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## Laboratory Rearing of the Diamondback Moth *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) with Artificial Diet

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The diamondback moth *Plutella xylostella* (L.) (DBM) is a serious pest of crucifers worldwide. Both augmentative biological control and autocidal genetic control of DBM require an efficient mass-rearing system. We developed a wheat germ based artificial diet with kale leaf powder (ADK) and one without the powder (AD) and compared development of DBM on these diets, together with cabbage (Cab), its natural host. We used both laboratory and wild strains of DBM. ADK diet supported significantly higher percentage survival to pupation in laboratory and wild strains, than cabbage and AD. Survival rates within diets were relatively higher in laboratory strain than in wild strain. Larval period was significantly shorter with ADK than with cabbage in the laboratory strain but the reverse was observed in the wild strain. Pupal weights were similar between diets in wild strain, although females were significantly heavier than males. ADK diet able to support growth and development of DBM optimally, with no microbial contamination, thus suitable for mass production of DBM in laboratory.

### INTRODUCTION

The diamondback moth (DBM), *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), is a serious pest of cruciferous crops worldwide, including cabbage, cauliflower and collards (Harcourt, 1957; Miyata *et al.*, 1982; Abro *et al.*, 1992; Talekar and Shelton, 1993; Potting *et al.*, 1999; Reddy *et al.*, 2004; Shelton, 2004). DBM is believed to have originated in the Mediterranean region (Harcourt, 1957) which is also the place of origin of some of the important crucifer crops (Tsunoda, 1980). Outbreaks of DBM in Southeast Asia sometimes cause more than 90% crop loss (Verkerk and Wright, 1996). Although insecticides have often been used to control DBM, this pest has been developing resistance to many different insecticides (Liu *et al.*, 1982; Shelton *et al.*, 1993; Sarfraz and Keddie, 2005). Both augmentative biological control and autocidal genetic control of DBM require an efficient mass-rearing system (Carpenter and Bloem, 2002).

Conventional laboratory rearing of DBM uses live plants such as seedlings or leaves of cruciferous crops (Koshihara and Yamada, 1976; Omar and Mansor, 1993; Shirai, 2000). However, this practice requires more labor space and greatly increases the chances for microbial contamination (Carpenter and Bloem, 2002). To alleviate these shortcomings, artificial diets for mass rearing of DBM have been developed. However, most of them are not sufficient to sustain large DBM colonies under laboratory conditions (Berger, 1963; Biever and Boldt, 1971; Hisao and Hou, 1978; Shelton *et al.*, 1991; Carpenter and

Bloem, 2002). Although an artificial diet developed by Miyasono *et al.* (1992, 1996) can effectively mass-rear DBM, the diet contains either fresh cabbage leaves or fresh foliar parts of germinating radish which may make the dietary effect inconsistent due to variation in water content of fresh leaves in each preparation. This also makes the diet prone to microbial contamination.

Our objective was to develop effective rearing method for DBM using artificial diet. Towards this goal, we developed an artificial diet using plant powder by modifying a wheat germ based diet (Miyasono *et al.*, 1996) for rearing DBM. We compared the performance of two DBM strains, laboratory and wild, on our artificial diets with fresh cabbage.

### MATERIALS AND METHODS

#### Test insects

Two different strains of DBM, laboratory and wild strains were used in this study. Laboratory strain was initially collected in Hisayama, Fukuoka Japan, in 2004, and had been continuously reared on artificial diet with dried kale leaf powder (ADK) (Table 1). The laboratory strain was more than 80 generations when this study was initiated. For the wild strain, DBM larvae and pupae collected from a cabbage field in Hisayama in 2007 were reared on cabbage and their F1 used in this study.

#### Artificial diets

The wheat germ based artificial diet (Miyasono *et al.*, 1996) was modified by adding dried kale leaf powder instead of foliar part of germinating radish (Table 1). We also used propionic acid instead of formaldehyde solution because propionic acid is one of the most effective mold inhibitor (Ghosh *et al.*, 1996). The kale leaves, dried for two days after extraction of the juice, were provided by Q'SAI farm aojiru, Shimane, Japan. The dried leaves

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**Table 1.** Artificial larval diet ingredients used in the study

Ingredient	Amount	
	ADK	AD
Kidney bean	50 g	50 g
Choline Chloride	0.05	0.05
L- Cysteine	0.2	0.2
Cholesterol	0.25	0.25
Wesson salt mixture	0.5	0.5
Methyl <i>p</i> -hydroxybenzoate	1.42	1.42
Sucrose	2	2
L- Ascorbic acid	2	2
Casein	3.5	3.5
Dried yeast <sup>1</sup>	20	20
Wheat germ <sup>2</sup>	50	50
Dried kale powder	5	–
Agar	5	5
Linoleic acid	0.17 ml	0.17 ml
Propionic acid	0.8	0.8
Distilled water	340	340

<sup>1</sup> HY-GY B, Nisshin Seifun Co., Tokyo, Japan.

