

## **CONTROL OF CHOCOLATE SPOT DISEASE IN FABA BEAN.**

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### **ABSTRACT**

Chocolate spot disease caused by *Botrytis fabae* and *Botrytis cinerea*, is one of the most important diseases on faba bean in Egypt. *Botrytis fabae*, *Botrytis cinerea*, *Alternaria alternata* and *Stemphylium botryosum* were isolated from infected leaves of two faba bean cultivars (Giza 461 and Giza 3 cvs.) throughout two growing seasons (2004-2005 and 2005-2006). These fungi were varied between cultivars and years, also, were isolated more frequently from Giza 3 cultivar compared with Giza 461 cultivar in both seasons especially in the second season. The sensitivity of *B.fabae* and *B.cinerea* to five fungicides (Topsin-M, Rovral, Ferrocopper, Dithane M-45 and Kocide 101) were studied under laboratory conditions. The results showed that *B.fabae* was more sensitive to these fungicides than *B.cinerea*, where EC<sub>50</sub> values were 1.0 and 1.7, 1.3 and 1.8, 2.1 and 2.9, 5.8 and 9.0 and 66.0 and 96 ug a.i/ ml, respectively. The field trails showed that Topsin-M and Rovral were the most effective fungicides in reducing disease incidence, severity and finally increased yield, followed by Ferrocopper and Dithane M-45 and the least effective were Kocide 101 and bioagent (Plant-Guard). Efficiency of these compounds increased with increasing the rate of application. These fungicides gave good control of chocolate spot disease and improved plant growth and increased grain yield. Also, the results showed that Giza 461 cultivar was more resistant to this disease than Giza 3 cultivar.

### **INTRODUCTION**

Faba bean (*Vicia faba* L.) is considered the fifth important food legume crops in the world after dry bean, dry pea, Chickpea and Lentil (Adak *et al.*, 1998). The great majority of low and middle income classes people in Asia, Latin America, and the Mediterranean and Nile Valley regions depend on faba bean as a major source of proteinaceous food (FAO production Yearbook, 1981). In other parts of the world, however, faba bean is used mainly as an animal feed (Hanounik and Robertson, 1988). In Egypt, this crop is the first legume crop and considered the main source of protein for the majority of people, which consumed either as fresh green pods or as dry mature seeds for human, also grains and straw were used as animal feed (El-Afifi, 2003 and El-Gammal 2005).

Faba bean plants is liable to attack by a wide range of diseases such as leaf spots, downy mildew, wilt and root rot. Among these diseases, chocolate spot disease caused by *Botrytis fabae* and/or *Botrytis cinerea* is considered one of the most destructive diseases to the crop especially in the northern parts of Delta, where suitable conditions (low temperature and high relative humidity) favour its spread and severity (Mohamed, 1982; Abou-Zeid *et al.*, 1990; Abou Zeid *et al.*, 1998; Nasr, 2003 and El-Gammal, 2005). The yield loss due to this disease reached more than 50% of the total yield especially under favourable conditions (Mohamed, 1982). Two severe epidemic of this disease were recorded in Egypt during 1987-1988 and 1990-1991 seasons, which the yield was reduced by 50% in both seasons (Nassib *et al.*, 1991). In the year of low infections, the yield loss reached 5-15% (Mansour *et al.*, 1976).

Previous works showed that the usage of fungicides (Hanounik and Bisri, 1991; Khalil *et al.*, 1993; Khaled *et al.* 1995; Mohamed *et al.*, 1996; Brazauskiene, 1998; Abou-Zeid *et al.*, 2002 and El-Gammal, 2005) or biological control agents such as *Trichoderma* spp. (Roulston and Lane, 1988; Bennett and Lane, 1992; Zimand *et al.*, 1996; About –Zeid 2000 and El-Afifi, 2003) were very effective in controlling chocolate spot disease in faba bean crop.

Accordingly, two objectives of this work were to determine (i) the sensitivity of *Botrytis fabae* and *Botrytis cinerea* to several fungicides under laboratory conditions and (ii) the efficiency of five fungicides and one bioagent for controlling the chocolate spot disease in faba bean crop in relation to final yield.

## **MATERIALS AND METHODS**

Laboratory tests were conducted in Department of Plant Protection, Fac. of Agric., Al-Azhar University and field experiments were carried out during the 2004-2005 and 2005-2006 faba bean growing season at Itay El-Baroud, Beharah Governorate.

### **A- Isolation and Identification**

According to a method described by Hanounik and Robertson (1988) with some modified, the naturally infected leaves of faba bean cultivars (Giza 461 and Giza 3 cvs) grown in the field experiments were collected during both seasons (2004-2005 and 2005-2006). The leaves were cut into small pieces (1 cm), then, were surface disinfested with 10% Sodium hypochlorite solution for 2 min and washed with sterilized distilled water three times. The small pieces were dried between two sterilized filter papers (Whatman No. 1), then, were plated on PDA medium with Streptomycin Sulphate at 130 µg/ml in Petri dishes ( 9.0 cm in diam) and incubated at 25 + 2°C for 5 days, then, subcultured until pure colonies of the growing fungi were obtained. The number of isolates for each fungus was counted, and the frequency percent of each fungus was calculated as follow:

$$\text{Frequency \%} = \frac{\text{Number of isolates for each fungus}}{\text{Total number of isolates of all fungi}} \times 100$$

The isolated fungi were identified in Department of Agricultural Botany (Branch of Plant Pathology) in the same faculty, and listed in Table (2).

