

A decorative border consisting of a repeating pattern of small, stylized flowers with multiple petals, arranged in a rectangular frame around the page content.

***RESULTS AND DISCUSSION***

## **4.RESULTS AND DISCUSSION**

### **4.1-Chemical Estimation of Pesticide Residues.**

Soil samples were mixed with five biofertilizers (Cerialin, Rizobacterin, Microbien, Nitrobiein and Phosphorin) and pesticide residues were determined in this soil [Carbamate oxime compound (Methomyl), Organophosphorus compound (Profenofos) and Pyrazole compound (Fenpyroximate)]. Samples were taken 0, 30, 60, 90, 120, 150 and 180 days after application.

The loss of parent compounds was used indicator for pesticide degradation by the multi-strain biofertilizers.

### **4.2-Residues analysis:-**

Samples were taken out in the fixed days for extraction, clean up and determination.

#### **4.2.1-Residues of Methomyl.**

Data in Table (2) & Fig.(1). indicate the pesticide residues in the soil which treated with five different biofertilizer at zero, 30, 60, 90, 120, 150, and 180 days respectively.

Data concerning the existence of detected Methomyl in initial of treatment was (10.13 ppm) The degradation in the soil samples after 30 days of treatment were (7.27, 9.16, 7.15, 8.38 and 7.64 ppm) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 60 days of treatment, methomyl residues were (6.16, 6.33, 5.20, 5.55 and 5.54 ppm) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 90 days of treatment pesticide residues were (4.89, 4.97, 3.91, 4.81 and 4.77 ppm) respectively. while after 120 days of treatment the

pesticide residues decreased to (4.81, 4.77, 2.69, 4.64 and 3.91 ppm) respectively.

Methomyl residues decreased to (4.53, 4.46, 2.46, 4.43 and 2.69 ppm) after 150 days of treatment while degradation increased to be (3.38, 3.75, 1.47, 3.61 and 2.46 ppm) after 180 days .when soil were treated with Cerialin, Rizobacterin, Microbin, Nitrobin and phosphorin respectively and compared to the control soil which contained ( 9.38, 8.16,7.09, 4.09, 4.81 and 3.97 ppm) after 30,60,90,120,150 and 180 days of treatment respectively.

Data in Table (2) . indicate the percentage breakdown of pesticide residues in the soil which treated with five different biofertilizer at zero, 30, 60, 90, 120, 150, and 180 days respectively.

Data concerning the existence of detected percentage breakdown of Methomyl in initial of treatment was ( 0.0 ) The percentage of methomyl breakdown in the soil samples after 30 days of treatment were ( 28.2, 9.6, 29.4, 17.3 and 24.6 %) in soils treated with ( Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 60 days of treatment percentage of methomyl residues breakdown were ( 39.2, 37.5, 48.7, 45.2 and 45.3 %) in soils treated with ( Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 90 days of treatment percentage of methomyl residues breakdown were ( 51.7, 50.9, 61.4, 52.5 and 52.9 %) respectively. while after 120 days of treatment the percentage of methomyl residues breakdown were ( 52.5, 52.9, 73.4, 54.2 and

68.5%) respectively. The percentage of methomyl residues breakdown were ( 55.3, 56.0, 75.7, 56.3 and 73.4 %) after 150 days of treatment. While the percentage of methomyl residues breakdown were ( 66.6, 63.0, 85.5, 64.4, and 75.7%) after 180 days .when soils were treated with Cerialin, Rizobacterin, Microbin, Nitrobin and phosphorin respectively. and compared to the control soil which contained ( 7.4, 19.4, 30.0, 51.6, 52.5 and 60.8 %) after 30, 60, 90, 120, 150 and 180 days of treatment respectively.

Data concerning the breakdown in control sample resulting in the microorganisms which founded in this soil such as *Aspergillus* (33 species), *Penicillium* (46), *Fusarium* (6 species) and *Mucor* (4 species) were high occurrence. Five genera were of moderate occurrence and these were *Humicola*, *Myrothecium*, *Rhizopus*, *Cochliobolus* and *Alternaria*. Fourteen genera were of low occurrence namely, *Cunninghamella*, *Chaetomium*, *Stachybotrys*, *Cladosporium*, *Syncephalastrum*, *Paecilomyces*, *Trichoderma*, *Scolecobasidium*, *Circinella*, *Curvularia*, *Ulocladium*, *Botryotrichum*, *Sepedonium* and *Gliocladium*. Fourteen species were of moderate occurrence and these were *A. sydowii*, *M. verrucaria*, *P. funiculosum*, *A. versicolor*, *H. grisea*, *F. oxysporum*, *A. nidulans*, *F. moniliforme*, *A. alternate*, *F. solani*, *M. hiemalis*, *A. candidus*, *R. nigricans* and *A. ochraceus*. Fungi was recorded, the number /g varied between 81 thousand to 101 thousands. While breakdown in soil sample which treated with biofertilizers resulting in microorganisms in this soil and microorganisms which founded in biofertilizers. The biofertilizers become very active through

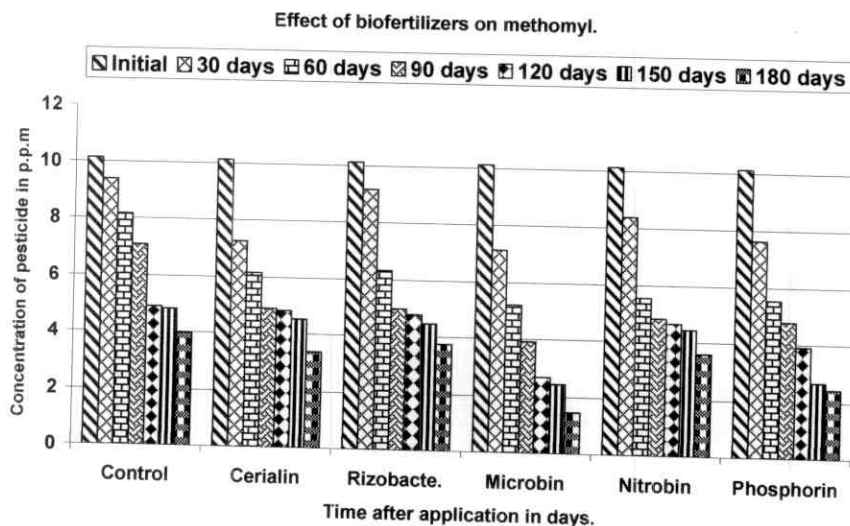
first 90 days of treatment. The biofertilizers increased activity of microorganisms which work to increased degradation of pesticide residues in the soil. All the break down or degradation of methomyl in the control soil sample which had no biofertilizers, is due to natural biodegradation and physical degradation. Many authors reported that more than 33 factors play role in the breaked down of pesticide residues in soil. Natural bioremediation was clear in the case of the samples which were not treated with biofertilizers. During the 180 days 60.8 % of the methomyl residues were degraded or break down naturally without adding any quantity of biofertilizers. While only 30% was degraded or break down after 90 days. While in the soil which was treated by the tested biofertilizers, the percentage of biodegradation and break down was increased to be the double of break down in control (61.4% in the case of soil treated with microbin while in the case of soil treated with cerialin, nitrobin and phosphorin the percentage of biodegradation was 52.7%, 52.5% and 52.9 % respectively. That means according to the content of these biofertilizers from microorganisms, the biodegradation or the bioremediation of methomyl increased.

