Journal of Entomology

ISSN 1812-5670

Academic Journals Inc.

www.academicjournals.com

Journal of Entomology 3 (3): 241-247, 2006
ISSN 1812-5670

Some Aspects on Oviposition Behaviour of *Spodoptera exigua* (Hübner) (Lepidoptera: Noctuidae)

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Abstract: *Spodoptera exigua* females oviposit on any host plant offered to them. However, they do show oviposition preference among the host plants. The hierarchy of oviposition preference in the three choice test was reflected in the five choice test. The oviposition hierarchy is shallot > long beans > chilly > cabbage > lady's finger. The females prefer to lay eggs on the host plant leaves especially on the abaxial compared to the other part of the plant. Besides that, the females also prefer to lay eggs on the cage. The leaves and leaf stalk position did not influence the oviposition preference.

Key words: *S. exigua*, oviposition behaviour, multiple host plant

Introduction

The beet armyworm, *Spodoptera exigua* (Hübner) is a serious pest of numerous cultivated and wild plants throughout the world. Host preference for oviposition is a very important component of the insect and its host plants interactions when multiple potential hosts are presence. There is not much information available in the literature on the egg laying behaviour of the *S. exigua* on multiple host plants. However, a few studies have been done in Texas (Greenberg *et al*., 2002; Showler, 2001). Further, no such information has been recorded in Malaysia. Nevertheless, several studies have addressed *S. exigua* oviposition behaviour in no-choice situations (Sappington *et al*., 2001; Showler and Moran, 2003; Smits *et al*., 1986; Yoshida and Parrella, 1991). A very important component of such interactions is that of host preference for oviposition, but information on this subject with respect to *S. exigua* is limited (Greenberg *et al*., 2002). Information is needed regarding relative preferences when in the presence of multiple potential hosts.

Whereas, an understanding of the within-plant distribution of eggs on the host plant provides insight manner how the host plant is utilized by a foraging adult female. In addition, the location of eggs within the plant is also important to ensure higher survivorship of its progeny. For *S. exigua*, such information is currently scarce. Undoubtedly, development of effective alternative strategies for managing the *S. exigua* will require a thorough knowledge of the biological interactions of the insect and its host plants. Information on this aspect is important to understand the nature of female behaviour in egg deposition on plants.

This study aimed to determine (a) the oviposition behaviour and preference of *S. exigua* on various plants and (b) within plant distribution of eggs on a plant. So, the objectives of this study are to evaluate *S. exigua* oviposition preferences and egg deposition patterns on host plant.

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Materials and Methods

Three Host Plants

The ovipositional preference of *S. exigua* on cabbage, shallot and chilly was investigated under laboratory condition with the temperature and relative humidity ranged from 24-26°C and 56-84% respectively. The leaves were obtained from plants grown in the Institute of Biological Sciences garden. These plants were selected based on their importance as cultivated crops in Malaysia or severely attacked by this pest as reported by the Malaysian Department of Agriculture. Intermediate leaves of cabbage (i.e., 4th leaf position on the plant from the terminal bud), shallot and chilly were excised near the axil with scissors for ovipositional substrate. Each of the leaves was immediately inserted into a glass vial with water and were placed in a cylindrical plastic cage (40 cm diam and 45 cm high) with a muslin cloth top. A newly emerged female and male were confined in each cage until the death of the female. Adults were supplied with a soaked cotton ball of 10% honey solution for feeding which was replenished daily.

Cages were examined daily to record the number of eggs laid and egg masses on each plant leaf. One cage containing leaves from three plant species was a replication and there were 10 replications.

Five Host Plants

The same procedure was repeated for ovipositional preference of *S. exigua* on five host plant leaves. The five host plants used are cabbage, shallot, chilly, long beans and lady's finger. One cage containing leaves from five plant species was a replication and there were 14 replications.

Within Plant Distribution of S. exigua Eggs on Cabbage

The within plant distribution of eggs of *S. exigua* was also examined under laboratory conditions, as mentioned earlier. Three pairs (3 females and 3 males) of one day old laboratory reared adult *S. exigua* were confined in a plastic cylindrical cage (40 cm diam and 45 cm high) with muslin cloth top, with a one and half month old intact cabbage plant. Each plant was trimmed to ten leaves before it is used in the experiment. The position of each leaf was labeled from 1 to 10 starting from the first fully opened leaf. Adults were supplied with a soaked cotton ball of 10% honey solution for feeding which was replenished daily.

