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POPULATION ECOLOGY OF DIAMONDBACK MOTH, *Plutella*

xylostella L. (LEPIDOPTERA: PLUTELLIDAE) AND ITS

PARASITOIDS IN ETHIOPIA

BY

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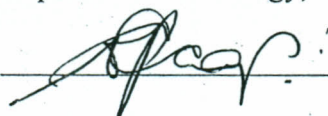
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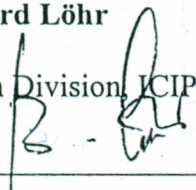


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DEDICATION

To my parents, Degitu Ashenefer and late Ayalew Chekol

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ABSTRACT

The objective of this study was to generate information on the ecology of Diamondback moth (DBM) (*Plutella xylostella* L.) and the parasitoids associated with it with emphasis on parasitism for the purpose of strengthening biocontrol in Integrated Pest Management (IPM) program. Prior to this study no or little information was available in Ethiopia on DBM despite its importance in brassica production. A series of studies were conducted between 2001 and 2002, including a survey covering the most important crucifer growing areas, within field distribution of larvae and pupae, yield loss assessment, suitability of wild and cultivated crucifers for DBM development and population dynamics (spatial and temporal) of DBM and its parasitoids.

Survey conducted in thirteen brassica producing areas showed that DBM density and level of damage were higher in the vegetable production system of central rift valley regions and Arsi high land. Seven hymenopteran parasitoid species from five families were recorded. Of these, *Oomyzus sokolowskii* (Hymenoptera: Eulophidae), *Diadegma* sp. (Hymenoptera: Ichneumonidae) and *Apanteles* sp. (Hymenoptera: Braconidae) were the dominant ones accounting for more than 90 per cent of the parasitoid complex. Level of parasitism by the indigenous parasitoid species was low in major brassica producing areas of the country, the Arsi highland and the rift valley areas.

Yield loss was assessed using natural infestation in a Randomized Complete Block Design with nine treatments. Treatments were pesticides,

Bacillus thuringiensis (*Bt*) and karate, applied at different growth stages of head cabbage, seedling, pre-heading, and heading. Yield loss ranged between 36.1 and 85.2 percent corresponding to 12 to 27.6 tons per hectare.

The developmental period of DBM on four cultivated brassica crops and one wild crucifer, *Erucastrum arabicum* Fisch. and Mey, was studied for two generations in the laboratory in ambient conditions (Temperature ranged between 23 and 30°C). Life table statistics including developmental periods indicated that cabbage was the most suitable host and the wild crucifer was as suitable as some of the cultivated crucifers.

Within field distribution of larvae and pupae was contagious in cabbage and kale. In Ethiopian mustard, only the young larvae showed contagious distribution. Spatial analysis showed that DBM is aggregated in field of its host plants. The influence of weather variables on aggregation varied with locations. In highland areas of Arsi, maximum temperature showed a significant influence ($R^2 = 0.407$, $P < 0.05$). In the lowland areas of Wonji, the aggregation index was significantly influenced by rainfall ($R^2 = 0.603$, $P < 0.05$).

Studies on temporal dynamics showed two to three generations per head cabbage growing season in the highland brassica production area and three to five generations in the lowland. Population fluctuated between 0 to 15.7 and 0 to 1.7 insects per plant in November/December and April planted trials, respectively, at the highland site; and between 0 to 3.2 and 0 to 8.5 at the lowland site. Lower DBM density was associated with higher rainfall and

lower maximum temperature in the highland experimental site. At both locations, three species of parasitoids were recorded. These were *Oomyzus sokolowskii*, *Diadegma* sp., and *Apanteles* sp. Average overall apparent (and potential) parasitism was 13.6 (14.0) % and 46.3 (51.7) % at the highland site; and 4.6 (4.6) % and 25.6 (26.3) % at the lowland site in the November/December and April planted experiments, respectively. *O. sokolowskii* was the dominant species at the low land site and *Diadegma* sp. at the highland site. One or more factors including host density, season of planting, location, and environmental variables, showed significant influence ($P < 0.5$) on parasitism level by the different parasitoid species.

Biocontrol of DBM in Ethiopia needs to target areas of major brassica production with higher level of DBM damage and low parasitism: the Arsi highland and the central rift valley region. In addition, currently used pesticides in these areas need to be replaced with safer alternatives with minimal effect on bio-control agents such as the use of neem based products and microbial pesticides such as *Bt*.

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CHAPTER 1

1.0 GENERAL INTRODUCTION AND LITERATURE REVIEW

1.1 INTRODUCTION

Crucifers, especially head cabbage and kale are among the major vegetables produced both by private farmers and the state farms in Ethiopia (Dessalegne *et al.*, 1994). Cabbage is the second most important vegetable crop next to red pepper (*Capsicum* spp.) both in the level of production and land allotment in the country (MOA, 2002). Land allotted for Ethiopian cabbage alone in the main rainy season (Meher) is estimated to be 14, 528 ha with a yield estimate of 143, 680 tons (MOA, 2002).

A number of insect species including the Diamondback moth *Plutella xylostella* L., cabbage aphid *Brevicoryne brassicae* (L.), mustard aphid *Lipaphis erysimi* (Kaltenbach), flea beetles *Phyllotreta* spp. and cabbage leaf miner *Chromatomyia horticola* (Goureau) inflict damage on brassica crops in Ethiopia (Abate and Ayalew, 1994). Of these, the Diamondback moth (DBM) is the most important both in distribution and severity of damage as it is in other parts of the world, especially in the tropics (Carl, 1992; Talekar and Shelton, 1993).

Although pesticide use in pest management is minimal in crop production in Ethiopia in comparison to several African countries (Abate *et al.*, 2000), vegetable growers depend on pesticides because of the commercial nature of vegetable production and quality requirement of the

markets (Carl, 1992). A large amount of pesticide is applied by growers against DBM and cabbage aphids in Ethiopia. Although satisfactory control of cabbage aphids is achieved through use of insecticides, DBM has become increasingly difficult to control by a range of available pesticides in the market just like in other parts of the world (Sun *et al.*, 1978; Liu *et al.*, 1981). Several possible causes for the increasing problem of DBM are known and are associated with pesticide use including development of pesticide resistance, destruction of natural enemies by insecticides, short generation cycle of the insect (Carl, 1992; Talekar and Shelton, 1993) among others. The ever-increasing difficulties associated with pesticide control of DBM led to the development of a biological control project in an IPM program (Carl, 1992; Talekar and Shelton, 1993).

Success on biological control of DBM has been reported by several workers (Alam, 1992; Mustata, 1992; Ooi, 1992; Talekar *et al.*, 1992). Carl (1992) equated the success of biocontrol of DBM in Cape Verde, to the classical example of successful biocontrol of the mealy bug, *Phenacoccus manihoti* Matile-Ferrero, that threatened the production of cassava in Africa. It has been a belief that DBM and other pests attacking crucifers should be among the first vegetable pests against which biological and integrated control should be applied in Africa (Carl, 1992) and other parts of the world (Talekar and Shelton, 1993). Knowledge on the complex of naturally occurring parasitoids and their efficacy in

regulating pest population is fundamental to the development of biocontrol program through conservation of the existing species or introduction. Nothing or little was known about the ecology of DBM, its population dynamics and the parasitoids complex associated with it in Ethiopia. No single species of natural enemies of DBM was documented in Ethiopia (Abate, 1991). Knowledge on the ecology of the pest such as temporal and spatial dynamics, among others, forms the basis for designing appropriate management in an Integrated Pest Management program. In addition, an inventory of the indigenous natural enemies and assessing their role in regulating DBM population would help to determine the biological control option that need to be followed.

1.2 LITERATURE REVIEW

1.2.1 Biology

The biology of DBM has been intensively studied in different parts of the world; Canada (Harcourt, 1957), Malaysia (Ho, 1965), India (Abraham and Padmanabhan, 1968), China (Lee, 1968). The time taken to complete its life cycle can vary from 9 to 10 days under favorable condition to as long as 110 days under unfavorable condition (Ko and Fang, 1979). According to Bhalla and Dubey (1986), a daily mean temperature ranging from 5 to 12° C and daily maximum temperature between 21° and 36° C favored the multiplication of the insect, resulting to a short generation cycle.

Most adults emerge during the first eight hours of photophase (Pivnic *et al.*, 1990). The moths mate at dusk on the day of emergence. The copulating pairs face opposite direction and hang downwards. Mating lasts 1-2 hours and females mate only once (Jayarathnam, 1977). Eggs are laid on different parts of the plant including leaf, stem and petiole. The majority of eggs are, however, laid on the leaf in the proportion of 3:2 on the upper and lower leaf surface (Chelliah and Srinivasan, 1986; Talekar and Shelton, 1993). The oviposition period lasts 3 to 10 days (Chelliah and Srinivasan, 1986). The peak oviposition occurs on the first day of emergence. Fecundity as low as 11 eggs and as high as 203 eggs per female has been reported (Chelliah and Srinivasan, 1986; Talekar and Shelton, 1993). Incubation period lasts 3 to 6 days depending on temperature (Abraham and Padmanabhan, 1968).

There are four larval instars. Each instar has different duration depending on the weather conditions. For example, Chelliah and Srinivasan (1986) reported a duration of 3, 4, and 5 days for the first instar larvae in hot, rainy and cold seasons, respectively. Total larval period ranges from 14 to 21 days (Abraham and Padmanabhan, 1968). Upon hatching neonate larvae start feeding on foliage. The first instar larvae mine in the spongy mesophyll tissues, whereas older larvae feed from lower leaf surface and usually consume all the tissues except the wax layer on the upper surface, thus creating a window in the leaf (Talekar and Shelton, 1993). Upon completion of feeding, the mature caterpillars form a

gauzy, loosely spun cocoon for pupation. The pupal color is yellowish-green first then changes to brownish. It attains a dark-brownish color by the time of adult emergence. The pupal period can vary from 4 to 15 days depending on temperature (Talekar and Shelton, 1993). Adults emerge in the evening and rarely in the morning hours (Chelliah and Srinivasan, 1986).

1.2.2 Ecology

The Diamondback moth is found almost everywhere in the world where its hosts grow. Its ability to fly long distance is one of the major factors for its wide distribution. Reports by Talekar and Shelton (1993) show that DBM can remain in a continuous flight for several days and cover distance of 1000 km per day. Wind is reported as a major factor in DBM migration from one area to the other in temperate areas although the mechanism of its migration is not clear (Honda, 1992; Talekar and Shelton, 1993). Honda (1992) suggested that density of source population, meteorological conditions when the adults take off, the physiological conditions of the migrants and the temperature of the upper air in which they migrate affect DBM migration. Seedlings contaminated with DBM have also been reported as sources of infestation in the United States (Talekar and Shelton, 1993).

The presence of crucifers throughout the year in the tropics and subtropics allows the pest to be active throughout the season and hence the

simultaneous occurrence of all stages of the insect. The mechanism of its occurrence in temperate areas where crucifers are not found year round is not clearly known (Talekar and Shelton, 1993).

The present knowledge regarding its survival in the temperate region is that the moths do not over winter in many temperate areas where it is a pest and that migration occurs by the adult moths moving on wind currents or by all stages arriving on contaminated seedlings (Talekar and Shelton, 1993).

1.2.3 Economic importance

The Diamondback moth is a serious and important pest of crucifers in many parts of the world, particularly in the tropics (Lim, 1992). Most of the chemical control in brassica production is directed towards DBM control. A conservative estimate of its control cost globally is USD 1 billion annually (Carl, 1992). Although such a large amount of money is spent for the control of the pest, the level of control to offset the expense is low because of the physiological peculiarity of the insect to acquire resistance to insecticides very quickly. As a result, pesticide abuse is very common practice, especially in tropical areas in a desperate attempt to achieve reasonable control. This practice rather increases the magnitude of DBM problem through decimating the associated natural enemies which can play a significant role in the natural control of the pest. The danger this

poses to the environment and the consumers is considerable too. Carl (1992) indicates that over 60 % of the market value of the cabbage crop is spent for the purchase of pesticides in Asian countries without considering cost of inputs including pesticide application.

1.2.4 Host range of DBM

Diamondback moth feeds on cultivated and wild plants belonging to the family Cruciferae (Talekar and Shelton, 1993). The cultivated species include cabbage (*Brassica oleracea* L. var. *capitata* L.), cauliflower (*B. oleracea* L. var. *botrytis* L.), broccoli (*B. oleracea* L. var. *italica* Plenck), radish (*Raphanus sativus* L.), turnip (*B. rapa* L. *pekinensis* (Lour)), Brussels sprouts (*B. oleracea* L. var. *gemmifera* Zenker), Chinese cabbage (*B. rapa* L. *pekinensis* (Lour)), kohlrabi (*B. oleracea* L. var. *gongylodes* L.), mustard (*B. juncea* L.), rapeseed (*B. napus* L.), collard (*B. oleracea* var. *acephala* DC., pro parte), watercress (*Nasturtium officinale* L.) and kale (*B. oleracea* L. var. *alboglabra* (L. H. Bailey) Musil (Talekar and Shelton, 1993). One or more uncultivated (weed) species in 27 different genera are also recorded from different parts of the world as hosts that sustain feeding and reproduction of DBM (Talekar and Shelton, 1993).

The association of DBM to plant species of crucifers is due to the presence of one or more glucosinolates, singrin, sinalbin and glucoheirolin, which act as specific feeding stimulant (Talekar and

Shelton, 1993). Works of Gupta and Thorsteinson (1960) suggested that occurrence of feeding inhibitors as well as the absence of essential feeding stimulants exclude some botanical species in crucifers from the food plant range. On the other hand, the existence in plants of effective feeding stimulants other than mustard oil, glucosides, without feeding inhibitors, have put some plant species of leguminosea in the food range of the pest. For example, Field pea, *Pisum sativum* L. (Fam. Leguminosae) was shown to be sufficiently palatable, nutritious and free of toxicants to support successive generations of *Plutella xylostella* larvae in the laboratory (Gupta and Thorsteinson, 1960). Löhr (2001) reported serious damage inflicted by DBM on sugar snap pea in Kenya under field condition and Löhr and Gathu (2002) showed that in only few generation of selection, a DBM strain can be produced that performs comparably well on crucifer and pea.

1.2.5 Control of DBM

The control practices against DBM range from cultural control to the recent and novel techniques which include the use of pheromones and growth regulators (Talekar and Shelton, 1993; Robert *et. al.*, 1996). The methods followed vary from area to area depending on the production scale and the availability of the control measures. In small scale farming of several tropical and subtropical areas, insecticides are the main control

tactic because of lack of proven alternatives and the availability of relatively cheap insecticides (Talekar and Shelton, 1993).

Attempt to use alternative control methods has been and is being made only in few countries in Asia. These alternatives include parasitoid introductions for biocontrol and sterile insect programs (Talekar and Shelton, 1993). On the other hand, in developed nations where crucifers are produced in large scale such as North America, Europe and New Zealand, insecticides form an integrated component in DBM IPM, and pesticide application is threshold based (Talekar and Shelton, 1993). Management options of DBM which include cultural, biological, insecticidal and IPM are highlighted below.

1.2.5.1 Cultural control

This method has been given greater attention in recent years because of the inadequacy of the available pesticides to control the pest. Some of the important cultural practices include inter-cropping, irrigation and trap-cropping (Talekar *et al.*, 1986; Srinivasan and Krishna Moorthy, 1992).

1.2.5.1.1 Inter-cropping

Incidence of DBM is reported to be low in cabbage inter-cropped with tomato, dill, garlic, safflower, oat and barley (Talekar *et al.*, 1986). Although it is a common practice among vegetable growers in several

developing countries, it is basically aimed to diversify production and to avert risk from natural factors instead of reduction of pest incidence.

1.2.5.1.2 Irrigation

Sprinkler irrigation has been reported to reduce DBM injury on crucifers. The physical disruption of flying activity, oviposition and to some extent wash-off of larvae and adults were reported as the major causes for DBM injury reduction using sprinkler irrigation (Talekar *et al.*, 1986).

1.2.5.1.3 Trap-cropping

Planting Indian mustard *Brassica juncea* (L.) as a trap crop in cabbage field (paired mustard rows at either end of 25 cabbage rows) was reported to reduce DBM and other cabbage pests significantly (Srinivasan and Krishna Moorthy, 1992).

1.2.5.2 Biological control

The problem of resistance development to insecticides and residues in harvestable products necessitate the promotion of biocontrol program in several countries. A large number of natural enemies including parasitoids, predators, and microbials attack DBM. Of these, parasitoids have been studied extensively and helped in the development of DBM biocontrol programs. Larval parasitoids belonging to two genera, *Diadegma*

(Hymenoptera: Ichneumonidae) and *Cotesia* (Hymenoptera: Braconidae) are the most predominant and effective. Success stories through the introduction and conservation of these parasitoids are reviewed by Talekar and Shelton (1993).

1.2.5.3 Use of insecticides in the control of DBM

Insecticidal control has remained the major method of DBM control despite resistance development and hence failure to achieve satisfactory control. Talekar and Shelton (1993) reviewed insecticides use pattern which vary over geographic locations and time. Although large number of chemical insecticides have been used against DBM, none has been found to remain effective for a quite longer period including the biopesticide, *Bacillus thuringiensis*.

1.2.5.4 Integrated Pest Management

The availability of several management options including cultural and biological coupled with problems associated with insecticide use in DBM management creates a good opportunity for promoting an IPM approach for cabbage and other brassicas. In small-scale agriculture, this requires a coordinated effort among brassica growers. For example, biocontrol through either conservation or introduction of parasitoids in an area need to be augmented by the use of safer pesticides with little or no

harm to the parasitoids as was observed in several Asian countries (Talekar and Shelton, 1993).

1.2.6 DBM parasitoids

1.2.6.1 Primary parasitoids

Over 90 parasitoid species are reported to attack Diamondback moth of which the larval parasitoids are the most predominant and effective (Talekar and Shelton, 1993). The most important families and representative parasitoid species based on stage of DBM they attack are presented in Table 1.1.

Table 1.1. Important Hymenopterous families and representative parasitoid species attacking the different stages of DBM

Stage	Family	Representative species
Egg	Trichogrammatidae	<i>Trichogramma spp</i>
Larva	Ichneumonidae	<i>Diadegma spp</i>
	Braconidae	<i>Cotesia plutellae</i> Kurdjumov
Larval-Pupal	Eulophidae	<i>Oomyzus sokolowskii</i> Kurdjumov
Pupal	Ichneumonidae	<i>Diadromus collaris</i> (Gravenhorst)

The range of parasitoid species associated with DBM varies depending on the diversity of brassica species. Although DBM is believed to have originated from Europe (Talekar and Shelton, 1993), several workers have questioned the origin of DBM considering the diversity of brassica species and the range of parasitoids found (Kfir, 1998; Liu *et al.*, 2000). Twenty eight species of primary parasitoids from four families of Hymenoptera in Romania (Mustata, 1992), thirteen species from four families of hymenoptera and one species from tachinidae (Diptera) in South Africa (Kfir, 1998), eight species from four families of hymenoptera in China (Liu *et al.*, 2000) have been reported.

1.2.6.2 Hyperparasitoids on DBM

A parasite that parasitizes a parasite of insect species is hyperparasitoid and sometimes called secondary parasitoid. This activity of hyperparasitoid limits the efficiency of primary parasitoids in controlling the pest species (Mustata, 1992). Several species of hyperparasitoids of DBM have been reported from several places (Alam, 1992; Mustata, 1992; Kfir, 1998; Liu *et al.*, 2000). *Oomyzus sokolowskii* Kurdjumov has been recorded as a primary parasitoid as well as a facultative hyperparasitoid of *P. xylostella* L. (Fitton and Walker 1992; Liu *et al.*, 2000). This is in disagreement with observations of Talekar and Hu (1996) who reported that *O. sokolowskii* Kurdjumov does not

parasitize *C. plutellae* Kurdjumov and thus is not a facultative hyperparasitoid.

1.2.7 Evaluation of the impact of natural enemies on DBM populations

The accurate measurement of parasitoid impact on Diamondback moth *P. xylostella* (L.) population is important to both the evaluation of exotic parasitoids for introduction, and to the integration of parasitoids with other cropping practices (Waage and Cherry, 1992).

A number of ways can be used to measure the impact of parasitoids on the host. These include insecticide check method to remove parasitoids (Lim *et. al.*, 1986), percent parasitism (Van Driesche, 1983), and recruitment method (Van Driesche and Bellows 1988; Van Driesche *et. al.*, 1991; Wage and Cherry, 1992). Percent parasitism is based on estimation of host and parasitoid density where as recruitment method is based on the measurement of recruitment to both the host susceptible stage and the pool of parasitized host (Van Driesche and Bellows, 1988).

Estimation on mortalities at the different stages of the insect obtained in one or more of these methods will help to construct a life table of the insect which is useful for studying the mechanisms underlying population dynamics which is considered as a prerequisite for the design of integrated pest management systems (Roux and Baumgärtner, 1995).

1.2.7.1 Percent parasitism

This method is one of the simplest methods of assessing the impact of parasitoids on host population although it has been known to produce biases in the estimation (Van Driesche, 1983). Problems associated with the use of this method in accurate estimation are discussed by Waage and Cherry (1992).

The major problems associated with the use of percent parasitism on parasitoids impact assessment are first, measurement is made with no reference to the density of the host population on which the parasitoid act. Secondly, the host stage used for calculating percent parasitism may not be correct which leads to either underestimation or overestimation of parasitism rate (Waage and Cherry, 1992). The other limitation of using this method in tropical countries is that it does not permit a quantitative measure of generational parasitism because of generation overlap of the host population (Lopez and Van Driesche, 1989). These problems have led the development of the recruitment method (van Driesche and Bellows, 1988) and others (Southwood, 1978; Waage and Chery, 1992).

In spite of the problems mentioned, percent parasitism is still widely used since the reliability and practicality of other methods including the recruitment method is questionable. For example, it has been indicated (Liu *et al.*, 2000) that for the assessment of parasitoids impact on *P. xylostella* L., the recruitment method requires not only much work but also produce estimates with biases of unknown magnitude. Despite biases

difficult to avoid in the estimation of parasitism rate using host and parasitoid density (Waage and Cherry, 1992), changes of rates of parasitism obtained by frequent sampling over seasons offer a good indicator of the general pattern of seasonal abundance of parasitoid activity (Liu *et al.*, 2000).

1.2.7.2 Recruitment method

This method estimates the rate at which host larvae are recruited into the stage susceptible to parasitoids over a particular period and the rate at which parasitoids are recruited into the adult parasitoid population from these larvae at a latter time (Van Driesche and Bellows, 1988; Waage and Cherry, 1992). In this method, a certain number of plants are stripped of hosts of susceptible stage at sampling occasion and reexamined after a short interval, counting all hosts present. The number of hosts encountered on the subsequent occasion is taken as the total recruitment during the interval between the two sampling occasions. Parasitism recruitment can be based on the parasitoid egg stage by dissection of the field-collected larvae as described by Van Driesche and Bellows (1988) or rearing the parasitoids from host larvae (Waage and Cherry, 1992).

1.3 HYPOTHESES

- DBM does not cause economic damage in cultivated brassica in Ethiopia
- DBM and DBM parasitoids population dynamics is independent of

season and agroecological zone.

- DBM density is independent of spatial variation
- Suitability of cultivated and wild crucifers for DBM development is the same
- Distribution and abundance of DBM and its parasitoids and their relationship is independent of agro-ecological zones and cropping systems

1.4 OBJECTIVES OF THE STUDY

Overall objective is to contribute to improved management of DBM in the production of brassica in Ethiopia. The specific objectives were:

- To determine parasitoid complex associated with DBM in different brassica producing areas of Ethiopia
- To assess influence of agro-ecological zones and cropping system on the DBM/parasitoid relationship.
- To assess population dynamics of DBM in relation to its parasitoids in different agro - ecological zones
- To determine the spatial relationship of trap catches of male DBM adult by dispersion indices and geostatistical method
- To assess damage caused by DBM on head cabbage (*Brassica oleracea* L. var. *capitata* L.)
- To compare suitability of wild and cultivated crucifer for DBM development

CHAPTER 2

2.0 **Within-field Spatial Distribution of Larvae and Pupae of Diamondback Moth, *Plutella xylostella* L. (Lepidoptera: Plutellidae) in Ethiopian Brassica Fields**

2.1 **Introduction**

The Diamondback Moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), is the most destructive insect of cruciferous plants throughout the world (Talekar and Shelton, 1993).

Diamondback moth (DBM) eggs are laid on different parts of the plant including leaf, stem and petiole. The majority of eggs, however, are laid on the leaf in the proportion of 3:2 on the upper and lower leaf surface (Talekar and Shelton, 1993). The oviposition period lasts three to ten days (Chelliah and Srinivasan, 1986) depending on environmental conditions. Peak oviposition occurs on the first day of emergence. Fecundity as low as 11 eggs and as high as 203 eggs per female has been reported (Chelliah and Srinivasan, 1986; Talekar and Shelton, 1993). The incubation period lasts three to six days depending on temperature (Abraham and Padmanaban, 1968). Upon hatching, neonate larvae start feeding on foliage. The first instar larvae mine in the spongy mesophyll tissue, whereas older larvae feed from lower leaf surface and usually consume all tissues except the epidermis of the upper surface, thus creating a window in the leaf. There are four larval instars. Total larval period ranges from 14 to 21 days

(Abraham and Padmanaban, 1968). Upon completion of feeding, the mature caterpillars construct on the host plant a gauzy, loosely spun cocoon for pupation. The pupa is yellowish-green first then changes to brownish. It attains a dark-brownish color by the time of adult emergence. The pupal period can vary from 4 to 15 days depending on temperature (Talekar and Shelton, 1993).

