

RESPONSE OF HAWTHORN TRANSPLANTS TO BIOFERTILIZERS AND POULTRY MANURE

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ABSTRACT

This study was conducted in lath house, Dept. of Horticulture and Landscape Gardening, Coll. of Agricultural Engineering Sciences – Univ. of Baghdad during 2017 / 2018 growing seasons to investigate the influence of biofertilizers and poultry manure on 2 year's old trees of hawthorn transplants. This study included two treatments: four types of biofertilizers control (B₁), *Azospirillum brasilense* (B₂), *Bacillus megatherium* (B₃) and *A. brasilense* + *B. megatherium* (B₄) and three levels of poultry manure , 0 (M₀), 250g.tree⁻¹ (M₂₅₀) and 500g.tree⁻¹ (M₅₀₀) and their interactions. Treatments were replicated four times at factorial experiment in a RCBD. The experimental results showed that biofertilizers treatment (B₄) gave the highest leaves number of 72.42 and 77.83 leaf.plant⁻¹, highest leaf nitrogen content of 1.62 and 1.75 % and highest leaf IAA content of 34.41 and 38.85 µg g⁻¹ FW, for both seasons, respectively. Results also showed that poultry manure at levels 500g.tree⁻¹ (M₅₀₀) was superiority on control treatment and gave the highest leaves chlorophyll content of 59.19 and 59.96 SPAD units and highest leaf GA content of 139.46 and 140.76 µg. g⁻¹ FW, for both seasons, respectively.

Key words: fertilizers, organic manure, growth, leaves mineral, fruit trees.

الحديثي

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استجابة شتلات الزعرور للأسمدة الحيوية و مخلفات الدواجن

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مدرس

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المستخلص

أجريت هذه التجربة في الظلة التابعة لقسم البستنة و هندسة الحدائق / كلية علوم الهندسة الزراعية / جامعة بغداد للموسمين 2017 و 2018 لمعرفة تأثير اضافة الأسمدة الحيوية و مخلفات الدواجن في شتلات الزعرور بعمر 2 سنة. تضمنت التجربة عاملين، الأول هو اضافة اربعة انواع من الأسمدة الحيوية هي المقارنة (B₁) و اضافة *Azospirillum brasilense* (B₂) و اضافة *Bacillus megatherium* (B₃) و اضافة *A. brasilense* + *B. megatherium* (B₄) و اضافة ثلاثة مستويات من مخلفات الدواجن هي صفر (M₀)، 250 غم.شتلة⁻¹ (M₂₅₀)، 500 غم.شتلة⁻¹ (M₅₀₀) والتداخل فيما بينهما. صممت التجربة بتصميم التجارب العاملية ضمن تصميم RCBD وبأربعة مكررات. اظهرت نتائج التجربة ان الأسمدة الحيوية (B₄) اعطت اعلى عدد للأوراق والبالغ 72.42 و 77.83 ورقة، اعلى محتوى للأوراق من النتروجين والبالغ 1.62 و 1.75 % ومن IAA والبالغة 34.41 و 38.85 مايكروغرام.غرام⁻¹ ووزن طري لموسمي الدراسة، بالتتابع. كما اظهرت النتائج ايضاً تفوق المعاملة (M₅₀₀) على معاملة المقارنة و اعطت اعلى محتوى للأوراق من الكلوروفيل والبالغ 59.19 و 59.96 وحدة Spad و اعلى محتوى للأوراق GA والبالغة 139.46 و 140.76 مايكروغرام.غرام⁻¹ ووزن طري لموسمي الدراسة، بالتتابع.

الكلمات المفتاحية: الأسمدة ، سمد عضوي، النمو، العناصر في الأوراق، اشجار الفاكهة.

