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BIO- AND PHYTOREMEDIATION TO DECONTAMINATE THE ATRAZINE RESIDUES IN WATER

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ABSTRACT

The efficiency of the bioremediation using two microorganisms, i.e. *Pseudomonas fluorescens* bacteria and *Trichoderma viride* fungus and the phytoremediation utilizing two plants, i.e. water hyacinth and giant reed was investigated to minimize the residues of the herbicide atrazine in water samples collected from Menofia Governorate. The obtained results of the bioremediation experiment carried out under laboratory conditions revealed that the bacteria was more potent than fungi. The statistical and mathematical analysis of the plotted degradation regression lines of the tested concentration (1 ppm) showed that the estimated half-life periods ($t_{1/2}$) were 4.95 and 7.08 days and the slope values were – 0.0865 and – 0.0429 for the utilized bacteria and fungi, respectively. Moreover, the phytoremediation of the same herbicide under open field conditions showed that the dissipation rate of atrazine was found to be slightly higher in case of using water hyacinth and moderately with the giant reed than the disappearance rate recorded by the two tested plants together (water hyacinth + giant reed). The estimated half-life periods ($t_{1/2}$) were 5.32, 6.96 and 7.026 days and the slope values of the degradation regression lines were – 0.0899, 0.0558 and – 0.0469, respectively for water hyacinth, giant reed and water hyacinth plus giant reed. The obtained results revealed the usefulness of the mentioned bacteria, fungi and plants to decontaminate the herbicide atrazine residues in water.

Keywords: Bioremediation, Phytoremediation, Atrazine residues, Water

INTRODUCTION

The presence of various pollutants such as pesticides in the River Nile near the metropolitan areas in Egypt are now well documented (**Osfor *et al* 1998 and Zidan *et al* 2003**). Such toxicants are not only detrimental to human health but to the health of the entire Ecosystem. The herbicide, atrazine is considered one of these pollutants because of their physicochemical characteristics. Beside the urgent need to the regular survey and monitoring the level of the pesticide residues in the environmental elements, the importance of the remediation procedures to

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decontaminate the ecosystem components is well documented. Since 1970s, it was found that the bioremediation is an attractive method for contaminated site restoration because it accomplishes the dual objective of contaminant removal and destruction. Scientifically, such methods depend on using microorganisms to biodegrade the contaminants in an aqueous environment through their enzymatic activity that is capable to degrading the contaminant and transforming it to a beneficial form, i.e. as carbon source **(Newcombe & Crowley, 1999 and Radosevich & Tuovinen 2004).**

On the other hand, phytoremediation was defined as the use of plants for environmental clean-up. In other words, phytoremediation (also called, green remediation) was identified as the use of plants to remediate either partially or substantially selected contaminants present in soil, sludge, sediment, ground water, surface water including waste water (**Xia-Huilong *et al* 2002; Belden *et al* 2004**).

Depending on the importance of such trend of investigation, the aim of the present work is directed to study the efficiency of remediation, i.e. bioremediation and the phytoremediation methods for the herbicide, atrazine in water.

MATERIAL AND METHODS

1- Pesticide used

Atrazine (6-chloro-N^2- ethyl- N^4-isopropyl-1, 3, 5-triazine- 2, 4-diamine) active ingredient (a.i., 95%) was used in the bioremediation experiment while the formulation form (Atrazex® 80% W.P) was used in the phytoremediation.

2. Design of the bioremediation experiment

2.1. Isolation, purification and identification of the pesticide utilizing microorganisms in sediment and water samples

The pesticide-utilizing microorganisms from water and sediment samples collected from Samadon, Ashmon and El-Ramla was used for isolating and cultivating. The mineral salt medium (MSM) was used as described by **Palittapongarnpim *et al* (1998).** The enumeration of total microorganisms in either water or sediment samples was recorded and calculated through the equation described by **Thiery and Francon (1997).** Ten grams of sediment or 10 ml of water sample were inoculated into each of the three replicates of 500 ml Erlenmeyer flasks containing 100 ml MSM and atrazine at concentration of 1 ppm. The flasks were shaken at room temperature of 25°C on a rotary shaker at 150 rpm. After seven days of enrichment, 0.1 ml of the supernatant was spread on selective MSM agar plates. The plates were incubated at 25 °C for 2-4 days growth period. Colonies with different characteristics were then repeatedly transferred to pesticide/MSM agar plates until pure culture of each kind of colonies was obtained. All purified culture of bacteria or fungi were kept on nutrient glucose agar (NGA) or (PDA) slants at 4°C as stock cultures for further work, and was transferred every month.