The association of DBM to plant species of crucifers appears to be due to the presence of one or more glucosinolates, sinigrin, sinalbin and glucocheirolin, which act as specific feeding stimulant (Talekar and Shelton, 1993). Work of Gupta and Thorsteinson (1960) suggests that occurrence of feeding inhibitors as well as the absence of essential feeding stimulants exclude some botanical species in crucifers from the food plant range. The ever-increasing difficulties associated with pesticide-based control of DBM led to the development of biological control projects within the framework of IPM programs (Carl, 1992; Talekar and Shelton, 1993). Currently, the International Center of Insect Physiology and Ecology (ICIPE) is promoting a biological control based IPM program against the pest. Knowledge on spatial distribution of an insect is an integral component in the promotion of IPM (Bennett, 1993) and the basis for sampling plan design.

The objective of the study was to describe the distribution of the larval and pupal stages of DBM on its cultivated host plants.

2.2 Materials and Methods

2.2.1 Sampling Procedures

Data on the density of the different life stages of DBM, young larvae, mature larvae and pupae were obtained from surveys conducted in 13 brassica producing areas of the Rift Valley, northern, western and eastern Ethiopia between February and June 2001 (Fig.2.1). Fields sampled were located along the main road. The number of fields sampled in areas 1 to 13 depended on crop availability and was 43, 19, 18, 8, 10, 12, 7, 11, 8, 4, 8, 33 and 13 respectively. Figure 2.1 depicts the areas surveyed and the number of fields sampled in each area. The sample unit was a plant. Accordingly, in each field, all leaves of ten randomly selected plants were examined for the presence of young larvae, mature larvae and pupae, and the counts were recorded. A sample here refers to all the units (plants) of a field at a given sampling date.

2.2.2 Statistical Analysis of the Spatial Distribution.

Davis (1994) observed that numerous indices, regression models, and distributions have been used to evaluate dispersion, but states that none of them are without some degree of criticism. Iwao's (1968) comprehensive mean crowding regression method was used. The method is based on Lloyd's (1967) mean crowding (mc).

$$mc = m + \frac{s^2}{m} - 1, \quad [2.1]$$

Where m is the population mean, and s^2 is the variance. The mean and the mean crowding statistics were calculated for each sample. The mean crowding mc was regressed on mean density m according to the equation

$$mc = \alpha + \beta m, \quad [2.2]$$

Estimates of the parameters α and β were obtained by the least square method using PROC REG of the SAS statistical package (SAS Institute, 1999). The regression parameters were estimated for each crop separately. Subsequently, an analysis of covariance was carried out to detect significant differences for the slope and intercept values of the mean crowding regression (equation 2.2) for the various life stages on the different brassica crops.

2.3 Results

2.3.1 Sample Distribution

The number of samples of each data set and the ranges of the sample means are given in Table 2.1. A total of 125 fields of cabbage (*Brassica oleracea* L. var. *capitata* L.), 41 fields of Ethiopian mustard (*Brassica carinata* Braun) and 29 fields of kale (*Brassica oleracea* L. var. *alboglabra*) were sampled. The overall density of DBM was higher in cabbage (2.51 ± 0.34) than both in kale (1.41 ± 0.50) and in the Ethiopian mustard (0.63 ± 0.11). Apparently, there was a higher number of mature larvae and pupae in cabbage than the other brassica crops. Differences for the density of young larvae between brassica crops appeared to be smaller than the other life stages. The number of pupae, however,

appeared to be higher in cabbage (1.14) than both in kale (0.34) and the Ethiopian mustard (0.10).

Table 2.2 shows the regression statistics for Iowa's mean crowding (equation [2.2]) for each crop species and life stage. The model gave a significant linear relationship ($p < 0.0023$) of the mean crowding to the mean density for the whole data set except for pupae in the Ethiopian mustard. It accounted for 65 to 76 %, 69 to 91% and 13 to 58% of the variation for young larvae, mature larvae, or pupae on cabbage, kale and the Ethiopian mustard, respectively. The slopes of the regression line were significantly greater ($p < 0.05$) than unity for all the three stages of DBM in cabbage and kale; and for young larvae in the Ethiopian mustard. Values of the intercept were significantly different from 0 only for mature larvae on cabbage ($\alpha = 0.64$, $p = 0.0019$) and pupa on kale ($\alpha = -0.540$, $p = 0.0070$) (Table 2.2).

According to the covariance analysis, slopes of the mean crowding to the mean relationship were not significantly ($P > 0.05$) different between the crucifer crops. Level of significance ranged from 0.361 to 0.624, 0.075 to 0.246, 0.607 to 0.909 and 0.390 to 0.530 for young larvae, mature larvae, pupae and all stages combined. Intercept values were not significant ($P > 0.05$) except for the mature larvae. Hence, data of the three host plant species were pooled and the parameters of equation (2.2) recalculated. Results are shown in Fig. 2.2. Distribution of the different life stages was contagious.

2.4 Discussion

Density of mature larvae and pupae was generally higher in cabbage than was in Kale and the Ethiopian mustard. Differences for the density of young larvae between brassica crops, however, appeared to be smaller than the other stages. The detection of young larvae is more difficult in cabbage than is in kale and Ethiopian mustard. This may introduce a systematic sampling error into the sampling program (LeRoux and Reimer, 1959; Schaub *et al.*, 1988) that, because of limited resources, I was unable to ascertain. A more detailed analysis of crop type, management and parasitoids effect on DBM density has been reported by Ayalew *et al.* (2004).

The slopes of the regression models were significantly greater than unity for all the three stages in cabbage and kale indicating distribution was contagious (Iwao, 1968). In the Ethiopian mustard, the slopes obtained for mature larvae and pupae were not significantly different from unity indicating a random distribution, but young larvae displayed aggregative distributions; the slope was significantly different from unity. The general low number of mature larvae and pupae may be responsible for the random distribution (Table 2.1). The Ethiopian mustard is cultivated mainly as intercrop with large cereals such as maize and sorghum. This may create favorable environment for natural enemies to reduce larval numbers and randomly disperse them during the late stages of development. At the early stage of larval development, mortality caused by parasitoids is low and the effect

of predators is minimal, because of feeding inside the leaf. Harcourt (1961) also reported an increase in randomness with the development of the insect.

Intercept values were not significantly different from zero for the whole data set with the exception of mature larvae in cabbage and pupae in kale indicating that in general, the basic component of the population are single individuals (Iwao, 1968) in most instances. The intercept was significantly higher than zero for mature larvae in cabbage and lower than zero for pupae in kale indicating that the basic components are groups of individuals in cabbage and single individuals in kale (Davis, 1994). The different growth patterns between kale and cabbage may be responsible for the differences between the intercepts. Leaves are fully exposed in kale exposing the insect to predation and removal by rain. Density-independent mortality is reported to alter the value of intercept (Southwood, 1978).

Turner and Gardner (1991) define scale as the spatial or temporal dimension of an object or process characterized by both grain and extent. Here, focus was on grain as the spatial resolution chosen to analyze a given data set, whereas extent is the size of the study on which measurements are made. In general, the observed distribution patterns depend on grain and extent size (Baumgärtner *et al.*, 2002). Here, the mean crowding-mean relationship appears to be constant over the crops considered although the sampling units occupy different ground surfaces and the fields vary in size. Likely, the differences in grain sizes in this study are small for demonstrating these effects. In brassica fields of several countries of Africa including Ethiopia, field size of less than one

tenth of a hectare is common. This restricted range may explain the absence of extent influence.

This information on within field distribution of the larval and pupal stages can be utilized to develop sampling plans for decision making in DBM management when threshold data are available that consider factors such as yield levels, price of product and costs of control measures.

Table 2.1. Density of the different stages of diamondback moth on three brassica crops in Ethiopia (sampling was carried out in 2001)

Brassica crops/ Diamondback moth stage	Range/ plant	Mean \pm SE
Cabbage (125)*		
Young larvae	0-5.7	0.56 \pm 0.08
Mature larvae	0-10.1	0.81 \pm 0.14
Pupae	0-11.5	1.14 \pm 0.16
Total	0-21.4	2.51 \pm 0.34
Ethiopian mustard (41)*		
Young larvae	0-2.5	0.33 \pm 0.08
Mature larvae	0-1.0	0.20 \pm 0.04
Pupae	0-0.4	0.10 \pm 0.02
Total	0-3.2	0.63 \pm 0.11
Kale (29)*		
Young larvae	0-7.5	0.57 \pm 0.27
Mature larvae	0-5.6	0.51 \pm 0.19
Pupae	0-6.6	0.34 \pm 0.23
Total	0-9.9	1.41 \pm 0.50

* Figures in parenthesis show number of fields sampled

Table 2.2. Statistics for Iwao's (1968) mean crowding to mean relationship relative to the distribution of three stages of Diamondback moth on major cultivated brassica crops in Ethiopia

Life stages and brassica crops	R ² (p-value)	$\alpha \pm SE$ (p-value)	$\beta \pm SE$ (p-value)
Young larvae			
Cabbage	0.64 (<0.0001)	0.15 \pm 0.24 (0.5464)	2.18 \pm 0.18 (<0.0001)
Ethiopian mustard	0.58 (<0.0001)	0.05 \pm 0.22 (0.8330)	1.80 \pm 0.30 (0.0056)
Kale	0.69 (<0.0001)	0.31 \pm 0.65 (0.6457)	1.76 \pm 0.32 (0.0125)
Mature larvae			
Cabbage	0.76 (<0.0001)	0.64 \pm 0.20 (0.0019)	1.64 \pm 0.10 (<0.0001)
Ethiopian mustard	0.34 (0.0023)	-0.15 \pm 0.21 (0.4816)	1.79 \pm 0.52 (0.0682)
Kale	0.91 (<0.0001)	-0.14 \pm 0.39 (0.7212)	3.04 \pm 0.26 (<0.0001)
Pupae			
Cabbage	0.65 (<0.0001)	0.27 \pm 0.31 (0.3840)	1.74 \pm 0.13 (<0.0001)
Ethiopian mustard	0.13 (0.0843)	-0.02 \pm 0.23 (0.9252)	1.82 \pm 1.01 (0.2114)
kale	0.78 (<0.0001)	-0.540 \pm 0.15 (0.0070)	4.20 \pm 0.35 (<0.0001)
All stages combined			
Cabbage	0.73 (<0.0001)	0.66 \pm 0.50 (0.1924)	1.71 \pm 0.10 (<0.0001)
Ethiopian mustard	0.75 (<0.0001)	0.0002 \pm 0.14 (0.9907)	1.51 \pm 0.14 (<0.0001)
Kale	0.78 (<0.0001)	-0.57 \pm 1.16 (0.6287)	2.67 \pm 0.35 (<0.0001)

α = intercept of the regression equation

β = slope of the regression equation

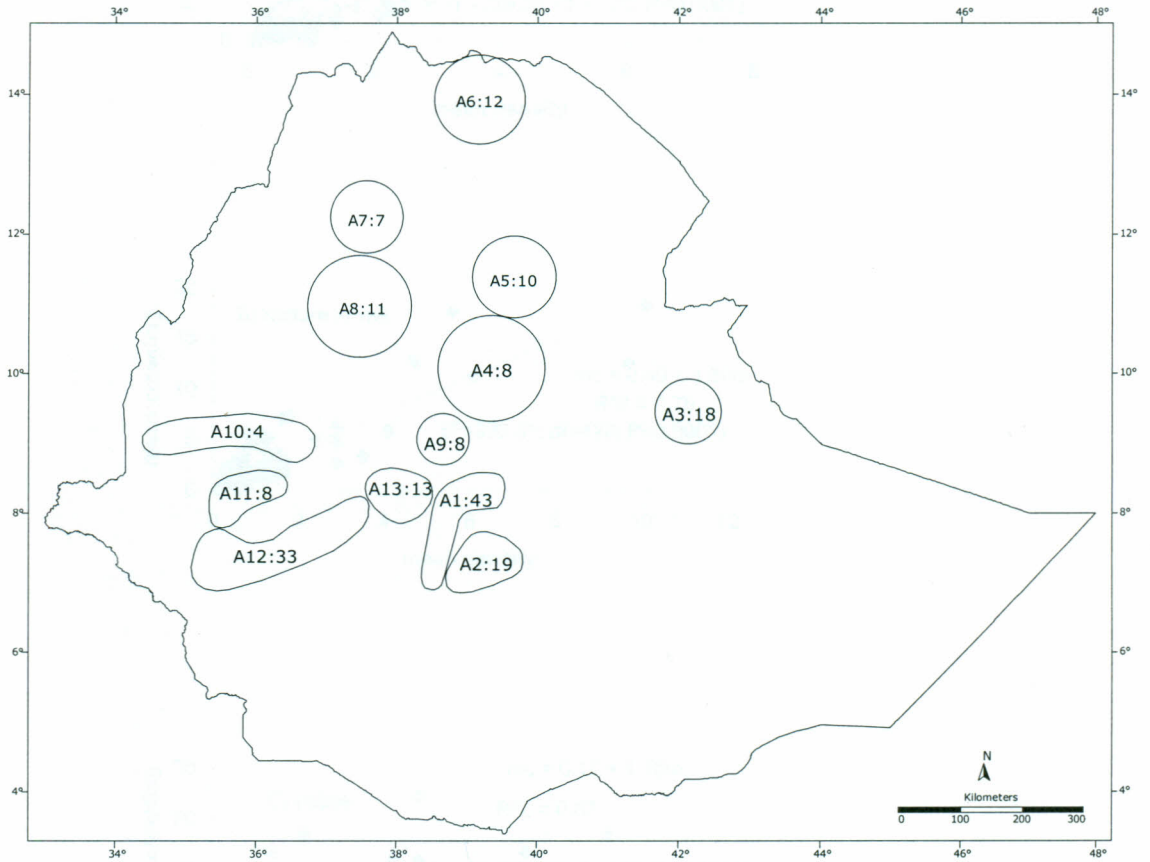


Fig. 2.1. Areas and number of fields surveyed for Diamondback moth in Ethiopia (A = Area; number after colon corresponds to number of fields sampled)

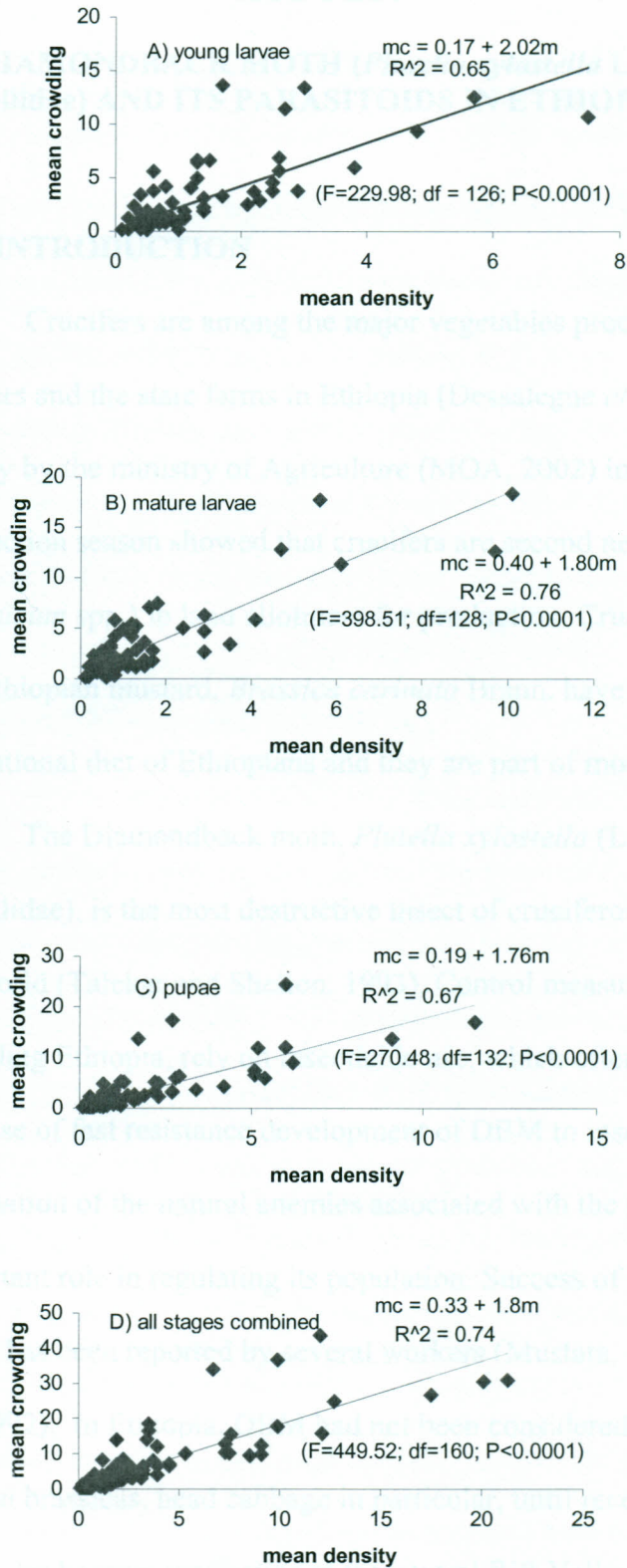


Fig. 2.2. The mean crowding to mean relationship for with-in field distribution of young larvae (A), mature larvae (B), pupae (C) and all stages combined (D) for the pooled data.

CHAPTER 3

3.0 DIAMONDBACK MOTH (*Plutella xylostella* L.) (Lepidoptera: Plutellidae) AND ITS PARASITOIDS IN ETHIOPIA

3.1. INTRODUCTION

Crucifers are among the major vegetables produced both by private farmers and the state farms in Ethiopia (Dessalegne *et al.*, 1994). Recent survey by the ministry of Agriculture (MOA, 2002) in the major production season showed that crucifers are second next to red pepper (*Capsicum* spp.) in land allotment for production. Crucifers, particularly the Ethiopian mustard, *Brassica carinata* Braun. have a dominant place in the national diet of Ethiopians and they are part of most meals.

The Diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), is the most destructive insect of cruciferous plants throughout the world (Talekar and Shelton, 1993). Control measures in Africa, including Ethiopia, rely on insecticide use, which often is ineffective because of fast resistance development of DBM to insecticides and the decimation of the natural enemies associated with the pest which play an important role in regulating its population. Success of biological control of DBM has been reported by several workers (Mustata, 1992; Talekar *et al.*, 1992). In Ethiopia, DBM had not been considered a serious insect pest on brassicas, head cabbage in particular, until recently when pesticides became ineffective in the central Rift Valley of Ethiopia. Although its occurrence as a common pest on head cabbage and other

brassica species has been reported (Abate and Ayalew, 1994), there has been little research on this insect pest in Ethiopia. No information was available on the distribution and severity of the pest and its natural enemies. No single species of natural enemy of DBM is documented in Ethiopia (Abate, 1991). In order to generate baseline information about DBM and the indigenous natural enemies in Ethiopia, a survey was conducted in 2001 in the major crucifer growing areas. This study presents results about the distribution and severity of DBM and its parasitoids on cultivated crucifers in brassica producing areas of Ethiopia.

3.2. MATERIALS AND METHODS

The survey was conducted in crucifer producing areas of Rift Valley, northern, western and eastern Ethiopia in 2001 (Fig. 2.1). Sampling was conducted on brassica field near the main roads. Global positioning system (GPS 300) was used to record the location and altitude of each field. Data records included DBM population, production practices including pesticide usage, intercropping practices and crucifer species/variety grown. In each field, ten randomly selected plants were examined for DBM larvae and pupae; and other pests.

All DBM larvae and pupae were collected and reared in the laboratory in a plastic Petri-dish for determination of parasitism. The Petri-dish was labeled with collection date and field number. When less than 20 DBM larvae and/or pupae were collected from the ten plants

sampled, additional plants were checked for DBM for a period of ten minutes to complement the collection.

In the laboratory, the field-collected material was sorted out, counted and labeled. For each farm, the materials were separated into three groups (pupae, mature larvae and small larvae). Larvae were provided with food (clean leaf of the host plant collected from) to enable the larvae complete their growth. Leaves were changed every other day for the mature and young larvae until pupation. To prevent condensation, a small piece of tissue paper was inserted in the Petri-dish. Emergence of adults and/or parasitoids was recorded every day from the date of collection until no further insects emerged.

3.2.1. Data Analysis

Percent parasitism was calculated by dividing the number of parasitoids by the combined number of emerged parasitoids and DBM adults. For the gregarious parasitoid, *O. sokoluskii*, an average of ten was considered emergent from a single larva or pupa based on a preliminary observation conducted in the laboratory as larvae and pupae of DBM were not kept singly. All adult parasitoids were preserved in ethanol (95%) and labeled with date, field code, site and host plant. Grouping of surveyed fields into areas was made based on proximity of fields along the main road in the survey. Fields in each area had similar features in terms of host plant species, cropping system, pesticide use, etc. Elementary summary

statistics was used to generate mean values of variables measured in each area. Mean values obtained were subjected to multivariate analysis.

A multivariate ANOVA which is appropriate for observational study (Mead *et al.*, 1993) was used instead of other statistical tests. Principal component analysis was used to determine principal factors that determine distribution of DBM and its parasitoids; and cluster analysis was used to cluster areas surveyed (Gray *et al.*, 2000) using SAS soft ware.

Dendrogram and Geographic Information System (GIS) soft ware were used to depict the clusters.

3.3. RESULTS

3.3.1 Diamondback moth distribution and damage severity

All important brassica producing areas of Ethiopia, central Rift Valley, eastern highland, northern, western and southwestern Ethiopia were visited. Between four and 43 fields were sampled in each of the thirteen areas (Fig. 2.1). Diamondback moth numbers ranged from 0.25 to 3.86 per plant (Table 3.2). Density of DBM was higher in central rift valley region, numbered 1, 2. These are mainly vegetable growing areas and cabbage is the dominant crucifer species. On the other hand, DBM density was lowest in west and southwestern part of Ethiopia; areas 10, 11 and 12. In these areas, the dominant crucifer is the Ethiopian mustard, *Brassica carinata* Braun and production is mainly in intercropping with large cereals such as sorghum and maize.

3.3.2 Parasitoids complex and parasitism level

A total of seven hymenopterous parasitoid species from five families were recorded (Table 3.1). Of these *O. sokolowskii*, *Diadegma* sp. and *Apanteles* sp. are the dominant species accounting for higher than 90 percent of the parasitoids complex. Level of parasitism by these major parasitoids in the thirteen crucifer production areas is presented in Table 3.3.

Overall parasitism ranged from 3.6 % (area 10) to 55.8 % (area 7). Parasitism by individual species appeared to associate with areas.

Oomyzus sokolowskii Kurdjumov presence was negligible in southwestern Ethiopia, areas 10,11,12 and 13. On the other hand *Apanteles* sp. and *Diadegma* sp. were largely confined to those areas (Table 3.3).

Level of parasitism and distribution of the three major parasitoids, viz. *O. sokolowskii*, *Apanteles* sp., *Diadegma* sp. and the total parasitism recorded on field bases of areas covered in the survey is shown in Figures. 3.2, 3.3, 3.4 and 3.5 respectively. Higher overall parasitism was observed in southwestern Ethiopia where *Apanteles* sp. and *Diadegma* sp. were the dominant parasitoid species accounting for over 90 percent of all the parasitoid species. *O. sokolowskii* was recorded only from two fields of that area and accounted only for less than one percent of the parasitoid complex recorded in southwestern Ethiopia. On the other hand, *O.*

sokoloskii was the major parasitoid in the central rift valley areas and northern Ethiopia.

3.3.3 Similarity of surveyed areas

3.3.3.1 Correlation Matrix

Correlation matrix from the principal component analysis is presented in Table 3.4. DBM numbers were strongly associated with pesticide use and inversely related to intercropping (Table 3.4). Overall parasitism had a weak negative relationship with pesticide use. Parasitism by *Apanteles* sp. and *Diadegma* sp., however, showed strongly negative relationship with pesticide use and strongly positive relationship with intercropping system. On the other hand *O. sokolowski* appeared to have no relationship with pesticide use and strong negative relationship with intercropping system. Higher level of parasitism by *Apanteles* sp. and *Diadegma* sp. was associated with intercropping and no/minimal pesticide use (Table 3.4).

3.3.3.2 Principal component analysis

The first principal component accounted for 46 % of the variation in the data set and the first two explained 70%. The principal factors for the groupings of areas surveyed were DBM density and pesticide use as can be seen from high positive load of these variables on the first principal component that accounted for 46 % of the variation in the data set (Table 3.5). Figure 3.1 shows the position of areas surveyed on the principal

component axes. Areas on the left bottom had higher level of DBM parasitism and lower DBM population (Fig 3.1a). On the other hand Areas on the top of the first two principal component axes (Fig. 3.1a) had higher DBM population and low level of parasitism.

3.3.3.3 Cluster analysis

Results of cluster analysis of areas surveyed for abundance of DBM and its parasitoids using Ward' s minimum variance method is shown in dendrogram (Fig. 3.6) and depicted in map (Fig. 3.7). Areas 1 and 4 are grouped under cluster 1; areas 3, 5, 7, 11 and 12 under cluster 2; area 2 alone under cluster 3; and areas 6, 8, 9, 10 and 13 under cluster 4. Areas within the same cluster show similar feature in DBM density and pesticide use. Table 3.5 shows higher positive load of DBM density and pesticide use in the first principal component indicating that they are major factors for clustering of areas.