**Table (2): Statistical analysis of degradation of Methomyl in soil treated with different biofertilizers. (SAS,2004)**

Types of Biofertilizers	Control		Certain		Rizobacterin		Microbin		Nitrobin		Phosphorin	
	% breakdown	Conc. in ppm	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m
Time after application in days												
Initial	0.0	10.13 <sup>a</sup> ± 0.18	0.0	10.13 <sup>a</sup> ± 0.18	0.0	10.13 <sup>a</sup> ± 0.18	0.0	10.13 <sup>a</sup> ± 0.18	0.0	10.13 <sup>a</sup> ± 0.18	0.0	10.13 <sup>a</sup> ± 0.18
30 days	7.4	9.38 <sup>ab</sup> ± 0.68	28.2	7.27 <sup>bcd</sup> ± 1.23	9.6	9.16 <sup>ab</sup> ± 0.04	29.4	7.15 <sup>bcd</sup> ± 0.25	17.3	8.38 <sup>abc</sup> ± 0.04	24.6	7.64 <sup>bcd</sup> ± 0.34
60 days	19.4	8.16 <sup>abc</sup> ± 0.26	39.2	6.16 <sup>cde</sup> ± 0.03	37.5	6.33 <sup>cde</sup> ± 0.97	48.7	5.20 <sup>ef</sup> ± 0.82	45.2	5.55 <sup>def</sup> ± 0.55	45.3	5.54 <sup>def</sup> ± 0.04
90 days	30.0	7.09 <sup>bcd</sup> ± 0.59	51.7	4.89 <sup>ef</sup> ± 0.11	50.9	4.97 <sup>ef</sup> ± 0.33	61.4	3.91 <sup>ijk</sup> ± 0.11	52.5	4.81 <sup>ef</sup> ± 0.01	52.9	4.77 <sup>gh</sup> ± 0.03
120 days	51.6	4.90 <sup>gh</sup> ± 0.4	52.5	4.81 <sup>ef</sup> ± 0.09	52.9	4.77 <sup>gh</sup> ± 0.43	73.4	2.69 <sup>lm</sup> ± 0.19	54.2	4.64 <sup>hijk</sup> ± 0.14	68.5	3.19 <sup>klm</sup> ± 0.31
150 days	52.5	4.81 <sup>hijk</sup> ± 0.19	55.3	4.53 <sup>hijk</sup> ± 0.03	56.0	4.46 <sup>hijk</sup> ± 0.26	75.7	2.46 <sup>mn</sup> ± 0.04	56.3	4.43 <sup>hijk</sup> ± 0.23	73.4	2.69 <sup>mn</sup> ± 0.19
180 days	60.8	3.97 <sup>ijklm</sup> ± 0.63	66.6	3.38 <sup>klmn</sup> ± 0.02	63.0	3.73 <sup>ijklm</sup> ± 0.6	85.5	1.47 <sup>n</sup> ± 0.09	64.4	3.61 <sup>ijklm</sup> ± 0.11	75.7	2.46 <sup>mn</sup> ± 0.16

-Minimum Significant Difference=2.3509

-Means with the same letter are not significantly different.



**Fig. (1). Degradation of Methomyl in soil treated with different biofertilizers.**

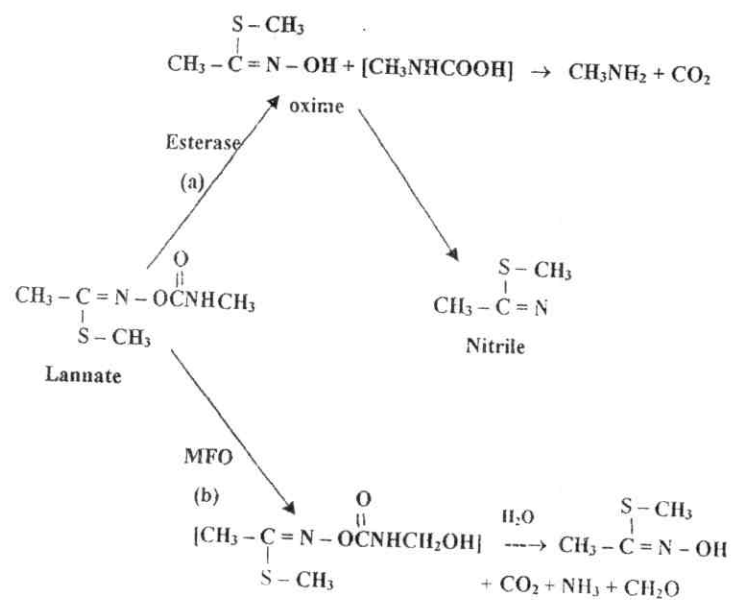
The above results are in agreement with **El-Kabbany (2002)** who reported that the isolated bacterial (*Achromobacter sp.* And *Pseudomonas sp.*) displaying hydrolase activities specific for different classes of carbamats (such as carbaryl, carbofuran and aldicarb). The obtained results indicated that during the first week after application with different biofertilizers the percent loss of carbaryl was about 58.5% and after 2 weeks a substantial loss of extractable residues reach to 86 % in treated soil. The rate of aldicarb disappearance in soil was found to be rapid in the first 7 days, furthermore, the percent recovery of aldicarb decreased faster to reach 93 % at 15 days after application. **Makboul et al., (1989)** reported that the soil fungi *Fusarium oxysporum* and *Rhizoctonia solani* can use meterbuzin and aldicarb as sole source of carbon in pure cultures. Both

species degraded metribuzin to DADK and oxidized aldicarb to its sulfoxide and then sulfone derivatives. **Talebi and Walker (1993)** studied that in laboratory experiments, soils pre-treated with carbofuran were found to degrade the chemical more rapidly than soils which had no carbofuran pre-treatment. When pre-treated soils were sterilized, the rate of carbofuran degradation was greatly reduced and was due to microbial action. The major metabolic route for carbofuran in pre-treated soils involves hydrolysis of the ester bond leading to the release of carbofuran phenol which rapidly binds to soil organic matter; and the release of the carbonyl moiety which quickly degrades to generate carbon dioxide. **Asgari et al., (1995)** studied that a *bacterial consortium* isolated from soil samples from Turkey with a history of aldicarb (Temik) application was tested for its ability to utilize aldicarb as the sole source of carbon and energy. **Kamanavalli and Ninnekar (2000)** studied that a bacterium capable of degrading propoxur was isolated from soil by enrichment cultures and was identified as a *Pseudomonas species*. The organism grew on propoxur as sole source of carbon and nitrogen, and accumulated 2-isopropoxyphenol as a metabolite in the culture medium. The cell free extract of *Pseudomonas sp.* grown on propoxur contained the activity of propoxur hydrolase. The results suggest that the organism degraded propoxur by hydrolysis to yield 2- isopropoxyphenol and methylamine, which was further utilized as a carbon source. **(El-Sharkawi. 2002).** *Pseudomonas putida* was able to metabolize lannate, and converted it to the corresponding oxime which degraded to the corresponding nitrile as shown in (scheme1,2), the degradation products of lannate observed after