Forty bacterial isolates differed in their cultural and morphological characteristics were purified on mineral salt agar medium according to the method described by (**Palittapongarnpim and *et al* 1998**). For fungi, the developing five

fungal colonies were purified by single spore technique. Identification of purified fungi was accomplished according to the keys given by **Gilman (1957) and Barnett & Hunter (1972)**. However, the identification of the representative isolates was confirmed in Plant Pathology and Bacteriology Department, Faculty of Agriculture, Ain Shams University, Cairo, Egypt.

2.2. Bioremediation of atrazine herbicide

To determine the efficiency of the isolated bacteria and fungi in the decomposition the atrazine, the purified bacterial isolate *Pseudomonas fluorescens* and the fungal strain *Trichoderma viride* were used. To study the biodegradation of atrazine herbicide by *Pseudomonas fluorescens* and *Tricoderma viride* an experiment was conducted as follows: 50 ml of mineral salts (MS) medium were dispensed into 108 conical flasks (100 ml), then the flasks were sterilized at 121°C for 20 minutes. After sterilization and cooling at room temperature, atrazine was added to the flasks under aseptic conditions to give final concentrations of 1 and 10 ppm. Some flasks were kept without atrazine as control. Flasks were inoculated with 1 ml of spore suspension of *Trichoderma viride* fungus and with *Pseudomonas flourocence* bacterium. The control was kept without inoculation. All flasks were incubated at 28°C for 45 days. At different intervals, i.e. 0, 1, 2, 4, 8, 10, 15, 30, and 45 days, three flasks were taken from each treatment as well as the control. Three replicates of each group were adjusted for residue analysis.

3. Phytoremediation of atrazine

The experimental work was carried out under open field conditions during summer 2004 in a private land located at Smadon village, Menofia Governorate, Egypt. The treated area was 100 m^2 with twelve plots each plot 1x1x1m. The plots were divided into three sites, each site contained 4 plots which included three replicates and one control. The first site was planted with giant reed (*Arundo donax* L.), the second was planted with water hyacinth (*Eichhornia crassipes* Mart.) and the third with both giant reed and water hyacinth. The application rate of the tested atrazine was (1mg/L) on each plot, which was equivalent to 1 ppm. Three replicates of each group have been taken for residue analysis.

3.1. Sampling technique

Thirty-six samples of water (each sample contained 1 liter), were collected from all sites with three replicates at different periods after treatment, i.e. 0, 1, 2, 4, 8, 10, 15, 30 and 45 days. The collected samples were transferred to the laboratory, prepared and subjected to residue analysis.

3.2. Extraction, clean-up and determination procedures

Both of the extraction and clean-up procedures of the collected and representative mineral salt (MS) medium and water samples were carried out according to the Method No.507, **(U.S. EPA, 1988)**. In such method, 50 ml of medium sample was taken for extraction. Two portions of methylene chloride (each 50ml) plus saturated sodium chloride were used for ex-

traction 500 ml water. Two portions of methylene chloride (each 100 ml) plus saturated sodium chloride were used. There after the extracts were evaporated to a final volume of 5ml for clean-up and determination by gas chromatography (GC) (Shimadzu 12-A) with Flame Ionization Detector (FID) at the following conditions: G.C column packed with 3% silicon OV-101 of chromosorb Q; Inj./Det. Temp. (250°C); Oven Temp. (220°C); N_2 1.5 (kg/cm^2 Flow: 30 ml/min); H_2 (1.0 kg/cm^2 Flow: 30 ml/min); air (1.0 kg/cm^2 Flow: 30 ml/min) and Attenuation (10 x 5).

