

COMPARATIVE DEVELOPMENT AND HOST SELECTION MECHANISM
OF THE PEA LEAF-MINER, PHYTOMYZA HORTICOLA GOUREAU ON
RAPESEED AND MUSTARD

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ABSTRACT

The development of the pea leaf-miner, Phytomyza horticola Goureau was investigated on three Brassica host plants in the screen-house. BSH 1 (B. campestris) served as the most suitable host for feeding, oviposition and development of the insect. B. tournefortii was least preferred. RLM 198 (B. juncea) was intermediate in its response to the leaf-miner. The antibiosis in B. tournefortii is attributed to higher levels of glucosinolates, phenolic compounds, calcium, magnesium and sulphur contents. Influence of these constituents of the plant on biology of the pest is discussed.

Experiments on the feeding and oviposition behaviour of the flies revealed that irrespective of the host plant and position of the leaf (lower, middle or upper) on a plant, there was a marked preference for (i) feeding in the central portion of the leaf and (ii) oviposition in the area along the leaf margins. Carotene pigment, hairiness and allylisothiocyanates in plant leaves have been detected as important cues in mediating the host seeking behaviour of the flies. Role of allylisothiocyanates (a plant kairomone) as oviposition excitant has been established.

INTRODUCTION

The pea leaf-miner, Phytomyza horticola Goureau is a polyphagous pest and feeds on 127 plant species belonging to 27 families (Singh & Mavi, 1982). Its damage to Brassica crops is often very high. Brassica species exhibit differential response to its attack. A strain of B. tournefortii proved as highly resistant and those of B. juncea as highly susceptible (Bakhettia & Sandhu, 1977). The field biology and behaviour of this leaf-miner in relation to Brassica plants was not understood very well and hence the present project was taken up.

MATERIAL AND METHODS

Host plant preference of the leaf-miner was studied in two experiments. In first experiment, BSH 1 (Brassica campestris Linn.), RLM 198 (B. juncea Coss.) and a strain of B. tournefortii Gouan were grown in 15.0 x 8.5 m plots. Larvae and pupae of the leaf-miner were counted from 25 plants per plot taking 6 leaves per plant (two each from top, middle and lower portion). Population was finally computed on unit area basis. In another experiment, nine cultivars namely, B. juncea strains RC 512, RC 978 and Blaze; B. campestris strains BSH 1 and Novini; B. napus strains Midas and Regent, and a strain of each of B. nigra and B. tournefortii, were grown in the field as well as in trays (50 x 36 cm) in screen-house in a latin square design in two replications. In the screen-house, mated females (one fly per plant) were released in the caged trays containing 45 plants of each host. The number of punctures made by the flies after 4 days were counted. However, in the field, larvae and pupae were counted on all the 45 plants.

The biology of the pest was studied for 4 generations on BSH 1, RLM 198 and B. tournefortii in the screen-house. Preference for oviposition site was studied on B. juncea strains RC 978 and RLM 198 in the field and screen-house as well.

The biochemical analyses of the leaves was made for chlorophyll (Arnon, 1949); phenols (Swain & Hillis, 1959); nitrogen (McKenzie & Wallace, 1954); crude fat and volatile isothiocyanates (AOAC, 1970); phosphorus and potassium (Jackson, 1967); glucosinolates (McGhee et al., 1965); sulphur (Chesnin & Yein, 1950), calcium and magnesium (Black, 1965).

To detect the visual stimuli for host selection, the flies were exposed to 15 cm² sized filter papers impregnated in one per cent glucose solution and wrapped with yellow, green and red butter papers, placed in small cages.

Fifteen leaf discs (0.50 cm²) of B. tournefortii with variable hair density (0-12 hair) were kept in a petridish covered by split cages. One mated female fly per disc was released and punctures made in each disc were counted after 24 hours. Response of the flies to non-hairy leaf discs of B. tournefortii was also compared with those of RLM 198 and BSH 1.

The chemical stimuli were detected by spraying B. juncea seed extract and standard allylisothiocyanate (Merck. Schuchardt brand) at 0.1-1.0 per cent concentrations on the potted plants of Primor (B. campestris) cultivar containing zero level glucosinolates. Single mated female was confined on each plant and number of punctures per plant were recorded after four days.