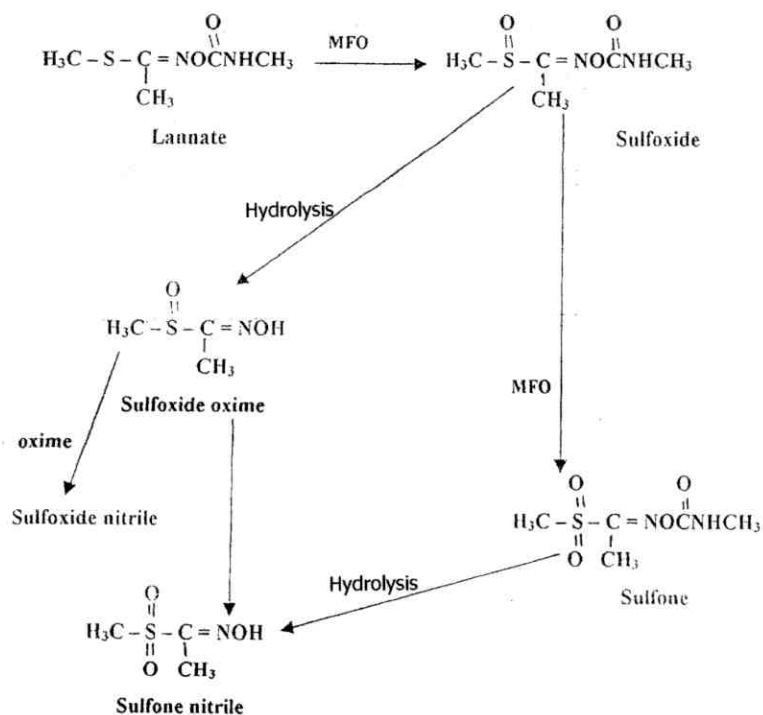
three days was oxime then nitrile was observed. This suggests that oxidation of lannate to its oxime is microbially mediated. Appearance of oximes followed the appearance of nitriles was minor and rarely observed. The oxidation of lannate to the sulfoxide and then the hydrolysis to sulfone was observed and it is the major degradation product. **Rahal et al., (2003)** studied that used of locally isolated microorganisms in bioremediation of the polluted soil with pesticides. Therefore, the study included isolation and identification of some microorganisms from pretreated soil by carbofuran and tested for their tolerance and ability to degrade carbofuran which is extensively used under Egyptian Agricultures against many pests. Those microorganisms were identified as *Streptomyces violaceusniger* and *Azospirillum brasilense*. Six metabolites were detected in *S. violaceusniger* culture by GLC and TLC. Three of them were identified as carbofuran phenol, 3-hydroxy carbofuran and 3-keto carbofuran, whereas the other three metabolites could not be identified. *Abrasifense* degraded carbofuran to only three metabolites; two of them could identify as carbofuran phenol and 3-keto carbofuran, while the third one could not be identified. **Zaghloul et al., (2003)** studied the persistence rate and decomposition of carbofuran and temik nematicides in autoclaved and non-autoclaved soil inoculated with *S. violaceusniger* or /and *A. brasilense*. Results indicated that the application of either carbofuran or temik to soil led to a decrease in total microbial counts, actinomycetes counts and dehydrogenase activity. The main compounds produced from carbofuran degradation were carbofuran-phenol; 3-keto carbofuran; 3-hydroxy-carbofuran and other unknown

compounds. Whereas, temik mainly degraded to temik sulfone, temik sulfoxide and other unknown compounds. **Das et al., (2005)** reported that an experiment has been conducted under laboratory conditions to investigate the effect of carbofuran (a carbamate insecticide). Application of carbofuran in general, induced growth and development of bacteria, actinomycetes, fungi, N<sub>2</sub>-fixing bacteria and phosphate solubilizing microorganisms in both the soils and the stimulation. Carbofuran on the other hand, augmented fungi and N<sub>2</sub>-fixing bacteria in laterite and actinomycetes in alluvial soil. Bacterial population was inhibited due to the application of carbofuran in alluvial soil.



(Scheme 1)

The oxime carbamate (Lannate) degradation, and thioester derivative in (scheme2), (El-Sharkawi, 2002).



(Scheme 2)

Lannate is rapidly metabolized by mixed-function oxidases (MFO) to the sulfoxide and much more slowly to the sulfone, (El-Sharkawi, 2002).

#### 4.2.2-Residues of Profenofos.

Data in Table(3) & Fig.(2). indicate the pesticide residues in the soil which treated with five different biofertilizer at zero, 30, 60, 90, 120, 150, and 180 days respectively.

Data concerning the existence of detected Profenofos in initial of treatment was (4.38 ppm) The degradation in the soil samples after 30 days of treatment were (3.19, 3.24, 3.06, 3.29 and 3.05 ppm) in soils treated with (Cerialin, Rizobacterin,

Microbin, Nitrobin and Phosphorin) respectively. After 60 days of treatment while profenofos residues were (2.18, 1.88, 1.25, 1.34 and 1.20 ppm) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 90 days of treatment pesticide residues were (1.48, 1.24, 0.85, 1.27 and 0.84 ppm) respectively. While after 120 days of treatment the pesticide residues decreased to (0.86, 0.87, 0.42, 0.40 and 0.12 ppm) respectively. Profenofos residues decreased to (0.47, 0.44, 0.12, 0.35 and 0.08 ppm) after 150 days of treatment while degradation decreased to be (N.D, 0.39, N.D, 0.09, and N.D ppm) after 180 days . When soil samples were treated with Cerialin, Rizobacterin, Microbin, Nitrobin and phosphorin respectively, compared to the control soil which contained ( 3.92, 2.39, 1.81, 1.49, 1.30 and 0.91 ppm) after 30, 60, 90, 120, 150 and 180 days of treatment respectively.

Data in Table (3) . indicate the % breakdown of pesticide residues in the soil which treated with five different biofertilizer at zero, 30, 60, 90, 120, 150, and 180 days respectively.

Data concerning the existence of detected percentage of Profenofos breakdown in initial of treatment was (0.0 ) The percentage of Profenofos break down in the soil samples after 30 days of treatment were ( 27.2, 26.0, 30.1, 24.9 and 30.4 %) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 60 days of treatment percentage of Profenofos breakdown were ( 50.2, 57.1, 71.5, 69.4 and 72.6 %) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 90 days of treatment percentage of Profenofos breakdown were ( 66.2, 71.7,

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## **RESULTS AND DISCUSSION**

80.6, 71.0 and 80.8 %) respectively. while after 120 days of treatment the percentage of Profenofos break down were ( 80.4, 80.1, 90.4, 90.9 and 97.3 %) respectively. The percentage of Profenofos break down were ( 89.3, 90.0, 97.3, 92.0 and 98.2 %) after 150 days of treatment. While the percentage of Profenofos break down were (92.7, 91.1, N.D, 97.9, N.D) after 180 days . When soil were treated with Cerialin, Rizobacterin, Microbin, Nitrobin and phosphorin respectively and compared to the control soil which contained ( 10.5, 45.4, 58.7, 66.0,70.3 and 79.2 %) after 30, 60, 90, 120, 150 and 180 days of treatment respectively.

Data concerning the breakdown in control sample resulting in the microorganisms which founded in this soil .While breakdown in soil sample which treated with biofertilizers resulting in microorganisms in this soil and microorganisms which founded in biofertilizers.

The biofertilizers become very active through first 90 days of treatment. The biofertilizers increased activity of microorganisms which work to increased degradation of pesticide residues in the soil.

The degradation of profenofos in the control soil sample which had no biofertilizers , is due to natural biodegradation and physical degradation. Natural bioremediation of profenofos was clear in the case of the samples which were not treated with biofertilizers. During the 180 days 79.2 % of the profenofos residues were degraded or break down naturally without adding

any quantity of biofertilizers. While only 58.6 % was degraded or break down after 90 days.

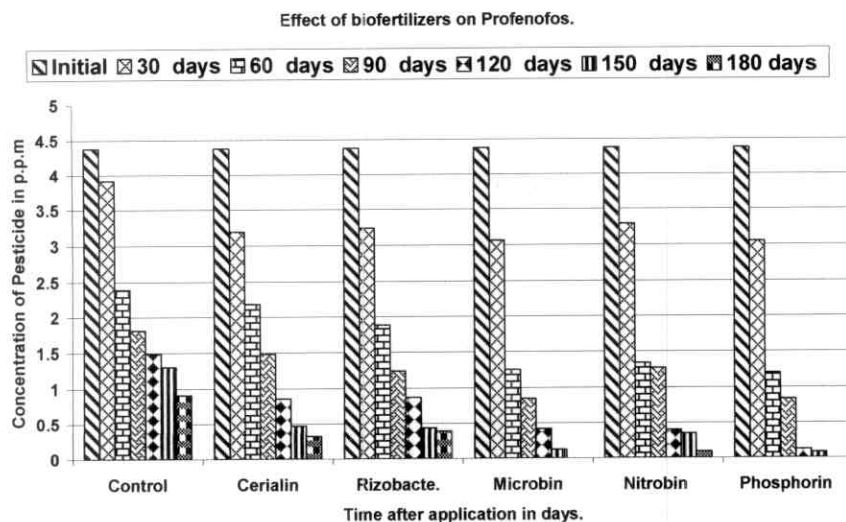
But in the soil which was treated by the tested biofertilizers, the percentages of biodegradation and break down were increased to be (80.6 % and 80.0 % in the case of soil treated soil with microbin and phosphorin respectively , while in the case of soil treated with cerialin, rizobacterin and nitrobin the percentage of biodegradation was 66.3 %, 71.7 % and 71.0 % respectively. That means according to the content of these biofertilizers from microorganisms, the biodegradation or the bioremediation of profenofos increased.