Cages were examined daily to record the egg's position and the number of eggs laid on the plant and non host plant for five days. One cage containing one intact cabbage plant was a replication and there were 10 replications.

Data Analysis

One way ANOVA (STATISTICA 6.0; StatSoft Inc., 1984-2001) were performed to detect effects of host plant on oviposition preference of *S. exigua* females. When significant F values were obtained, means were separated using the Duncan's multiple range test. Besides one way ANOVA, t-test for independent samples by groups were performed to detect effects of host plant or part of host plant on oviposition preference of *S. exigua* females. Homogeneity of variances was tested before the data were analyses with ANOVA. If the variances were not homogenous, then the data were transformed first, as the following.

Data for number of eggs laid and proportion of eggs on plant and cluster of eggs on five host plants were log + 0.5 and square-root transformed, respectively. Data on number of eggs on leaf surface, cluster of eggs on leaf surface, cluster of eggs on host plant and non host plant, number of eggs between leaves and cluster of eggs between leaves were square-root transformed. While number of eggs on different parts of plant and number of eggs between leaf stalks were double square-root transformed. Data on number of eggs on host plant and non host plant was log + 1 transformed.

Results

In a choice test of three host plants, the number of eggs laid was significantly different ($F = 3.49$; $df = 2, 27$; $p<0.05$) among the tested leaves. The number of eggs laid on the shallot (48%) was higher compared to the cabbage (11%) leaves, while chilly (41%) was intermediate (Table 1). There was a significant difference ($F = 4.18$; $df = 2, 27$; $p<0.05$) in the proportion of eggs laid on a plant to the total that were deposited on all plants tested. The proportion of total eggs laid also showed the same hierarchy as the number of eggs laid (Table 1). The number of egg cluster also was significantly different ($F = 4.25$; $df = 2, 27$; $p<0.05$) among the three host plants. There are more egg clusters on both chilly and shallot compared to cabbage (Table 2).

In a choice test of five host plants, the number of eggs laid was significantly different ($F = 3.31$; $df = 4, 65$; $p<0.05$) among the tested leaves. The number of eggs laid on the long beans (39%) and shallot (33%) higher compared to the cabbage (8%) and lady's finger (6%), while chilly (14%) was intermediate (Table 3 and 4). There was a significant difference ($F = 2.76$; $df = 4, 65$; $p<0.05$) in the proportion of eggs laid on a plant to the total that were deposited on all plants tested. When simultaneously presented with all five plant species, the proportion of total eggs laid on cabbage was not significantly different from chilly (Table 3), but was significantly higher than the proportion for lady's finger. Whereas, long beans and shallot have the highest proportion. The number of egg cluster also was significantly different ($F = 2.86$; $df = 4,65$; $p<0.05$) among the five host plants. There were more egg clusters on shallot compared to chilly, cabbage and lady's finger, while long beans was intermediate (Table 4).

Based on location on the plant the number of eggs laid on the leaf lamina was significantly higher ($F = 18.98$; $df = 2, 27$; $p<0.05$) than those on the leaf stalk and stem, where 89.04% of the eggs were laid on the leaf (Table 5).

Table 1: Number and proportion of eggs laid on three host plants

Host plant leaves	Eggs/day (Mean±SE)	Proportion of eggs on plant/total laid on plant
Cabbage	2.54±1.42a	0.11a
Shallot	8.73±1.94b	0.50b
Chilly	6.88±1.70ab	0.38ab

*Mean accompanied by the same letter within test or column are not significantly different

Table 2: Percentage and number of cluster of eggs laid on three host plants

Host plant leaves	% eggs laid on the leaves	No. of egg cluster/day (Mean±SE)
Cabbage	11.31	0.12±0.06a
Shallot	47.58	0.43±0.9b
Chilly	41.11	0.49±0.13b

*Mean accompanied by the same letter within test or column are not significantly different

