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PLANT NUTRIENTS AS CONTROLLING OF CHOCOLATE SPOT, RUST DISEASES AND IMPROVING THE PERFORMANCE OF FABA BEAN

Morsy,S. M. A.* and A. A. Abd EL – Razek**
***Plant Path. Res. Inst., Agric. Center, Giza, Egypt.**
****Dep.of Crops, Fac. of Agric .Univ.of Tanta**

ABSTRACT

Fungal disease i.e.,chocolate spot (*Botrytis fabae* Sard.)and rust (*Uromyces vicia fabae* (Pers.) Schart) are the most destructive diseases of faba bean and causes considerable dramatic damage losses on faba bean yield worldwide Therefore, the use of plant nutrients (leaves fertilizers) i.e., Ascobein(As), Citreen(Ci) and Potassen (Po) to both diseases (chocolate spot and rust) in *Vicia fabae* ,gave evidence for controlling these diseases. Results showed that ,spraying faba bean plants with As+Ci+Po significantly decreased chocolate spot disease with 3.5% and 3.81% while ,rust was by decreased 5.2% and 5.77% for the two growing seasons 2010/2011 and 2011/2012.

Total chlorophyll reached to the maximum values when using Potassen (1.933mg/g f.w.) while ,As+Ci+Po as combination revealed the most superior effect on total phenolics compounds .All treatments were found to be most effective in increasing growth characters and yield of faba bean.

Keywords: Faba bean ,chocolate spot ,rust ,chemical composition ,plant nutrients ,growth characters.

INTRODUCTION

Faba bean (*Vicia fabae* L.) is one of the major pulse crops grown in Egypt and many countries.Its recorded history in Egypt returned back to 1580 .C (Abdalla,1989).Due to its high nutritive value in both energy and protein contents, it is a primary source of protein (Nassib *et al.*,1991).It is amulti–purpose crop that plays an important role in the socio-economic life of farming communities (Agegnehu & Fessehaie, 2006). In addation, it is an excellent candidate crop to provide nitrogen input into temperate agricultural systems, moreover, it makes a significant contribution to soil fertility restoration as a suitable rotation crop that fixes atmospheric nitrogen (Samuel *et al* .,2008).Diseases are among the important biotic constrains that limit the production of faba bean crop .chocolate spot (*Botrytis fabae* Sard.)and rust (*Uromyces vicia fabae* (pers.) Schart) are of the economically important diseases that damage the foliage ,limiting photosynthetic activity and reduce faba bean production (Awaad *et al.*,2005 and El-Bramawy & Abdul Wahid, 2005).

The disease appears as lesions on flowers, leaves and stems with oblong elliptical of reddish to chocolate brown colour , with darker margins that are fairly defined and often a concentric circular pattern.As lesions increase in number , generally on the upper leaf side ,they may remain small

expand or merge (Harrison ,1988)The spots led to harmful effects on growth , physiological activities and yield (Khaled *et al.*, 1995).

The infection by *Uromyces vicia faba*e first appear as minute, slightly raised ,white to cream colored spots on leaves and to a lesser extent on stems .As spots enlarge the epidermis ruptures, releasing mases of dark drown spores (urediospores)to form characteristic pustules (uredia). The pustules are often surrounded by aring of yellow tissue. On highly susceptible cultivars , rust can bulid up rapidly until most of leaves are covered with pustules .Severely infected leaves rapidly dry up and premature defoliation may occur (Benier *et al.*,1993).

In the middle and Near east, losses due to both diseases reached about 60-80%in case of chocolate spot disease (Dereje & Yaynu,2001 ,Herath *et al* , 2001 and Bouhassan *et al.*,2004)and 50-70% in case of rust disease (Stoddard&Herath,2001, Avila *et al.*, 2003) on susceptible cultivars.

The problem of adequately protecting plant against the two studied diseases by using fungicides has been complicated by the development of fungicidal resistance and/or adverse effect on growth an productivity of the host plant as well as on the accompanying microflora (Khaled *et al.*,1995).

Because of the hazards of fungicide on public health and environmental balance , it was hypothesized that bio, organic (plant nutrients) might reduce or nullify the negative effects of chocolate spot and rust on the growth photosynthesis pigments ,phenolic content and yield of faba bean. aimed to study the effect of plant nutrients on some pathological,morphological.physiological traits and productivity of faba bean .