<sup>2</sup> Dried Yeast Ebios, Asahi Food & Health Co. Ltd., Tokyo, Japan.

were then ground and the resultant powder stored in a freezer until it was used. We prepared two artificial diets; a diet contained the dry kale powder (5 g/500 ml) (ADK) and one that did not contain the powder (AD).

### Rearing Methods

The two DBM strains were reared in separate oviposition cages. The pupae in a Petri dish (9 cm–diameter) were placed in a plexiglass oviposition cage (30×30×30 cm), one side of which had a slide door (13×13 cm) at the center, and two other sides of which were covered with fine nylon mesh, at 25 °C and 16L: 8D. After emergence, adults were allowed to mate and oviposit in the oviposition cage. They were provided with a piece of cotton saturated with 10% honey–water solution. Plastic cups (7.8 cm×5.5 cm) containing (50 mg) kale leaf powder were placed on the bottom of the rearing cage as an oviposition substrate for 24 h. Thereafter, the cups with newly laid eggs were collected and neonate larvae reared with cabbage, ADK and AD diets.

To compare immature development of DBM with artificial diets and cabbage leaves, 10 newly hatched (less than 6 h–old) first–instar larvae were transferred with a fine brush into a plastic cup (7.8 cm×5.5 cm) containing either 2 g of ADK diet, 2 g of AD diet, or 2 g of cabbage leaf. The cup was covered with tissue paper and then with a perforated plastic lid. The diets were renewed every two days. At the onset of pupation, the number of pupae per day per cup was recorded, and each pupa was weighed. Pupae were then individually kept in a 1.5 ml centrifugal tube. Adult emergence was daily checked, and the date of adult emergence and sex of emerging adults were recorded. The experiment was conducted at 25±1 °C and a photoperiod of L16: D8 condition. There were 10 replications (10 larvae per replicate) for each treatment.

### Statistical Analysis

Effects of diet and sex on survival of DBM were ana-

lyzed using a two–way analysis of variance (ANOVA), following arcsine transformation, and means were separated by use of Tukey–Kramer HSD test. Survival rates, developmental times and pupal weight of DBM between the laboratory and wild strains were compared by a two–sample t–test. Alpha was set at 0.05 for all analyses. Statistical analyses were done using the statistical software package, Stat View (SAS Institute, 1998).

## RESULTS

### Survival of the laboratory and wild strains of DBM on different diets

Survival of DBM was significantly affected by diet and strain, with a significant interaction between the two factors (Survival up to pupation: diet,  $F_{2,54}=21.0$ ;  $P<0.0001$ ; strain,  $F_{1,54}=25.9$ ;  $P<0.0001$ ; interaction between diet and strain,  $F_{2,54}=5.6$ ;  $P=0.006$ . Survival up to adult emergence: diet,  $F_{2,54}=7.6$ ;  $P=0.001$ ; strain,  $F_{1,54}=7.9$ ;  $P=0.007$ ; interaction between diet and strain,  $F_{2,54}=5.0$ ;  $P=0.010$ ). ADK supported significantly higher percentage survival to pupation of both laboratory and wild strains than cabbage. Similarly, survival to adult emergence followed the same trend, although the difference in emergence did not significantly differ between cabbage and ADK in the laboratory strain (Table 2). ADK and AD supported similar survival rates of the laboratory strain of DBM to pupation and adult emergence. However, survival rates to pupation and adult emergence were significantly higher with ADK than with AD in the wild strain.

Generally survival rates of DBM within diets were relatively higher in the laboratory strain than the wild strain. However, with regards to survival to pupation these differences were only significant with cabbage and AD diets while percentage emergence was only significant with AD diet ( $p<0.05$ ) (Table 2).

