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Bio-Remedaition of Fluazifop-P-Butyl Herbicide Contaminated Soil with Special Reference to Efficacy of Some Weed Control Treatments in Faba Bean Plants

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Abstract: The objective of this investigation aimed to study the effect of fluazifop-p-butyl alone or in combination with hand hoeing on the weed infestation and on nodulation, growth and yield components of faba bean plants. The potential of inoculation *Rhizobium* to degrade fluazifop-p-butyl herbicide as well as the role of broad bean cultivation and hand hoeing practice on persistence of the herbicide in soil was also studied. Number and dry weight of narrow leaved weeds were significantly decreased by different treatments, the most effective treatments were: cultivated and inoculated soil + fluazifop-p-butyl (3L/ fed), cultivated and inoculated soil + fluazifop-p-butyl (2L/Fed.)+ one hand hoeing and cultivated and inoculated soil + fluazifop-p-butyl(2L/Fed.). Cultivated and inoculated soil + two hand hoeing followed by cultivated and inoculated soil + fluazifop-p-butyl + one hand hoeing recorded the maximum values of number and dry weight of nodules / plant, some physiological parameters, yield and yield attributed as well as protein percentage. While cultivated soil with *Rhizobium* + fluazifop-p-butyl successively was the most treatments in increasing root, shoot and total dry weight of faba bean plant after 60 and 90 days from sowing. The initial deposits of fluazifop-p-butyl were 35.62, 36.13, 31.26, 30.45, 39.89 and 57.54 ppm for uncultivated and uninoculated soil; uncultivated and inoculated soil; cultivated soil, cultivated and inoculated soil; cultivated and inoculated soil followed by one hand hoeing as well as cultivated and inoculated soil at rate of tested herbicide 3 L./ fed, respectively. The residual loss amounted to be 22.85, 19.73, 24.5, 25.68, 23.26 and 27.77 % one day after application, respectively. These values declined to 6.17, 3.63, 0.12, undetected, undetected and 1.3 ppm recording to the rates of loss 82.68, 89.95, 99.62, > 99.99, > 99.99 and 97.74 % at 28 days after spraying, respectively. The results revealed that the residues dissipated rapidly in cultivated and inoculated soil followed by one hand hoeing and in case of cultivated and inoculated soil (undetectable residues at 21 and 28 days after application, respectively). In other words, obtained results clearly showed that microorganism living in soil and in root nodules have an obvious role in pesticides biodegradation, also, hand hoeing process playing the same role.

Key words: Faba bean, annual grasses, *Rhizobium* inoculation, hand hoeing, fluazifop-p-butyl, bioremediation

INTRODUCTION

Faba bean (*Vicia faba*, L.) as the most popular seed legume, is an important source of protein in the Egyptian food. It also improves the fertility of the soil via providing a substantial input of N_2 fixation. The reduction of faba bean yield is mainly due to weed infestation which reached 30 to 44 %[12]. Thus, weed control is one of the essential cultural practices for raising faba bean yield and improving its quality. Two hand hoeing are recommended for effective weed control in faba bean[13,14,29].

On the other hand, various herbicides are used for controlling different species of weeds. Application of fluazifop-p-butyl at the rate of 2L./ fed significantly reduced dry matter of narrow weeds and increased growth and seed yield[20,27]. However, the direct or indirect application of these pesticides may enter or more into soil, air and water compartments of the environment[7,11].

A variety of microorganism (bacteria and fungi) has been used in soil inoculations intended to improve the supply of nutrients to crop plants, to stimulate plant growth, to control or inhibit the activity of plant pathogens and to improve soil structure. Recently, other objectives such as introduction of microorganism into soil to mineralize of organic pollutants are also considered (bioremediation of polluted soils, Van Veen *et al.*[30]). The bioremediation technology of the contaminants is nowadays in progress to dispose the

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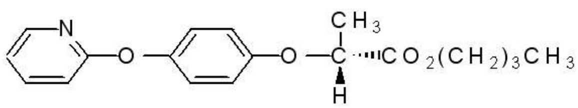
soil pollutes and maintains the environment cleaner and safer as well.[15,23,26].