Such procedures were used to establish the standard calibration curve of atrazine and recovery tests. The mean percentage recovery of atrazine from spiked medium at 1.0 ppm was 88.3 %, whereas it was 83.70 % from spiked water sample at the same concentration. All detected concentrations of atrazine were corrected based in the rate of recovery. Statistical and mathimatical analysis of the obtained data was carried out according to **Timme and Freshes (1980).**

RESULTS AND DISCUSSION

1. Bioremediation efficiency

Two microorganisms were used in this experiment, i.e. bacteria (*Pseudomonas fluorescens*) and fungus (*Trichoderma viride*) to investigate the possibility to biodegrade the most common herbicide e.g. atrazine. The design of such part of study is divided into two main experiments, the first is to isolate and identify the microorganisms that already exist in the collected samples Secondly, to study the capability of the most frequently isolated microorganism to biodegrade the herbicide atrazine under laboratory conditions.

1.1. Isolation, identification of bacterial and fungal microorganisms

Data in Table (1), revealed that water and sediment samples contained different isolates of bacteria and fungi. From water samples collected from Samadon, two bacterial isolates as were identified, i.e., *Bacillus subtilis* and *Pseudomonas* sp. Water samples from both of Ashmon and El Ramla sites, showed two bacterial isolate, i.e., *Bacillus subtilis* and *Pseudomonas* sp. In samples of sediments collected from Samadon, four isolates of bacteria were identified, i.e. *Bacillus subtilis*, *Pseudomonas* sp., B4 and B5. Ashmon sediment samples contained three bacterial isolates, i.e. *Bacillus subtilis*, B3 and B6 while sediment samples of EL Ramla site contained one bacterial isolate, i.e. *Pseudomonas* sp.

In case of the fungal isolates, *Trichoderma viride* was isolated from water samples of Samadon and *Asparagellus* sp from Ashmon water samples. In sediment samples, two fungal isolates were found in Samadon sediment samples, i.e. *Trichoderma viride* and *Asparagellus* sp. while one fungal isolate from sediment samples was collected from Ashmon, i.e. *Trichoderma viride* and two fungal isolates from El Ramla site, i.e. *Trichoderma viride* and *Penicillium* sp. According to the bacterial and/or fungal isolates, it was found that such isolates represent most microorganisms that are capable to biodegrade the pesticides (**Patil *et al* 1970 and Fradette *et al* 1994**). The biochemical and physiological functions of the mentioned bacterial isolates are listed in **Table (2).**

Table 1. Isolated and identified microorganisms from water and sediment samples collected from Samadon, Ashmon and El-Ramla, Menofia Governorate, Egypt.

Sample	Site		
	Smadon	Ashmon	El-Ramla
Water	*Trichoderma viride* *Penicillum sp* Bacillus subtilis *Pseudomonas sp*	*Asparagellus sp* *Bacillus subtilis*	*Pseudomonas sp*
Sediment	*Trichoderma viride* *Asparagellus sp* *Penicillum sp* *Bacillus subtilis* *Pseudomonas sp* *Penicillum sp* B4 B5	*Trichoderma viride* *Bacillus subtilis* *B3* B6	*Trichoderma viride* *Penicillum sp* *Pseudomonas sp*

Table 2. Biochemical and physiological characters of bacteria isolated from water and sediment samples collected from Samadon, Ashmon and El-Ramla village, Menofia Governorate, Egypt.

Test	Isolate	
	Bacillus subtilis	Pseudomonas fluorescens biovar III or V
KOH 3%	-	+
Catalase	+	+
Oxidase	+	+
Starch hydrolysis	+	-
Gelatin liquefication	+	+
Lipolytic activity	-	-
Voges- Proskauer (V.P)	+	-
Indol production	-	-
Arginin dihydrolase	-	-
Nitrate reduction	+	+
Fluorescence pigment *	-	+
Levan formation	-	-
Anaerobic growth	-	-
Acid production from		
Mannose	+	+
Glucose	+	+
Lactose	+	+
Sorbitol	+	+
Arabinose	+	-
Sucrose	+	-

