

EXPERIMENTAL RESULTS

Distribution of barley spot blotch in Egypt from 1979-81:

To determine the presence and spread of barley spot blotch in Egypt, forty seven Barley Trap Varieties were grown in different Governorates of Egypt. Data obtained and tabulated in Table (4) show that the percentage of infection and disease severity differed from season to season and from locality to another. Percentage of infection of these varieties to this disease is shown in Table (4) indicating that the disease was found in Delta and Middle Regions. High level of spot blotch infection was observed at Kafr El-Sheikh, Alexandria, Domietta and decreased gradually towards the south where no infection was observed at Fayoum Governorate and Southwards and also to some extent in Governorates of Beheira, Sharkia and Gharbia.

Isolation and identification of the causal organism:

Twelve isolates of Helminthosporium sativum were isolated from diseased leaves showed typical spot blotch symptoms. They were sampled from Kafr El-Sheikh (isolates No. 1,,2, 3, 4 and 5), Damietta (isolates No.

Table (4): Distribution of spot blotch on 47 Barley Trap Varieties at 14 different locations in Egypt during (1978/79)-(1980/81)

			1978/1979			1979/1980			1980/1981		
No.	Governorate	Location	N.I.V.	P.	S.	N.I.V.	P.	S.	N.I.V.	P.	S.
a- Delta Region:											
1	Alexandria	Alexandria	20	42.6	60	34	72.4	10	30	63.9	70
2	Beheira	Kom-Hammada	5	10.7	30	10	21.3	30	4	8.6	20
3	Kafr El-Sheikh	Sakha	34	72.4	70	38	80.9	80	35	74.5	80
4	Damietta	Damietta	13	27.7	20	18	38.4	50	15	31.9	40
5	Dakahlia	Talkha	5	10.7	30	2	4.3	10	3	6.4	20
6	Sharkia	Hehya	3	6.4	30	8	17.1	20	6	12.8	40
7	Gharbia	Samanoud	3	6.4	20	7	14.9	30	6	12.8	20
8	Menoufia	Quesna	0	0	0	2	4.3	10	4	8.6	20
9	Kalubia	Bahtim	10	21.3	20	12	25.6	30	8	63.9	20
b- Middle Region:											
10	Giza	Giza	6	12.8	20	10	21.3	20	7	14.9	20
11	Fayoum	Fayoum	0	0	0	0	0	0	0	0	0
12	Minia	Mallawi	0	0	0	5	10.7	30	0	0	0
c- Southern Region:											
13	Assiout	Manfalot	0	0	0	0	0	0	0	0	0
14	Soheg	El-Manshah	0	0	0	0	0	0	0	0	0

* N.I.V. = Number of infected varieties.

P = Percentage of infection

S = Severity of infection

6, 7 and 8), Alexandria (isolate No. 9), Kalubia (isolate No. 10) and Giza (isolates No. 11 and 12).

Identification of these isolates was carried out according to the key of Helminthosporium species reported by Luttrell (1951), in the USA depending on conidiospores characters, i.e., colour, number and germination of conidiospores cells, shape, measurements, as well as, number of pseudoseptates.

These isolates were, also, compared with other isolates previously identified at Mycol. Res. Div., plant pathology Institute, Agric. Res. Center, Giza.

Colonies were at first white becoming dark olivaceous brown with spore production. Conidiophores pale to mid-dark brown, 150-180 U long, 6-10 U thick, Conidia curved or straight, tapering toward the ends, ends rounded, olivaceous, 90-130 x 15- 23 U. Most commonly 6-10 pseudoseptated,

Also, isolation from the grains showing black point symptoms and particularly in case of the heavy infected plants, show that Helminthosporium sativum was frequently isolated from these infected grains.

Pathogenicity test:

Twelve isolates of Helminthosporium sativum, were used to inoculate the plants of four barley varieties (Table 5) in seedling stage. Data were recorded after 6 days of inoculation.

Results indicate that isolate No. 4 of Helminthosporium sativum was more virulent than the other isolates where it infected three varieties. On the other hand, isolate No. 12 showed the lowest reaction of infection. As regards to the affected barley varieties, Traill followed by Giza 117 were the most affected ones. The highest disease reaction was observed on Traill variety inoculated with isolates No. 4, No. 7 and No. 11, whereas, the lowest disease reaction was recorded on Mianwali barley variety inoculated with isolates No. 9, No. 10, No. 11 and No. 12. Reisolation of the causals was undertaken as before and the isolated fungi were matched with the original isolates.

Host range:

Results of the host range of Helminthosporium sativum demonstrated that Avena fatua, Cynodon dactylon, Cyperus rotundus, Echinochloa colonum, E. crusgalli, Hordeum vulgare (Giza 117), Lolium perenne, L. temulerum, Oryza sativa (Arabi), polypogon monspeliensis, setaria viridis, Sorghum algare

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Table (5): Reaction of different barley varieties
to twelve isolates of H. sativum

Isolate No.	Varieties and disease reaction*			
	Traill	Giza 117	C.I. 7123	Mianwali
1	4	3	2	2
2	3	4	1	2
3	4	3	2	1
4	5	5	3	2
5	4	4	2	1
6	3	4	1	1
7	5	3	2	1
8	4	3	1	2
9	3	4	2	1
10	3	3	2	1
11	5	4	1	1
12	2	2	1	1

* Disease reactions are given on 1 to 5 basis.

1 = (R) = Being resistant.

2 = (MR) = Moderately resistant

3 = (MS) = Moderately susceptible

4-5 = (S) = Susceptible

Table (6): Reaction of different hosts to H. sativum

No.	Host	Reaction*
1	<i>Avena fatua</i>	+
2	<i>Cynodon dactylon</i>	+
3	<i>Cyperus rotundus</i>	+
4	<i>Echinochloa colonum</i>	+
5	<i>Echinochloa crusgalli</i>	+
6	<i>Hordeum vulgare</i> (Giza 117)	+
7	<i>Lolium perena</i>	+
8	<i>Lolium temulentum</i>	+
9	<i>Oryza setiva</i> (Arabi)	+
10	<i>Phalaris minor</i>	-
11	<i>Phalaris paradoxa</i>	-
12	<i>Polypogon monspeliensis</i>	+
13	<i>Setaria viridis</i>	+
14	<i>Sorghum vulgare</i> (Giza 15)	+
15	<i>Triticum vulgare</i> (Giza 139)	+
16	<i>Zea maize</i> (Hybrid 186).	+

(*) - = No visible symptoms
+ = Typical symptoms of leaf spot.

perena, L. temulentum, Oryza sativa (Arabi), polypogen monspeliensis, setaria viridis, Sorghum vulgare (Giza 15), Triticum vulgare (Giza 139) and Zea maize (Hybrid 186) were susceptible (Table 6). On the other hand, Phalaris minor and P. paradoxa were immune. These results indicated that Helminthosporium sativum has a wide host range in Egypt. Reisolation of the causal fungus was undertaken as before and the isolated fungi were matched with the original fungus.

Viability of Helminthosporium sativum:

Barley plants of Giza 117 variety were grown and inoculated at seedlings stage using the cultures of different ages of Helminthosporium sativum.

Data in Table (7), show that the susceptible variety Giza 117 was infected regardless of the storage periods of cultures. The highest disease severity was recorded when the inoculum was taken from 1 month old culture followed by that taken from two months old culture. However, no significant differences were noticed between the two periods of 3 or 6 months. In this respect, disease severity gradually increased when the age of culture was elongated from 15 days to 1 month, whereas, it decreased by elongating the storage periods. The lowest percentage of the infected area

were observed when the culture was stored for 18 months. The results of these studies indicate that conidia of Helminthosporium sativum could remain viable for at least one year without significant reduction in their viability.

Table (7): Effect of different storage periods of cultures on the viability of Helminthosporium sativum.

Storage periods (Month)	15 days	1 month	2 month	3 month	6 month	12 months	18 months	Control [*]
Percentage of infection (Leaf area)	60	80	75	70	65	40	30	0.0
L.S.D. 0.05	13.79							

* Sprayed with water only.

Enzymes activity:

A- Production of pectulolytic and cellulolytic enzymes by three isolates of *Helminthosporium sativum* and effect of some fungicides on their activity in the fungal filtrate:

The polygalacturonase (PG), cellulase (CX) and Pectin Methyl Estrase (PME) activities in fungal filtrates of isolates No. 4, 5 and 12 of *Helminthosporium sativum* grown in pectin and CMC media were studied. Results in Table (8) and Fig. (), show that the activity of each of enzymes (PG and CX) increased as the time of incubation period increased. Also, results indicated that the highest activity of PG and CX enzymes was in the filtrate of isolate No. 4 (the most pathogenic isolate) and the lowest activity was in filtrate of isolate 12 (the least pathogenic isolate). Also, it was observed that the relative loss in viscosity of pectin and CMC solutions of different isolates was faster in the first period of incubation.

Also, results in Table (8), indicated that the highest activity of Pectin Methyl Estrase (PME) enzyme was in fungal filtrates of isolates 4 followed by isolates No. 5 and 12.

Table (6): Determination of pectulclytic and cellulolytic enzymes activities for certain isolates of *H. sativum* causing spot blotch of barley.

Isolates	% cf loss in viscosity of PG and Cx activity after periods										Relative PME activity
	Polygalacturonase (PG)					Cellulase (Cx)					
	5	10	15	30	60	5	10	15	30	60	
(A) Highly pathogenic	9.65	34.02	44.68	59.90	72.59	34.12	37.89	41.18	48.24	51.77	1.2
(B) Moderately "	5.61	7.48	26.17	33.18	50.94	6.76	21.23	31.68	43.73	46.12	0.9
(C) Least "	18.89	21.76	30.00	31.12	46.12	7.18	14.11	22.06	27.70	39.90	0.8

- (A) Isolate No. 4
(B) Isolate No. 5
(C) Isolate No. 12

* Mean values of 0.1% NaOH milliliters required to neutralize the carboxylic groups produced from 0.5% pectin solution at pH 7.0 after 24 hours inocubation with crude enzyme.

The effect of the different concentrations of Plantavax, Bayleton, Wolfen thiram and Dithane M₂₂ on polygalacturonase (PG) and cellulolytic (CX) enzymes activities, in fungal filtrates of the three isolates of Helminthosporium sativum, grown on Pectin and Carboxy methyl cellulase (CMC) media, at pH 5.6, was studied according to Maxwell and Bateman (1967) method.

Results in Table (9) and Fig. (3) indicated that the activity of enzymes depends on the fungicidal concentration and on the isolate of Helminthosporium sativum used. In other words this activity differed from one treatment to another. For example the highest activity of PG enzyme was found in fungal filtrate of isolate 4 followed by isolate 5 and 12 respectively.

In general, these results indicated that each concentration of each one of all fungicides used had different effect on the activity of PG enzyme in their fungal filtrates. For example in fungal filtrate of isolate 4, 5 and 12 the results indicated that PG enzyme activity was reduced when the concentration of the tested fungicides increased specially in case of Plantavax and Bayleton. Also, it was clear from results that the concentrations of Wolfen thiram and Dithane M₂₂ had little effect on the activity of PG enzyme in

Table (9) Effect of different concentrations of some fungicides in fungal filtrates by using Talboys and Bush medium on PG activity (loss in viscosity) by the three isolates of H. sativum.