INTRODUCTION

Hawthorn, (*Crataegus azarolus* L.) is a fruit tree, belong to Rosaceae family. Medium-sized trees are taller than 8m. Its white flowers and spherical fruits contain 2-3 seeds. Hawthorn consists of about 100 species, are found in temperate regions in Central Europe. In Iraq, there are two types of Hawthorn in the wild *Crataegus azarolus* L. and *C. monigyna* L, in addition to other rare species *C. meyeri*, *C. pentagyna* and *C. netrophylla* (7). Hawthorn juice contains vitamin C, which is a favorite fruit for children. The fresh fruit contains citric acid, pectin and sugar (glucose, fructose). Their wood can be used in manufacture of furniture. Studies showed that hawthorn contains antioxidants (Procyandins and quercetin) , which fight against free radicals that cause cell membranes damage and genetic mutations, and can also cause cell death. Scientists consider these free radicals responsible for many events that affect the body with aging such as wrinkles, as well as many diseases such as cancer, cardiovascular disease (CVD) (18 and 22). As a result of increase chemical fertilizers in the recent years and the likelihood of negative effects on soil pollution, groundwater and atmosphere, and potential damage to human health , animal and microorganisms, as well as economic losses due to loss of chemical fertilizers. Scientists have sought alternative methods for chemical fertilization which are safer on human health and do not cause environmental pollution. An alternative is use of biotechnology to solve these problems. Biotechnologies include the use of organisms, part of a living organism, or organic and inorganic products from an organism used to perform a specific function (16). Biofertilizers are microbial fertilizers often increasing the biological activity of the soil. The most important function of these organisms is to convert atmospheric nitrogen into plant-usable forms, biodegradable bacterial fertilizers reducing the rate of chemical phosphate fertilization, reducing soil and environmental pollution as well as increasing production, quality, and increasing soil fertility (5). Several studies have been conducted to determine the role of biofertilizers in growth, leaf mineral and hormonal content, Nithya *et al*, 2011 (19)

mentioned that *Azotobacter* and *Aspergillus* spp caused a significant increase in vegetative growth on (pairedV-1) mulberry trees. Al-Hadethi, 2015 (3) , studied the effect of five of fertilizer sources (no application, 15 g.tree⁻¹ of Phosphorene biofertilizer, 15 g.tree⁻¹ of Nitrobeine biofertilizer , 15 g.tree⁻¹ of both Phosphorene + Nitrobeine and NPK fertilizer as recommended on apricot trees) and found Phosphorene + Nitrobeine and NPK caused significant increases in stem diameter, shoot growth , leaf chlorophyll content, leaf N, P, K contents and leaves endogenous hormones (IAA, GA₃ and Zeatin) in apricot trees. Abo AL-Mikh, (2) Recorded that, highest plant height, leaves number, leaves dry weight and leaves N, P, K, content it was in Wonderful pomegranate trees treated biofertilizers. Al-Hadethi *et al*, 2017 (4), found that the addition of bio-fertilizers to Peento peach transplants gave the highest leaves area, leaves chlorophyll content and increase in stem diameter. Organic fertilizers play a role in reducing the nitrogen loss during decomposition and facilitate the movement of nutrients to the roots to be absorbed and benefit from them, poultry manures have the most concentrated nutrient content compared to other animal manures, and are cheaper in cost (6). Poultry manures affect the growth of fruit trees, AL- Obaidi, (8) mentioned that the poultry manure applied with 4, 6 and 8 kg.tree⁻¹ caused a significant increase in vegetative growth, leaves dry weight and leaves mineral content compared to the control treatment from his study on apricot trees (cv. Zaini). Hamad and Abd, (14) found that poultry manure at 1, 2 and 3 kg.tree⁻¹ caused significant increases in leaves dry weight, chlorophyll content, leaves area and leaves potassium content for two pomegranate types Salimi and Wonderful. Kakehzadeh *et al*, (15) recorded that the highest increase in trunk diameter, annual shoot length and tree height was in the apple trees treated with poultry manure. Due to the absence of studies on hawthorn transplants and the effect of organic and bio fertilizers on their growth, the present study was conducted.