**Table 2.** Survival of DBM reared on Cab, ADK and AD diets

Diet	No. of insects examined	% survival from first instar up to	
		Pupation	Adult emergence
Wild strain			
Cab	100	61a A	58a A
ADK	100	96b A	85b A
AD	100	49a A	42a A
Laboratory strain			
Cab	100	83a B	69a A
ADK	100	99b A	84a A
AD	100	87ab B	77a B

The values with different small letters in the same columns within strain were significantly different by Tukey–Kramer HSD test ( $P<0.05$ ). The values with different capital letters in the same column with corresponding diets between strains were significantly different by t–test ( $P<0.05$ ).

### Developmental time

Diet significantly affected larval, pupal and total developmental periods in the wild DBM strain (Larval period,  $F_{2,179}=61.0$ ;  $P<0.0001$ ; pupal period,  $F_{2,179}=14.0$ ;

$P<0.0001$ ; total period,  $F_{2,179}=68.7$ ;  $P<0.0001$ ) while sex did not (Larval period,  $F_{2,179}=0.1$ ;  $P=0.78$ ; pupal period,  $F_{2,179}=3.9$ ;  $P=0.05$ ; total period,  $F_{2,179}=0.9$ ;  $P=0.34$ ), with no interaction between diet and sex (Larval period,  $F_{2,179}=0.5$ ;  $P=0.59$ ; pupal period,  $F_{2,179}=0.1$ ;  $P=0.95$ ; total period,  $F_{2,179}=0.4$ ;  $P=0.66$ ). Larval period was significantly shorter with ADK than with cabbage in the laboratory strain but the reverse was observed with the wild strain in both sexes (Tables 3 and 4). However, there was no difference between ADK and AD in the laboratory strain in both sexes but the larval period was significantly shorter in the former in the wild strain. Similarly, the pupal and total developmental periods were significantly shorter with cabbage than ADK and AD in wild strain while they were not different with the three diets in the laboratory strain among the males. However, the pupal period was significantly shorter with cabbage than ADK and AD in both laboratory and wild strains among the

**Table 3.** Developmental period (days) of male DBM fed on Cab, ADK and AD diets

Diet	No. of insects examined	Larval period	Pupal period	Total period
		(mean $\pm$ SEM)	(mean $\pm$ SEM)	(mean $\pm$ SEM)
Wild strain				
Cab	34	12.2 $\pm$ 0.2a A	4.4 $\pm$ 0.2a A	16.6 $\pm$ 0.3a A
ADK	44	14.6 $\pm$ 0.2b A	5.0 $\pm$ 0.1b A	19.6 $\pm$ 0.3b A
AD	22	16.4 $\pm$ 0.5c A	5.1 $\pm$ 0.2b A	21.6 $\pm$ 0.5c A
Laboratory strain				
Cab	37	10.0 $\pm$ 0.3b B	4.1 $\pm$ 0.1a B	14.1 $\pm$ 0.3a B
ADK	38	9.4 $\pm$ 0.1a B	4.2 $\pm$ 0.1a B	13.5 $\pm$ 0.2a B
AD	38	9.6 $\pm$ 0.2ab B	4.1 $\pm$ 0.1a B	13.7 $\pm$ 0.2a B

The values with different small letters in the same columns within strain were significantly different by Tukey–Kramer HSD test ( $P<0.05$ ). The values with different capital letters in the same column with corresponding diets between strains were significantly different by t-test ( $P<0.05$ ).

**Table 4.** Developmental period (days) of female DBM fed on Cab, ADK and AD diets

Diet	No. of insects examined	Larval period	Pupal period	Total period
		(mean $\pm$ SEM)	(mean $\pm$ SEM)	(mean $\pm$ SEM)
Wild strain				
Cab	24	12.3 $\pm$ 0.3a A	4.2 $\pm$ 0.1a A	16.5 $\pm$ 0.3a A
ADK	41	14.2 $\pm$ 0.2b A	4.7 $\pm$ 0.1b A	18.9 $\pm$ 0.2b A
AD	20	16.6 $\pm$ 0.8c A	4.9 $\pm$ 0.2b A	21.4 $\pm$ 0.8c A
Laboratory strain				
Cab	32	9.6 $\pm$ 0.2b B	3.6 $\pm$ 0.1a B	13.2 $\pm$ 0.3a B
ADK	46	9.2 $\pm$ 0.1a B	4.0 $\pm$ 0.0b B	13.3 $\pm$ 0.1a B
AD	39	9.5 $\pm$ 0.1ab B	4.0 $\pm$ 0.1b B	13.4 $\pm$ 0.2a B

The values with different small letters in the same columns within strain were significantly different by Tukey–Kramer HSD test ( $P<0.05$ ). The values with different capital letters in the same column with corresponding diets between strains were significantly different by t-test ( $P<0.05$ ).

females. Total developmental period followed the same trend among the females but the difference was only significant in the wild strain. Larval, pupal and total developmental periods were significantly shorter with all the three diets in the laboratory strain than in the wild strain in both sexes.