Fungicides	Conc.	Isolate 12				Isolate 5				Isolate 4			
		5 min.	15	30	60	5	15	30	60	5	15	30	60
Plantavex 20	50	11.31	13.50	14.29	17.36	14.11	28.83	41.72	53.99	22.50	25.00	34.36	50.63
	100	10.43	11.61	13.98	17.18	12.36	23.31	40.49	52.76	19.38	25.00	33.38	48.50
	200	10.02	11.43	13.45	14.11	12.91	20.25	32.52	44.79	17.63	26.59	31.13	43.13
	400	9.24	10.65	11.83	14.11	11.36	14.11	31.29	41.40	13.13	16.25	30.00	41.50
	800	7.04	9.52	10.83	10.43	10.04	10.43	14.11	40.49	11.63	13.75	21.25	49.75
Bayleton	50	19.05	23.81	27.62	33.33	16.00	25.00	34.00	45.00	20.00	31.70	36.59	46.82
	100	16.19	21.43	24.76	32.38	14.00	20.00	29.00	40.00	15.12	24.88	32.20	41.95
	200	14.29	19.05	23.81	38.10	12.00	17.00	26.00	39.00	13.17	22.93	30.37	41.16
	400	14.29	19.05	23.81	26.19	10.00	14.00	24.00	37.50	12.20	22.93	31.20	40.98
	800	18.10	12.38	7.14	16.67	15.00	16.00	26.00	40.00	20.98	27.80	35.61	45.37
Wolfen-thiram	50	11.56	32.81	53.91	60.50	31.09	41.70	44.54	64.71	27.78	48.61	58.33	61.11
	100	15.63	45.31	64.72	62.41	32.52	47.20	51.30	66.39	26.22	45.39	53.38	51.38
	200	17.19	44.53	57.03	64.84	35.88	47.10	56.26	67.34	28.39	41.67	51.94	46.67
	400	21.88	50.00	57.81	67.84	39.24	49.50	56.79	68.03	20.00	37.50	48.78	43.56
	800	29.69	53.91	61.72	68.75	40.84	50.34	56.79	62.18	48.61	47.22	59.72	59.72
Dithane-M ₂₂	50	12.18	14.00	19.64	28.00	33.35	45.53	49.42	58.75	11.63	24.65	44.19	50.70
	100	18.00	15.64	20.00	30.91	30.07	36.58	42.41	54.47	19.76	29.53	46.40	58.84
	200	21.82	19.27	27.45	33.64	21.01	32.68	48.64	59.53	30.23	34.88	48.60	57.91
	400	22.91	24.55	27.28	33.82	17.90	29.96	43.58	55.64	34.65	37.67	50.04	59.65
	800	24.55	25.45	28.73	31.55	15.96	28.02	40.86	52.53	42.56	56.98	59.97	61.86
Control	0.0	23.70	36.42	55.20	61.80	35.12	47.22	54.15	65.17	32.11	49.70	61.13	66.18

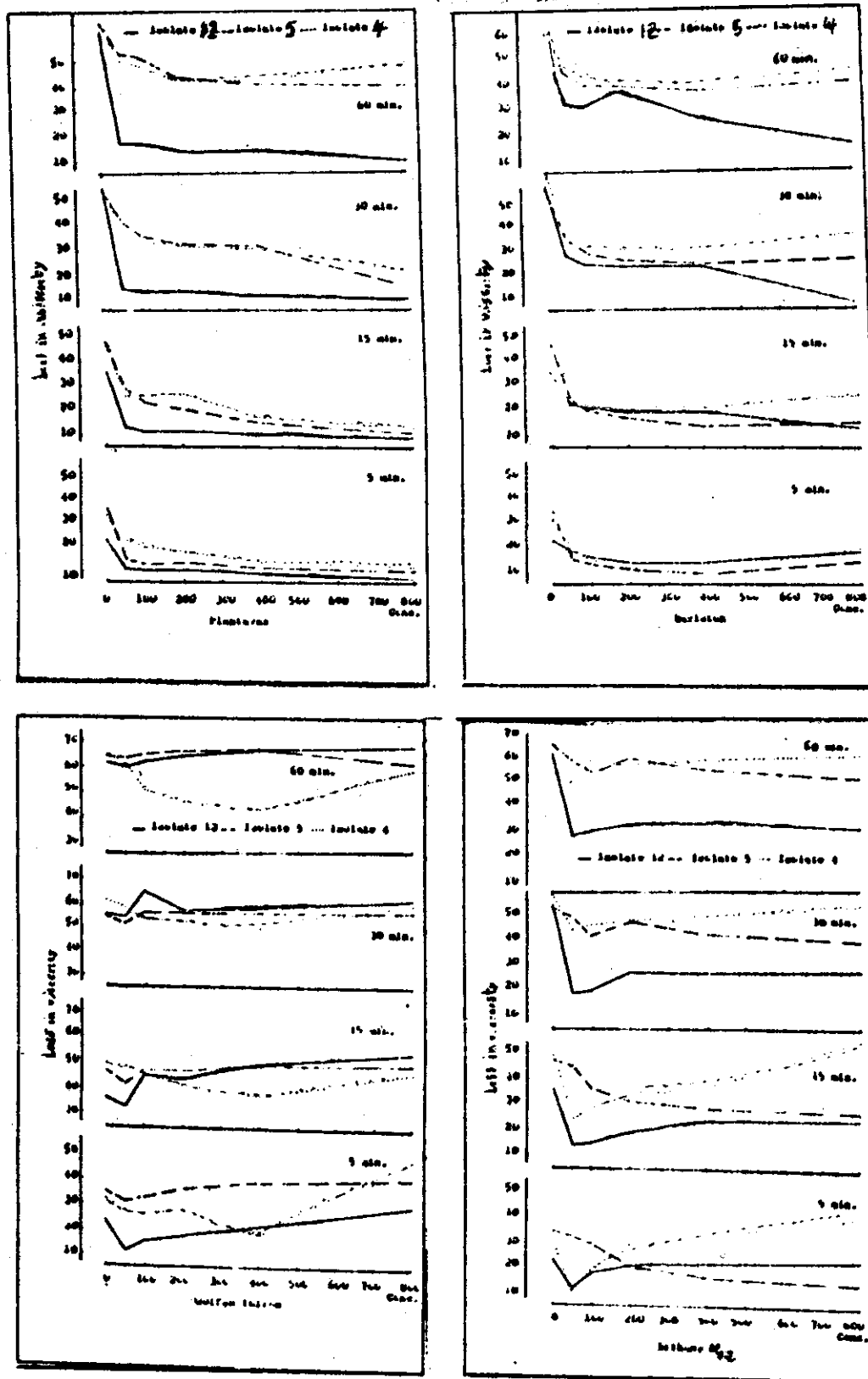


Fig.(3) Effect of different concentrations of some fungicides in fungal filtrates by using Talboys and Bush medium PG activity (loss in viscosity) by the three isolates of H. sativum.

the fungal filtrate of isolates 4, 5 and 12 descendingly, and increased the activity of PG enzyme in the fungal filtrates of isolates in contrast arrangement as isolates 12, 5 and 4 respectively.

Results in Table (10) and Fig. (4) indicate that the different concentrations of the tested fungicides had varying effect on the activity of CX enzymes in the fungal filtrates of the different isolates of Helminthosporium sativum. For example, for fungal filtrate of isolates 4 and 5, results indicated that generally CX enzyme activity was reduced by increasing the concentrations of all fungicides except in case of Wolfen thiram with isolate 4.

B-Activities of oxidative enzymes:

The activity of Polyphenol oxidase, peroxidase, catalase and ascorbic acid oxidase were determined in both fungal filtrates and mycelial matrix of the two isolates of Helminthosporium sativum grown on glucose peptone medium for 15 and 30 days. Results are shown in Tables (11, 12, 13 and 14).

Polyphenoloxidase activity:

Data obtained in Table (11) show that clear differences in Polyphenoloxidase activity were found

Table (10) Effect of some fungicides in fungal filtrates by using Talboys and Bush medium on cellulolytic activity (less in viscosity) by the three isolates of H. sativum

		Isolate 12						Isolate 5						Isolate 4					
		5 min.		15	30	60	5	15	30	60	5	15	30	60	5	15	30	60	
Fungicides	Conc.																		
	50	13.39	17.32	19.95	23.88	15.48	28.57	44.05	58.33	17.20	19.14	20.43	32.90	33.33	35.48	37.63	34.40		
	100	21.26	23.88	29.13	33.08	28.10	29.76	46.14	47.14	44.52	47.31	50.54	58.28	44.52	47.31	50.54	58.28		
	200	28.14	33.39	37.32	40.73	34.76	36.67	48.57	53.81	24.73	23.66	22.58	25.48	24.73	23.66	22.58	25.48		
	400	20.26	25.51	29.45	30.39	24.29	19.52	27.86	25.95	20.22	16.45	15.05	19.68	20.22	16.45	15.05	19.68		
	800	15.51	19.45	21.07	26.01	19.33	15.48	23.81	27.14										
Plantavax																			
	50	18.75	22.86	40.71	44.29	18.79	29.38	39.97	42.80	14.48	37.93	31.03	28.00	17.24	32.76	26.21	29.56		
	100	21.43	25.71	31.43	34.29	23.73	30.79	40.68	43.50	29.93	31.31	22.83	33.66	29.93	31.31	22.83	33.66		
	200	27.86	31.43	37.86	41.43	35.03	36.45	37.85	39.27	25.52	28.28	36.55	37.29	25.52	28.28	36.55	37.29		
	400	24.29	29.29	35.00	40.00	21.19	29.38	32.20	35.03	41.38	43.45	45.52	50.34	41.38	43.45	45.52	50.34		
Bayleton	800	20.00	25.00	30.00	37.86	17.49	16.67	28.81	32.20										
	50	18.27	15.38	26.93	48.08	12.17	21.74	34.78	43.47	21.74	47.83	56.52	63.04	29.35	47.83	56.52	63.04		
	100	17.31	15.38	28.85	38.46	13.81	26.09	39.13	47.83	36.96	52.17	54.35	63.04	36.96	52.17	54.35	63.04		
	200	11.54	15.38	30.77	40.38	15.22	26.09	36.96	47.83	28.26	45.65	56.52	65.23	28.26	45.65	56.52	65.23		
Wolfen- thiram	400	23.08	21.15	32.69	44.23	13.04	26.09	39.13	47.83	39.13	47.83	58.70	65.23	39.13	47.83	58.70	65.23		
	800	28.85	21.15	30.77	42.31	13.04	30.43	41.30	50.00										
Dithane-M ₂₂	50	12.05	15.51	24.96	25.20	27.27	27.27	38.73	40.00	4.42	9.73	15.04	23.00	15.04	23.89	28.32	32.74		
	100	19.95	18.11	23.62	32.28	30.00	34.55	39.09	43.64	23.89	29.20	35.40	43.36	23.89	29.20	35.40	43.36		
	200	22.83	21.92	36.22	40.16	39.09	45.45	52.73	60.00	18.58	23.89	28.32	32.74	18.58	23.89	28.32	32.74		
	400	21.57	32.28	37.80	41.73	17.27	26.36	32.73	41.82	11.50	25.66	20.97	30.51	11.50	25.66	20.97	30.51		
	800	15.75	21.26	27.56	35.43	14.91	27.27	30.91	34.55										
Control	0.0	26.17	24.60	32.16	45.12	25.11	36.16	43.35	52.70	29.40	39.80	47.60	58.67						