Data of the biochemical and physiological functions of the identified isolates indicate that *Bacillus subtilis* gave negative result with KOH 3%, lipolytic activity, indol production, arginin dihydrolase, fluorescence pigment on King'B medium, levan formation and anaerobic growth. Conversely, such isolate gave positive results with catalase, oxidase, starch hydrolysis, gelatin liquefication Voges-Proskauer (VP), nitrate reduction, utilization of mannose, Glucose Lactose sorbitol Arabinose and Sucrose. In addition, the second isolate of *Pseudomonas fluorescens* biovar III and/or V gave negative results with starch hydrolysis, lipolytic activity, indol production, arginin dihydrolase, levan formation and utilization of Sucrose. At the same time, such isolates gave positive results with KOH 3%, catalase oxidase, gelatin liquefication, nitrate reduction, fluorescence pigment on King'B medium, and utilization of mannose, glucose, lactose & sorbitol. Fungal isolates were identified as *Trichoderma viride*, *Aspargellus niger* and *Penicillium* sp according to the developing five colonies purified by single spore technique. Identification of the purified fungi was accomplished according to the keys give by **Gilman (1957) and Barnett & Hunter (1972).**

1.2. Persistence and biodegradation of atrazine in liquid culture of *Pseudomonas* bacteria

Before carrying out the bioremediation experiment of the herbicide atrazine by *Pseudomonas* bacteria, it was necessary to investigate the persistence of such compound in the liquid culture of *Pseudomonas* sp. Data concerning the atrazine biodegradation in such liquid culture are tabulated in **Table (3).** The results showed that a considerable dissipation of atrazine during the first week occurred reduced its concentration to 0.23 and 6.54 ppm from the initial applied concentrations, 1 and 10 ppm, respectively.

Table 3. Bioremediation of atrazine herbicide with the bacteria *Pseudomonas* sp (10^6cfu).

Days after treatment	Atrazin residues			
	1 ppm	% Dissipation	10 ppm	% Dissipation
0	1	0	10	0
1	0.87	13	9.42	5.8
2	0.69	31	9.01	9.9
4	0.48	52	8.79	12.1
8	0.23	77	6.54	34.6
10	0.19	81	4.97	50.3
15	0	100	2.05	79.5
30	0	100	0.78	92.2
45	0	100	0	100

In addition, data in **Table (4)** and **Figure (1)** indicate the degradation regression line of atrazine as affected by the liquid culture of *Pseudomonas* sp. Such data revealed that the time intervals for 50 % dissipation, i.e. the half-life periods ($t_{1/2}$) were estimated to be 4.95 and 19.02 days. Also, the tenth -life periods ($t_{1/10}$) were calculated to be 13.12 and 28.73 days with 1 and 10 ppm atrazine, respectively. In addition, it was observed that the dissipation, rate of atrazine was slightly lower at the lowest rate of application (1 ppm).

Table 4. Statistical and mathematical analysis of the of atrazine residues by the bacteria *Pseudomonas* sp.

Parameters*	Concentration Applied	
	1 ppm	10 ppm
Intercept of degrada-	0.1233	1.0664
Slope (b)	-0.0865	-0.0719
Degradation rate (k)	0.282	2.37×10^{-4}
Coefficient of de-	0.869	0.9357
Half-life ($t_{1/2}$ days)	4.95	19.02
Tenth-life ($t_{1/10}$ days)	13.12	28.73

* Source: Timme and Frehse, 1980

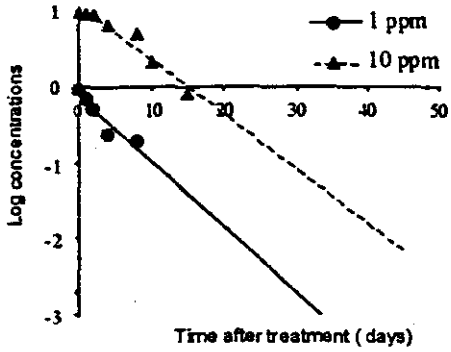

Fig. 1. Degradation regression lines of atrazine by liquid culture of *Pseudomonas* sp.

In other words, the detected concentrations were 0.19 ppm and 0.78 ppm after 10 and 30 days of application of 1 and 10 ppm. Furthermore, no atrazine residues were detected after 15 days when it had been applied at 1 ppm while in case 10 ppm, no residues were detected till the end of the experiment (45 days).

