Research Article

Comparing the Effect of Selected Cabbage Varieties on the Fitness of *Diadegma insulare* Cresson Parasitizing *Plutella xylostella* Linnaeus under Controlled Conditions

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Abstract | *Plutella xylostella* (Plutellidae: Lepidoptera) is the one of most destructive and critical cosmopolitan insect herbivores of brassica crops. Globally efforts have been undertaken to develop integrated management strategies for its control, based principally on manipulation of its parasitoids including *Diadegma insulare*. The research study was conducted to investigate the effects of two cabbage varieties on fitness parameters (i.e. percent parasitism, offspring sex ratio and developmental periods) of *Diadegma insulare*, a larval parasitoid of *Plutella xylostella*. All experiments were carried out under controlled conditions maintained at 23±2 ºC, 60±5% RH and 16L: 8D photoperiod. The findings revealed that developmental period was lower on Asha while higher on Golden acre whenever both second and third instars larvae were released on cabbage varieties. Parasitism efficacy was lower on third instars larvae as compared to the second. Higher parasitism (53.53%) was assessed on Asha while lower parasitism (44.95%) was observed on Golden acre when second instars larvae were exposed. Parasitism was higher (61.17%) on Asha while lower (50.1%) on golden acre when third instars larvae were exposed. Offspring sex ratio differed on stages of parasitized larvae. Higher male offspring emerged when second instars larvae were parasitized while more female offspring were emerged when third instars larvae were parasitized by *D. insulare* reared on both varieties. It was concluded that Asha was recognized as most suitable variety, as developmental period of *D. insulare* was shorter as well as parasitizing efficacy was also recorded higher on Asha.

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Introduction

Consistent struggles have been made by researchers to increase the productivity of cabbage crop. Practically all methods and strategies for the management of pest have been initiated to control *P. xylostella* (Glare and Gallagham, 2000). Among those proper selection of suitable varieties and management practices of crop are the important factors that contribute to growing cost-effective crucifers (Zerkoune, 2000). *P. xylostella* has also been managed by using chemical pesticides however it showed resistant among chemicals but control gone in vain in various parts of USA. To overcome this problem various other control tactics such as biological control, genetic control and cultural control is used (Shelton *et al.*, 1993). Biological control agents such as parasitoids, predators and plenty of diseases are crucially effective for managing *P. xylostella* (Sarfraz *et al.*, 2005). Among biological control agents, parasitoids are very important and about 135 species of parasitoids have been listed which parasitize on different life forms of *P. xylostella* (Billqvist and Ekbom, 2001). Parasitoids play a vital role in suppressing *P. xylostella* populations (Lohr *et al.*, 2007). Most important parasitoids belong to genus *Diadegma*, *Oomyzus*, *Cotesia* and *Diadromus* of order Hymenoptera, which have also been reported from Pakistan by Qadeem *et al.* (2015). *D. insulare* Cresson (Ichneumonidae: Hymenoptera) plays a vital role in managing *P. xylostella* infestation globally, as its parasitizing efficacy is highest as compared to other biological control agents. Current research has been designed on the importance biological control aspects of *Plutella xylostella* mainly focusing on *D. insulare,* as an effective control agent of *P. xylostella*.

The current study aimed to investigate the impact of selected cabbage varieties on the fitness parameters (developmental duration, percent parasitism and sex ratio) of *D. insulare* when *P. xylostella* completes its life cycle on selected cabbage varieties.

Materials and Methods

Culture of cabbage varieties

Two cabbage varieties (viz. Golden Acre and Asha) were used during current experimental studies. Each variety was grown in four seedling trays (i.e., 16×8 holes/ tray) separately in glass house of Entomology Department, PMAS, Arid Agriculture University, Rawalpindi. After thirty-five days of sowing, seedlings were transplanted into plastic pots filled with a mixture of compost and soil at 2:1 ratio. Transplanted cabbage plants from each variety (5-7 leaf stage) were utilized for further studies.

Culture of P. xylostella

P. xylostella in all life stages (e.g., egg, larval, pupal and adult) were collected from farmers' field of cabbage crop located at Chak Shehzad, Islamabad. Larvae were collected in plastic jars covered with muslin cloth, provided with fresh leaves and honey solution-soaked cotton plugs. Collected population was brought into Bio-control laboratory of Entomology Department, Pir Mehr Ali Shah- Arid Agriculture University, Rawalpindi (PMAS-AAUR). The larvae were then transferred into rearing cages (45×30×35 cm) made up of transparent plastic sheet and ventilated through openings covered with muslin cloth. In rearing cages, potted cabbage varieties were placed singly for maintaining insect culture of *P. xylostella*. Culture was developed under controlled conditions at 23±2°C, 60±5% RH and 16L: 8D photoperiod.