3.4 DISCUSSION

Diamondback Moth density in crucifer fields of Ethiopia was observed to be influenced by one or more factors including the level of insecticide usage, parasitism , and cropping system. In general, higher DBM density was associated with heavy pesticide usage and low level of parasitism. High level of DBM density in pesticide use areas as opposed to minimal or no pesticide use areas can be explained by the development of

resistance by the pest to the pesticides being used in the area as is observed in several countries (Tabashnik *et al.*, 1987; Cheng *et al.*, 1992; Shelton and Wyman, 1992; Syed, 1992; Kibata 1996). Low level of parasitism because of the pesticide effect on the parasitoids (Sastrosiswojo and Sastrodihardjo, 1986; Ooi, 1992; Talekar and Shelton, 1993) also explain the observed higher DBM density in pesticide use areas. Recent observation in Kenya of higher DBM population (Macharia, Personal comm.) in plots treated with the pyrethroid insecticide, Karate, compared to pesticide free plots corroborates this observation.

The extremely low number of *O. sokolowskii* in south western Ethiopia where the highest parasitism by *Apanteles* sp. was observed could partly be due to competition between these two larval parasitoids for DBM. Liu *et al.*, 2000 reported negative relationship in parasitism level between *O. sokolowskii* and braconid *Cotesia plutellae*. The dominant brassica species sampled in south western Ethiopia was the Ethiopian mustard, *Brassica carinata* Braun with intercropping system that might have contributed to the observed variation in host parasitoid relationship. Plant species influence on host parasitoid relationship has been reported (Verkerk *et al.*, 1998; Liu *et al.*, 2000). However, estimation of their influence is difficult without a controlled experiment (Liu *et al.*, 2000).

The present study has revealed that a number of insect parasitoids are associated with DBM in Ethiopia. Of these, *O. sokolowskii*, *Apanteles*

sp. and *Diadegma* sp. are the most important ones accounting for more than 90 % of the parasitoid complex.

In east and central African countries, *D. mollipla* is a common DBM parasitoid (Löhr and Kfir, 2003). The *Diadegma* sp. recorded from DBM in Ethiopia has however been reported to be different from *D. mollipla* and other known species of the genus (Wagner *et al.*, 2002) suggesting that it can be a new addition to the species complex of the genus *Diadegma*. *D. mollipla* was earlier recovered in Ethiopia from the Potato Tuber Moth (PTM), *Pthorimaea operculella*, (Negasi *et al.*, 1986). Considering the brassica production situation in Ethiopia especially in the vegetable cropping system where potato are grown side by side with brassica crops, the total absence of *D. mollipla* from DBM in Ethiopia is not clear. One possible reason could be that the new species yet to be confirmed might have replaced it.

The *Apanteles* sp recorded could also be new to DBM (Gérard Delvare, Personal communication). It was earlier misidentified and reported as *Cotesia plutellae* (Ayalew *et al.*, 2004) which is a common DBM larval parasitoid in southern African countries (Lohr and Kfir, 2003)

The level of parasitism by the indigenous parasitoid species is generally low and the DBM density is high in the major brassica producing areas of the country: the Arsi highland (Cluster 3) and the rift valley area (cluster 1).

Biocontrol of DBM in Ethiopia needs to target the central Rift Valley region and the Arsi highland through both the introduction of effective exotic parasitoids and conservation of the indigenous ones. Currently used pesticides that are harmful to the parasitoids need to be replaced with safer alternatives such as the use of the neem based products and microbial pesticides such as *Bacillus thuringiensis*.

<i>Exochus</i> sp.	Ichneumonidae	Major
<i>Apanteles</i> sp.	Braconidae	Minor
<i>Brachymeria</i> sp.	Chalcididae	Minor
<i>Pentapleura angustifrons</i>	Ichneumonidae	Minor
<i>Imperialia</i> sp.	Ichneumonidae	Minor
<i>Mesochorus</i> sp.	Proctosptera	Minor

Table 3.1. Hymenopteran parasitoids of DBM recorded from Ethiopia, 2001.

Species	Family	status
<i>O. sokolowskii</i>	Eulophidae	Major
<i>Diadegma</i> sp.	Ichneumonidae	Major
<i>Apanteles</i> sp.	Braconidae	Major
<i>Brachchymeria</i> sp	Chalcididae	Minor
<i>Pediobius angustifrons</i>	Eulophidae	Minor
<i>Itoplectis</i> sp.	Ichneumonidae	Minor
<i>Mesopolobus</i> sp.	Pteromalidae	Minor

Table 3.2. Diamondback moth density in 13 crucifer growing areas of Ethiopia

Area	No. fields Sampled	DBM per plant Mean (\pm SE)	Damage Score Mean (\pm SE)	Altitude (m a.s.l.) Mean (\pm SE)
1	43	3.12 (0.63)	2.86 (0.24)	1668 (16)
2	19	3.86 (1.14)	3.58 (0.36)	2468 (90)
3	18	2.22 (0.94)	2.72 (0.36)	2024 (9)
4	8	2.50 (1.42)	2.62 (0.59)	1603 (173)
5	10	0.23 (0.11)	1.20 (0.59)	1794 (64)
6	12	2.39 (1.08)	2.58 (0.47)	2130 (69)
7	7	1.69 (0.53)	2.43 (0.43)	1977 (59)
8	11	1.66 (0.40)	2.18 (0.32)	2214 (125)
9	8	2.15 (0.87)	2.50 (0.38)	2200 (59)
10	4	0.70 (0.17)	2.00 (0.41)	2140 (119)
11	8	0.69 (0.25)	1.87 (0.35)	1800 (41)
12	33	0.70 (0.15)	1.73 (0.19)	1860 (36)
13	13	0.25 (0.06)	1.15 (0.10)	2212 (126)

Table 3.3. Diamondback moth parasitism rates (percent) by major parasitoids in 13 crucifer growing areas of Ethiopia

Area	Percent parasitism of DBM by the major parasitoid species			
	<i>O. sokolowskii</i>	<i>Diadegma</i> sp.	<i>Apanteles</i> sp.	Over-all
1	11.80 (3.29)	0.54 (0.32)	0.86 (0.57)	13.21 (3.41)
2	23.35 (6.07)	1.44 (0.91)	2.86 (1.73)	27.66 (7.08)
3	11.48 (6.89)	7.96 (4.03)	0 (0)	19.44 (7.36)
4	15.54 (4.60)	1.74 (0.91)	1.19 (1.19)	18.47 (5.44)
5	54.46 (4.60)	25.00 (25.00)	0 (0)	79.46 (13.55)
6	11.28 (4.20)	0 (0)	0 (0)	11.28 (4.20)
7	55.88 (18.22)	0 (0)	0 (0)	55.88 (18.22)
8	28.13 (11.49)	2.81 (2.23)	0 (0)	30.93 (10.97)
9	22.13 (10.26)	8.65 (3.27)	3.74 (2.07)	34.52 (11.93)
10	0 (0)	1.78 (1.78)	1.78 (1.78)	3.57 (3.57)
11	0 (0)	15.13 (6.49)	9.43 (5.64)	24.57 (9.58)
12	0.26 (0.18)	17.08 (4.09)	13.62 (3.18)	30.96 (5.040)
13	0 (0)	9.95 (5.49)	18.97 (12.48)	28.92 (12.49)

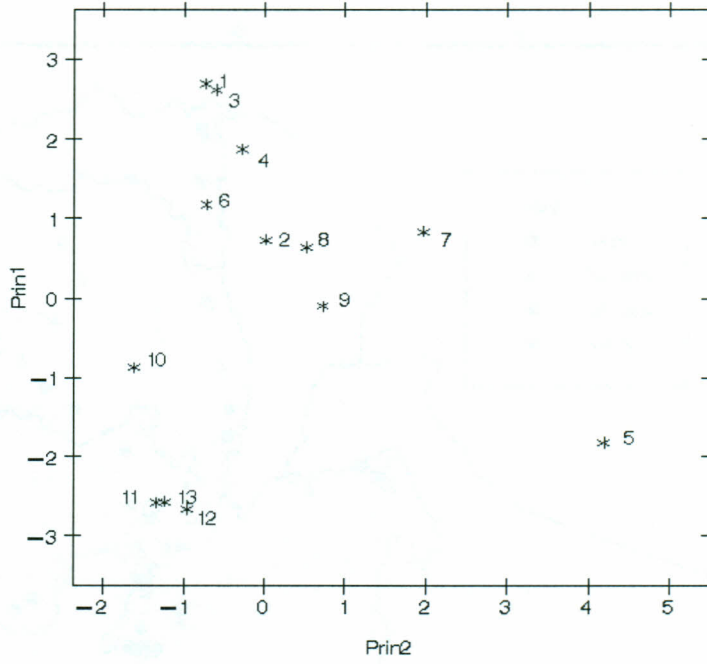
Table 3.4 Correlation matrix of physical environment, Diamondback moth population density and crop management of brassica fields of thirteen areas of Ethiopia , 2001

	FRE	INS	TOT	APA	DIAD	OM	DBMIN	AL
Intercropping (IN)	-0.353	-0.513	-0.269	0.704	0.293	-0.640	-0.610	-0.125
Altitude (AL)	-0.348	-0.244	-0.146	0.096	-0.189	-0.050	0.170	
DBM intensity (DBMIN)	0.505	0.617	-0.351	-0.445	-0.603	0.002		
Parasitism by <i>O. sokolowskii</i> (OM)	-0.118	0.026	0.862	-0.516	0.277			
Parasitism by <i>Diadegma</i> sp. (DIAD)	-0.356	-0.491	0.635	0.380				
Parasitism by <i>Apanteles</i> sp. (APA)	-0.446	-0.562	-0.047					
Over-all parasitism (TOT)	-0.336	-0.262						
Insecticide usage (INS)	0.950							
Frequency of insecticide application (FRE)								

Table 3.5 Eigenvectors of the first three principal components of variables measured in brassica fields of Ethiopia, 2001

Variable	Prin 1	Prin 2	Prin 3
Intercropping	- 0.363	- 0.364	0.225
Altitude	- 0.033	-0.060	- 0.750
DBM intensity	0.419	- 0.062	- 0.250
Parasitism by <i>O. sokolowkii</i>	0.055	0.615	- 0.060
Parasitism by <i>Diadegma</i> sp.	-0.352	0.270	0.282
Parasitism by <i>Apanteles</i> sp.	-0.381	- 0.268	0.028
Parasitism (Total)	-0.169	0.570	0.063
Pesticide use	0.465	- 0.027	0.263
Pesticide application frequency	0.414	- 0.096	0.399

a)



b)

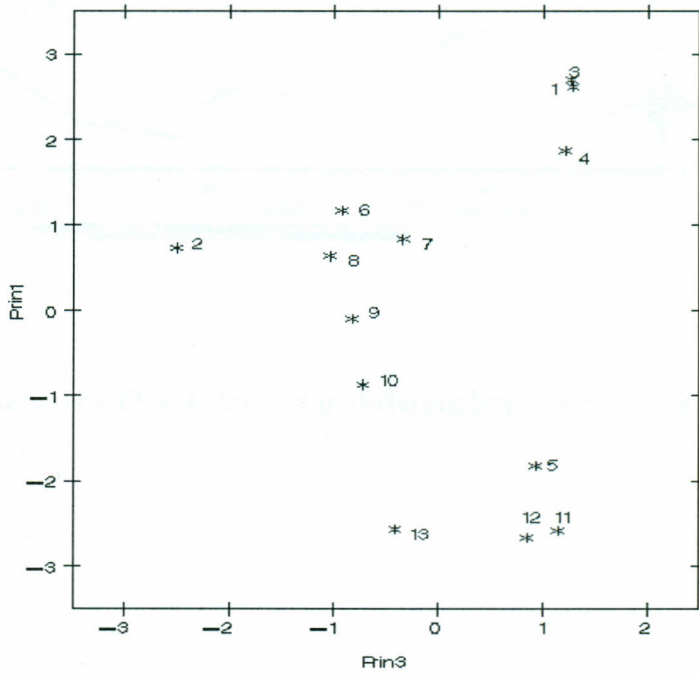


Fig. 3.1 Plot of a) the first and second; and b) the first and third axes from the principal component analysis showing similarity of areas surveyed for DBM and its parasitoids on brassica fields of Ethiopia, 2001

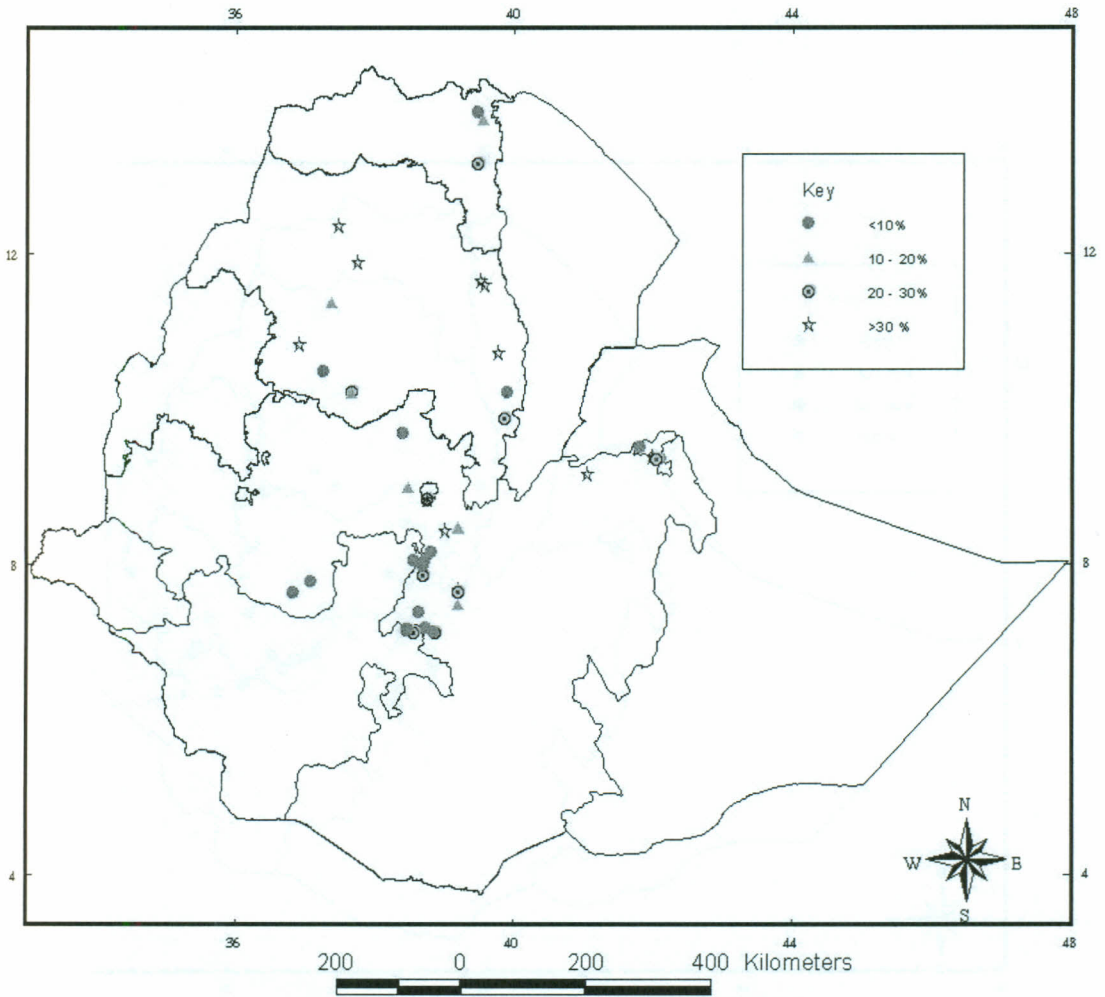


Fig 3.2. Level of DBM parasitism by *O. sokolowskii* in different brassica producing areas of Ethiopia

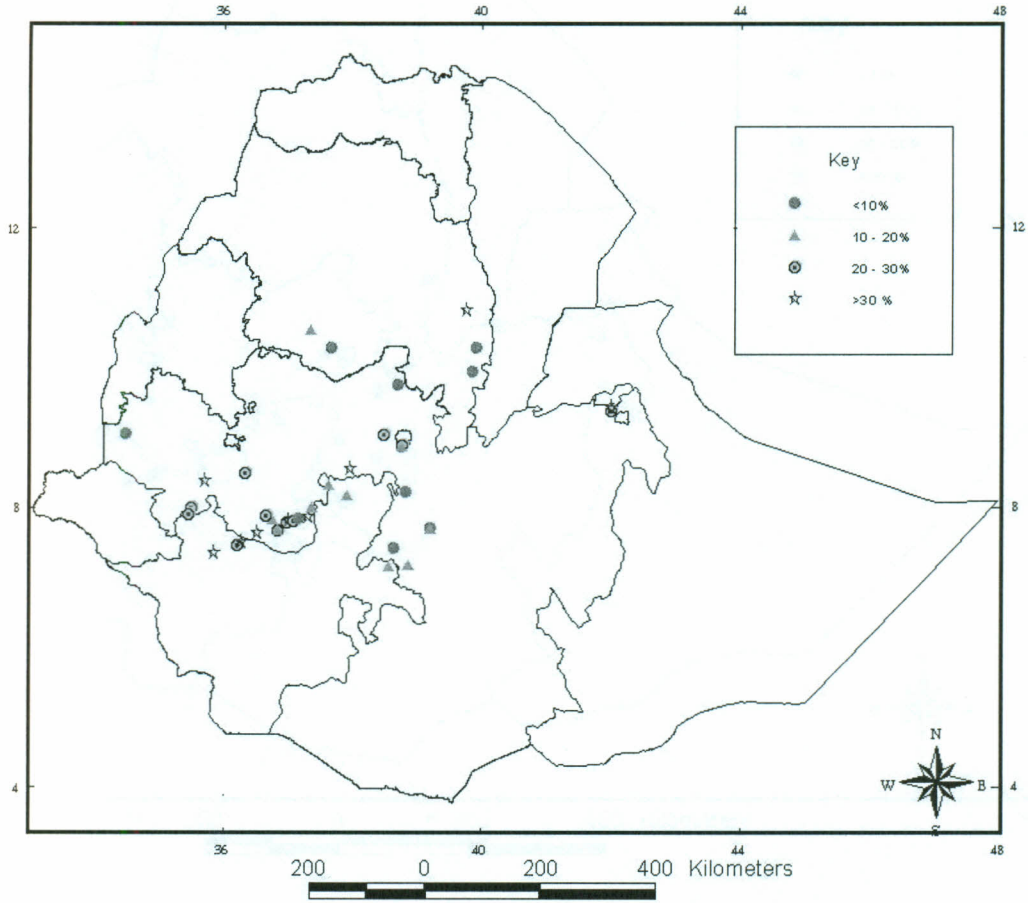


Fig 3.3. Level of DBM parasitism by *Diadegma* sp. in different brassica producing areas of Ethiopia

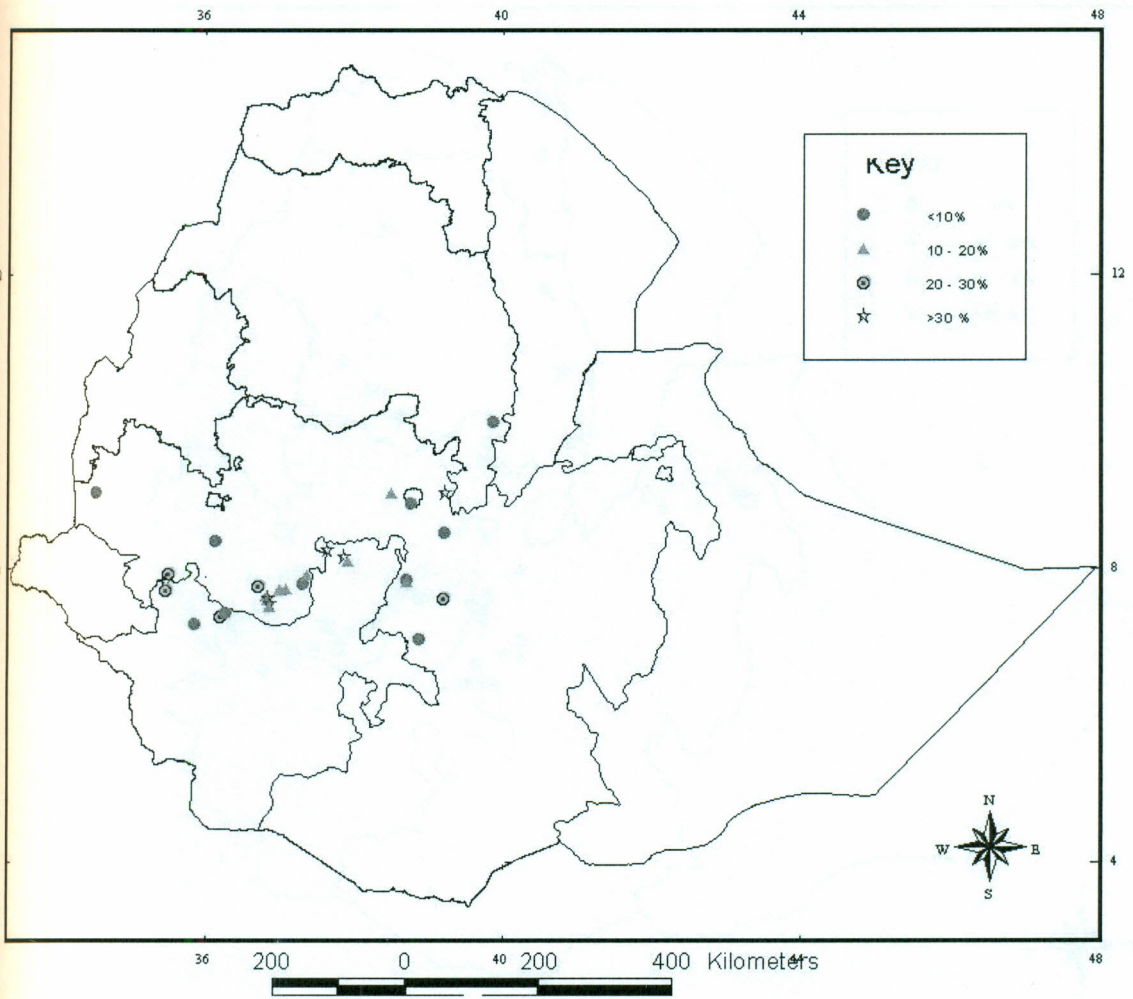


Fig. 3.4 Level of DBM parasitism by *Apanteles* sp. in different brassica producing areas of Ethiopia

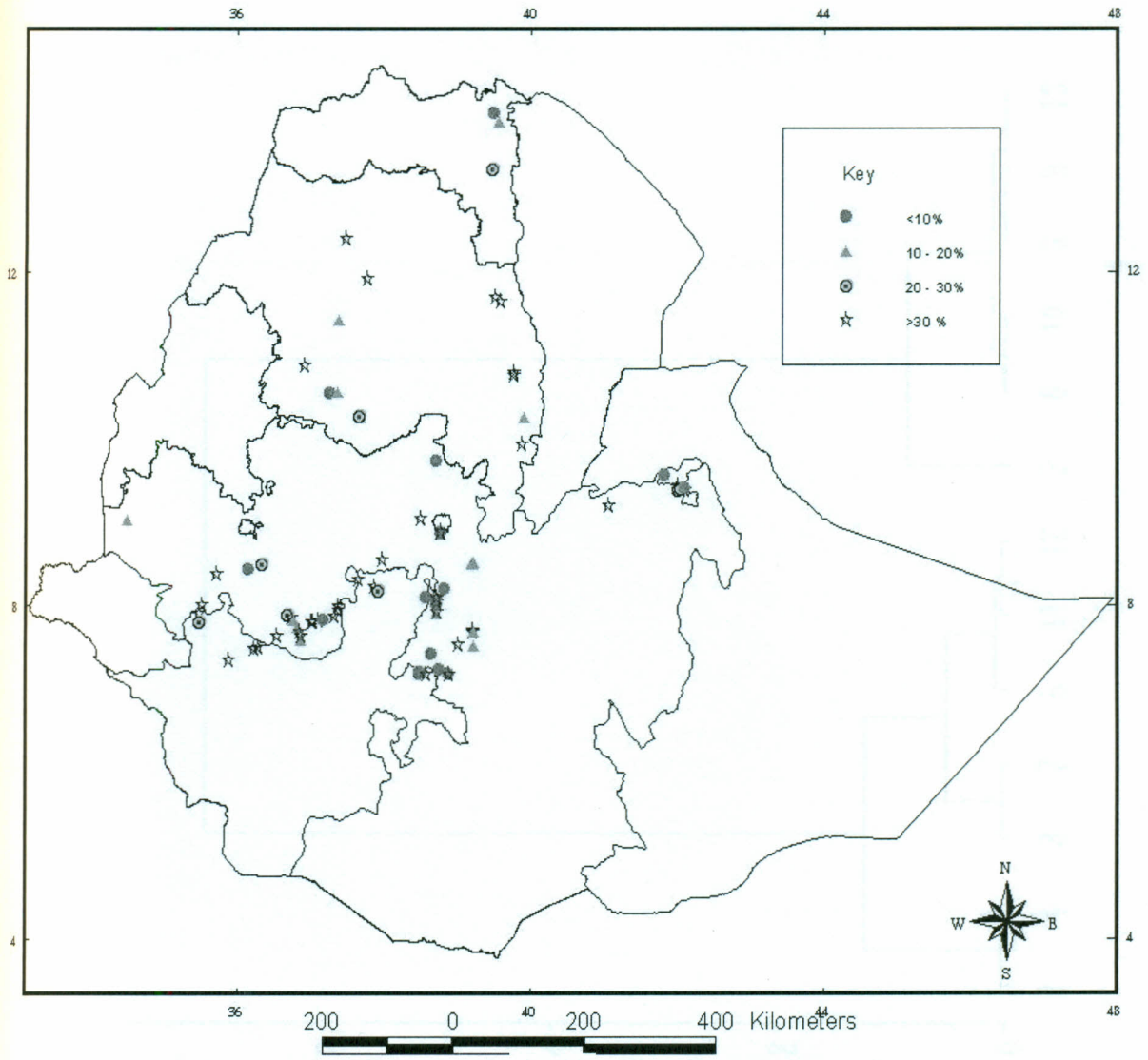


Fig. 3.5 Over-all level of DBM parasitism by its parasitoids in different brassica producing Areas of Ethiopia