MATERIALS AND METHODES

1-Field experiments:

The experiments were carried out at Itay EL-Baroud Agric. Res. St. In tow successive seasons , 2010/2011and 2011/2012.Faba bean seeds (*Vicia faba*) cv.Misr 1 were sown ,1st November, in 4m long rows and four rows represented a replicate .Each treatment was replicated three times in a complete randomized block design .Plant nutrients (leaves fertilizers) as Ascobein (As), Citreen(Ci) and Potsseen(Po) were used in the experiments.Plants were treated with the plant nutrients at rate of 4 ml,4 ml and 12 g/l,respectively. Plants were sprayed with tap water only served as check .All agricultural practices were carried out according to the recommendation of Ministry of Agriculture, Egypt.

Chocolate spot severity was recorded in 15 January and then every two weeks (a= 15 January, b= 31 January,c=16 Febrauary and d=2 March).

Rust severity was recrded on 20 Febrauary and then every two weeks (a=20 Febrauary, b = 5 March ,c = 20 March and d = 5 april)

Diseae assessment :

The plants developed from each assigned treatment were sprayed with Ascobein Strean and potassen three times with 15 day intervals beginning from 45 day after sowing using a hand atomizer till dripping The plants were rated for disease severity (D.S), the former as the presence or absence of

disease (percentage of infected leaves on the plant) and the latter as % of disease severity.

The disease severity (D.S) of chocolate spot disease was estimated at 55 and 75 days from sowing under natural infection using the Scale of Bernier *et al.*, (1993) as follow:

1=No disease symptoms or very small specks (highly resistance)
3=few small discrete lesions (resistance)
5=some coalesced lesions with some defoliation (moderate resistance).
7=large coalesced sporulating lesians,50% defoliation and some dead plant (susceptible).
9=Extensive lesions on leaves ,stems and pods ,severe defoliation ,heavy sporulation, stem girdling, blackening and death of more than 80% of plants (highly susceptible)

The severity of rust disease was recorded at 100 days from sowing according to the standard scale suggested by Bernier *et al* .,(1993)as follows:

1=No pustules or very small non sporulating flecks (highly resistant)
3=few scattered pustules covering less than 1%of leaf area ,and few or no pustules on stem (resistant).
5=pustules common on leaves covering some pustules on stem (moderately resistant).
7=pustules very common on leaves covering 4-8% leaf area, some defoliation and many pustules on stem (Susceptible).
8=Extensive pustules on leaves, petioles and stems covering 8-10%of leaf area, many dead leaves and severe defoliation (highly susceptible).

Percentage of chocolate spot and rust disease severity were calculated using the formula adapted by (Hanounik, 1986):

$$\text{Disease severity \%} = \frac{\sum(NPC \times CR)}{(NIP \times MSC)} \text{ X } 100$$

Where NPC=No. Of plants in each class rate
CR=Class rate
NIP=No. of infected plants.
MSC=Maximum severity class rate.

Leaf chlorophyll determination:

Leaf chlorophyll content was determined spectrophotometrically at 645mm and 663mm according to Grodzinsky and Grodzinsky(1973).

Chlorophyll (a) and (b) concentrations in mg/g leaves were calculated as follows:

Ch. a = [(12.7 O.D.663)-(2.69 O.D.645)]0.2
Ch. b = [(22.9 O.D.645)-(4.68 O.D.663)]0.2

Determination of total phenolic compounds:

Total phenolics were determined after 75 days from sowing in fresh leaves using of Folin–Ciocalteau reagent (Singlelon and Rossi, 1965). Samples (2g) were homogenized in 80 % aqueous ethanol at room temperature and centrifuged at 10000 rpm for 15 min under cooling. The residues were re-extracted twice with 80% ethanol and supematants were pooled ,put into evaporating dishes and evaporated to dryness at room temperature. Residues were dissolved in 5ml of distilled water. One –hundred

micro -liters of this extract were diluted to 3ml, with water and 0.5 ml of Folin-Ciocalteau reagent was added. After 3 min,2ml of 20%sodium carbonate was added and the contents were mixed thoroughly.

The developed color was measured spectrophotometrically at 650 nm after 60 min ,using catechol as a standard. The results were expressed as catechol/g fresh weight material.

Morphological characters and seed yield.

Samples were taken at 90 days from planting to estimate growth Parameter as: plant height, number of branches /plant, number of pods /plant and seed yield were recorded.

