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Controlling of *Alternaria* Leaf Spot Disease on Faba Bean Using some Growth Substances

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ABSTRACT

Experiments have been designed to determine the effect of growth substances, which included Indole Butyric Acid (IBA), Gibberellic acid (GA_3), Ethrel and Paclobutrazol (PBZ) at 50, 100 and 150 ppm in minimizing the damage of *Alternaria alternata* in faba bean compared to Tridex-80% fungicide. *In vitro*, results revealed that all concentration of PBZ and high Ethrel concentrate (150 ppm) completely inhibited the linear growth of *Alternaria alternata*. *In vivo*, all growth substances at any concentration reduced significantly severity and incidence of *Alternaria* leaf spot disease. The maximum reduction was recorded with PBZ followed by Ethrel. All photosynthetic pigments significantly increased by IBA, Ethrel and PBZ treatments. PBZ was more effective followed by Ethrel. Whilest, GA_3 and Tridex-80% decreased chlorophylls a and b. Total phenols and total soluble carbohydrates are reached to the maximum values when using Ethrel followed by PBZ. Plant height was increased significantly with growth promoter (IBA and GA_3), GA_3 was the most effective. On the other side, using of growth retardants (Ethrel and PBZ) increased significantly number of branches/plant, paclobutrazol (PBZ) came first. Yield components (pods number/plant and weight of 100 seed) increased significantly with any level of growth substances. The maximum values occurred under Ethrel followed by PBZ. This research clearly showed that there is no significant difference between the efficacy of growth substances (Ethrel and PBZ "at 150 ppm") and fungicide tridex-80% in reducing *Alternaria* leaf spot disease of faba bean plant.

Key words: *Alternaria* leaf spot, faba bean, growth substances

INTRODUCTION

Faba bean (*Vicia faba* L.), which has common names (broad bean, fava bean, horse bean and field bean) native to North Africa and Southwest Asia. It is grown mainly for its green pods and dried seeds, which are rich in protein, vitamins, carbohydrates, dietary fibers, minerals and secondary metabolites such as phenolics. Thus, it is an important component of the human nutritional consumption (Randhir *et al.*, 2002; Sahile *et al.*, 2011). Furthermore, it improves the soil fertility by fixing atmospheric nitrogen leads to reducing costs and minimizing impact on the environment. Therefore, increasing the plant crop production is one of the major targets of Egypt Agriculture policy (Mahmoud *et al.*, 2004; Bendahmane *et al.*, 2012).

Faba bean is suffering from many destructive diseases. It is attacked by more than 100 pathogens in the Mediterranean region. Diseases can inflict great losses in faba bean production. During the last years, *Alternaria* leaf spot disease caused by *Alternaria alternata* cause serious damage to the plant and decrease of yield production due to changing in temperature degrees (by increasing from natural values) in Egypt (Honda *et al.*, 2001; Reis *et al.*, 2007; Juroszek and von Tiedemann, 2011). The lesions first appear on the lower leaves as small, brown circular spots which

slowly increase in size and develop concentric brown rings with dark margins. Occasionally, an old rust pustule is found in the center of lesion, indicating that the leaf spot fungus may have entered through dead pustule tissue. *Alternaria* leaf spot generally occurs late in the season as plants start to mature (Bernier *et al.*, 1993).

It is an urgent need to apply alternative safe efficient methods against fungal plant diseases due to the hazard of fungicides on public health and environmental balance. It is well known that normal plant growth and development are partially controlled by chemical substance produced by the plant. Recently, attempts have been made to use growth regulator substances not only to increase yield but also to control or minimizing the damage and loss caused by plant pathogens. For this reasons, both growth promoters (IBA and GA₃) and growth retardants (Ethrel and PBZ) used as fungicides that control fungal plant diseases (Abdalla, 2001; Abd El-Hai, 2001; Khalifa, 2003; Metwally *et al.*, 2006; Abd El-Hai *et al.*, 2010).

The aim of this research is an attempt to study the effect of growth substances on some morphological, physiological traits and yield. In addition to reducing the negative effects of *Alternaria* leaf spot disease on faba bean.

MATERIALS AND METHODS

Plant material and growth substances: Faba bean seeds cv. Maser 1 were obtained from Legume Crops Research Depart, Agricultural Research Center, Giza, Egypt. Meanwhile, Indole butyric acid 99% ("IBA" Aldrich Chemical Company, England), Gibberellic acid 92% ("GA₃" Berelex tablets, Imperical Industries Plc, plant protection division "ICI", England), Ethrel (2-chloroethyl phosphoric acid) obtained from BDH chemicals LTD, U.K and Paclobutrazol (PBZ) "2RS-3RS-1-(4-chlorophenyl)-4,4-dimethyl -2-(1,2,4-triazol-1-yl)-pentan-3-01) as well as chemical fungicide (Tridex-80% wp "Mancozeb group") were used in this investigation.