The objectives of the present study aimed to investigate the following aspects:

a- The effect of the fluazifop-p-butyl herbicide alone or in combination with hand hoeing on the weed infestation, nodulation, growth and yield of the crop.
b- Potentials of *Rhizobium* inoculation to degrade fluazifop-p-butyl herbicide as well as the role of faba bean cultivation and hand hoeing practice on persistence of the herbicide into soil.

MATERIALS AND METHODS

I- Herbicide Used: Fluazifop- P-butyl (Fusalide Super E.C. 12.5 %)



Uses: Post-emergence control of wild oats, volunteer cereals and annual and perennial grassy weeds in oilseed rape sugar beet, fodder beet, potato, vegetables, cotton, soybean, pome fruit, stone fruit, bush fruit, vines, citrus fruit, pineapples, banana, strawberry, sunflower, alfalfa, ornamentals and other broad-leaved crops[3].

II- Field Experiments: Two field experiments were carried out during the two successive seasons (2004/2005 and 2005/2006) at the Agricultural Experimental station of National Research Centre, Egypt. Experimental soil was a clay loam with organic matter 1.78 %, pH 7.79, total N. 0.079 % and available P 14.2 ppm. The experiments were laid out in Randomized Complete Blocks Design with four replicates for each treatment. The experimental unit was 3.5 X 3.0 m. Faba bean seeds (Giza 40) were inoculated with the specific *Rhizobium* strain and immediately sown in hills 25 cm apart on both sides of the ridge. Sowing dates were November, 5 and 7 for the two seasons 2004/2005 and 2005/2006, respectively. The herbicide fluazifop-p-butyl was applied as foliar application 4 weeks from sowing by using knapsack sprayer equipped with one nozzle boam. Broad-leaved weeds only were controlled by hand weeding 6 weeks after sowing. The normal cultural practices of growing faba bean plants were followed normally. Harvesting was performed in April 21 and 25 for the first and second seasons, respectively. Treatments were arranged randomly as follows:

1- Cultivated soil + fluazifop-p-butyl (2 L/ fed).
2- Cultivated and inoculated soil+ tested herbicide (2 L/ fed).
3- Uncultivated soil + tested herbicide (2 L/ fed).
4- Uncultivated and inoculated soil + tested herbicide (2 L/ fed).
5- Cultivated and inoculated soil + tested herbicide (2 L/ fed) + one hand hoeing after 6 weeks from sowing.
6- Cultivated and inoculated soil + tested herbicide (3 L/ fed).
7- Cultivated and inoculated soil.
8- Cultivated and inoculated soil + two hand hoeing after 3 and 6 weeks from sowing. 9- Cultivated soil only (control).

Data Recorded:

III- On Weeds: After 60 days from sowing and at harvest in both seasons, weed samples from one square meter area were randomly collected from each plot. Number and dry weight of weeds were determined after drying in a forced draft oven at 70 °C to constant weight. The most dominant weeds in both growing seasons were: Wild oat (*Avena fatua* L.), Ryegrass (*Lolium temulentum* L.); Bermudagrass (*Cynodon dactylon* L.) and Purple nutsedge (*Cyperus rotundus* L.).

IV- On Faba Bean Plants:

A- Vegetative Growth Parameters: After 60 and 90 days from sowing in both seasons samples of five random plants were taken from experimental plots to estimate the following characteristics:

1- Total number of nodules bacterial / plant.
2- Dry weight of nodules bacterial / plant (g.). 3- Plant height (cm)
4- Root dry weight (g). 5- Shoot dry weight (g). 6- Plant dry weight (g).
7- Number of branches / plant. 8- Leaf area index (LAI).
9- Specific leaf area (SLA, cm^2/mg). 10- Net assimilation rate (NAR, gm/dm^2/week).
11- Relative growth rate (RGR, gm/ gm / week).
12- Crop growth rate (CGR, gm/ m^2 / day).

(LAI, SLA, NAR, RGR and CGR were determined according to Watson[31]).

B- Yield and Yield Attributes: At harvesting, the following data were recorded:

1- Number of pods / plant. 2- Pods dry weight / plant (g). 3- Seeds weight / plant (g).

4- Number of seeds / plant. 5- Seed yield (ton / fed.) for the last traits the two central ridges of each experimental unit were devoted the determination.