Statistical analysis:

All data were subjected to the proper statistical analysis of variance of randomized complete block design by Gomez and Gomez, (1984).Mean values of treatments were differentiated using LSD according to procedure outlined by Steel and Torrie, (1980).

RESULTS AND DISCUSSION

1) Disease assessment:

a. Chocolate spot.

Data of disease severity of chocolate spot disease of faba bean were recorded in Table (1) and Fig. (1 and 2). Generally disease severity was reduced and Ascobein, citreen and potassen alone or combination had the best effect. The severity increased in 16 February and 2 March (c and d) in the two growing seasons 2010 and 2011.It was found that in the first season (AS + Ci + Po) had the best effect chocolate spot severity to 3.5% and 3.81% in the two growing seasons 2010 and 2011, respectively.

Table (1): Effect of some plant nutrients as spray on the severity of chocolate spot disease on faba bean cv. Misr 1.

Plant nutrients	Chocolate spot severity% 2010				Mean	Chocolate spot severity % 2011				Mean
	A	B	C	D		A	B	C	D	
Ascobein (As)	2.56	2.15	4.1	9.04	4.46	3.04	3.54	6.32	9.23	4.96
Citreen (Ci)	2.2	2.25	4.6	8.57	4.41	3.01	2.65	6.1	8.48	5.06
Potassen (Po)	1.88	1.9	4.03	6.9	3.68	2.7	2.25	5.5	7.61	4.52
As + Ci	2.29	2.16	4.34	8.18	4.24	3.49	2.13	5.5	8.1	4.81
As + Po	2.04	1.95	4.3	7.06	3.84	2.87	2.15	4.73	7.5	4.31
Ci + Po	2.03	1.89	3.94	6.2	3.57	2.86	2.5	4.53	6.5	4.05
As + Ci + Po	1.95	1.95	2.99	7.1	3.5	2.59	2.08	4.1	6.45	3.81
Control (non treated)	3.33	3.18	5.65	11.28	5.86	4.21	4.8	8.75	12.75	7.63
L.S.D 0.05%	0.33	0.24	0.52	0.81		0.29	0.55	0.61	1.05	

Also, Ci + Po as a combination reduced chocolate spot by 3.57% and 4.05% for the two growing seasons. While, the highest disease severity 4.46% was obtained when applied Ascobein during season 2010 on the other hand Citreen 5.06% in growing season 2011,

compared with control which reached 5.86% and 7.63% in both growing seasons 2010 and 2011, respectively.

The role of plants nutrients effects of chocolate spot in faba bean plant may be due to the increase in chitinase activity or stimulation plant growth through increased cell division, as well as optimized uptakes of nutrients and water (Abd-El-Kareem, 2007 and Chen *et al.*, 2004).

Also El-Garhy (2002) studied that concerning the effect of bio-fertilizer, garlic and yeast extracts on chocolate spot disease, data showed that, garlic extract gave the lowest percentage of chocolate leaf spot 26.5% and 19>1% in Giza 843 while , bio-fertilizer gave reduction between about 16.3% to 6.6% during two growing seasons 1999/ 2000 and 2000 / 2001 respectively.

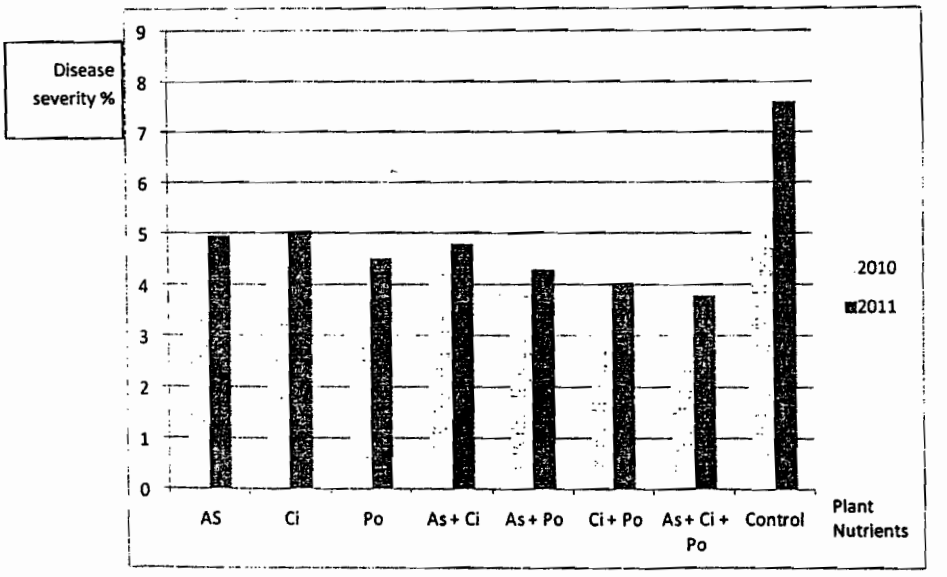

Fig. (1): Effect of some plant nutrients as spray on the severity chocolate spot disease on faba bean cv. Misr 1.

