HOST PREFERENCE AND LIFE CYCLE PARAMETERS OF *CHROMATOMYA HORTICOLA* GOUREAU (DIPTERA: AGROMYZIDAE) ON CANOLA CULTIVARS

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ABSTRACT: *Chromatomyia horticola* Goureau (Diptera: Agromyzidae) is a pest of canola crop, *Brassica napus* L., in the Ardabil region. The resistance of six canola cultivars to *C. horticola* was studied in the two following experiments: (1) host feeding selection and (2) life cycle study. The host feeding selection of females, were carried out in a greenhouse at 23±1°C, 50±5% RH and 14L:10D; the number of punctures and mines on Hyola401, RGS003 and Opera was significantly lower than that on Talayh. In the life cycle study, the development, survivorship and reproduction of *C. horticola* raised on five canola cultivars, was studied under defined condition. The female lifetime of leafminer was 27.43d on Talayh, 28.67d on Zarfam, 29.8d on RGS003, 30.2d on Hyola401 and 32.5d on Opera; the female lifetime on Opera, Hyola401 and RGS003 was significantly longer than that on Talayh. The mean lifetime fecundity of leafminer on Talayh, Zarfam, RGS003, Hyola401 and Opera was 80.15, 82.18, 74.4, 76.1 and 67.57 eggs, respectively. Based on these results, we conclude that Talayh is the most suitable and Opera is the least suitable host plant for *C. horticola*.

KEY WORDS: *Chromatomyia horticola*, host suitability, life-history parameters, leafminer

Chromatomyia horticola Goureau (Diptera: Agromyzidae) is a pest of canola crop, *Brassica napus* L., in the Ardabil region. Saljoqi et al. (2006) reported that *C. horticola* is a major pest of canola crop in Pakistan. Different species of leafminers might gradually become a serious pest of cultivated crops, due to high fecundity, short generation time, a wide range of host plants and their dispersal ability (Leibee, 1984; Parrella, 1987; Minkenberg, 1988; Mason et al., 1989; Zou et al., 1998; Wei et al., 2000; Saljoqi et al., 2006). Punctures caused by females of different species of leafminers during the feeding and oviposition processes can result in a stippled appearance on foliage, especially at the leaf tip and along the leaf margins (Parrella et al., 1985; Parrella, 1987; Wei et al., 2000). However, the major form of damage is the mining of leaves by larvae, which results in destruction of leaf mesophyll. The mine becomes noticeable about three to four days after oviposition and becomes larger in size as the larva matures. The pattern of mining is irregular. Both leaf mining and stippling can greatly depress the level of photosynthesis in the plant (Leibee, 1984; Parrella, 1987; Zou et al., 1998; Capinera, 2008).

Growers depend on insecticides for suppression of leafminers. However, different species of leafminers are capable of becoming resistant to insecticides, due to high fecundity and short generation time (Parrella, 1987; Zou et al., 1998; Wei et al., 2000). Moreover, the negative environmental impacts of insecticides have promoted the other alternative approaches such as host plant resistance. Host plant resistance is part of an effective strategy of integrated pest management programs that can reduce leafminer damage. So, the application of resistance plant cultivars has economical, ecological and environmental benefits (Smith, 1989).

Regardless of broad publications about plant characteristics that provide mechanisms of resistance, no study has been carried out about the resistance of canola (*Brassica napus* L.) cultivars to *C. horticola*. The purpose of this research was to the determination the host feeding selection of females on canola cultivars, and the study of the life cycle parameters of this leafminer on canola cultivars.

MATERIAL AND METHODS

Plant Collections

Two seeds of each cultivar were planted in pots, 20cm in diameter, filled with suitable soil at 10 replicates. Plants were grown at 23±1°C, 50±5% RH and 14L:10D. No pesticides were applied to the plants. The canola cultivars at seedlings stage with six leaves were used for these experiments; due to damage of leafminer to mature plants they have no influence on the growth of the plant. In contrast, seedlings are severely weakened by leafminer damage.

Insect and General Rearing Procedure

The primary population of leafminer was collected on cabbage in the Agricultural Station of the University of Mohaghegh Ardabili in June-2008. Leaves with larvae were collected and kept in culture dishes. To avoid a collected plant effect on the following experiments, the leafminer were reared on cabbage, *brassica oleracea* L. The leafminer colony was maintained in a polypropylene box (50cm diameter and 50cm depth) under laboratory defined conditions. A 40cm diameter hole was cut into the box lid, and this hole was covered by a nylon screen (ca. 0.1mm mesh). After two or three generations, 2-d old females were used for the experiments.

Host Feeding Selection

Host feeding selection of leafminer females was tested for six canola cultivars including: Zarfam, RGS003, Opera, Option500 and Hyola401 with Talayh as control cultivar. These experiments were conducted at seedlings stage with six leaves in a greenhouse at 23±1°C, 50±5% RH and 14L:10D. One test plant (*N* 10 for each cultivar) was placed in one circle route in a plastic box (2m diameter and 70cm depth). Ten females, 2-d old, from the stock leafminer colony were released in this box. The top of this box was covered using a nylon screen and kept for 4 day. After 4 days, the number of feeding punctures and mines on each cultivar in box was counted. Each treatment was replicated 10 times.