C- Total Protein Content: Total nitrogen content was determined by Kjeldahl method[21]. N values were multiplied by 6.25 to calculate protein content.

V- Residues Determination of Fluazifop-p-Butyl Herbicide:

A- Soil Samples: A sample of four replicates (250-500 grams each) were collected from different treatments of the contaminated soil. The soil samples were collected randomly from the soil surface (depth of 10 cm) surrounding the treated plants at zero time (one hour after application), 1, 7, 14, 21 and 28 days post application. Clean polyethylene bags were used for preservation of the collected samples. All samples were stored at -20 ̊C in a deepfreeze until time of analysis.

B- Extraction and Clean- up: Fifty grams of soil were shaken mechanically (one hour) with 100 ml methanol. The extract was filtered through Buchner funnel containing filter paper (Whatman No.1). This step was repeated twice with 50 ml methanol. The filtrate was collected and transferred to separately funnel (250 ml), then 100 ml ethyl acetate was added and shook for 2 min. After separation and equilibration of the layer, the organic layer was transferred to conical flask (250 ml) and dried over anhydrous sodium sulfate[5]. The extract was evaporated using a rotary evaporator at 40 ºC. The residues were ready for HPLC determination.

C- Quantitative Analysis: Fluazifop-p-butyl residues were quantitatively analyzed using a high performance liquid chromatography (HPLC) according to the technique of El-Mahy[8]. The chromatographic system consisted of Hewlett Packard (HP series 1100), Quaternary pump model (G 1314A) monitored at 217 nm. An ODS-Hypersil 5 µm (20 cm X 4.6 mm i.d.) was used and the column temperature was 40 ̊C. The residues were eluted isocratically with acetonitrile – water (80: 20 v/v). A 20 µl injector was used at a flow rate 1.0 ml / min. Under these conditions, the retention time for fluazifop- p-butyl was 4.364 min.

The obtained data were corrected according to the rate of recovery which was 89.2 %. The residual half-lives (RL_{50}) values were calculated using Moye's *et al.*[18] equation.

VI- Statistical Analysis: The data obtained were subjected to analysis of variance (ANOVA) according to Gomez and Gomez[10].

RESULTS AND DISCUSSIONS

I- Effect of Weed Control Treatments:

On the Weeds: Data in Table 1 revealed that all weed control treatments under investigation significantly inhibited the number and dry weight of narrow leaved weeds as well as the percentage of control as compared to unweeded check (control) after 60 days from sowing as well as at harvest. The results also indicated that cultivated and inoculated soil + fluazifop-p-butyl treatment (3 L. / fed) gave the best weed control when compared to other weed control treatments. It recorded dry weight of weed than cultivated soil only (control) by 90.16 and 87.88 % at 60 day from sowing and at harvest, respectively. With regard to weed control treatments, data cleared that the highest efficiency in decreased dry weight of narrow leaved weeds were obtained by cultivated and inoculated soil + fluazifop-p-butyl (2 L./ fed) + one hand hoeing, cultivated and inoculated soil + fluazifop-p-butyl (2 L./ fed) and cultivated and inoculated soil+ two hand hoeing treatments, respectively. These treatments decreased dry weight of narrow leaved weeds than cultivated soil only (control) by 88.57, 86.60, 86.29 and 82.86 % at 60 days from sowing as well as 87.58, 85.78, 84.68 and 83.60 % at harvest, respectively.

While, the highest dry weight of narrow leaved weeds after 60 days from sowing and at harvest were observed with cultivated soil (control) treatment followed by that of cultivated soil with *Rhizobium* inoculated treatments. Generally, the results recorded in Table 1 indicated that fluazifop-p-butyl alone or inoculated soil + one hand hoeing treatments decreased significantly number and dry weight of faba bean narrow leaved weeds as compared to control. These reductions may be due to the inhibition effect of herbicidal and hand hoeing treatments on growth and development of weeds. These results are in harmony with those obtained by Metwally[17], Singh and Wright[28], Singh and Jolly[29], El-Mahy[8] and Abd El-Razik[1].