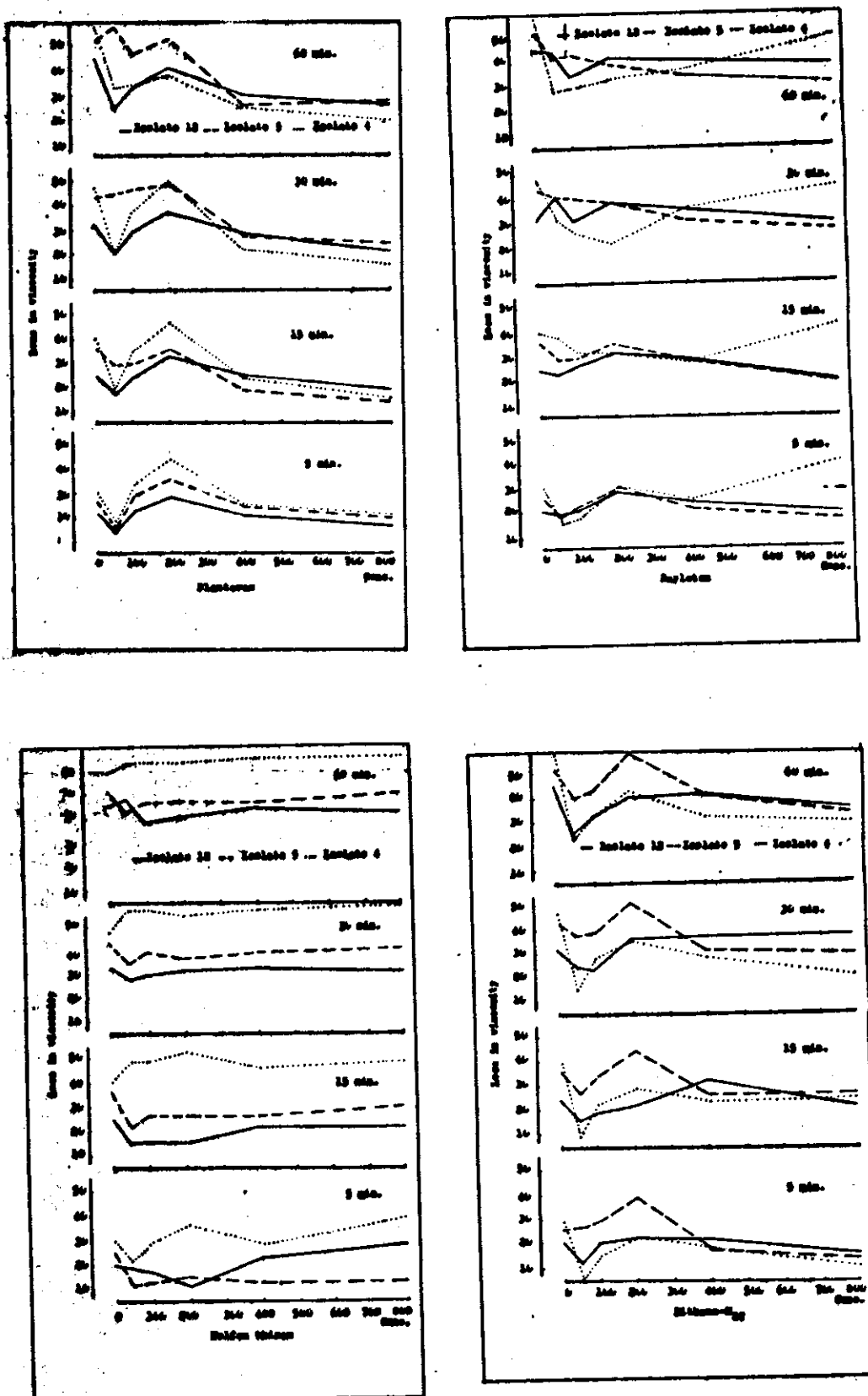


Fig. (4) Effect of some fungicides in fungal filtrates by using Talboys and Bush medium on cellulolytic activity (loss in viscosity) by the three isolates of *H. sativum*.

between the two tested isolates of the fungus in both the culture filtrates and mycelial matrix.

Mycelial matrix exhibited higher activity levels than the filtrates specially in the mycelial matrix and filtrate of the highly pathogenic isolate after 30 days.

Peroxidase activity:

Data presented in Table (12), indicate that peroxidase activity was relatively higher in mycelial matrix than in the filtrates in the two isolates. Data, also revealed that the highly pathogenic isolate showed the highest levels of enzyme activity in the mycelial matrix and filtrates. However, the highest levels of enzyme activity in both pathogenic isolates was noticed after 30 days from inoculation.

Catalase activity:

Data obtained in Table (13), show that catalase activity was higher in mycelial matrix than in filtrates for both isolates No. 4 and 12. However, Catalase activity was higher in isolate 4 (highly pathogenic) than isolate 5. Data, also revealed that the highest level of enzyme activity was observed after 30 days from incubation except in the filtrate of the least pathogenic isolate 5.

Table (11): Activity of polyphenol oxidase in both filtrates and mycelial matrix of isolates 4 and 12 of H. sativum for 2 and 4 weeks.

Isolates	Polyphenoloxidase activity/minute as optical density			
	filtrate after		Mycelial matrix after	
	15 days	30 days	15 days	30 days
highly pathogenic (a)	0.130	0.093	0.640	0.780
least pathogenic (b)	0.090	0.145	0.570	0.620

(a) Isolate No. 4.

(b) Isolate No. 12.

Table (12): Activity of peroxidase in both filtrate and mycelial matrix of isolates 4 and 12 of H. sativum for 15 and 30 days.

Isolates	Peroxidase activity/minute as optical density			
	filtrate after		Mycelial matrix after	
	15 days	30 days	15 days	30 days
highly pathogenic (a)	0.42	0.150	0.72	0.160
least pathogenic (b)	0.48	0.111	0.67	0.120

(a) Isolate No. 4

(b) Isolate No. 12

Table (13): Activity of Catalase in both filtrates and mycelial matrix of isolates 4 and 12 of H. sativum for 15 and 30 days.

Isolates	Catalase activity/minute as optical density			
	filtrate after		Mycelial matrix after	
	15 days	30 days	15 days	30 days
highly pathogenic ^(a)	0.447	0.499	0.509	0.592
least pathogenic ^(b)	0.415	0.363	0.389	0.460

(a) Isolate No. 4
(b) Isolate No. 12

Table (14): Activity of ascorbic acid oxidase in both filtrates and mycelial matrix of isolates 4 and 12 of H. sativum for 15 and 30 days.

Isolates	Ascorbic acid oxidase/minute as optical density			
	filtrate after		Mycelial matrix after	
	15 days	30 days	15 days	30 days
highly pathogenic ^(a)	0.284	0.312	0.302	0.380
least pathogenic ^(b)	0.302	0.265	0.315	0.365

(a) Isolate No. 4
(b) Isolate No. 14

Ascorbic acid oxidase:

Data in Table (14), show that ascorbic acid oxidase activity was higher in the highly pathogenic isolate 4, than the least pathogenic isolate. Also, the activity of the enzyme was higher in the mycelial matrix than in fungal filtrates. The activity of ascorbic acid oxidase increased by increasing the incubation periods in both mycelial matrix and fungal filtrates except in the filtrate of the least pathogenic isolate 4 which decreased from after 30 days.

Determination of oxidative enzymes activities in the extracts of plants:

Polyphenoloxidase activities:

Data presented in Tables (15 and 16) and Fig. (5) indicate that adult plants showed high levels of enzymes activity as compared with the seedling stage. Infection with H. sativum increased in enzyme activity specially in the resistant varieties Mianwali and C.I. 7123. Also data show that the enzyme activity gradually increased in the seedling stage specially in the resistant varieties then slightly decreased with disease development in the adult stage.

Peroxidase activities:

It is clear from data presented in Tables (15 and 16) that infection with H. sativum increased the activity of peroxidase enzyme as compared with healthy tissues. This increase was more pronounced in resistant varieties C.I. 7123 and Mianwali followed by the susceptible ones Giza 117 and Traill. Also, it is clear that the adult plants showed the highest level of enzyme activity. However, decrease in enzyme activity was found after 2 days from inoculation. Ten days after inoculation, peroxidase activity increased again in the adult leaves.

Catalase activity:

Data presented in Tables (15 and 16) show that catalase activity was higher in seedling and adult leaves of the tested varieties and it is clear that enzyme activity was higher in the resistant varieties than in susceptible ones. Data, also, show that infection with H. sativum exhibited higher catalase activity specially in the resistant varieties C.I. 7123 and Mianwali than in the susceptible varieties. Giza 117 and Traill.

Table (15): Determination of oxidative enzymes activity, polyphenol oxidase, peroxidase, catalase and ascorbic acid oxidase in leaves of four berley varieties inoculated with H. sativum at seedling stage in vivo.

Enzymes	Polyphenol oxidase				Peroxidase				Catalase				Ascorbic acid oxidase			
	2 days after inoc.		10 days after inoc.		2 days after inoc.		10 days after inoc.		2 days after inoc.		10 days after inoc.		2 days after inoc.		10 days after inoc.	
	pre-inoculation	inoculation	pre-inoculation	inoculation	pre-inoculation	inoculation	pre-inoculation	inoculation	pre-inoculation	inoculation	pre-inoculation	inoculation	pre-inoculation	inoculation	pre-inoculation	inoculation
Varieties																
Uninoculated:																
Traill	S	0.340	0.270	0.390	0.31	0.26	0.25	2.278	2.642	2.668	0.615	0.724	0.791	0.605	0.689	0.760
Giza 117	S	0.360	0.300	0.370	0.32	0.28	0.26	2.177	2.309	2.357	1.241	1.532	1.717	1.241	1.811	1.950
C.I. 7123	R	0.370	0.390	0.450	0.34	0.35	0.42	2.440	3.187	3.198	1.623	1.721	1.815	1.623	1.767	1.934
Mainwali	R	0.400	0.410	0.460	0.37	0.87	0.40	2.516	2.982	3.202						
Inoculated:																
Traill	S	0.340	0.360	0.410	0.31	0.36	0.40	2.278	2.677	2.817	0.615	0.862	0.859	0.605	0.699	0.848
Giza 117	S	0.360	0.320	0.380	0.32	0.35	0.42	2.177	2.356	2.886	1.241	1.811	1.950	1.241	1.811	1.950
C.I. 7123	R	0.370	0.390	0.490	0.34	0.42	0.50	2.440	3.861	3.894	1.623	1.767	1.934	1.623	1.767	1.934
Mainwali	R	0.400	0.430	0.500	0.37	0.39	0.48	2.516	2.527	3.584						

Table (16): Determination of oxidative enzymes activity, polyphenol oxidase, peroxidase, catalase and ascorbic acid oxidase in leaves of four barley varieties inoculated with *H. sativum* at adults stage in vivo.

Enzymes	Polyphenol oxidase				Peroxidase				Catalase				Ascorbic acid oxidase			
	pre-inoculation		2 days after inoc.		pre-inoculation		2 days after inoc.		pre-inoculation		2 days after inoc.		pre-inoculation		2 days after inoc.	
	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.	10 days after inoc.	2 days after inoc.
Varieties																
Uninoculated:																
Freall	S	0.600	0.590	0.320	0.56	0.55	0.45	2.171	2.129	2.201	0.460	0.761	0.915			
Giza 117	S	0.580	0.400	0.340	0.50	0.29	0.37	2.973	2.926	2.994	0.540	0.556	0.928			
C.I. 7123	R	0.700	0.350	0.350	0.71	0.62	0.66	3.111	3.171	3.448	1.183	1.359	1.768			
Mainwali	R	0.950	0.480	0.460	0.90	0.77	0.78	2.671	3.130	3.327	1.667	1.730	1.897			
Inoculated:																
Freall	S	0.600	0.570	0.510	0.56	0.48	0.52	2.171	3.340	2.914	0.460	0.769	1.039			
Giza 117	S	0.580	0.500	0.460	0.50	0.40	0.46	2.973	3.171	3.286	0.540	1.123	1.236			
C.I. 7123	R	0.700	0.580	0.530	0.71	0.57	0.68	3.111	3.536	3.564	1.183	1.756	1.838			
Mainwali	R	0.950	0.580	0.570	0.90	0.78	0.82	2.671	3.563	3.734	1.667	1.938	1.976			