### **B- Fungicides toxicity *in Vitro* :**

The isolates of *Botrytis fabae* sard. and *B.cinerea* Pers. (The causal organisms of chocolate spot disease in faba bean) were tested for their sensitivity to five commercially formulated fungicides (Table 1) under laboratory conditions according to the method described by LaMondia and Douglas (1997). Fungicide concentrations of 0.00, 0.01, 0.05, 0.1, 0.5, 1.0, 5.0, 10.0, 50.0, 100.0, 250 and 500 µg a.i./ml (ppm) were prepared by adding the appropriate amount of stock solutions to 100 ml portions of autoclaved PDA medium cooled to about 50°C. The fungicide amended and unamended PDA were poured into five Petri dishes (9.0 cm in diam.) and a 2mm

mycelium plug was cut from the edge of an actively growing culture (7 day old) on PDA, invested and placed in the center of each fungicide amended and unamended plate. All plate were incubated in the dark at 25± 2°C. The mean diameter growth of colony was measured after 7 days from inoculation by which time the untreated controls for just covered the plate. Percentage of growth inhibition was calculated according to the equation described by Toppes and Wain (1957) as follow:

$$I\% = \frac{A - B}{A} \times 100$$

Where:

I % = percent of inhibition.

A = mean diameter growth in the control.

B = mean diameter growth in a given treatment.

**Table (1): List of the used compounds.**

Trade name	Commen name or active ingredient	Chemical name (IUPAC)	Rate of application/ 100 L. water
Dithane M-45 80 % w.p.	mancozeb (78% maneb + 2% zinc ion)	manganese ethylenebis (dithiocarbamate)(polymeric) complex with zinc salt.	-150 gram -200 gram -250 gram
Kocide -101 77% w.p.	cupric hydroxide; copper hydroxide; copper (II) hydroxide	cupric hydroxide; copper (II) hydroxide; copper hydroxide.	-150 gram -200 gram -250 gram
Rovral 50% w.p.	iprodione	3-(3-5-dichlorophenyl)-N-isopropyl-2,4-dioximidazolidine-1-carboxamide.	-150 gram -200 gram -250 gram
Ferrocopper 90% w.p.	mancozeb 40% + dicopper oxide 45%+ferrous sulfate 5% (Iron (II) sulfate	manganese ethylene bis (dithiocarbamate) (polymeric) complex with zinc salt + dicopper oxide (copper (I) oxide)+ iron (II) sulfate.	-150 gram -200 gram -250 gram
Topsin-M 70% w.p.	thiophanate-methyl	dimethyl 4-4' (o-phenylene) bis (3-thioallophanate).	-150 gram -200 gram -250 gram
Plant-Guard 30 million spore/cm <sup>3</sup>	<i>Trichoderma harzianum</i> L.	Egyptian strains of fungus <i>T.harzianum</i> each one cm <sup>3</sup> of the liquid contains 30 million organisms.	-150 cm <sup>3</sup> -200 cm <sup>3</sup> -250 cm <sup>3</sup>

Linear regression equations were fitted to logarithmic-propability data of fungicide concentrations and percent growth inhibition of each fungicide-fungus-treatment so that slope values and EC<sub>50</sub> values (concentration giving 50% linear growth inhibition) were interpolated (Finney, 1971).

### **C- Field experiments :**

Field trials were carried out during the two consecutive seasons (2004-2005 and 2005-2006) at Itay El-Baroud, Beharah Governorate. These experiments were designed by randomized complete block design (RCBD) with four replicates for each treatment. The size of each plot was 42m<sup>2</sup> (6 x 7m). The seeds of two faba bean cultivars (Giza 461 and Giza 3) were obtained from Central Administration of Seeds ARC, Ministry of Agriculture and Land Reclamation. The planting dates were 20 November 2004 and 13

November 2005 at the rate of 60 kg/feddan (4200m<sup>2</sup>). The seeds were planted on rows in hills (one seed/hill) and the distance between hills 20 cm. Recommended agronomic practices were followed in all plots including the untreated control (check).

Five commercial fungicides and one biological control agent (BCA) were applied with a knapsack sprayer (CP 3) with an adjustable one-nozzle calibrated to deliver 200 liter/feedan of the diluted spray. Three rates for each treatment (Table 1) were applied at 3 times from Mid January to the start of March in both seasons (Nasr, 2003). The interval between first, second and third applications ranged between 15-17 days, depending on suitable spraying conditions.

In both growing seasons, at the end of March, three hundred leaves were randomly collected from 10 plants /each plot and the following results were recorded:

- 1- Number of infected leaves with chocolate spot disease.
- 2- Disease incidence % (% of infected leaves)

$$= \frac{\text{Number of infected leaves}}{\text{Total number of examined leaves}} \times 100$$

- 3- Efficacy of treatments on disease incidence % (% reduction) was assessed by equation described by Rewal and Jtooty (1985).

Efficacy % =

$$\frac{\% \text{ infection in control} - \% \text{ infection in treatment}}{\% \text{ infection in control}} \times 100$$

- 4- Disease severity was recorded according to Hanounik and Rebertson (1988) as follow:

**Numerical grade**      **Description**

- |   |   |   |
|---|---|---|
| 1 | = | no disease symptoms or very few lesions covering up to 1% of the leaf surface;  |
| 3 | = | a few small, discrete lesions covering 1.1-2% of the leaf surface;  |
| 5 | = | lesion common, some coalesced, covering 2.1-5% of the leaf surface and poor sporulations;   |
| 7 | = | large, coalesced sporulating lesions, covering 5.1-10% of the leaf surface, some defoliation, and intermediate sporulations; and                          |
| 9 | = | extensive lesions on leaves covering more than 10% of the leaf surface, severe defoliation, abundant sporulation, stem girding, and death of most plants. |

- 5- Disease severity % was calculated by the following equation:-

$$\text{Disease severity \%} = \frac{n \times V}{9 \times N} \times 100$$

Where:

n= number of plants in every grade (V). V = numerical grade.

9 = Maximum disease grade. N= Total number of examined plants.

