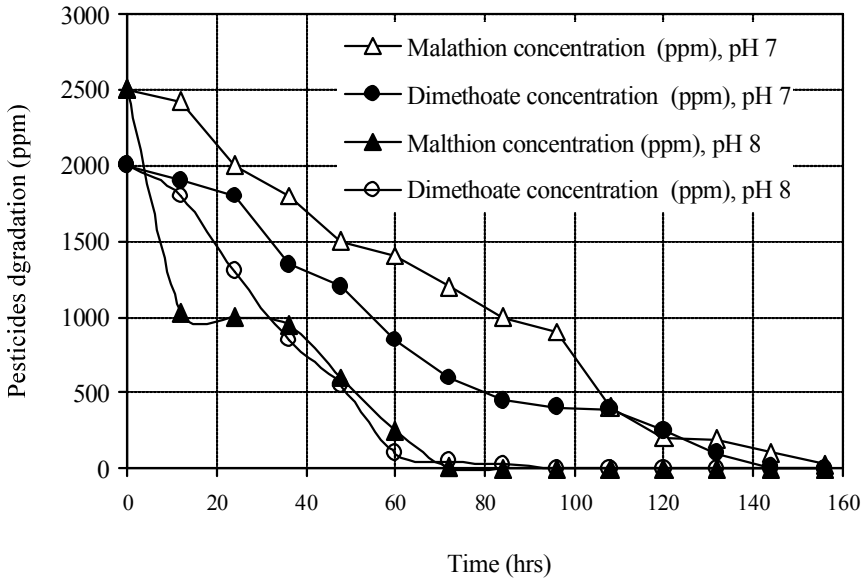


Fig. 4: Time course of malathion and dimethoate biodegradation and by *Pseudomonas frederiksbergensis*

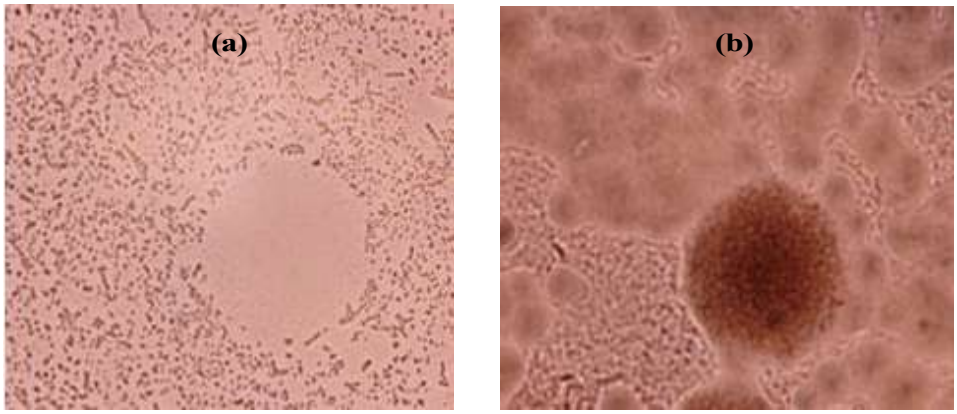


Fig. 5: Fluorescent photomicrograph of *Pseudomonas frederiksbergensis* growth after 3 days
(a): mimethoate MSM, (b): malathion MSA