Culture of D. insulare

Pupae of *P. xylostella* were collected from farmers field of cabbage crop located at Chak Shehzad, Islamabad. Larvae were collected in glass jars singly covered with muslin cloth and provided with honey solution (20%) soaked cotton plugs and placed. After adult emergence, *D. insulare* were released in separate rearing cages for seventy-two hours, provided with honey solution (20%) soaked cotton plugs for mating (2:1). Whereas, emerged adults of *P. xylostella* in numbers were released on each cabbage variety separately for oviposition and culture development for 24 hours. Five mated female *D. insulare* adults were released in rearing cages having larval population of *P. xylostella* on each cabbage variety separately for parasitization. After 24 hours, adult *D. insulare* were removed from the rearing cages and placed in other cages for further parasitization.

Assessment of D. insulare fitness associated with P. xylostella on selected cabbage varieties

To assess the fitness of *D. insulare* on two cabbage varieties (viz. Golden Acre and Asha), second and third instar larvae of *P. xylostella* were utilized for the experimental studies. Twenty potted plants from each variety of cabbage were placed singly in rearing cages. One pair of three days old mated *D. insulare* were released for 24 hours on ten plants of each variety

whereas other ten plants received insect pest only (control). Rearing cages were kept under controlled conditions i.e., 23±2°C, 60±5% RH and 16L:8D. Insects were observed after every 48 hours and the number of surviving larvae of *P. xylostella* were noted until pupation. Pupae were collected in labeled glass vessels and placed under above mentioned controlled conditions.

Duration of development

To investigate developmental time taken by *D. insulare*, eight parasitized second and third instars larvae were isolated and kept in ventilated plastic jars containing leaves of cabbage varieties. For maintaining sanitize environment, the leaves were replaced on daily basis. Experiments were carried out for both second and third instars.

Parasitism percentages of D. insulare

Eight exposed larvae from each separate study were kept and reared in separate plastic jars in the laboratory until the emergence of *D. insulare* or *P. xylostella* adults. Number of emerged parasitoids were counted and recorded on daily basis. Number of adult *P. xylostella* in both cases and control were recorded as well as the dead ones. Natural mortality of *P. xylostella* larvae and pupae in the experiment were standardized.

Sex ratio of D. insulare

For this purpose, eight second and third instars larvae of *P. xylostella* were kept in separate closed container and mated female were released for parasitization. Sex ratio of *D. insulare* was expressed from the progeny adults that emerged from eight exposed larvae of *P. xylostella*. To measure the sex ratio of *D. insulare*, emerged males and females were counted separately. Females were simply identified with the presence of ovipositor at the end of abdomen.

Statistical analysis

Analysis of the collected data was performed using Statistics 8.1 statistical software. Completely randomized design (CRD) was followed for analysis of variance and means of significant treatments were compared using Tukey's HSD test at 5% level of significance. Graphical demonstrations were made using Microsoft Excel 2013 and statistical program Microcal Origin Graphic Package.

Results and Discussion

Assessment of D. insulare fitness associated with P. xylostella on selected cabbage varieties

Experimental studies were performed to assess the fitness of *D. insulare* on two cabbage varieties (Asha and Golden acre) in associated with *P. xylostella* when second and third larval instars were used. Parameters of percent parasitism and sex ratio of emerged parasitoids as well as developmental time of parasitized 2nd and 3rd instars larvae were observed and the findings are given below.

Duration of development

Development of *D. insulare* was significantly different on host larvae reared on selected cabbage varieties. The findings revealed that developmental time taken to reach larvae and then to pupae was shortest on Asha and longest on Golden acre. Similarly, the time taken by pupa to reach adult stage was shortest on Asha and longest on Golden acre (Table 1). Total developmental period from second instars to adult was shortest for parasitoid on Asha (12.11±0.34 days) and significantly different from Golden acre (14.3±1.13 days). While total developmental duration from third instars to adult was longest on Golden acre (8.68±0.54 days) and shortest on Asha (10.9±0.13 days) (Table 2).