6- Efficacy of treatments on disease severity % (% reduction in disease severity) was assed by the following equation:

$$\text{Efficacy} = \frac{\% \text{ of disease severity in control} - \% \text{ of disease severity in treatment}}{\% \text{ of disease severity in control}} \times 100$$

At harvest, the plants were left to dry for 7 days under natural conditions, then, the grain yield of faba bean from each plot was determine as kg/plot. Increase % in grain yield were calculated as follow:

$$\% \text{ increase} = \frac{\text{grain yield in treatment} - \text{grain yield in control}}{\text{grain yield in treatment}} \times 100$$

The results were statistically analyzed according to Snedecor and Cochran (1969), and L.S.D. values were performed at 0.05 and 0.01.

## RESULTS AND DISCUSSION

### A- The isolation and identification.

The isolation was performed from naturally infected leaves in two faba bean (*Vicia faba* L.) cultivars (Giza 461 and Giza 3) through two consecutive growing seasons (2004-2005 and 2005-2006). The results in Table (2) showed that four fungi were isolated and identified as: *Botrytis fabae*, *botrytis cinerea*, *Alternaria alternata* and *Stemphylium botryosum*. Such results are in accordance with those obtained by Sinha *et al.*, (1988); Abd El-Moity *et al.*, (1990); Bouzhad *et al.*, (1998); El-Shazly, (2001) and El-Gammal (2005). The isolated fungi varied in number of isolated and frequently % between cultivars and also between years. These results clearly indicated that *B.fabae* was the most frequently isolated fungus followed by *B. cinerea*, *A.alternata* and *S.botryosum* (Harrison, 1988; Habib, 1990; Heweid, 1993, and El-Afifi, 2003). The differences between cultivars in this respect may be due to the presence of some nutrient substances on leaves of cultivar than another one. Also, Giza 3 cultivar was more susceptible to the infection by these fungi than Giza 461 cultivar (Abd El-Moity *et al.*, 1990; Abou-Zeid *et al.*, 1998; and El-Afifi, 2003).

**Table (2): The number of isolates and frequency % of the isolated fungi from leaves of two faba bean cultivars during two growing seasons (2004-2005 and 2005-2006).**

Isolated fungi	Giza 461cv.				Giza 3 cv.			
	Number of isolates		Frequency %		Number of isolates		Frequency %	
	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
<i>B. fabae</i>	23	34	56.09	57.63	28	36	57.14	59.01
<i>B. cinerea</i>	13	19	31.71	32.20	16	20	32.65	32.79
<i>A. alternata</i>	3	4	7.32	6.78	3	3	6.12	4.92
<i>S. batryosum</i>	2	2	4.88	3.39	2	2	4.08	3.28
Total	41	59	100	100	49	61	100	100

The differences between years may be attributed to suitable conditions for favorite the infection in the second season compared with in the first season. Walker (1975) reported that differences between cultivars in their susceptibility might be due to differences in their genetic make up and there effect on some morphological characters and chemical components of plants. Also, differences in genetic make up between cultivars as well as the environmental conditions might affect his pathogen relationship, which might play a role in cultivar susceptibility.

**B- Fungicides toxicity in Vitro.**

The sensitivity of *B.fabae* and *B.cinerea* to five fungicides (Topsin-M, 70% W.P., Rovral, 50% W.P., Ferrocopper, 90% W.P., Dithane M-45, 80% W.P. and Kocide 101, 77% W.P.) were studied under laboratory conditions, and the values of EC<sub>50</sub> of these fungicides were listed in Table (3). The results showed that *B.fabae* was more sensitive to these fungicides than *B.cinerea*. Generally, Topsin-M was the most effective followed by Rovral, Ferrocopper, Dithane M-45 and Kocide 101, respectively. The EC<sub>50</sub> values of these fungicides were 1.0 and 1.7, 1.3 and 1.8, 2.1 and 2.9, 5.8 and 9.0 and 66 and 96 ug a. i./ml for *B.fabae* and *B.cinerea*, respectively.

**Table (3): Fungicide sensitivity (EC<sub>50</sub> on fungicide-amended potato dextrose agar for 7 days in dark at 25+2°C) of *Botrytis fabae* and *Botrytis cinerea* isolated from leaves of faba bean.**

Fungi fungicides	Dithane M-45		Kocide 101		Rovral		Ferrocopper		Topsin-M	
	EC <sub>50</sub> *	slope	EC <sub>50</sub>	slope	EC <sub>50</sub>	slope	EC <sub>50</sub>	slope	EC <sub>50</sub>	slope
<i>B. fabae</i>	5.8	0.66	66.0	1.35	1.3	0.79	2.1	0.75	1.00	0.93
<i>B. cinerea</i>	9.0	0.67	96.0	1.52	1.8	0.77	2.9	0.83	1.7	0.71

\* EC<sub>50</sub> is the concentration of fungicide a.i (µg/ml) that suppresses the growth rate to half that of fungus on fungicide-free agar, calculated form the regression of normal growth rate and log concentration of fungicide.

These findings are in agreement with those reported by several authors. For Topsin-M, Virk and Grover (1975) reported that Deuteromycetous fungi were relatively more sensitive, exception, being *H. australiense* and *A.alternata* to thiophanate methyl (TPM). Mansour (1992) and El-Afifi (2003) found that growth of *B.fabae* was completely inhibited with Topsin-M at 10.0 ppm. For Rovral (Dicarboximides), several authors showed that Dicarboximide fungicides were effective on growth of *B.fabae*. Vyas (1984) reported that Dicarboximides which are specifically active against members of sclerotineaceae family such as *sclerotinia* spp., *Monilinia* sp. and *Botrytis* spp. Mohamed *et al.*, (1996) found that, both Ronilan and Sumisclex (Dicarboximides), inhibited *B.fabae* growth at 1.0 ug/ml. they added that the EC<sub>50</sub> values on susceptible strain of *B.fabae* were 0.013 and 0.073 ppm, while with tolerant strain were 1.65 and 1.50 ppm for Ronilan and Sumisclex, respectively.