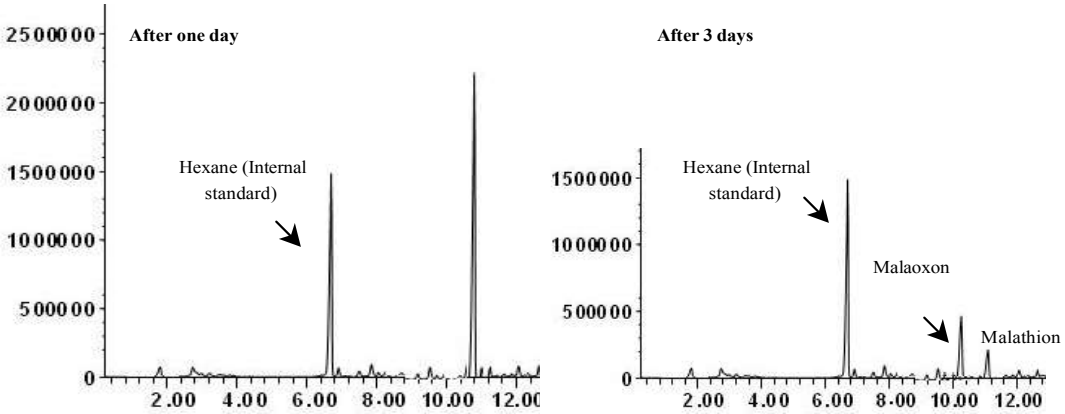


Fig. 6: Quantification of GC-MS analysis of malathion degradation by *Pseudomonas frederiksbergensis*

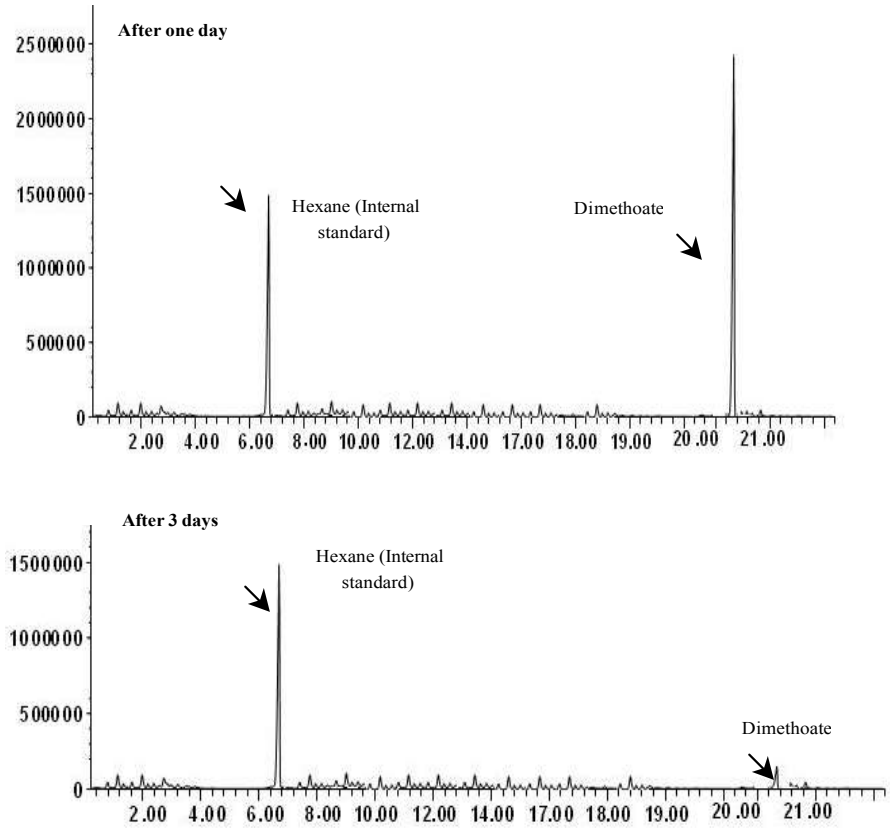
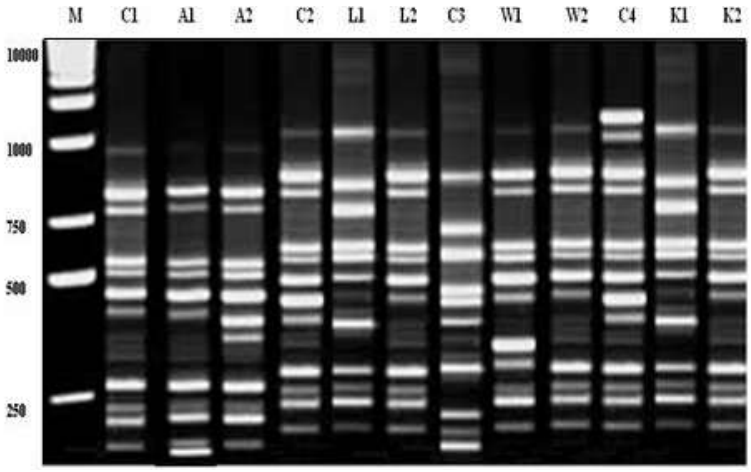