MATERIALS AND METHODS

This study was conducted in lath house, Department of Horticulture and Landscape

Gardening, College of Agricultural Engineering Sciences – University of Baghdad- Al-Jadriya during 2017 / 2018 growing seasons to investigate the influence of biofertilizers and poultry manure on 2 year's old trees of hawthorn transplants. This study included two treatments: four types of biofertilizers **control (B₁)**, *Azospirillum brasilense* (**B₂**), *Bacillus megatherium* (**B₃**) and *Azospirillum brasilense* + *Bacillus megatherium* (**B₄**) and three levels of poultry manure , **0 (M₀)**, **250g.tree⁻¹ (M₂₅₀)** and **500g.tree⁻¹ (M₅₀₀)** and their interactions. Treatments were replicated four times at factorial experiment in a RCBD. The number of trees used was 48 trees. The following parameters were determined in the two successive seasons:

1- Leaves number.

2- Stem diameter increase (mm): Stem diameter was measured using a (Vernier) at the beginning and end of the experiment, and calculating the difference between them for both seasons

3- Average of shoots length (cm): four branches were measured using metric tape from each experimental unit at the beginning and end of the experiment and calculating the difference between them for both seasons

4- Leaf chlorophyll contents (SPAD unit).

5- Leaf dry weight (%): Various leaves were taken from the trees was weighing then drained and calculated the percentage of dry matter by dividing weight after drying on weight before drying× 100

6- Leaf carbohydrates content (%): was determined according to Dubois *et.al*, (11).

7- Leaves mineral content: Leaves samples were collected for chemical analysis at the 2nd

week of June. Each sample consisted of 10 leaves. Tree⁻¹. Leaves were washed with tap water, rinsed with distilled water, and then dried at 70 c⁰ until a constant weight, ground and digested according (Chapman, and Pratt, 1978). Nitrogen was estimated by micro-kjeldahl method of (1). Phosphorus was estimated using a spectrophotometer by Estefan *et.al*, (12). Potassium was determined using Flame photometer according to (12).

8- Leaves hormonal content (IAA, gibberellins and zeatin) were assayed according to Ūnyayar *et al.*, (23).

The obtained results were subjected to analysis of variance according to Elsahookie and Wuhaib (12) using L.S.D 0.05 for comparing differences between various treatment means.

RESULTS AND DISCUSSIONS

Effects of biofertilizers and poultry manure on leaves number, stem diameter increase and shoots length: Data concerning the effect of treatments on leaves number, stem diameter increase and shoots length during the two experimental seasons are listed in (Table -1). Data indicated that, *Azospirillum brasilense* + *Bacillus megatherium* (**B₄**) treatment significantly increased leaves number of 72.42 and 77.83 leaf.plant⁻¹, stem diameter increase of 12.49 and 13.38 mm and shoots length of 15.50 and 17.42 cm for both seasons, respectively. Table (1) also revealed that the poultry manure at level 500g.tree⁻¹ (**M₅₀₀**) gave the highest leaves number of 74.61 and 80.06 leaf.plant⁻¹, stem diameter increase of 12.36 and 13.46 mm and shoot length of 15.15 and 17.30 cm for both seasons, respectively. Interaction treatment (**B₄M₅₀₀**) significantly affected in all studied traits in (Table -1).

Table 1. Effects of biofertilizers and poultry manure on leaves number, increased of stem diameter and shoots length of hawthorn transplants during 2017 and 2018 seasons