The effect Dithane M-45 on mycelial growth was also reported, Mansour (1992) found that completely inhibited growth *B.fabae* at 50 ppm of

Dithane M-45. Mohamed *et al.*, (1996) found the same trend but at 20 ppm, and they found that EC<sub>50</sub> on susceptible strain was 11.0 ppm, while with tolerant strain was 36.0 ppm. El-Gammal (2005) found that the mycelial growth of *B.fabae* was 5.0 mm with 200 ppm of Dithane M-45. On the other hand, the result indicated that Kocide 101 was the least effective on mycelial growth of *B.fabae*. El-Afifi (2003) reported that mycelial growth of *B.fabae* was 2.45 mm with Kocide 101 at 250 ppm. Nasr (2003) cited that the mycelial growth of *B.fabae* was 8.0 mm with Kocide 101 at 200 ppm. El-Gammal (2005) found that the mycelial growth of this fungus was 26.67 mm at 200 ppm from Kocide 101.

The effect of fungicides on mycelial growth of *B.cinerea* was also reported by several authors. Yourman and Jeffers (1999) mentioned that the EC<sub>50</sub> of thiophanate methyl (TPM) and vinclozolin (Dicarboximides) were 0.1 and 0.3 ug/ml for mycelial growth of *B.cinerea*, respectively. Similar trend was also found by Yourman *et al.*, (2001). Lennox and Spotts (2003) reported that the baseline sensitivity levels (Mean EC<sub>50</sub> values) of a wild type *B.cinerea* populations to iprodione (Rovral) was 0.56 mg/L.

### **C. Field experiments.**

Field experiments were carried out during two consecutive growing seasons (2004-2005 and 2005-2006) in two faba bean cultivars (Giza 461 and Giza 3 cvs.) to study the efficiency of five commercial fungicides and one biological control agent (BCA) at three rates (Table 1) on chocolate spot disease incidence and severity in relation to crop yield.

#### **1- Effect of treatments on disease incidence.**

The results in Tables (4 and 5) showed the effect of treatments on disease incidence (number of infected leaves, % of infected leaves and % reduction in disease incidence). Generally, all treatments, at any rates in both cultivars, were significantly reduced the number of infected leaves with disease compared with the control.

Also, when the rate of application increased, the number of infected leaves was significantly reduced, plus, cultivars varied in response to these treatments. It was observed that these treatments were significantly reduced the number of infected leaves in Giza 461 compared with Giza 3. Topsin-M, Rovral, Ferrocopper and Dithane M-45 were significantly more effective than Kocide 101 and Plant-Guard. In the first season, on Giza 461 cultivar, the results indicated that at the first rate (150g/100L), Topsin-M and Rovral were significantly (at  $p = 0.05$ ) the most effective fungicides. At the second rate (200g/100L), the same trend was observed. At the third rate (250g/100L.), Topsin-M, Rovral and Ferrocopper were significantly reduced the number of infected leaves than Dithane M-45, Kocide 101 and Plant-Guard.

However, there were no significant differences between Topsin-M and Rovral and also between Rovral and Dithane M-45, and between Ferrocopper and Dithane M-45. At the third rate, no significant differences between Topsin-M, Rovral and Ferrocopper were occurred. On the other hand, on Giza 3 cultivar, the results indicated that, at the first rate, Topsin-M was significantly the most effective treatment. At the second rate, Topsin-M and Rovral were significantly the most effective.

**Table (4): Effect of fungicides or bioagent on chocolate spot disease incidence in two season (2004-2005 and 2005-2006).**

Treatments	Rate of application /100 L. water	* Average number of infected leaves		* % of infected leaves		** Average number of infected leaves		** % of infected leaves	
		Giza 461	Giza 3	Giza 461	Giza 3	Giza 461	Giza 3	Giza 461	Giza 3
		Dithane M-45 80% W.P.	150 g	16.00	20.00	5.33	6.67	19.00	21.25
	200 g	7.75	12.25	2.58	4.08	10.50	16.25	3.50	5.42
	250 g	6.00	7.75	2.00	2.58	5.50	8.25	1.83	2.75
Kocide-101 77% W.P.	150 g	23.25	28.25	7.75	9.42	20.50	30.00	6.83	10.00
	200 g	15.50	20.25	5.17	6.75	16.50	21.00	5.50	7.00
	250 g	11.00	14.75	3.66	4.92	11.75	16.50	3.92	5.50
Rovral 50% W.P.	150 g	10.75	17.00	3.58	5.66	14.00	16.75	4.00	5.58
	200 g	6.00	9.25	2.00	3.08	7.00	10.75	2.33	3.58
	250 g	2.50	5.00	0.83	1.67	3.75	6.00	1.25	2.00
Ferrocopper 90% W.P.	150 g	14.00	18.50	4.67	6.17	15.00	18.75	5.00	6.25
	200 g	8.25	13.75	2.75	4.58	8.50	14.00	2.83	4.67
	250 g	3.00	5.25	1.00	1.75	5.25	7.50	1.75	2.50
Topsin-M 70% W.P.	150 g	10.25	14.25	3.42	4.75	13.75	17.00	4.58	5.67
	200 g	5.25	8.50	1.75	2.83	6.25	10.75	2.08	3.58
	250 g	2.00	5.25	0.67	1.75	3.50	6.50	1.17	2.17
Plant-Guard 30 million spore/cm <sup>3</sup>	150 cm <sup>3</sup>	29.75	37.50	9.92	12.50	30.50	38.75	10.17	12.92
	200 cm <sup>3</sup>	19.75	24.25	6.58	8.08	23.50	27.50	7.83	9.17
	250 cm <sup>3</sup>	14.25	18.25	4.75	6.08	16.25	20.25	5.42	6.75
Untreated(check)	00.00	39.00	54.20	13.00	18.07	43.75	58.00	14.58	19.33

\* = first season

\*\* = second season

L.S.D. at	5%	1%	5%	1%
Treatments (T.)	1.79	2.36	1.97	2.61
Rates (R.)	1.17	N.S.	1.50	N.S.
Culivars (C.)	0.96	1.26	1.05	1.39
T.X.R.	3.09	4.09	3.42	4.52
T.X.C.	N.S.	N.S.	N.S.	N.S.
R.X.C.	1.66	2.19	1.83	2.42
T.XR.XC.	4.38	N.S.	N.S.	N.S.