### Pupal weight

Pupal weights of the wild strain were affected by sex and not diet, with a significant interaction between diet and sex (sex,  $F_{1,179}=51.73$ ;  $df=$ ;  $P<0.0001$ ; diet,  $F_{2,179}=1.10$ ;  $P=0.33$ ; interaction  $F_{2,179}=3.24$ ;  $P=0.04$ ). These weights were however not significantly different among the three diets in both sexes, although females were significantly heavier than males (Table 5). Conversely, pupal weights were significantly affected by both diet and sex, with no interaction between the two factors among the laboratory strain (diet,  $F_{2,224}=62.4$ ;  $P<0.0001$ ; sex,  $F_{1,224}=103.7$ ;  $P<0.0001$ ; interaction  $F_{2,224}=1.38$ ;  $P=0.25$ ). Pupal weights were not significantly different between diets, for both males and females, in the wild strain. However, they were significantly lighter with cabbage than with ADK and AD in the laboratory strain and were significantly heavier with all diets in the laboratory strain than in the wild strain for both sexes (cabbage: male,  $P=0.03$ , female,  $P<0.0001$ ; ADK: male,  $P<0.0001$ , female,  $P<0.0001$ ; AD: male,  $P<0.0001$ , female,  $P<0.0001$ ) (Table 5).

**Table 5.** Pupal weight of DBM fed on Cab, ADK and AD diets

Diet	Pupal weight (mg) <sup>b</sup>			
	Male		Female	
	N <sup>a</sup>	Mean $\pm$ SEM	N <sup>a</sup>	Mean $\pm$ SEM
Wild strain				
Cab	34	4.5 $\pm$ 0.1 a A	24	5.0 $\pm$ 0.1 a A
ADK	44	4.1 $\pm$ 0.1 a A	41	5.2 $\pm$ 0.1 a A
AD	22	4.2 $\pm$ 0.2 a A	20	5.6 $\pm$ 0.4 a A
Laboratory strain				
Cab	37	4.9 $\pm$ 0.2 b B	32	6.0 $\pm$ 0.2 b B
ADK	38	6.4 $\pm$ 0.2 a B	46	7.9 $\pm$ 0.2 a B
AD	38	6.4 $\pm$ 0.2 a B	39	7.9 $\pm$ 0.2 a B

<sup>a</sup> The numbers of insects examined.

<sup>b</sup> The values with different small letters in the same columns within strain were significantly different by Tukey–Kramer HSD test ( $P<0.05$ ). The values with different capital letters in the same column with corresponding diets between strains were significantly different by t-test ( $P<0.05$ ).

## DISCUSSION

An appropriate artificial diet is one that satisfies the chemical, physical and nutritional requirements of the insects (Friend, 1958). This is important for maintenance of laboratory colonies of insects which are subsequently used for different studies. There are therefore continuous efforts to both develop new and improve the already existing diets that not only ensure higher survival rates of the target insects, but that are cost-effective and reduce/eliminate the problems of contamination.

A number of diets, based on live parts of natural host



plants or artificial have been developed for mass rearing of DBM, with a number of inherent limitations (Hou, 1986). In the current studies, ADK diet not only supported significantly higher survival rates to pupation of DBM than cabbage but also removed the problem of diet contamination. Survival rates of DBM on most available diets have tended to be below 80%. We therefore consider the rates observed with ADK (>95%) in this study is the highest among artificial diet ever obtained. The fact that survival rates of DBM on this diet was similarly higher than cabbage in both laboratory and wild strains indicate that the reason for exemplary performance of the diet is not as a result of adaptation of the insect to the diet. This implies that either the diet is nutritionally more superior to cabbage or that the conversion of the diet into nutritionally utilizable units by the insect is more efficient. Moreover, microbial contamination which is a major problem in insect rearing facilities (Sikowski *et al.*, 1994) was not observed with ADK diet, while this proved to be one of the reasons for lower survival of DBM with fresh cabbage.