**Table (3): Statistical analysis of degradation of Profenofos in soil treated with different biofertilizers. (SAS,2004)**

Types of biofertilizers	Control		Cerialin		Rizobacterin		Microbin		Nitrobin		Phosphorin	
	% breakd own	Conc. in ppm	% breakd own	Conc. in p.p.m	% breakd own	Conc. in p.p.m	% breakd own	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakd own	Conc. in p.p.m
Initial	0.0	4.38 <sup>a</sup> ± 0.37	0.0	4.38 <sup>a</sup> ± 0.37	0.0	4.38 <sup>a</sup> ± 0.37	0.0	4.38 <sup>a</sup> ± 0.37	0.0	4.38 <sup>a</sup> ± 0.37	0.0	4.38 <sup>a</sup> ± 0.37
30 days	10.5	3.92 <sup>ab</sup> 0.09 ±	27.2	3.19 <sup>abc</sup> ± 0.01	26.0	3.24 <sup>abc</sup> ± 0.26	29.4	3.06 <sup>abcd</sup> ± 0.99	17.3	3.29 <sup>abc</sup> ± 0.21	24.6	3.05 <sup>abcde</sup> ± 0.06
60 days	45.4	2.39 <sup>bcdef</sup> ± 0.25	50.2	2.18 <sup>bcddefg</sup> ± 0.84	57.1	1.88 <sup>cdefgh</sup> ± 0.67	71.5	1.25 <sup>defgh</sup> ± 0.60	69.4	1.34 <sup>defgh</sup> ± 0.61	72.6	1.20 <sup>fgh</sup> ± 0.1
90 days	58.7	1.81 <sup>cdefgh</sup> ± 0.13	66.2	1.48 <sup>cdefgh</sup> ± 0.07	71.7	1.24 <sup>cfigh</sup> ± 0.25	80.6	0.85 <sup>fgh</sup> ± 0.05	71.0	1.27 <sup>defgh</sup> ± 0.03	80.0	0.84 <sup>fgh</sup> ± 0.15
120 days	66.0	1.49 <sup>cdefgh</sup> ± 0.18	80.4	0.86 <sup>fgh</sup> ± 0.09	80.1	0.87 <sup>fgh</sup> ± 0.02	90.4	0.42 <sup>gh</sup> ± 0.80	90.9	0.40 <sup>gh</sup> ± 0.05	97.3	0.12 <sup>h</sup> ± 0.07
150 days	70.3	1.30 <sup>defgh</sup> ± 0.36	89.3	0.44 <sup>gh</sup> ± 0.02	90.0	0.44 <sup>gh</sup> ± 0.09	97.3	0.12 <sup>fgh</sup> ± 0.12	92.0	0.35 <sup>h</sup> ± 0.15	98.2	0.08 <sup>h</sup> ± 0.01
180 days	79.2	0.91 <sup>fgh</sup> ± 0.06	92.7	0.32 <sup>h</sup> ± 0.09	91.1	0.39 <sup>h</sup> ± 0.007	N.D	N.D	97.9	0.09 <sup>h</sup> ± 0.02	N.D	N.D

-Minimum Significant Difference=1.8123 -Means with the same letter are not significantly different. -N.D=non detected.



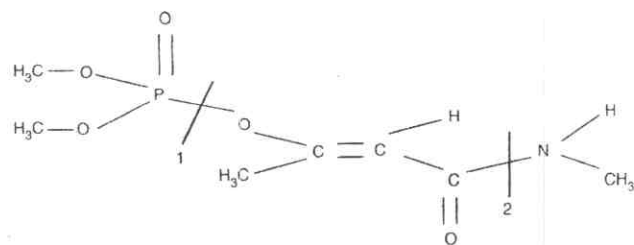


**Fig. (2). Degradation of Profenofos in soil treated with different biofertilizers.**

Results were in agreement with **Ramanathan et al., (1999)**. Organophosphorus insecticides are widely used in agriculture. Reports on the mineralization of a spectrum of these insecticides by a single potential strain are scarce. *Pseudomonas sp. A3*, was isolated through enrichment technique, able to degrade methyl parathion [parathion-methyl] (MP), malathion, monocrotophos, and diazinon. The potential of this strain to mineralize MP as a carbon and/or phosphorus source has been evaluated. During the breakdown of MP, nitrite was released as a catabolic by product. **Dimitrios and Allan (2000)** reported that enhanced biodegradation of ethoprophos was evident in a soil from a previously treated field in Northern Greece. It is suggested that degradation of ethoprophos in the soil from the previously treated field proceeds via hydrolysis of the P-S bond in the -S-propyl moiety of the ethoprophos molecule.

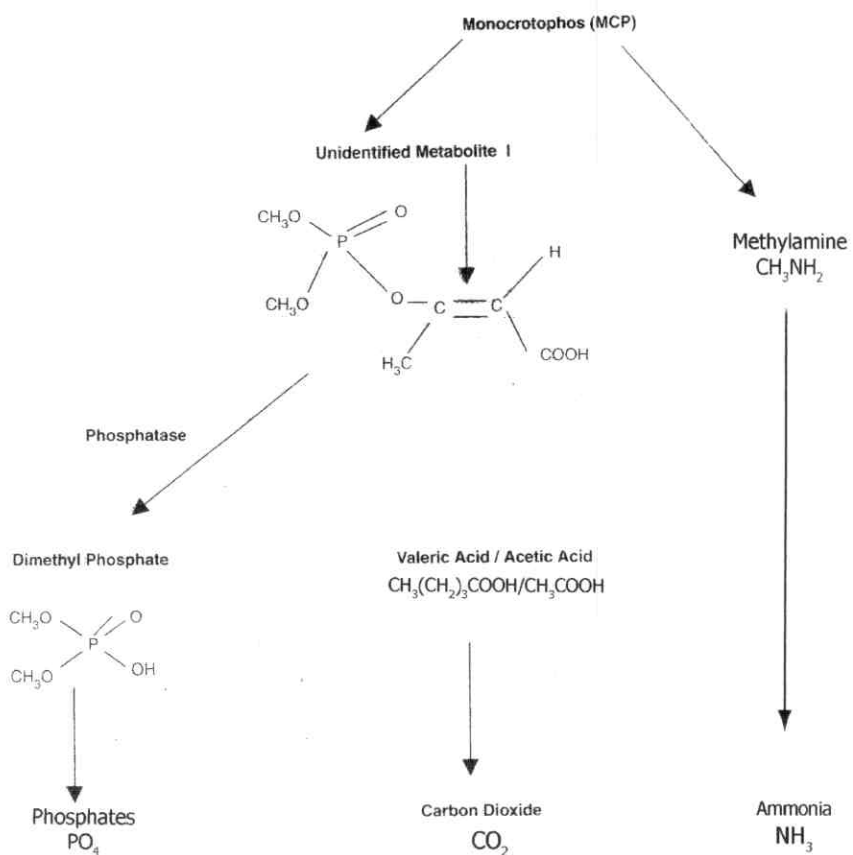
**Bhadbhade et al., (2002)** reported that biomineralization of monocrotophos (MCP) and the metabolites formed during biodegradation was studied. The cultures degraded MCP to carbon dioxide, ammonia and phosphates through formation of one unknown compound metabolite I, valeric or acetic acid and methylamine, as intermediate metabolites. The enzymes phosphatase [phosphoric monoester hydrolase] and esterase, reported to be involved in biodegradation of organophosphorus compounds, were detected in both the organisms. *A. atrocyaneus* MCM B-425 and *B. megaterium* MCM B-423 isolated from soil exposed to MCP were able to mineralize MCP to carbon dioxide, ammonia and phosphates, (Scheme 3).

