HOST DISCRIMINATION IN *RHAGOLETIS BERBERIS* (DIPTERA:TEPHRITIDAE)

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ABSTRACT

Following oviposition, females of *Rhagoletis berberis* Curran (Tephritidae), appear to deposit host marking pheromones on the surface of their host fruit, *Mahonia* (Berberacidae), and discriminate against such marked hosts when choosing oviposition sites. Marking is accomplished by dragging the ovipositor on the fruit surface, resulting in the deposition of a fluid trail. In addition to these findings, females were observed feeding on the juice of host fruit through punctures made with their ovipositors. Therefore, the incidence of fly feeding was compared with successful and unsuccessful oviposition.

INTRODUCTION

Host discrimination is defined as the ability to detect conspecifics (Salt, 1934) and is demonstrated in several entomophagous and phytophagous parasitic insects (Prokopy, 1982). In some members of the tephritid fruit fly genus, *Rhagoletis*, for example, host discrimination is mediated by the deposition of host marking pheromones (HMPs) which are laid down in a fluid trail over the fruit surface following egg-laying. Females foraging for suitable oviposition sites detect the presence of HMPs through contact with receptors on their foretarsi and generally reject such marked hosts (Prokopy, 1981). The present study examines the host discrimination behaviour of females of the tephritid species, *Rhagoletis berberis* Curran, as part of a long term study on the population dynamics of *R. berberis* and its host *Mahonia* (Berberacidae) in British Columbia.

Rhagoletis berberis is found in the Okanagan Valley, on Vancouver Island and in Lower Mainland regions of B.C. The species is easily distinguished from other members of the genus by its entirely black body, distinctive karyotype and wing pattern. Its narrow host range includes several species of northwestern *Mahonia*, notably *M. aquifolium* and *M. nervosa*, commonly known as mountain grape and Oregon grape respectively (Bush, 1961). Adult flies emerge in early summer and can be found at host sites for several weeks. During this period, mated females lay eggs in nearly ripe fruit. The pupating larvae drop from rotting fruit and overwinter in the soil beneath the host. In the following summer, adults emerge from the soil, initiating a new cycle of insect-host interaction. Our rationale for studying host discrimination in *R. berberis* is based upon the following:

First, *R. berberis* larvae are unable to move between host fruit. Therefore their success as larvae is dependent upon the choice of host fruit by their mothers. As food and space within hosts is limited, competition among larvae within the fruit may be important to larval survival. Thus, females that mark hosts and avoid laying eggs in already-occupied fruit may enhance their reproductive fitness.

Second, HMPs are known to operate in at least ten other species of the *Rhagoletis* genus (Prokopy, 1981).

Third, certain species of fruit infesting tephritids are among the world's most damaging agricultural pests (Prokopy & Roitberg, 1984). Although not an economic pest, *R. berberis* is closely related to the cherry fruit fly, *R. cerasi*, a current pest in B.C. and the apple maggot fly *R. pomonella*, a pest present in Washington State and feared to be spreading to B.C. Thus, knowledge gleaned from this system may be utilized in management of those related deleterious pests.

Finally, elucidation of the oviposition behaviour of *R. berberis* should promote our understanding of the population dynamics of this fly-fruit system. In addition to host discrimination behaviour, we report observations of related behaviour.

METHODS

The present research consisted of field observations and laboratory experiments for which we utilized two groups of *R. berberis* females: wild flies, reared and observed in nature and flies of wild origin, reared and observed in the lab.

Field Observations

Three field sites, located in two suburbs of the B.C. Lower Mainland, were chosen based on host and fly presence. At each site, we followed wild females individually as they moved among fruit clusters, documenting their search, oviposition and fruit surface-dragging behaviour with a tape recorder and stop watch. Visited fruit were dissected in the lab. From the dissections we tabluated the number of successful ovipositions (egg(s) found), unsuccessful ovipositions (no egg found) and the number of eggs found per fruit. In addition, we noted the number of fruits dragged upon following oviposition.

Lastly, we picked a random sample of fruit in the field. Individual infested fruit from this sample and their emergent flies were assigned paired numbers. In each pair, the weight, head capsule width and pronotal width of the fly were compared with the diameter of the fruit.

Lab Experiments

Flies of wild origin were reared in the lab. We obtained larvae, from rotting fruit picked the previous summer, in the following manner: gathered ripe fruit clusters were brought into the lab and spread out on wire mesh screens set over trays of moist vermiculite and fine sand. Pupating larvae dropped from the rotting fruit into the vermiculite mixture. Collected larvae, stored at 3°C overwintered until required for the summer's experimentation. Following a warming period, emergence and a maturing period (*ca.* 8 days), mated females were separated from males and placed collectively in a 25 x 25 cm plexiglass-mesh cage. The flies were fed on a diet of water, sucrose and yeast hydrolysate (Prokopy, 1971) and were maintained under fluorescent light, 16L:8D. We conducted lab experiments 6 hours after lights-on to approximate the time females would forage for oviposition sites in nature. Lab-reared flies were used for the experiments because wild flies collected at our field sites did not acclimatize to lab conditions.