2- On Faba Bean Characters:

a- Number and Dry Weight of Nodules/Plant: Data presented in Table 2 showed that number and dry weight of nodules / plant were significantly increased with inoculated of *Rhizobium* and recommended rate of tested herbicide + one hand hoeing treatment as compared to the uninoculated soil or higher concentrations of fluazifop-p-butyl treatment. Cultivated and inoculated soil + fluazifop-p-butyl (2 L./ fed) + one hand hoeing treatment recorded the highest values of number and dry weight of nodules / plant followed by that of cultivated and inoculated soil+ two hand hoeing and cultivated soil

Table 1: Average of number dry weight of narrow leaves weeds (g/m^2) after 60 days from sowing and at harvest as affected by some weed control treatments (Average of the two seasons).

Parameters	At 60 days			At harvest		
Treatments	Number of narrow weeds / m^2	Dry weight of narrow weeds / m^2	Inhibition % of control	Number of narrow weeds / m^2	Dry weight of narrow weeds / m^2	Inhibition % of control
1	6.21	4.22	86.60	11.12	15.17	85.78
2	6.36	4.32	86.29	12.30	17.42	84.68
3	10.32	7.01	77.75	19.82	88.07	75.31
4	9.87	6.9	78.10	19.40	27.80	75.54
5	5.17	3.6	88.57	10.72	14.12	87.58
6	4.40	3.10	90.16	10.32	13.78	87.88
7	43.12	30.18	4.19	80.75	107.60	5.36
8	8.11	5.40	82.86	14.18	18.65	83.60
9	45.90	31.50	--	82.62	113.70	--
F-test	**	**	--	**	**	--
LSD 5 %	0.76	0.42	--	1.12	1.75	--
1 %	1.14	0.63	--	1.68	2.63	--

1- Cultivated and uninoculated soil + tested herbicide (2 L/ Fed.).
2- Cultivated and inoculated soil + tested herbicide (2 L/ Fed.).
3- Uncultivated and uninoculated soil + tested herbicide (2 L/ Fed.).
4- Uncultivated and inoculated soil + tested herbicide (2 L/ Fed.).
5- Cultivated and inoculated soil followed by one hand hoeing + tested herbicide (2 L/ Fed.).
6- Cultivated and inoculated soil + tested herbicide (3 L/ Fed.).
7- Cultivated and inoculated soil.
8- Cultivated and inoculated soil followed by two hand hoeing.
9- Cultivated and uninoculated soil (control).

with *Rhizobium* + fluazifop-p-butyl (2 L / fed), respectively. On the other hand, the lowest number and dry weight of nodules / plant were recorded with uncultivated soil (control) followed by that of cultivated and inoculated soil + fluazifop-p-butyl (3 L. / fed) treatments. Similar results were obtained by Attia[4] and Singh and Wright[28].

b- Vegetative Growth: In average of two seasons, weed control treatments with *Rhizobium* inoculated had significant effects on vegetative growth of faba bean as shown in Table 2. Cultivated and inoculated soil + fluazifop-p-butyl (2 L./ fed) + one hand hoeing, followed by cultivated with inoculated soil + two hand hoeing and cultivated soil with *Rhizobium* + tested herbicide (2 L / fed) significantly increased root, shoot and total dry weight of faba bean plants after 60 days from sowing as well as root, shoot, total dry weight of plant, number of branches / plant and plant height after 90 days from sowing as compared to other treatments. In contrast, the lowest values of growth characters were observed in the cultivated soil with uninoculated *Rhizobium* (control). Generally, it can be concluded that the highest increase in growth of faba bean plants was achieved from plots inoculated with *Rhizobium* and fluazifop-p-butyl + one hand hoeing.

The high numbers of nodules, higher nodule and root weight in faba bean probably resulted in higher aboveground plant growth of this treatment (*Rhizobium* and fluazifop-p-butyl + one hand hoeing), as greater nodulation should have resulted in high rated of nitrogen fixation and better root growth can enable plants to absorb more nutrients and water for shoot growth as well as number of branches and plant height. Similar results were obtained by Jat and Gaur[13], Jat *et al.*[14], Singh and Jolly[29] and Abd EL-Razik[1].

c- Physiological Parameters: Results also indicated that all physiological parameters of plants i.e. LAI at 60 and 90 days from sowing, NAR, RGR and CGR were significantly increased by application of weed control treatments (Table 3). Cultivated and inoculated soil + two hand hoeing followed by that of cultivated and inoculated soil + fluazifop-p-butyl and one hand

Table 2: Average number of nodules, dry weight and some growth characters of faba bean plants after 60 and 90 days from sowing as affected by some weed control treatments (Average of the two seasons).