At the third rate, Topsin-M, Rovral and Ferrocopper were significantly the most effective treatments than Dithane M-45, Kocide 101 and Plant-Guard. However, no significant differences at first rate between Rovral and Ferrocopper and between Ferrocopper and Dithane M-45 were recorded. At the second rate, no significant differences were observed between Topsin-M and Rovral and between Ferrocopper and Dithane M-45. Also, at the third rate, no significant differences between Topsin-M, Rovral and Ferrocopper were occurred. In the second season, the results in Table (4) showed that, with Giza 461 cultivar, Topsin-M, Rovral and Ferrocopper were significantly ( $p=0.05$ ) more effective than other treatments, at the first and second rates, but at the third rate, only Topsin-M and Rovral were significantly more effective than other treatments. However, no significant differences were found between Topsin-M, Rovral and Ferrocopper, and also between Dithane M-45 and Kocide 101 (at the first and second rates).



**Table (5): Efficacy of fungicides or bioagent on % reduction of disease incidence in both seasons (2004-2005 and 2005-2006).**

Treatments	Rate of application /100 L.	Giza 461			Giza 3		
		1st season	2nd season	Mean	1st season	2nd season	Mean
Dithane M-45 80% W.P.	150 g	59.00	56.58	57.79	63.09	63.37	63.23
	200 g	80.15	75.99	78.07	77.42	71.96	74.69
	250 g	84.61	87.45	86.03	85.72	85.77	85.74
Kocide-101 77% W.P.	150 g	40.38	53.15	46.76	47.87	48.27	48.07
	200 g	60.24	62.28	61.26	62.64	63.79	63.21
	250 g	71.85	73.12	72.48	72.77	71.55	72.16
Rovral 50% W.P.	150 g	72.46	72.56	72.51	68.68	71.13	69.90
	200 g	84.61	84.02	84.31	82.96	81.48	82.22
	250 g	93.61	91.43	92.52	90.76	89.65	90.20
Fero-copper 90% W.P.	150 g	64.08	65.71	64.89	65.85	67.66	66.75
	200 g	78.85	80.59	79.72	74.65	75.84	75.24
	250 g	92.37	87.99	90.15	90.31	87.07	88.69
Topsin-M 70% W.P.	150 g	73.69	68.59	71.14	73.71	70.67	72.19
	200 g	86.54	85.74	86.14	84.34	81.48	82.91
	250 g	94.85	91.97	93.41	90.31	88.77	89.54
Plant-Guard 30million spore/cm <sup>3</sup>	150 cm <sup>3</sup>	23.69	30.25	26.97	30.82	33.16	31.99
	200 cm <sup>3</sup>	49.39	46.30	47.84	55.28	52.56	53.92
	250 cm <sup>3</sup>	63.46	62.82	63.14	66.35	65.08	65.71

At the third rate, no significant between Topsin-M and Rovral was observed. On the other hand, on Giza 3 cultivar, Topsin-M and Rovral were significantly more effective than other treatments (in the first and second rate), but at third rate, Topsin-M, Rovral and Fero-copper were significantly the most effective. However, at the first and second rates, no significant differences were observed between Topsin-M and Rovral, but at the third rate, no significant between Topsin-M, Rovral and Fero-copper and also between Fero-copper and Dithane M-45. The results in Table (5) clearly indicated that all treatments were reduced the % of infected leaves. In general, Topsin-M and Rovral were the most effective treatments, and gave the highest reduction in this respect, followed by Fero-copper, Dithane M-45, Kocide 101 and Plant-Guard, respectively. The same trend was observed on both cultivars (Giza 3 and Giza 461) but at higher rate, the Rovral fungicide gave the best results on Giza 3 cultivar.

**2- Effect of treatments on disease severity.**

The results in Table (6) showed the efficiency of treatments on disease severity (% necrosis and % reduction in disease severity). All treatments in Table (6), except plant Guard at the first rate on Giza 461 cultivar, were significantly ( $p=0.05$ ) reduced disease severity (% necrosis) compared with untreated control. Also, increased rate from 150 to 250 g/100L. resulted in significant reduction in disease severity. These treatments reduced the disease severity on Giza 461 more than on Giza 3 cultivars. In the first season, on Giza 461 cultivar, the results in Table (6) indicated that Topsin-M, Rovral and Fero-copper were significantly more effective than others (at  $p = 0.05$ ) with the first and second rates, but at the third rate, Topsin-M and Rovral were significantly the most effective. However, no significant differences were observed at the first rate between Topsin-M,

Roveral and Ferrocopper and between Dithan-M-45, Kocide 101, and between Kocide 101 and plant- Guard. At the second rate, no significant differences were observed between Topsin-M , Rovral and Ferrocopper and also between dithane M-45 and Kocide 101. At the third rate, no significant were observed Topsin-M and Rovral, also between Ferrocopper and Dithane M-45 also between Dithane M-45 and Kocide 101 .