season Biofertilizers	2017				2018			
	0	Poultry manure (M)		mean	0	Poultry manure (M)		mean
		250	500			250	500	
	Leaves number							
B ₁	64.50	66.50	70.00	67.00	66.75	70.75	74.25	70.58
B ₂	66.00	72.25	76.75	71.67	69.00	79.50	81.75	76.75
B ₃	67.25	68.75	74.00	70.00	67.25	76.00	80.75	74.67
B ₄	67.75	71.75	77.75	72.42	70.50	80.00	83.50	77.83
mean	66.38	69.81	74.61		68.38	76.56	80.06	
LSD 0.05	M	B	M×B		M	B	M×B	
	1.81	2.09	3.62		2.04	2.36	4.08	
	Stem diameter (mm)							
B ₁	10.22	10.96	11.43	10.87	11.78	11.86	12.00	11.88
B ₂	10.90	11.75	11.92	11.52	11.96	12.11	12.73	12.27
B ₃	11.36	12.53	12.89	12.26	11.80	12.36	13.89	12.68
B ₄	11.40	12.87	13.19	12.49	12.03	12.88	15.22	13.38
mean	10.97	12.03	12.36		11.89	12.30	13.46	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.43	0.50	0.86		0.47	0.54	0.92	
	Shoots length (cm)							
B ₁	12.10	12.56	13.07	12.58	13.26	13.82	15.67	14.25
B ₂	14.21	14.90	15.93	15.01	15.00	16.29	17.78	16.36
B ₃	12.80	13.55	14.66	13.67	14.21	15.03	16.30	15.18
B ₄	14.51	15.06	16.94	15.50	15.72	17.11	19.44	17.42
mean	13.41	14.02	15.15		14.55	15.56	17.30	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.55	0.64	1.10		0.70	0.81	1.40	

Effects of biofertilizers and poultry manure on chlorophyll content, dry weight and carbohydrates content in leaves:

Results in (Table- 2) indicated that, *Azospirillum brasilense* + *Bacillus megatherium* (B₄) treatment significantly increased leaves chlorophyll content of 59.31 and 59.79 SPAD units and the highest leaves carbohydrates content of 9.68 and 10.48 % for both seasons, respectively. Leaves dry weight was not affected in the first year, while the same treatment (B₄) gave the highest leaves

dry weight of 33.45 % for the second year. Table (2) also shows that the poultry manure at level 500g.tree⁻¹ (M₅₀₀) was superior compared to control treatment and gave the highest leaves chlorophyll content of 59.19 and 59.96 SPAD units, leaves dry weight of 31.53 and 33.35 % and leaves carbohydrates content of 9.58 and 10.37 % for both seasons, respectively. Interactions treatment (B₄M₅₀₀) significantly had an effect on all studied traits in table (2).

Table 2. Effects of biofertilizers and poultry manure on chlorophyll content, dry weight and carbohydrates content in leaves of hawthorn transplants during 2017 and 2018 seasons

season Biofertilizers	2017				2018			
	0	Poultry manure (M)		mean	0	Poultry manure (M)		mean
		250	500			250	500	
	Leaf chlorophyll content (SPAD units)							
B ₁	53.37	55.18	56.92	55.16	53.91	55.70	57.48	55.70
B ₂	57.11	58.29	60.03	58.48	56.82	58.58	60.62	58.67
B ₃	53.48	56.00	58.48	55.99	54.94	57.11	59.54	57.20
B ₄	57.67	58.92	61.34	59.31	57.95	59.22	62.20	59.79
mean	55.41	57.10	59.19		55.91	57.65	59.96	
LSD 0.05	M	B	M×B		M	B	M×B	
	1.38	1.59	2.76		1.52	1.76	3.04	
	Leaves dry weight (%)							
B ₁	30.14	32.01	31.18	31.11	31.23	31.80	32.54	31.86
B ₂	31.58	31.13	30.87	31.19	32.29	32.86	33.76	32.97
B ₃	30.38	31.19	31.81	31.13	31.98	32.08	32.96	32.34
B ₄	31.38	30.17	32.24	31.26	32.77	33.45	34.14	33.45
mean	30.87	31.13	31.53		32.07	32.55	33.35	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.18	N.S	0.35		0.32	0.37	0.64	
	Leaves carbohydrates content (%)							
B ₁	8.38	8.93	8.85	8.72	8.44	9.02	9.44	8.97
B ₂	9.14	9.49	9.83	9.49	9.52	10.13	10.86	10.17
B ₃	8.56	9.02	9.44	9.01	8.94	9.67	10.04	9.55
B ₄	9.19	9.66	10.20	9.68	9.92	10.38	11.14	10.48
mean	8.82	9.28	9.58		9.21	9.80	10.37	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.24	0.28	0.48		0.36	0.42	0.71	