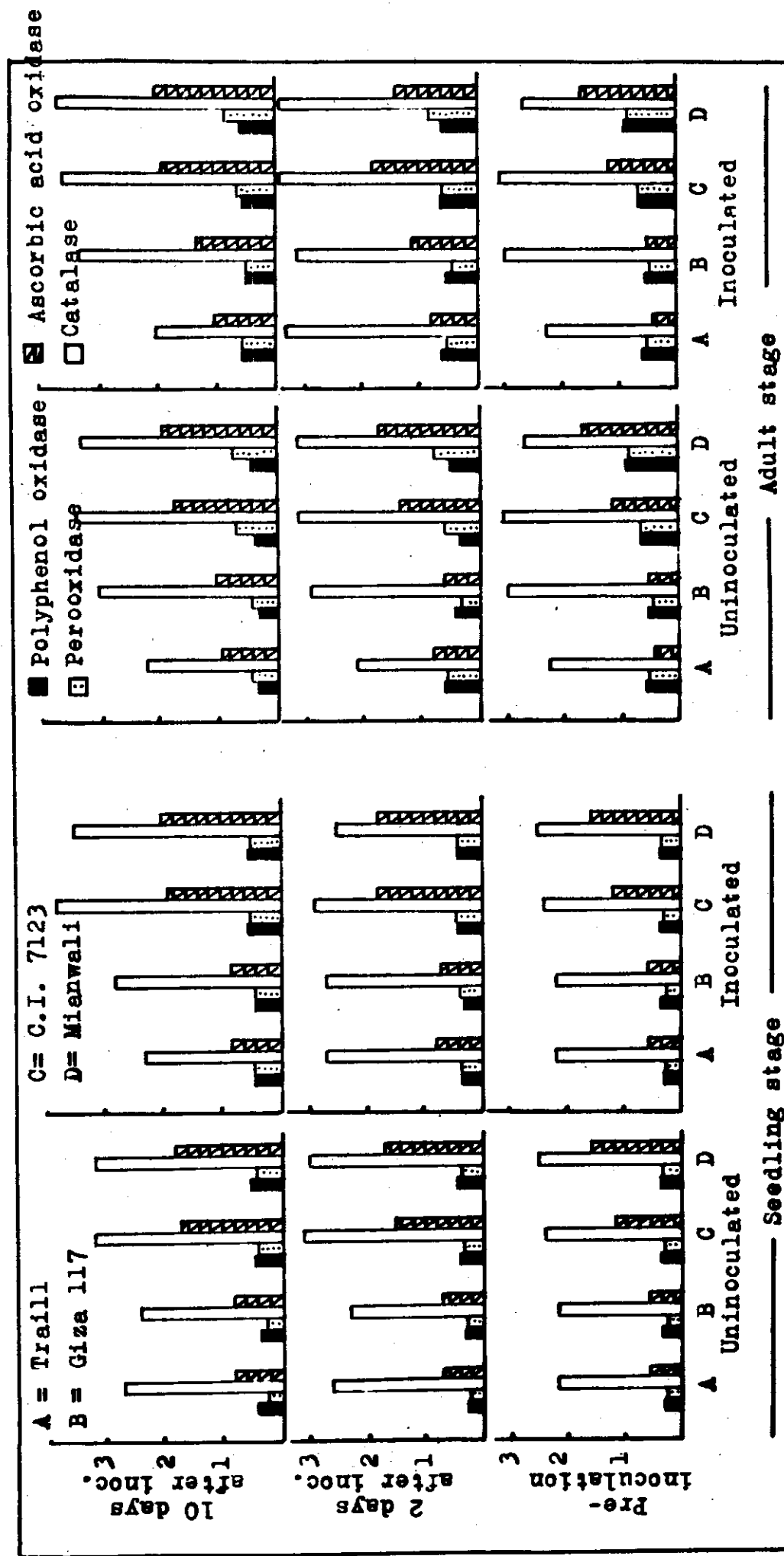


Fig. (5) Determination of oxidative enzymes activity, polyphenol oxidase, peroxidase, catalase and ascorbic acid oxidase in leaves of four barley varieties inoculated with *H. sativum* at seedling and adult stages.

Ascorbic acid oxidase:

Results in Tables (15 and 16) show that healthy leaves from seedling and adult plants of the resistant varieties Mianwali and C.I. 7123 exhibited higher activity of the enzyme than that in the susceptible Triall and Giza 117 varieties. Inoculation with H. sativum resulted an increase in ascorbic acid oxidase activity specially in the resistant varieties than in the susceptible ones and also, in adult plants than in the seedling stage.

Biochemical changes associated with resistance to spot blotch disease:

A- Phenolic compounds:

Total, free and conjugated phenolic contents, as well as, ortho-dihydroxyphenols were determined as mg equivalent of 3,4-dihydroxy benzene (protocatechol = catechol) per 10 gm fresh weight of barley leaves from a catechol standard curve. The obtained data are presented in Tables (17 and 18) and illustrated in Fig. (6). The four barley varieties differed greatly with respect to amounts of phenols in their leaves at seedling and adult stages of growth. Generally, the amounts of phenols were higher in leaves of adult plants than those of the seedling stage.

Total phenols were found with higher amounts in leaves of the resistant variety Mianwali than the other tested varieties. Generally, the resistant varieties (Mianwali and C.I. 7123) contained higher amounts of total phenols than the susceptible varieties (Traill and Giza 117). It was found also that, inoculation with H. sativum increased the amount of total phenols in resistant as well as in susceptible ones. It was clear that the total amounts of phenols increased gradually in both inoculated and healthy leaves of resistant and susceptible varieties and exhibiting the highest amounts after 10 days from inoculation (completely disease development).

Concerning free phenols Tables (17, 18) and Fig. (6). It was found that the healthy leaves of the resistant barley variety Mianwali exhibited the highest amounts of free phenols comparing with the other tested varieties. It was clear that the same trend of results was observed in both seedling and adult plant stages. Generally, free phenols were found to be higher in inoculated tissues than the uninoculated ones and the accumulation of free phenols, due to inoculation, was higher in the resistant varieties than the susceptible ones especially the resistant variety Mianwali in the seedling stage.

Table (18): Total, Free and conjugated phenols and ortho-dihydroxyphenols in leaves of four barley varieties inoculated with H. sativum at adult stage calculated as mg/10 g fresh weight.

calculated as mg/10 g fresh weight														
Varieties	Infection type	Total after inoculation			Free after inoculation			Conjugated after inoculation			Ortho-dihydroxy phenol after inoculation			
		pre-inoculation			pre-inoculation			pre-inoculation			pre-inoculation			
		2 days	10 days	20 days	2 days	10 days	20 days	2 days	10 days	20 days	2 days	10 days	20 days	
<u>Uninoculated:</u>														
Traill	S	9.87	11.88	13.23	6.11	7.48	10.08	3.76	4.40	3.15	1.72	2.34	3.11	
Giza 117	S	13.45	19.27	23.74	9.54	17.26	18.02	3.91	2.01	5.72	2.70	2.86	2.55	
C.I. 7123	R	15.55	24.51	26.99	10.17	19.25	21.27	5.38	5.26	5.72	3.18	3.04	2.51	
Mianwali	R	14.40	26.33	26.11	10.40	18.77	18.04	4.00	7.26	8.07	3.29	3.46	2.05	
<u>Inoculated</u>														
Traill	S	9.87	13.56	24.22	6.11	10.90	18.51	3.76	2.66	5.71	1.72	2.34	4.85	
Giza 117	S	13.45	19.81	32.51	9.54	16.04	24.69	3.91	3.77	7.82	2.70	3.49	3.80	
C.I. 7123	R	15.55	28.64	36.04	10.17	19.67	21.52	5.38	8.97	14.52	3.18	4.04	5.08	
Mianwali	R	14.40	28.43	33.40	10.40	20.69	24.40	4.00	7.74	9.00	3.29	4.18	5.45	

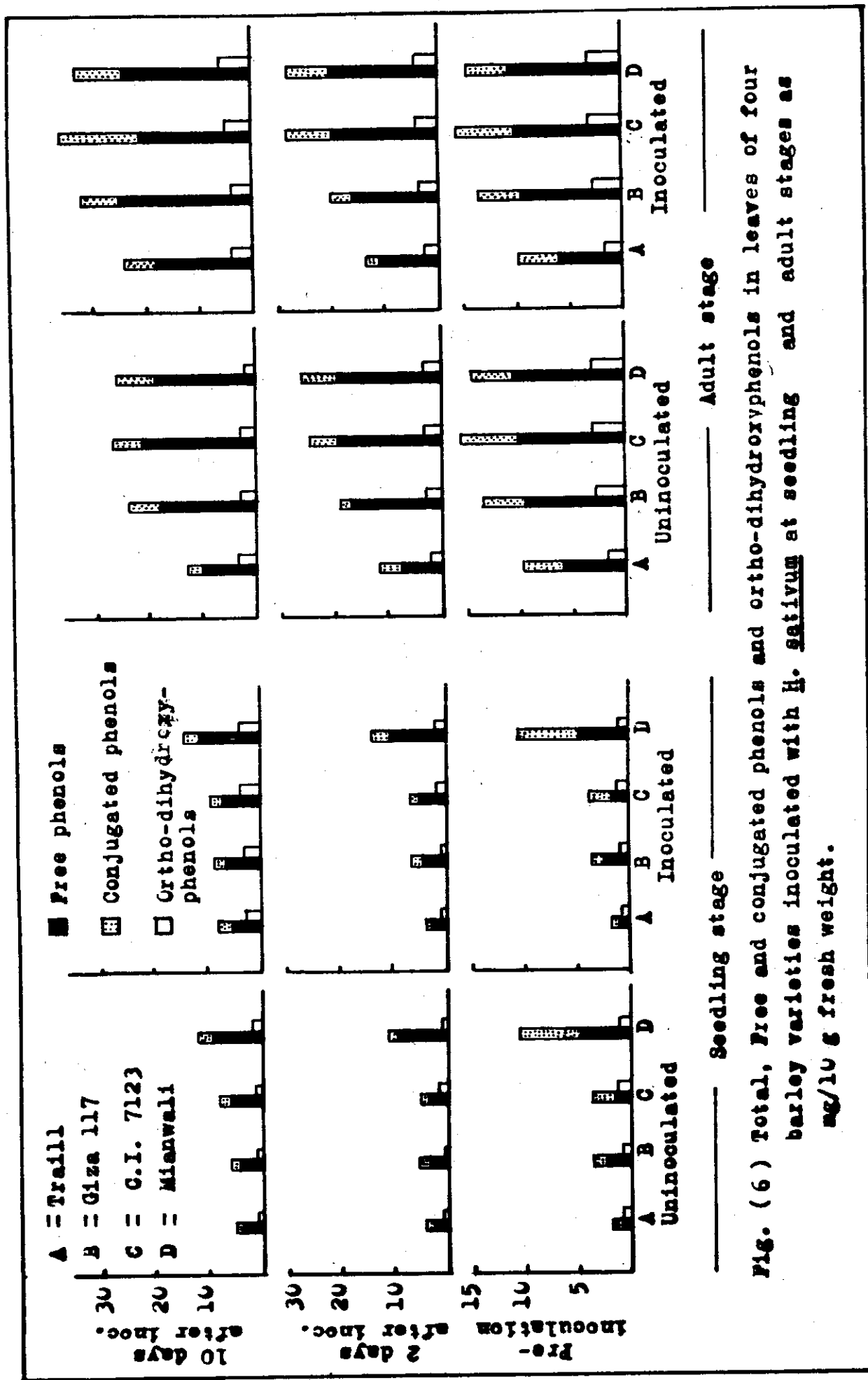


Fig. (6) Total, Free and conjugated phenols and ortho-dihydroxyphenols in leaves of four barley varieties inoculated with *H. sativum* at seedling and adult stages as mg/10 g fresh weight.

Conjugated phenols accumulated faster after inoculation in leaves of the resistant varieties in both seedling and adult stages. As regards the conjugated phenols it was noticed that uninoculated leaves contained higher levels than the inoculated ones in the two susceptible varieties.

However, at the time of complete disease development (10 days after inoculation) conjugated phenols decreased in inoculated tissues of susceptible variety Giza 117 and resistant variety G.I. 7123 in the seedling stage.

Regarding the ortho-dihydroxyphenols it was clear that inoculated leaves contained higher levels of ortho-dihydroxyphenols than the uninoculated ones in all tested varieties.

Ortho-dihydroxyphenols increased continuously with disease development and reached the maximum level after 10 days from inoculation in all resistant and susceptible varieties. It is of much importance to record that the higher amounts of ortho-dihydroxyphenols were noticed in inoculated leaves of Mianwali and C.I. 7123 resistant varieties in the adult stage after 10 days of inoculation as compared with the other tested varieties.

B- Soluble sugars:

Total, reducing and non-reducing sugars were quantitatively determined in seedling and adult plants leaves of the four tested barley varieties before or after 2 and 10 days from inoculation with H. sativum as mg equivalent of glucose per 10 gm fresh weight, using a standard curve according to the methods recorded by Thomas and Dutcher (1924).

Data are presented in Tables (19, 20) and Fig. (7) show that total sugars contents were higher in healthy as well as inoculated leaves of the adult plants than in seedling leaves of the four tested barley varieties. Total sugar contents, in seedling leaves of the susceptible varieties, were higher in susceptible varieties leaves than the resistant ones.

On the other hand, inoculation with H. sativum caused pronounced decrease in the amount of total sugars after 2 and 10 days in seedling or adult plants of susceptible varieties, except in Giza 117 seedling after 10 days. On the contrary, inoculation of the resistant Mianwali and C.I. 7123 varieties induced a considerable increase in the amounts of total sugars in the leaves from seedlings and adult plants after 2 and 10 days except Mianwali adult plants after 2 days from inoculation.