RESULTS AND DISCUSSION

Host plant preference : In the field, BSH 1 harboured the maximum population of larvae and pupae whereas B. tournefortii remained almost free from leaf-miner damage (Table 1). Pest

population on RLM 198 was also quite high but significantly lower than that on BSH 1. Another free-choice test (Table-2) revealed that BSH 1, RC 512 and RC 978 were highly preferred by flies for making punctures for feeding and egg laying which indicated the presence of some visual/chemical stimuli in the plants of these cultivars. However, the population of larvae and pupae per plant was very high in case of BSH 1 and very low or rather zero in other hosts. It suggested that BSH 1 was most preferred host for the leaf-miner.

Comparative development of leaf-miner : The data on different biological parameters of the leaf-miner are given in Table 1. The development of the leaf-miner was fastest on BSH 1, slowest on B. tournefortii and intermediate on RLM 198. Survival and growth index of larvae, fecundity, fecundity-cum-viability and longevity of male and female flies were highest on BSH 1 followed by that on RLM 198 and B. tournefortii. The weight and size of larva, pupa and flies cultured on BSH 1 was significantly more than those reared on B. tournefortii and RLM 198.

BSH 1 contained highest amount of protein (34.75%), crude fat (7.70%) and P (0.66%), and lowest amount of glucosinolates (1.86%), phenols (0.79%), K (2.35%), S (300 ppm), Ca and Mg (8.0 and 4.2 mg/100 g respectively). On the other hand B. tournefortii possessed highest amount of glucosinolates (2.85%), phenols (1.0%), K (3.14%), S (425 ppm), Ca and Mg (26.0 and 11.4 mg/100 g respectively), and lowest amount of protein (22.88%), crude fat (6.29%) and P (0.48%).

Phenolic constituents in B. tournefortii and B. napus (1.0 and 1.4%) were higher than that in BSH 1 and RLM 198 (0.75%). The population of larvae and pupae was also lower on the former group of hosts as compared to that on the latter group (Table 2). It was observed that the larvae did only localize feeding in B. napus and left the mine before pupation. Higher levels of Ca and Mg in B. tournefortii provided rigidity to cell wall which proved unfavourable for the growth and development of the leaf-miner larvae.

On the basis of the above mentioned facts, it can be inferred that BSH 1 was nutritionally superior and proved most suitable host for P. horticola whereas B. tournefortii was nutritionally inferior and least suited. It corroborates the earlier report of Singh *et al.* (1981). The status of other host plants tested needs further investigation.

Feeding and oviposition punctures : Two types of punctures in the leaves were identified such as feeding and oviposition punctures. The feeding punctures (called pseudo-punctures by Tandon, 1972) were creamish-white specks and were crowded mainly in the central area of the leaf. The egg containing punctures were yellowish-brown protuberances and were made sparsely in a linear fashion along the leaf margins. Irrespective of (i). The host plant and (ii) the position of the leaf (upper/middle/lower) on a plant, more punctures (58.06-72.41%) were made in the marginal area of the leaf (Table 3).

Detection of plant stimuli :

Leaf colour : In bioassay studies, the flies showed a higher preference for yellow over green and red colour. It was also noticed that highly preferred varieties namely BSH 1, RC 512, RC 978 and RLM 198 possessed higher level of carotene that is 0.41-0.56 mg/g (Table 4). This pigment is known to exhibit a yellow colour and thus provided a positive visual stimuli for host selection. However, chlorophyll did not seem to act as an important stimuli.

Hairiness : A significant and negative correlation between punctures and hair density ($r = -0.78^{**}$) revealed that hairiness acted as deterrent to the flies for feeding and oviposition. Non-hairy leaf discs of BSH 1, RLM 198 and B. tournefortii, when exposed to mated female flies in cages, bore 4.4 ± 2.03 , 4.2 ± 2.70 and 0.6 ± 0.80 punctures, respectively. It suggested that besides hairiness, some chemical stimuli in BSH 1 and RLM 198 was responsible for their high preference by the flies.

Chemical stimuli : The volatile isothiocyanates ranged from 0.10 to 0.35 per cent in B. juncea strains (RC 512, RC 978 and RLM 198) and B. campestris (BSH 1) whereas it was negligible or absent in B. tournefortii and B. napus. Their concentration was more in margins of the leaves than that in the central portion (Table 3). It was due to this fact that the leaf margins received much more puncturing by the flies (Table 2).

Spray of allylisothiocyanate (ATC) on Primor plants stimulated the flies for puncturing activity. The number of punctures were 46 with water, 266 with B. juncea seed extract (containing 0.38% ATC) and 45,167,219,375,92 and 37 punctures with 0.1,0.2,0.3,0.5,0.7, and 1.0 per cent ATC spray respectively. It therefore suggested that ATC at 0.2 to 0.5 per cent acted as an excitement to the flies for feeding and oviposition. Optimum concentration of ATC was 0.5% and the activity got suppressed at higher concentrations. These findings are in conformity with those of Finch (1978), that ATC acted as plant kairomone for many insects.