(a)



Monocrotophos (MCP)

(b)



(Scheme 3) Examples for degradation of organophosphorus compound (Monocrotophos), (Bhadbade et al.,2002).

#### **4.2.3-Residues of Fenpyroximate.**

Data in Table(4) &Fig.(3). indicate the pesticide residues in the soil which treated with five different biofertilizer at zero, 30, 60, 90, 120, 150, and 180 days respectively.

Data concerning the existence of detected Fenpyroximate in initial of treatment was (9 ppm). The degradation in the soil samples after 30 days of treatment were (4.20, 6.21, 4.82, 4.59 and 5.37 ppm) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively, after 60 days of treatment. While fenpyroximate residues were (3.52, 4.72, 2.87, 3.67 and 4.45 ppm) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 90 days of treatment pesticide residues were ( 2.49, 4.41, 2.57, 3.52 and 3.57 ppm) respectively. While after 120 days of treatment the pesticide residues decreased to (2.35, 4.19, 2.45, 3.52 and 3.19 ppm) respectively. Fenpyroximate residues decreased to (2.06, 3.69, 2.12, 2.99 and 2.63 ppm) after 150 days of treatment while degradation decreased to be (1.35, 3.00, 2.09, 2.73 and 2.26 ppm) after 180 days, when soil were treated with Cerialin, Rizobacterin, Microbin, Nitrobin and phosphorin respectively, compared to the control soil which contained (6.43 , 5.67 , 5.50 , 5.48, 4.67 and 4.34 ppm) after 30, 60, 90, 120, 150 and 180 days of treatment respectively.

Data in Table (4) . indicate the % breakdown of pesticide residues in the soil which treated with five different biofertilizer at zero, 30, 60, 90, 120, 150, and 180 days respectively.

Data concerning the existence of detected percentage of Fenpyroximate break down in initial of treatment was ( 0.0 ) The

percentage of break down in the soil samples after 30 days of treatment were ( 53.3, 31.0, 46.4, 49.0, 40.3 %) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 60 days of treatment percentage of fenpyroximate residues break down were ( 60.9, 47.6, 68.1, 59.2 and 50.6 %) in soils treated with (Cerialin, Rizobacterin, Microbin, Nitrobin and Phosphorin) respectively. After 90 days of treatment percentage of fenpyroximate residues break down were ( 72.3, 51.0, 71.4, 60.9 and 60.3 %) respectively. while after 120 days of treatment the % breakdown of pesticide residues were ( 73.9, 53.4, 72.8, 60.9 and 64.6 %) respectively. The percentage of fenpyroximate residues break down were ( 77.1, 59.0, 76.4, 66.8 and 70.8 %) after 150 days of treatment. While the percentage of fenpyroximate residues break down were ( 85.0, 66.7, 76.8, 69.7 and 75.6) after 180 days .when soil were treated with Cerialin, Rizobacterin, Microbin, Nitrobin and phosphorin respectively and compared to the control soil which contained ( 28.6, 37.0, 38.9, 39.1, 48.1 and 51.8 %) after 30, 60, 90, 120, 150 and 180 days of treatment respectively.

Data concerning the break down in control sample resulting in the microorganisms which founded in this soil .While breakdown in soil sample which treated with biofertilizers resulting in microorganisms in this soil and microorganisms which founded in biofertilizers.

The biofertilizers become very active through first 90 days of treatment. The biofertilizers increased activity of

microorganisms which work to increased degradation of pesticide residues in the soil.

Nearly same results were obtained in the case of fenpyroximate in soil treated with the same five biofertilizers. Natural biodegradation and break down in the untreated soil with biofertilizers was 38.9 % after 90 days while it was 51.8 % after 180 days.

Biodegradation of all pesticide residues tested by cerialin and microbin was nearly the double in control after 90 days of treatment. While it was 51.0 and 60.9 and 60.3 % in the case of rizobacterin , nitrobin and phosphorin respectively.

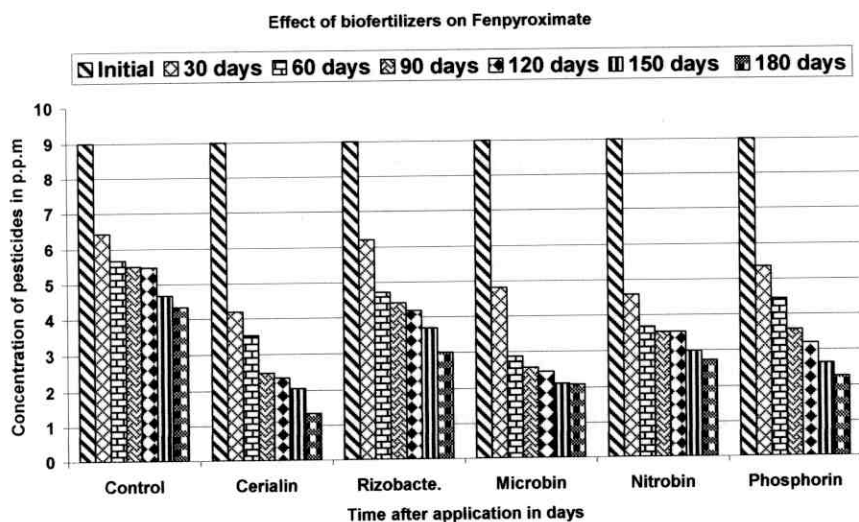
Cerialin headed all the tested biofertilizers in biodegradation of fenpyroximate after 180 days (followed by microbin 76.8 % and phosphorin 74.9 %).

**Table (4): Statistical analysis of degradation of Fenpyroximate in soil treated with different biofertilizers. (SAS.2004)**

Types of Biofertilizers	Control		Ceriatin		Rizobacteria		Microbin		Nitrobin		Phosphorin	
	% breakdown	Conc. in ppm	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m	% breakdown	Conc. in p.p.m
Initial	0.0	9.00 <sup>a</sup> ± 0.20	0.0	9.00 <sup>a</sup> ± 0.20	0.0	9.00 <sup>a</sup> ± 0.20	0.0	9.00 <sup>a</sup> ± 0.20	0.0	9.00 <sup>a</sup> ± 0.20	0.0	9.00 <sup>a</sup> ± 0.20
30 days	28.6	6.43 <sup>ab</sup> ± 0.52	53.3	4.20 <sup>bcd</sup> ± 0.16	31.0	6.21 <sup>abc</sup> ± 1.89	46.4	4.82 <sup>bcd</sup> ± 0.96	49.0	4.59 <sup>bcd</sup> ± 0.38	40.3	5.37 <sup>bcd</sup> ± 1.47
60 days	37.0	5.67 <sup>abcd</sup> ± 0.73	60.9	3.52 <sup>bcd</sup> ± 0.79	47.6	4.72 <sup>bcd</sup> 590. ±	68.1	2.87 <sup>bcd</sup> ± 0.57	59.2	3.67 <sup>bcd</sup> ± 0.83	50.6	4.45 <sup>bcd</sup> ± 0.74
90 days	38.9	5.50 <sup>abcd</sup> ± 0.35	72.3	2.49 <sup>bcd</sup> ± 0.11	51.0	4.41 <sup>bcd</sup> 570. ±	71.4	2.57 <sup>bcd</sup> 90.1 ±	60.9	3.52 <sup>bcd</sup> ± 1.13	60.3	3.57 <sup>bcd</sup> ± 0.37
120 days	39.1	5.48 <sup>abcd</sup> ± 0.73	73.9	2.35 <sup>cde</sup> ± 0.42	53.4	4.19 <sup>bcd</sup> 950. ±	72.8	2.45 <sup>bcd</sup> 80.1 ±	60.9	3.52 <sup>bcd</sup> ± 0.79	64.6	3.19 <sup>bcd</sup> ± 0.87
150 days	48.1	4.67 <sup>bcd</sup> ± 0.88	77.1	2.06 <sup>de</sup> ± 0.31	59.0	3.69 <sup>bcd</sup> ± 1.12	76.4	2.12 <sup>de</sup> 0.73 ±	66.8	2.99 <sup>bcd</sup> ± 0.98	70.8	2.63 <sup>bcd</sup> 0.16 ±
180 days	51.8	4.34 <sup>bcd</sup> ± 0.75	85.0	1.35 <sup>e</sup> ± 0.46	66.7	3.00 <sup>bcd</sup> ± 0.39	76.8	2.09 <sup>de</sup> 820. ±	69.7	2.73 <sup>bcd</sup> ± 0.22	75.6	2.26 <sup>cde</sup> ± 0.23