These results are attributed to the role of biofertilizers in improving soil biological and physical properties as well as the chemical properties resulting from the release of larger quantities of nutrients available for absorption by the roots and thus influence the physiological processes such as increasing the efficiency of photosynthesis in the leaves (24) and increase its output such as carbohydrates and thus increase vegetative growth. Also may be due to increase the microbial potential of biofertilizers and organic manure to produce plant growth regulators such as auxin, cytokines and gibberellins (Tables 4), which affect growth and increase uptake of soil nutrients (21). Generally, these results are in harmony with those reported by (17) on pear trees and (3) on apricot trees.

Effects of biofertilizers and poultry manure on leaves N, P, K content:

Data concerning the effect of treatments on leaves nitrogen, phosphor and potassium content are listed in Table (3). The data cleared that *Azospirillum brasilense* + *Bacillus megatherium* (B₄) treatment significantly increased and gave the highest leaf nitrogen and phosphor content, while biofertilizers treatments did not affect on leaf potassium content. Table (3) also shows that the poultry manure at level 500g.tree⁻¹ (M₅₀₀) significantly superiority of the control treatment and gave the highest leaf nitrogen and phosphor content. The interaction between biofertilizers and poultry manure treatments significantly affected especially at *Azospirillum brasilense* + *Bacillus megatherium* and poultry manure at level 500g.tree⁻¹ (B₄M₅₀₀) as it gave the highest leaf nitrogen content of 1.83 and 1.96 % and leaf phosphor content of 0.41 and 0.48 %, for both seasons respectively. Values in Table (3) showed that leaf potassium content was not affected by biofertilizers and poultry manure treatments.

Effects of biofertilizers and poultry manure on leaves IAA, GA₃ and Zeatin content:

Data concerning the effect of treatments on leaves IAA, GA and zeatin content are listed in Table (4). The data cleared that *Azospirillum brasilense* + *Bacillus megatherium* (B₄) treatment gave the, highest leaf IAA content of 34.41 and 38.85 µg g⁻¹ FW and highest leaf GA content of 133.22 and 135.86 µg g⁻¹ FW and highest leaf zeatin content of 35.87 and 36.96 µg g⁻¹ FW, for both seasons respectively. Table (4) also shows that poultry manure at levels 500g.tree⁻¹ (M₅₀₀) gave the highest leaf IAA content of 34.57 and 38.49 µg g⁻¹ FW and highest leaf GA content of 139.46 and µg g⁻¹ FW and highest leaf zeatin content of 35.99 and 36.63 µg g⁻¹ FW, for both seasons respectively. The interaction between biofertilizers and poultry manure significantly affected all studied parameters. The reason for these results is that the addition of bio and organic fertilizer to the soil has led to a greater concentration of these elements in the soil solution increasing its availability and thus increasing their transmission and thus increase the concentration of these elements in the leaves. It is also due to the increase leaves number and leaves chlorophyll content (Tables 1 and 2) resulting in the absorption of these elements to meet their vegetative needs (5). Also increase leaves mineral and hormonal content by adding organic fertilizer, especially poultry manure may be due to increased concentration in poultry manure and thus increases its concentration in leaves (20). These results are in harmony with those obtained by (4) who worked on biofertilizers in apricot and peach trees, (20) are found that the application of organic fertilizer gave the highest leaves mineral content, (9) who worked on organic fertilizers in peach trees.