**Table (6): Efficacy of fungicides or bioagent on chocolate spot disease severity and % reduction in both seasons (2004-2005 and 2005-2006).**

Treatments	Rate of application /100 L.	Disease severity (% necrosis)*				% Reduction					
		1st season		2nd season		Giza 461			Giza 3		
		Giza 461	Giza 3	Giza 461	Giza 3	1st season	2nd season	Mean	1st season	2nd season	Mean
Dithane M-45 80% W.P.	150 g	12.42	15.12	12.07	16.64	22.03	28.71	25.37	17.74	20.46	19.10
	200 g	9.40	10.12	10.99	11.09	40.99	35.09	38.04	44.94	46.99	45.96
	250 g	5.28	5.52	6.32	6.29	66.85	62.67	64.76	69.97	69.93	69.95
Kocide-101 77% W.P.	150 g	13.42	15.68	14.41	16.89	15.76	14.89	15.32	14.69	19.26	16.97
	200 g	10.77	12.21	12.57	13.92	32.39	25.75	29.07	33.57	33.46	33.51
	250 g	6.36	6.86	7.77	7.50	60.07	54.10	57.08	62.68	64.15	63.41
Rovral 50% W.P.	150 g	10.98	11.79	10.58	13.34	31.07	37.51	34.29	35.85	36.23	36.04
	200 g	8.04	10.09	8.75	10.90	49.53	48.32	48.92	45.10	47.90	46.50
	250 g	2.29	3.68	3.63	4.21	85.63	78.56	82.09	79.98	79.87	79.92
Ferrocopper 90% W.P.	150 g	10.43	11.17	11.98	14.16	34.53	29.24	31.88	39.23	32.31	35.77
	200 g	7.45	8.59	7.35	9.05	53.24	56.58	54.91	53.26	56.74	55.00
	250 g	4.20	4.67	5.14	5.06	73.64	69.64	71.64	44.59	75.81	75.20
Topsin-M 70% W.P.	150 g	10.15	11.74	11.40	12.87	36.28	32.66	34.47	36.13	38.48	37.30
	200 g	6.85	8.43	6.61	8.26	56.99	60.96	58.97	54.14	60.52	57.33
	250 g	1.66	2.30	2.35	3.53	89.58	86.12	87.85	87.49	83.13	85.31
Plant-Guard 30 million spore/cm <sup>3</sup>	150 cm <sup>3</sup>	14.77	16.26	15.25	17.86	7.28	9.92	8.60	11.53	14.63	13.08
	200 cm <sup>3</sup>	12.97	14.82	13.10	14.96	18.58	22.62	20.60	19.37	28.49	23.93
	250 cm <sup>3</sup>	10.08	11.02	10.45	11.69	36.72	38.27	37.49	40.04	44.12	42.08
Untreated (check)	00.00	15.93	18.38	16.93	20.92	00.00	00.00	00.00	00.00	00.00	00.00

\* 0.00% = No necrosis on leaves, 100% = all of leaf tissue necrotic.

L.S.D. at	5%	1%	5%	1%
Treatments (T.)	1.88	2.49	1.81	2.40
Rates (R.)	1.24	1.63	1.19	1.57
Culivars (C.)	1.01	N.S.	0.97	N.S.
T.X.R.	3.27	4.32	3.14	4.17
T.X.C.	N.S.	N.S.	N.S.	N.S.
R.X.C.	1.30	2.31	1.68	2.22
T.XR.XC.	N.S.	N.S.	4.44	N.S.

On the other hand, in case of Giza 3 cultivar, significant differences were observed between Topsin-M, Rovral and Ferrocopper compared with other treatments, at the first rate. At the second rate, significant differences were observed ( $p = 0.05$ ) with Topsin-M and Rovral compared with others. At the third rate, significant differences were observed between all treatments and plant-Guard. Also, no significant differences was observed between Topsin-M, Rovral and Ferrocopper, and between Dithane M-45 Kocide 101 and Plant-Guard in the first rate. Also, between Topsin-M, Rovral and Ferrocopper and also between Ferrocopper and Dithane M-45, at the second rate. At the third rate, no significant differences ( $p = 0.05$ ) were observed between Topsin-M and Rovral and also between Rovral and Ferrocopper, and between Ferrocopper and Dithane M-45, and between Dithane M-45 and Kocide 101.

In the second season, on Giza 461 cultivar, the results in Table (6) indicated that most treatments (except plant-Guard at the first rate) significantly ( $p=0.05$ ) reduced the disease severity compared with untreated control. Also, increased rates of application significantly reduced the severity of the disease (except from Dithane M-45 at 150 to 200g./100L). The results also showed that at the first rate, Topsin-M, Rovral, Ferrocopper and Dithane M-45 were significantly reduced disease severity greater than Kocide 101 and Plant-Guard. Topsin-M was significantly the most effective treatment (at second rate). Also, no significant differences were observed between Topsin-M, Rovral, Ferrocopper and Dithane M-45 and between Kocide 101 and Plant-Guard (at the first rate). However, no significant differences between Rovral, Ferrocopper and between Kocide 101 and Plant-Guard. At the third rate, no significant differences were observed between Ferrocopper and Dithane M-45 and between Dithane M-45 and Kocide 101. The effect of treatments on disease severity, on Giza 3, also were shown in Table (6) in the second season. The results showed that all treatments were significantly reduced disease severity compared with the untreated check. However, increased rates resulted in higher effective on disease severity. At the first rate, Topsin-M, Rovral and Ferrocopper were significantly reduced disease severity than Dithane M-45, Kocide 101 and Plant-Guard. Also, Topsin-M was significantly the most effective at the second rate, where, at the third rate, both Topsin-M and Rovral were significantly the most effective ones. However, no significant differences were observed between Topsin-M, Rovral, and between Rovral, Ferrocopper, and between Dithane M-45, Kocide 101 and Plant-Guard (at the first rate). At the second rate, also between Rovral, Ferrocopper, Rovral, and Dithane M-45 as well as between Kocide 101 and Plant-Guard, but the third rate, also no significant differences were observed between Topsin-M, Rovral, and Rovral, Ferrocopper. On the other hand, it was observed that, in some cases, no significant differences were observed between cultivars such as Dithane M-45 at the second rate, also Rovral, Ferrocopper, Dithane M-45 and Kocide 101 at third rate. The results in Table (6) also indicated that Topsin-M and Rovral were the most effective in controlling the disease severity followed by Ferrocopper, Dithane M-45, Kocide 101 and plant-Guard, respectively.