			At 60 days			At 90 days				
	Total No. of nodules /plant	Dry weight of nodules /plant (g)	Root dry weight /plant(g)	Shoot dry weight /plant(g)	Total dry weight /plant(g)	Root dry weight /plant(g)	Shoot dry weight /plant(g)	Total dry weight /plant(g)	Number of branches /plant	Plant height (cm)
1	51.89	0.43	1.54	7.13	8.67	2.56	16.73	19.29	3.11	92.99
2	55.11	0.46	1.78	7.54	9.32	2.74	19.79	22.53	3.53	93.22
3	-	-	-	-	-	-	-	-	-	-
4	-	-	-	-	-	-	-	-	-	-
5	57.11	0.50	1.95	7.70	9.65	3.40	21.93	25.33	4.11	95.11
6	38.89	0.28	1.52	6.80	8.32	2.42	15.69	18.11	3.08	84.33
7	43.44	0.35	1.40	6.47	7.87	2.48	13.87	16.35	2.92	92.00
8	58.78	0.59	1.82	7.55	9.37	3.12	21.74	24.86	3.75	94.00
9	32.34	0.25	1.31	5.62	6.93	2.11	13.64	15.75	2.27	81.67
F test	**	**	**	**	**	**	**	**	**	**
LSD 5 %	3.21	0.06	0.12	0.52	0.64	0.30	2.11	1.02	0.36	4.37
1 %	4.82	0.09	0.18	0.78	0.95	0.45	3.17	1.52	0.54	6.52

Table 3: Average of physiological parameters, yield and yield attributes as well as protein percentage as affected by some weed control treatments (Average of the two seasons).

Treat -ments	LAI at 60 days	LAI at 90 Days	SLA cm^2/mg at 60 days	SLA cm^2/mg at 90 days	NAR gm/dm^2 /week	RGR gm/gm week	CGR/ gm/m2 days	N. of Pods /plant	Pod dry w/ plant(g)	Seed w./ plant(g)	No. of seeds /plant	Seed yield ton/fed	Prote %
1	20.18	4.38	28.77	26.22	0.076	8.50	0.094	20.66	45.43	32.17	52.22	2.15	26.80
2	2.67	4.40	31.43	28.11	0.094	11.64	0.098	20.11	46.08	34.12	55.12	2.40	28.24
3	-	-	-	-	-	-	-	-	-	-	-	-	-
4	-	-	-	-	-	-	-	-	-	-	-	-	-
5	2.70	4.75	30.99	27.24	0.090	12.32	0.112	21.90	50.18	36.12	58.17	2.77	28.70
6	2.47	4.12	28.90	25.65	0.087	9.76	0.089	18.89	44.66	31.78	46.44	2.00	26.12
7	2.26	3.20	26.84	24.74	0.075	7.70	0.086	17.00	37.05	27.20	45.00	1.75	25.73
8	2.89	5.30	31.73	27.36	0.096	13.23	0.115	23.44	51.25	37.11	59.33	2.88	29.11
9	1.79	3.00	25.14	23.76	0.070	6.40	0.083	16.17	34.08	25.00	41.99	1.54	25.15
F test	**	**	NS	NS	**	**	**	**	**	**	**	**	**
5 %	0.36	0.50	--	--	0.004	1.25	0.006	3.76	2.60	2.22	1.40	0.20	0.35
1 %	0.54	0.75	--	--	0.006	1.86	0.009	-	3.90	3.33	2.10	0.30	0.53

hoeing treatments markedly increased the pervious parameters as compared with other treatments. On the other hand, the lowest values of previous characters were recorded with the unweeded plots (control). The treatments proved to be effective in controlling weeds and consequently the competition was limited and lighter, water and nutrients were available to promote the faba bean growth if compared to the other treatments. These results are in good agreement with those recorded by Mahmoud[16] and Ramadan and Saad El-Din[24]. Data also indicated that no significant differences were found among the treatments in SLA through the two successive seasons.