-Minimum Significant Difference=3.9879

-Means with the same letter are not significantly different.



**Fig. (3). Degradation of Fenpyroximate in soil treated with different biofertilizers.**

The same trend was in agreement with **Guang and Raikookana. (2002)** who studied that degradation of fipronil. Three metabolites of fipronil (desulfinyl, sulfide, and sulfone derivatives) were identified from soils after treatment. Microorganisms in soil accelerated the degradation of fipronil to sulfide and sulfone derivatives. The desulfinyl derivative degraded rapidly in field soils with a half-life of 41–55 days compared with an average half-life of 132 days for fipronil. The half-life of the (total toxic component) (fipronil and its metabolites) in field soil was 188 days on average.

### **Biodegradation or bioremediation of methomyl, profenofos and fenpyroximate by biofertilizers tested.**

Natural biodegradation or bioremediation was very clear in the soil which was not treated by biofertilizers. As it well known, pesticide residues persisted from few days to several



years in the soil. These residues can be affected with 33 factors ,i.e., soil moisture, temperature, cation exchange capacity, organic matter, microorganisms....etc.

In the present study natural biodegradation was clear in the untreated soil sample with biofertilizers. The percentage of biodegradation was 7.4 %,10.5 % and 28.6 after 30 days , while these percentages of the biodegradation or bioremediation increased to be 30.9 % , 58.7 % and 38.9 after 90 days. The maximum natural biodegradation recorded after 180 days were 79.2 %. That means that biodegradation of pesticide can be increase more than 79.2 % after 180 days and it may be 100 % after one year or more according to the type of pesticide residues in the tested soils.

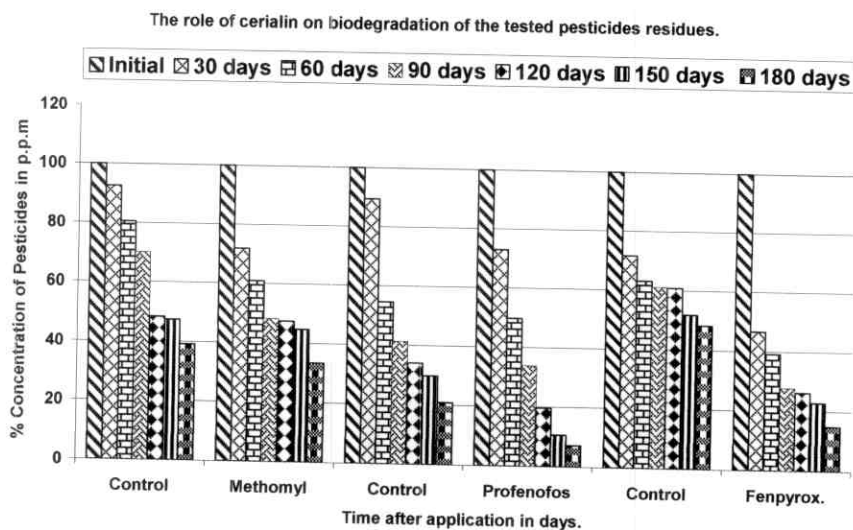
#### **4.3.1-The role of Cerialin on biodegradation of the tested pesticide residues.**

Data in Table (5) &Fig. (4). indicate the percentage of pesticide residues in the soil which treated with Cerialin. This biofertilizer contains (*Bacillus polymyxa*) and others.

Data indicate that percentage of concentration of pesticide residues were lower in this case Profenofos than in this case Fenpyroximare than Methomyl.

**Table (5): The role of Certalin on biodegradation of the tested pesticides residues.**

Types of Biofertilizers	Control		Methomyl		Control		Profenofos		Control		Fenpyroximate	
	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m
Initial	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100
30 days	7.4	92.6	28.2	71.8	10.5	89.5	27.2	72.8	28.6	71.4	53.3	46.7
60 days	19.5	80.6	39.2	60.8	45.5	54.5	50.2	49.8	37.0	63.0	60.9	39.1
90 days	30.0	70.0	51.7	48.3	58.7	41.3	66.2	33.8	38.9	61.1	72.3	27.7
120 days	51.3	48.7	52.5	47.5	66.0	34.0	80.4	19.6	39.1	60.9	73.9	26.1
150 days	52.5	47.5	55.5	44.7	70.3	29.7	89.3	10.7	48.1	51.9	77.1	22.9
180 days	60.8	39.2	66.6	33.4	79.2	20.8	92.7	7.3	51.8	48.2	85.0	15.0



**Fig. (4). The role of Cerialin on biodegradation of the tested pesticides residues.**

The same trend was in agreement with **Omar et al., (2003)** they reported that *Bacillus polymyxa* was more resistant to Topsin M70% and showed that fungicides adversely affected the activity of nitrogen fixing bacteria. **Dellamatrice and Monteiro (2004)**, reported that two *Bacillus spp.* were isolated in medium containing diuron as the only carbon source. Only *A. johnsonii* was able to grow alone in medium with diuron as the only carbon source. **Bennaceur et al., (1999)** found that the rate of lindane disappearance increased with the increase in the frequency of application. After the fourth application of lindane, more than 85% of applied chemical was lost within 8 weeks (70% in control soil). The rapid loss of lindane was detected in the field during the long-term experiment. *Bacillus sp.* was identified as a lindane decomposing microorganisms.