Isolation of *Alternaria* leaf spot causal pathogen: *Alternaria alternata* was isolated from naturally infected faba bean leaves collected from different fields of Dakhlia Governorate. The isolated fungus was identified based on cultural and microscopic morphological characters according to the key given by Barnett and Hunter (1977).

***In vitro*:** This experiment aimed to study the effect of growth substances and Tridex-80 % on linear growth of *A. alternata*. The tested growth substances (IBA, GA₃, Ethrel and PBZ) at 50, 100 and 150 ppm as well as Tridex-80% at 2.5 g L⁻¹ were separately mixed with PDA before solidification, then poured in a sterile petri dishes. Four plates for each concentration were inoculated with fungal disc of *A. alternata* which had been cut from the periphery of 7 days old culture. Four plates inoculated with *A. alternata* only without any amendment served as control. The plates were incubated at 20°C, linear growth was observed daily and diameter of fungal colonies were recorded when particular control filled of petri dishes with fungal mycelia.

***In vivo*:** Experiments were carried out at Tag El-Ezz Agricultural Research Station, Dakahlia, Egypt under natural infection during 2013/2014 and 2014/2015 winter seasons. The experiments aimed to study the effects of foliar spray treatments of growth substances and Tridex-80% on reduce *Alternaria* leaf sot disease of faba bean and some physiological aspects, growth and yield. Seeds of faba bean were sown in 23 and 15 November in the first and second seasons, respectively. At the age of 50, 65 and 80 days in tow seasons, the plants were sprayed until dripping using small

pressure pump with one of the concentration of treatments. A complete randomized block design with three replicates was used in the two mentioned seasons.

Disease assessment: Disease Severity (DS) of *Alternaria* leaf spot was recorded at 90 days from sowing using the scale of Vakalunakis (1990), based on the leaf area infected (class rate from 0-4), then calculated as disease severity %, using the following formula:

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

Where:

NPC = No. of plants in each class rate

CR = Class rate

NIP = No. of infected plants

MSC = Maximum severity class rate

The Disease Incidence (DI) of *Alternaria* leaf spot as, a disease percentage was determined according to the following formula:

$$\text{Disease incidence} = \frac{\text{Number of infected leaflets}}{\text{Total number of tested leaflets}} \times 100$$

Physiological traits: After 80 days from sowing, the following physiological activities were determination:

- Photosynthetic pigments (chlorophyll a, b, total chlorophylls and carotenoids) were determined in the blade of terminal leaflet of third leaf from plant tip according to Mackinney (1941)
- Total phenolic compounds were determined in fresh shoot using the Folin-Ciocalteu reagent according to Malik and Singh (1980)
- Total soluble carbohydrate were extracted from 2 g shoot dry matter by ethanol 70% for overnight at laboratory temperature (Kayani *et al.*, 1990) then determined by the Anthron method (Sadasivam and Manickam, 1996)

Morphological characters and yield components: At harvest, plant height, number of branches/plant, pods number/plant and weight of 100 seed were recorded.

Statistical analysis: All data were subjected to statistical analysis of software CoStat (2005) using analysis of variance technique (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

In vitro

Antifungal activity of growth substances: Great variations in the linear growth of the pathogen were recorded with different types of growth substances (Fig. 1). PBZ at any concentration and high concentrate of Ethrel (150 ppm) completely inhibited the growth of

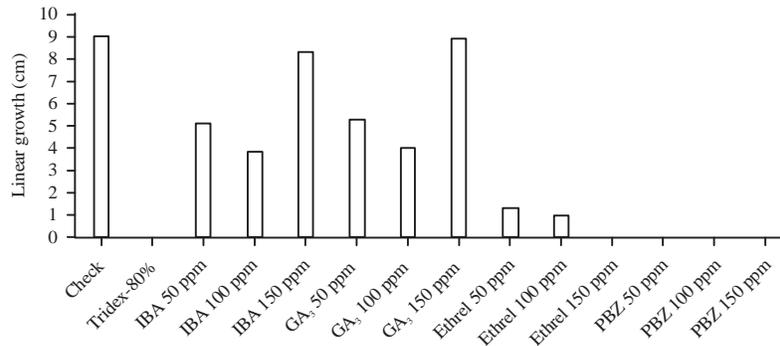


Fig. 1: Effect of growth substances on *Alternaria alternata* growth

Table 1: Effect of growth substances on disease severity and disease incidence of *Alternaria* leaf spot in faba bean plant