d- Yield and Yield Attributes: It is obvious from the data in Table 3 which reveal that weed control treatments significant influence on yield and yield attributes. Cultivated and inoculated soil + hand hoeing twice or fluazifop-p-butyl with one hand hoeing significantly increased number of pods / plant, pods dry weight / plant, seeds weight / plant and number of seeds / plant as compared to the other treatments. Vice-versa, the lowest values of pervious characters were recorded with control plots. The increase in yield attributes by different weed control treatments may be due to good control of faba bean weeds and minimizing weed competition which gave good chance of faba bean growth and improved good characters. The promoting effect of weed control treatments on growth characters of plants may be reflected on increasing the yield attributes of faba bean. These results are in coinciding with those detected by Saad El-Din and El-Metwally[24], Sharara *et al.*[27] and Abd El-Razik[1].

With regard to seed yield / fed, data in Table 3 show significant differences in yield as an average of the two seasons. Weed control treatments with *Rhizobium* inoculation markedly produced higher seed yield than control. Hand hoeing twice with *Rhizobium* inoculated, fluazifop-p-butyl with *Rhizobium* + one hand hoeing treatments recorded the highest seed yield / fed. as compared with other treatments. Such superior treatments increased the average of seed yield than control treatment by about 87.01 and 79.87 % as an average of the two seasons. On the other side, the lowest values of faba bean yield were obtained when plots were left unweeded and uninoculated. The high yields obtained with some of the treatments (Table3) were the result of effective weed control, as can be seen from the weed control efficiency (Table1). The treatments giving low yields had poor weed control efficiency. However, high weed control efficiency and consequently high seed yield have been reported with use of fluazifop-p-butyl[17] and two hand hoeing[1]. Further, compared to the sole application of herbicides, the integration of post-emergence herbicides with one hand hoeing resulted in lower weed dry matter and higher seed yield of faba bean[25]. The same conclusion was mentioned by Singh and Jolly[29], Sharara *et al.*[27] and Abd El-Razik[1].

e- Total Protein Content: These results revealed that all weed control treatments significantly increased protein percentage as shown in Table 3. The highest protein percentage was recorded with inoculation and two hand hoeing treatment. While, the lowest protein percentage resulted from untreated plots. The increase in total protein content may be due to inoculated which due to promote the rate of nitrogen fixation and better growth can enable plants to absorb more nutrients and water for shoot growth and faba bean plants may be reflected on seed protein content. Also, these results may be due to less competition for nutrients water and light through limiting weeds infestation with two hand hoeing or herbicidal treatments due to increasing the uptake of different nutrients. Similar results were obtained by Saad El-Din[25] and Abd El-Razik[1].

II- Bioremediation of Fluazifop-p-Butyl-Contaminated Soil: Results in Table 4 revealed that the initial deposits of fluazifop-p-butyl herbicide in case of uncultivated and uninoculated soil were 35.62 ppm, one hour after spraying. Only, 22.85 % of this amount can be degraded within the first day. Obtained data also indicated that the percent residues loss was continued on prolonging the time, where the percent loss amounted to 44.92, 65.69, 77.46 and 82.68 % after 1, 2, 3 and 4 weeks from spraying, respectively.

Addition of *Rhizobium* to uncultivated soil (inoculated) had negligible effect on degradation of fluazifop-p-butyl in soil, compared to the non *Rhizobium* treated case. The data obtained cleared that the initial deposit of fluazifop-p-butyl was 36.13 ppm, this amount dropped to 29.0 ppm within 24 hours. The residues continued to decrease 16.16, 9.37, 5.97 and 3.63 ppm after 7,14, 21 and 28 days from spraying, respectively. The data also revealed that the percent loss in case of addition *Rhizobium* to the soil was much higher (89.95 %) compared with uncultivated and uninoculated soil (82.68 %). Moreover, calculated half life period when the compound applied solely to the soil was 6.26 days, while this value was 5.8 days when the soil mixing with *Rhizobium* (Fig.1). These results are in agreement with those obtained by Abdel-Rahman[2], who reported that degradation of some pesticides in soil not only due to the chemical hydrolysis but might, also due to the biological activity of soil microorganisms. Also, Cutright and Lee[6] concluded that all bacteria that use complex aromatic hydrocarbons, as the carbon / energy source in cometabolism with the transfer of respiratory electrons.