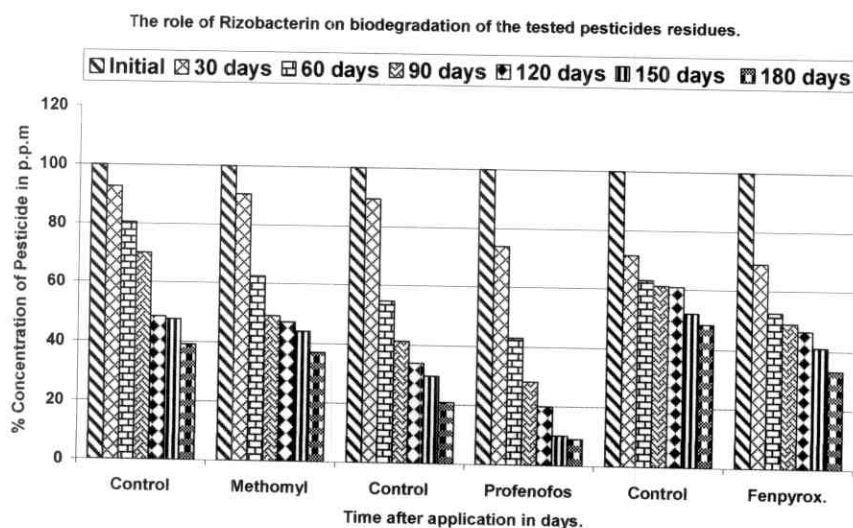
**Table. (6): The role of Rizobacterin on biodegradation of the tested pesticides residues.**

Types of Biofertilizers	Control		Methomyl		Control		Profenofos		Control		Fenpyroximate	
	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m
Time after application in days												
Initial	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100
30 days	7.4	92.6	9.6	90.4	10.5	89.5	23.6	76.4	28.6	71.4	31.0	69.0
60 days	19.5	80.6	37.5	62.5	45.5	54.5	57.1	42.9	37.0	63.0	47.6	52.4
90 days	30.0	70.0	50.9	49.1	58.7	41.3	71.7	28.3	38.9	61.1	51.0	49.0
120 days	51.6	48.4	52.9	47.1	66.0	34.0	80.1	19.9	39.1	60.9	53.4	46.6
150 days	52.5	47.5	56.0	44.0	70.3	29.7	90.0	10.1	48.1	51.9	59.0	41.0
180 days	60.8	39.2	63.0	37.0	79.2	20.8	91.1	8.9	51.8	48.2	66.7	33.3

#### 4.3.2- The role of Rizobacterin on biodegradation of the tested pesticides residues.

Data in Table (6) & Fig. (5). indicate the percentage of concentration of pesticide residues in the soil which treated with Rizobacterin, this biofertilizer contains (*Rizubium sp.*) and others.

Data indicate that percentage of pesticide residues were lower in Profenofos than Fenpyroximare than Methomyl.



**Fig. (5).** The role of Rizobacterin on biodegradation of the tested pesticides residues.

Omar et al., (2003), Dart and Persley (1990) and Smith et al., (2005) reported that *Rizubium sp.* can breakdown different pesticide residues and use these pesticide residues as carbon source.

**Table (7):The role of Microbin on biodegradation of the tested pesticides residues.**

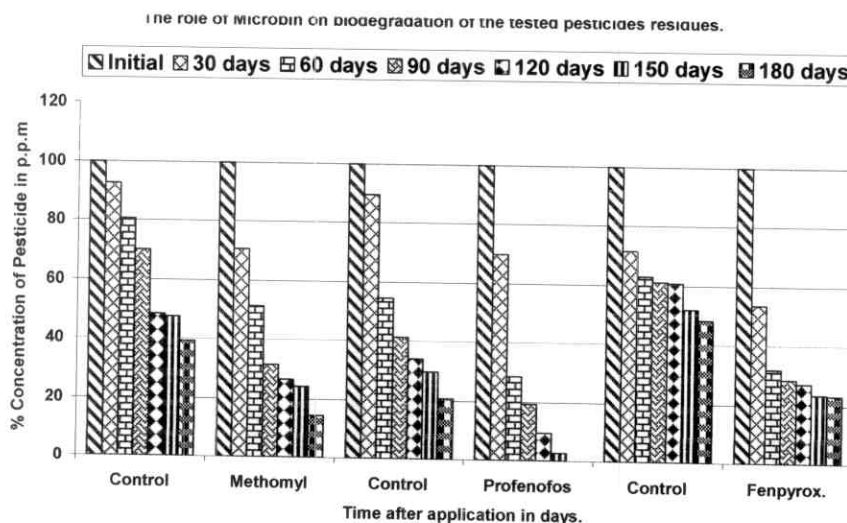
Types of Biofertilizers	Control		Methomyl		Control		Profenofos		Control		Epproximate	
	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m
Initial	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100
30 days	7.4	92.6	29.4	70.6	10.5	89.5	30.1	69.9	28.6	71.4	46.4	53.6
60 days	19.5	80.6	48.7	51.3	45.5	54.5	71.5	28.5	37.0	63.0	68.1	31.9
90 days	30.0	70.0	68.5	31.5	58.7	41.3	80.6	19.4	38.9	61.1	71.4	28.6
120 days	51.6	48.4	73.5	26.6	66.0	34.0	90.4	9.6	39.1	60.9	72.8	27.2
150 days	52.5	47.5	75.5	24.3	70.3	29.7	97.3	2.7	48.1	51.9	76.4	23.6
180 days	60.8	39.2	79.2	14.5	79.2	20.8	N.D	N.D	51.8	48.2	76.8	23.2

**N.D =non detected**

### 4.3.3- The role of Microbin on biodegradation of the tested pesticides residues.

Data in Table (7) & Fig. (6). indicate the percentage of pesticide residues in the soil which treated with Microbin. This biofertilizer contains (*Pseudomonas sp.*) and others .

Data indicate that percentage of pesticide residues were lower in the case Profenofos than Methomyl than in the case Fenpyroximare.



**Fig. (6). The role of Microbin on biodegradation of the tested pesticides residues.**

Ragini et al., (1998) isolated two strains of *Pseudomonas spp.* from soil . The strains were evaluated for their ability to degrade organo chlorinated pesticides. The degradation of DDT and HCH was measured by monitoring the release of organically bound chloride ions and the presence of DDT and HCH residues in the growth medium. Both isolates were capable of utilizing

DDT and HCH as carbon source. **Mathurap and Damodharan (1999)** isolated *Pseudomonas sp. A3* from soil. This strain was able to degrade methyl parathion (MP), malathion, monocrotophos, and Diazinon. The potential of this strain to mineralize MP as a carbon and / or phosphorus source. **Kamanavalli and Ninnekar (2000)** found a *Pseudomonas species* capable of degrading propoxur. The organism grew on propoxur at 2 g/litre as sole source of carbon and nitrogen, and accumulated 2-isopropoxyphenol as a metabolite in the culture medium. **Bhadbhade et al., (2002)** reported that *Pseudomonas mendocina* isolated from soil degraded an insecticide, Monocrotophos (MCP). **(El-Sharkawi. 2002)**. *Pseudomonas putida* was able to metabolize lannate, and converted it to the corresponding oxime which degraded to the corresponding nitrile as shown in (scheme1, 2), the degradation products of lannate observed after three days was oxime then nitrile was observed. This suggests that oxidation of lannate to its oxime is microbially mediated. Appearance of oximes followed the appearance of nitriles was minor and rarely observed. The oxidation of lannate to the sulfoxide and then the hydrolysis to sulfone was observed and it is the major degradation product. **Grant et al., (2002)** showed that the potential for the biodegradation of synthetic pyrethroids (SPs) showed that *Pseudomonas sp.* used this pesticides and depend on the availability of other carbon sources and nutrients. **Walia et al., (2003)** repored that *pseudomonas putida OU83* is a soil organism capable of degrading 3-nitrotoluene (3-NT), 2, 4-dinitrotoluene (DNT), 2, 6-DNT, and mono and polychlorinated biphenyls. An investigation of the nitro group dependent



biotransformation of nitrotoluenes in *P. putida* revealed accumulation of two metabolic products, a and b from each of the three tested NT isomers (2-, 3-, and 4-NT) results show that the degradation of nitrotoluene in *P. putida* was influenced by the position of the nitro group on the benzene ring. **Hong et al., (2005)** reported that *Pseudomonas sp.* strain WBC-3 utilizes methyl parathion (MP) or *p*-nitrophenol (PNP) as the sole source of carbon, nitrogen and energy.