Tandon (1972) reported that more oviposition punctures were made in the leaf-margins because of fewer sub-marginal veins. However, in present studies, the leaves of RLM 198 were highly reticulated in the marginal area, even then these received very high oviposition.

Thus it can be deduced that carotene pigment, hairiness and allylisothiocyanates (0.20 to 0.50%) served as important cues for the leaf-miner flies in the process of host selection. The antibiosis factor may be attributed to higher amounts of glucosinolates, phenols, K, S, Ca and Mg contents.

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Table 1 : Field population and different biological parameters of *P.horticola* on some *Brassica* plants.

Biological Parameters	BSH 1	RLM 198	B.t.
Population in field (15.2.80)			
(Larvae + Pupae/100 cm ²)	312.40	50.0	0.16
Total Developmental period (days)	25.20	31.23	40.37
Per cent survival of larvae	85.83	60.70	21.60
Growth index	11.57	5.92	2.10
Fecundity (Eggs/Female)	307.36	256.89	1.54
Fecundity-cum-viability	254.21	148.86	3.14
Per cent egg viability	80.53	59.16	51.77
Longevity of male flies (days)	9.61	4.96	1.81
Longevity of female flies (days)	19.28	13.33	3.24
Weight of larva (mg)	0.90	0.71	0.39
Length of larva (mm)	3.37	2.47	1.98
Breadth of larva (mm)	0.86	0.61	0.55
Weight of pupa (mg)	0.73	0.57	0.43
Length of pupa (mm)	2.25	1.76	1.70
Breadth of pupa (mm)	1.09	0.89	0.80
Weight of male fly (mg)	0.23	0.19	0.15
Weight of female fly (mg)	0.40	0.33	0.20

Note : 1. Each value is the mean over four generations

2. Weight and size measurements based on 75 & 25 individuals respectively.

B.t. = *Brassica tournefortii*.

Table 2 : Relative population of and host preference by *P. horticola* flies.

Host	In Field		In Screen House	
	Mean L + P		Mean punctures/plant	
	A : 10.2.82	B : 11.2.82	A : 15.2.82	B : 2.3.82
<u>B. juncea</u>				
RC 512	1.41	3.48	28.00	21.22
RC 978	1.73	1.56	15.77	18.88
Blaze	0	0	0.11	0
<u>B. campestris</u>				
BSH 1	17.67	22.18	12.44	13.00
Novini	0	0	0	0
<u>B. napus</u>				
Regent	0	0	0.22	0
Midas	0.02	0	0	0
<u>B. tournefortii</u>	1.53	0.07	0.88	0.77
<u>B. nigra</u>	0.45	0.22	0.22	0
S. EX	0.58	1.63	2.90	2.80
LSD (p = 0.05)	1.63	4.52	8.05	7.76

L = Larva, P = Pupa - A and B indicate two different experiments.

Table 3 : Site of puncturing for feeding and oviposition by *P. horticola* flies on two strains of *B. juncea*.

Leaves from given plant portion	RC 978			RLM 198		
	TP	% punctures in		TP	% punctures in	
		M	C		M	C
Upper	36	63.88	36.11	31	58.06	41.93
Middle	45	64.44	35.55	33	69.69	30.31
Lower	29	72.41	27.58	26	69.23	30.76

TP = Total punctures, M = Marginal area, C = Central area

Table 4 : Leaf pigments and volatile isothiocyanate (ITC) contents in the leaves of some *Brassica* species.

Cultivar	Chlorophyll (mg/g)			Carotene	% ITC	
	a	b	Total	(mg/g)	17.1.82	4.3.82
BSH 1	0.64	0.47	1.30	0.42	0.13	0.22
RC 512	0.80	0.63	1.59	0.41	0.10	0.20
RC 978						
Total leaf	0.76	0.58	1.66	0.41	0.13	0.30
Marginal area	—	—	—	—	0.27	0.35
Central area	—	—	—	—	0.01	0.29
RLM 198						
Total leaf	0.89	0.55	1.83	0.56	0.10	0.26
Marginal area	—	—	—	—	—	0.32
Central area	—	—	—	—	—	0.28
Blaze	0.60	0.40	1.15	0.34	—	—
<i>B. tournefortii</i>	0.98	0.95	2.17	0.36	0.00	0.04
Regent	0.69	0.51	1.44	0.33	0.01	0.06

Total leaf taken where not mentioned otherwise.