Distinguishing Larval Instars of the Vegetable Leaf-Miner Liriomyza huidobrensis (Diptera: Agromyzidae)

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Introduction

The vegetable leaf-miner, *Liriomyza huidobrensis* (Diptera: Agromizidae) is a highly polyphagous species known to attack ten different plant families including economic crops and ornamental plants (Wijesekara, 1997).

Larval parasitoid, *Diglyphus iseae*, has been introduced and has recorded higher levels of parasitism of the vegetable leaf -miner (Nugaliyadde *et al.*, 2000). *Diglypus isaea* females oviposit on larger hosts but reject or feed on smaller hosts (Parrella, *et al.*, 2005). Therefore, a need has arisen to develop an easy method to determine the correct time duration of different larval instars during the life cycle of vegetable leaf-miner, without help of sophisticated methods. The Major objective of this study, was to identify a simple and easy method to distinguish different larval instars of the *Liriomyza huidobrensis* under glass house conditions and specific objectives were, to identify the different larval instars of *Liriomyza huidobrensis* and to study the relationship between larval development time (in days) and larval instars of *Liriomyza huidobrensis.*

Materials and methods

Chinese cabbage (*Brassica chinensis*) nurseries were prepared using nursery trays for maintenance of host plants for *Liriomyza huidobrensis*. After 2 to 3 weeks seedlings were transplanted in to the pots. Pupae were collected and they were introduced in to the sand trays in insect proof cages. Adults emerging from pupae were used for oviposition process. Chinese cabbage lettuce plants were introduced daily into an insect proof cage with 400 leaf miner adults for oviposition and hatched live larvae were collected from third day onwards. This was continued at every 24 hours interval up to 12^{th} day of the oviposition. 400 live larvae were collected (40 larvae from each age) and they were preserved in 70% alcohol. Preserved larvae of each age were put into a separate vial containing 80% HCl for cleaning of the body contents and temporary slides were prepared.

Results and discussion

From the study, results were obtained on larval development time, length of mouth hooks and cephalpharyngeal skeleton and growth ratios.

Correlation studies

Correlation analysis between larval instar numbers with the length of cephalopharyngeal skeleton and length of mouth hooks is presented in Figure 1. According to the results significant positive correlation was observed between the lengths of cephalopharyngeal skeleton and mouth hooks and the larval instars.

Differentiation of larval instars of *Liriomyza huidobrensis*

Regression studies on length of cephalopharyngeal skeleton and the length of mouth hooks against presumed larval instar number gave significant difference.

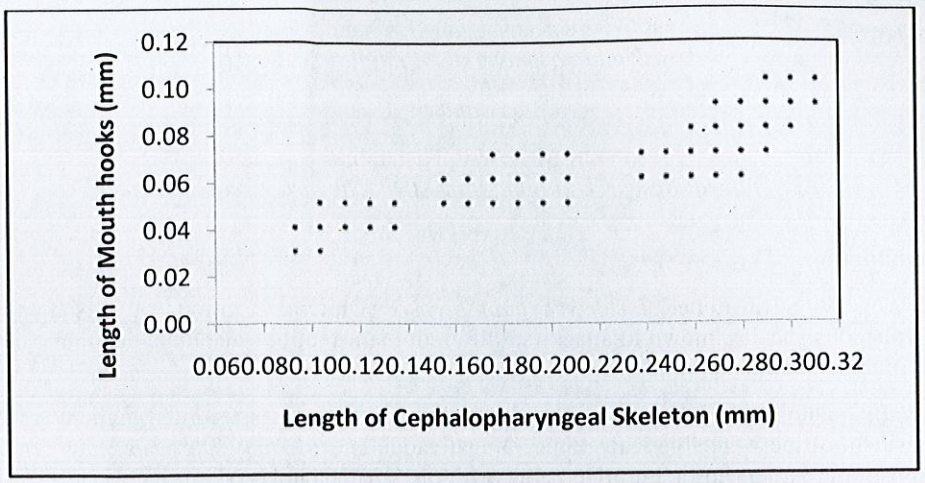

Figure 1: Relationship between length of mouth hooks and cephalopharyngeal skeleton for, first, second and third instars of *Liriomyza huidobrensis* larvae