**3- Effect of treatments on grain yield.**

The results in Table (7) showed the effect of treatments on grain yield (Kg/plot) and % of increase in grain yield compared with untreated control. In the first season, on Giza 461 cultivar, the results indicated that most treatments at any rate (except Kocide 101 and Plant-Guard at the first rate) were significantly increased grain yield compared with the untreated control.

**Table (7): Efficacy of fungicides or bioagent on grain yield of faba bean in both seasons (2004-2005 and 2005-2006).**

Treatments	Rate of application /100 L.	Grain yield (kg/plot)				% increase in grain yield					
		1st season		1st season		Giza 461			Giza 3		
		Giza 461	Giza 3	Giza 461	Giza 3	1st season	2nd season	Mean	1st season	2nd season	Mean
Dithane M-45 80% W.P.	150 g	14.77	14.61	14.67	14.45	4.45	3.88	4.16	6.43	7.13	6.78
	200 g	15.33	15.10	15.28	15.11	7.76	7.72	7.74	9.47	11.18	10.32
	250 g	15.47	15.37	15.42	15.20	8.59	8.56	8.57	11.06	11.71	11.38
Kocide-101 77% W.P.	150 g	14.43	14.19	14.51	14.15	2.01	2.82	2.41	3.66	5.16	4.41
	200 g	15.30	14.51	14.94	14.61	7.58	5.62	6.60	5.79	8.14	6.96
	250 g	15.49	15.08	15.28	14.95	8.71	7.72	8.21	9.35	10.23	9.79
Rovral 50% W.P.	150 g	15.47	14.92	15.11	14.78	8.59	6.68	7.63	8.38	9.21	8.79
	200 g	15.95	15.66	15.92	15.41	11.35	11.43	11.39	12.70	2.91	12.80
	250 g	16.16	15.73	16.06	15.61	12.50	12.20	12.35	13.09	14.03	13.56
Ferrocopper 90% W.P.	150 g	14.90	14.81	15.09	14.71	5.10	6.56	5.83	7.70	8.77	8.23
	200 g	15.51	15.48	15.66	15.43	8.83	9.96	9.39	11.69	13.03	12.36
	250 g	15.59	15.56	15.84	15.71	9.30	10.98	10.14	12.15	14.58	13.36
Topsin-M 70% W.P.	150 g	15.67	15.13	15.52	15.08	9.76	9.15	9.45	9.65	11.01	10.33
	200 g	16.04	15.83	15.92	15.81	11.84	11.43	11.63	13.64	15.12	14.38
	250 g	16.76	16.69	16.71	16.61	15.63	15.62	15.62	18.09	19.20	18.64
Plant-Guard 30 million spore/cm <sup>3</sup>	150 cm <sup>3</sup>	14.35	14.19	14.38	13.98	1.46	1.95	1.70	3.66	4.01	3.83
	200 cm <sup>3</sup>	15.18	14.34	14.80	14.42	6.86	4.73	5.79	4.67	6.93	5.80
	250 cm <sup>3</sup>	15.29	14.62	15.22	14.75	7.52	7.36	7.44	6.50	9.02	7.76
Untreated(check)		00.00	14.14	13.67	14.10	13.42	00.00	00.00	00.00	00.00	00.00

L.S.D. at	5%	1%	5%	1%
Treatments (T.) =	0.35	0.46	0.34	0.45
Rates (R.) =	0.23	0.30	0.22	0.30
Culivars (C.) =	N.S.	N.S.	N.S.	N.S.
T.X.R =	N.S.	N.S.	N.S.	N.S.
T.X.C. =	N.S.	N.S.	N.S.	N.S.
R.X.C. =	0.32	0.42	0.31	0.42
T.XR.XC. =	N.S.	N.S.	N.S.	N.S.

At the first rate, Topsin-M and Rovral were significantly ( $p = 0.05$ ) more effective than other treatments. The same trend was also observed on second rate, but at the third rate, the results showed that Topsin-M was significantly the most effective compared with other tested compounds. Also, no observed significant differences between Ferrocopper, Dithane- M-45, and Dithan, Kocide 101 and between Kocide 101 and Plant-Guard. The same trend was also observed on second rate and also third rate. Also, no significant differences between second and third rates in Ferrocopper, Dithane M-45, Kocide 101 and plant-Guard in grain yield of faba bean. Also, on Giza 3, the results indicated that all treatments increased grain yield significantly compared with untreated control, also, Topsin-M, Rovral and