Table 3: Number and proportion of eggs laid on five host plants

Host plant leaves	Eggs/day (Mean±SE)	Proportion of eggs on plant/total laid on plant
Cabbage	2.77±1.40a	0.41ab
Shallot	13.51±4.60b	0.29b
Chilly	5.18±2.33ab	0.17ab
Long beans	12.85±6.28b	0.24n
Lady's fingers	1.23±0.55a	0.06a

* Mean accompanied by the same letter within test or column are not significantly different

Table 4: Percentage and number of cluster of egg laid on five host plants

Host plant leaves	% eggs laid on the leaves	No. of egg cluster/day (Mean±SE)
Cabbage	8.07	0.25±0.09a
Shallot	33.2	1.15±0.41b
Chilly	13.5	0.26±0.06a
Long beans	39.17	0.67±0.18ab
Lady's finger	6.00	0.28±0.11a

*Mean accompanied by the same letter within test or column are not significantly different

There was also a significant difference ($p<0.05$) in the number of eggs laid on the surface of the leaf. It was found that the female of *S. exigua* laid more eggs on the lower (abaxial) compared to the upper (adaxial) surface of the leaf (Table 6), where 77.19% was laid on the abaxial. Further, the number of egg clusters laid on the lower surface was significantly more ($p<0.05$) than on the upper (Table 6).

It was also found that the *S. exigua* female also laid many eggs on the cage (26%) besides the plant leaves (Table 7). The number of eggs laid on the cage was significantly more ($p<0.05$) than on the leaf stalk, stem and food place (Table 7). There was a significant difference ($F = 17.46$; $df = 4, 45$; $p<0.05$) in the number of egg clusters laid on both host plant and non host plant. This study has found that the number of egg clusters laid on both leaf and cage were significantly higher than food place, leaf stalk and stem (Table 7).

There was no significant difference ($p>0.05$) in the number of eggs laid between the leaves on the plant (Table 8), although relatively a higher percentage of eggs were laid on the leaf number 4 (19%). There was also no significant difference ($p>0.05$) in the number of egg clusters laid between the leaves, although relatively eggs were frequently laid on the leaf number 4 (Table 8).

There was no significant difference ($p>0.05$) in the number of eggs laid between the leaf stalks on the plant (Table 9), although relatively a higher percentage of eggs were laid on the leaf stalk number 10 (25%). There was also no significant difference ($p>0.05$) in the number of egg clusters laid between the leaf stalks.

Table 5: Mean and percentage of egg's laid on various part of the host plant

Location on plant	No. of eggs laid (Mean±SE)	% of total eggs laid
Leaf	74.76±17.58a	89.04
Leaf stalk	5.18±1.25b	6.17
Stem	4.02±1.55b	4.79

*Mean accompanied by the same letter within test or column are not significantly different

Table 6: Mean and percentage of egg's laid on leaf surface

Leaf surface	No. of eggs laid (Mean±SE)	% of total eggs laid	No. of egg cluster (Mean±SE)
Upper/adaxial	24.28±7.03b	22.81	4.6±1.12a
Lower/abaxial	82.2±17.94a	77.19	11.0±2.94b

*Mean accompanied by the different letter within test or column are significantly different

Table 7: Mean and percentage of eggs laid on plant and non host plant

Location	No. of eggs laid (Mean±SE)	% of total eggs laid	No. of egg cluster (Mean±SE)
Leaf	74.76±17.58a	61.03	15.6±3.53a
Leaf stalk	5.18±1.25b	4.23	2.2±1.07bc
Stem	4.02±1.55b	3.28	1.6±0.65c
Cage	31.76±3.96a	25.93	15.2±1.83a
Food place	6.78±1.55b	5.53	5.6±1.45b

*Mean accompanied by the same letter within test or column are not significantly different

Table 8: Number and percentage of eggs laids and number of egg clusters between leaves

Leaf No.	No. of eggs laid (Mean±SE)	% of total eggs laid	No. of egg cluster (Mean±SE)	Total No. of egg cluster
1st	4.44±2.68	5.94	0.8±0.59	8
2nd	6.68±2.95	8.94	1.5±0.82	15
3rd	3.94±1.96	5.27	0.8±0.47	8
4th	14.36±5.26	19.21	3.1±1.85	31
5th	4.38±1.99	5.86	1.0±0.42	10
6th	11.94±4.99	15.97	2.2±0.79	22
7th	8.88±3.67	11.88	1.6±0.58	16
8th	10.3±3.22	13.78	2.4±0.82	24
9th	7.62±2.64	10.19	1.4±0.43	14
10th	2.22±1.09	2.97	0.8±0.39	8

