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Studies on Broomrape (orobanche spp.)

Avoidance and Control

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PROGRESS REPORT (not for publication)

SOIL FUMIGATION WITH TELONE II FOR BROOMRAPE CONTROL

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Introduction

Ethylene dibromide (EDB) was banned for agricultural use in the USA and several european countries. In Israel, the compound was removed from the market by the manufacturer two years ago, just about when the first commercial applications for crenate broomrape control were to take place at the Bet Shean valley. Against that background, the interest in dichloropropane (Telone II) for broomrape control grew and preliminary experiments started at mid-1988.

Materials and Methods

The first two experiments were conducted in closed jars.
In both experiments, the effect of various concentrations and time of exposure on the survival of three broomrape species were tested. EDB was also introduced into the first experiment for comparion in order to evaluate the prospects of Telone II.

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The seeds were pre-treated, i.e. kept wet on small (10 mm diam.) glass filter paper discs for ten days, before being exposed to the fumigant. Before pre-treatment, the seeds were surface sterilized with 2% sodium hypochloride solution for five minutes. The sterilization solution contained 0.1% surfactant Triton x-100. After exposure to the fumigant, the seeds were triggered to germinate with 1 ppm GR 24 solution.

In the first experiment, various Telone II concentration were tested as presented in Table 1, and exposure time was 48 hours, the compound was injected into the jars through a rubber stoper in the lid. The compound was diluted in xylene and thus the effect of xylene alone on the broomrape seeds was also tested. The effect of Ethylene dibromide (EDB) on the broomrape seeds was tested under similar conditions. The experiment with Telone II had four replication and the one with EDB was replicated twice.

In the second experiment fewer of the rates tested in the first experiment were tested again at different times of exposure, as presented in Table 2.

Third experiment - this experiment consisted of small field plots (1.8 m x 1.8 m). Telone II was injected into 20 cm deep holes in the ground. The holes were set in squars and were 30 cm apart. Broomrape seeds of three species were placed in small bags made of a synthetic fabric. The bags were fastened to a nylon string. Two bags were fasten on each string, 15 cm from each other. The bags on the string were placed in the ground so that the upper bag was at depth of 5 cm and the lower bag was at a depth of 20 cm. The string with the bags was placed in the center of the square formed by four holes into which the fumigant was injected. The seeds in the bags were kept wet for two weeks prior to placing them into the ground. The field was irrigated before the microplots were prepared in a field that was

cultivated with a subsoiler followed by a rototiller. After the injection of the fumigant, the field was again watered shortly with sprinklers in order to somewhat seal the upper layer to slow down diffusion of fumigant out of the soil. The bags with the seed were pulled out of the soil about one week after the fumigation and kept in wet sand until they were ready to be tested for germination.

Results and Discussion

The purpose of the first experiment was to determine whether Telone II has any phytotoxic effect on broomrape seeds and whether there is any differential response to the compound among the broomrape species as we discovered several years ago with respect to EDB.

In order to evaluate the prospects of the compound as a potential broomrape controlling agent, EDB was introduced into this study. The results of that experiment are presented in Table 1. They reveal several facts. First, Telone II is indead toxic to broomrape seeds, nearly as much as EDB. Similarly to EDB, crenate broomrape is more sensitive to the Telone II, than the branching broomrapes, Egyptian and Muteli. 0.001 cc/l of EDB killed crenate broomrape seed while Telone did not, and 0.005 cc/l killed crenate broomrape and did not kill the seed of the branching broomrape, similarly to EDB. The results of the second experiment confirm again and even more clearly that crenate broomrape is more sensitive than Egyptian and Muteli broomrape to Telone II (Table 2). All crenate broomrape seeds were killed by 0.005 cc/l after 24 hours of exposure while in that concentration, some germination was still recorded after 12 hours of exposure. Also, seeds of the branching broomrapes remain vaible for a long time at a concentration of 0.01 cc/l of Telone II. The results of those two

experiments were encouraging and lead to the third experiment in which Telone was first evaluated in the soil for broomrape control. Unfortunately, germination even in the control was relatlively low (Table 3).

The results indicate complete control of the branching broomrapes by 450 l/ha. Crenate broomrape seeds were killed by 1/3 of that rate.

Very obvious weed control was observed in the treated plots. Table 4 present the number of the different weeds that were counted in the plots. There were not only fewer weed in the treated ploots but they were also more retarded.

Table 1. Effect of Telone II and EDB on the germination of the seed of three broomrape species after exposure of 48 h. in closed jars.