Females were pre-tested prior to the experiments to ensure their readiness and motivation for egg-laying. To qualify for the experiments each fly was required to lay a single egg in each of two uninfested fruits. We transferred pre-tested females to individual plastic, numbered Dixie®-cup cages. Each qualified female was offered, randomly, three types of *M. aquifolium* fruit atached singly to the end of a coded probe and placed inside her cage. The three types offered were: 1. Uninfested fruit (– –), 2. Egg-infested fruit with surface dragging (+ +) and 3. Egg infested fruit without surface dragging (+ –) which we obtained by removing females from the fruit surface immediately following oviposition. This was a necessary step because our field observations indicated that females will generally drag the fruit surface after oviposition (see Results).

During the experiments, females that rejected a random fruit, i.e., left without attempting oviposition, were offered an uninfested fruit to ensure that rejection occurred due to fruit quality and not the motivational state of the fly. If the uninfested fruit was rejected as well, the previous data for the fly were eliminated. Females rested 5 minutes between each experiment. We dissected the offered fruit after each experiment.

We recorded the females' search times on all three fruit types. We observed that occasionally, females would feed on the juice of the offered fruit through punctures made with their ovipositors. The incidence of fly feeding was therefore compared with successful and unsuccessful ovipositons.

RESULTS

Field Observations

Females were active in the field from 1100 to 1400 hours and made short flights to nearby fruit clusters or longer flights to distant bushes. Males, by contrast, were present from 900 to 1700 hours. They stationed themselves on fruit within single clusters and apparently waited for females. Sightings of both sexes were considerably fewer on overcast or rainy days as compared to days of full sunlight. Females did not attempt to oviposit on every fruit they encountered. Females that did attempt oviposition followed one of two sequences, both of which began with a search of the fruit surface. After searching, flies either left the fruit or initiated oviposition. Following oviposition, they either dragged their ovipositor over the fruit surface or left. We documented 25 ovipositions, the mean duration of which was 123.9 s (S.E. = 8.17 s). The mean duration of ovipositor dragging was 21.6 s (S.E. = 3.7 , N = 17). On occasion, we observed a fine, thread-like, fluid trail on the fruit surface after dragging occurred.

Results of the fruit dissections (Table 1) show ovipositor dragging following in 80.6% of successful ovipositions (egg found). Conversely, ovipositor dragging following in 40% of unsuccessful ovipositions (no egg). In only one of the 15 successful ovipositions was a fruit found to contain more than one egg.

Females in the field oviposited in a wide range of fruit sizes (range: 7.0 - 13.0 mm diameter). No significant

TABLE 1. Comparison by fruit dissection of successful and unsuccessful oviposition attempts and their associations with HMP dragging by *R. berberis* in the field.

OVIPOSITION	POST-OVIPOSITION BEHAVIOUR			
	Drag	No Drag	Total	
Successful (egg)	13	2	15	G-test
Unsuccessful (no egg)	4	6	10	$p < .02$
Total:	17	8	25	

TABLE 2. Comparison of size of emerged adult flies of *R. berberis* with the diameter of the fruits in which they developed.

SEX	SIZE PARAMETER	r_s *
Male	Weight	0.028
"	Head Capsule Width	0.077
"	Pronotal Width	0.34
Female	Weight	0.18
"	Head Capsule Width	0.21
"	Pronotal Width	0.068
Male & Female	Weight	0.14
" "	Head Capsule Width	0.3
" "	Pronotal Width	0.043

* Spearman's Rank Correlation Coefficient

correlation, however, exists between fly size and the diameter of the fruit from which each fly emerged. (Table 2). During the process of measuring fly size and fruit diameters we did not encounter more than one fly emerging per fruit (N = 48).

Lab Experiments

Results (Table 3) indicated that 84% of females rejected the egg-infested and dragged fruit (+ +) while they generally accepted both egg-infested only (+ −) and uninfested fruit (− −), 87% and 92% respectively. Every female that accepted an uninfested or infested only fruit successfully oviposited and followed egg-laying with ovipositor dragging.

Females readily climbed onto and spent similar amounts of time searching the surface of all three fruit types: 17.6 s (SE = 2.9 s) on type (+ +), 18.4 s (SE = 2.0 s) searching type (+ −) and 23.6s (SE = 2.6 s) on type (− −). Statistically, no significant differences exist between the search times (Mann-Whitney U-test: $p = <.05$).

Females initiated feeding behaviour by puncturing the fruit surface with their ovipositors. They then turned

TABLE 3. Response of *R. berberis* females to host fruit types offered in the lab.