Table 9: Number and percentage of egg laid and number of egg clusters between leaf stalk

Leaf stalk No.	No. of eggs laid (Mean±SE)	% of total eggs laid	No. of egg cluster (Mean±SE)	No of egg cluster
1st	0.26±0.26	5.02	0.1±0.1	1
2nd	0.08±0.08	1.54	0.1±0.1	1
3rd	0.18±0.18	3.47	0.1±0.1	1
4th	0.74±0.47	14.28	0.3±0.15	3
5th	0.94±0.59	18.15	0.3±0.21	3
6th	0.16±0.13	3.09	0.2±0.2	2
7th	0.60±0.37	11.58	0.3±0.15	3
8th	0.36±0.25	6.95	0.2±0.2	2
9th	0.58± 0.37	11.19	0.3±0.21	3
10th	1.28±0.64	24.71	0.3±0.15	4

Discussion

The beet armyworm's polyphagous nature has been interpreted as evidence that the moth oviposits indiscriminately on any available plant species (Atkins, 1960). This study has shown that the *S. exigua* females do oviposit on any plant offered to them. However, results of this study have shown that oviposition preferences existed among the three host plants (Table 1 and 2) and among the five host plants tested (Table 3 and 4). Greenberg *et al*. (2002) also reported the same phenomenon for their oviposition preference study on cabbage, cotton, bell pepper, pigweed and sunflower. In their study, the highest proportion of total eggs laid was on pigweed, followed by cotton and bell pepper, while sunflower and cabbage were the least. Similar hierarchy of female *S. exigua* oviposition preference on cabbage was found in both studies, where it was the less attractive.

In general, the hierarchy of oviposition preference shown in the three choice test was reflected in the five choice test. The highest number of eggs laid was on shallot followed by chilly and cabbage was the least. In the five choice test, the number of eggs laid follows the same hierarchy i.e., shallot > long beans > chilly > cabbage > lady's finger. However, long beans seem to be one of the preferred host plants besides shallot and chilly was still intermediate (Table 3). However, when referring to the number and proportion of eggs laid, lady's finger was the most not appealing to the *S. exigua* female compared to cabbage for oviposition when all the five host plant were existed together. Hence, this confirmed the existence of host preference of the *S. exigua* female within the tested host plants.

From both choice tests, it was reasonable to state that shallot was the most preferred host plant for oviposition. Whereas, cabbage was clearly less attractive and this was also supported by Greenberg *et al*. (2002).

In the five choice test, the number of eggs per mass averaged 55% less on cabbage (6.68) than on shallot (14.84) and long beans (14.78) and 46% less than on chilly. While, in the three choice test, the number of eggs per mass was 54% less on cabbage (7.88) than on shallot (17.14) and 43% less than on chilly (13.75). This may be related to be apparent oviposition preference of beet armyworm for long beans, shallot and chilly over cabbage. Sappington *et al*. (2001) also reported the same phenomenon occurred between cotton and pigweed, where the number of egg per mass averaged 29% less on cotton (58.6±2.75) than on pigweed (82.7±5.48) and that beet armyworm preferred pigweed over cotton.

The nature of the host plant cues recognized by beet armyworm females is unknown but could be chemical, tactile, visual or some combination. However, dependance of differential changes in oviposition behaviour towards a given host on the species of a nearby host suggests that chemical cues maybe more important than tactile cues and perhaps more important than visual cues in host plant discrimination by females (Greenberg *et al*., 2002). Thus, experiments designed to specifically test these hypotheses will be necessary to dissect the relative importance of potential cues influencing oviposition preference in these different context.

It is no doubt that knowledge of hierarchies of host plant oviposition preference by beet armyworm females will be useful in understanding the population dynamics of this important agricultural pest and for developing effective monitoring and management strategies (Greenberg *et al*., 2002).