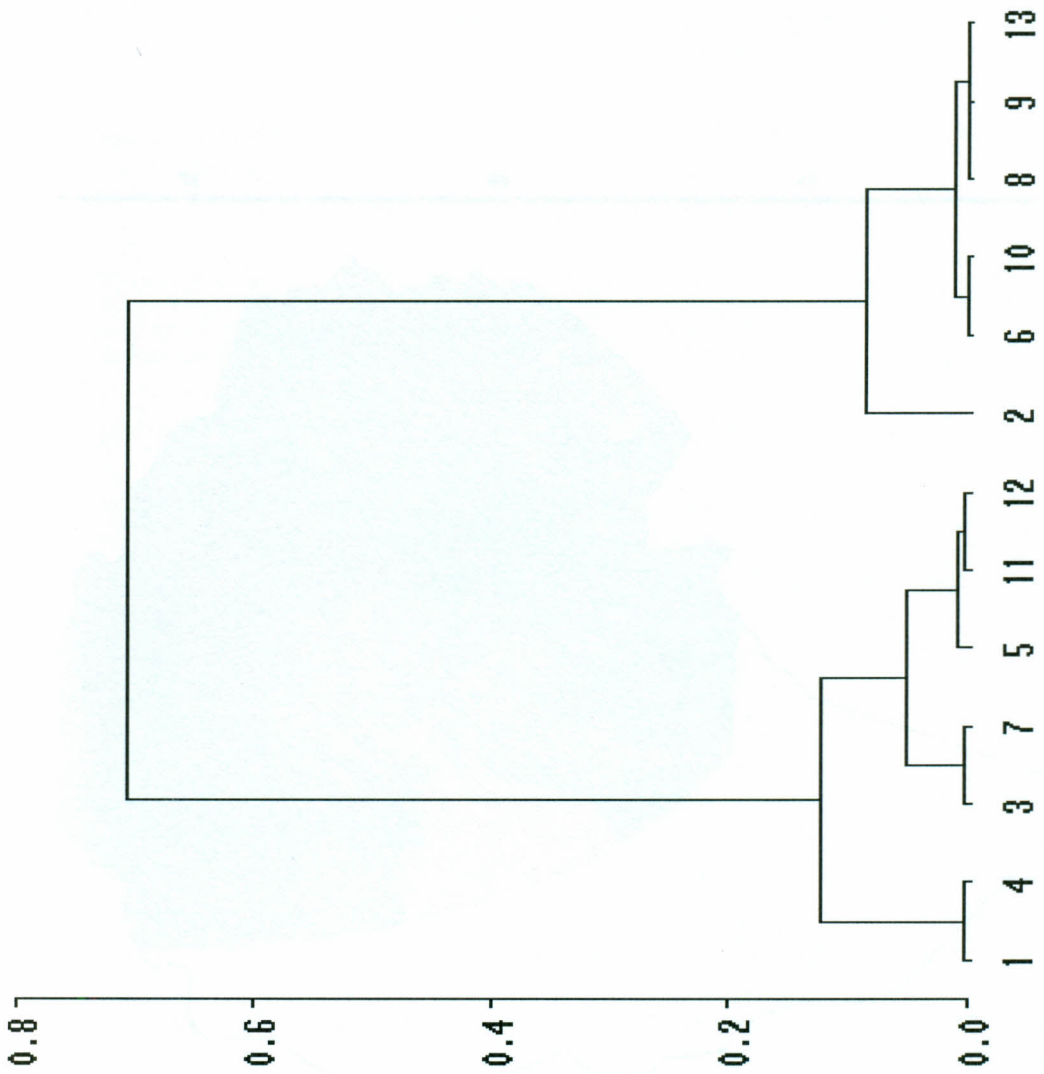


Fig 3.6. Dendrogram showing clusters of thirteen areas of Ethiopia surveyed for DBM and its parasitoids; Ward's minimum variance method

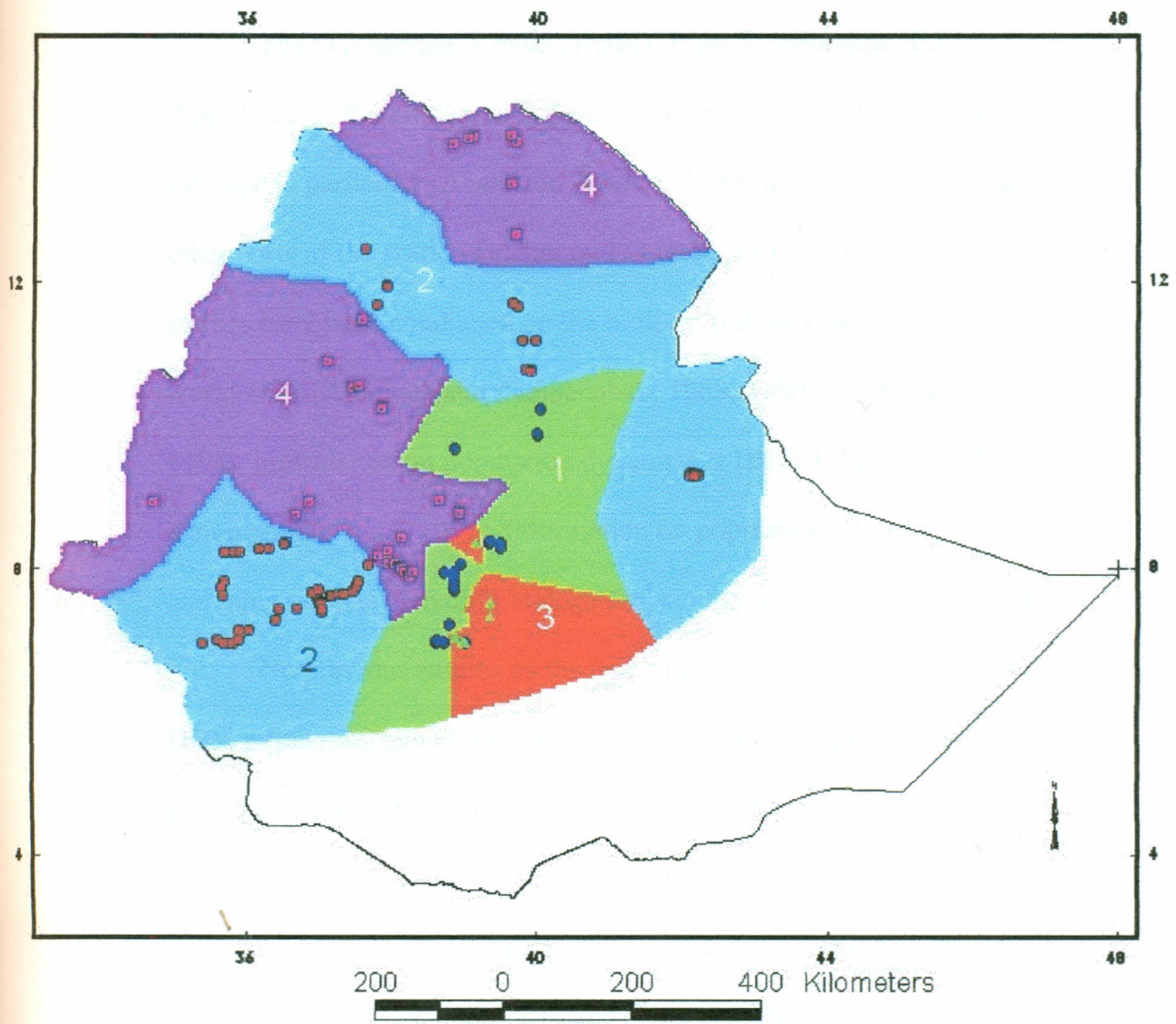


Fig. 3.7 Four areas of Ethiopia for DBM and parasitoids distribution; Results of cluster analysis

CHAPTER 4

4.0. POPULATION DYNAMICS OF DIAMONDBACK MOTH [(*Plutella xylostella* (L.): (Lepidoptera: Plutellidae) AND ITS PARASITOIDS IN CENTRAL ETHIOPIA

4.1 INTRODUCTION

The Diamondback moth (DBM), *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), is a destructive insect pest of brassica, head cabbage in particular (Abate and Ayalew, 1994) in Ethiopia, as it is in other parts of the world (Talekar and Shelton, 1993). Although extensive studies have been conducted on the biology and the control of this pest world wide (Abraham and Padmanaban, 1968; Talekar and Shelton, 1993), information on population dynamics is limited (Harcourt 1963; Keinmeesuke *et al.*, 1992). Information on population dynamics of DBM and other aspects including biology and control are lacking in Ethiopia.

No previous studies on the role of insect parasitoids on regulating DBM population have been conducted in Ethiopia. Reports on insect parasitoids associated with DBM in Ethiopia were not available until recently (Ayalew *et al.*, 2004). Three major insect parasitoids, namely *Oomyzus sokolowskii* (Hymenoptera: Eulophidae), *Diadegma* spp. (Hymenoptera: Ichneumonidae), and *Cotesia plutellae* (Hymenoptera: Braconidae) are reported to be associated with DBM in major cultivated brassica crops in Ethiopia (Ayalew *et al.*, 2004). Knowledge of parasitoids impact on their host would help to promote biological control as observed in several countries (Talekar and Shelton, 1993). In general, the

understanding of the fluctuation pattern of the insect population and its parasitoids helps in the design of appropriate management in an Integrated Pest Management program (Jervis and Kidd, 1996)

This study was performed with the aim of understanding the fluctuation trends of DBM in different production seasons in the high and low elevation brassica production areas of Ethiopia. The role of meteorological factors (temperature, rainfall and relative humidity) and insect parasitoids in the DBM population fluctuation and their importance as natural control factors of the pest was assessed.

4.2 MATERIALS AND METHODS

The experiment was carried out at Melkassa center (8° 24' N; 39° 21'E, alt. 1550) representing the low-lying brassica producing areas and at Holeta center (9° 02' N; 38° 30'E, alt 2400) representing the highland brassica producing areas. Both centers are part of the Ethiopian Agricultural Research Organization (EARO). Data were collected during two seasons at each site. The first season lasted between November/December 2001 and March/April 2002, and the second season between April/May and August 2002. Seed of the widely grown cabbage variety, Copenhagen Market was used. Seeding and (transplanting) was made on 10 October 2001(4 December 2001) and 4 March 2002 (20 April 2002) in December and April plantings respectively at Holeta and on 30 September 2001(12 November 2001) and 10 March 2002 (16 April 2002) at Melkassa. The size of the experimental field was 44 X 40

m at Melkassa and 30 X 20 m at Holeta in both seasons. Spacing between rows and plants was 60 and 40 cm respectively. Cultural practices (fertilization, irrigation, weeding etc.) were followed as per recommendation of the horticulture division of the research center. No pesticide was applied during the entire period of the crop growth.

4.2.1 Sampling Procedure and Data Collected

Fields were divided into 12 plots, each of 20 rows and 10 m long in both seasons at Melkassa. One plant per row of each plot was sampled randomly giving a total of 240 plants per sampling period. At Holeta, fields were divided into 12 plots, each of 8 rows and 10m long in both seasons. Two plants per row were sampled from the December 2001 planted cabbage with a sample size of 192 per sampling period and a single plant per row from the April planted field.

Sampled plants were examined for the different stages of the insect; young larvae, mature larvae and pupae, twice weekly (three to four days interval) in most cases and records were made.

During each sampling, the different stages of the insect, young larvae, mature larvae and pupae were collected for parasitoids rearing. The materials were sorted by stages and kept in plastic petri-dishes in the laboratory. Larvae (both young and mature) were provided with food (clean leaf of the host plant collected from to enable them complete their growth). Leaves were changed every other day until adult DBM and/or parasitoids emergence. Recording for emerged adults and/or parasitoids

were made every day from the date of collection until no further emergence occurred. Percent parasitism for each sampling period was calculated by dividing total number of parasitoids with the numbers of emergent parasitoids and DBM. To determine parasitism by the gregarious parasitoid species, *O. sokolowskii*, ten individuals were regarded as emergent from one larva or pupa (Ayalew *et al.*, 2004). Meteorological data (rainfall, relative humidity, minimum and maximum temperature) were obtained from Meteorology Division of the respective research centers.

4.2.2 Statistical Analysis

Mean insect number per plant was calculated for each stage of the insect per sampling period to examine the population fluctuation for the different seasons and locations.

Potential parasitism i.e. parasitism level by each parasitoid species in the absence of others was determined using the method used by Dreyer and Baumgärtner (1995) which was proposed by Carey (1989). The following assumptions were considered in the calculation of apparent and potential parasitism (Dreyer and Baumgärtner, 1995).

If D_A and D_B denote the fractions of DBM that were parasitized by parasitoid A and B, then the probability of surviving DBM by both parasitoids is given as

$$P_{AB} = 1 - (D_A + D_B) = (1 - q_A) (1 - q_B)$$

where q_A and q_B are the probabilities of parasitism by parasitoid A and B respectively. Assuming that the ratio of the observed fractions (D_A/D_B) equals the ratio of the probabilities (q_A/q_B), substitution of the corresponding terms in the above equation yields the quadratic equation:

$$aq_A^2 + bq_A + c = 0$$

where $a = D_B$, $b = -(D_A + D_B)$ and $c = D_A(D_A + D_B)$

Hence, the probability of DBM being parasitized by parasitoid A, q_A , is

$$q_A = (-b - (b^2 - 4ac)^{0.5}) / 2a$$

and q_A is calculated via the above relationship between D_A/D_B and q_A/q_B .

Probabilities for three and more parasitoids are determined by applying the quadratic to two factor cases, e.g. *O. sokolowskii* vs (*Diadegma* sp. + *Apanteles* sp.).

The proportion of parasitized hosts (P_i = arcsin transformed proportions) by the i -th parasitoid species (*Diadegma* sp. $i=1$, *Oomyzus sokolowskii* $i=2$, *Apanteles* sp. $i=3$) was expected to be influenced by location ($L=0$ for Holeta, and $L=1$ for Melkassa), plant age A [Days after planting], level of parasitism by other parasitoid (P) species, season [$S=0$ early planting and $S=1$ late planting], D = host density and weather condition and developed the following model that I subjected to step-wise linear regression analysis.

$$P1 = a_1 + a_{12}L + a_{13}A + a_{14}S + a_{15}D + a_{16}P2 + a_{17}P3 + a_{18}R + a_{19}H + a_{110}T_{\max} + a_{111}T_{\min} \quad [4.1a]$$

$$P2 = a_{21} + a_{22}L + a_{23}A + a_{24}S + a_{25}D + a_{26}P1 + a_{27}P3 + a_{28}R + a_{29}H + a_{210}T_{\max} + a_{211}T_{\min} \quad [4.1b]$$

$$P3 = a_{31} + a_{32}L + a_{33}A + a_{34}S + a_{35}D + a_{36}P1 + a_{37}P2 + a_{38}R + a_{39}H + a_{310}T_{\max} + a_{311}T_{\min} \quad [4.1c]$$

where S = season of planting, D = density of DBM, P = parasitoid species

(1 = *Diadegma* sp. , 2 = *Oomyzus sokolowskii*, 3 = *Apanteles* sp.), R =

rainfall (mm), H = daily mean humidity between successive sampling,

T_{\max} =daily mean maximum temperature between consecutive sampling,

T_{\min} = daily mean minimum temperature between consecutive sampling.

'Mean' means 'average sample values'

I used a significance level of 0.3 for variable entry and stay in the stepwise regression model.

DBM population density and weather data of the two locations were compared using student- t test. Analysis was performed using SAS statistical package (SAS institute, 1999).

4.3. RESULTS

4.3.1 *Population dynamics of DBM and its Parasitoids at Holeta*

(highland production site)

Diamondback moth population dynamics and level of parasitism by its insect parasitoids are shown in Fig. 4.1 for December 2001 plantings and Fig. 4. 2 for April 2002 plantings. Population of DBM fluctuated between 0 to 15.7 and 0 to 1.7 insects per plant in December and the April planted fields respectively.

Three parasitoids were recorded in both seasons. These included *Oomyzus sokolowskii* (Kurdjumov) (Hymenoptera: Eulophidae), *Diadegma* sp. (Hymenoptera: Ichneumonidae), and *Apanteles* sp. (Hymenoptera: Braconidae). Overall parasitism ranged from 0 to 66% and 0 to 86% respectively. Parasitism by *O. sokolowskii*, *Diadegma* sp. and *Apanteles* sp. ranged between 0 and 22.2%, 0 and 56.2% and 0 and 11% in the December planted field. In the April planted field, it ranged between 0 and 21%, 0 and 86% and 0 and 30% respectively. Of the total of 2142 insects reared from the collection of the December planted field, 128 *O. sokolowskii*, 103 *Diadegma* sp. and 63 *Apanteles* sp. were recorded. This corresponds to a parasitism level of 5.9, 4.8 and 2.9 per cent respectively. From the planting made in April, a total of 523 insects were reared of which 42 were *O. sokolowskii*, 160 were *Diadegma* sp. and 41 were *Apanteles* sp. corresponding to a parasitism level of 8.0, 30.6 and 7.8 per cent respectively. Overall average parasitism was 13.7% in the December planting and 46.6% in the April planting.

Group comparison of means of meteorological data of December and the April planting showed significantly higher maximum temperature in December planting than the April planting ($df = 14$; $F = 5.06$ $P = 0.0058$) and significantly higher rainfall in the April planting than the December planting ($df = 14$; $F = 4.15$; $P = 0.0145$).

4.3.2 Population dynamics of DBM and its parasitoids at Melkassa (lowland production site)

Diamondback moth density at Melkassa fluctuated between 0 to 3.2 insects in the December planted field (Fig. 4.4) and between 0 to 8.5 insects per plant in the April planted one (Fig. 4.5). Estimation of the number of generations using physiological time showed occurrence of three generations in the December planted field peaking on 20th January, 14th February and 3rd March. In the April planted field, the number of generations observed both from population peaks (Fig. 4.5) as well as degree days calculated showed occurrence of five generations. Based on the physiological time, peaks of the five generations occurred on June 7, June 23, July 9, July 28 and August 18. The level of parasitism in the December planted cabbage was low (0 to 24%) and mainly was due to *O. sokolowskii*. *Diadegma* sp. was not recorded at all and *Apanteles* sp. was observed on the 8th sampling (8%) which is not shown in Fig. 4.4b. Of the total of 997 insects reared, only 44 were parasitised by *O. sokolowskii* and two by *Apanteles* sp.. Overall average parasitism was 4.8 per cent; 96.5% of which was attributed by *O. sokolowskii*. On the other hand, parasitism in the trial planted in April was much higher. Here too, *O. sokolowskii* was the dominant parasitoid, followed by *Apanteles* sp.. Parasitism by *Diadegma* sp. was low. Overall parasitism ranged between 0 and 81 %. Parasitism by *O. sokolowskii*, *Apanteles* sp. and *Diadegma* sp. ranged

between 0 and 62.7%, 0 and 20% and 0 and 3.3% respectively. Overall mean parasitism was 25.8% *O. sokolowskii* accounted for 88.7%, *Apanteles* sp. for 8.8% and *Diadegma* sp. for 2.2% of the overall mean parasitism of the season.

Potential parasitism, i.e. parasitism by each parasitoid species in the absence of others calculated from the observed overall average parasitism with all species occurring concurrently for each location and season is shown in Table 4.7. The difference did not appear to be larger. Overall percent increment was 5.4 % at Holeta in the second season. This was mainly attributed to *Diadegma* sp. that exhibited an apparent parasitism of 30.5% and potential parasitism of 34.7 % at Holeta in the second season.

4.3.3 Regression of parasitism by the three major parasitoids on host density and meteorological variables

A. Diadegma sp.

The final model is shown in equation 4.2a.

$$P1_i = 0.05 - 0.12L + 0.13S - 0.02D - 0.18P2 - 0.21P3 + 0.003H - 0.007T_{\min} \quad [4.2a]$$

Location, season, host density and relative humidity showed a significant influence ($P < 0.05$) on parasitism by *Diadegma* sp. (Table 4.4). Table 4.3 shows the stepwise procedure for selection of variables. Host density effect can be interpreted using derivatives as follows:

$\frac{DP1}{dD} = -0.02$, an increase in host density by a unit or one decreases

parasitism level by *Diadegma* sp. by 0.02. This indicates an inversely density dependent relationship between *Diadegma* sp. and DBM density.

Similarly, the effect of relative humidity can be interpreted as

$\frac{DP1}{dH} = 0.003$, an increase in the relative humidity by one increase

parasitism by *Diadegma* sp. by 0.003.

B. *Oomyzus sokolowskii*

The final model is shown in equation 4.2b. Table 4.2 shows the stepwise procedure for selection of variables.

$$P2_i = -0.47 + 0.13S + 0.01D - 0.17P1 + 0.002H + 0.02T_{\max}$$

[4.2b]

Planting season, parasitism by *Diadegma* sp. and maximum temperature influenced parasitism by *O. sokolowskii* significantly (Table 4.5).

An increase in the level of parasitism by *Diadegma* sp. by one decreases parasitism level by *O. sokolowskii* by 0.17. Maximum temperature showed a positive effect on parasitism level; an increase by one in maximum temperature resulted in increase in parasitism level by 0.02. A significant positive coefficient of season indicates significantly higher level of parasitism in the second season than the first season.

C. Apanteles sp.

The final model is shown in equation 4.2c. Table 4.1 shows the stepwise procedure for selection of variables.

$$P3 = -0.03 + 0.06S + 0.001A - 0.006D - 0.05P1 + 0.001H - 0.005T_{\min}$$

[4.2c]

Plant age, host density, planting season, relative humidity and minimum temperature influenced parasitism by *Apanteles* sp. significantly ($P < 0.05$) (Table 4.6). An increase in plant age by a day showed an increase in parasitism level by 0.001. Relationship with host density was negative; increase in DBM number by one decreased parasitism by 0.006 indicating an inversely density dependent relationship. An increase in relative humidity by one increased parasitism by 0.001 and an increase in minimum temperature by one decreased parasitism level by 0.005.

4.4 DISCUSSION

DBM population buildup started slowly and reached its first, second and third peaks on February 8, March 15, and March 29 respectively in the highland experimental site in the December planted trial. Diamondback moth requires 518 degree days to complete one generation with a lower developmental threshold of 7.2 °C (Phillips ND). Degree-days calculated using the average method basing the first generation of young larvae observed on February 1 showed that the 2nd and 3rd generations could be expected on March 9 and April 9. This fairly

coincides with the observed peaks (Fig. 4.1) suggesting occurrence of three generations in the December planted field of the highland site.

On the other hand, parasitism level was higher during the early stage of the crop growth with the highest level of 66% on January 22 when DBM population was generally low. This higher level of parasitism at low host population density could be due to a good searching capacity of *Diadegma* sp. as reported for *Diadegma insulare* (Martínez-Castillo *et al.*, 2002) and favorable environment for the parasitoid activity.

The generally observed lower density of DBM in the second season and a decline from early sampling period until harvest could be explained by higher rainfall and lower temperature in this season compared to the first one (Fig 4.3). Reports on the effect of rainfall on abundance of DBM (Harcourt 1963; Shigekazu *et al.*, 1992) and higher parasitoid activity in rainy period (Martínez-Castillo *et al.*, 2002) are similar to my observation in the highland experimental site.

Although the population of DBM generally remained low, three distinct peaks were observed on June 11, July 9 and July 26 in the April planted trial in the highland site. However, estimation of the number of generations based on physiological time showed occurrence of two generations peaking on June 6 and July 10. Lower number of the insect and higher parasitism activity in the highland production site in the April planted trial suggest the potential of conservation practices as an integral component of DBM IPM in the rain-fed brassica production of Ethiopia (Martínez-Castillo *et al.*, 2002).

Higher DBM density in the April planted trial than in the December planted trial at Melkassa unlike that of Holeta was due to differences in meteorological conditions in the two sites. As pointed out above, maximum temperature was significantly lower and rainfall was higher in the April plantings at Holeta resulting in lower population. However, at Melkassa, although differences were not significant between the seasons both in maximum temperature and rainfall; maximum temperature was slightly higher and rainfall was lower in the trial planted in April (Fig. 4.5).

A negative slope of location (dummy variable $L = 0$ for Holeta and 1 for Melkassa) and positive slope of season (dummy variable $S = 0$ first planting and $S = 1$ second season planting) indicates significantly higher level of parasitism at Holeta and in the second season. A negative coefficient of the other two parasitoids, *Oomyzus sokolowskii* and *Apanteles* sp. indicates an inverse relationship between them and *Diadegma* sp. probably due to competition effect. Negative relationship for *Diadegma insulare* (Cresson) (Martínez-Castillo *et al.*, 2002), *D. semiclausum* (Poelking, 1992), *Apanteles* sp. (Morallo- Rejesus and Sayaboc, 1992) with DBM population have been reported. On the other hand Nagarkatti and Jayanth (1982) found a density dependent relationship between *Apanteles plutellae* Kurdjumov and DBM population.