Fig. 7: Quantification of GC-MS analysis of dimethoate degradation by *Pseudomonas frederiksbergensis*

Mutagenic effects of dimethoate and malathion on phytoremediated plants: Random amplified polymorphic DNA-polymerase chain reaction (RAPD-PCR) patterns of plants DNA with both random primers exhibits the effects of malation and dimethoate in plants' DNA. These results revealed sever mutagenic effects on plants´ DNA. The developed bands with various molecular weights were studied. For Amaranth, it was observed that dimethoate has slight effects on the genomic DNA, while malathion has sever effects represented in two three alteration and mutation points in DNA. In contrast, it could be observed that dimethoate has a sever effects on Lettuce compared to malathion treatment. For Watercress, pesticides have severed alterations of genomic DNA, some proteins were synthesized, and some were not compared to the control. It could be observed more or less occurred for Kidney bean treatment and some



M	1 kb		
C1	Amaranth DNA (control)	C3	Water cress DNA (control)
A1	Amaranth with dimethoate	W1	Water cress with dimethoate
A2	Amaranth with malathion	W2	Water cress with malathion
C2	Lettuce DNA (control)	C4	Kidney bean DNA (control)
L1	Lettuce with dimethoate	K1	Kidney bean with dimethoate
L2	Lettuce with malathion	K2	Kidney bean with malathion

Fig. 8: Mutagenic effects of malathion and dimethoate on Plants' DNA. Random amplified polymorphic DNA-polymerase chain reaction (RAPD-PCR) using the phytoremediated plants. Agarose gel electrophoresis (1.25%) [Plant samples were taken after 28 days from cultivation]

mutations occurred with the treatments of the pesticides. In general, malathion has sever effects on DNA in most of the phytoremediated plants.

In fact, the change occurred in the DNA bands may reflect the decreased in total protein in the transcriptional system and this is due to mutations synthesis of DNA resulted from the treatment of these pesticides. Our results are consistent with the result reported in literature [26]. These mutations occurred in DNA could be explain by the hydrophobic nature and small molecular size of dimethoate and malathion that could pass through the cell membrane and reache the nucleus. It is suggested that within the nucleus these pesticides bind to DNA through the reactive groups of their active moieties, leading to destabilization as well as unwinding of the DNA, which could be a possible mechanism for its genotoxicity [27].

On the other hand, these pesticides may induce oxidative stress and generation of reactive oxygen species (ROS) in insect systems [28]. It has been demonstrated that ROS may cause DNA damage, which could lead to single-strand breaks and mutation. Our results clearly show that the pesticides treatments induce DNA mutations in different sites of the plants, comparing with the untreated check. These data suggest that direct binding of dimethoate and malathion to DNA is unlikely to be a mechanism through which any genotoxic effects are produced. This study suggests also that these pesticides have the capacity to alter the genetic material particularly chromosomes in the tested plants.

This investigation shows the isolated bacteria from a soil could assist in plants to increase their ability to degrade dimethoate and malathion. In the analysis of malathion and dimethoate degradation in plants, soil and growth media, there was a reduction of total pesticide residues in the cultivated soil compared with the non cultivated one. This could be because of the natural activity of the microorganisms in the rhizospheric region [29]. Plants have evolved interactions and association with micoorganisms that can cause accelerated breakdown or transformation of certain pollutants in the plant root zones to products that no longer pose environmental hazards. [30] reported that the understanding pesticide metabolism in plants and microorganisms is necessary for pesticide development, for safe and efficient use, as well as for developing pesticide bioremediation strategies for contaminated soil and water. Pesticide biotransformation may occur via multistep processes known as metabolism or cometabolism [31].