However the observed high dissipation rate of atrazine in liquid culture of *Pseudomonas* sp, may be attributed to dehalogenation and hydroxylation reactions by the bacterial enzymes [dioxygenases (C120)], amidohydrolase and chlorohydrolase as mentioned by **Hinteregger *et al* (1992) and Rodosevich & Tuovinen (2004)** who showed that such enzymatic system permit bacteria to use such compounds as a source of carbon and nitrogen for nutrition. In this respect, similar findings were obtained by **Topp (2001) and Dec & Bollag (2001).**

On the other hand, when atrazine was added and incubated with the liquid culture of *Pseudomonas* sp. at rates of 1 and 10 ppm during 2, 4, 15, 30 and 45 days, it was observed that the remained concentration of such compound was decreased. Such reduction in atrazine concentration may be due to the biodecomposition of C = N groups in its aromatic ring. Such interpretation was supported by the finding of **Radosevich *et al* (1997).** Conversely, **Weidner (1974)** found that atrazine was highly persistent. He observed that ^{14}C – atrazine was stable in aerobic water samples incubated for 15 months at 25°C in dark without adding the *Pseudomonas* bacteria. Similar results were observed by **Warnock & Leary (1978) and Belluck *et al* (1991).**

However, several studies revealed that the principal reactions in atrazine metabolism by *Pseudomonous* sp. involved a dealkylation at C–4 and C–6 position of the molecule. There is also some evidence of dechlorination at the C–2 as mentioned by **Bakke *et al* (1972).** In addition, **Wolf and Martin (1975)** reported the possibility of the microbial docomposition of ring [^{14}C] atrazine. Also,

Hauswirth (1988) indicated that dechlorination of atrazine ring and *N*–dealkylation are the major metabolic pathways while oxidation of the alkyl substituent appears to be minor and secondary metabolic route. Such interpretation was supported by **De-souza *et al* (1997) and Dec & Bollag (2001).**

2.3. Persistence and biodegradation of atrazine in liquid culture of *Trichoderma viride* fungus

Data of atrazine persistence in the liquid culture of *Trichoderma viride*, expressed as percent of the detectable initial concentrations of atrazine are listed in **Table (5)**. The obtained results show a considerable dissipation of atrazine during the first week, which amounted to 0.42 to 7.34 ppm with 1 and 10 ppm, respectively. Such disappearance was estimated to be 58%and 26.6%, respectively.

Table 5. Bioremediation of atrazine herbicide in the fungi liquid culture of *Trichoderma viride*.

Days after treatment	Atrazin residues			
	1 ppm	% Dissipation	10 ppm	% Dissipation
0	1	0	10	0
1	0.96	4	9.8	2
2	0.92	8	9.11	8.9
4	0.73	27	8.82	11.8
8	0.42	58	7.34	26.6
10	0.35	65	7	30
15	0.187	81.3	5.01	49.9
30	0.059	94.1	3.67	63.3
45	0	100	1.02	89.8

As shown in **Table (6)** and **Figure (2)**, the time intervals for 50% dissipation, i.e. the half-life periods ($t_{1/2}$) for such herbicide were 7.08 and 64.9 days, while the tenth-life periods ($t_{1/10}$) were determined to be 23.37 and 99.25 days of the applied atrazine concentrations, i.e. 1 and 10 ppm, respectively. As expected, it was observed that disappearance rate of atrazine was slightly lower at the highest rate of application (10 ppm). The detected amount was 1.02 ppm of the applied concentration at the end of the experiment (45 days), whereas no amount was detectable at the same interval when it had been applied at the lower concentration (1 ppm). However, the detected amounts of atrazine indicate that the dissipation was higher in the case of higher concentrations of application. The rapid disappearance of atrazine in *Trichoderma viride* culture may be attributed to biochemical hydrolysis, i.e. hydrolytic enzymes produced by such fungus to use this compound as a source of carbon and nitrogen for nutrition.

Table 6. Statistical and mathematical analysis of the degradation of atrazine residues by the fungus *Trichoderma viride*.