Treatments	2013/2014		2014/2015	
	Disease severity	Disease incidence	Disease severity	Disease incidence
Check	35.00 ^a	19.67 ^a	33.00 ^a	18.33 ^a
Tridex-80%	4.33 ⁱ	2.67 ^h	3.67 ^g	3.00 ^g
IBA (ppm)				
50	12.33 ^e	9.67 ^e	12.00 ^e	9.00 ^c
100	19.00 ^c	11.67 ^d	21.67 ^d	9.00 ^c
150	30.67 ^b	15.00 ^b	30.33 ^b	17.00 ^a
GA₃ (ppm)				
50	11.67 ^e	9.67 ^e	12.67 ^e	8.67 ^c
100	16.33 ^d	13.33 ^c	26.67 ^c	11.33 ^b
150	31.00 ^b	16.33 ^b	33.00 ^a	18.33 ^a
Ethrel (ppm)				
50	9.33 ^f	8.33 ^e	9.00 ^f	7.67 ^{cd}
100	7.00 ^{gh}	5.67 ^f	5.67 ^g	5.33 ^{ef}
150	6.00 ^{hi}	4.67 ^{fg}	5.67 ^g	4.00 ^{fg}
PBZ (ppm)				
50	8.67 ^{fg}	5.67 ^f	8.00 ^f	6.00 ^{de}
100	6.00 ^{hi}	4.00 ^{gh}	5.33 ^g	3.67 ^{fg}
150	5.00 ^{hi}	3.33 ^{gh}	4.00 ^g	3.33 ^g

Different letters within a column indicate significant difference at $p \leq 0.05$

Alternaria alternata. These results come online with this recorded on the fungicide Tridex-80%. While, the high level of IBA and GA₃ (150 ppm) had no significant effect in fungal growth. The inhibitory effect of growth retardants (Ethrel and PBZ) on fungal growth may be due to the inhibition of ergosterol biosynthesis and blocking C-14 demethylation reaction in fungi (Fletcher and Hofstra, 1990). Ergosterol is essential for maintenance of fungal membrane structure and without it the physical stability of membranes, as well as, the activity of membrane bound enzymes can be affected adversely (Brenneman and Murphy, 1991). Meanwhile, plant growth promoter (IBA and GA₃) increased both of pathogen growth and host growth (Al-Masri *et al.*, 2002). Moreover, Reddy and Strzelczy (1989) stated that GA₃ is growth stimulators for hyphae and stimulated spore germination of many pathogenic fungi.

In vivo

Disease assessment: Data of disease severity and disease incidence of *Alternaria* leaf spot of faba bean under field condition were recorded in Table 1. Application of growth substances at any concentration used and Tridex-80% significantly reduced *Alternaria* leaf spot DS and DI as

compared with check. Except for fungicide Tridex-80%, the maximum reduction of disease severity and incidence were recorded by PBZ at 150 ppm followed by 100 ppm then Ethrel at 150 ppm. While, IBA and GA₃ at 150 ppm came to late in both seasons. It is worthy to mention that, no significant differences among Tridex-80%, PBZ (100 or 150 ppm) and Ethrel 150 ppm.

Similar results were previously reported that using triazoles in controlling of potato early blight. PBZ increased epicuticular-wax which acts as protective waxy layer on the surface of the plant organs and decrease the size of stomatal openings (Fletcher and Hofstra, 1990). The outer thick cuticle of plant organs seem to be the barriers to inward movement of fungal mycelium (Singh *et al.*, 1980). Moreover, PBZ increased activity of antioxidants enzymes that protect cell organelles from oxidative damage (Sankhla *et al.*, 1992). In addition, PBZ increased Ca content that increase the rigidity of plant cell walls by complexing with wall matrix polysaccharides, which lead to be more resistant against fungal invasion (Abdalla, 2001). In addition to, the inhibitory effects growth substances on fungal diseases may be due to its effects on fungal growth, sporulation and sclerotial formation (Khalifa, 2003). Abd El-Hai *et al.* (2010) added that, Ethrel and IBA increased chlorophyll, total phenol and proline content. Proline and phenolics are used as indicator for controlling any stress conditions. Moreover, chlorophyll content is a good parameter reflecting the health condition of any plant. More, phenolic are well known as antifungal, antibacterial and antiviral. Gogoi *et al.* (2001) reported that, the first step of plant defence mechanism involves a rapid accumulation of phenols at the infection site, which restricts or slows the growth of the pathogen. With respect to Ethrel effects, Bovio *et al.* (1987) stated that, ethylene releasing from Ethrel induced plant resistance. Chowdhury (2003) found that treated plants with growth regulators lead to increase in total phenol, calcium content and chatechol oxidase activity, which protect plants against pathogen stress. Meanwhile, growth substances reduced the activity of pectolytic enzymes. The GA₃ decrease cell size that makes them denser, thus increasing resistance by forming a biological barrier (Ketykova *et al.*, 1985). In addition to, GA₃ at high concentration of gibberellic acid caused increasing in activity of lytic enzymes (Reddy and Strzelczy, 1989).