In fact, metabolic pathway diversity depends on the chemical structure of the compounds, the organisms, environmental conditions, metabolic factors and the

regulating expression of these biochemical pathways. In general, GC analyses revealed the amount of pesticides adsorbed on soil particles could not be detected and the results may be related to the organic carbon contents of soils according to the previous study [32].

On the other hand, plant-microbe interactions may be beneficial or harmful to the plant depending on the specific microorganisms and plant involved. Plant beneficial interactions can be divided into three categories [33]. The first interaction includes microorganisms, which, in association with the plant, increase the supply of mineral nutrients to the plant. This is the case of the symbiotic dinitrogen-fixing bacteria of leguminous plants as *Rhizobium*, *Bradyrhizobium* species or of monocots as *Azospirillum brasiliense* and the free nitrogen-fixing bacteria such as *Klebsiella pneumoniae* [34]. Secondly, there are microorganisms as *Pseudomonads fluorescent* that stimulate plant growth indirectly by the production of antibiotics, siderophores, volatile compounds or hydrolytic enzymes, which prevent the growth or activity of plant pathogens. Nowadays, these bacteria are used as biocontrol agents. Thirdly, there are the plant growth-promoting rhizobacteria as *Azospirillum*, *Azotobacter*, *Pseudomonas* and *Bacillus* that stimulate directly plant growth by the production of phytohormones [35]. Detrimental interactions within the rhizosphere involve deleterious *rhizobacteria, which* inhibit shoot or root growth without causing any other visual symptoms by the production of phytotoxins such as cyanide [36] or phytohormones. Microorganisms play role in the degradation of pesticide in nature. Bacterial strains isolated from nature are able to degrade a variety of pesticides. However, reports on microbial degradation of dimethoate are very scanty. [1, 37] reported on dimethoate degradation using fungi, *Aspergillus niger*. [34] also used Pseudomonas strain from the coastal water of Haldia Port, India, in their petroleum degradation studies.

Even though the low metabolism efficiency for the absorbed malathion, the high efficiency of the evaluated plants played an important role in degradation of the pesticides. This explained the low level of malathion in the tissue of plants. In addition, effective degradation of malathion in the plants tissue can keep the plants healthy, which is essential for phytoremediation. The improved degradation of OPs pesticides by plants and microorganisms highlights a new way to combat such diffuse contamination. Soil microflora is one of the basic agents for detoxification of pesticides. Some investigators found that soil contaminated with pesticides could be possible decontaminated by inoculation with specifically adapted microorganisms [38]. Enzymatic detoxification of OPs insecticides by some bacterial species has received a considerable attention. Varieties of OPs pesticide-degrading bacteria have been isolated from environments that have met these chemicals [15].

Knowledge obtained from this study could help in understanding the biodegradation of dimethoate and malathion in contaminated sites by plants and microorganisms, as well as to design efficient biocatalyst allowing transformation of pesticides into non toxic compounds. On the other hand, the isolation of *Pseudomonas frederiksbergensis* has a great significance in understanding the role played together with plants in rhizospheric area. More research is necessary to understand the fundamental mechanisms of enhancing and inhibition in the microbial degradation of super high concentration of toxic compounds. However, bacteria could be used very effectively for in situ bioremediation in an environment, which is highly contaminated with pesticides. However, further research could be carried out in these isolate and the plants, on genetic manipulation for improvement and exploitation as bioremediation vehicles.

CONCLUSION

Our results make possible solution to phytoremediate and detoxify these OPs from the environment. This phyto-technology using Amaranth, *Amaranthus caudate*, Lettuce, *Lactuca sativa*, Watercress, *Nasturtium officinale* and Kidney bean, *Phaseolus vulgaris* could detoxify and degrade malathion and dimethoate during cultivation periods. Even though, the mutagenic effects explained the plants tolerance against these toxic compounds. Further studies should be conducted to investigate the mechanisms by which the plants, microorganisms and their enzymes can assimilate these compounds. Knowledge of these enzymatic processes, especially concepts related to pesticides mechanism of action, resistance, selectivity, tolerance and environmental fate, has advanced our understanding of pesticide science and of plant and microbial biochemistry and physiology.

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