	Xylene	Telone II cc/liter volume					
Broomrape	0.01 cc/l	0.000	0.001	0.005	0.01	0.05	0.1
Crenate	28±1.3[1]	22±8.3	31±4.1	0	0	0	0
Egyptian	94±1.3	89±6	90±1.4	7±0.4	0	0	0
Muteli	89±1.3	89±3	87±2.5	12±3	0	0	0
		EDB cc/liter volume					
Crenate	30±1	36±1.4	0	0	0	0	0
Egyptian	96±0.7	98±0.5	80±0.6	28±0.2	0	0	0
Muteli	93±0.8	94±0.3	66±1.6	5±0.5	0	0	0

1) Standard error

Table 2. Effect of Telone II concentration and length of exposure on % seed germination of three broomrape species.

Telone II	Hours of exposure				
cc/liter	3	6	12	24	48
Crenate broomrape					
0.000	59±5.3[1]	23±1	21±1.7	18±3	28±1.3
0.005	67±1.6	8±0.8	4±0.8	0	0
0.01	21±1.4	0	0	0	0
0.05	0	0	0	0	0
0.1	0	0	0	0	0
Egyptian broomrape					
0.000	85±1.8	87±1.4	85±0.7	86±2.6	86±5.1
0.005	84±1.3	81±1.5	69±1.6	57±0.2	7±0.4
0.01	80±2.6	64±0.6	11±4.1	0	0
0.05	0	0	0	0	0
0.1	0	0	0	0	0
Muteli broomrape					
0.000	84±1	73±1.6	75±1.3	85±1.1	89±2.9
0.005	85±1.4	71±3.3	50±2.3	61±1.9	12±3
0.01	67±4.4	41±1.5	13±2	0	0
0.05	0	0	0	0	0
0.1	0	0	0	0	0

1) Standard error

Table 3. Effect of soil fumigation with various rates of Telone II on the viability of broomrape seed of three species.

Treatments		% Germination		
Telone l/ha	depth of seeds	Crenate broomrape	Egyption broomrape	Muteli broomrape
0	5	10.2±4.8	4.3±2.6	16.8±4.6
	20	8.5±3	2.1±2.1	14.9±2.6
150	5	9	1.2±0.8	8.9±4.5
	20	0	0.3±0.3	1.4±1.4
300	5	0	1.9±1.8	6.7±3.5
	20	0	0	0
450	5	0	0	0
	20	0	0	0
600	5	0	0	0
	20	0	0	0

1) Standard error

Table 4. Effect of soil fumigation with various rates of Telone II on the number of weed plant in the plots (2.25 m^2)

Weed species	Telone II 0	15	liter/hectare 30	45	60
Diplotaxis	2	1			
Avena	3	4			
Sinapsis	14	7	3	1	1
Raphanus	1	1			1
Ridolfia segetum	8		1		1
Phalari	28		2	6	2
Rhagadiolus				1	
Calendula			1		
Total number of weed plants	55	13	7	8	7

BROOMRAPE CONTROL WITH IMIDAZOLINONE HERBICIDES

Tomato

The results of the first experiment were reported in the previous report. Two herbicides, namely, Imazaquin (Scepter) and Imazetapir (AC-499) were sprayed once with several rates ranging from 0 to 80 grams a.i/ha. No broomrape control was observed.

This experiment was repeated in the same way as the previous one.

Materials and Methods

The experiment was conducted in 2 L. pots filled with potting soil mixture ((1/3 claysoil, 1/3 sand and 1/3 tuff (grounded porous volcanic stone)). Each pot had 20 mg. of Egyptian broomrape seeds.

Tomato plants (var VF139) were planted on September 8, 1988. The pots were placed into the ground in the field and irrigated with drip irrigation. The experiment was sprayed once on September 28 with several rates (Table 1), and a spray volume of 300 L/ha with 0.1% serfectant Triton X-100. The experiment was terminated on November 23, 1988. Plant response to the herbicide was visually evaluated on a 1-5 scale (Table 1). Broomrape was categorized into groups according to size and the number of the broomrape plants in each group was determined. (Table 1). The experiment was designed to split plots with seven replications.

Results and Discussion

The experiment was conducted in a period in which prevention of yellow dwarf virus disease was close to impossible. The situation in the late summer of 1988 was particularly difficult. Most of the tomato plants were indeed infected. Therefore the second application of the herbicides that was initially planned was cancelled. The results are presented in Table 1.

Table 1: Effect of a single application of Scepter and AC499 on tomato plants and broomrape infection grown in pots.