Data showed in Fig.(2). Indicated high reduction in faba bean chocolate leaf spot disease severity during the growing seasons, due to the application of plant nutrients.

The correlation analysis between chocolate spot disease severity % and months in the two growing seasons 2010 and 2011. Results indicated that it is a positive and significant by values of 0.922 and 0.913 compared to the control severity of 0.898 and 0.963 in the two successive seasons 2010 and 2011, respectively.

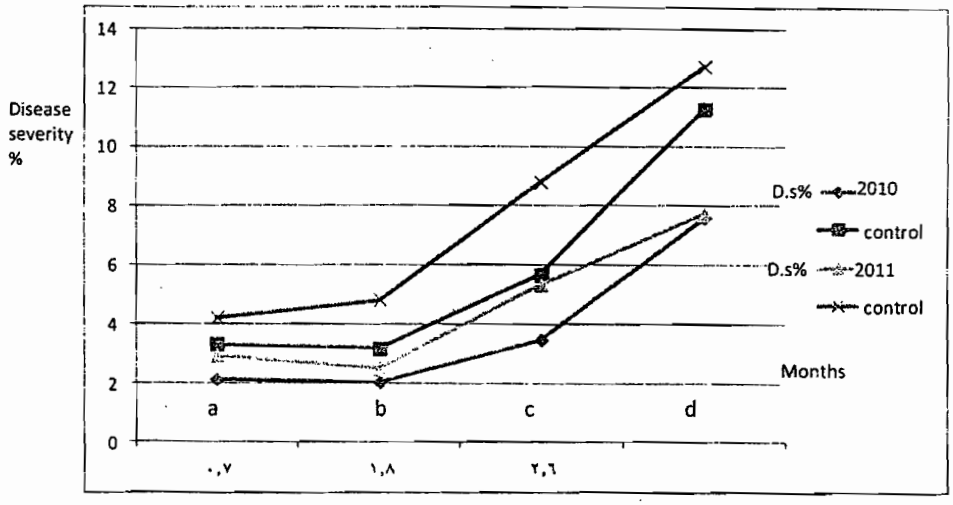

Fig. (2) :Effect of plant nutrients on chocolate spot disease of faba bean Miser 1 during the two growing seasons.

b. Faba bean rust disease severity :

Data presented in Table (2) and Fig (3 and 4) show that, spraying faba bean with AS, Ci and Po decreased significantly the D.s% of rust dsease. It is clear that the lowest D.s were 5.2% and 5.77% when plants treated with Ascobein, Citreen and potassen as a combination in the two growing seasons 2010 and 2011, respectively. Also, Po + Ci decreased D.S to 5.44% and 6.26% in contrast with the high percentage of D.S 7.67% and 7.68%when used A.s alone in the two seasons 2010 and 2011, respectively.

The plants nutrients containing microelements play roles in plant resistance by regulating the levels of auxin in plant tissues and activating the auxin oxidase system (Maschner, 1986). Also, lead to increase in total phenal, calcium content these materials protect plants against pathogen stress (Chowdhury, 2003).

The correlation analysis between rust disease severity% and months indicate that a positive and significant by values of 0.943 and 0.940 compared to the control severity of 0.982 and 0.966 in the two successive seasons 2010 and 2011, respectively.