Fig.4. 1 (a) Diamondback moth density per plant; (b) level of parasitism by its parasitoids and (c) minimum and maximum temperature ($^{\circ}\text{C}$) (average of the consecutive sampling periods) and (d) rainfall (mm) and relative humidity (%) (average of the consecutive sampling dates) at Holeta in December 2001 cabbage planted field

Fig. 4.2. (a) Diamondback moth density per plant; (b) level of parasitism by its parasitoids and (c) minimum and maximum temperature ($^{\circ}\text{C}$) (average of the consecutive sampling periods) and (d) rainfall (mm) and relative humidity (%) (average of the consecutive sampling dates) at Holeta in April 2002 cabbage planted field

Fig. 4. 4 (a) Diamondback moth density per plant; (b) level of parasitism by *O. sokolowskii* (c) minimum and maximum temperature ($^{\circ}\text{C}$) (average of the consecutive sampling dates) and (d) rainfall (mm) and relative humidity (%) (average of the consecutive sampling dates) at Melkassa in November 2001 cabbage planted field

Fig. 4.5. (a) Diamondback moth density per plant; (b) level of parasitism by parasitoids (c) minimum and maximum temperature ($^{\circ}\text{C}$) (average of the consecutive sampling dates) and (d) rainfall (mm) and relative humidity (%) (average of the consecutive sampling dates) at Melkassa in April 2002 cabbage planted field.

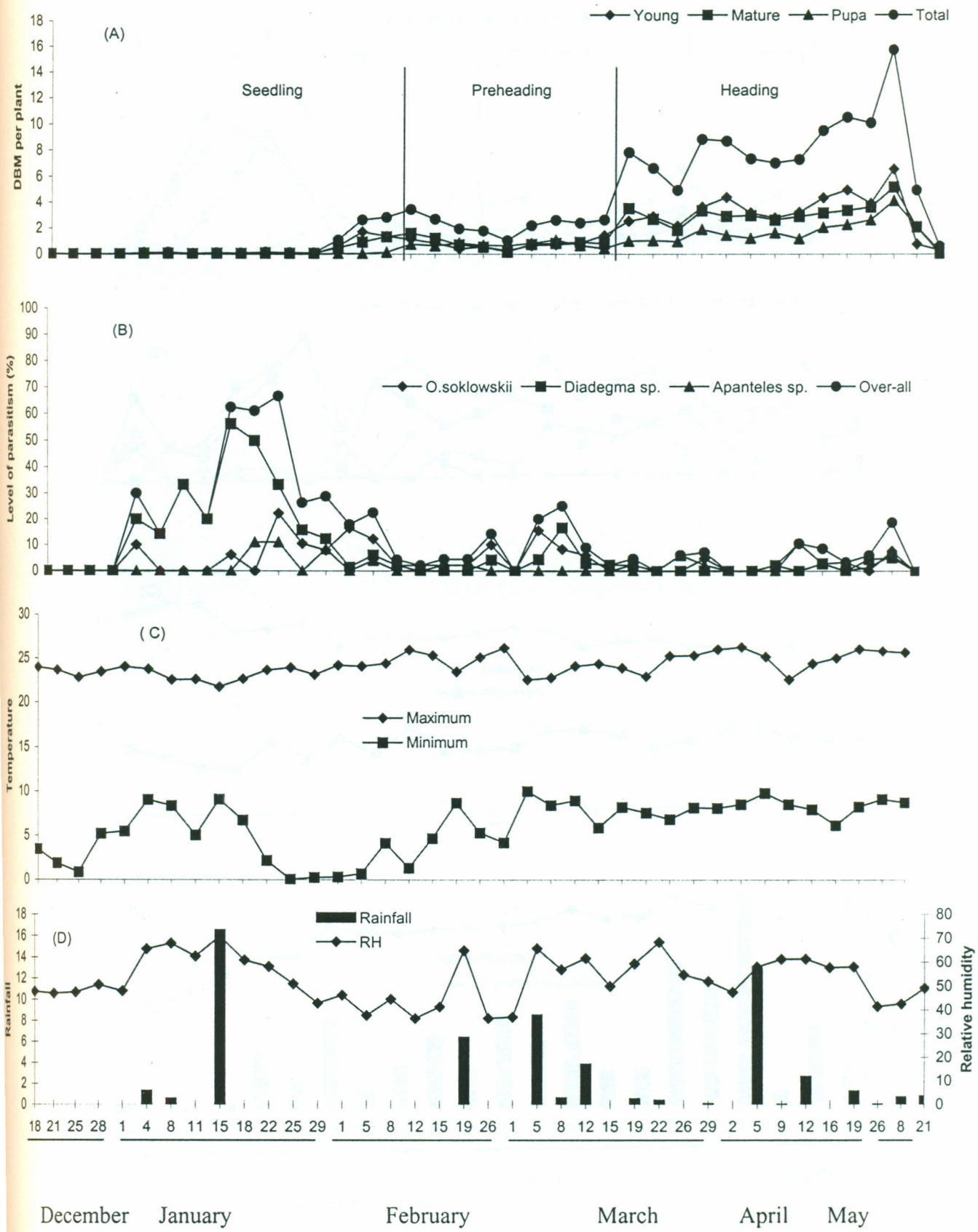


Fig. 4.1 (See page 65 for the caption)

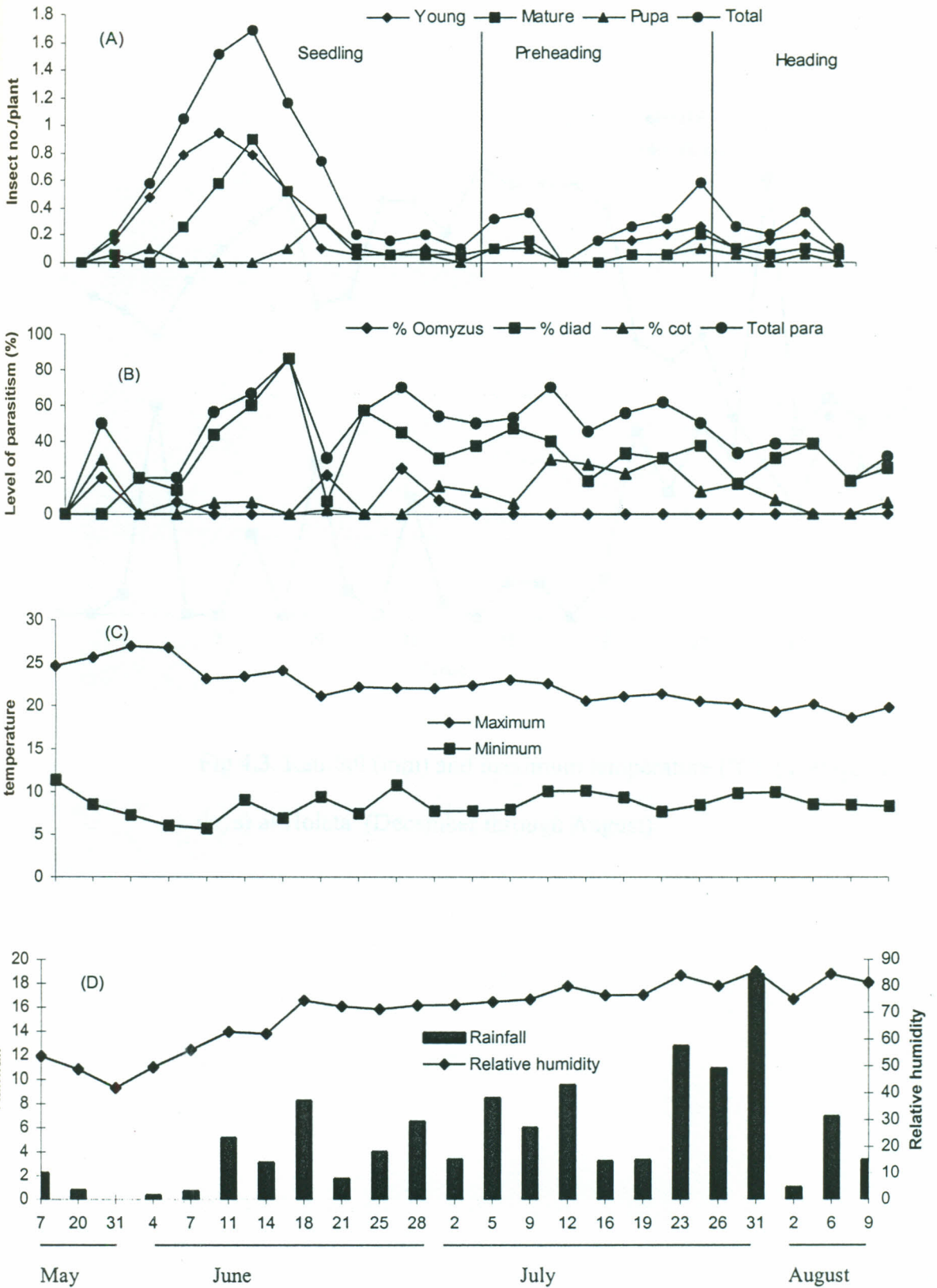


Fig. 4.2 (See page 65 for the caption)

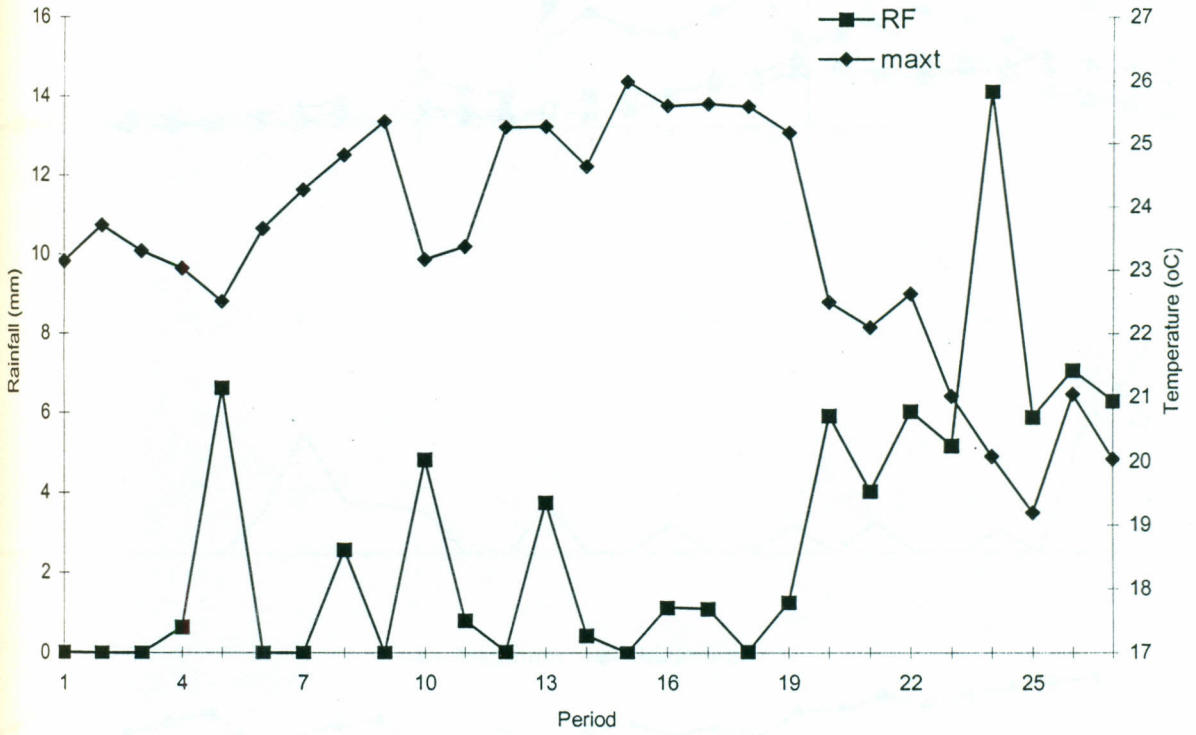


Fig 4.3. Rainfall (mm) and maximum temperature ($^{\circ}\text{C}$) (average of 10 days) at Holeta (December through August)

◆ Young ■ mature ▲ pupa ● total

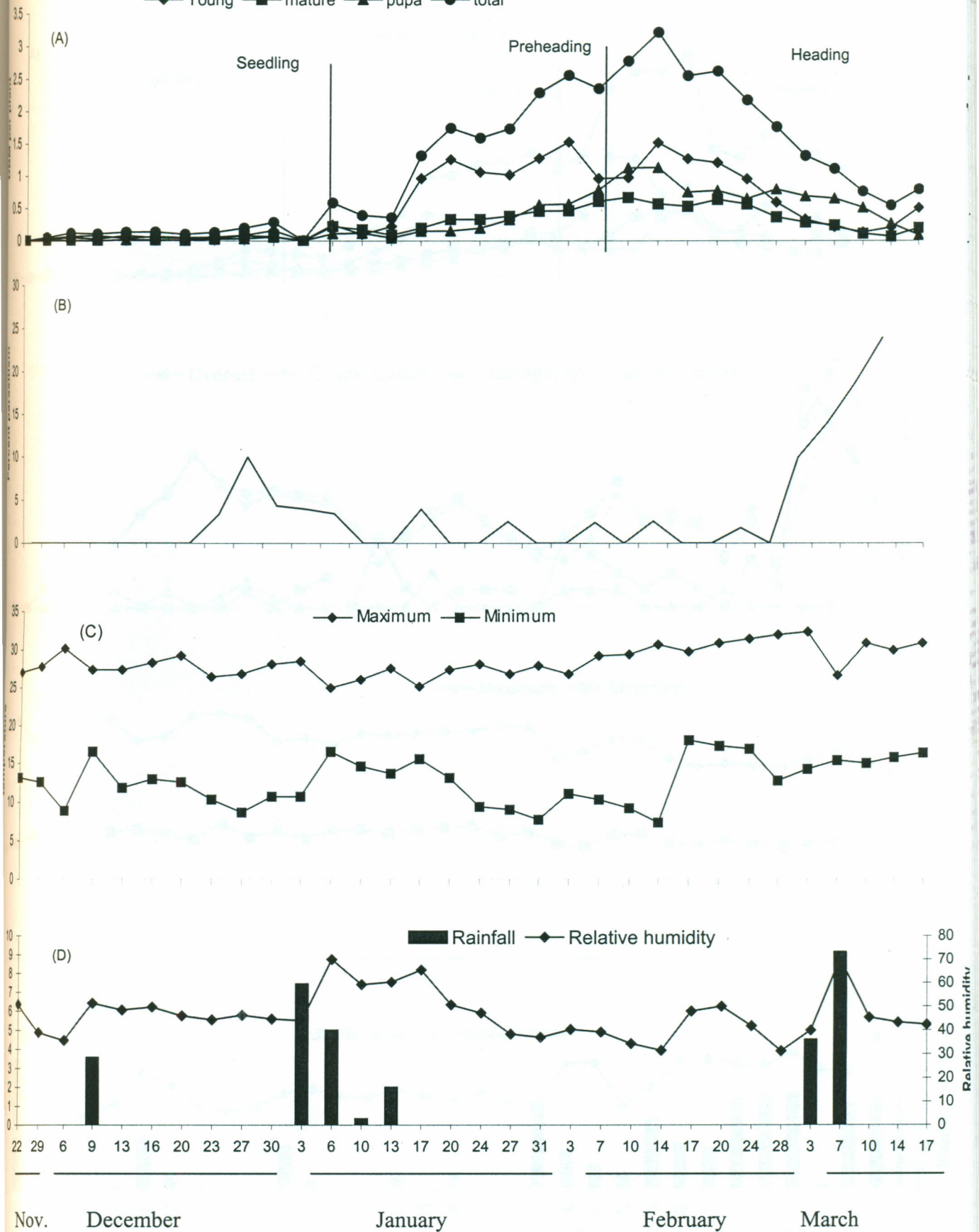
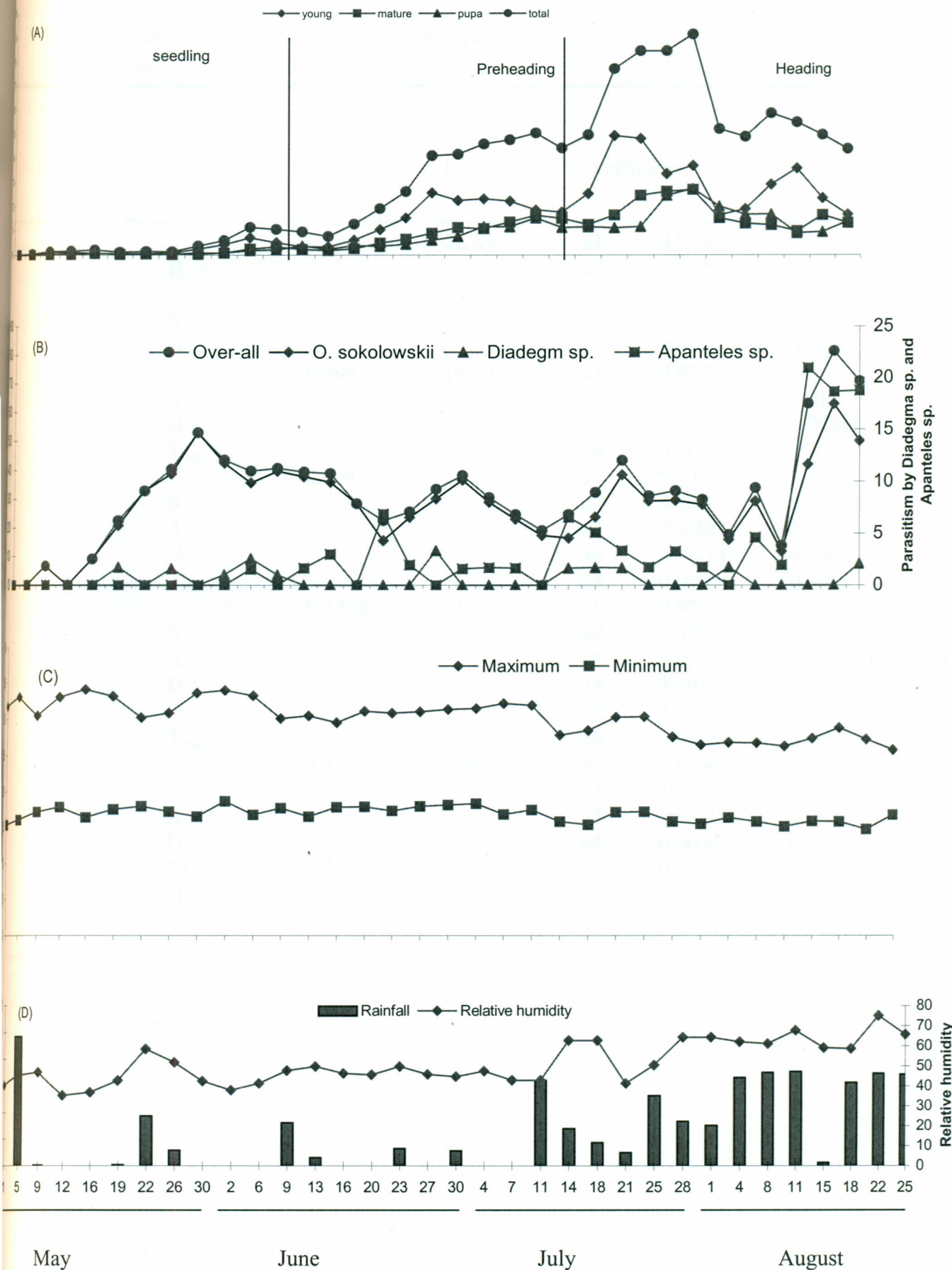


Fig 4.4. (See page 65 for the caption)



May June July August
 Fig. 4.5 (See page 65 for the caption)

Table 4.1. Summary of the stepwise linear regression analysis of parasitism level by *Apanteles* sp.

steps	variable	R ²	F	coefficient	Standard error		
1	H	0.194	28.65	a1	-0.088		
				a2	0.002	0.0004	
2	S	0.252	19.88	a1	-0.082		
				a2	0.035	0.115	
3	H	0.296	16.39	a3	-0.002	0.0007	
				a1	-0.032		
				a2	0.052	0.013	
4	S	0.3045	12.70	a3	0.0015	0.0001	
				T _{min}	a4	-0.003	0.0013
				A	a1	-0.038	
				H	a2	0.054	0.013
5	T _{min}	0.3276	11.20	a3	0.0002	0.0002	
				A	a4	0.0013	0.0005
				D	a5	-0.0036	0.0013
				H	a1	-0.036	
				T _{min}	a2	0.056	0.013
				A	a3	0.0005	0.0002
6	D	0.3357	9.60	a4	-0.005	0.003	
				H	a5	0.0011	0.0005
				T _{min}	a6	-0.0038	0.0013
				S	a1	-0.0346	
				A	a2	0.062	0.014
				D	a3	0.0005	0.0002
				P1	a4	-0.0056	0.003
H	a5	-0.050	0.042				
T _{min}	a6	0.0013	0.0005				
				a7	-0.005	0.002	

Table 4.2. Summary of the stepwise linear regression analysis of parasitism level by *Oomyzus sokolowskii*

steps	variable	R ²	F	coefficient	Standard error	
1	T _{min}	0.26	41.47	a1	-0.068	
				a2	0.012	0.002
2	S T _{min}	0.35	31.79	a1	-0.065	
				a2	0.098	0.240
3	S P1 T _{min}	0.41	26.83	a3	0.011	0.0025
				a1	-0.002	
				a2	0.142	0.026
4	P1 T _{min} S D	0.42	20.83	a3	-0.246	0.073
				a4	0.006	0.003
				a1	-0.038	
				a2	0.140	0.027
5	P1 T _{min} S D P1 T _{max} T _{min}	0.43	17.28	a3	0.005	0.004
				a4	-0.212	0.077
				a5	0.007	0.003
				a1	-0.171	
				a2	0.142	0.264
				a3	0.006	0.004
6	P1 T _{min} S D P1 H T _{max} T _{min}	0.44	14.62	a4	-0.163	0.083
				a5	0.006	0.004
				a6	0.004	0.004
				a1	-0.494	
				a2	0.013	0.023
				a3	0.006	0.004
				a4	-0.175	0.084
7	H T _{max} T _{min} S D P1 H T _{max}	0.44	17.69	a5	0.002	0.002
				a6	0.016	0.010
				a7	-0.0004	0.005
				a1	-0.474	
				a2	0.133	0.028
				a3	0.006	0.004
				a4	-0.173	0.081
a5	0.002	0.001				
a6	0.015	0.005				

Table 4.3. Summary of the stepwise linear regression analysis of parasitism level by *Diadegma* sp.

steps	variable	R ²	F	coefficient	Standard error	
1	T _{max}	0.32	55.91	a1	0.812	
				a2	-0.027	0.004
2	S	0.42	42.84	a1	0.80	
				a2	0.11	0.03
3	T _{max}	0.49	37.05	a3	-0.03	0.003
				a1	0.83	
4	S	0.54	33.53	a2	0.105	0.024
				a3	-0.015	0.004
5	D	0.55	28.04	a4	-0.028	0.003
				a1	-0.038	
6	L	0.56	23.57	a2	-0.14	0.039
				a3	0.109	0.023
7	S	0.55	28.39	a4	-0.018	0.004
				a5	-0.014	0.005
8	T _{max}	0.56	23.57	a1	0.481	
				a2	-0.125	0.038
9	L	0.55	28.04	a3	0.133	0.026
				a4	-0.016	0.004
10	S	0.56	23.57	a5	-0.181	0.099
				a6	-0.0122	0.005
11	D	0.55	28.04	a1	0.211	
				a2	-0.145	0.043
12	P2	0.56	23.57	a3	0.116	0.031
				a4	-0.016	0.004
13	H	0.55	28.04	a5	-0.191	0.099
				a6	0.002	0.002
14	T _{max}	0.56	23.57	a7	-0.005	0.009
				a1	0.052	
15	L	0.55	28.04	a2	-0.163	0.027
				a3	0.109	0.028
16	S	0.56	23.57	a4	-0.016	0.004
				a5	-0.200	0.097
17	D	0.55	28.04	a6	0.003	0.001
				a6	0.003	0.001

Table 4.3 cont'd

steps	variable	R ²	F		coefficient	Standard error
8		0.56	23.93	a1	0.061	
	L			a2	-0.117	0.049
	S			a3	0.122	0.031
	D			a4	-0.015	0.004
	P2			a5	-0.190	0.098
	H			a6	0.003	0.001
	T _{min}			a7	-0.006	0.005
9		0.56	20.71	a1	0.055	
	L			a2	-0.116	0.049
	S			a3	0.132	0.032
	D			a4	-0.016	0.004
	P2			a5	-0.180	0.098
	P3			a6	-0.210	0.194
	H			a7	0.003	0.001
	T _{min}			a8	-0.007	0.005

Table 4.4. Regression statistics for parasitism level by *Diadegma* sp.

Variable	b _j	t-value	Significance level	mean
L	-0.116	-2.35	<0.05	0.52
S	0.132	4.13	<0.05	0.45
D	-0.016	-3.99	<0.05	2.30
P2	-0.180	-1.83		0.11
P3	-0.210	-1.08		0.034
H	0.003	2.95	<0.05	53.91
T _{min}	-0.007	-1.30		11.06
N	121			
Mean	0.092			
R ²	0.56			
Intercept (standard error)	0.055 (0.06)			

Table 4.5. Regression statistics for parasitism level by *Oomyzus sokolowskii*

Variable	bj	t-value	Significance level	mean
S	0.132	4.81	<0.05	0.448
D	0.006	1.62	<0.05	2.301
Pl	-0.173	-2.13	<0.05	0.092
H	0.002	1.52		53.91
T _{max}	0.015	2.79	<0.05	26.52
N	121			
Mean	0.11			
R ²	0.43			
Intercept (standard error)	-0.47 (0.21)			

Table 4.6. Regression statistics for parasitism level by *Apanteles* sp.