Table 3. Effects of biofertilizers and poultry manure on leaves N,P,K content of hawthorn transplants during 2017 and 2018 seasons

season Biofertilizers	2017				2018			
	0	Poultry manure (M)		mean	0	Poultry manure (M)		mean
		250	500			250	500	
				N (%)				
B ₁	1.22	1.36	1.55	1.38	1.27	1.39	1.72	1.46
B ₂	1.37	1.51	1.70	1.53	1.43	1.66	1.84	1.64
B ₃	1.30	1.41	1.49	1.40	1.38	1.53	1.77	1.56
B ₄	1.44	1.59	1.83	1.62	1.55	1.74	1.96	1.75
mean	1.33	1.47	1.64		1.41	1.58	1.82	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.11	0.13	0.22		0.13	0.15	0.26	
				P (%)				
B ₁	0.18	0.20	0.21	0.20	0.20	0.22	0.28	0.23
B ₂	0.21	0.24	0.29	0.25	0.24	0.31	0.33	0.29
B ₃	0.23	0.31	0.40	0.31	0.29	0.41	0.46	0.39
B ₄	0.24	0.33	0.41	0.33	0.30	0.41	0.48	0.40
mean	0.22	0.27	0.33		0.26	0.34	0.39	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.06	0.07	0.12		0.05	0.06	0.10	
				K (%)				
B ₁	1.19	1.31	1.25	1.25	1.33	1.47	1.41	1.40
B ₂	1.23	1.29	1.25	1.26	1.40	1.45	1.37	1.41
B ₃	1.22	1.20	1.33	1.25	1.28	1.39	1.51	1.39
B ₄	1.26	1.21	1.28	1.25	1.49	1.32	1.40	1.40
mean	1.23	1.25	1.28		1.38	1.41	1.42	
LSD 0.05	M	B	M×B		M	B	M×B	
	N.S	N.S	N.S		N.S	N.S	N.S	

Table 4. Effects of biofertilizers and poultry manure on leaves IAA, GA and Zeatin content of hawthorn transplants during 2017 and 2018 seasons

season Biofertilizers	2017				2018			
	0	Poultry manure (M)		mean	0	Poultry manure (M)		mean
		250	500			250	500	
				Leaves IAA content (µg g ⁻¹ FW)				
B ₁	30.48	30.96	32.04	31.16	31.33	32.76	36.04	33.38
B ₂	31.18	33.15	35.48	33.27	33.51	35.97	39.06	36.18
B ₃	30.82	32.08	33.90	32.27	33.12	34.48	37.38	34.99
B ₄	32.56	33.80	36.86	34.41	36.16	38.92	41.48	38.85
mean	31.26	32.50	34.57		33.53	35.53	38.49	
LSD 0.05	M	B	M×B		M	B	M×B	
	1.13	1.30	2.26		1.69	1.95	3.38	
				Leaves GA content (µg g ⁻¹ FW)				
B ₁	102.70	109.86	121.19	111.25	107.27	112.82	126.22	115.26
B ₂	108.88	117.26	150.65	125.60	118.18	120.28	148.89	129.12
B ₃	105.67	112.42	133.36	117.15	116.28	126.20	137.39	126.62
B ₄	115.16	131.78	152.72	133.22	121.32	135.70	150.55	135.86
mean	108.10	117.83	139.46		115.76	123.75	140.76	
LSD 0.05	M	B	M×B		M	B	M×B	
	7.18	8.29	14.35		6.42	7.41	12.84	
				Leaves Zeatin content (µg g ⁻¹ FW)				
B ₁	32.81	33.32	33.94	33.36	32.42	33.08	34.22	33.24
B ₂	33.68	34.52	36.84	35.01	33.80	35.12	37.12	35.35
B ₃	33.20	33.90	35.08	34.06	32.96	33.62	35.98	34.19
B ₄	33.90	35.60	38.11	35.87	34.43	37.28	39.18	36.96
mean	33.40	34.34	35.99		33.40	34.78	36.63	
LSD 0.05	M	B	M×B		M	B	M×B	
	0.74	0.85	1.48		0.69	0.80	1.38	

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