HOST TYPE	ACCEPT	REJECT	
Uninfested (- -)	12	1	
Egg-infested only (+ -)	13	2	n.s.
Egg-infested & dragged (+ +)	4	21	$p = <.001$

(+ -) and (+ +) fruit were each compared to the control (- -) with a G-test.

TABLE 4. Response of *R. berberis* females to fruit surface punctures following oviposition attempts.

OVIPOSITION	FEED	NO FEED	
Successful (egg)	5	26	G-test
Unsuccessful (no egg)	14	6	$p = < .001$

about and placed their mouth parts into the puncture. Data from feeding observations (Table 4) show that 70% of unsuccessful ovipositions were followed by feeding while only 18% of successful ovipositions were followed by feeding.

DISCUSSION

First, both field observations and lab experiments indicate that *R. berberis* females generally follow egg-laying with ovipositor dragging of the host fruit surface. Most importantly, lab results indicate that it is not the presence of an egg but rather the dragging that enables females to discriminate. Thus, it follows that females detect a substance deposited on the fruit during dragging. Several factors give weight to this conclusion: firstly, evidence for the existence of this substance comes from our observation of a fine, thread-like trail on the fruit, visible briefly, following ovipositor dragging. Secondly, the fact that all pre-tested females readily climbed onto and searched each fruit type equally, indicates that physical contact with the fruit surface is necessary for determination of its quality. Thirdly, contact pheromone markers are used by several species within the *Rhagoletis* genus including *R. pomonella, R. cerasi, R. completa, R. fausta, R. cingulata, R. indifferans, R. mendax, R. cornivora, R. tabellaria* and *R. basiola* (Prokopy, 1981). Thus, we conclude that *R. berberis* employs a contact marking pheromone to aid in host discrimination.

The usage of host marking pheromones is functionally significant in several ways. HMPs appear to be the only means by which *R. berberis* females can detect the presence of an egg after it has been laid. HMPs signal egg presence to other foraging females enabling them to avoid conspecific competition and thereby enhancing their reproductive fitness. Recent theoretical studies (Roitberg & Prokopy, 1986) however, suggest the functional significance of HMPs is that they signal to the female that laid the egg initially that it has already exploited a particular fruit. Therefore, additional eggs should not be laid in the same fruit to avoid sibling competition for limited food and space. In either case, as our data suggest, single *M. aquifolium* fruits support only one larva, so that rejection of marked fruit should enhance the fitness of parents through increased offspring survival. In addition, Price (1970), suggested that females' response to HMPs enhances foraging efficiency via dispersal of females away from areas already heavily exploited.

Second, our lack of correlation between fruit diameter and fly size indicates that competition between larvae may be far more deleterious than variation in fruit size. Thus, it is not surprising that females do not appear to discriminate between different sized fruit for oviposition sites.

Third, results suggest that the females' feeding behaviour, at fruit surface punctures, has a single functional significance, that of obtaining nutrients. If this phenomenon were related to offspring survival we might expect to observe a high correlation between oviposition and feeding. In fact, feeding rarely followed oviposition.

Finally, we hope knowledge of this HMP system will help us to reach an overall understanding of tephritid marking systems. Such an understanding will aid in future management of both harmless and damaging *Rhagoletis* species. Already, recent computer simulation studies (Roitberg & Angerilli, 1986) show employment of HMPs in orchards, in conjunction with traps, may provide effective population control at rates comparable to chemical biocides.

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REFERENCES

Bush, G.L. 1961. *Rhagoletis* in North America. Bulletin of the Museum of Comparative Zoology. 134, No. 11: 508-511.

Price, P.W. 1970. Trail odours: Recognition by insects parasitic on cocoons. Science. 170: 546-547.

Prokopy, R.J. and E.F. Boller. 1971. Artificial egging system for the European cherry fruit fly. Journal of Economic Entomology. 63: 1413-1417.

Prokopy, R.J. 1981. Oviposition deterring pheromone system of apply maggot flies in: (E.R. Mitchell, Ed.) *Management of Insect Pests with Semiochemicals*. Plenum Publishing Corporation 477-494.

Prokopy, R.J., Roitberg, B.D. and A. Averill. 1982. Chemical mediation of resource partitioning in insects in: (R. Carde and W. Bell, Eds.) *Chemical Ecology of Insects*. Chapman and Hall. London. 301-330.

Prokopy, R.J. and B.D. Roitberg. 1984. Foraging behaviour of true fruit flies. American Scientist. 72: 41-49.

Roitberg, B.D. and N. Angerelli. 1986. Management of temperate orchard pests: Applied behavioural ecology. Agricultural Zoology Reviews. 1: (in press).

Roitberg, B.D. and R.J. Prokopy. 1986. Marking pheromones. Bioscience. (in press).

Salt, G. 1934. Experimental studies in insect parasitism. II. Superparatisism. Proceedings of the Royal Society of London. Series B. 114: 455-476.