Parameters*	Concentration Applied	
	1 ppm	10 ppm
Intercept of degradation line (A)	0.0028	1.0249
Slope (b)	-0.0429	-0.0204
Degradation rate (k)	0.141	0.06737
Coefficient of determination (r^2)	0.9872	0.9567
Half-life ($t_{1/2}$ days)	7.08	64.9
Tenth-life ($t_{1/10}$ days)	23.37	99.25

* Source: Timme and Frehse (1980)

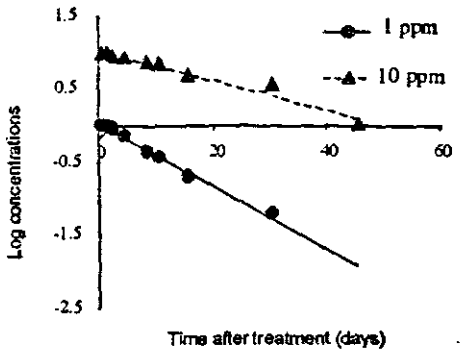

Fig. 2. Degradation regression lines of atrazine herbicide by the fungus *Trichoderma viride*.

Similar results were obtained by several investigators using other fungi in biodegradation of triazine (**Bortels *et al* 1967 and Kaufman & Kearney, 1970**).

On the other hand, the biodecomposition of atrazine as a result of its addition to the liquid culture of *Trichoderma viride* at rates of 1 and 10 ppm was investigated 2, 4 , 8 ,15 , 30, 45 days after incubation. According to the results obtained from the degradation regression lines of atrazine (**Figure 3**) and their statistical and mathematical analysis (**Table 6**), it is clear that the fungus *Trichoderma viride* exhibited its ability to bioremediate the tested concentration of atrazine, i.e. 1 and 10 ppm. Similar results obtained by other researchers reported that the capacity of such fungi to degrade atrazine might be due to its ability to dealkylate the alkyl group from its bound with nitrogen atom (N-dealkylation). Such interpretation was supported by the results obtained by the chromatographical analysis of labeled atrazine to prove that more than 97% of applied atrazine (5 ppm) was metabolized to two metabolites, i.e. 2-chloro-4- amino-6- isopropylamino-s-triazine and 2-chloro-4-ethylamino-6-amino-triazine during a period of 32 days by the biodegradadtion effect of *Trichoderma viride* (**Kaufman and Blake, 1970**).

To compare the biodegradation efficiency of atrazine by the bacteria *Pseudomonas* sp. and the fungus *Trichoderma viride*, some estimated parameters related with the statistical and mathematical analysis of the plotted degradation regression lines of atrazine were used. From such parameters, i.e. half-life ($t_{1/2}$) data indicated that bioremediation of atrazine by the bacteria *Pseudomonas* exhibited higher degradation efficiency 4.95 and 19.02 days than that recorded by the fungus *Trichoderma viride* 7.08 and 64.90 days after adding 1 and 10 ppm atrazine, respectively. Also, the determined tenth-life ($t_{1/10}$) showed the same trend related to the higher efficacy of the bacteria than the fungus to biodegrade the applied atrazine. Such parameter was 13.12 and 28.73 days with the bacteria while the same parameter showed 23.37 and 99.25 days with the fungus in case of 1 and 10 ppm of atrazine, respectively. In addition, the higher bioremediation capacity of the bacteria *Pseudomonas* than the fungus *Trichoderma viride* was confirmed through the estimated slope values from the plotted degradation regression lines. Such values were (-0.086) and (-0.072) as obtained by the degradation line caused by the bacteria whereas the slope values were (-0.043) and (-0.020) as obtained by the tested fungi. Such differences and/or high slope values of the regression lines by the bacteria demonstrate the rapidity of the degradation capacity than that obtained by the tested fungi.

Actually, similar results were obtained by other investigators, i.e. Newcombe and Crowley (1999) who found that *Pseudomonas* sp. minerilized more than 90 % of atrazine during 35 days, while **Bordjiba *et al* (2001)** showed that atrazine demonstrated a slight inhibition effect on the growth of fungus *Trichoderma viride*. Such data are in contrast to that obtained by **Kaufman and Blake (1970)** who found that such fungus is capable to degrade the herbicide atrazine during 32 days. However, it was demonstrated that the efficacy of the biodegradation of any chemical compound depends upon two main factors, i.e. biomass concentration and duration of biotreatment periods **(Fradette *et al* 1994)**. The low capacity of the fungus to biodegradae atrazine compared with the bacteria *Pseudomonas* may be due to the utilized and noninhibited fungal biomass in such study.