Variable	bj	t-value	Significance level	mean
S	0.062	4.50	<0.05	0.449
A	0.0005	2.19	<0.05	74.81
D	-0.006	-2.19	<0.05	2.30
Pl	-0.050	-1.18		0.09
H	0.001	2.58	<0.05	53.91
T _{min}	-0.005	-3.20	<0.05	11.06
N	121			
Mean	0.03			
R ²	0.34			
Intercept (standard error)	-0.03 (0.03)			

Table 4.7. Probabilities of Diamondback moth parasitism by its insect parasitoids, *O. sokolowskii*, *Diadegma* sp. and *Apanteles* sp. in the presence (apparent parasitism - bold) and absence (potential parasitism - normal) of other parasitoids at Holeta and Melkassa in December (SI) and April (SII) planted cabbage.

Location and season	Parasitoid species			Total
	<i>O. sokolowskii</i>	<i>Diadegma</i> sp.	<i>Apanteles</i> sp.	
Holeta SI	0.059	0.048	0.029	0.136
	0.061	0.049	0.030	0.140
Holeta SII	0.080	0.305	0.078	0.463
	0.086	0.347	0.084	0.517
Melkassa SI	0.044	0.000	0.002	0.046
	0.044	0.000	0.002	0.046
Melkassa SII	0.229	0.005	0.022	0.256
	0.235	0.005	0.023	0.263

CHAPTER 5

5.0. Geostatistical analysis of the spatial distribution of Diamondback Moth, *Plutella xylostella* (Lepidoptera: Plutellidae)

5.1 INTRODUCTION

Diamondback Moth (DBM), *Plutella xylostella* L.

(Lepidoptera: Plutellidae) is the most destructive insect pest of brassicas with an estimated management cost of USD 1 billion annually (Talekar and Shelton, 1993). Although such a large amount of money is spent for the control of the pest, the level of control to offset the expense is low because of the physiological peculiarity of the insect to acquire resistance to insecticides very quickly (Chen and Sun, 1986). This has necessitated search for alternatives including cultural control, biological control and use of pheromones and growth regulators, (Talekar and Shelton, 1993; Schroeder *et al.*, 2000). Success of biological control of DBM has been reported by several workers (Alam, 1992; Mustata, 1992; Ooi, 1992; Talekar, *et al.*, 1992). An important component in the promotion of biological control and IPM in general is knowledge on the dynamics of the pest both in space and time (Nestle and Klein, 1995; Brenner, *et al.*, 1998). Although DBM is a well studied insect pest, information on its dynamics, especially spatial dynamics, is scarce. Available relevant information are on its long distance wind assisted migration (Chu, 1986) and local dispersal (Mo *et al.*, 2003).

Geostatistics has been used for understanding spatial processes in insect ecology (Schotzko and O'keeffe, 1989; Arbogast *et al.*, 1998;

Brenner *et al.*, 1998; Odulaja *et al.*, 2001; Sciarretta *et al.*, 2001, Tobin and Pitts, 2002). This study was conducted to assess the spatial variation in the density of adult DBM at local (single field) and regional (brassica production areas) scales in the Ethiopian brassica fields as part of an IPM program.

5.2 MATERIALS AND METHODS

5.2.1 Study Area

The study was conducted in a single head cabbage (*Brassica oleracea* var. *capitata*) field of Arsi highland (7° 01' N, 38° 53' E) (Fig. 5.1) and, at regional scale, in brassica producing areas of Wonji, located in central Rift Valley region of Ethiopia (8° 27' N, 39° 13' E) (Fig. 5.2).

5.2.1.1 Single field study

Observations on the distribution of adult male DBM was made by regularly placing pheromone traps of delta type with a rubber lure impregnated with a synthetic sexual blend containing Z 11-16:Ald, Z11-16:AC and Z11-16:OH, at a space of c 20 m from each other. The pheromone was obtained from pherobank (Wageningen, the Netherlands). One of the traps was kept at the centre of the cabbage field with the remaining distributed equidistant in the four directions, north, east, south and west. Hence the minimum and the maximum distances between two traps were 20 and 80 m respectively with a mean of 37.13. Traps were placed 30 cm above the crop surface. Three

of the eleven traps were located within the cabbage field with the remaining distributed around either in open field or non-brassica crop (Fig. 5.1). The study was conducted for a period of one and half months (November 5 through December 20, 2001). Count of catches was made twice a week, at three to four days interval, and there were a total of eleven counts. Weather data (minimum temperature, maximum temperature and relative humidity) were recorded from a data logger (Hobo) fitted with temperature and relative humidity probes and installed on the experimental field.

5.2.1.2. Regional Scale study

Nineteen traps of the same type as described for single field study were randomly distributed in brassica producing areas of Wonji in central rift valley region (Fig. 5.2). The minimum distance between any two traps was 96.7 m and the maximum was 16,020 m with a mean of 7618.6 m. Traps were placed either in brassica fields, non-brassica crops fields or open field as shown in Fig 5.2. Counts were made weekly for a period of 6 weeks between May to July 2002.

5.2.2 Data Analysis

5.2.2.1 Spatial analysis

Spatial analysis for the single field study was carried out using Surfer version 8.02 (Golden software, Golden, CO) as described by Sciarretta *et al.* (2001), with x,y representing the coordinates, and z the trap counts. The interpolation algorithm used is linear kriging with zero nugget.

At regional scale, trap catches were square root transformed ($\sqrt{x + 0.5}$) and subjected to Principal Component Analysis (PCA) to delimit areas with similar trap catches (Odulaja *et al.*, 2001). Because of missing data, only counts from the 2nd to the 5th week were considered for the PCA. Trap number 6 was not included in the analysis because data on the second week were not available.

5.2.2.2. Evaluation of environmental variables

Aggregation index for each count was calculated using SADIE software (Perry *et al.*, 1996), at both local and regional scale. The calculation is based on the effort needed by single individuals to reach maximum crowding compared with that needed to reach maximum randomness.

Influence of weather data on aggregation pattern was analyzed using stepwise regression analysis of the SAS software (SAS Institute, 1999). To allow for curvilinear effects (Odulaja *et al.*, 2001), the squares of the variables were included as independent variables.

5.2.2.3 Assessment of distance

Effect of distance was assessed by regressing Pearson correlation coefficient of trap catches between traps on their distance, at both local and regional scale. Trap counts were square root transformed ($\sqrt{x + 0.5}$) to stabilize variance (Odulaja *et al.*, 2001), before correlation analysis was carried out.

5.2.2.4 *Autocorrelation analysis*

Spatial autocorrelation was measured by Moran's I index (Cliff and Ord, 1981) which was computed by the SAAP software (Exeter Software, Setauket, NY). Autocorrelograms were constructed for each sampling period of both the single field study and regional scale study. Equal frequency correlograms were built, using an equal number of point pairs for each distance class.

5.3 *Distribution of Field-measured variables*

5.3 RESULTS

5.3.1 Single field study

5.3.1.1 *Spatial analysis*

Figure 5.3 shows the distribution of trap catches for the 11 sampling periods. High level of catches was recorded from traps installed in the field throughout the study period. Patterns for variation in the three central traps was similar until the final sampling period (December 20), which was highest in the central trap. Counts were low in all border traps throughout the study period. The relationship between correlation coefficients of trap catches and distance between traps was not significant ($P > 0.05$), with an R square very low (Table 5.2).

5.3.1.2 *Autocorrelation analysis*

The autocorrelation analysis resulted in 5 class intervals at 20, 28, 40, 44 and 80 m. Moran's I index shows significant autocorrelation on the 1st (5 November), 4th (16 November) 7th (26 November) and 8th (30 November) survey. Correlograms (Fig. 5.7) indicate

significant negative values ($P < 0.05$) at 28 m distance class and significant positive value at 40 m distance on the sampling dates mentioned. Pattern of autocorrelations, in all sampling periods, had similar trend: it decreased in the first lag distance to reach negative values, then increased in the second lag distance to positive values and remained close to zero in the last lag distances.

5.3.1.3 Evaluation of Environmental variables

Only maximum temperature showed significant influence ($P < 0.05$) on the aggregation index (Table 5.3). Fig 5.5(a) shows that aggregation index increased with increasing maximum temperature. It accounted for 41% of the variation in the aggregation index. Maximum temperature during the study period fluctuated between 19 °C and 24 °C and minimum temperature between 3 °C and 7 °C.

5.3.2 Regional scale

5.3.2.1 Spatial analysis

Figure 5.4 shows similarity of the trapping positions. The first principal component explained 85 % of the variance and the first two together explained 97 %. Groupings of traps appeared to relate with host crop type. Table 5.1 shows the crop type, growth stage and surrounding crop types in relation to traps position. Traps 1 to 9 were located in the main brassica growing areas with the exception of trap 4, which was installed in the nearby sugarcane plantation. All these traps appeared at the top on the principal axes space (Fig 5.4). The rest of the traps, positioned either on open field or non-brassica crops with the

exception of trap number 15, appeared on the bottom area. The groupings, particularly in the non-brassica producing location (trap number 10 to 19), indicate a strong relationship between patterns in trap catches between traps in close geographical proximity. Similar to the single field study, no correlation was observed between correlation coefficients of trap catches and distance between traps (Table 5.2).

5.3.2.2 Autocorrelation analysis

The autocorrelation analysis resulted into 7 class intervals at 778, 1,760, 2,971, 11,487, 13,006 and 13,666 and 16,026 m (Fig. 5.6). In the short distance class (intervals 1 to 3), values are positively significant, indicating similarity of captures. The medium distance class (interval 4) was not significant and near to 0, indicating absence of autocorrelation. For class intervals 5 to 7, correlation was negatively significant indicating that catches for higher distances were different.

5.3.2.3. Evaluation of Environmental variables

Only rainfall showed significant influence ($P < 0.05$) on the aggregation index (Table 5.3). Fig 5.5(b) shows that aggregation index increased with decreasing rainfall. It accounted for 60% of the variation in the aggregation index. Mean daily rainfall during the study period fluctuated between 0 and 3.5 mm.

5.4 DISCUSSION

At a local scale, kriging shows that adult DBM males were higher in the centre of the field. Correlogram also showed negative significant values at 28 m, distance where most of inside and outside traps were compared, indicating a discontinuity in trap catches corresponding to the edge of the field, that is a border effect (Sciarretta *et al.*, 2001). From local dispersal study using mark recapture technique, Mo *et al.* (2003) showed that in the active growing season, DBM population does not move out of cabbage fields. Although the location of traps at the border was within the local dispersal range (Mo *et al.*, 2003), lower DBM number in traps installed out of the field could be due to this behaviour of the insect not to move out from its host field. Caprio and Tabashnik (1992) also noted lower DBM numbers in traps located in the fallow field surrounding cabbage fields. According to their observations, the proportion of catches in the surrounding field was 7.8 % of those recorded within field, which is in agreement with the observation in this study.

On the other hand, significant positive values at 40 m, distance where most of the outside traps were compared among them, indicates similarity of the trap catches located out of the border.

The observation at a regional scale in terms of DBM association with its host is similar to the single field study. The DBM captures were higher within the cabbage field of the brassica crops producing locality (traps 1 to 9). Traps 1 to 9 were located in vegetable crops producing locality where crucifers are among the most important ones. On the other hand, DBM captures were lower in traps located in

open field and non-brassica producing locality (traps 10 to 19). Although trap number 15 was installed in cabbage field in the non-producing locality, DBM captures were as low as those found in traps installed in the surrounding. This indicates influence of geographic location or cropping system on the distribution of DBM, although the insect is known to migrate longer distances (Chu, 1986). On the other hand, Mo *et al.* (2003) observed that DBM moths flew close to the ground and below the plant canopy, suggesting trivial movement and limited dispersal range. The higher DBM density in cabbage field of crucifer producing areas, compared to cabbage field of non producing areas observed in this study, could be due to this limited dispersal range. The practical implication of this observation is that in new areas where crucifers were not cultivated, damage inflicted by DBM can be minimal. This, however, can be influenced by the presence of wild crucifers that can sustain development and reproduction of DBM. Insecticidal control in such areas can also give effective control because of the limited migration of pesticide resistance population. Significant positive values at short distance class interval, within a range of 1,760 m for outside-outside comparison and 2,971 m for inside-inside comparison, indicates similarity of trap catches in those lag distances. At the 4th class interval, the autocorrelation tends to zero and no correlation was observed. The interval between 5 and 7 represent most of the traps compared between brassica and non-brassica fields. Autocorrelation decreased linearly in this class interval indicating a negative correlation of trap catches between traps inside brassica fields and non-brassica fields. Principal component analysis

also confirmed similarity of trap catches among traps within brassica fields and non-brassica fields indicating dissimilarity in trap catches between traps of brassica and non brassica fields. Absence of correlation between correlation of trap catches and distance could partly be due to this dissimilarity between sites. This was similar with the observation at a local scale; no correlation was observed between correlation of trap catches and distance because of dissimilarity of trap catches between inside and outside which masked the effect of distance.

At local and regional scale, environmental variables appear to influence in different manner the aggregation index. This could be due to climatic difference in the two areas. Kofele, where the single field study was carried out, located in a highland brassica production sites (alt. 2,450 m a.s.l.), is a cool and wet area; Wonji, where the regional scale study was carried out, is located in the low land brassica producing region (alt. 1,550 m a.s.l.) and the environment is hot and dry.

As pointed out, at Kofele maximum temperature influenced the aggregation index positively and at Wonji, it was negatively influenced by rainfall. Temperature has been reported to have influence on the initiation of flight activity and flight duration (Goodwin and Danthanarayana, 1984; Shirai, 1991). The optimal flight temperature range of DBM is 18 to 28 °C (Shirai, 1991). The maximum temperature at Kofele fluctuated between these temperature ranges. This may explain the dependency of aggregation on the maximum temperature at Kofele. On the other hand, in the regional scale study,

the maximum temperature is beyond the optimum, ranging between 28.5 °C and 33.5 °C, suggesting the reason for the absence of correlation with aggregation index. At this scale, rainfall resulted to be the only environmental variable with significant effect on aggregation index.

Table 5.1 Location details of traps installed at Wonji

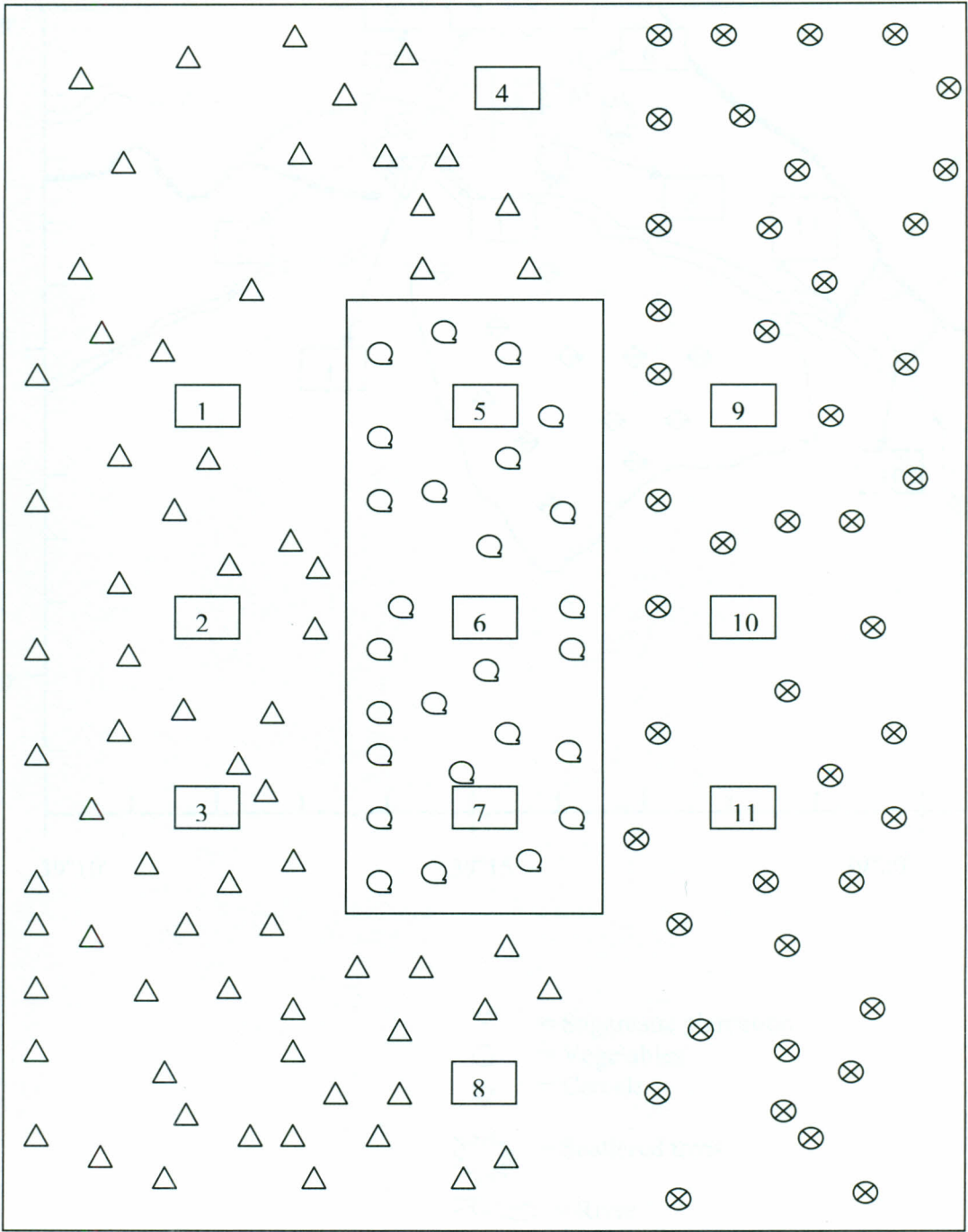
Trap no.	Location		Crop fields	Crop stage	Surrounding plant
	Latitude	Longitude			
1.	08°28'08''N	039°13'21''E	Cabbage	Seedling	Kale, tomato, cabbage, tree
2.	08°28'15''N	039°13'21''E	Cabbage	Seedling	Ttomato, cabbage
3.	08°28'20''N	039°13'30''E	Tomato	Flower	Tree (Eucalyptus)
4.	08°27'05''N	039°13'65''E	Sugar cane	Seedling	Sugarcane, banana, tree
5.	08°28'38''N	039°14'17''E	Maize	Seedling	Maize, Road
6.	08°28'65''N	039°14'29''E	Tomato	Early fruiting	Pepper, tomato
7.	08°29'10''N	039°13'70''E	Cabbage	Seedling	Pepper, cabbage
8.	08°28'80''N	039°13'33''E	Cabbage	Preheading	Onion, maize
9.	08°28'99''N	039°13'60''E	Maize	Seedling	Cabbage, tomato, maize
10.	08°24'00''N	039°19'87''E	Onion	Seedling	Onion
11.	08°23'99''N	039°20'09''E	Open field	—	Beans
12.	08°24'96''N	039°19'75''E	Open field	—	Open field
13.	08°24'58''N	039°19'38''E	Onion	—	Onion
14.	08°24'63''N	039°20'00''E	Open field	—	Open field
15.	08°24'89''N	039°19'59''E	Cabbage	Seedling	Tomato
16.	08°25'00''N	039°19'63''E	Open field	—	Open field
17.	08°24'90''N	039°19'62''E	Open field	—	Open field
18.	08°24'85''N	039°19'67''E	Open field	—	Open field
19.	08°24'84''N	039°19'58''E	Open field	—	Open field

Table 5.2 . Linear regressions of correlation coefficients between traps on distance between traps (m) (in parentheses are reported standard errors of the corresponding parameters).

Location	Intercept	Slope	R ²
Kofele	0.201 (0.124)	-3.0 X 10 ⁻³ (3.0 X 10 ⁻³)	0.02
Wonji	0.0331 (0.561)	(1.94 X 10 ⁻⁶) (5.87 X 10 ⁻⁶)	0.0006

Table 5.3 Aggregation index described by maximum temperature (°C) at Kofele, and rainfall (mm) at Wonji, as independent environmental variables with significant effect (slope is different from 0 with P<0.05).

Location	Variable	Intercept	Slope	R ²
Kofele	Max. Temperature (°C)	-0.65 (0.76)	0.09 (0.037)	0.407
Wonji	Rainfall (mm)	2.80 (0.17)	-0.306 (0.11)	0.603



- Cabbage
- ⊗ Enset (*Enset ventricosum* L.)
- △ Grass field

Fig. 5.1. Map of the study area at Kofele showing the 11 trapping positions (20 m X30m)

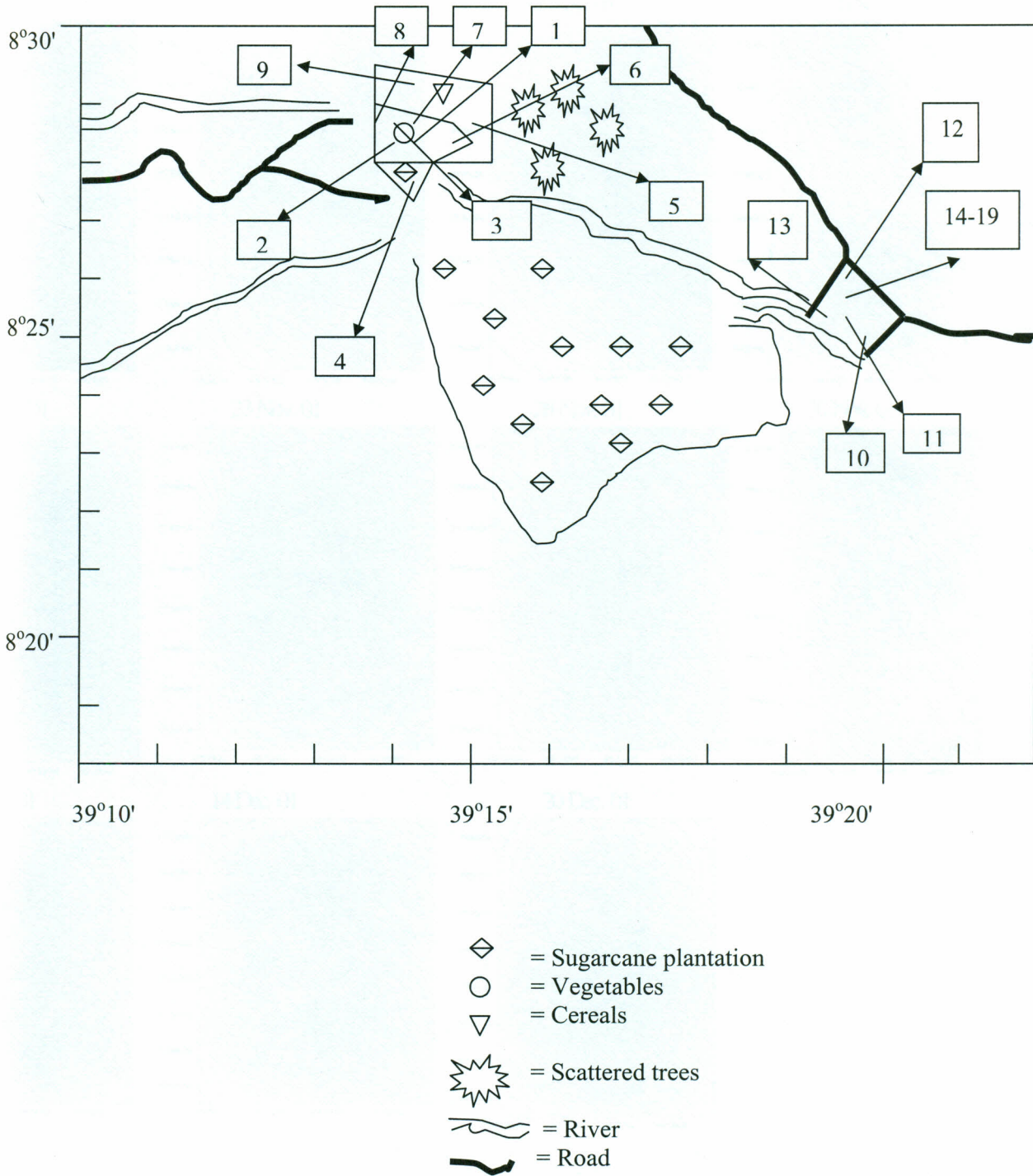


Fig. 5.2. Map of the study area at Wonji showing the 19 trapping positions

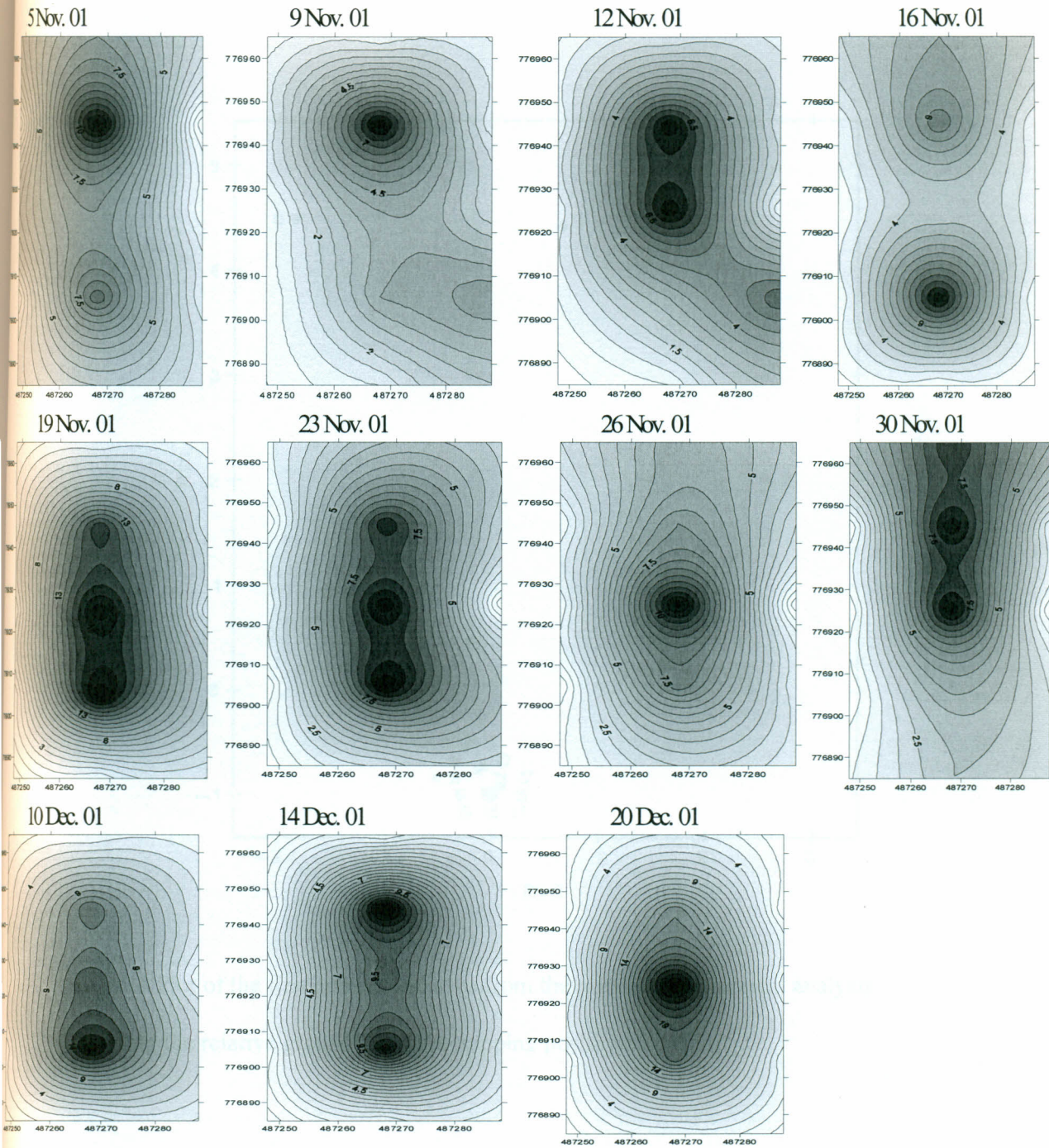


Fig . 5.3 Contour lines showing trap catch distribution of male *Plutella xylostella* at Kofele for the 11 sampling periods in one single cabbage field.