Table (19): Total, Reducing and Non-reducing sugars content in leaves of four barley varieties inoculated with H. sativum at seedling stage calculated as mg/10 g fresh weight.

mg/10 g fresh weight.										
Varieties	Infection type	Total after inoculation			Reducing after inoculation			Non-reducing after inoculation		
		pre-	2 days	10 days	pre-	2 days	10 days	pre-	2 days	10 days
<u>Uninoculated:</u>										
Trail	S	65.37	45.55	35.23	46.04	29.08	20.07	19.33	17.47	15.16
Giza 117	S	52.93	41.58	28.93	30.78	23.43	19.44	22.15	18.15	9.49
C.I. 7123	R	32.58	52.20	61.02	20.06	28.37	31.45	12.52	23.83	29.57
Mianwali	R	29.02	51.23	57.18	20.08	39.07	43.05	8.94	12.16	14.13
<u>Inoculated:</u>										
Trail	S	65.37	35.41	33.15	46.04	21.04	20.41	19.33	14.36	12.74
Giza 117	S	52.93	38.41	32.21	30.78	80.05	19.43	22.15	18.36	12.78
C.I. 7123	R	32.58	66.06	86.74	20.06	46.03	58.15	12.52	20.03	28.49
Mianwali	R	29.02	79.53	94.25	20.08	57.02	68.07	8.94	22.51	26.18

Table (20): Total, Reducing and Non-reducing sugars content in leaves of four barley varieties inoculated with H. sativum at adult stage calculated as mg/10 g fresh weight.

as mg/10 g fresh wt											
Varieties	Infection type	Total after in-oculation			Reducing after inoculation			Non-reducing after inoculation			
		pre-	2 days	10 days	pre-	2 days	10 days	pre-	2 days	10 days	
<u>Uninoculated:</u>											
Trail	S	95.40	88.89	67.47	76.70	64.05	41.37	18.70	24.84	26.10	192
Giza 117	S	87.21	85.64	56.72	75.93	56.19	50.23	11.28	29.45	6.49	1
C.I. 7123	R	56.52	87.45	86.44	43.48	54.08	76.74	13.04	33.37	9.70	1
Mianwali	R	49.23	89.89	101.34	38.76	59.39	58.76	10.47	30.50	42.67	1
<u>Inoculated:</u>											
Trail	S	95.40	65.44	64.30	76.70	44.74	48.70	18.70	20.70	15.60	1
Giza 117	S	87.21	68.19	56.53	75.93	46.81	41.36	11.28	21.38	15.17	1
C.I. 7123	R	56.52	88.14	110.32	43.48	58.43	77.57	13.04	29.71	32.75	1
Mianwali	R	49.23	87.46	125.40	38.76	69.39	78.63	10.47	18.07	46.77	1

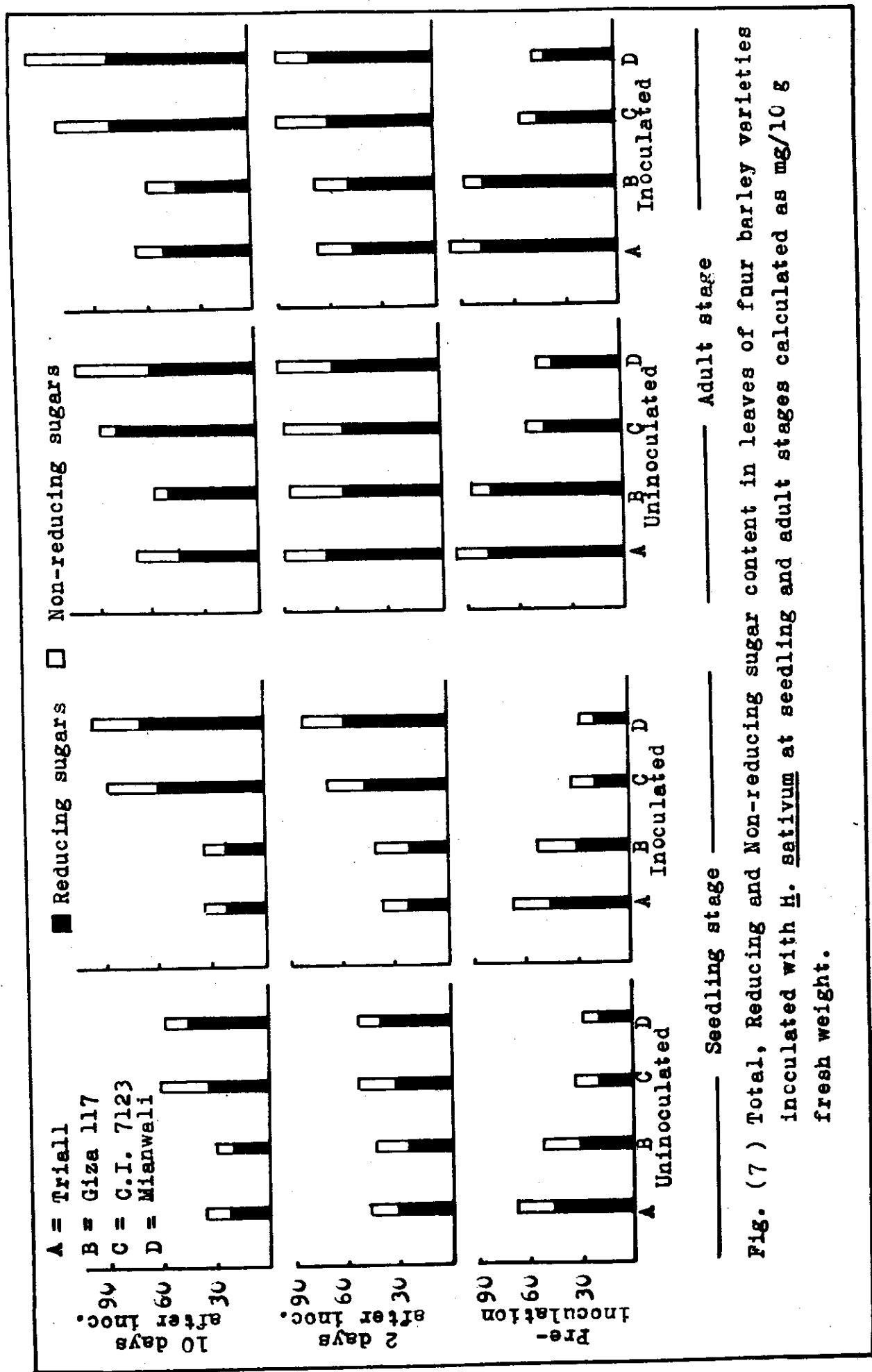


Fig. (7) Total, Reducing and Non-reducing sugar content in leaves of four barley varieties inoculated with *H. sativum* at seedling and adult stages calculated as mg/10 g fresh weight.

Same trend of results was noticed as regards the reducing sugars in the four barley varieties which indicate that inoculated leaves of the resistant varieties Mianwali and C.I. 7123 contained higher levels of reducing sugars than uninoculated ones as a result of infection with H. sativum in both seedling and adult plants.

Concerning non-reducing sugars the results were similar to that found in reducing sugars in both seedling and adult plants with few exceptions. Generally, inoculated tissues contained less amounts of non-reducing sugars than healthy tissues except the resistant barley variety Mianwali.

Individual sugars:

Qualitative analysis:

Chromatographic analysis of leaf extracts indicate the presence of Maltose, Sucrose, Glucose, Galactose and Fructose in inoculated and uninoculated leaves of all the tested varieties in both seedling and adult plants.

Quantitative analysis:

Data presented in Table (21) show that the adult plants contained higher levels of individual sugars than in the seedling stage.

Table (21): Individual sugars (mg/10 g fresh weight) in inoculated and uninoculated leaves with H. sativum of barley varieties in both seedling and adult stages.

Varieties	Infection type	Sugar %	Seedling stage						Adult stage						
			Healthy			Inoculated			Healthy			Inoculated			
			Days after inoculation						Days after inoculation						
			0	2	10	0	2	10	0	2	10	0	2	10	
Gize 117	S	*	A	1.23	2.16	2.08	1.23	2.32	2.16	6.16	8.32	10.32	6.16	4.16	7.38
			B	3.75	5.50	6.20	3.75	4.25	4.15	9.48	12.59	10.48	9.48	10.32	10.28
			C	3.50	3.16	3.16	3.50	3.48	2.32	12.32	13.48	15.32	12.32	11.32	11.19
			D	2.49	1.50	2.24	2.49	2.16	3.24	5.32	8.32	7.18	5.32	6.32	9.19
Mainwali	R	*	A	2.25	1.75	2.17	2.25	3.10	4.32	8.28	10.32	11.35	8.28	10.16	13.48
			B	3.31	6.50	7.23	3.31	9.21	9.48	10.68	14.48	16.46	10.68	14.48	19.16
			C	4.25	5.25	5.24	4.25	8.13	10.16	14.42	16.48	18.21	14.42	13.32	17.48
			D	1.23	2.21	3.48	1.23	3.10	4.16	6.68	9.64	13.25	6.68	9.32	11.16

B = Sucrose

D = Fructose

* A = Maltose

C = Glucose + Galactose

The level of Maltose slightly increased after inoculation specially in the resistant variety whereas, it decreased in the susceptible variety.

Sucrose increased gradually after inoculation specially in the resistant variety but it decreased in the susceptible one.

Glucose and Galactose increased after inoculation in the seedling and adult stages of mainwali resistant variety and decreased after 2 or 10 days from inoculation in the susceptible Giza 117 variety.

As regards the amounts of Fructose it varied from one variety to another and between the inoculated and uninoculated leaves with no general trend.

Generally, inoculation with H. sativum cause an increase of different sugars in the resistant variety Mianwali whereas, it decreased in the susceptible variety Giza 117 and there is no clear correlation with resistance or susceptibility of barley varieties to spot blotch infection.

C- Free amino acids:

Quantitative and qualitative analysis:

Quantitative determination of total free amino acids in extracts of barley leaves was made according

to the method of buffer acetate (Rosen, 1957). The intensity of colour was measured at 520 nm on Spectrocolourmeter (Carl, Zeiss, Jena). Also, qualitative analysis was estimated by paper chromatograms. The solvent consisted of n-butanol : acetic acid:distilled water at the rate of 4 : 1 : 5 by volume, respectively according to Heftmann (1967). Colouration was obtained by spraying the chromatograms with a solution of 0.2 nin -hydrin in acetone (W/V), Smith (1969). Thereafter each spot was dissolved in 50% methanol and was determined at 520 nm for all amino acids except proline which was determined at 430 nm.

Total free amino acids was evaluated as mg/10 g fresh weight by using the modified colourimetric nin-hydrin method, in leaves extracts of healthy and inoculated barley plants with H. sativum. Data presented in Table (22) show that the total free amino acids were highest in the extracts of all varieties in adult stage as compared with seedling stage. It was also observed that inoculation of seedlings and adult plants of the four varieties with H. sativum produced appreciable increase in the amount of total amino acids during 2 and 10 days after inoculation. Also, the amounts of total free amino acids were higher after 10

Table (22): Total-free amino acids in leaves of four barley varieties inoculated with *H. sativum* at seedling and adult stages calculated as mg/10 g fresh weight.

Varieties	Infection type	pre-inoculation	Days after inoculation			
			Uninoculated		Inoculated	
			2 days	10 days	2 days	10 days
<u>In seedling stage:</u>						
Traill	S	30.26	33.68	45.81	39.50	56.49
Giza 117	S	32.96	32.85	43.18	39.24	53.80
C.I. 7123	R	29.31	32.00	34.93	35.46	37.79
Mianwali	R	27.79	30.86	37.85	36.35	43.13
<u>In adult stage:</u>						
Traill	S	51.56	49.77	66.94	57.38	76.88
Giza 117	S	56.90	53.88	58.15	60.49	83.94
C.I. 7123	R	51.42	45.90	40.99	51.44	52.31
Mianwali	R	49.60	47.89	42.12	49.20	51.13

days from inoculation than 2 days. However, susceptible varieties (Traill and Giza 117) contained higher amounts of total free amino acids than the resistant varieties (C.I. 7123 and Mianwali) and also the increase which resulted after inoculation was more than resistant varieties.