Physiological traits: Data in Table 2 show that all photosynthetic pigments (chl. a, b and carotenoids) increased significantly under application of IBA, Ethrel and PBZ. There are apposite

Table 2: Effect of growth substances on photosynthetic pigments (mg g⁻¹ fresh weight) of faba bean leaves

Treatments	2013/2014				2014/2015			
	Chlorophyll a	Chlorophyll b	Total chlorophyll	Carotenoids	Chlorophyll a	Chlorophyll b	Total chlorophyll	Carotenoids
Check	1.01 ^e	0.43 ^d	1.44 ^g	0.21 ^h	1.18 ^f	0.59 ^f	1.77 ^h	0.26 ^j
Tridex-80%	0.80 ^g	0.30 ^g	1.10 ^j	0.28 ^g	1.05 ^g	0.50 ^{gh}	1.54 ⁱ	0.32 ^{hi}
IBA (ppm)								
50	1.08 ^d	0.45 ^d	1.53 ^f	0.32 ^{ef}	1.24 ^e	0.63 ^e	1.87 ^g	0.35 ^{fg}
100	1.18 ^c	0.49 ^c	1.67 ^{cd}	0.36 ^d	1.33 ^d	0.68 ^d	2.01 ^e	0.36 ^{ef}
150	1.22 ^{bc}	0.51 ^c	1.72 ^c	0.40 ^{a-c}	1.38 ^c	0.72 ^c	2.10 ^{cd}	0.38 ^{de}
GA₃ (ppm)								
50	0.94 ^f	0.38 ^e	1.32 ^h	0.22 ^h	1.05 ^g	0.52 ^g	1.57 ⁱ	0.27 ^j
100	0.91 ^f	0.34 ^f	1.24 ⁱ	0.23 ^h	0.95 ^h	0.50 ^{gh}	1.45 ^j	0.30 ⁱ
150	0.73 ^h	0.30 ^g	1.03 ^k	0.26 ^g	0.91 ⁱ	0.47 ^h	1.38 ^k	0.32 ^{si}
Ethrel (ppm)								
50	1.10 ^d	0.49 ^c	1.59 ^{ef}	0.34 ^{de}	1.33 ^d	0.63 ^e	1.96 ^{ef}	0.34 ^{fh}
100	1.24 ^b	0.60 ^b	1.84 ^b	0.40 ^{bc}	1.44 ^b	0.71 ^{cd}	2.15 ^c	0.41 ^c
150	1.31 ^a	0.65 ^a	1.96 ^a	0.42 ^{ab}	1.53 ^a	0.81 ^a	2.35 ^a	0.49 ^a
PBZ (ppm)								
50	1.12 ^d	0.49 ^c	1.61 ^{de}	0.31 ^f	1.32 ^d	0.60 ^{ef}	1.92 ^{fg}	0.32 ^{si}
100	1.21 ^{bc}	0.52 ^c	1.73 ^c	0.39 ^c	1.41 ^{bc}	0.68 ^d	2.08 ^d	0.40 ^{cd}
150	1.31 ^a	0.61 ^b	1.92 ^a	0.42 ^a	1.50 ^a	0.76 ^b	2.26 ^b	0.46 ^b

Different letters within a column indicate significant difference at $p \leq 0.05$

relationship between concentrations and its effects. The PBZ followed by Ethrel was more effective in this respect. While, GA₃ at any concentrate and Tridex-80% decreased chl. a and b, but increased carotenoids content. The high level of GA₃ (150 ppm) was more effective.

Carotenoids are known as major endogenous plant antioxidants. In addition, chlorophyll is a good parameter of plant healthy. The reduction in chlorophylls (a and b) by GA₃ application may be due to destruction of pigments, reduced rate of synthesis, or dilution of pigments as a result of increased cell size and rate of cell expansion induced by GA₃. In addition, GA₃ induced chloroplast degradation and apparently interferes directly with chlorophyll metabolism (Straub and Lichtenthaler, 1973). On the other side, the increase in photosynthetic pigments due to Etherl may be explained by the stimulation of chlorophyll synthesis and the delay of chlorophyll destruction by increased cytokinin content in the leaves. Cytokinin causes an increase in the number of chloroplasts in the leaf by increasing both intensity of cell growth phytohormones and the activity of cytoplasm ribosome, thus chlorophyll synthesis is stimulated (El-Zawily *et al.*, 1994). Paclobutrazol also increased cytokinin content (Izumi *et al.*, 1988), which delaying chlorophyll destruction (Dalziel and Lawrence, 1984). Moreover, Fletcher and Arnold (1986) stated that the increase in photosynthetic pigments by PBZ may be due to a simulation of chlorophyll production due to higher level of cytokinins content, reduction in chlorophyll degradation and delaying the senescence.

Total phenols and total soluble carbohydrate in faba bean plants were recorded in Table 3. The application of growth substances at any concentrate increase significantly total phenols and total soluble carbohydrates compared with check. Ethrel came to first followed by PBZ then IBA, hence GA₃ came late. In contrast, fungicide Tridex-80% decreased significantly total soluble carbohydrate in faba bean shoot in both seasons. While, total phenols increased using fungicide treatment.