Table (2) : Effect of some plant nutrients as spray on the rust disease severity of faba bean cv. Misr 1.

plants Nutrients	Rust severity% 2010				Mean	Rust severity % 2011				
	A	B	C	D		A	B	C	D	
Ascobein (As)	4.17	7.47	9.35	9.68	7.67	5.08	7.65	8.85	9.13	7.68
Citreen (Ci)	5.41	6.48	8.38	9.23	7.38	5.8	6.95	8.38	8.98	7.53
Potassen (Po)	342	4.21	6.75	8.33	5.68	4.38	6.7	6.85	7.48	6.35
As + Ci	3.93	5.55	8.53	9.18	6.8	5.3	7.48	9.0	8.3	7.52
As + Po	3.09	4.01	6.98	7.78	5.47	4.63	6.45	7.7	8.15	6.73
Ci + Po	3.13	4.38	6.7	7.55	5.44	3.85	6.1	7.6	7.5	6.26
As + Ci + Po	2.87	4.38	6.48	7.05	5.2	3.83	5.6	6.43	7.2	5.77
Control	6.66	8.28	10.93	11.73	9.48	7.78	10.13	11.03	11.9	10.22
L.S.D 0.05%	0.6	0.73	0.71	0.72		0.73	0.94	0.94	0.97	

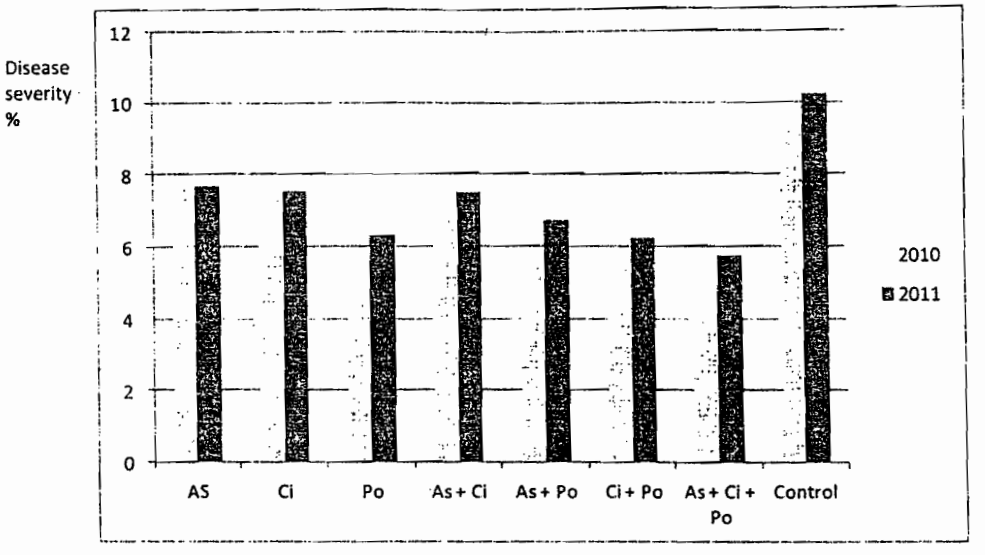

Fig. (3): Effect of some plant nutrients as spray on the severity rust disease of faba bean cv. Misr 1.

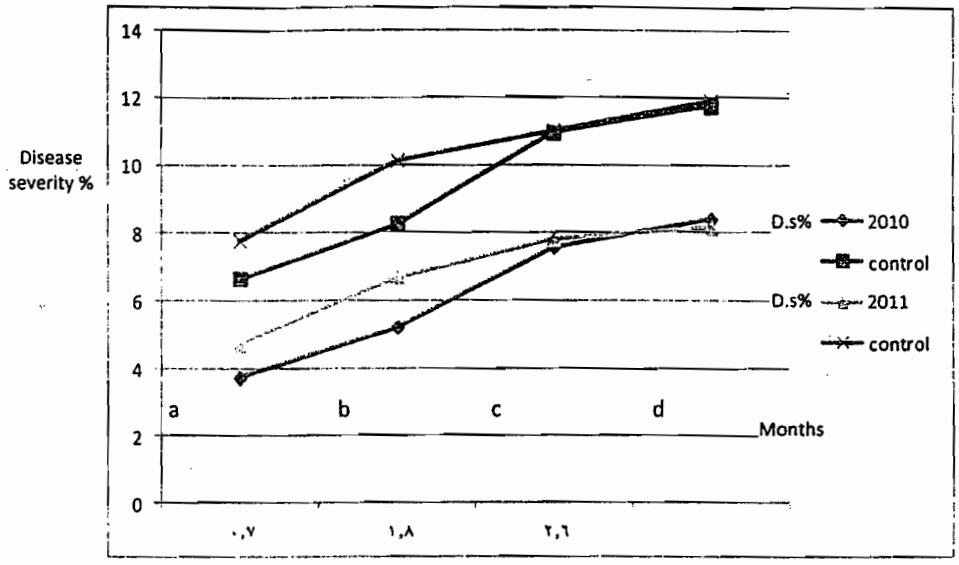

Fig.(4): Effect of plant nutrients on rust disease of faba bean Misr 1 during the two growing seasons.