Ferrocopper were the most effective followed by Dithane M-45, Kocide 101 and Plant-Guard, respectively. The same trend was also observed on other rates. The results clearly indicated that all treatments were significantly increased grain yield compared with plant-Guard and untreated control. Topsin-M was the most effective at higher rate, with no significant between Rovral, Ferrocopper, and between Ferrocopper, Dithane M-45 as well as between Dithane M-45 and Kocide 101. Similar trend was observed in second season. In case of Giza 461 cultivar, The results showed that most treatments at any rate (except plant-Guard at first rate) were significantly increased grain yield compared with other treatments. Generally, Topsin-M, Rovral and Ferrocopper were significantly increased grain yield compared with other treatments. However, no significant differences were observed between some treatments such as between Topsin-M and Rovral, similar trend was also observed on second and third rate and also on Giza 3 cultivar. Also, no significant differences were found between rates especially the second and third rates in Rovral, Ferrocopper and, Dithane M-45. The results also in Table (7) showed the effect of treatments on % increase in grain yield. Topsin-M gave the best results followed by Rovral, Ferrocopper, Dithane M-45, Kocide 101 and plant-Guard, respectively, increasing the rate of application resulted in high grain yield than the untreated control. Also, increasing yield was more in Giza 3 cultivar than in Giza 461 cultivar. These results pointed out the chocolate spot disease was the most serious on faba bean plants and reduced grain yield. Similarly, Abd El-Latif (1984) found that a positive correlation between these disease severity and yield. Spraying with fungicides gave best results and reducing the incidence and severity of this disease improved plant growth and finally increased grain yield. These results also indicated that Topsin-M, Rovral, Ferrocopper and Dithane M-45, respectively, gave the best results on this disease and increased grain yield. While, Kocide 101 and bioagent (Plant-Guard) were the least effective. Abd El-Monem (1981) found that Dithane M-45 effectively limited infection by *B.fabae*. Hanounik (1981) reported that Ronilan at 2 g./L. increased grain yield than control in field trials under inoculation with *B.fabae* and *B.cirerea*. Nene and Thaplial (1982) found that both thiophanate and thiophanate methyl show a combination of protective, curative and systemic fungitoxic activity. Also, they have effective against *Botrytis* mold of various crops. Abou-Zeid *et al.*, (1990) found that Rovral followed by Dithane M-45 and Cuprozan Super 311 were the best in control chocolate spot disease. Giltrap (1991) found that Rovral and Corbendazim gave good control of chocolate spot disease and significantly increased grain yield. Mansour (1992) reported that Topsin-M followed by Dithane M-45 were the most effective in reducing the chocolate spot disease and increased grain yield. Hegab and Beshir (1994) reported that Topsin-M followed by Dithane M-45 and Kocide 101 decreased infection percentage of chocolate spot disease. Abou-Zeid *et al.* (2002) reported that Dithane M-45 significantly decreased infection with this disease El-Afifi (2003) found Topsin-M -70 was more effective than Kocide 101 in field trials during 1999-2000 and 2000-2001. Nasr (2003) indicate that fungicides such as Galbin copper or Acrobat copper were more effective than plant-Guard in control this disease. El-Gammal (2005) found that Dithane M-

45 was more effective than Kocide 101 on chocolate spot disease in faba bean.

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مكافحة مرض التبقع الشيكولاتي في الفول البلدي  
رمضان مصطفى عبده الخولي  
قسم وقاية النبات- كلية الزراعة بالقاهرة- جامعة الأزهر

يعتبر مرض التبقع الشيكولاتي (البنّي) الذي يحدثه فطر *Botrytis fabae* and *Botrytis cinerea* واحداً من أهم الأمراض الفطرية على الفول البلدي في مصر. تم عزل فطريات *Botrytis fabae*, *Botrytis cinerea*, *Alternaria alternata* and *Stemphylium botryosum* من أوراق فول مصابة (صنفي جيزة ٤٦١ وجيزة ٣) خلال موسمي الدراسة (٢٠٠٤-٢٠٠٥ و ٢٠٠٥-٢٠٠٦). وهذه الفطريات قد اختلفت في عدد العزلات بين الأصناف وأيضاً بين المواسم ، ولقد تم عزلها بصورة اكبر من صنف جيزة ٣ عن صنف جيزة ٤٦١ وأيضاً في الموسم الثاني عن الموسم الأول. ولقد تم دراسة حساسية فطريات *Botrytis fabae* and *Botrytis cinerea* لخمسة من مبيدات الفطريات تحت الظروف المعملية وأوضحت النتائج بصفة عامة أن فطر *Botrytis fabae* كان أكثر حساسية من فطر *Botrytis cinerea* كانت قيم  $EC_{50}$  للمبيدات المختبرة كالتالي: توبسين م. ٧ (١,٠ و ١,٧ ، روفرال (١,٣ و ١,٨) فيروكوبير (٢,١ و ٢,٩) ، دياتين م-٤٥ (٥,٨ و ٩) وكوسيد ١٠١ (٦٦ و ٩٦ ميكروجرام مادة فعالة/مل) على كل من فطري *Botrytis fabae* and *Botrytis cinerea* على الترتيب. وأيضاً تم عمل تقييم لخمسة مبيدات فطريات وأيضاً احد المبيدات الحيوية وهو بلانت- جارد تحت الظروف الحقلية بثلاثة معدلات وهي ١٥٠ ، ٢٠٠ ، ٢٥٠ جرام منتج لكل ١٠٠متر ماء وذلك لمكافحة هذا المرض. وأوضحت النتائج أن مبيدات توبسين م. ٧ وروفرال قد أعطت أحسن النتائج في مكافحة هذا المرض سواء في وجود المرض أو شدته وأدى ذلك لزيادة المحصول ثم يأتي بعدها مبيدات فيروكوبير ثم دياتين م-٤٥ وقد أعطت مكافحة مرضية بينما كانت مبيدات كوسيد ١٠١ والمبيد الحيوي بلانت جارد هي اقل المعاملات فاعلية ضد هذا المرض. ولقد كانت الفاعلية تزداد بزيادة المعدل المستخدم في كل المعاملات. وعموماً فإن هذه المعاملات عندما تم رشها على أوراق الفول البلدي قد أعطت مكافحة مرضية للمرض وقد أدى ذلك لزيادة نمو النباتات كما أدى إلى زيادة المحصول النهائي للحبوب- وقد أظهرت النتائج المتحصل عليها أن صنف جيزة ٤٦١ أكثر مقاومة للمرض من صنف جيزة ٣ خلال موسمي الدراسة.

*El-Kholy, R. M.A.*

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