In addition, seventeen amino acids, i.e., Cystine, Lysine, Histidine, Arginine, Aspartic acid, Glycine, Serine, Glutamic, Threonine, Alanine, Proline, Tyrosine, Methionine, Valine, Phenylalanine, Leucine and Iso-leucine were quantitatively determined, using paper chromatography, in barley leaves obtained from seedlings and adult plants either before or after 2 and 10 days from inoculation with H. sativum.

Samples of the four barley varieties were taken from inoculated and uninoculated leaves of seedlings (14 days old) and adult plants (at heading stage) at 3 periods (pre, 2 and 10 days from inoculation). In this method the amounts of amino acids as mg/10 gm fresh weight in all treatments are shown in Tables (23, 24, 25, 26 and 27).

The seventeen amino acids were found in different levels in leaves of both seedlings and adult plants and varied greatly in all treatments. At seedling and adult

Table (23): Free amino acids contents in leaves of four barley varieties inoculated with *H. sativum* at seedling and adult stages pre-inoculation calculated as mg/10 g fresh weight.

Amino acids		Infection type											
		Phenyl-alanine											
		Methionine + Valine											
		Tyrosine											
		Proline											
		Alanine											
		Glutamic + Therionine											
		Glycine + serine											
		Asparatic acid											
		Arginine											
		Histidine											
		Lycine											
		Cystine											
		Leucine + Isoleucine											
Varieties													
In seedling stage:													
Triall	S	1.74	1.43	1.33	2.22	0.91	0.93	4.82	5.20	4.96	1.23	1.95	0.48 1.86
Giza 117	R	1.57	1.61	2.69	3.90	0.51	1.13	5.90	3.29	4.15	1.52	2.65	0.69 2.35
G.I. 7123	R	1.35	1.97	1.39	3.43	0.36	1.09	2.56	2.26	3.45	5.54	2.87	2.82 1.52
Mainwall	R	1.26	1.12	1.16	3.43	0.39	1.87	2.08	2.30	2.75	6.26	1.43	1.26 1.48
In adult stage:													
Triall	S	4.86	3.61	3.52	3.43	1.78	1.56	10.51	5.90	5.95	3.22	3.52	1.35 2.35
Giza 117	S	4.96	2.93	4.26	4.04	1.29	2.78	9.68	7.46	4.81	2.95	3.52	2.43 3.61
G.I. 7123	R	3.87	1.20	2.67	4.13	1.91	3.01	8.68	4.34	3.62	6.07	3.54	4.82 2.56
Mainwall	R	3.43	1.52	2.74	4.36	0.94	3.48	7.51	4.00	2.97	8.52	2.43	5.35 1.35

Table (24): Free amino acids contents in leaves of four barley varieties inoculated with *H. sativum* at seedling stage after 2 days of inoculation calculated as mg/10 g fresh weight.

Amino acids		Leucine + Isoleucine	Phenyl-alanine	Methionine + Valine	Tyrosine	Proline	Alanine	Glutamic + Therionine	Glycine + serine	Aspartic acid	Arginine	Histidine	Lycine	Cystine	Infection type
Varieties															
<u>Uninoculated:</u>															
Triall	S	3.26	1.35	2.52	2.36	2.34	1.65	6.08	3.65	3.35	1.30	1.61	1.43	1.78	
Giza 117	S	3.61	1.74	1.52	2.34	1.00	1.39	5.64	2.95	3.15	2.69	2.17	1.78	1.87	
C.I. 7123	R	1.97	1.26	1.30	2.30	2.78	0.69	3.64	2.78	2.94	5.78	0.78	2.59	2.69	
Mianwali	R	1.58	1.35	1.17	2.04	5.82	0.26	2.26	1.65	2.54	6.23	0.35	3.35	2.26	
<u>Inoculated:</u>															
Triall	S	4.55	2.04	1.73	5.16	1.17	1.20	13.12	0.43	3.41	1.22	3.12	1.17	1.78	
Giza 117	S	3.90	1.97	2.87	2.35	5.03	1.04	10.20	1.04	3.54	1.43	1.13	1.62	2.26	
C.I. 7123	R	2.17	1.34	1.30	2.30	3.65	3.04	5.08	1.74	1.54	6.04	2.52	2.78	2.26	
Mianwali	R	1.93	1.65	1.39	2.17	3.51	1.43	4.47	3.39	1.73	7.78	1.69	3.65	3.56	

Table (25): Free amino acids contents in leaves of four barley varieties inoculated with *H. sativum* at seedling stage after 10 days of inoculation calculated as mg/10 g fresh weight.

Varieties	Amino acids												
	Infection type												
	Leucine + Isoleucine	Phenyl-alanine	Methionine + Valine	Tyrosine	Proline	Alanine	Glutamic + Therionine	Glycine + serine	Asparatic acid	Arginine	Histidine	Lycine	Cystine
<u>Uninoculated:</u>													
Traill	S	5.17	2.52	3.22	2.78	2.17	2.43	8.69	5.21	3.94	1.85	1.74	1.50
Giza 117	S	4.17	2.61	2.65	2.80	2.69	2.34	8.69	6.08	3.84	2.85	2.34	1.82
G.I. 7123	R	1.26	1.04	1.52	2.08	0.35	5.21	5.60	1.04	2.97	7.78	0.70	3.43
Mienwali	R	1.52	0.78	1.65	2.17	1.95	3.78	4.10	5.20	2.88	8.34	1.30	2.78
<u>Inoculated:</u>													
Traill	S	6.35	2.21	3.30	5.64	4.47	7.29	12.91	1.39	4.12	1.48	1.87	1.17
Giza 117	S	5.74	2.82	2.89	6.86	2.56	4.60	13.47	5.15	3.70	1.61	1.69	1.13
G.I. 7123	R	2.43	1.04	1.80	2.08	2.43	1.74	6.94	2.78	2.70	8.12	1.13	3.78
Mienwali	R	1.59	1.67	1.92	3.32	3.71	2.93	4.30	2.25	1.99	9.34	2.34	4.34

Table (26): Free amino acids contents in leaves of four barley varieties inoculated with *H. sativum* at adult stage after 2 days of inoculation calculated as mg/10 g fresh weight.

Varieties	Amino acids													
	Leucine + Isoleucine	Phenyl-alanine	Methionine + Valine	Tyrosine	Proline	Alanine	Glutamic + Therionine	Glycine + serine	Asparatic acid	Arginine	Histidine	Lycine	Cystine	Infection type
Uninoculated:														
Trail	S	4.87	2.35	3.43	3.52	3.12	1.91	8.25	5.56	4.91	4.69	3.48	1.26	2.52
Giza 117	S	4.66	2.54	4.01	3.91	3.30	2.82	8.68	5.04	5.36	3.87	3.91	1.74	3.04
C.I. 7123	R	3.63	3.21	2.08	2.61	4.95	3.34	2.00	4.61	3.57	5.91	2.91	2.69	2.39
Mianwali	R.	3.52	1.52	1.26	2.25	8.68	4.95	2.95	4.69	2.98	6.65	2.48	3.48	1.48
Inoculated:														
Trail	S	1.04	2.65	4.82	5.52	4.74	1.48	11.68	8.42	4.25	4.74	4.39	1.26	3.39
Giza 117	S	0.52	2.17	5.73	4.81	6.60	2.78	10.87	7.09	6.37	5.99	5.39	1.69	3.78
C.I. 7123	R	0.61	1.69	2.26	2.68	7.81	7.64	5.26	3.30	2.01	8.82	2.00	3.95	2.41
Mianwali	R	0.35	1.26	1.30	1.34	8.68	5.90	6.48	3.43	1.33	10.38	1.60	4.61	1.54

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varieties than the susceptible ones and increased with increasing the age of plants before and after inoculation with pathogen. The amounts of aromatic amino acids increased in the uninoculated susceptible seedling varieties from 2 to 10 days while it decreased in the resistant ones. Also, in the seedling stage inoculation with H. sativum increased the level of aromatic amino acids in the resistant variety after 2 and 10 days. On the other hand, it decreased in the susceptible ones. In the adult stage the amounts of aromatic amino acids decreased in the uninoculated susceptible varieties from 2 to 10 days while it increased in the resistant C.I. 7123 variety only. The inoculation of H. sativum increased the amounts of aromatic amino acids in the resistant and susceptible varieties after 2 and 10 days except phenylalanine in the susceptible varieties after 2 days which it decreased and not change in Giza 117 and Traill varieties, respectively.

The inoculation with the pathogen increased Methionine and Valine amino acids in the resistant varieties after 2 and 10 days in the seedling stage, on contrary, it decreased in the adult stage. The amounts of Methionine and Valine were appeared with no general trend in the susceptible varieties.

The other amino acids increasing and decreasing in the susceptible and resistant varieties with no general trend in the seedling after 2 and 10 days from inoculation. Also, the results in the adult stage show the same trend.

Table (28): Effect of amino acids on the growth and dry weight of H. sativum in vitro.

Dry weight in gm.	Amino acids
1.1355	Lycine
1.1033	Histidine
1.1767	Arginine
1.2082	Tyrosine
1.1339	Phenyl- alanine
1.1553	Proline
1.1329	Lycine + Histidine + Arginine
1.0717	Tyrosine + phenyl- alanine
1.1272	Lycine + Histidine + Arginine + Tyrosine + phenyl- alanine + Proline
1.1172	Control*

* Without amino acids.

Data in Table (28) show clearly that the growth of H. sativum is affected by the amino acids. In this respect Arginine and proline enhanced the growth greatly on contrast with the combination between Tyrosine and Phenylalanine. Also, except in case

of Histidine and the combination between Tyrosine and phenylalanine the tested amino acids increased the growth than the control.

Toxicological studies:

1. Effect of different concentrations of 15 or 30 days crude culture filtrates of two isolates of *H. sativum* on the percentage of seed germination, coleoptile elongation and length of roots:

Two isolates of *H. sativum* differing in their virulence, were grown on potato-dextrose broth (PDB) and incubated at 25°C, for 30 days. PDB-medium was inoculated with equal discs of each isolate. Twenty conical flasks (250 ml.) were used as replicates for each isolate. Culture filtrates were obtained after 15 or 30 days from inoculation and were filtered, then the filtrates were centrifuged at 3000 rpm for 20 min. (Lindberg 1970).

Different concentrations of the filtrate of each isolate i.e. crude filtrate, 75, 50, 25% and sterile distilled water as control were used. Hundred grains of each of Giza 117 (susceptible) and C.I. 7123 (Resistant) varieties were used and divided into ten dishes

as 10 replicates for each variety. The grains in each dish were watered with each of the prepared dilutions of filtrates.

Grain germination percentages, coleoptile elongation and roots lengths were recorded as mentioned before. Results are shown in Table (29).

Data in Table (29), indicate that the crude filtrate of the two isolates induced inhibitory effect on the percentages of seed germination and lengths of coleoptile and roots on both Giza 117 and C.I. 7123 barley varieties. However, the susceptible variety Giza 117 was more sensitive to the toxic effect of the crude filtrate and its dilutions more than the resistant one. The high concentration of crude filtrate was the most effective in reducing the percentage of seed germination, coleoptile elongation and root growth of plants of both tested varieties.

Data also show that the crude filtrate isolated from 30 days old cultures, was the most effective in this respect than those obtained from 15 days old cultures, proving that the inhibitory effect increased by ageing of the cultures.

There was also a correlation between the virulence of the isolate and its toxic effect, in this regard

Table (29): Effect of different concentrations of 15 or 30 days crude culture filtrate of two isolates of H. sativum on the percentage of seed germination, coleoptile elongation and length of roots (cm.).