Treatment gra.ℓ/ha	Visual evaluation: Tomato condition[2]	Visual evaluation: Broomrape condition	Number of broomrapeplants/pot: Size 4	Size 3	Size 2	Size 1
Scepter						
+0[1]	3.3 ab[4]	3.0 ab	1.0 ab	1.6 bc	3.3 bc	9.3 a
5	3.5 ab	2.7 ab	1.1 ab	2.1 bc	5.7 ab	5.1 ab
10	3.5 ab	4.2 a	2.0 a	4.4 a	8.0 a	9.7 a
20	3.3 ab	1.5 bcd	0.9 b	1.0 bc	1.9 bc	4.4 ab
40	3.2 ab	2.2 ab	0.7 bc	2.7 ab	3.3 ab	7.6 a
80	2.9 c	1.3 cd	0.3 b	1.0 bc	3.1 bc	4.4 ab
-0[1]	4.3 a					
Ac499						
+0	3.3 ab	3.0 a	1.0 ab	1.6 bc	3.3 abc	9.3 bc
5	2.4 b	2.2 ab	0.7 ab	1.4 bc	4.3 ab	6.4 cd
10	2.9 b	2.8 a	1.1 a	5.0 a	7.0 a	17.6 a
20	2.7 b	2.6 a	1.1 a	3.7 ab	6.6 a	15.4 ab
40	2.5 b	1.3 bc	0.3 bc	2.3 ab	4.9 ab	14.1 ab
80	2.2 b	0.0 c	0.0 b	0.3 c	0.9 bc	13.6 ab
-0	4.3 a					

[1] 0, -0 control with and without broomrape respectively

[2] evaluation 1-5 1 - tomato plant strongly retarded
5 - normal growth

[3] broomrape sizes: 1 - plants not emerged
2 - plants up to 2 cm height
3 - 2 cm - before flowering
4 - flowering plants

[4] values alone columns, followed by similar letter do not differ

significantly (p = .05) according to Duncan Multiple Range Test.

Some effect can be seen on the larger and older broomrape-ranked 3 and 4. Those broomrapes developed earlier at the time when the plants contained the herbicides. It can be assumed that the herbicides were gradually inactivated in the plants, and as no additional herbicide was applied, new broomrape infection could occur. As seen in the data, a lot of young broomrape plants were counted in all treatments of both herbicides (broomrape ranked 1 and 2), while a trend of reduction in the number of the older brromrapes (ranked 4) can be observed. 40 and 80 gr. AC 499 and 80 gr/ha of Scepter reduced the number of the older broomrapes. This trend of reduction of broomrape can also be seen in the visual evaluation of the broomrape plants. Unfortunately, it also seems that the herbicides have some phytotoxic affect on the tomato plants.

The results of the present experiment indicate that those two herbicides have some effect on broomrape control in tomatoes, and that research has to continue along this line, particularly considering the severity of the problem in this most important crop.

BROOMRAPE NURSERY

As indicated in the research proposal, one of the main objectives of this project is to determine the non-host range of the important broomrape species. This objective falls under the more general topic of broomrape avoidance. We have a fairly good knowledge (not more than that!) on crop families, genus or species that are not parasitized by broomrape. However, the issue becomes more complex with crop plants parasitized by crenate broomrape and not by Egyptian broomrape or vetch (Vicia sativa L) parasitized by both Egyptian and crenate broomrapes and the vetch-Vicia atropurpurea - parasitized by neither. The fact that host: parasite relationships in broomrape are also strongly affected by environmental conditions, adds another complexity to the situation. Israel is also unique in this respect that all the economically important broomrape species except one Orobanche minsor sm, are found to the extent of economical significance. That statement does ignore the possible existence of subspecies of broomrape. The practical implication of this study is to create the knowledge that will enable the professionals to advise the farmer which crop he can or should not grow in his field known to be infested with a certain broomrape species.

Description of the nursery

The nursery includes six plots, each infested with a different broomrape species, namely:

1. Orobanche aegyptiaca from a carrot field "winter Egyptian broomrape"
2. Orobanche cumuna from a sunflower field
3. Orobanche crenata from a carrot field
4. Orobranche muteli from a potato field
5. Orobanche cernua from a tomato field
6. Orobanche aegyptiaca from a tomato field "summer Egyptian broomrape"

Each plot is 4 metres wide and 36 metres long. The plots are 4 metres apart from each other.

Fences will be contracted between the plots and around the whole nursery.

A watering system was constructed so that each plot can be irrigated separately.

Inoculation was done by mixing the seeds with dry sand and "seeding" the mixture with a hand drivnen Plant Jr. breeder.

We planted the following crops: carrots, vetch, peas, chickpeas, cabbage and celery in all six plots.

Progress Report (July 1,1988 - December 31,1988)

on

the Project: Studies on Broomrape(Orobanche Spp.) Avoidance and Control

by

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CROP LOSS ASSESSMENT

There is a general practice of cultivating Baltori (Brassica campestris) along with Kalotori (B. nigra) in the Kathmandu Valley. Farmers start planting seeds of both kinds of mustards together in the same field in the month of September. Baltori, having shorter life span becomes ready for harvest by the beginning of the month of December, leaving Kalotori alone in the field. The latter is harvested in the month of February.