3. Phytoremediation of atrazine

In this study, the possibility to use two plants, i.e. giant reed *(Arundo donax* L.) and water hyacinth (*Eichhornia crassipes* Mart.) to phytoremdiate atrazine from water under open field conditions was investigated. Data concerning the detected amounts of atrazine residues in water after the phytoremediation by the mentioned plants are tabulated in **Table (7)**. The obtained results are expressed as percent of the initial concentration detected during the period of experiment. In terms of figures, during the first week, a considerable concentration of atrazine, i.e. 0.652 ppm which remained from 1 ppm as effect of water hyacinth and 0.624 ppm remained from 1 ppm as effect of giant reed. At the same time, when the two mentioned plants are planted together, the remained concentration of 1 ppm atrazine was 0.596 ppm. Such amounts resulting from the mentioned plants, i.e. water hyacinth, giant reed and both of them (water hyacinth + giant reed) demonstrated a dissipation percent of 34.8, 37.6 and 40.4%, respectively.

In addition, the estimated values in **Table (8)** and shown in **Figure (3)**, the time intervals for 50% dissipation, i.e. the half-life periods ($t_{1/2}$) were 7.026, 5.32 and 6.96 days for the water hyacinth giant reed and water hyacinth and giant reed, respectively. Also, the tenth -life periods($t_{1/10}$) were found 21.92, 13.09 and 19.49 days as effected by the tested plants and both of the two plants, water hyacinth and giant reed, respectively.

However, the dissipation rate of atrazine was found to be slightly higher in case of using water hyacinth and moderately with giant reed than that recorded by the two tested plants together (water hyacinth + giant reed). Such results are in agreement with those observed by **Rice (1997)** who reported that atrazine concentrations were significantly reduced in presence of *Ceratophyllum dermersum, Elodea Canadensis* and *Lemna minor* by 41.3 %, 53.2 % and 85 % of applied atrazine, respectively. In addition, the lower activity of the two tested plants when planted together to remove the herbicide atrazine than the water hyacinth alone may be due to the competition between the two plantes for the uptake through the rhizofiliration of the available atrazine molecules, which affect negativley the total absorption of the tested herbicide.

Table 7. Phytoremediation of atrazine herbicide in water by water hyacinth and gaint reed.

Days after treatment	Atrazin residues in water (ppm) as effect of the tested plants							
	water hyacinth+ gaint reed		water hyacinth		giant reed		control	
	l ppm	% Dissi-pation	l ppm	% Dissi-pation	l ppm	% Dissi-pation	l ppm	% Dissi-pation
0	1	0	1	0	1	0	1	0
1	0.921	7.9	0.972	2.8	0.944	5 6	1	0
2	0.873	12.7	0.953	4.7	0.913	8.7	1	0
4	0.733	26.7	0.798	20.2	0.795	20.5	1	0
8	0.596	40.4	0.652	34.8	0.624	37.6	0.966	3.4
10	0.225	77.5	0.441	55.9	0.497	50.3	0.961	3.9
15	0	100	0.033	67	0.113	87	0.923	7.7
30	0	100	0	100	0	100	0.875	12.5
45	0	100	0	100	0	100	0.844	15.6

Table 8. Statistical and mathematical analysis of phytoremediation of atrazine (1ppm) as affected by teasted plants.

Parameters*	Tested plants		
	water hyacinth + gaint reed	water hyacinth	gaint reed
Intercept of degradation line (A)	0.0285	0.1774	0.0876
Slope (b)	0.0469-	0.0899-	0.0558 -
Degradation rate (k)	0.15478	0.29669	0.18415
Coefficient of determination (r^2)	0.96	0.7677	0.8459
Half-life ($t_{1/2}$ days)	7.026	5.32	6.96
Tenth-life ($t_{1/10}$ days)	21.92	13.09	19.49