Concentration	Var.	Giza 117				C.I. 7123			
		4		12		4		12	
		Isol.		Isol.		Isol.		Isol.	
		Days	15	30	15	30	15	30	15
Crude filtrate %	s.g.	45.0	22.5	57.5	37.5	57.5	50.0	65.0	65.0
	c.e.	1.3	3.5	1.3	10.5	1.0	4.1	2.4	2.6
	r.l.	0.8	3.1	0.9	4.8	1.0	4.6	1.2	1.7
75	s.g.	50.0	47.5	60.0	55.0	77.5	70.0	87.5	72.5
	c.e.	2.2	8.7	2.4	11.2	3.8	5.3	2.5	3.4
	r.l.	1.8	4.1	1.7	7.3	1.8	4.8	1.5	4.7
50	s.g.	70.0	55.0	75.0	72.5	85.0	65.0	87.5	72.5
	c.e.	4.5	13.1	4.1	13.7	6.7	7.4	5.7	5.7
	r.l.	3.7	5.7	3.9	8.4	2.9	3.8	2.3	4.0
25	s.g.	82.5	75.0	95.0	85.0	97.0	87.5	97.5	90.0
	c.e.	8.1	18.1	7.5	19.8	9.7	9.8	8.6	13.4
	r.l.	6.5	8.4	6.7	10.3	4.0	4.6	3.2	10.0
Control	s.g.	100	100	100	100	100	100	100	100
	c.e.	9.6	19.8	9.6	19.8	13.2	18.9	13.2	18.9
	r.l.	9.7	11.2	9.7	11.2	9.2	11.2	9.2	11.2

s.g. = % of seed germination c.e. = Coleoptile elongation
r.l. = root length.

<u>L.S.D. at 5%</u>	<u>for seed germination</u>	<u>for coleoptile</u>	<u>for root length</u>
Between varieties	2.228	0.401	0.550
,, periods	2.490	0.788	0.276
,, Isolates	1.575	0.283	0.389
,, Interaction	3.522	1.416	1.038

According to Turner (1971), these crystals were identified as Helminthosporal which is one of the phytotoxins produced by H. sativum, and Sativene an intermediate compound. The chemical structure of Helminthosporal is ($C_{15}H_{24}O_2$), and Sativene is ($C_{15}H_{24}$).

However the quantities of the crystalized toxin from each of the two isolates of H. sativum were weighted. Results as shown in Table (30) and Fig. (9).

Table (30) : Amounts of isolated toxins from two isolates of H. sativum differing in their pathogenicity to barley plants.

Isolate No.	Toxin gm./liter of culture filtrate
4 (Virulent)	4.633
12 (Less virulent)	2.461

Data indicate that the two isolates of H. sativum, differing in their pathogenicity differed also in the production of their toxins. In this respect isolate No. 4 the most virulent isolate produce higher amounts of toxin than the low virulent isolate No. 12.



Fig. (8): Rod crystals of toxin produced by
H. sativum.

3. Effect of different concentrations of the purified toxin on the percentage of seed germination, coleoptile elongation and length of roots:

The effect of different concentrations of isolated toxin of the aforementioned two isolates of H. sativum was studied on seed germination percentages, coleoptile elongation and root lengths.

Results in Table (31), indicated that the toxin was effective on both barley varieties and decreased greatly the percentages of germination, coleoptile elongation and root lengths. In this regard, there was a positive correlation between the concentration and its toxic effect and the lowest percentage of seed germination and shortest coleoptiles and roots were obtained in the highest concentrations of toxins. Crude toxin was the most effective in reducing and results show clearly that the inhibitory effect of the toxin decreased proportionally by decreasing its concentration.

Control studies:

There are two primary approaches to solve the problem of spot blotch control, the first is to select resistant parents for breeding resistant varieties which will exhibit immunity to all races of the causal

Table (31): Effect of different concentrations of the purified toxin of two isolates of H. sativum on the percentages of germination, coleoptile elongations and lengths of roots (cm.).

Concentration	Var.	Giza 117		C.I. 7123	
	Iso.	4	12	4	12
Crude Toxin	s.g.	0	47.50	0	0.00
	c.e.	0	1.68	0	2.48
	r.l.	0	5.60	0	1.20
75%	s.g.	27.50	57.50	35.00	40.00
	c.e.	1.55	3.05	0.75	1.40
	r.l.	1.85	2.60	1.83	3.18
50%	s.g.	55.00	75.00	65.00	72.50
	c.e.	6.05	6.10	3.78	5.53
	r.l.	5.20	4.78	2.38	3.10
25%	s.g.	70.00	87.50	77.50	90.00
	c.e.	13.63	13.78	11.63	11.45
	r.l.	8.08	9.45	7.13	6.83
Control	s.g.	100	100	100	100
	c.e.	15.53	15.70	12.75	12.95
	r. .	14.15	13.35	10.23	10.33

s.g. = % of seed germination.
r.l. = Root length.

c.e. = coleoptile elongation

<u>L.S.D. at 5%</u>	<u>For seed germination</u>	<u>For coleoptile</u>	<u>For root length</u>
Between varieties	1.900	0.241	0.547
,, isolates	1.900	0.153	1.320
,, concentration	2.010	1.324	0.030
,, Interaction	5.010	1.872	1.353

organism even under adverse climatic conditions. The second is to suppress disease development by means of treating seeds or spraying plants with specific fungicides. Studies of these two approaches were undertaken during the course of this study as follows:

A- Varietal resistance:

1. Screening varieties in seedling stage:

Thirty seven varieties were tested in seedling stage under greenhouse conditions, the plants were artificially inoculated and control plants were sprayed with water only. After 6 days of inoculation, reaction of infection on the different varieties were recorded according to Cook and Timian (1962).

Data in Table (32), indicate that 16 varieties were highly resistant and 5 varieties were moderately susceptible, while 16 cultivars were susceptible.

The resistant varieties were Roho, Palmella blue, CM 67, Bursat, C.I. 7792, A Hor 1179/59, A Hor 1182/59, A Hor 1187/59, AS 139/60, MCU 3832, IV/94, Mianwali, Anoidium, C.I. 4638 and Baladi 16.

2. Screening varieties in adult stage:

Forty three varieties were tested to study their resistance in field at Sakha Station, where heavy

Table (32): Reaction of barley varieties inoculated with H. sativum in seedling stage under greenhouse conditions.

Variety	Source	Spot blotch reaction	Variety	Source	Spot blotch reaction
Roho	Portugal	R	A Hor 1214/61	Turkey	S
Palmella Blue	"	R	A Hor 1176/61	"	S
CM 67	"	R	H Hor 1059/59	"	S
Lechtaler	"	MS	AS 139/60	Germany	R
Burat	USDA	R	A Hor 1126/61	"	S
C.I. 7792	"	R	A Hor 1169/59	"	S
C.I. 7123	"	R	AS 3205/61	"	S
G.I. 4838	"	R	MCU 3832	Colombia	R
Anoidium	"	R	Iv/94	Yugoslavia	R
C.I. 8821	"	S	Mianwali	India	R
Trell	"	S	C.I. 7207 x Oli-10 cr	PON	S
A Hor 1179/59	Turkey	R	268121/2		
A Hor 1182/59	"	R	IAR/H/485	RDISEN	S
A Hor 1187/59	"	R	Nutance - 22	USSR	S
A Hor 1184/59	"	MS	Dier Alla 108	Jordan	MS
A Hor 1345/61	"	MS	Baladi 16	Egypt	R
A Hor 741/59	"	MS	Sahrawy	"	S
A Hor 638/61	"	S	Giza 117	"	S
C.I. 8135	"	S	Balder	Sweden	S

R = Resistant.

S = Susceptible

MS = Moderately susceptible.

PON = Preliminary Observation Nursery.

USDA = United States Department of Agriculture.

RDISEN = Regional Disease and Insect Screening Nursery.

Table (33): Reaction of 44 barley varieties to H. sativum in adult stage
under field conditions.

Variety	Source	Spot blotch reaction	Variety	Source	Spot blotch reaction
C.I. 7835	USDA	R	H Hor 1058/59	Turkey	VS
C.I. 8517	"	R	H Hor 1065/61	"	VS
Dubois	"	R	H Hor 1081/81	"	VS
C.I. 8673	"	R	H Hor 401/59	"	VS
Opal B.	"	R	Delisa	Holland	R
C.I. 7123	"	R	Martha	Austrilia	R
Mensing	"	S	Cernigovskiu K 20329	USSR	MS
C.I. 7889	"	S	Nutance 6955	"	S
Traill	"	VS	Peroline	"	S
Mianwali	India	R	Wise	"	S
Hege 11965	"	R	Agio	"	S
MCU 3834	Colombia	R	Viner	"	S
MCU 3833	"	VS	R.T. Ramage	Portugal	S
Ricardo	Uruguay	R	IV/64	Yugoslavia	R
Mansholts groninger	Germany	R	IV / 93	"	VS
H Hor 1360/61	"	R	Club mariout	Egypt	MS
Mut. 4009	"	R	Bahtim	"	MS
A Hor 1172/59	"	MS	Giza 117	"	S
Mut. 3479	"	S	Giza 24	"	S
H Hor 402/59	Turkey	R	Local Borg El-Arab	"	S
H Hor 1068/61	"	R	Giza 122	"	S
H Hor 1082/58	"	R	Gold	Sweden	R

R = Resistant
MS = Moderately susceptible
VS = Very susceptible.
MR = Moderately resistant.
S = Susceptible

natural infection is usually observed. Seeds of each variety were sown in single row (3.5 m.), 20 cms. apart within each row. Reactions of infection were recorded about 2 weeks after plant heading.

Data in Table (33), indicate that the behaviour of 44 selected varieties under field conditions in adult plant stage were a high level of resistance to disease under investigation observed on 19 cultivars, whereas 4 varieties were moderately susceptible and 13 were susceptible while 8 varieties were very susceptible. The resistant varieties were C.I. 7835, C.I. 8517, Dubois, C.I. 8673, Hege 11965, Gold, Recardo, Mansholts groninger, H Hor 402/59, MCU 3834, H Hor 1068/61, H Hor 1082/58, H Hor 1360/61, Opal B., C.I. 7123, Mut. 4009, Delisa and Martha. These resistant varieties can be used as parents in breeding program for developing new resistant varieties to this disease.

3. Effect of host age on spot blotch resistance:

Studies on the effect of plant age on spot blotch resistance under artificial condition in seedling stage in an air-conditioned greenhouse and in adult stage in the field at Sakha Experiment Station under natural infection.

Data in Table (34), indicated that host age appears to play an important role on barley resistance to this disease. As regards the correlation between the behaviour of 45 varieties in both seedling and adult plant stages, it was found that these varieties could be put in two groups as shown in Table (33).

A- The first group in which there was no effect of plant age on the varietal resistance or susceptibility as follows:

1. Seven varieties namely CM 67 - Apam, B 106 x Research, Avt-Aths, A Hor 1168/59, H. Hor 1056/59, Suvenir k 20919 and IV/63 were resistant in both seedling and adult stages.
2. Fourteen varieties namely Galt, Enir, C.I. 7890, H. Hor 1067/59, Clipper, C.I. 7968, H. Hor 1055/59, Atlas 46, Narym, Giza 119, C.I. 7791, Polong, Presto and Giza 118 were susceptible in both seedling and adult stages.

B- The second group in which there was an important effect of plant age on the varietal resistance or susceptibility as follows:

1. Eleven varieties namely A. Hor 1171/61, A. Hor 1182/59, Bakaw 17, Lenta, C.I. 5286, Speciale,

Table (34): Effect of plant age on the varietal resistance to spot blotch

Variety	Source	Spot blotch reaction		Variety	Source	Spot blotch reaction	
		Seedlings	Adults			Seedlings	Adults
CM67-Apam	Syria	R	R	Galt	Portugal	MS	MS
B106 x Research	RDIN	R	R	Emir	"	MS	MS
Avt - Aths	PCW	R	R	Bonus	"	MS	R
A Hor 1168/59	Germany	R	R	Bakaw 17	USDA	MR	R
A Hor 1171/61	"	MR	R	Lenta	"	MR	R
Szekacs 31	"	MR	R	C.I. 5286	"	MR	R
A Hor 1123/61	"	S	R	Speciale	"	MR	R
A Hor 117C/59	"	MS	VS	C.I. 9539	"	MR	R
AS 140/60	"	MS	VS	C.I. 7890	"	MS	MS
H Hor 1056/59	Turkey	R	R	C.I. 7968	"	S	S
A Hor 118C/59	"	MR	R	Narym	"	S	S
A Hor 1182/59	"	MR	R	C.I. 7791	"	VS	VS
H Hor 1057/58	"	MR	R	Polong	"	VS	VS
A Hor 1177/61	"	MS	R	Presto	"	VS	VS
Tiroler landgerste	"	MS	R	Reka 1	Australia	MR	S
A Hor 639/61	"	S	R	Clipper	"	S	S
A Hor 1213/59	"	VS	R	Atlas 46	Africa	S	S
A Hor 1175/61	"	MS	VS	Local Marsa Matrouh	Egypt	MS	MR
A Hor 1186/51	"	MS	VS	Giza 120	"	MR	S
H Hor 1067/59	"	MS	MS	Giza 121	"	MR	S
H Hor 1055/59	"	S	S	Giza 119	"	S	S
Suvenir K 20919	USSR	R	R	Giza 118	"	VS	VS
IV/63	Yugoslavia	R	R				

A. Hor 1180/59, H. Hor 1057/ 58, Szekacs 31, C.I. 9539 and local Marsa Matrouh showed increasing resistance with age.

2. Six varieties namely A. Hor 639/61, A Hor 1123/61, Tiroler landgerste, A. Hor 1177/61, Bonus, and A. Hor 1213/59 were susceptible in the seedling stage and became resistant with age.