2)- Effect of plants nutrients on the chlorophyll and phenolic contant of faba bean leaves:

a). Total chlorophyll contents:

The effect of the different plants nutrients on the chlorophyll content of leaves was significant Table (3). Po + Ci, and As + Po showed the highest concentrations of the chlorophyll (a) of leaves which were 0.84 and 0.823 mg/g leaves, respectively while, (As + Ci + Po), (As + Po) and (Po) showed the highest concentrations of the chlorophyll (b) of leaves which were 0.63, 0.623 and 0.613 mg/g leaves, respectively. The total chlorophyll content of leaves showed the highest concentrations in case of (As + Ci + Po) as a combination, compared with (As + Po) and (Po) alone which were 1.45, 1.446 and 1.433 mg/g leaves, respectively. These results are agreement with Ayman *et al.*, (2009) who found that in chlorophyll content, all treatments of Amino acid (AA) and humic acid (HA) gave significant increase. In this respect, highest increase was observed with the combination of HA (2000 ppm) + AA (2000 ppm), followed by HA (1000 ppm) + AA (1000 ppm),they added that fungicides applications had no significant effects on these parameters.

b)-Total phenolic contents:

Data Table (3) showed that the total phenolic contents of the faba bean leaves of treated and non-treated with plants nutrients were.

1. Total phenolics contents were increased in the leaves treated compared with non-treated by plant nutrients.
2. The highly contents 816.89 mg/100 g fresh weight were found when treated by AS + Ci + Po as a combination, followed by AS + Ci 769.33 mg/100 g fresh weight. While, the lowest total contents of phenolics were obtained 576.66 mg/100 g fresh weight when treated faba bean with Po alone.
3. Phenol play an important role in plant defense such phenols that are essential for biosynthesis of lignin which considered an important structural component of plant cell walls (Hahlbrock and Scheel , 1989).

Table (3): Effect of some plants nutrients as spray on the chlorophyll a,b, total content (mg /g leaves) and total phenolics in faba bean.

Plants nutrients	Chlorophyll (a)	Chlorophyll (b)	Total chlorophyll (a+b)	Total phenol (mg $100g^{-1}$ fresh weight)
Ascobein (As)	0.80	0.543	1.343	657.11
Citreen (Ci)	0.79	0.557	1.347	755.11
Potassen (Po)	0.82	0.613	1.933	576.66
As + Ci	0.78	0.54	1.32	769.33
As + Po	0.823	0.623	1.446	642.33
Ci + Po	0.84	0.577	1.417	618
As + Ci + Po	0.82	0.63	1.45	816.89
Control	0.76	0.507	1.267	546.45
L.S.D 0.05%	0.017	0.029	-	46.56

2) Effect of plants nutrients on some agronomic characters on faba bean cv. Misr1.

a. Plant height:

Data in Table (4) showed that significant effects on the height plant at the two growing seasons. In two growing season 2010/2011 As + Ci + Po as a combination were the most effective, followed by Ascobein alone. However Citreen alone and AS + Po as combination gave the lowest height plant 86.7 and 86.1 cm respectively. These results are in agreement with Bozorgi *et al.*, (2011) who reported that, the highest plant height was obtained with foliar zinc spraying.

b. Number of branches / plant

The highest number of branches / plant was obtained with Ascobien + potassen (3.6 branches / plant) while , Ascobien as alone gave lowest number of branches / plant (2.2) as average of two growing seasons 2010 and 2011.

c. Number of pods / plant :

Data presented in Table (4) showed that foliar applications with Ascobein, Citreen and Potassen as alone or combination significantly effect number of pods / plant. The highest No. of pods / plant were obtained by AS + Ci + Po as combination (17.8 pod / plant) followed by As + Po (17.15 pod/ plant) as average of two growing seasons. However, Ascobein gave the lowest No. of pods / plant (12.05) as compared with control (10.65 pods / plant).

d)-Seed yield /Faddan.

It is evident from Table (4) that spraying with As + Ci + Po showed the highest seeds yield/ faddan. While, the lowest values were recorded with spraying Ci + Po as average of two growing seasons 2010 and 2011.