In the first phase (Jan/Feb. 1988) of the project work we could collect some crop loss assessment data in the Kalotori crop in last season at Badhikhel, Kathmandu Valley, as the Baltori crop had already been harvested. This season we found broomrape (O. aegyptiaca) infecting an appreciable number of Baltori plants in the fields. So we collected data (Table I) to show the extent of influence of broomrape infection on the seed yield and general growth of individual Baltori plants.

Table I

Influence on Baltori Plants (B. campestris) due to Broomrape (O. aegyptiaca) at Badhikhel, Kathmandu.

Parameters	Uninfected Plants	Infected plants	Loss (%)
Height (cm)	48.5 ± 8.65	32.25 ± 6.82	33.51
No. of branches	4.1 ± 1.79	1.09 ± 0.73	53.51
No. of Siliqua	35.6 ± 17.27	3.04 ± 1.77	90.45
Length of siliqua	4.6 ± 0.53	2.97 ± 0.80	64.04
No. of seeds/ siliqua	15.68 ± 2.14	7.78 ± 4.38	50.35
No. seeds/plant	539.82 ± 238.91	30.02 ± 34.22	94.04
Wt. of 100 seeds (gm)	0.519 ± 0.008	0.286 ± 0.011	44.09

The data show that there is a tremendous loss in the seed yield which appears to be due to the decreased number and size of siliqua. As far as the effect on the individual plant is concerned, the broomrape influence is found to be more pronounced in Baltori compared to the Kalotori. In contrast to the individual broomrape influence, the population of Kalotori had more mass infestation than the Baltori. The reason for the less mass infestation on the Baltori might be due to the fact that the soil conditions and temperature were not perfectly ideal to the broomrape infestation in the Baltori.

Studies on seed Germination

Following studies on the seed germination of Orobanche spp. were done during the second phase (June-Dec 1988):

1. The effect of preconditioning with water for varying lengths of time on the seed germination of O. aegyptiaca.

2. The effect of GA3 on the seed germination of O. aegyptiaca and O. solmsii.

3. The effect of root extract of mustards (B. campestris and (B. nigra) on the seed germination of O. aegyptiaca and O. solmsii.

4. The effect of stimulants- GR24, GA3, root extract of mustard separately and in combination with GA3 on the seed germination of O. aegyptiaca and O. solmsii.

Materials and Methods:

1. *Collection and storage of broomrape seeds*: Seeds of O. aegyptiaca were collected from mustard (Brassica campestris var.toria) fields at Nawalpur, while seeds of O. solmsii were collected from tobacco (Nicotiana tabaccum) field at Belachapi, in Feb. 1988.

2. *Surface sterilization*: Seeds were surface sterilized by immersion in a dil. solution of sodium hypochloride (1%) for 5 min. and washed thoroughly with distilled water.

3. *Preconditioning of Seeds*: Seeds as such or surface-sterilized were soaked in a small volume of distilled water for desired periods. Seeds were put either on the Whatman filter paper disks or glass fiber filter paper (GFFP) disks, which in turn were placed in the petri-dishes containing double layer of moistened filter papers; and incubated at 20-25°C in the dark for a desired period.

4. *Pretreatment of seeds with GA3* : In our earlier experiments, preconditioned seeds were pretreated by soaking in 50 ppm GA3 solution for a desired period. In later experiments, the preconditioned seeds were transferred to the fresh GFFP disks soaked in 100 ppm GA3 solution and incubated for a desired period at 20-25°C in the dark.

5. *Preparation of root extract*: Roots of mustards (B. campestris and B. nigra) were collected and stored in the refrigerator at -20°C. Roots were macerated with ethylacetate (1:2 w/v). The supernatant was collected and stored in the refrigerator at -20°C.

6. *Preparation of GR24 solution*: A known weight of GR24 was dissolved in a small volume of Dimethylsulphoxide and then diluted to desired concentrations adding distilled water.

7. *Use of germination stimulants*:

(a) Use of GR24: Seeds previously preconditioned with distilled water were transferred to fresh GFFP disks soaked with a definite volume of desired concentrations of GR24 solutions and incubated in dark at 20-25°C for a week. Then germination was observed under microscope.

(b) Use of root extract: Definite volume of ethylacetate root extract was applied in 50u aliquots on the filter paper disks allowed to evaporate. Seeds previously pretreated with GA3 were put on the disks and incubated in the dark at 20-25°C for a week and observations were taken with the help of microscope.