Developmental times were relatively longer with the artificial diets than cabbage in the wild strain, supporting the generally held view that larval development is often slower with artificial diets (Shelton *et al.*, 1991). The reverse was however observed with the laboratory strain. Similarly, we observed that the laboratory strain had generally higher survival rates and shorter developmental times than the wild strain, on all diets. These results are in agreement with those of Slansky and Wheeler (1992) who found that field strain larvae of *Anticarsia gemmatilis* (Hübner) exhibited longer developmental periods in artificial diets than a laboratory colony. Similarly, the laboratory-adapted DBM colony performed better on most artificial diets when compared to wild strains (Carpenter and Bloem, 2002).

Insect strain and degree of laboratory adaptation can influence the ability of an insect to develop on different diets, including its natural host plant (Carpenter and Bloem, 2002). Insects must be adapted to artificial rearing methods for successful mass rearing, because the goal of mass rearing program is to produce the maximum number of target insects as rapidly (Finney and Fisher, 1964) as possible without affecting quality (Chambers, 1977). From our studies, it is evident that DBM, in the laboratory strain having been continuously reared on the ADK diet for over 80 generations is well adapted to this diet. Its rather poor performance on cabbage, one of its host plants, and comparable performance between the two strains goes further to indicate suitability of ADK diet for mass production of DBM.

In conclusion, compared to the wild strain, laboratory strain had higher survival rate, produced heavier pupae and required shorter developmental times when reared not only on artificial diet but also on its natural food, cabbage. Comparison with cabbage leaves, the ADK diet was able to support growth and development of DBM optimally with no microbial contaminations. Using this diet DBM would be effectively mass produced with no microbial contaminations in the laboratory.

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## REFERENCES

- Abro, G. H., R. A. Soomro and T. S. Syed 1992 Biology and behavior of diamondback moth, *Plutella xylostella* (L.). *Pakistan. J. Zool.*, **24**: 7–10
- Berger, R. S. 1963 Laboratory techniques for rearing *Heliothis* species on artificial medium. U. S. Department of Agriculture ARS (Report) Series 33–84
- Biever, K. D. and P. E. Boldt 1971 Continuous laboratory rearing of the diamondback moth and related biological data. *Ann. Entomol. Soc. Am.*, **64**: 651–655
- Carpenter, J. E. and S. Bloem 2002 Interaction between insect strain and artificial diet in diamondback moth development and reproduction. *Entomol. Exp. Appl.*, **102**: 283–294
- Chambers, D. L. 1977 Quality Control in Mass Rearing. *Ann. Rev. Entomol.*, **22**: 289–308
- Finney, G. L. and T. W. Fisher 1964 Culture of entomophagous insects and their hosts. pp. 328–355. In P. DeBach and E. T. Schlinger [Eds], Biological control of insect pests and weeds. Chapman and Hall, London
- Friend, W. G. 1958 Nutritional requirements of phytophagous insects. *Ann. Rev. Ent.*, **3**: 57–74
- Ghosh, M. K., A. Chhabra, P. P. Atreja, and R. C. Chopra 1996 Effect of treating with propionic acid, sodium bisulfite and sodium hydroxide on the biosynthesis of aflatoxin on groundnut cake. *Anim. Feed Sci. Technol.*, **60**: 43–49
- Harcourt, D. G. 1957 Biology of the diamondback moth, *Plutella maculipennis* (Curt.) (Lepidoptera: Plutellidae), in eastern Ontario, Life-history, behaviour, and host relationship. *Can. Entomol.*, **89**: 554–564
- Hou, R. F. 1986 Mass rearing of diamondback moth. pp. 89–95. In N. S. Talekar and T. D. Griggs [Eds], Diamondback moth management. Proceeding of 1<sup>st</sup> international workshop, Asian vegetable research and development center, Shunhua. Taiwan
- Hsiao, J. H. and R. F. Hou 1978 Artificial rearing of the diamondback moth and, *Plutella xylostella* (L.), on a semi-synthetic diet. *Bull. Inst. Zool. Acad. Sin.*, **17**: 97–102
- Koshihara, T. and H. Yamada 1976 A simple mass-rearing, technique of the diamondback moth, *Plutella xylostella* (L.), on germinating rape seeds. *Jpn. J. Appl. Entomol. Zool.*, **24**: 6–12 (In Japanese with English summary)
- Liu, M. Y., Y. J. Tseng and C. N. Sun 1982 Insecticide resistance in diamondback moth. *J. Econ. Entomol.*, **75**: 153–155
- Miyasono, M., K. Ohba, M. Masuko and M. Doteuchi 1996 Improvement of aseptic rearing of diamondback moth, *Plutella xylostella* (L.) with artificial diet, *Jpn. J. Appl. Entomol. Zool.*, **40**: 302–305 (in Japanese with English summary)
- Miyasono, M., M. Yamamoto, K. Ohba, T. Koshihara, T. Ishiguro and Y. Hayashi 1992 Mass rearing of the diamondback moth, *Plutella xylostella* (L.) with an improved artificial diet. *Jpn. J. Appl. Entomol. Zool.*, **36**: 193–196 (in Japanese with English summary)
- Miyata, T., H. Kawai, and T. Saito 1982 Insecticide resistance in the diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae). *Appl. Entomol. Zool.*, **17**: 539–542
- Omar, D. and M. Mansor 1993 Effect of sub sterilization doses of radiation on the biology of diamondback moth. pp. 3–9. In