Life Cycle Study

The life cycle parameters of leafminer were studied on a middle leaf at seedling stage of five canola cultivars. One egg, selected randomly from eggs laid on a leaf and the other eggs were removed from the test plant leaves. Then each leaf with one egg of leafminer was restricted using a clip cage (6cm diameter and 1.5cm depth). Each clip cage has a mesh lid (2cm diameter) for ventilation. The margin of clip cages were covered with sponge to suppress injury to leaf tissue when linked to the leaf. The plants were then maintained under defined conditions in a greenhouse. The clip cages were monitored daily to egg hatching and to measure development time and survival of larvae and pupa. These observations were recorded until emergence of adults. The sex of emerged adults on each cultivar was recorded. At the emergence of adult, one pair leafminer was transferred to a new cage for ovipositing. The leaf in the cage was cut daily, and the oviposited eggs in the leaf were counted under a stereomicroscope. One pair leafminer was placed in new cage each day. Daily observations were made until the death of all adults. If a female died within the first 24h, she was replaced with a newly emerged and mated female. In this experiment, the number of eggs hatched, incubation period, and development time, longevity of female and fecundity on each cultivar were recorded. Each treatment was replicated 25 times.

Data Analysis

Prior to analysis, in order to correct for the heterogeneity of variance, all the data were log-transformed log(x+2). In the laboratory experiments, the data of the host feeding selection and the life cycle parameters of leafminer on canola cultivars were analyzed by one-way ANOVA, and the differences were compared by the Tukey's HSD test or Student-Newman-Keuls test (PROC ANOVA, SAS Institute 1999).

RESULTS

Host Feeding Selection

In the host feeding selection of leafminer, the lowest and highest number of punctures was significantly observed on Hyola401 and on Talayh, respectively. The number of punctures on Option500 and Zarfam was significantly higher than that on RGS003 and Opera (df= 5, 54; F= 34.05; P= 0.0001) (Table 1). The number of mines on Hyola401, RGS003 and Opera was significantly lower than that on Talayh. The number of mines on Zarfam and Option500 was moderate (df= 5, 54; F= 4.05; P= 0.0034) (Table 1).

Life Cycle Study

The life cycle parameters of leafminer reared on the five canola cultivars are summarized in Table 3. The incubation period of leafminer ranged from 2.41d to 2.82d and was not significantly different among the five canola cultivars (df= 4, 80; F= 0.92; P= 0.454) (Table 2).

The development time of larvae on Opera was significantly longer than that on Talayh and Zarfam. The development time of larvae on RGS003 and Hyola401 was significantly shorter than that on Opera, and was longer compare to that on Talayh and Zarfam (df= 4, 56; F= 15.47; P= 0.0001) (Table 2).

The development time of pupa was not significantly different among the five canola cultivars (df= 4, 46; F= 0.86; P= 0.49) (Table 2).

The longevity of female and male was not significantly different among the five cultivars (df= 4, 23; F= 2.4; P= 0.07 and df= 4, 18; F= 0.56; P= 0.69). The females reared on Opera had a significantly longer lifetime than those reared on Talayh and Zarfam. The females reared on RGS003 and Hyola401 was moderate lifetime among the five cultivars (df= 4, 23; F= 8.14; P= 0.0003). The males reared on RGS003, Hyola401 and Opera had a significantly longer lifetime than those reared on Talayh and Zarfam (df= 4, 18; F= 7.6; P= 0.0009) (Table 2).

The lifetime fecundity on Talayh and Zarfam was significantly higher compared to that on Opera and RGS003. The lifetime fecundity on Hyola401 was moderate among the five cultivars (df= 4, 46; F= 4.28; P= 0.005) (Table 2).

The egg-to-adult survival of leafminer was decreased in the following order: Talayh, Zarfam, Hyola401, RGS003 and Opera, respectively (Table 2).

The ratio of females that emerged in the development experiment was 0.54 on Talayh, 0.53 on Zarfam, 0.55 on RGS003, 0.56 on Hyola401 and 0.57 on Opera, respectively (Table 2).

DISCUSSION

In this study, the resistance of six canola cultivars to *Chromatomia horticola* was studied in the two following experiments: (1) host feeding selection and (2) life table study. In the host feeding selection experiments, the number of punctures and mines on Hyola401, RGS003 and Opera was significantly lower than that on Talayh. The canola cultivars with more feeding punctures and mines were more favored by females of leafminer. Therefore, Talayh was more favored and Opera was less favored by females of leafminer. It was reported that the number of feeding punctures and live larvae in mines in leaves was considered as an indicator of host feeding selection of *Liriomyza huidobrensis* (Blanchard) (Wolfenbarger, 1954; Wolfenbarger & Wolfenbarger, 1966; Zehnder & Trumble, 1985; Wei et al., 2000).