Results and discussions:

Our studies on Orobanche seed germination indicated that plant growth hormones (GA3, NAA, BAP), root exudate and aqueous extract of roots of mustard (B. campestris) and tomato (Lycopersicon esculentum) separately, could not induce germination of seeds of O. aegyptiaca. When the seeds were pretreated with GA3, germination took place in the presence of ethylacetate extract of roots of mustard and tomato.

The experiments, performed in the hope to find optimum period for preconditioning of seeds in the distilled water showed that the seed preconditioned for a period of six days and then, pretreated With 100 ppm GA3 gave the highest percentage of germination in the presence of root extract (Fig. 1).
The experiment on seed germination of O. aegyptiaca with root extract of mustard showed that there is an optimum concentration of root extract which, if exceeded, cause inhibitory effect on the seed germination. This suggests that the root extract of mustard has stimulatory as well as inhibitory effects an seed germination (Fig.2)

In a trial experiment conducted to find a comparative effects of GR24, GA3, root extract separately, and GA3 (100 ppm) in combination with the root extract, we found interesting results. Over all, the surprising part of the result was that O. solmsii seeds showed the highest percentage (70%) of germination in the preconditioning medium (dist. water) compared to all stimulants tested. In the experiment, GR24 gave statistically more or less similar percentage of germination in the concentrations: 10 ppm, 25ppm, 50ppm, indicating 10ppm GR24 to be an optimum concentration (Table II). Similar results were obtained with GA3 in the concentrations: 10ppm, 20ppm, 50ppm and 100ppm, indicating 10ppm GA3 to be an optimum concentration for the seed germination of O. solmsii (Table II).

The response of seeds of O. solmsii to the root extract of mustard (B. campestris) is found to be the lowest compared to other stimulants used and the percentage of germination decreased with the increasing concentration of the root extract, apparently showing an inhibitory effect (Table II). This is in agreement with the fact that mustard is not a natural host of O. solmsii in Nepal so far we know.

TABLE II

Effect of various treatments on germination of O. solmsii.

Treatment	Percentage germination
Control (Water)	70.0
GR24	
1 ppm	2.4
5 ppm	37.4
10 ppm	57.4
25 ppm	63.1
50 ppm	61.3
GA3	
1 ppm	18.1
10 ppm	27.1
20 ppm	25.0
50 ppm	29.0
100 ppm	24.0
B. Nigra Root extract	
100 ul	6.0 (24.1)
200 ul	3.0 (17.6)
300 ul	0 (15.2)
400 ul	0.8 (14.1)
500 ul	1.5 (21.0)

Seeds were pretreated for 10 days in water as described in Materials and Methods. Values in parantheses were obtained for seeds that were pretreated with 100 ppm GA3 in place of water.

The picture was different with the seeds of O. solmsii pretreated with GA3 (100ppm) and then post-treated with the root extract as it showed a bit higher percentage of germination compared to the seeds treated with the root extract only and it was a bit lower for the seeds pretreated with GA3 only. (Table II). Here, we might assume that the root extract of mustard has an inhibitory effect on the seed germination of O. solmsii in all levels as this broomrape does not include the mustard in its host range.

That the seeds of O. solmsii can germinate in the laboratory condition in an appreciable percentage without the host (tobacco) root extract, reveals the possibility of its control in the tobacco fields by using the field (which had the infestation of the broomrape) for cultivation of other crops except mustard for a few years before reusing the field for tobacco cultivation.

We have observed that incubation of O. aegyptiaca seeds at 28°C severely prevented germination even in the presence of stimulants. In the next phase we will be conducting experiments to study effects of temperature on Orobanche seed germination.

Field Experiments

We hesitated in conducting field experiments in full scale this year due to the lack of sufficient Orobanche seeds. Still, we are conducting experiments along this line at Badhikhel (KTM) where infection of Orobanche was found to be severe in the previous season (1987) in mustard crop. Therefore, we are depending on the seeds naturally present in the field in order to carry out these experiments. Following are the experiments being conducted at the moment.

(1) **Effect of planting date on the Parasitism of mustard (B. nigra) by O. aegyptiaca.**

For the experiment we took an area of 150 sq.m. (15m x 10m). The area was divided in three blocks and each block in five plots. The planting dates were 24 Sept., 9 Oct., 24 Oct., 8 Nov., 23 Nov. and 8 Dec.. Details of the experiment and its result will be given in the next report.

(2) **Study of host (B. nigra) - parasite (O. aegyptiaca) Phenology**

Area and design of the field taken was same as in the former experiment. Mustard (B. nigra) plants were planted in each plot.

We have been recording observations after two months of planting seeds. Details of the experiment will be given after completion of the observation.

(3) **Broomrape control with foliarly applied glyphosate on black mustard (B. nigra)**

In the trial experiment, we took an area of 131 sq. meter, dividing it into five blocks and each block in five plots. Each plot has about 30 - 40 mustard plants. We have already given first treatment of glyphosate in the field. Details of the experiments will be given after the completion of the treatment and final observation.