Phenols accumulation rapidly at the infection site for restricts or slows the growth of the pathogen due to its action as antioxidants, antimicrobial and photoreceptor (Lamba *et al.*, 2008). In our study, growth substances (IBA, Ethrel and PBZ) increased photosynthetic pigments consequently increased total soluble carbohydrates. In addition, Ethrel and PBZ increased cytokinin content in plant that accelerates the translocation of C-14 assimilated to storage parts

Table 3: Effect of growth substances on total phenol (mg/100 g fresh weight) and total soluble carbohydrate (mg g⁻¹ dry weight) of faba bean plant

Treatments	2013/2014		2014/2015	
	Total phenol	Total soluble carbohydrate	Total phenol	Total soluble carbohydrate
Check	410.00 ^k	82.50 ^{hi}	556.33 ^h	91.15 ^f
Tridex-80%	411.33 ^k	66.33 ^k	573.33 ^h	82.22 ^h
IBA (ppm)				
50	506.00 ⁱ	87.31 ^g	613.67 ^{f,h}	102.11 ^d
100	645.00 ^e	90.95 ^f	677.00 ^{d,f}	104.31 ^d
150	709.67 ^c	94.36 ^e	782.67 ^{a-c}	111.17 ^{bc}
GA₃ (ppm)				
50	476.00 ^j	85.37 ^{gh}	586.67 ^{gh}	94.29 ^f
100	576.33 ^h	81.47 ⁱ	657.00 ^{g-g}	87.14 ^g
150	515.67 ⁱ	74.41 ^j	592.67 ^{gh}	101.61 ^{de}
Ethrel (ppm)				
50	611.00 ^f	97.78 ^d	708.33 ^{c-e}	108.26 ^c
100	714.33 ^c	103.45 ^c	794.00 ^{ab}	120.05 ^a
150	787.67 ^a	112.35 ^a	846.67 ^a	98.69 ^e
PBZ (ppm)				
50	590.00 ^g	96.84 ^{de}	665.00 ^{d-g}	104.24 ^d
100	696.67 ^d	101.89 ^c	743.00 ^{b-d}	114.42 ^b
150	728.67 ^b	107.67 ^b	715.00 ^{b-e}	91.15 ^f

Different letters within a column indicate significant difference at p ≤ 0.05

Table 4: Effect of growth substances on some morphological characters of faba bean plant

Treatments	2013/2014		2014/2015	
	Plant height (cm)	No. branches/plant	Plant height (cm)	No. branches/plant
Check	115.67 ^f	4.00 ^{c-e}	117.33 ^{sh}	3.67 ^{ef}
Tridex-80%	119.67 ^{ef}	4.00 ^{c-e}	120.67 ^s	3.67 ^{ef}
IBA (ppm)				
50	124.33 ^e	4.00 ^{c-e}	126.33 ^f	3.67 ^{ef}
100	133.67 ^d	4.00 ^{c-e}	135.33 ^e	3.67 ^{ef}
150	141.67 ^c	3.67 ^{d-f}	147.67 ^c	3.00 ^f
GA₃ (ppm)				
50	135.00 ^d	3.67 ^{d-f}	141.00 ^d	3.67 ^{ef}
100	150.00 ^b	3.33 ^{ef}	154.67 ^b	3.33 ^{ef}
150	161.00 ^a	3.00 ^f	168.33 ^a	3.00 ^f
Ethrel (ppm)				
50	110.00 ^g	4.33 ^{cd}	112.67 ^h	4.00 ^{de}
100	104.00 ^h	4.67 ^{bc}	102.33 ⁱ	4.67 ^{cd}
150	93.33 ⁱ	5.33 ^{ab}	92.67 ^j	5.33 ^{bc}
PBZ (ppm)				
50	88.33 ^j	4.67 ^{bc}	85.67 ^k	5.00 ^c
100	77.33 ^k	5.33 ^{ab}	72.00 ^l	6.00 ^{ab}
150	71.00 ^l	6.00 ^a	68.33 ^l	6.33 ^a

Different letters within a column indicate significant difference at $p \leq 0.05$

(Mauk *et al.*, 1986). Moreover, Rashad (1998) reported that GA₃ increase the activity of amylase, invertase and sucrose synthesis. These enzymes play an important role in increasing hydrolysis and translocation of carbohydrate from shoot to root. Because the accumulation of carbohydrates in shoot may lead to inhibition of photosynthesis (Paul *et al.*, 1992).

Morphological characters and yield components: As shown in Table 3 and 4, spraying faba bean plants with growth promoter (Ethrel and GA₃) increased significantly plant height, while growth retardants (Ethrel and PBZ) inverse this. The highest values of plant height occurred under GA₃ at 150 ppm. In contrast, PBZ give the lowest values. In the other side, number of branches/plant increased significantly under the application of growth retardants (Ethrel and PBZ), PBZ was the most effective.

Concerning the effects of growth substances on number of pods and weight of 100-seed, data in Table 5 shows that number of pods/plant was increased significantly by any level of growth substances used. Ethrel give the maximum values followed by PBZ then GA₃. Moreover, weight of 100-seed was increased significantly under all growth substances used except, the low level of IBA (50 ppm) which showed no significant effect. PBZ give the highest values followed by Ethrel.