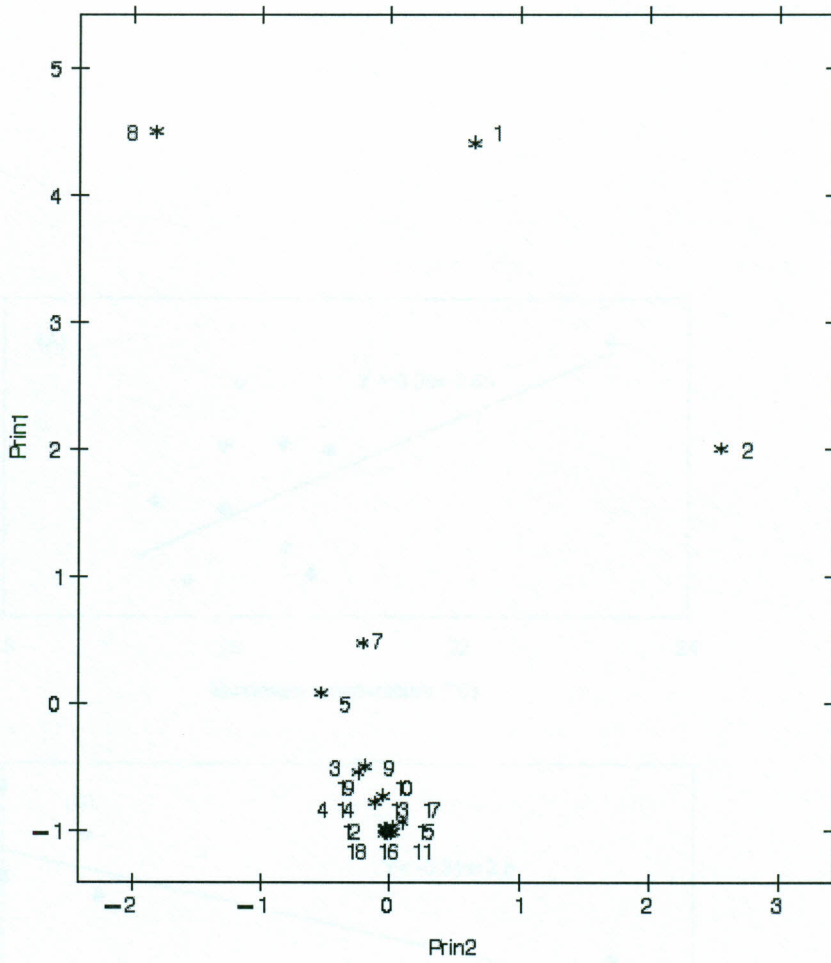


Fig. 5.4 Plot of the first and second axes from the principal component analysis showing the relative groupings of the trapping positions at Wonji

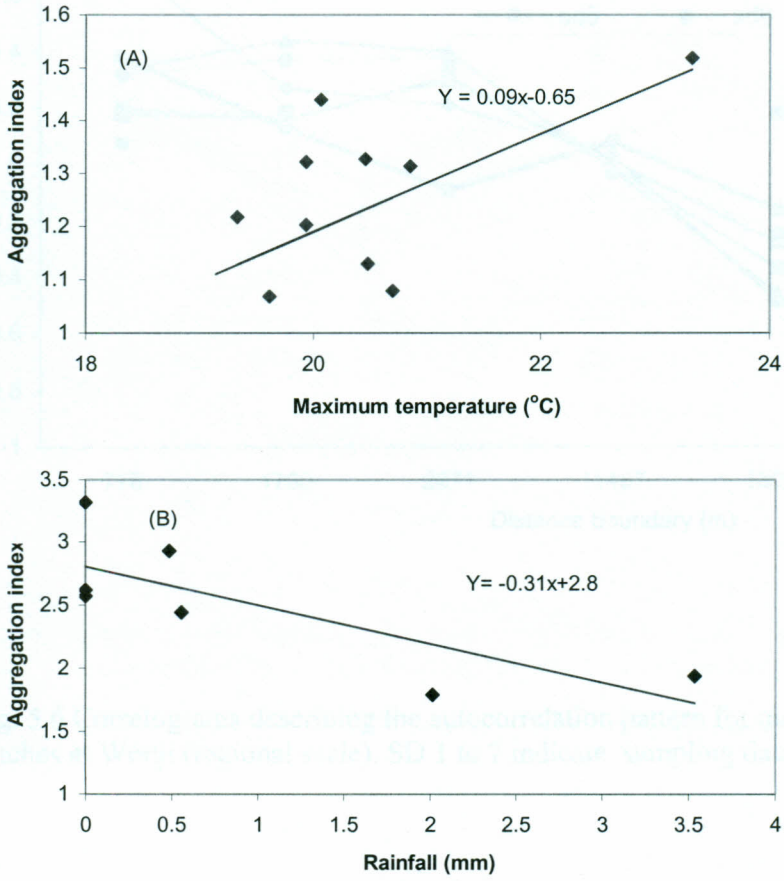


Fig. 5.5. Relationship between aggregation index and environmental variables with a significant effect ($P < 0.05$), at Kofele (A) and Wonji (B).

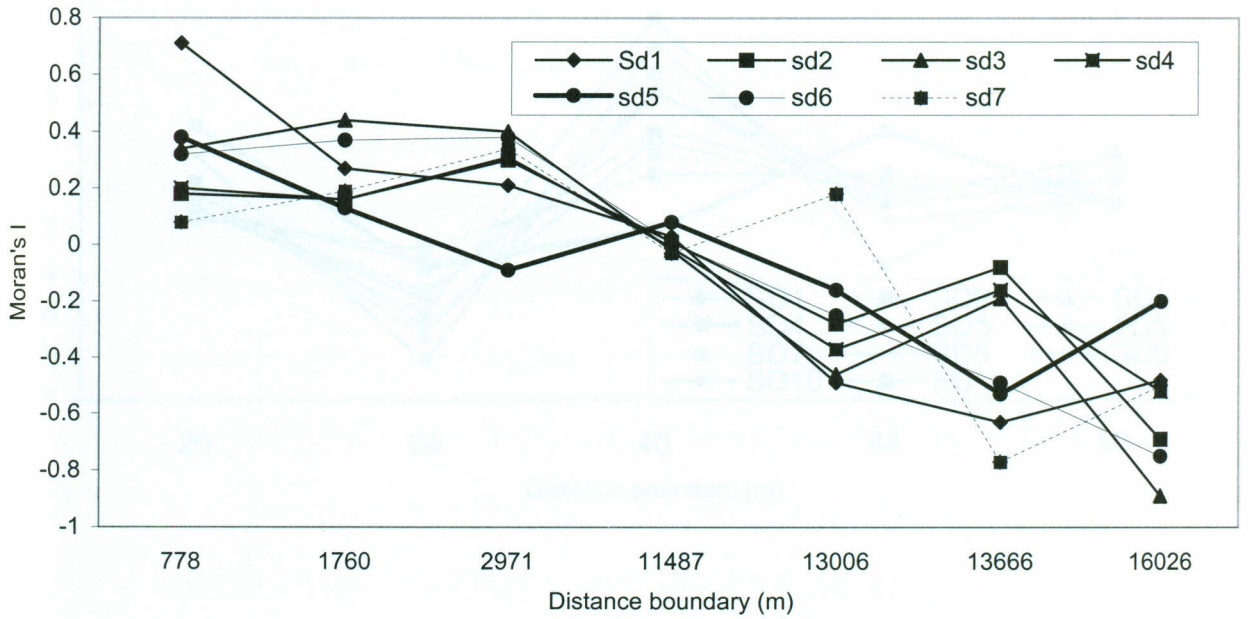


Fig. 5.6 Correlograms describing the autocorrelation pattern for male DBM trap catches at Wonji (regional scale). SD 1 to 7 indicate sampling dates

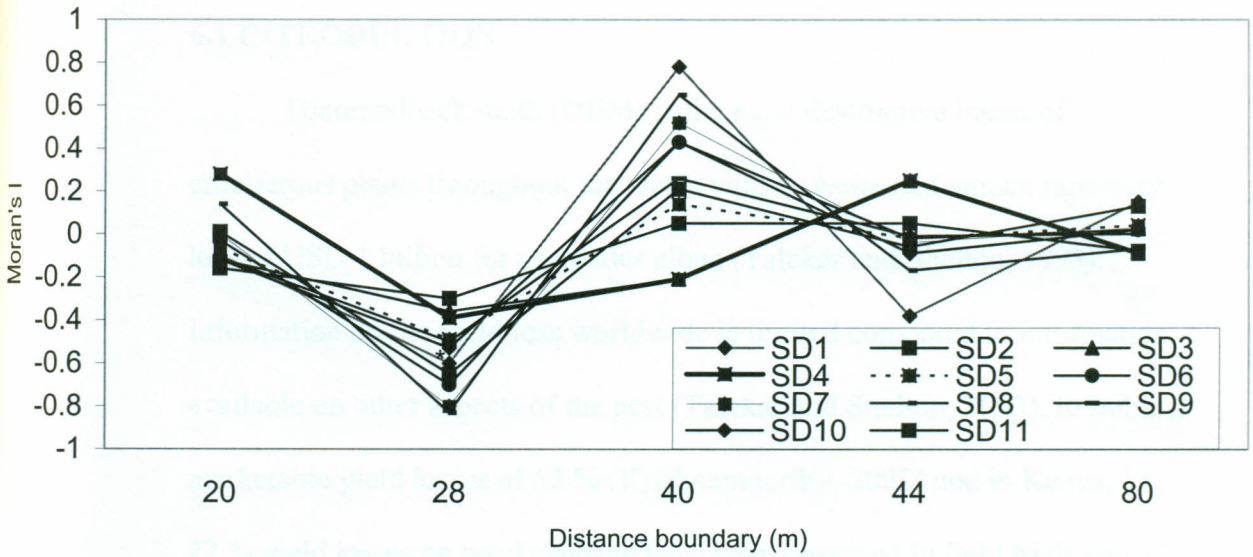


Fig. 5.7 Correlograms describing the autocorrelation pattern for male DBM trap catches at Kofele (single field study). SD 1 to 11 indicate sampling dates.

CHAPTER 6

6.0 Yield loss of cabbage caused by the Diamondback moth, *Plutella xylostella* L. (Lepidoptera: Plutellidae)

6.1 INTRODUCTION

Diamondback moth (DBM) is the most destructive insect of cruciferous plants throughout the world with an estimated annual monetary loss of USD 1 billion for pesticides alone (Talekar and Shelton, 1993). Information on the yield loss worldwide is limited compared to information available on other aspects of the pest (Talekar and Shelton, 1993). In India, a marketable yield losses of 52 % (Krishnamoorthy, 2002) and in Kenya, 11 - 22 % yield losses on head cabbage have been observed in field trials and 37 % losses were established from farmers' interviews (Macharia, personal comm.).

The insect inflicts heavy damage in brassica , cabbage in particular in central Rift Valley region of Ethiopia. Complete crop failure is common in seasons of heavy infestation. However, no data was available in yield loss caused by DBM in Ethiopia because of the absence of experimental work in the past. This experiment was conducted to estimate yield loss due to DBM on head cabbage and critical period of infestation under natural infestation.

6.2 MATERIALS AND METHODS

The experiment was conducted during the cold dry season (November planting) and hot dry season (April planting) between 2001 and 2002 at the Melkassa center of the Ethiopian Agricultural Research Organization

(EARO). The most popular head cabbage variety, Copenhagen Market, was used. Seeding and (transplanting) was made on 30 September 2001(20 November 2001) and 10 March 2002 (19 April 2002) respectively. Cultural practices (fertilization, irrigation, weeding etc.) were carried out as per recommendation of the horticulture division of the research center. Plot size was eight rows of 4 m long each with a spacing of 40 cm between plants and 60 cm between rows. A Randomized Complete Block Design with nine treatments replicated 4 times was used. Each replicate had two untreated plots. Treatments were a microbial product *Bacillus thuringiensis* (*Bt*) var. *kurstaki* and a pyrethroid insecticide, lambda cyhalothrin (Karate) applied at different growth stages, seedling, pre-heading, heading and throughout the crop-growing period in addition to the untreated check. To compare the two pesticides and their interaction with the treatments, a factorial RCBD was used by excluding the untreated plot. *Bt* was applied at the rate of 0.5 kg per ha and Karate (5 EC) at 320 ml per ha. Dates of pesticides application for the different treatments for the two experimental seasons are shown in Table 6.1.

Prior to pesticide application on each date, five randomly selected plants from border rows of each plot were examined for young larvae, mature larvae and pupae of DBM and records were made. At the same time, intensity of aphid (*Brevicoryne brassicae*) was estimated based on aphid colonies present (0 – no aphid, 1 – one colony, 2 – two colonies; and 3 - three and above colonies). DBM Feeding/damage score was taken on a 1 to 5 scale (1 = no damage, 2 = up to 25 per cent damage, 3 = 25 to 50 per cent damage, 4 =

50 to 75 per cent damage and 5 = greater than 75 per cent damage) once at the end of each growth stage. At harvest, from central two rows, stand count, and marketable yield weight were recorded.

6.2.1 Statistical analysis

Analysis of variance (ANOVA) was used for analysis of insect count at different sampling dates. The counts were subjected to square root ($\sqrt{x + 1}$) transformation before analysis. In addition, mean number of DBM (pooled for all the sampling dates) and aphid colonies (*Brevicoryne brassicae* L.) per plant were analyzed. Non-categorical data such as yield were analyzed using ANOVA; and categorical data such as scores were analyzed using non-parametric test (Spearman Correlation coefficient). Means were separated using S-N-K test.

Yield loss was estimated by calculating the differences in yield between the treatments with the highest yield with the yield of the untreated plot:

$$Yieldloss(\%) = \left[\left(\frac{YBP - YUP}{YBP} \right) * 100 \right]$$

where YBP = yield of plot with the highest yield

and YUP = Yield of the untreated plot

6.3 RESULTS

6.3.1. Number of DBM at different growth stages

The number of DBM per plant for November and April trials are presented in Tables 6.2 and 6.3 respectively. Overall, insect populations were relatively low in both seasons ranging between 0 and 4.1 in November and between 0 and 10.9 in April. In majority of the cases, there were no significant differences ($P>0.05$) between treatments at early and mid growth stages in both planting seasons. However, differences between treatments appeared to be significant after mid growth stages. Plots treated with *Bt* or karate throughout the growing period had significantly lower ($P<0.05$) DBM counts than the untreated check from the eighth week in November. A similar trend was observed in the April planted trial, though this was not consistent in some cases. Table 6.4 shows the mean number of DBM per plant (average of samples taken for a period of 13 weeks weekly). The highest overall mean of DBM number per plant among treatments including the untreated check ranged between 0 and 1.2 for the November planting and between 0.4 and 2.9 for the April planting (Table 6.4). The highest DBM number per plant of 1.2 and 2.9 was recorded from the untreated plot in November and the April planted trials respectively. The data in Table 6.4 does not show the DBM number recorded from the untreated check. As pointed out in the materials and methods part of this chapter, data from the untreated plot was excluded to compare the two pesticides and their interaction with the treatments using a factorial RCBD. The DBM number was significantly higher in the *Bt* treated

plots than plots treated with Karate in the November planted trial. In the April planted trial, mean number of DBM was equal for both pesticides. The highest number of DBM was recorded from the untreated plot in both seasons. On the other hand the lowest number was recorded from the plots treated weekly throughout the growing season. DBM number was lower in plots treated during the pre-heading stage than those treated during the seedling and heading stage in both seasons.

6.3.2 Yield

Yield data are presented in Tables 6.5 and 6.6 for November and April plantings respectively. The lowest mean yield in both seasons was recorded from the untreated plot and the highest yield was recorded from plots treated with karate weekly throughout the growing period. In the November plantings, significant difference in yield was observed only between the untreated check and plots treated with karate weekly throughout the growing period. Among *Bt* plots, the highest yield was recorded from plots treated weekly throughout the growing season followed by plots treated at pre heading stage only. Similarly, in this season, the highest yield among karate plots was recorded from plots treated weekly throughout the growing period followed by plots treated at pre-heading stage only (Table 6.5). Comparison of yield between the untreated check and the best treatment shows a yield loss of 36.1% in the *Bt* treated plots and 49.4% in the karate treated plots in the November planting. These percentages corresponded to yield losses of 12

tons/ha in the *Bt* treated plot and 20.7 tons/ha in the karate treated plot.

Results of the April planting show significant difference among treatments (Table 6.6). Here too, the lowest yield was recorded from the untreated plot and the highest yield was recorded from plots treated with karate throughout the growing season. Similar to the November planting, the highest yield in *Bt* plots was recorded from the plot treated weekly throughout the growing season. However, in this planting season, yield from the plot treated at seedling stage only was higher than the plot treated at pre heading stage only and at heading stage only with significant differences among the karate treated plots. Differences were not significant between *Bt* plots. Yield difference between the untreated check and the best treatment shows a yield loss of 85.2 % in the *Bt* treated plots and 91.2% in the karate treated plots in April planted trial. These percentages corresponded to yield losses of 27 tons/ha in the *Bt* treated plot and 48.7 tons/ha for the karate treated plot.

Results of correlation analysis of yield with DBM damage scores taken at the seedling, pre-heading and heading stages are presented in Table 6.7. In both November and April planting seasons, no significant ($P > 0.5$) relationship was observed between yield and damage score at the seedling stage. At pre-heading stage, relationship was negatively significant both in the November ($P < 0.001$) and April plantings ($P < 0.01$). Relationship was negatively significant at heading stage ($P < 0.05$) only in the November planted trial.

6.4 DISCUSSION

DBM population growth showed a similar trend in both planting seasons. It remained low at the early stage of crop growth (Seedling stage) and showed a faster development starting from pre-heading stage. Hence, DBM density between treatments at the early crop growth stage did not show high variability between treatments. This is evident when differences between treatments applied at seedling stage only compared with the untreated check. In both seasons, no significant differences were observed between *Bt* applied at seedling stage only with the untreated check. However, plots treated with karate at seedling stage only gave significantly lower DBM numbers on week 3, 4, 7 and 8 of the April planted trial (Table 6.3) and on weeks 8, 9, 11 and 12 of the November planting (Table 6.2). This significantly lower number of DBM in plots treated with karate than the untreated check on some weeks at late stage of crop growth unlike that of the *Bt* treated plots might be due to differences in persistence of the two pesticides. In general, in both seasons, significant differences between the untreated check and the rest of the treatments was observed starting from week 7 which coincided with the treatment application at the pre-heading stage indicating that the pre-heading was the critical period of infestation in this study. Tables 6.5 and 6.6 also show that overall mean number of DBM was lower for both pesticide types and seasons in plots treated at pre heading stage than plots treated at seedling stage only and at heading stages only. No significant interaction between

pesticide types and growth stages was observed for DBM density. In the April planting when DBM population was higher than in November, mean DBM density was equal for both pesticide types. In the November planted trial, however, it was lower in karate than *Bt* treated plots. This was partly due to differences in DBM numbers in plots treated at seedling stage only. As pointed out above and can be seen from Table 6.2, DBM density was two fold in *Bt* plot compared to the karate plot for treatments at seedling stage only. Yield was consistently higher in plots treated at pre-heading stage than seedling and heading stages in the November planted trial for both *Bt* and the karate treated plots. In the April plantings, however, it was higher in plots treated at seedling stage than treatments at pre-heading and heading stage. This does not show consistency with the observed lower DBM density for both pesticides in the April planted trial which was lower in plots treated at pre-heading stage than the other stages. This could be due to time of aphid infestation and their effect on yield in the two planting seasons. In the April planted trial, aphid colonies per plant were significantly lower in the plots treated at seedling stage only than both plots treated at pre-heading stage only and heading stage only which could explain the observed higher yield in the plots treated at seedling stage than the other stages. In the November planted trial, aphid density was similar in the karate treated plots at pre-heading stage only and seedling stage only. This suggests that pre-heading stage is a critical period of DBM infestation. The observed variation in yield in the karate treated plots particularly in the April planted trial was therefore due to both

DBM and aphids. On the other hand, the variation in yield in the *Bt* treated plots could be regarded mainly due to DBM, as it did not affect aphids population, which is expected because of its mode of action.

A period of one month from transplanting has been reported as a critical period of DBM infestation (Palis, 1983) from experiments carried out using artificial infestation. Lumban and Raros (1975) on the other hand reported from studies conducted under natural infestation that effect of DBM on yield is minimal during early and late growth stages which agree with results of this study. Under natural infestation, DBM population development shows a logistic growth curve (Guilloux *et al.*, 2003). Hence population at early growth stage can be too low to inflict heavy damage. This is particularly true if there are no near by brassica fields infested with DBM. Results of the temporal dynamics (Chapter 4) confirm this observation. This picture could be different in farmers' brassica fields where continuous planting is practiced and infestation is higher. Results of the correlation analysis (Table 6.7) also confirmed the importance of pre-heading stage in relation to DBM infestation. Stronger and highly significant negative relationship ($P < 0.01$) between DBM damage and yield in both seasons was observed at pre-heading stage.

These findings show that yield loss on cabbage due to DBM ranges between 36.1 and 85.2 % depending on the season of production. This corresponds to a yield loss of 12 to 27.6 tons per ha. These figures were calculated from the *Bt* treated plot only as it did not show any effect on aphids. When aphids are added, the yield loss ranged from 36.1 to 91.2 %

corresponding to losses of 12 to 48 tons/ha. Although the yield loss percentage from the *Bt* treatment appeared similar to that of karate, the actual yield loss in tons per ha which shows marked difference indicates the level of damage by the aphids and the efficacy of karate in controlling the pest. This figure, though high, can be regarded as low when compared with the level of damage DBM inflicts on brassica crops in general in the central Rift Valley region of Ethiopia (Ayalew *et al.*, 2004) where total crop failure or up to 100 per cent loss is common in a season of heavy infestation. Assessment of loss using pesticides as described in this study, however, is difficult under farmers' field of brassica producing areas with a long history of pesticide use. Because of pesticide resistance development and decimation of the bio-control agents, DBM infestation and damage is higher in heavy pesticide use areas and lower in no or minimal pesticide use areas (Ayalew *et al.* 2004). Experiments conducted under farmers field in Kenya also showed higher DBM population in the karate treated plot than untreated plot (Macharia, Personal comm.). *Bt* may be used to assess DBM damage under farmers' field condition. However reports from elsewhere (Kirsch and Schmutterer, 1988) where *Bt* is extensively used show absence of effect by *Bt* on controlling DBM. According to these authors, no increment in yield level was observed through the application of *Bt* over the untreated control. Yield loss on the cabbage assessed under farmers' condition using *Bt* in Kenya (Macharia, pers comm.) showed 12 to 22 %. This variation could be due to resistance development to *Bt* (McGaughey, 1985; Kirsch and Schmutterer, 1988), similar to other

synthetic insecticides (Chen and Sun, 1986). Until threshold levels are available for use in the integrated management of DBM, these results indicate that in areas where insecticides can give effective control, the pre-heading stage (Meier, 1997) need to be protected to minimize DBM damage.