Study of Pollen Morphology

Morphological heterogeneity was found to be pronounced in the populations of O. aegyptiaca. Moreover, the Orobanche species on different hosts, particularly tobacco and leafy Brassicas were found to be morphologically different (in terms of their height and branching pattern, color etc.) from those infecting mustards (B. campestris and B. nigra). We have collected other two species of Orobanche and their taxonomic status is yet be confirmed. Hoping that pollen morphology gives an additional taxonomic evidence to ascertain the delimitation of taxa and to draw the phylogeny of this taxa, Orobanche, We undertook this study to investigate the different spp. of the genus occurring in Nepal. We are carrying out this part of investigation in collaboration with Dr. R. P. Chaudhary, Central Dept. of Botany, Tribhuvan University, KTM, Nepal.

Materials and Methods

The samples of O. aegyptiaca, collected from fields of mustard (B. campestris), black mustard (B. nigra) and tobacco (N. tabaccum), and one sample of O. solmsii, collected from tobacco field, were taken. Polliniferous material was taken out of the flowers and soaked in 70% alcohol. After 24 hrs. in the alcohol, the material was crushed and transferred to glass tubes. The tubes were centrifuged and alcohol was decanted off. The sediment was again centrifuged in glacial acetic acid which was again decanted off. Then about 5 ml. of freshly prepared acetolysis mixture (acetic acid anhydride + Conc. sulphuric acid, 9:1 v/v) was put in each tube and heated in hot (70°C) water for 3-5 minutes. After that the tubes were centrifuged and the 'mixture' was decanted. The sediment was washed with 90% alcohol, centrifuged and decanted. The sediment material was mounted in glycerin jelly for microscopic observation. (Details of observation will be given in the next report).

Study of development of parasite (broomrape) on the host (B. campestris) by sandwich method

A trial experiment was conducted to study the parasite for its germination, attachment and development on the roots of the host plant.

Materials and Method

Glass plates of two different sizes (20 cm.x11 cm. and 13 cm.x11 cm.), foam pieces (13 cm.x11 cm.), synthetic cloth (11 cm.x11 cm.) and plastic containers were the basic materials. Mustard plants were used as a host. Seeds of O. aegyptiaca and O. solmsii collected from mustard (B. campestris) and tobacco (N. tabaccum) fields respectively were taken for the experiment order to inspect the development of the parasite. Seeds of mustard were sterilized with sodium hypochlorite (1%) for 5 min. and washed thoroughly. Then the seeds were allowed to germinate on the germination paper. When the seedlings had gained

sufficient growth, they were sandwiched between glass plates along with synthetic cloth which was layered on the foam. The sandwich plates containing the seedlings were put in the plastic containers containing dist. water. Containers were put under light at the room temperature (fluctuating between 15 to 20°C). After a week, the dist.water was replaced by Hoagland solution. After a week Seeds of Orobanche were put on the mustard root. Observations were taken after two weeks.

Observation

The Brassica plants showed a poor growth; a majority of them were not more than 5 cm. in height with 2-3 leaves only. Some of the plants died shortly after the transfer. Most of them started bearing flower buds in 3-4 leaf stage, but the flower buds did not open. Orobanche seed germination showed a good response even in such a poor state of host plants. There was a fairly good percentage of seed germination in O. aegyptiaca. The parasite also exhibited it's early developmental stages like attachment with the host root and development into precauline stage. But we are not sure if we could be able to see their further development due to the stunted growth of host plants. Furthermore, the experiment also indicated that seeds of O. aegyptiaca, collected from tobacco field, are able to infect mustard. Another important part of the experimental result was that seeds of O. solmsii (infecting tobacco) did not germinate even though they are able to germinate in dist. water. This is supporting our earlier finding that mustard cause inhibitory influence on the seed germination of O. solmsii.

Dr.Jha's visit to Israel

Dr. P. K. Jha has visited the Volcani Center, Israel for a period of ten weeks (16.7.1988 to 28.9.1988) for training on various aspects of broomrape research. During his stay at Volcani Center with Dr. R. Jacobsohn, he learnt the following techniques:

- Broomrape seed germination.
- Inoculation of host plants with broomrape in a soilless system.
- Quantitative and qualitative determination of broomrape seeds in the soil.
- Broomrape experiments in the field and problem involved.
- Quantitative application of herbicides in the field and problems involved.
- Quantitative application of herbicides in the field. Proper equipments and necessory computation.
- Quantitative determination of broomrape seeds in crop seeds.