- Radiation Induced F1 Sterility in Lepidoptera for Area-Wide Control. IAEA STI/PUB/929. Vienna, Australia
- Potting, R. P. J., G. M. Poppy and T. H. Schuler 1999 The role of volatiles from cruciferous plants and pre-flight experience in the foraging behavior of the specialist parasitoid *Cotesia plutellae*. *Entomol. Exp. Appl.*, **93**: 87–95
- Reddy, G. V. P., E. Tabone and M. T. Smith 2004 Mediation of host selection and oviposition behavior in the diamondback moth *Plutella xylostella* and its predator *Chrysoperla carnea* by chemical cues from cole crops. *Biol. Cont.*, **29**: 270–277
- Sarfraz, M. and B. A. Keddie 2005 Conserving the efficacy of insecticides against *Plutella xylostella* (L.) (Lepidoptera: Plutellidae). *J. Appl. Entomol.*, **129**: 149–157
- SAS Institute. 1998. Stat View 5. O. J. SAS Institute Inc., Cary, NC
- Shelton, A. M., J. L. Robertson, J. D. Tang, C. J. Pérez, S. D. Eigenbrode, H. K. Preisler, W. T. Wilsey and R. J. Cooley 1993 Resistance of diamondback moth (Lepidoptera, Plutellidae) to *Bacillus thuringiensis* subspecies in the field. *J. Econ. Entomol.*, **86**: 697–705
- Shelton, A. M., R. J. Cooley, M. K. Kroening, W. T. Wilsey and S. D. Eigenbrode 1991 Comparative analysis of two rearing procedures for diamondback moth (Lepidoptera: Plutellidae). *J. Entomol. Sci.*, **26**: 17–26
- Shelton, A.M. 2004. Management of the diamondback moth: déjà vu all over again? pp. 3–8. In N. M. Endersby and P. M. Ridland [Eds.], The management of diamondback moth and other crucifer pests proceedings of the fourth international workshop, 26–29 November 2001. Department of natural resources and environment, Melbourne. Australia
- Shirai, Y. 2000 Temperature tolerance of the diamondback moth, *Plutella xylostella* (Lepidoptera: Yponomeutidae) in tropical and temperate regions of Asia. *Bull. Entomol. Res.*, **90**: 357–364
- Sikorowski, P. P. and A. M. Lawrence 1994 Microbial contamination and insect rearing. *Am. Entomol.*, **40**: 240–253
- Slansky, F. Jr. and G. S. Wheeler 1992 Feeding and growth responses of laboratory and field strains of velvet bean caterpillars (Lepidoptera: Noctuidae) to food nutrient level and allelochemicals. *J. Econ. Entomol.*, **85**: 1717–1730
- Talekar, N. S. and A. M. Shelton 1993 Biology, ecology, and management of the diamondback moth. *Annu. Rev. Entomol.*, **38**: 275–301
- Tsunoda, S. 1980 Eco-physiology of wild and cultivated forms in Brassica and allied genera. In; Brassica crops and wild allies, Biology and breeding, (Eds) S. Tsunoda, K. Hinata and C. Gomez-Campo, PP – 109 Scientific Societies Press, 20. Tokyo, Japan. pp. 354
- Verkerk, R. H. J. and D. J. Wright 1997 Field-based studies with the diamondback moth tritrophic system in Cameron Highlands of Malaysia: implications for pest management. *Int. J. Pest Manag.*, **43**: 27–33