3. Seven varieties namely Reka 1, Giza 120, Giza 121, A. Hor 1170/59, A. Hor 1175/61, A. Hor 1186/51 and AS 140/60 showed increasing susceptibility with age.

B- Chemical control:

1. In the laboratory:

Effect of different concentrations of some fungicides on the linear growth of *H. sativum*:

The effect of 11 different systemic and non-systemic fungicides each with 9 concentrations i.e., 0, 10, 25, 50, 100, 200, 400, 800 and 1600 ppm on the linear growth of *H. sativum* was investigated in vitro and four petri dishes were used for each concentration and also the control. Czapek's medium was used in this experiment.

Table (35) Effect of different concentrations of some fungicides on the linear growth of *H. sativum* (cm.)

Fungicides	Concentrations in ppm						
	Control	10	25	50	100	200	400 800 1600
Bavistin	8.50	8.50	7.88	7.50	7.50	6.88	7.13 6.50 6.13
Vitavax	8.50	7.00	6.50	6.00	5.63	3.63	0.50 0.50 0.38
Wolfen Thiram	8.50	4.00	3.25	0.38	0.00	0.00	0.00 0.00 0.00
Brassicol	8.50	4.13	2.75	2.50	1.63	1.25	1.00 1.00 0.50
Sperguson	8.50	5.38	4.63	3.50	2.63	2.50	2.13 1.38 1.00
Dexon	8.50	8.50	8.50	6.75	4.75	2.63	0.00 0.00 0.00
Plantvax	8.50	7.13	4.50	4.50	3.63	2.25	0.75 0.00 0.00
Bayleton	8.50	5.25	4.13	3.38	2.63	1.88	1.75 1.13 0.88
Kilazen	8.50	6.75	6.25	5.75	4.50	2.25	1.50 1.75 1.00
Mithane M ⁴⁵	8.50	4.00	3.75	3.00	2.13	1.88	0.63 0.00 0.00
Dithare ²²	8.50	6.75	6.50	5.13	4.13	2.75	1.63 0.25 0.00

L.S.D.	0.05%	0.01%
Fungicides (F):	0.15	0.21
Concentrations (C):	0.14	0.19
Fx G	0.46	0.62

Data in Table (35) indicate that all the fungicides except Bavistin and Spergon almost stopped the linear growth of H. sativum at relatively low concentrations of 10 ppm. This effect increased with the increase in concentration of the fungicide specially Wolfenthiram, Dexon, Plantavax and Dithane M₄₅ completely inhibited the fungal growth at 50, 200, 400 and 400 ppm. respectively. On the other hand, the least effective fungicide was Bavistin and seemed to be unsatisfactory.

2. Greenhouse experiment:

a- Seed treatment:

The effect of the recommended dose of different fungicides on spot blotch disease of barley under greenhouse conditions is estimated and results are recorded in Table (36). Ten seeds were sown in every pot and ten replicates were carried-out for each treatment.

Generally, the tested fungicides increased the percentage of seed germination, the number of ears per pot, the weight of grains in gm. per pot and 100 grains weight. The severity of infection was reduced as the fungicides were applied as seed dressing specially Brassicol, Vitavax and Wolfen thiram. In this respect

Table (36): Effect of different fungicides as seed treatment on the control of spot blotch of barley under greenhouse conditions.

Fungicides	Percent- age of germin- ation	Severity of infection	Number of ears/ pot	Weight of grains (gm./ pot)	Weight of 100 grains (gm.)
Wolfen thiram	90.667	13.334	24.000	20.827	3.467
Sp ergon	100.000	20.000	21.667	16.237	3.030
Brassicol	90.667	10.000	16.334	9.308	3.170
Bavistin	90.000	40.000	14.334	9.989	2.777
Vitavax	80.000	13.000	17.334	11.389	2.831
Dexon	80.000	20.000	19.667	16.340	3.093
Control	70.000	70.000	12.000	7.742	1.960
L.S.D. 0.05	9.327	13.820	7.820	5.377	0.490

Bavistin was the least effective fungicide.

Results also indicate that the highest number of ears per pot was obtained when the grains were treated with Wolfen thiram followed by Spergon. Similarly the heaviest grains were obtained from those treated with Wolfen thiram followed by Brassicol, Dexon and Spergon.

b- Foliage spraying:

Foliage spraying of the plants that reached flowering stage of growth was carried out by using five different fungicides. The seeds were sown on 15th of November in pots (25 cm in diameter). Ten seeds were sown in every pot and ten replicates for each treatment were used. The fungicides were applied at the rate recommended for use and artificial infection was carried out by spraying spore suspension of H. sativum. After 4 weeks of inoculation the upper two leaves of each plant were used to estimate the average damage of leaf area infected by spot blotch and Data obtained is recorded in Table (37).

Data in Table (37) show clearly that the severity of infection was reduced by spraying the plants with different fungicides specially plantavax and Bayleton. The weight of grains per pot and 100 grains weight significantly increased by spraying the fungicides after

Table (37): Effect of different fungicides as spray on the control of spot blotch of barley under greenhouse conditions.

Fungicides	Severity of infection	Number of ears/pot	Weight of grains (gm/ pot)	Weight of 100 grains (gm.)
Dithane M ₄₅	13.334	14.000	13.488	3.137
Plantavax	8.334	16.667	18.101	3.320
Kitazen	26.667	15.667	9.913	2.510
Bayleton	10.000	19.667	10.714	2.946
Dithane M ₂₂	13.334	17.000	12.476	3.246
Control	76.667	12.000	5.398	1.772
L.S.D. 0.05	6.920	N.S.	3.648	0.710

inoculation specially by plantavax, Dithrane m₂₂ and Dithane m₄₅. As regards the number of ears per pot slightly increased by fungicidal applications.

3. Field experiment:

a- Seed treatment:

The effect of the recommended dose of different fungicides as seed treatment on spot blotch disease of barley in 1980-81 and 1981-82 seasons was obtained and results are tabulated in Tables (38 and 39).

Data in Tables 37 and 38 clearly indicate that generally, all the tested fungicides reduced the severity of infection. Also both the yield and 1000 grains weight significantly increased as a result of applying all the tested fungicides. In this respect, Wolfen thiram, Dexon and Vitavax were superior to the other tested fungicides.

Results of Sakha were almost similar to those obtained at Bahtim since they showed the same trend.

Wolfen thiram, Dexon and Vitavax significantly reduced the severity of infection and increased the yield and 1000 grains weight in both seasons. While Brassicol and Spergon followed the aforementioned fungicides in their effect. However the other tested fungicide namely Bavistin had little effect.

Table (38) Effect of different fungicides as seed treatment on the control of spot blotch of barley at Sakha and Bahtim in season 1980-81.

Fungicides	Sakha			Bahtim		
	Severity of infection	Weight of grains (kg./plot).	Weight of 1000 grains (g.)	Severity of infection	Weight of grains (kg./plot).	Weight of 1000 grains (g.)
Wolfen thiram	8.334	2.787	50.370	5.000	3.863	48.248
Spergon	18.334	1.987	46.050	16.667	2.631	45.630
Brassicol	13.334	2.295	46.200	13.334	2.863	46.486
Bavistin	43.334	1.825	42.940	33.334	2.450	43.028
Vitavax	18.334	2.400	50.940	8.334	3.206	47.540
Dexon	11.666	2.450	48.900	6.667	2.887	47.780
Control	63.334	1.637	41.270	60.000	2.030	42.492
L.S.D. 0.05	6.728	0.474	0.248	10.665	0.790	1.560

Table (39): Effect of different fungicides as seed treatment on the control of spot blotch of barley at Sakha and Bahtim in season 1981-1982.

Fungicides	Sakha			Bahtim		
	Seve- rity of infec- tion	Weight of grains (kg./ plot)	Weight of 1000 grains (gm.)	Seve- rity of infec- tion	Weight of grains (kg./ plot)	Weight of 1000 grains (gm.)
Wolfen thiram	5.000	2.625	50.705	5.000	3.834	50.280
Sperguson	16.667	1.610	47.465	20.000	3.056	47.255
Brassiccol	11.667	2.350	48.644	10.000	3.500	46.663
Bavistin	40.000	1.515	44.710	36.667	2.389	44.705
Vitavax	10.000	2.565	46.522	8.334	3.445	49.588
Dexon	8.334	2.495	49.968	5.000	3.667	48.455
Control	66.667	1.360	41.284	56.667	2.056	42.083
L.S.D. 0.05	6.441	0.260	0.833	4.890	0.820	1.272

b- Foliage Spraying:

The susceptible variety Giza 117 was grown in plots 3 x 3.5 m. and four plots were prepared for each treatment as replicates and four plots without treatment as control.

Artificial infection was carried out by spraying spore suspension of Helminthosporium sativum and fungicides were sprayed after 24 hr. from inoculation and data obtained is recorded in Tables (40 and 41).

The severity of infection was reduced as the fungicides were applied. The yield for each plot and 1000 grains weight significantly increased by spraying the fungicides after inoculation.

Dithane M₄₅ (Mancozeb) and Plantavax were superior in this respect to the other tested fungicides.

Results of Sakha were almost similar to those obtained at Bahtim experiment Station and showed the same trend.

Dithane M₄₅ and Plantavax significantly reduced the severity of infection and increased the yield and 1000 grains weight and were more effective in disease control. While Dithane M₂₂ (Maneb) and Bayleton were moderately effective, but significantly increased the yield. At last, Kitazen was of a little effect in controlling spot blotch disease of barley.

Table (4C): Effect of different fungicides as spraying on the control of spot blotch of barley at Sahka and Bahtim in season 1980-1981.

Fungicides	Sakha			Bahtim		
	Seve- rity of infec- tion	Weight of grains (kg./ plot)	Weight of 1000 grains (gm.)	Seve- rity of infec- tion	Weight of grains (kg./ plot)	Weight of 1000 grains (gm.)
Dithane M ₄₅	15.000	2.330	53.167	6.667	3.500	47.111
Plantvax	8.334	2.500	52.590	5.000	3.312	48.580
Kitazin	38.334	1.750	46.080	30.000	2.737	42.641
Bayleton	33.334	2.470	50.386	13.334	2.700	45.681
Dithane M ₂₂	18.334	2.160	49.959	10.000	3.043	46.397
Control	63.334	1.750	41.937	60.000	2.062	42.038
L.S.D. 0.05	7.668	0.370	0.035	9.463	0.340	0.830

Table (41): Effect of different fungicides as spraying on the control of spot blotch of barley at Sakha and Bahtim in season 1981-1982.

Fungicides	Sakha			Bahtim		
	Seve- rity of infec- tion	Weight of grains (kg./ plot)	Weight of 1000 grains (gm.)	Seve- rity of infec- tion	Weight of grains (kg./ plot)	Weight of 1000 grains (gm.)
Dithane M ₄₅	13.334	2.485	52.320	5.000	3.500	50.290
Plantvax	6.667	2.905	51.754	5.000	4.056	50.868
Kitazin	36.667	1.080	45.683	23.334	2.500	45.163
Bayleton	23.334	2.615	49.218	10.000	3.695	50.073
Dithane M ₂₂	15.000	2.385	48.269	8.334	3.556	48.613
Control	66.667	1.030	41.100	60.000	2.100	41.503
L.S.D. 0.05	8.664	0.190	0.420	8.972	0.604	2.110