The stimulatory effect of GA₃ on plant height may be due to increase in cell division, number of internodes and/or elongation of each internode (Bruce, 1990; Abdel-All, 1994; Deotale *et al.*, 1998). Whilest, the suppression in plant height due to Ethrel my have been resulted from a reduction of cell elongation and/or through inhibition of cell division and elongation of sub apical meristems (Sen and Naik, 1977). Moreover, Anderson (1971) stated that the decrease in plant height and increase in number of branches due to Ethrel might be attributed to the liberation of ethylene. Ethylene is a natural growth regulator, which causes among many biological effects, inhibition of terminal bud growth and stimulation of lateral shoot development (Helaly *et al.*, 1984). In addition to, Ethrel decreasing auxin levels which in turn inhibition of both cell division and elongation (Ibrahim *et al.*, 1990). They added that the effect of GA₃ on plant height closely related to an increase in IAA. The increase in number of branches by the application of PBZ may be due to the role of PBZ on stimulation of lateral shoot development (Davis *et al.*, 1988).

Table 5: Effect of growth substances on yield components of faba bean plants

Treatments	2013/2014		2014/2015	
	No. of pods/plant	Weight of 100 seeds (g)	No. of pods/plant	Weight of 100 seeds (g)
Check	28.33 ^b	63.00 ^h	32.33 ^g	62.00 ^h
Tridex-80%	31.67 ^e	61.00 ^{hi}	34.33 ^{fg}	61.67 ^h
IBA (ppm)				
50	34.00 ^{fg}	62.67 ^{fi}	37.33 ^{ef}	64.00 ^g
100	36.67 ^f	63.67 ^{g-g}	39.33 ^e	65.33 ^{fg}
150	41.33 ^{de}	65.67 ^{de}	44.00 ^d	67.00 ^{de}
GA₃ (ppm)				
50	40.00 ^e	63.00 ^h	39.67 ^e	61.33 ^h
100	43.00 ^d	61.33 ^{g-i}	45.00 ^d	60.67 ^{hi}
150	48.00 ^c	60.33 ⁱ	51.33 ^{bc}	59.67 ⁱ
Ethrel (ppm)				
50	40.00 ^e	65.00 ^{d-f}	44.33 ^d	66.33 ^{ef}
100	48.00 ^c	67.00 ^{cd}	49.67 ^c	68.33 ^{cd}
150	56.33 ^a	69.33 ^{bc}	60.67 ^a	70.33 ^b
PBZ (ppm)				
50	40.00 ^e	65.67 ^{de}	43.33 ^d	67.67 ^{de}
100	47.67 ^c	70.00 ^{ab}	54.67 ^b	69.33 ^{bc}
150	53.33 ^b	72.33 ^a	53.00 ^{bc}	75.00 ^a

Different letters within a column indicate significant difference at $p \leq 0.05$

The GA₃ increased number of pods/plant due to increase in flowering period (Castro *et al.*, 1987). Moreover, the stimulating effect of Ethrel on number of pods/plant might be related to enhancing photosynthetic pigments formation in the leaves and cyclic photophosphorylation. Ethrel also stimulating axillary buds development giving rise to more lateral branches. The increase in seed weight due to Ethrel may be the result of a certain role on stimulating the formation of conducting elements allowing the passing of more nourishing materials to be accumulated in the developing seeds (Arafa, 1982). The PBZ and GA₃ increased number of inflorescences/plant, number of pods and seed number (Abd El-Hai, 2001), thus increased plant yield. Also, PBZ increased cytokinin content which delay chlorophyll destruction and senescences leading to increase plant yield (Izumi *et al.*, 1988).

It could be concluded that foliar spraying of PBZ, Ethrel and IBA at 150 ppm during the growth of faba bean is recommended for reducing *Alternaria* leaf spot disease and improving growth and yield.

REFERENCES

- Abd El-Hai, K.M., 2001. Physiological and pathological studies on soybean plants under different levels of salinity. Ph.D. Thesis, Faculty of Agriculture, Mansoura University, Egypt.
- Abd El-Hai, K.M., M.A. El-Metwally and S.M. El-Baz, 2010. Reduction of soybean root and stalk rots by growth substances under salt stress conditions. *Plant Pathol. J.*, 91: 149-161.
- Abdalla, M.E., 2001. Effect of paclobutrazol on growth, yield and anatomical structure of peanut plant infected with pod and root rot pathogens. *Phytopathology*, 91: S181-S181.
- Abdel-All, F.S., 1994. Effect of salinity and some growth regulators on growth and yield of *Phaseolus vulgaris* plant. M.Sc. Thesis, Faculty of Agriculture, Mansoura University.
- Al-Masri, M.I., M.S. Ali-Shtayeh, Y. Elad, A. Sharon, P. Tudzynski and R. Barakat, 2002. Effect of plant growth regulators on white mould (*Sclerotinia sclerotiorum*) on bean and cucumber. *J. Phytopathol.*, 150: 481-487.
- Anderson, A.S., 1971. Plant growth modification by 2-chloroethyl phosphonic acid (ethrel). II-Interaction between ethrel and gibberellins in the growth and fruiting of pea plants. *Horticult. Abstract*, 41: 6733-6733.