This study demonstrated that the type of canola cultivars had a significant effect on the development, survival and reproduction of leafminer. The development time, survivorship and fecundity of insects, reflects the suitability of the host plant (Smith, 1989; Panda & khush, 1995). However, the survival and lifetime fecundity of leafminer varied across cultivars of canola. In this study, development time was shorter and survival was higher in leafminer reared on Talayh than those reared on RGS003, Hyola401 and Opera. Moreover, our measurement of fecundity of leafminer reared on Talayh (80.15 eggs) and Zarfam (82.18 eggs) was higher than those reared on Opera (67.57 eggs), RGS003 (74.4 eggs) and Hyola401 (76.1 eggs). These results obtained in our study suggest that Talayh is the most suitable cultivar and Opera is the least suitable cultivars for *C. horticola*. The generation times of *Liriomyza trifolii* (Burgess) was reported 19d to 28d depending on host plant and temperature (Parrella et al., 1983; Leibee, 1984; Minkenberg, 1988; Capinera, 2008). However, to obtain further precise life-history parameters of *C. horticola*, we should study the effect of host plant leaf stages on the development and reproduction of *C. horticola*. Differences in the population growth rate of a pest reared on different host plants influence the effectiveness of using natural enemies such as biological control agents (Yano, 2004).

No study in the resistance of plant to *C. horticola* has been carried out. Host feeding selection of different species of leafminers was influenced by the differences in plant materials (Parrella et al., 1983; Carolina et al., 1992). Leafminers preferred to feed and deposit eggs on plants with high nitrogen content (Minkenberg & Fredrix, 1989; Minkenberg & Ottenheim, 1990). It was reported that distribution, density and length of leaf trichome affect the host selection of leafminers, the mobility and its feeding activities; high trichome density acts as a physical deterrent to *L. trifolli* (Fagoonee & Toory, 1983; Knodel-Montz et al., 1985; Alanerb et al., 1993). Whereas, Wei et al. (2000) were found that the density and length of leaf trichomes are not the main factors that influence the host feeding selection of *L. huidobrensis*. They concluded that the leaf physical structure, such as its thickness, thickness of the epidermis wall, densities of the palisade and spongy tissues, and so on, can play very important roles in the feeding and ovipositing by female leafminers and mining and development of larvae *L. huidobrensis*. Thickness of the epidermis wall most significantly correlated with host feeding selection by female *L. huidobrensis*, among characteristics of leaf tissue structure (Wei et al., 2000). The differences in these reported results may be due to the difference of the tested plant materials.

In this study, we conclude that Talayh is the most suitable and Opera is the least suitable host plant for *C. horticola*. These results can be used in integrated pest management.

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Table 1. Mean (±SE) number of punctures and mines of *Chromatomia horticola* on six canola cultivars.

Cultivar	The number of punctures per leaf	The number of mines per leaf
Talayh	16.9±1.02 a	1.9±0.22 a
Option500	14.4±1.06 b	1.2±0.44 ab
Zarfam	13.3±1.15 b	1.3±0.47 ab
RGS003	12.4±1.16 c	0.9±0.4 b
Opera	10.4±1.01 d	1±0.57 b
Hyola401	8.9±1.12 e	0.7±0.34 b

Means followed by the different letter within a column are significantly different (Tukey's HSD test; P<0.05)

Table 2. Mean (±SE) of life cycle parameters of *Chromatomia horticola* on five canola cultivars.

life cycle parameters	Talayh	Zarfam	RGS003	Hyola401	Opera
The incubation period (day)	2.41±0.35 a	2.53±0.44 a	2.64±0.55 a	2.76±0.58 a	2.82±0.57 a
The development time of larvae (day)	3.86±0.46 c	4.46±0.54 c	5.17±0.66 b	5.33±0.69 b	6.5±0.68 a
The development time pupa (day)	8.08±0.45 a	8.09±0.66 a	8.4±0.95 a	8.3±0.75 a	8.86±0.64 a
The longevity of female (day)	12.86±0.95 a	13.67±0.36 a	14.2±0.59 a	14.33±0.57 a	14.25±0.89 a
The lifetime of female (day)	27.43±1.2 c	28.67±0.96 bc	29.8±1.05 b	30.2±1.13 b	32.5±1.22 a
The longevity of male (day)	4.17±0.53 a	4±0.5 a	3.8±0.59 a	4±0.57 a	3.33±0.81 a
The lifetime of male (day)	22.5±0.97 b	23.8±1.26 b	26.4±1.54 a	27±0.58 a	28±1.87 a
The lifetime fecundity	80.15±6.7 a	82.18±4.6 a	74.4±4.41 b	76.1±3.2 ab	67.57±6.2 b
The total survival (%)	71	65	53	59	47
The female sex ratio	0.54	0.53	0.55	0.56	0.57

Means followed by the different letter within a row are significantly different (Student-Newman-Keuls test; P<0.05)