Abraham *et al.*, 2009 found that growth promoting ability of coir pith based cyanobacterial (CB + CP) basal and foliar bio-fertilizer application on *Hibiscus esculents* was evaluated. The results indicated the presence of plant growth promoting substances in coir pith basal and foliar bio-fertilizer. In the trend Subramaniyan and Malliga, (2011) found that The effect of basa, spray and combined application of basal + spray of cyanopith biofertilizer on Zea mays were determined by analysis of the morphological, biochemical and yield of the control and test plants. The result showed that the combined application of Basal 25g + spray 0.4% of cyanopith biofertilizer significantly increase the morphological, biochemical parameters and yield.

Table (4): Effect of some plants nutrients on growth characters of faba bean cv. Misr 1 in the two growth seasons 2010 and 2011.

Plants nutrients	Plant Height (cm)		Mean	No. of branches / plant		Mean	No. of pods / plant		Mean	Yield / fed. (ton)		Mean
	2010	2011		2010	2011		2010	2011		2010	2011	
Ascobein (As)	95.5	99.7	97.6	2.2	2.4	2.3	12.1	12.0	12.05	3.52	2.41	3.47
Citreen (Ci)	85.2	88.2	86.7	3.2	2.3	2.75	14.2	14.9	14.55	3.4	3.10	3.25
Potassen (Po)	89.4	94.1	91.75	3.4	2.8	3.1	15.3	13.7	14.5	3.13	3.67	3.4
As + Ci	94.7	95.4	95.05	2.4	2.2	2.3	13.7	13.8	13.75	3.33	3.5	3.42
As + Po	87.5	84.7	86.1	3.9	3.3	3.6	17.0	17.3	17.15	3.08	3.2	3.14
Ci + Po	93.5	93.6	93.55	2.9	2.6	2.75	14.95	16.3	15.63	2.95	2.92	2.94
As + Ci + Po	104.8	102.2	103.4	3.5	3.3	3.4	17.5	18.1	17.8	3.77	3.6	3.69
Control	83.7	82.9	83.3	2.2	1.8	2.00	10.9	10.4	10.65	2.99	2.7	2.85
L.S.D 0.05%	7.4	5.62		0.77	0.62		3.32	2.26		0.24	0.30	

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تطبيق إستخدام المغذيات النباتية لمقاومة أمراض التبقع الشيكولاتى والصدأ وتحسين صفات الفول البلدى

صابر محمد على مرسى * و أسامة أحمد عبد الرازق **

***معهد بحوث أمراض البناتات – مركز البحوث الزراعية – الجيزة – القاهرة .**

****** قسم المحاصيل – كليه الزراعة – جامعه طنطا**

أجريت تجربة حقلية خلال موسمين (٢٠١٠/٢٠١١ ، ٢٠١١/٢٠١٢) بمحطة بحوث إيتاى البارود بهدف دراسة تاثير المغذيات النباتية (الأسمدة الورقية) على مرضى التبقع الشيكولاتى والصدأ فى الفول البلدى و تقديرصفات النمو .

أوضحت النتائج أن استخدام المغذيات النباتية مثل الأسكوبين والسترين والبوتاسين رشا على نباتات الفول البلدى اعطت نتائج معنويه فى مقاومة هذين المرضين حيث انخفضت نسبة الاصابه بمرض التبقع الشيكولاتى الى ٣,٥% ، ٣,٨١% بينما انخفضت الى ٥,٢% ، ٥,٧٧% فى حالة مرض الصدأ وذلك عند استخدام الثلاث مركبات معاً خلال موسمي النمو على التوالى .

قدر المحتوى الكلورفيللى أ ، ب والمحتوى الكلى حيث اظهرت النتائج ان اعلى قيمة كانت عند إستخدام البوتاسين (١,٩٣٣ مللى / جم وزن رطب) .

بينما اظهرت النتائج ان استخدام الاسكوبين والستريين و البوتاسين معا نتائج معنوية فى زيادة المحتوى الفينولى مقارنة بباقى المعاملات .

ثبت من البحث أن استخدام المركبات السابقة ذو تأثيراً ايجابيا على صفات النمو والمحصول للفول البلدى.

قام بتحكيم البحث

أ.د / سعد احمد المرسى	**كلية الزراعة – جامعة المنصورة**
أ.د / عبد الحميد محمد طرابيه	**كلية الزراعة – جامعة الاسكندريه**