Parasitism (%) by D. insulare on two larval instars of P. xylostella

Eight second and third instars larvae were released on both varieties (Golden Acre and Asha). Furthermore, larval parasitoids were released in a container containing immature of *P. xylostella*. Figure 1 showed that maximum parasitism level (53.53%) was assessed on Asha while minimum parasitism level

Table 1: *Developmental period (days) of parasitized P. xylostella reared on two cabbage varieties under laboratory conditions.*

Host varieties	Larval period (days)			Pupal Period (days)	Developmental time (days) from 2nd instars to Adult
	2nd instar	3rd instar	4th instar		
Asha	2.8±0.13a	2.9±0.15a	2.8±0.26c	3.61±0.13b	12.11±0.34b
Golden Acre	2.75±0.31a	3.32±0.35b	3.6±0.2d	4.68±0.8a	14.35±1.13a

Means sharing common letters in each column are not significantly different from each other.

Table 2: *Developmental period (days) of parasitized P. xylostella reared on two cabbage varieties under laboratory conditions.*

Host varieties	Larval period (days)		Pupal period (days)	Developmental time (days) from 3rd instars to Adult
	3rd instar	4th instar		
Asha	2.87±0.26a	3.2±0.18c	3.21±0.24ab	8.68±0.54b
Golden acre	3.16±0.28b	3.6±0.2d	4.68±0.15cd	10.9±0.13a

Means sharing common letters in each column are not significantly different from each other.

(44.95 %) was observed on Golden acre when *D. insulare* were reared on second instars larvae of *P. xylostella*. Figure 2 showed that maximum parasitism level (61.36 %) was assessed on Asha while minimum parasitism level (50.35 %) was observed on Golden acre when *D. insulare* were reared on third instars larvae of *P. xylostella*. The experiment was performed under controlled conditions. ANOVA table is also attached.

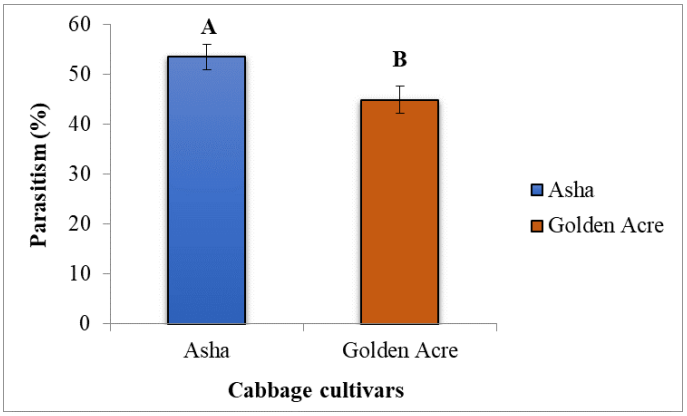

Figure 1: *Comparative parasitism (%) level of D. insulare on second instars larvae of P. xylostella reared on two varieties under controlled laboratory conditions.*

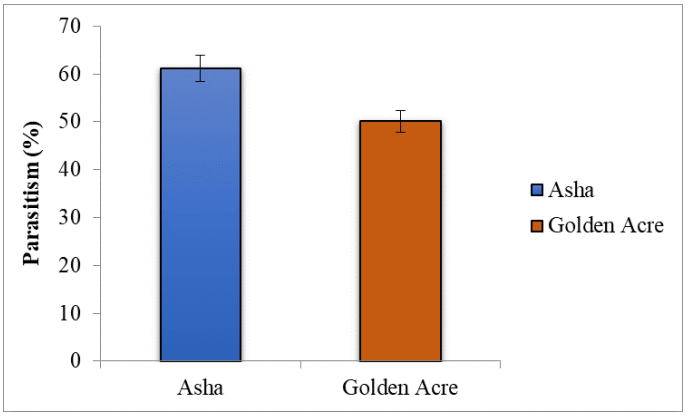

Figure 2: *Comparative parasitism % level of D. insulare on third instars larvae of P. xylostella reared on two varieties under controlled laboratory conditions.*

Sex ratio of F_1 progeny of D. insulare parasitizing two larval instars of P. xylostella

Figure 3 showed number of males to female ratio when second instars larvae were parasitized by larval parasitoid reared on two cabbage varieties (Asha and Golden acre). Figure 4 represents that number of males to female offspring ratio when third instars larvae were parasitized reared on two cabbage varieties.