- Arafa, A.A., 1982. Growth structure, some physiological characters and yield of faba beans as affected by certain growth substances. *J. Agric. Sci. Mansoura Univ.*, 16: 1176-1188.
- Barnett, H.L. and B.B. Hunter, 1977. *Illustrated Genera of Imperfect Fungi*. Burgess Publishing Co., Minnesota, Pages: 241.
- Bendahmane, B.S., D. Mahiout, I.E. Benzohra and M.Y. Benkada, 2012. Antagonism of three *Trichoderma* species against *Botrytis fabae* and *B. cinerea*, the causal agents of chocolate spot of faba bean (*Vicia faba* L.) in Algeria. *World Applied Sci. J.*, 17: 278-283.
- Bernier, C.C., S.B. Hanounik, M.M. Hussein and H.A. Mohamed, 1993. Field manual of common Faba bean diseases in the Nile Valley. International Center for Agricultural Research in the Dry Areas (ICARDA). Information Bulletin No. 3.
- Bovio, M., I.A. Gentile and A. Matta, 1987. Release of ethylene following abiotic and biotic stimuli that induce resistance to *Fusarium* wilt of tomato. *Phytopathol. Medit.*, 26: 177-182.
- Brenneman, T.B. and A.P. Murphy, 1991. Activity of tebuconazole on *Cercosporidium personatum*, a foliar pathogen of peanut. *Plant Dis.*, 75: 699-703.
- Bruce, A.P., 1990. The use of plant growth regulators to enhance yield and production efficiency of soybean (*Glycine max* L. Merrill). *Dissertation Abstracts Int. B Sci. Eng.*, 51: 2678B-2678B.
- Castro, P.R.C., W. Dathe, R. Lopez and G. Sembdner, 1987. The effect of gibberellins GA3 on the flowering and fruiting of different soybean (*Glycine max*) cultivars. *Soybean Abstract*, 10: 505-505.
- Chowdhury, A.K., 2003. Control of *Sclerotium* blight of groundnut by plant growth substances. *Crop Res.*, 25: 355-359.
- CoStat, 2005. CoStat program, version 6.311. CoHort Software, Monterey, CA., USA., September 25, 2005.
- Dalziel, J. and D.K. Lawrence, 1984. Biochemical and Biological Effect of Kaurene Oxidase Inhibitors Such as Paclobutrazol. In: *Biochemical Aspects of Synthetic and Naturally Occurring Plant Growth Regulators*, Menhenett, R. and D.K. Lawrence (Eds.). British Plant Growth Regulator Group, New York, USA., ISBN-13: 9780906673096, pp: 43-57.
- Davis, T.D., G.L. Steefens and N. Sankhia, 1988. Triazole plant growth regulators. *Horticult. Rev.*, 10: 63-105.
- Deotale, R.D., V.G. Maske, N.V. Sorte, B.S. Chimurkar and A.Z. Yerne, 1998. Effect of GA3 and NAA on morphological parameters of soybean. *J. Soils Crops*, 8: 91-94.
- El-Zawily, A.I., S.A. Ibrahim and E. Zayed, 1994. Effect of ethrel and plant spacing on onion (*Allium cepa* L.). *J. Agric. Sci. Mansoura Univ.*, 9: 734-741.
- Fletcher, R.A. and V. Arnold, 1986. Stimulation of cytokinins and chlorophyll synthesis in cucumber cotyledons by triadimefon. *Physiol. Plant.*, 66: 197-201.
- Fletcher, R.A. and G. Hofstra, 1990. Improvement of uniconazole-induced protection in wheat seedlings. *J. Plant Growth Regulat.*, 9: 207-212.
- Gogoi, R., D.V. Singh and K.D. Srivastava, 2001. Phenols as a biochemical basis of resistance in wheat against karnal bunt. *Plant Pathol.*, 50: 470-476.
- Gomez, K.A. and A.A. Gomez, 1984. *Statistical Procedures for Agricultural Research*. 2nd Edn., John Wiley and Sons Inc., New York, USA., ISBN-13: 9780471879312, pp: 95-109.
- Helaly, M.N., A.M. Salama and A.A. Arafa, 1984. Effect of some growth substances on growth, certain physiological aspects and yield of fennel plants. *J. Agric. Sci. Mansoura Univ.*, 9: 262-278.
- Honda, Y., M.Z. Rahman, S.Z. Islam and N. Muroguchi, 2001. Leaf spot disease of broad bean caused by *Alternaria tenuissima* in Japan. *Plant Dis.*, 85: 95-95.