Dr. Jha also visited several research stations in Israel including Weed Research Station at Neweyaar. He observed the polybag system of Orobanche germination and morphological observations of seeds under fluorescence microscope. Dr. Jha met the science Attache in U.S. Embassy of Tel Aviv, and discussed the program and problems regarding the on going project.

Morphological study

Sample no....Date:.....
Host.....
Loc.....

Scale leaves
Length.....breadth.....surface.....
Margin.....colour.....arrangement.....
Remark.....

Inflorescence
Length.....density.....remark.....

Flowers
Bracteoles.....bract: length.....
Divided half posteriourly only or divided at both sides: entire/bifid
Margin at the tip.....arrangement (viewing from top):spiral/straigh

Corolla
Length.....colour.....lobes:entire/emarginate;constriction.....
Remarks.....

Stamens
Length of filament.....;smooth/hairy/sparsely hairy;insertion above the base of corolla;anthers:hairy/sparsely hairy/smooth;maturing time: before or after anthesis.
Remark.....

Gynoecium
Ovary: length.....breadth.....surface .....shape.....
Style: length.....surface.....
Stigma:lobes-partially united/completely united,surface-hairy/smooth, colour.....,curvature-before .....after .....
Remarks.....

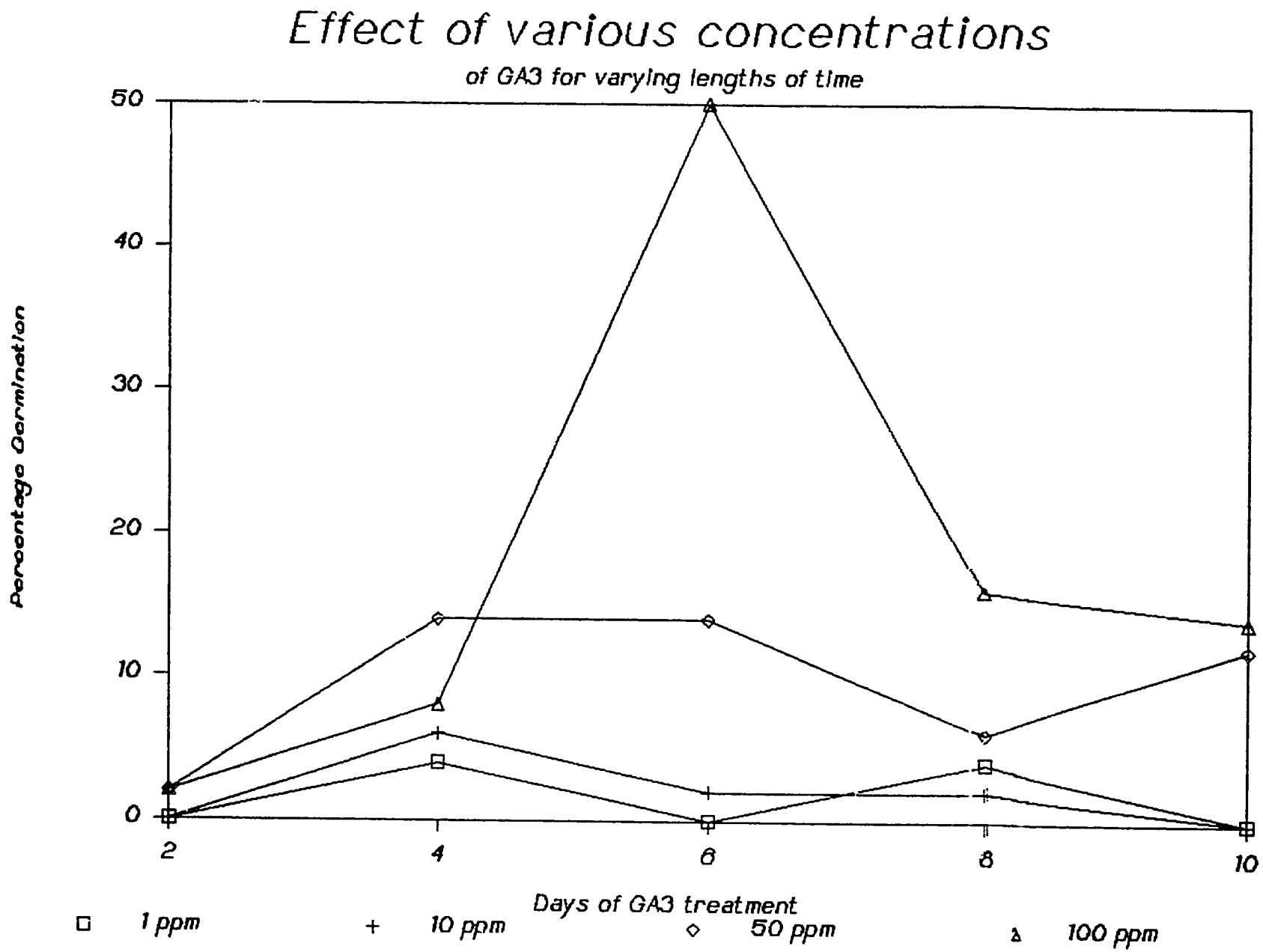
Effect of various concentrations
of GA3 for varying lengths of time
Percentage Germination
50
40
30
20
10
0
2
4
6
8
10
Days of GA3 treatment
1 ppm
10 ppm
50 ppm
100 ppm