Data on Figure 01 revealed that a clear separation exists among the three larval instars of *Liriomyza huidobrensis*. There was no overlapping observed in length of mouth hooks and cephalopharyngeal skeleton among three instars. When expected larval instar number was regressed with length of cephalopharyngeal skeleton ($r = 0.93$) and length of mouth hooks ($r = 0.75$) no unexpected deviation occurred. This clearly explained that no larval instar number was missed in this study. In the research, length of mouth hooks and cephalopharyngeal skeletons among the three larval instars were recorded as, 0.11 (0.09-0.14), 0.19 (0.16-0.21), and 0.27 (0.24-0.31) mm respectively. These values do not show a greater deviation as compared with the early studies, done by Parrella *et al.* (2005).

Growth ratios of the length of cephalopharyngeal skeleton

Table 1: Growth ratios of the length of the cephalpharyngeal skeleton of *Liriomyza huidobrensis* as compared with larvae of other *Liriomyza* species

Ratio of Instar	Ratio of length of cephalopharyngeal skeleton				
	L. huidobrensis	*L. sativa* [A]	*L. pictella* [B]	*L. trifolii* [C]	*L. brassicae* [D]
2/1	1.65	1.75	1.72	1.75	1.98
2/3	1.52	1.56	1.55	1.55	1.56

[A]Petit F. L. 1990 [B] Oatman & Michelbacher 1958 [C] Parella & Bethke 1988 [D] Beri 1974

According to the results, the growth ratio of cephalopharyngeal skeleton of first and second instar larvae of *Liriomyza huidobrensis* does not showa greater deviation, compared to the ratios of *Liriomyza sativa, Liriomyza pictella*, *Liriomyza trifolii* and *Liriomyz brassicae* species (Petitt, 1990).

Relationship between larval instar number and the larval development time

Figure 2 shows the relationship between the instar numbers and the larval development time. First instar larval period ended in 4 days after oviposition, whereas the 2nd larval

instar duration ranged from 5th to 8th day after oviposition and 3rd larval instar from 9th to 12th days after oviposition. Larvae were pupated in 12 days after oviposition.

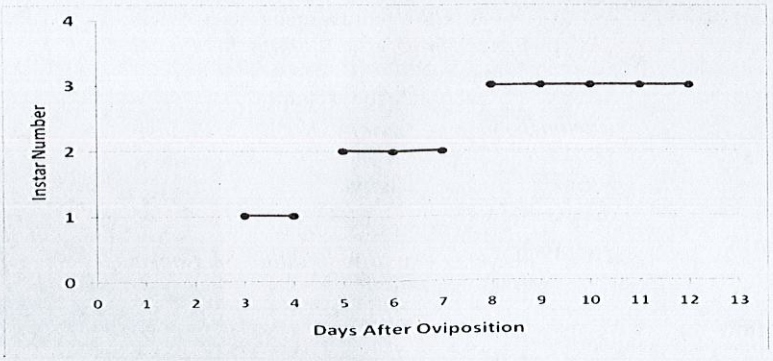

Figure 2: Relationship between larval instar and the larval development time (days after oviposition)

Conclusions

Liriomyza huidobrensis completed its larval stage within 12 days after oviposition at a temperature range of 13.2 - 19.2 ^{0}C and RH range from 80- 88% in the Nuwara - Eliya region. The length of mouth hooks and the cephalopharyngeal skeleton has a reciprocal relationship. This research is very helpful to determine the correct time duration of the different larval instars. Therefore, it helps to introduce the correct larval stages of the *Liriomyza huidobrensis* to the *Diglypus isaea* female parasites to increase the female ratio in the offspring populations in laboratory multiplication.

Acknowledgement

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