- Ibrahim, D.M., M.A. Khafagy and A.M. Abo El-Kheer, 1990. Some growth substances affecting the growth, chemical composition and alkaloidal content of *Lupinus termis* L. Egypt. J. Applied Sci., 5: 367-381.
- Izumi, K., S. Nakagawa, M. Kobayashi, H. Oshio, A. Sakurai and N. Takahashi, 1988. Levels of IAA, cytokinins, ABA and ethylene in rice plants as affected by a gibberellin biosynthesis inhibitor, Uniconazole-P. Plant Physiol., 29: 97-104.
- Juroszek, P. and A. von Tiedemann, 2011. Potential strategies and future requirements for plant disease management under a changing climate. Plant Pathol., 60: 100-112.
- Kayani, S.A., H.H. Naqvi and I.P. Ting, 1990. Salinity effects on germination and mobilization of reserves in jojoba seed. Crop Sci., 30: 704-708.
- Ketykova, E.K., D. Babaev and A.P. Chopanov, 1985. Research on gibberellins-like substances of the pathogen of *Fusarium wilt* and cotton under inoculation. Izvestiya Akademii-Nauk Turkmenskoi SSR, Biologicheskikh, Nuk, 5: 9-15.
- Khalifa, M.M.A., 2003. Pathological studies on charcoal rot disease of sesame. Ph.D. Thesis, Fac. Agric., Moshtohor, Zagazig Univ., Egypt.
- Lamba, P., S. Sharma, G.D. Munshi and S.K. Munshi, 2008. Biochemical changes in sunflower plants due to seed treatment/spray application with biocontrol agents. Phytoparasitica, 36: 388-399.
- Mackinney, G., 1941. Absorption of light by chlorophyll solutions. J. Biol. Chem., 104: 315-322.
- Mahmoud, Y.A.G., M.K.H. Ebrahim and M.M. Aly, 2004. Influence of plant extracts and microbioagents on physiological traits of faba bean infected with *Botrytis fabae*. J. Plant Biol., 47: 194-202.
- Malik, C.P. and M.B. Singh, 1980. Plant Enzymology and Histoenzymology. Kalyani Publishers, New Delhi, India, pp: 53.
- Mauk, C.S., M.G. Bausher and G. Yelenosky, 1986. Influence of growth regulator treatments on dry matter production, fruit abscission and ¹⁴C-assimilate partitioning in citrus. J. Plant Growth Regulat., 5: 111-120.
- Metwally, A.H., E.Y. Mahmoud, S.Y.M. Shokry and Z.N. Hussin, 2006. Effect of growth regulators in controlling of peanut root rot diseases and compared to fungicides treatment. J. Agric. Sci. Mansoura Univ., 31: 3537-3548.
- Paul, M.J., S.P. Driscoll and D.W. Lawlor, 1992. Sink-regulation of photosynthesis in relation to temperature in sunflower and rape. J. Exp. Bot., 43: 147-153.
- Randhir, R., P. Shetty and K. Shetty, 2002. L-DOPA and total phenolic stimulation in dark germinated fava bean in response to peptide and phytochemical elicitors. Process Biochem., 37: 1247-1256.
- Rashad, M.H., 1998. Influence of gibberellic acid and potassium interaction on some cabbage cultivars under salt stress. J. Agric. Sci. Mansoura Univ., 23: 1525-1540.
- Reddy, M.N. and E. Strzelczy, 1989. Influence of plant growth regulators on growth and virulence of *Rhizoctonia solani* (Kuhn) pathogenic to groundnut seedlings (*Arachis hypogaea* L. var. TMV 2). J. Phytopathol., 125: 187-191.
- Reis, R.F., T.F. de Almeida, E.S. Stuchi and A. de Goes, 2007. Susceptibility of citrus species to *Alternaria alternata*, the causal agent of the Alternaria brown spot. Scientia Horticulturae, 113: 336-342.
- Sadasivam, S. and A. Manickam, 1996. Biochemical Methods. 2nd Edn., New Age International (P) Ltd. Publishers, New Delhi, India.

- Sahile, S., P.K. Sakhuja, C. Fininsa and S. Ahmed, 2011. Potential antagonistic fungal species from Ethiopia for biological control of chocolate spot disease of faba bean. *Afr. Crop Sci. J.*, 19: 213-225.
- Sankhla, N., A. Upadhyaya, T.D. Davis and D. Sankhla, 1992. Hydrogen peroxide-scavenging enzymes and antioxidants in *Echinochloa frumentacea* as affected by triazole growth regulators. *Plant Growth Regul.*, 11: 441-443.
- Sen, S.K. and J. Naik, 1977. Growth and flowering response of pinched and unpinched chrysanthemum to growth regulators treatments. *Indian J. Horticult.*, 34: 86-90.
- Singh, D., S.B. Mathur and P. Neergaard, 1980. Histological studies of *Alternaria sesamicola* penetration in sesame seed. *Seed Sci. Technol.*, 8: 85-93.
- Straub, N. and H.K. Lichtenthaler, 1973. The effect of GA3 and kinetin on the formation of photosynthetic pigments in *Raphanus* seedlings. *Horticult. Abstract*, 44: 8534-8534.
- Vakalunakis, D.J., 1990. Host range of *Alternaria alternata* f. sp. *cucurbitae* causing leaf spot of cucumber. *Plant Dis.*, 84: 227-230.