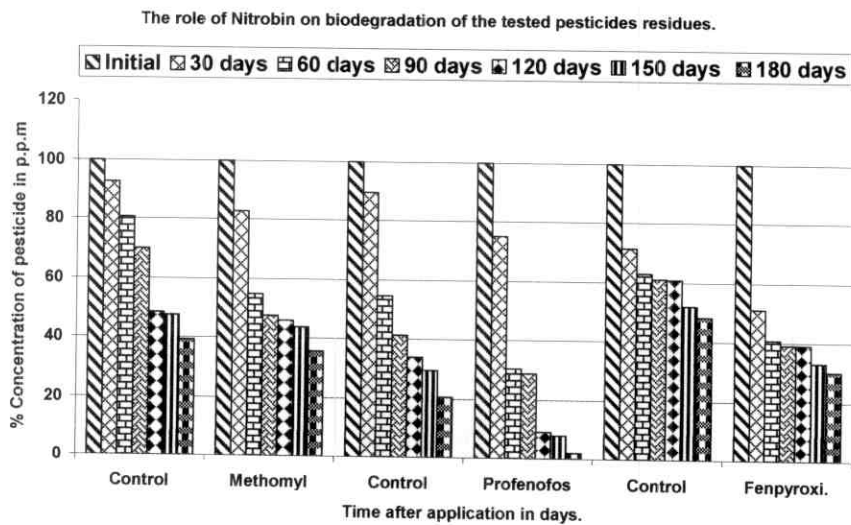
**Table (8): The role of Nitrobin on biodegradation of the tested pesticides residues.**

Types of Biofertilizers	Control		Methomyl		Control		Profenofos		Control		Fenpyroximate	
	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m	% breakdown	% Conc. in p p m
Time after application in days												
<b>Initial</b>	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100
<b>30 days</b>	7.4	92.6	17.3	82.7	10.5	89.5	24.9	75.1	28.6	71.4	49.0	51.0
<b>60 days</b>	19.5	80.6	45.2	54.8	45.5	54.5	69.4	30.9	37.0	63.0	59.2	40.8
<b>90 days</b>	30.0	70.0	52.5	47.5	58.7	41.3	71.0	29.0	38.9	61.1	60.9	39.1
<b>120 days</b>	51.6	48.4	54.2	45.8	66.0	34.0	90.9	9.1	39.1	60.9	60.9	39.1
<b>150 days</b>	52.5	47.5	56.3	43.7	70.3	29.7	92.0	8.0	48.1	51.9	66.8	33.2
<b>180 days</b>	60.8	39.2	64.4	35.6	79.2	20.8	98.0	2.1	51.8	48.2	69.7	30.3

#### 4.3.4- The role of Nitrobin on biodegradation of the tested pesticides residues.

Data in Table (8) & Fig. (7). indicate the percentage of pesticide residues in the soil which treated with Nitrobin. This biofertilizers content (*Azotobacter sp.*) and others.

The percentage of pesticide residues were lower in the case of Profenofos than Fenpyroximare than in the case Methomyl.



**Fig. (7). The role of Nitrobin on biodegradation of the tested pesticides residues.**

Omar et al., (2003) reported that *Azotobacter chroococcum* was more resistant to Topsin M70% and showed that fungicides adversely affected the activity of nitrogen fixing bacteria.

**Table (9): The role of Phosphorin on biodegradation of the tested pesticides residues.**

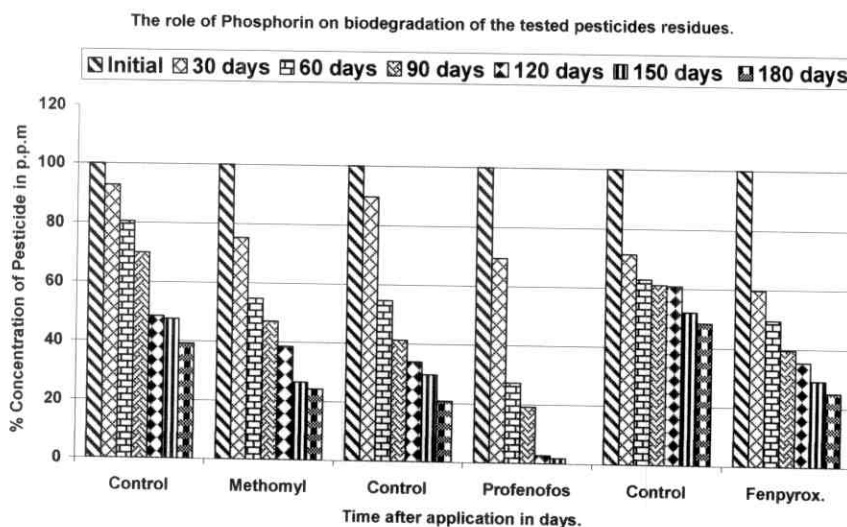
Types of Bioherbers Time after application in days	Control		Methomyl		Control		Profenofos		Control		Fenpyroximate	
	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m	% breakdown	% Conc. in p.p.m
Initial	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100	0.0	100
30 days	7.4	92.6	24.6	75.4	10.5	89.5	30.4	69.6	28.6	71.4	40.3	59.7
60 days	19.5	80.6	45.3	54.7	45.5	54.5	72.6	27.4	37.0	63.0	50.6	49.4
90 days	30.0	70.0	52.9	47.1	58.7	41.3	80.8	19.2	38.9	61.1	60.3	39.7
120 days	51.6	48.4	61.4	38.6	66.0	34.0	97.3	2.7	39.1	60.9	64.6	35.4
150 days	52.5	47.5	73.5	26.6	70.3	29.7	98.2	1.8	48.1	51.9	70.8	29.2
180 days	60.8	39.2	75.7	24.3	79.2	20.8	N.D	N.D	51.8	48.2	74.9	25.1

**N.D =non detected**

#### 4.3.5- The role of Phosphorin on biodegradation of the tested pesticides residues.

Data in Table (9) & Fig. (8). indicate the percentage of pesticide residues in the soil, which treated with Phosphorin. This biofertilizer contains (*Bacillus megaterium*) and others.

Percentage of pesticide residues were lower in the case of Profenofos than Methomyl, than in the case of Fenpyroximate.



**Fig.(8). The role of Phosphorin on biodegradation of the tested pesticides residues.**

Bhadbhade et al., (2002) reported that *B. megaterium* MCM B-423 isolated from soil exposed to MCP was able to mineralize MCP to carbon dioxide, ammonia and phosphates. Omar et al., (2003) reported that *Bacillus megaterium* as phosphate dissolving bacteria decreased the recommended dose

of fungicides. This decrease did not inhibit the growth of the pathogenic fungi but increased bacterial number.

In the recent experiment it is clear that the pesticides residues in the tested soil varied clearly in their concentration in the soil and also in their chemical contents.

The concentration of pesticide residues in the tested soil varied between 4.38 ppm profenofos, 9.00 ppm fenpyroximate and 10.13 ppm methomyl in all the tested soils in control and in the soil samples each treated with one of the following biofertilizers (Cerialin, rizobacterin, microbin, nitrobin and phosphorin).

The Natural biodegradation and break down of pesticides was clear in the samples which were not treated with biofertilizers and the percentage of this natural remediation was clear from the first 30 days of treatment to the end of treatment. It was slowly after the beginning of the experiment but increased at the end of the experiment.

The effect of biofertilizers tested was very clear on the biodegradation of pesticide residues.

Microbin, phosphorin and nitrobin headed all the tested biofertilizers in biodegradation of profenofos. The percentage of remediation was 100 %, 100 % and 98.0 % respectively at the end of experiment (after 180 days).

Cerialin gave 92.7 % remediation of profenofos after 180 days of treatment., It was responsible for 85.0 % bioremediation of fenpyroximate and only 66.6 % bioremediation of methomyl residues in the tested soil.

While rizobacterin gave 91.1 % remediation of profenofos after 180 days of treatment. It was responsible for 66.7 %

bioremediation of fenpyroximate and only 63.0 % bioremediation of methomyl residues in the tested soil.

The results indicate that all biofertilizers tested play good role on the biodegradation of the tested pesticide residues due to their contents of microorganisms or due to their side good effects on the living organisms which are available in the tested soil.