Obtained data (Table 4) revealed that cultivation was efficient in the removal of most tested herbicide residues from the treated soil. The initial deposit was 31.26 ppm; this amount was dropped to 23.6 ppm indicating 24.5 % loss within 24 hours of application. The residues of the compound decreased after wards

Table 4: Dissipation rate of fluazifop-p-butyl herbicide after soil application at of different treatment.

Days	Zero time	1day		7 days		14 days		21 days		28 days	
Treatments	ppm	ppm	Loss%	ppm	Loss%	ppm	Loss%	ppm	Loss%	ppm	Loss%
Uncultivated+Uninoculated	35.62	27.48	22.85	19.62	44.92	12.22	65.69	8.03	77.46	6.17	82.68
Uncultivated+Inoculated	36.13	29.0	19.73	16.16	55.27	9.37	74.07	5.97	83.48	3.63	89.95
Cultivated+Uninoculated	31.26	23.6	24.5	15.61	50.06	4.67	85.06	2.63	91.59	0.12	99.62
Cultivated+Inoculated	30.45	22.63	25.68	12.26	59.74	3.72	87.78	0.99	96.75	ND	>99.99
Culti.+inoc.+h. hoeing	39.89	30.59	23.26	11.27	71.75	4.31	89.2	ND	>99.99	ND	>99.99
Cult.+inoc. (3 L/fed)	57.54	41.56	27.77	28.75	50.03	6.12	89.36	2.34	95.93	1.3	97.74

Zero time = One hour after application, ND = Nonedetected
Cult.= Cultivation, Inoc. = Inoculation

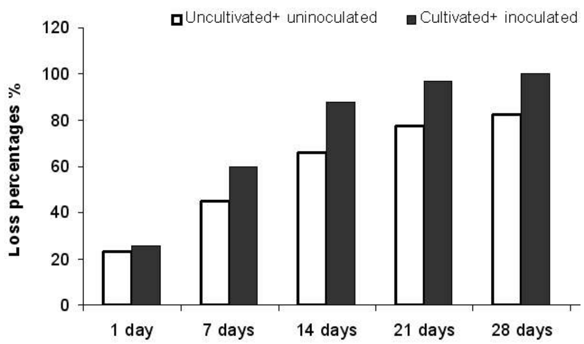


Fig. 1: The role of cultivation and inoculation on persistence of fluazifop-p-butyl in contaminated -soil.

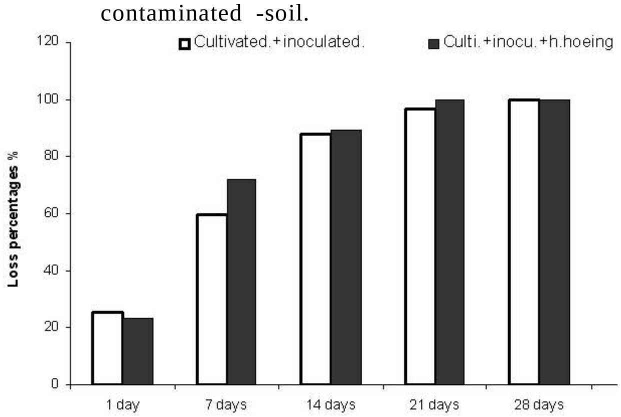


Fig. 2: The role of cultivation and inoculation followed by hand hoeing on persistence of fluazifop-p-butyl in contaminated -soil.

to 15.61, 4.67, 2.63 and 0.12 ppm in soil samples collected at 7, 14, 21 and 28 days after spraying, revealing 50.06, 85.06, 91.59 and 99.62 % loss. As a matter fact, *Rhizobium* bacteria species are able to fix atmospheric nitrogen while living symbiotically in root nodules legumes. However, *Rhizobium* species are free-living organisms and mobile in soil, and as such, are unable to fix atmospheric nitrogen[22]. So, these microorganisms may be use the degradable pesticides as a food source.