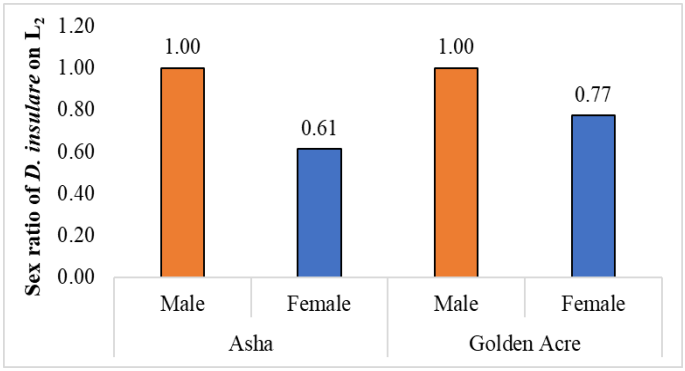

Figure 3: *Comparative sex ratio of D. insulare on second instars larvae of P. xylostella reared on two cabbage varieties.*

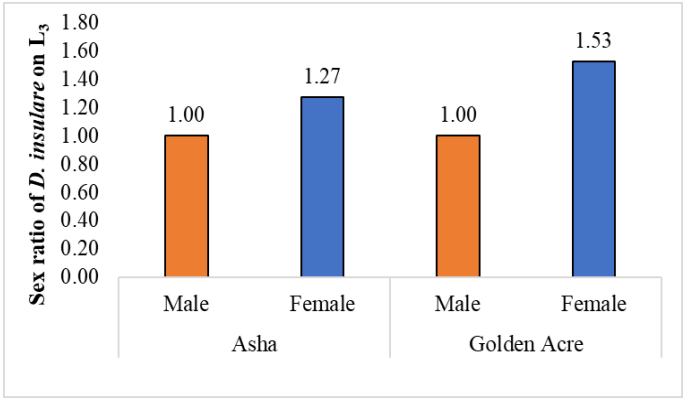

Figure 4: *Comparative sex ratio of D. insulare on third instars larvae of P. xylostella reared on two cabbage varieties.*

Host plants greatly affect insect pests which have positive impacts on biological control agents (Price, 1997). As parasitoid is an important natural enemies against insect herbivores in many cropping systems (Hawkins *et al.*, 1999), majority of studies have revealed that varieties reduce immature consumption rate and maximize their developmental period which is ideal for parasitoid than those one which reduces pest survival rate (Verkerk *et al.*, 1998). Moreover, phases of larval instars that is parasitized by parasitoids effect

on sex ratio. When second instars are parasitized more male formed and when third and fourth were parasitized more female are formed (Monnerat *et al*., 2002). During current study, the findings showed that maximum male offspring emerged when second instars larvae were parasitized while more female offspring were emerged when third instars larvae were parasitized by larval parasitoid reared on both varieties. While maximum female emerged when third instars larvae were parasitized reared on both varieties. The influence of selected varieties on fitness of the parasitoid is as important as parasitism rates to biological control. In this study when second instars larvae were used for parasitization against *D. insulare* on selected cabbage varieties, maximum parasitization (53.53 %) was assessed on Asha variety while minimum (44.95%) was observed on Golden acre. *D. insulare* were reared on third instar larvae of *P. xylostella* on two cabbage varieties. Maximum parasitization (61.17 %) was assessed on Asha while minimum (50.1 %) was observed on golden acre. Further it is concluded that parasitism level depends upon larval instars as our outcomes revealed that third instars were parasitized more as compared to second one.

SOV	DF	SS	MS	F	P
Analysis of variance for second larval instars					
Treatment	1	368.08	368.082	5.14	0.036
Error	18	1290	71.667		
Total	19	1658.08			
Variety	Mean	STDEV	S.E.		
Asha	53.53	8.16	2.58	A	
Golden acre	44.95	8.76	2.77	B	
Analysis of variance for third larval instars					
Treatment	1	612.72	612.725	9.35	0.0068
Error	18	1180	65.556		
Total	19	1792.72			
Variety	Mean	STDEV	S.E.		
Asha	61.17	8.76	2.77	A	
Golden Acre	50.1	7.38	2.33	B	

Figure 5: *Analysis of Variance for selected larval instars.*

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Novelty Statement

It is concluded that Asha variety is considered as best for cultivation as the parasitism level of D. insulare on Asha is maximum and developmental period of parasitized *P. xylostella* is minimum on Asha. So Asha variety is recommended to farmers for securing and enhancing the yield.

Author's Contribution

Riaz Hussain: Conducted research trials Data collection and overall management of the article.
Ata-ul-Mohsin: Conceived the idea and technical input at every step.
M. Farooq Nasir and Zahid Akram: Technical input during the study.
Muhammad Sajid Qureshi: Defined the methodology, SPSS analysis and reviewed the article.
Abdul Mannan Hamzah: Laboratory inputs and managed references.

Conflict of interest

The authors have declared no conflict of interest.

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