* Source: **Timme and Frehse, 1980.**

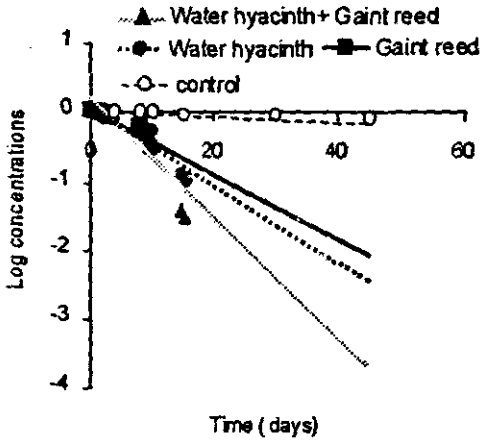

Fig. 3. Degradation regression lines of atrazine herbicide showing the effect of the tested plants, gaint reed, water hyacinth and gaint reed + water hyacinth comparing with the control.

Also, the obtained results were supported by the finding of Wilson *et al* (1999) and Xia – Huilong (2002) who studied the use of interaction between *Canna hybrida* and the herbicide simazine, water hyacinth with ethion, dicofol and cyhalothrin, respectively. Moreover, the reduction of pesticide residues was discussed by **Dietz and Schnook (2001)** who reported that phytoremediation is governed by physico-chemical properties of the compounds and moderately hydrophilic chemicals are most likely to be bioavailable to rooted and vascular plants. In addition, it was reported that some hydrophilic compounds may also be taken by plants via hydrogen bonding with transpiration water and organic chemicals that pass through plant cellular membranes and translocated to stem and leaf tissues. These sites that may be converted (e. g. oxidized by cytochrome P-450), conjugated by glutathione molecules or amino acids and/or compartmentalized in plant tissues as bound residues **(Dietz and Schnook 2001 and Yamada *et al* 2002).**

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المؤتمر العاشر لبحوث التنمية الزراعية، كلية الزراعة، جامعة عين شمس، القاهرة، مصر، ٢٠٠٦
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المعالجة الحيوية-النباتية لإزالة تلوث المياه بمتبقيات الأترازين

[١٩]

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١. قسم وقاية النبات- كلية الزراعة- جامعة عين شمس- شبرا الخيمة- القاهرة- مصر

أجرى هذا البحث بغرض دراسة كفاءة المعالجة الحيوية باستخدام نوعين من الكائنات الدقيقة هما بكتريا (*Pseudomonas flourescens*) وفطر (*Trichoderma viride*)، والمعالجة النباتية باستخدام نوعين من النباتات ورد النيل (*Eichornia crassipes*) و البوص (*Arundo donax*) فى تقليل تلوث عينات المياه المجمعة من محافظة المنوفية بمتبقيات مبيد الحشائش أترازين. دلت النتائج المتحصل عليها تحت الظروف المعملية أن البكتريا كانت أكثر فاعلية من الفطر وأكد على ذلك نتائج التحليل الإحصائى والرياضى لخطوط الهدم للتركيز المختبر من المتبقى (١ جزء فى المليون) والتى تشير إلى أن فترة نصف العمر ($t_{1/2}$) كانت ٤,٩٥ و ٧,٠٨ يوماً، وأن قيم الميل كانت (- ٠,٠٨٦٥) و (-٠,٠٤٤٩) لكل من البكتريا والفطر، على الترتيب. وعلاوة على ذلك، فإن نتائج المعالجة النباتية لمتبقيات المبيد فى الماء تحت الظروف الحقلية تشير إلى أن معدل إختفاء المبيد كان أعلى قليلاً فى حالة إستخدام نبات ورد النيل ومتوسطاً باستخدام البوص وذلك عنه بمعدل الإختفاء المسجل باستخدام كلا النباتين معاً (ورد

النيل + البوص). وقد بلغت قيم نصف العمر ٥,٣٢، ٦,٩٦، ٧,٠٢٦ يوماً بينما كانت قيم الميل (-٠,٠٨٩٩)، (-٠,٠٥٥٨) و (-٠,٠٤٦٩) على الترتيب باستخدام ورد النيل، البوص و ورد النيل+ البوص.

وتؤكد النتائج المتحصل عليها على فائدة كل من البكتريا والفطر والنباتات المختبرة فى إزالة تلوث المياه بمتبقيات مبيد أترازين.

الكلمات الدالة: المعالجة الحيوية والنباتية، متبقيات، أترازين، مياه

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