Similarly, the results in Table 4 indicated that the persistence of tested herbicide in cultivated soil was influenced by mixing faba seeds with the microorganisms more than that the corresponding uncultivated and uninoculated soil (Fig.1), or inoculated soil and / or the treated cultivated soil. It is obvious that the initial deposit was 30.45 ppm, after one day 22.63 ppm was detected with a 25.68 % loss, it was gradually decreased to 12.26 ppm after with 59.74 % of total loss. Further decrease of the residues was observed after 14 and 21 days from application giving the values of 3.72 and 0.99 ppm, respectively. The corresponding percentages of total loss were 87.78 and 96.75 % , respectively. No residues were detected after 28 days from application.

The data clearly showed that mixing the seeds with *Rhizobium* (cultivated + inoculated soil) and applying the hand hoeing process was very efficient in degradation of the residues when compared with the other treatments (Fig.2). The results pointed out that the initial deposit was 39.89 ppm, this amount was decreased to 30.59 ppm recording 23.26 % loss after 24 hours of application. Then the residues were dropped to 11.27 and 4.31 ppm in soil samples collected 7 and 14 days from spraying, giving 71.75 and 89.2 % loss. The results also revealed undetectable residues in soil samples collected at 21 and 28 days after application.

Treated soil with the tested compound at 3 L. / fed showed high levels of residues at all intervals (Table 4), so, the initial deposit of this herbicide was 57.54 ppm, then gradually decreased to 41.56 ppm at one day after application revealing 27.77 % loss. This value declined to 1.3 ppm recording to the rate of loss 97.74 % at 28 days after spraying. Naser and Hegazy[19] reported that extreme amounts of the pesticide residues as well as the great variation between their deposits could be

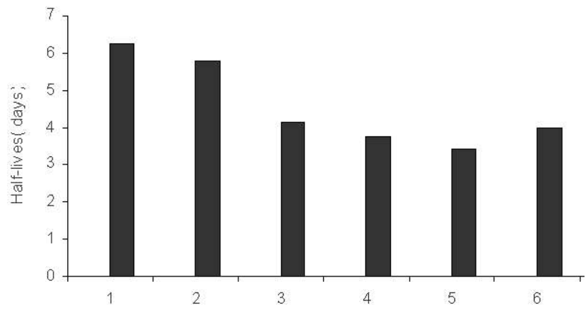


Fig. 3: Residual half-lives (RL_{50}) of fluazifop-p-butyl from soil of different treatments.

1- Uncultivated and uninoculated soil.
2- Uncultivated and inoculated soil.
3- Cultivated and uninoculated soil. 4- Cultivated and inoculated soil.
5- Cultivated and inoculated soil followed by one hand hoeing.
6 Cultivated and inoculated soil (3 L/ Fed.).

attributed to the differences in their applied rate. Also, El-Sayed *et al.*[9] stated that the amounts of deposits depended on the rate of application, the nature of the treated surface and the relation between the surface treated and its weight.

Furthermore, the estimated residual half-lives (RL_{50}) (Fig. 3) of uncultivated soil was 6.26 days, uncultivated soil with *Rhizobium* was 5.8 days, cultivated soil was 4.16 days, mixing the seeds with *Rhizobium* was 3.75 days, mixing seeds with *Rhizobium* followed by one hand hoeing was 3.4 days, while, in case of mixing seeds with Rhizobium followed by tested herbicide at rate 3 L./ fed was 3.97 days.

Generally, application of fluazifop-p-butyl + *Rahizobium* + one hand hoeing decreased significantly the dry weight of the weeds. These treatments also increased number and dry weight of nodules, yield and yield attributed of bean plants.

The obtained data also revealed that mixing the seed with *Rhizobium* followed by hand hoeing was the most active treatment for reducing fluazifop-p-butyl residues in soil, then cultivation broad bean in inoculations soil with *Rhizobium*, and then cultivation broad bean in non inoculations soil and addition microorganisms to soil without cultivation in comparison with spraying non inoculations soil by the tested herbicide, respectively. In other words, the obtained results clearly showed that microorganism lives in soil and in root nodules have an obvious role in pesticides biodegradation; also, hand hoeing practice plays the same role.

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