The within plant distribution of eggs study has shown that the *S. exigua* female prefer to lay eggs on the host plant leaves compared to the other part of the plant. Smits *et al*. (1986) also has found that *S. exigua* females prefer to lay eggs on the chrysanthemum (97%), tomato (100%), gerbera (98%) and geranium (91%) leaves compared to their stem, flower and fruit. Zalom *et al*. (1983) stated that more eggs were laid on the leaf especially the one immediately interior to the inflorescence. Other species of *Spodoptera* i.e., *S. frugiperda* also were found to lay more eggs on the corn, sorghum, cotton and soybean leaves (Pitre *et al*., 1983).

The female prefer to lay eggs on the abaxial of the leaf. This finding was supported by Smits *et al*. (1986) and Zalom *et al*. (1983) who stated that over 89% of the *S. exigua* eggs batches were laid on the underside of leaves and 85.4% of the proportion of total eggs was laid on the ventral leaf surface, respectively. Besides that, Dibiyantoro (1996) also reported that *S. exigua* eggs were laid underneath the leaf. Pitre *et al*. (1983) also stated that the under surface of leaves was preferred for egg deposition when compared with the upper surface. The percentage of egg masses on the under surface of leaves was 100% for cotton and soybean, 92% for sorghum and 70% for corn. While Sappington *et al*. (2001) reported that of 267 egg masses deposited on cotton, all were found on the underside of leaves except six on the top of leaves, one on a leaf petiole and one on a boll. Whereas, of 137 egg masses oviposited on pigweed, all were located on the underside of leaves except three masses laid on the main stem of the plant (Sappington *et al*., 2001).

Other examples of insect species that exhibited a strong preference for ventral leaf surface as an oviposition site are *Heliothis zea*, *Manduca* spp., *Trichoplusia ni* and *Spodoptera frugiperda*. The proportion of total eggs for each species laid on the ventral leaf surface was 81.8% (*H. zea*), 86.0% (*Manduca* spp.), 83.3% (*T. ni*) (Zalom *et al*., 1983) and 92.4% (*S. frugiperda*) (Ali *et al*., 1989).

Thomson and All (1982) stated that *S. frugiperda* female not only deposits eggs onto host plant, but also onto objects and non host plants in the vicinity of the host plants. The same phenomenon has occurred in this study where the females prefer to lay eggs on the cage besides the plant leaves. Perhaps this is due to cue from the cabbage plant was not simply nonstimulating but were less attractive than the cage, as shown in the three and five choice tests. Greenberg *et al*. (2002) also reported that even when given no choice of host plant, females tended to lay more eggs on the cage than on cabbage. Besides that, Pitre *et al*. (1983) stated that the *S. frugiperda* females may also oviposit on nonplant material despite the presence of a host plant nearby.

In this study, the female does not seem to discriminate the leaves position for oviposition, which was supported by Sappington *et al*. (2001) who also found that neither height above the ground nor relative vertical regions of the canopy seem to be important factors determine egg mass placement within the pigweed canopy. Moreover, Burgess and Jarret (1976) stated that the oviposition pattern of beet armyworm is different from the other noctuids moths, which showed no particular preference for oviposition in any foliage zone. Nevertheless, this was not in concurrence with Smits *et al*. (1986) who stated that the eggs were predominantly found on the lower leaves of all plants. In cotton, about 80% of the egg masses were consistently located in the upper 50% of the cotton canopy (Sappington *et al*., 2001). Wheeler *et al*. (1998) reported that *Spodoptera pectinicornis* egg masses were found on leaves number 1 until 12, with majority on leaves No. 5 to 8. The females also did not show any preference on the leaf stalk position for oviposition.

Sappington *et al*. (2001) reported that spatial distribution of beet armyworm egg mass deposition within plant varies with host plant species and sometimes with developmental stage of the host. Further, Smits *et al*. (1986) stated that the distribution of oviposition sites on plants appeared to be

independent of the plant species, cultivar or age. However, within the oviposition zone, the moths can discriminated between plants or plant parts as it is shown in this study.

Acknowledgements

We would like to thank Dr. Lim Guan Soon for his suggestion and initiative of this project. We are grateful to CAB International resources for technical assistance and support. We also would like to thank our support staffs who have been kindly helping in this project in every other ways. This work is supported by the Malaysian government research grant, IRPA No. 01-02-03-0695.

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