FIG. 1

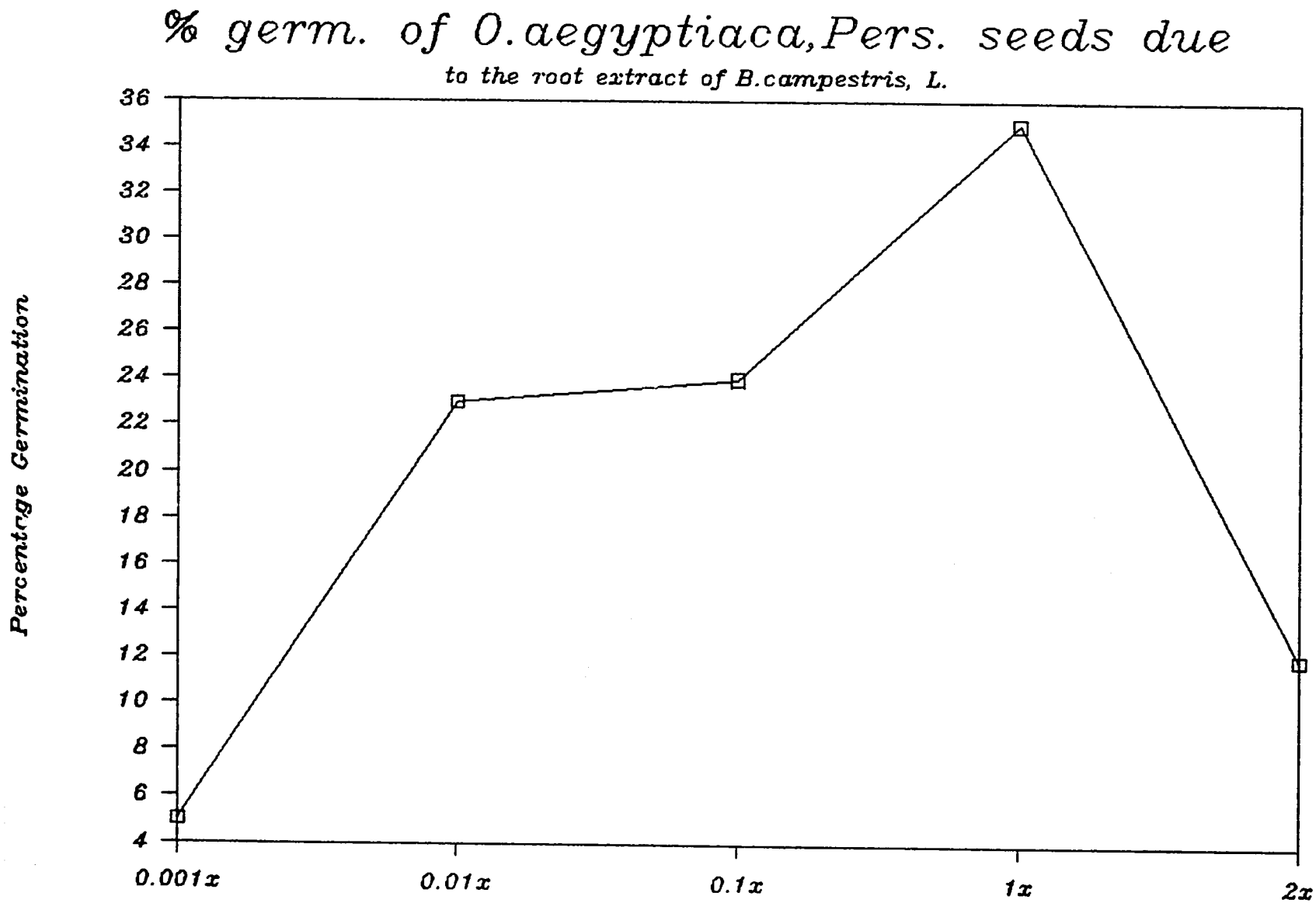
% germ. of O.aegyptiaca,Pers. seeds due
to the root extract of B.campestris, L.
Percentrge Germination
36
34
32
30
28
26
24
22
20
18
16
14
12
10
8
6
4
0.001x
0.01x
0.1x
1x
2x

Fig. 2