Stage		
Heading (S)	2, 18, 25 Dec 2009	9, 14, 21, 27 May and 3 June
Heading (P)	1, 8, 11, 17 Jan 2010	11, 17, 23 June and 3 July
Heading (H)	29 Jan, 5, 12 Feb 2010	4, 14 and 23 July
DBM	All the above dates	All of the above dates

Meier (1997) scale

Table 6.1 Treatment details of the DBM yield loss assessment experiment, Melkassa, 2001/2001

Growth stages of head cabbage*	Application dates of pesticides (Bt and Karate)	
	November planting	April planting
1. Untreated	—	—
2. Seedling (S)	12, 18, 25, Dec. 2001	9, 14, 21, 27 May and 3 June
3. Pre-heading (PH)	1, 8, 15, 22, Jan 2002	11, 17, 25 June and 2 July
4. Heading (H)	29 Jan, 5, 12 Feb 2002	9, 16 and 23 July
5. S+PH+H	All the above dates	All of the above dates

* = Meier (1997) scale

Table 6.2. Effect of cabbage growth stage and pesticide used on the numbers of diamondback moth/plant in the

November 2001 planting at Melkassa, Ethiopia.

Weeks untreated	<i>Bt</i>					Karate			
	Seedling	Pre-heading	Heading	throughout	Seedling	Pre-heading	Heading	Throughout	
1	0.10A	0.00A	0.00A	0.15A	0.25A	0.10A	0.20A	0.05A	0.05A
2	0.10A	0.15A	0.30A	0.10A	0.20A	0.05A	0.00A	0.50A	0.05A
3	0.05BC	0.10BC	0.25AB	0.15BC	0.05BC	0.05BC	0.00C	0.50A	0.05BC
4	0.05AB	0.00B	0.05AB	0.30A	0.15AB	0.00B	0.40A	0.10AB	0.00B
5	0.20A-C	0.15BC	0.30AB	0.40A	0.05C	0.00C	0.10BC	0.15A-C	0.00C
6	0.25AB	0.20AB	0.20AB	0.35A	0.00B	0.00B	0.15AB	0.05AB	0.00B
7	0.20A	0.05A	0.15A	0.20A	0.05A	0.00A	0.05A	0.20A	0.00A
8	1.70A	1.45AB	0.35BC	2.10A	0.40BC	0.45BC	0.00C	1.25AB	0.05C
9	2.00BC	2.30AB	0.35E-F	3.35A	0.40D-F	0.65DE	0.25EF	1.10CD	0.00F
10	1.80AB	2.70A	0.15C	1.45AB	0.15C	2.65A	0.00C	0.55BC	0.05C
11	2.40A	1.25AB	0.40CD	0.50CD	0.15D	1.00BC	0.00D	0.25D	0.00D
12	4.15A	2.15AB	0.75CD	0.30CD	0.00D	1.05BC	0.00D	0.10CD	0.00D
13	2.65A	2.45A	1.60AB	0.25BC	0.10C	1.25AB	0.00C	0.10C	0.00C

Means within rows followed by the same letter are not significantly different at $P < 0.05$ (Means were separated by S-N-K test)

Table 6.3 Effect of cabbage growth stage and pesticide used on the numbers of diamondback moth/plant in the April

2002 planting at Melkassa, Ethiopia.

Week	<i>Bt</i>					Karate			
	untreated	Seedling	Pre-heading	Heading	Throughout	Seedling	Preheading	Heading	Throughout
1	0.05AB	0.10AB	0.15A	0.00B	0.00B	0.00B	0.00B	0.00B	0.05AB
2	0.15A	0.15A	0.05A	0.10A	0.00A	0.10A	0.00A	0.00A	0.05A
3	0.40AB	0.10BC	0.10BC	0.65A	0.00C	0.00C	0.15C	0.30A-C	0.20A-C
4	0.35A	0.15AB	0.10AB	0.05B	0.05B	0.00B	0.15AB	0.20AB	0.00B
5	0.15AB	0.10AB	0.05AB	0.25A	0.05AB	0.00B	0.05AB	0.15AB	0.05AB
6	0.20AB	0.35AB	0.25AB	0.30AB	0.60AB	0.10B	0.35AB	1.15A	0.50AB
7	1.50A	0.15A-C	0.40A-C	1.05AB	0.40A-C	0.00C	1.00BC	0.95A-C	0.40A-C
8	0.75AB	0.30BC	0.10C	0.85A	0.15C	0.00C	0.30BC	0.30BC	0.05C
9	4.45AB	2.85A-C	1.05A-C	5.10A	0.95BC	3.70A-C	0.65C	2.45A-C	0.70C
10	6.10AB	7.95A	0.65C	5.40AB	0.60C	5.90A	0.85C	6.90A	1.95BC
11	7.00A	6.90A	0.90C	5.20AB	0.85C	6.70A	1.25C	5.30A	1.60BC
12	5.80A	4.95AB	1.60CD	2.70BC	0.70D	7.20A	0.40D	1.70CD	0.95CD
13	10.90A	6.30AB	5.50ABC	2.65BC	1.55C	6.30AB	5.10BC	4.35BC	2.45BC

Means within rows followed by the same letter are not significantly different at $P < 0.05$ (Means were separated by S-N-K test)

Table 6.4. Mean number of DBM per plant on cabbage treated with *Bacillus thuringiensis* (Bt) and Karate at different growth stages in the November 2001 and April 2002 plantings at Melkassa, Ethiopia

Treatments	November		April	
	Bt	Karate	Bt	Karate
	Applied at seedling stage only	1.0a	0.5a	2.3a
Applied at pre-heading stage only	0.4bc	0.1b	0.8bc	0.8b
Applied at heading stage only	0.7ab	0.3ab	1.9ab	1.8a
Applied throughout	0.1c	0.0b	0.4c	0.7b
Mean	0.6a	0.3b	1.4a	1.4a

Values with in a column followed by the same letter other than the mean are not significantly different at $P < 0.05$) (Means were separated by S-N-K test)

Table 6.5. Pest density and yield of head cabbage treated with *Bt* and karate at different growth stages in the November 2001 planting, at Melkassa, Ethiopia.

Treatments	Yield (t/ha)	DBM/plant	Aphid colonies/plant
Untreated	21.2b	1.3a	2.2a
Bt at seedling stage only	25.9ab	1.0ab	2.2a
Bt at pre-heading stage only	26.8ab	0.4b-d	2.3a
Bt at heading stage only	22.7ab	0.7bc	2.2a
Bt applied throughout	33.2ab	0.1cd	2.2a
Karate at seedling stage only	27.2ab	0.5b-d	1.7b
Karate at pre-heading stage only	34.0ab	0.1cd	1.4b
Karate at heading stage only	27.1ab	0.3b-d	2.1a
Karate applied throughout	41.9a	0.0d	0.4c

Means with in a column followed by the same letter are not significantly different at $P < 0.05$) (Means were separated by S-N-K test)

Table 6.6. Pest density and yield of head cabbage treated with *Bt* and karate at different growth stages in the April 2002 planting, Melkassa

Treatments	Yield (t/ha)	DBM/plant	Aphid colonies/plant
Untreated	4.7d	2.8a	2.4a
Bt at seedling stage only	18.93cd	2.3ab	2.3a
Bt at pre-heading stage only	17.21cd	0.8b	2.1a
Bt at heading stage only	11.48cd	1.9ab	2.4a
Bt applied throughout	31.71bc	0.4b	2.2a
Karate at seedling stage only	37.35b	2.3ab	0.9c
Karate at pre-heading stage only	18.12cd	0.8b	1.6b
Karate at heading stage only	15.00cd	1.8ab	2.2a
Karate applied throughout	53.4a	0.7b	0.5d

Means with in a column followed by the same letter are not significantly different at $P < 0.05$ (Means were separated by S-N-K test)

Table 6.7. Correlation matrix of DBM scores taken at Seedling, Pre-heading and Heading stages with yield (Marketable and total yield) in the November and April planted experiment.

	Seedling	Pre-heading	heading
November			
Marketable	-0.224ns	-0.524***	-0.357*
Total	-0.282ns	-0.588***	-0.361*
April			
Marketable	-0.1704ns	-0.4788**	-0.2890ns
Total	-0.1698ns	-0.4143**	-0.2455ns

ns = not significant ($P > 0.05$)

* = $P < 0.05$

** = $P < 0.01$

*** = $P < 0.001$

CHAPTER 7

7.0 Suitability of cultivated and wild crucifers for the development of Diamondback moth, *Plutella xylostella* L

7.1. INTRODUCTION

Diamondback moth (DBM), *Plutella xylostella* L., feeds on cultivated and wild plants belonging to the family Cruciferae. One or more uncultivated species in 27 different genera of this family are recorded from different parts of the world as hosts that sustain feeding and reproduction of DBM (Talekar and Shelton, 1993). The association of DBM to crucifer species is due to the presence of one or more glucosinolates, singrin, sinalbin and glucoheirolin, which act as specific feeding stimulant (Talekar and Shelton, 1993). Work of Gupta and Thorsteinson (1960) suggested that occurrence of feeding inhibitors as well as the absence of essential feeding stimulants exclude some botanical species in crucifers from the food plant range. On the other hand, the existence of effective feeding stimulants other than mustard oil, glucosides, and absence of feeding inhibitors, have put some species of Leguminosae in the food range of the pest. For example, field pea, *Pisum sativum* L. was shown to be sufficiently palatable, nutritious and free of toxicants to support successive generations of *Plutella xylostella* larvae in the laboratory (Gupta and Thorsteinson, 1960). Löhr (2001) reported serious damage inflicted by DBM on sugar snap pea in Kenya under field condition and Löhr and Gathu (2002) showed that in only few

generations of selection, a DBM strain can be produced that performs comparably well on crucifer and pea. Cruciferous weeds are able to sustain feeding and reproduction of DBM, and play an important role in maintaining DBM population in the absence of cruciferous crops (Harcourt, 1986). The objective of this experiment was to compare developmental period and reproduction of DBM in common wild and cultivated crucifers grown in Ethiopia under laboratory conditions.

7.2 MATERIALS AND METHODS

Seeds of two cultivated brassica crops, head cabbage (*Brassica oleracea* var. *capitata* variety Copenhagen Market) and Ethiopian mustard (*Brassica carinata* Braun) were purchased from commercial outlets and the other two, *Brassica nigra* Koch. accession number 231212 and *Brassica nigra* Koch. accession number 229938 were obtained from Ethiopian Biodiversity Institute. Seeds of the wild crucifer, *Erucastrum arabicum* Fisch. and Mey, were collected from field in the vicinity of Melkassa Research Center of the Ethiopian Agricultural Research Organization (EARO). Plants were raised in the green house at Melkassa center of EARO. Seeds were seeded in seedling raising tray and allowed to grow for two to four leaf stage and transplanted to a pot (13 cm diameter and 15 cm height). Compost and sand loam soil in a proportion of 1:1 was used as a growth medium. Leaves were used for testing after the 8-leaf stage. Because of poor germination of the wild crucifer,

collection of leaves was made from the near by field. The study was conducted in the laboratory in ambient condition (Temperature ranged between 23 and 30 °C).

A stock of culture of DBM was established with pupae collected from cabbage fields in the vicinity of Melkassa Research Center. Emerged adults were kept in plastic containers (30 cm X 30 cm X 30 cm) and allowed to mate. A young cabbage leaf was used for egg laying. The petiole of the leaf was immersed in a beaker (6 cm diameter and 10 cm height) filled with water to avoid quick drying of the leaf. Egg laying was checked daily and leaves were changed daily. Newly hatched larvae from the stock culture were introduced individually into a plastic vial (2.5 cm in diameter and 6 cm in height) and supplied with a piece of leaf size 5 cm long and 2.5 cm wide as food for each brassica plant and wild hosts. Twenty larvae were observed. To keep the food fresh, a piece of wet filter paper was placed in each vial. Food was changed every other day. Larvae were kept in the vial through pupation. Pupae were weighed when two days old and replaced back to the respective vial until adult emergence. The developmental period of immature stages was determined from daily observation. Individual pairs from each tested crucifer species were placed into a plastic cage measuring 30 cm X 30 cm x 30 cm. They were supplied with a piece of leaf of the test plants from where they were reared and 5% sugar solution to estimate the fecundity, fertility and incubation period. The larvae hatched were reared again in a similar fashion until adult

emergence. Pupal weight from each brassica plant was recorded to examine its relation with fecundity. Suitability of the test brassica plants was compared in generation one and two using survival rate, fecundity, fertility and longevity of the different stages; egg, larva, pupa and adult; and population growth statistics (Maia *et al.*, 2000)

7.2.1 Statistical analysis

Data were subjected to ANOVA using PROC GLM of the SAS software (SAS, 1999). Fertility data were subjected to arcsine transformation before analysis. Means were separated using Student-Newman-Keuls (SNK) test. Relationship of pupal weight and fecundity was examined using simple linear regression.

Population growth statistics, the net reproductive rate (R_0), the intrinsic rate of increase (r_m); the mean generation time (T), the finite rate of increase (λ) and the Doubling time (Dt) were calculated and compared using the method of Maia *et al.* (2000) for both generations. Parameters were estimated using the following formulae:

Net reproductive rate (R_0) (the average number offspring an individual in a population will produce in her life time = $\sum l_x m_x$

Intrinsic rate of natural increase (r_m) is the maximum rate of increase a population could experience in the absence of any limitations for resource abundance or predators, which can be approximated from

$$\sum e^{-r_m x} l_x m_x = 1$$

Generation time (T) (The average age at which a female gives birth to her

$$\text{offspring} = \frac{\log_e R_o}{r_m}$$

Doubling time (Dt) (The time it will take a population to double) =

$$= \frac{\text{Ln}(2)}{r_m}$$

Finite rate of increase (λ) (The amount that the population must be

multiplied by to give the population size in the next time unit) = e^{r_m}

where x is adult female age (days); lx is proportion of surviving female at age x; mx is the number of female offspring per adult female in the age interval x, ln is the natural logarithm and e is the base of natural logarithm (2.7183).

7.3 RESULTS

7.3.1 Developmental period, pupal weight and survival rate

Developmental period, pupal weights and survival of the first and second generations are presented in Tables 7.1 and 7.2 respectively. Egg, larval period, pupal period and pupal weight ranged between 2.8 to 3.5, 7.8 to 9.63, 5.15 to 5.58 days, and 6.1 to 6.7 mg respectively. In the first generation, significant variation was observed only for larval period where it was significantly longer on *B. nigra* acc. no. 229938 and *B. nigra* acc. no 231212 than the rest of the cultivated species and the wild crucifer. Although differences were not significant ($P>0.05$), developmental period was shorter in cabbage for all stages than the rest of the test plants. Egg

and larval period in the wild crucifer was intermediate between the cultivated crops.

In the second generation, developmental period, egg, larval period, pupal period and pupal weight ranged between 2.6 to 4.0, 9.2 to 10.7, 5.7 to 6.8 days and 6.5 to 8.2 mg respectively. Egg period was significantly longer in the wild crucifer than two of the cultivated crops; namely, cabbage and Ethiopian mustard. Larval period was significantly shorter in cabbage and significantly longer in the wild crucifer than the rest. Pupal period was significantly longer in *B. nigra* acc. no. 229938 and shorter in cabbage than the rest of the test plants. Pupal weight was significantly higher in cabbage and Ethiopian mustard than the rest.

7.3.2 Fecundity, fertility and adult longevity

Fecundity, fertility and adult longevity data are presented in Table 7.3 for the first generation and Table 7.4 for the second generation. Similar to the developmental period in the first generation, variation in fecundity, fertility and adult longevity was not significant ($P>0.05$). Fecundity ranged between 140 to 293 eggs per female and fertility (Arcsine transformed) ranged between 0.36 to 0.99. Fecundity on cabbage and Ethiopian mustard was about two fold compared to the wild crucifer and *B. nigra* acc. no. 229938. Similarly fertility in cabbage and Ethiopian mustard was higher than two fold compared to the wild crucifer. Adult

longevity did not show high variability; it ranged between 10.8 and 11.2 days for females and between 9.5 and 11.4 days for males.

In the second generation, fecundity per female, female and male longevity ranged between 63 to 320 eggs, 9.7 to 11.3 and 10.2 to 11.0 days. Fertility ranged between 0.55 to 0.99. Fecundity in cabbage was about four-fold and significantly higher than all cultivated species. It was also higher in the wild crucifer than the rest of the cultivated species except cabbage though differences were not significant. Variation in fertility was not significant ($P > 0.05$) but was higher in cabbage and Ethiopian mustard than the rest as observed in the first generation.

The relationship between weight of pupae and fecundity was positive and significant ($P < 0.05$) in both generations. However, these relationships were weak as only 28 % in the first generation and 18 % in the second generation of the variation in fecundity was explained by the weight of pupae (Fig. 7.1)

7.3.3 Population growth parameters

Population growth parameters are presented in Table 7.5 (for the first generation) and Table 7.6 (for the second generation). Intrinsic rate of increase (r_m), net reproductive rate (R_o) and finite rate of increase (λ) were highest and doubling time was lowest (Dt) in cabbage in both generation one and two than the rest. On the other hand r_m , R_o , and λ were lowest and Dt was highest in *Brassica nigra* (Both accession no. 231212 and

229938). Values of these parameters were intermediate for the Ethiopian mustard and the wild crucifer, *E. arabicum*. Figure 7.2 and 7.3 show fecundity at different age of female in the first and second generations respectively. Egg laying pattern is clear in cabbage and decreases with female age in both generations. In the first generation pattern of egg laying in the Ethiopian mustard was similar to cabbage; it decreased with the age of the female.

7.4. DISCUSSION

Results showed that cabbage followed by Ethiopian mustard is more suitable for the development of DBM; and the wild crucifer is as suitable as the rest of the cultivated brassica crops. Non significant difference in developmental periods of immatures (Yamada, 1983) in wild and cultivated crucifers has been reported although it was prolonged in the wild crucifers. In a similar study with one cultivated species (cabbage) and five wild crucifers, Muhamad *et al.* (1994) reported a shorter developmental period of immatures and highest early fecundity in cabbage. In this study, both in the first and second generations, developmental period of immatures was shorter and fecundity was higher in DBM raised on acabbage. Absence of significant variability in the fecundity of the first generation between the tested brassica plants despite higher differences in the number of eggs laid could be explained by high variability between replicates within treatment.

Positive relationship between pupal weight and fecundity observed is in agreement with several reports (Yamada and Umeya, 1972; Moller, 1988 and Muhamad *et al.*, 1994). The population growth statistics in both generations clearly show that cabbage was the most suitable host and *B. nigra* (both accessions) was the least suitable with the other cultivated brassica, Ethiopian mustard and the wild crucifer being intermediate. This was similar to results of the developmental periods of the different stages as well as fecundity and fertility observed in both generations. However, comparison for population growth parameters in the first generation showed significant differences between the tested plants. On the other hand, differences were not significant when comparison was made for developmental period and reproductive potential in the first generation. This could be due to differences observed in fertility (Per cent egg hatched) as the population growth analysis considers all variables in the estimation of population growth parameters. Although only one wild crucifer which is widely found as weeds in crop fields was compared with the cultivated species in this study, the development of DBM on wild crucifer was found to be comparable with some of the cultivated brassica crops, *Brassica nigra*. Sixty-one species of crucifers in 21 genera are reported to occur in Ethiopia (Jonsell, 2000). Their influence in the spatio-temporal dynamics of DBM and their parasitoids is expected to be high as observed in several brassica production regions (Talekar and Shelton, 1993). Future study in this line should focus on the testing of larger

number of wild crucifers for the development of both DBM and the commonly associated parasitoids. Assessment of the chemical constituent of the crucifer species would help to better understand mechanism of host suitability. Assessing DBM population and parasitism level by the different parasitoids under field condition on the commonly found wild crucifers would also help to understand their role in the population dynamics of DBM which in turn is helpful in the development of appropriate management program.

1 - Accession number 731212, 2 - Accession number 234938

Means followed by the same letter in a column are not significantly different at P

0.05, SNK test

Table 7. 1. Performance of the first generation of Diamondback moth on the common cultivated and wild crucifers grown in Ethiopia

Crucifer species	Developmental period (days)			Pupal weight (mg)	Survival (%)
	Egg	Larval	Pupal		
Cabbage	2.8±0.16a	7.85±0.11c	5.25±0.10a	6.55±0.27a	83.4±2.7a
Eth. mustard	3.0±0a	7.80±0.22c	5.58±0.12a	6.10±0.16a	79.3±3.9a
<i>Brassica nigra</i> ¹	3.4±0.24a	9.10±0.10b	5.23±0.14a	6.20±0.27a	28.1±13.0b
<i>Brassica nigra</i> ²	3.5±0.29a	9.63±0.16a	5.13±0.17a	6.10±0.17a	25.8±12.1b
<i>E. arabicum</i>	3.2±0.25a	8.05±0.13c	5.40±0.15a	6.70±0.23a	53.0±14.2ab

1 = Accession number 231212; 2 = Accession number 229938

Means followed by the same letter in a column are not significantly different at P = 0.05, SNK test

Table 7.2. Performance of the second generation of Diamondback moth on the common cultivated and wild crucifers grown in Ethiopia

Crucifer species	Developmental period (days)			Pupal weight (mg)	Survival (%)
	Egg	Larval	Pupal		
Cabbage	2.60±0.24b	9.20±0.09c	5.70±0.13c	8.20±0.22a	83.33±3.7a
Eth. mustard	2.67±0.21b	10.30±0.15b	5.94±0.12bc	7.75±0.23a	68.24±8.6a
<i>Brassica nigra</i> ¹	3.33±0.33ab	10.15±0.08b	6.35±0.12b	6.50±0.20b	59.14±1.9a
<i>Brassica nigra</i> ²	3.67±0.33ab	10.50±0.12ab	6.83±0.11a	6.66±0.20b	47.07±6.6a
<i>E. arabicum</i>	4.00±0.32a	10.70±0.13a	6.35±0.16b	6.75±0.33b	53.56±17.0a

1 = Accession number 231212; 2 = Accession number 229938

Means followed by the same letter in a column are not significantly different at P = 0.05, SNK test

Table 7.3. Reproductive potential and adult longevity of the first generation of Diamondback moth on the common cultivated and wild crucifers grown in Ethiopia

Crucifer species	Eggs/female	Fertility*	Longevity	
			Female	Male
Cabbage	293.5±23.8a	0.99±0.04a	11.2±0.5a	10.3±0.5a
Eth. mustard	257.3±62.4a	0.99±0.08a	11.0±0.6a	10.3±0.3a
<i>Brassica nigra</i> ¹	226.0±54.4a	0.36±0.17a	12.4±0.5a	11.4±0.4a
<i>Brassica nigra</i> ²	146.2±41.45a	0.37±0.17a	10.8±0.6a	9.5±0.6a
<i>E. arabicum</i>	140.0±51.9a	0.73±0.21a	11.5±1.04a	10.2±0.48a

1 = Accession number 231212; 2 = Accession number 229938

* = values are arcsine transformed

Means followed by the same letter in a column are not significantly different at P=0.05, SNK test

Table 7.4. Reproductive potential and adult longevity of the second generation of Diamondback moth on the common cultivated and wild crucifers grown in Ethiopia

Crucifer species	Eggs/female	Fertility	Longevity (days)	
			Female	Male
Cabbage	320.6±38.2a	0.99±0.06a	10.2±0.4a	10.2±0.7a
Eth. mustard	85.7±20.2b	0.83±0.12a	10.0±0.4a	10.7±0.4a
<i>Brassica nigra</i> ¹	63.0±4.2b	0.77±0.03a	9.7±0.7a	10.0±0.6a
<i>Brassica nigra</i> ²	83.3±28.8b	0.55±0.08a	11.3±0.9a	11.0±0.6a
<i>E. arabicum</i>	132.4±41.5b	0.64±0.25a	11.4±0.7a	10.4±1.1a

1 = Accession number 231212; 2 = Accession number 229938

* = values are arcsine transformed

Means followed by the same letter are not significantly different at P=0.05, SNK test

Table 7. 5. Population growth parameters of the first generation of Diamondback moth developed on the common cultivated and wild crucifers grown in Ethiopia

Crucifer species	Life table statistics				
	r_m	R_o	λ	T	Dt
Cabbage	0.31a	146a	1.37a	15.8a	2.20a
Ethiopian mustard	0.29ab	91ab	1.34a	15.5ab	2.38a
<i>Brassica nigra</i> ¹	0.16c	21c	1.17a	18.9c	4.30a
<i>Brassica nigra</i> ²	0.18c	26c	1.19a	18.0c	3.81a
<i>Erucastrum arabicum</i>	0.26b	87b	1.30a	17.0b	2.63a

1 = Accession number 231212; 2 = Accession number 229938

Means followed by the same letter in a column are not significantly different at P = 0.05 (Means were separated using student t-test mean comparison (Maia et al., 2000)).

Table 7. 6. Population growth parameters of the second generation of Diamondback moth developed on the common cultivated and wild crucifers grown in Ethiopia

Crucifer species	Life table parameters				
	r_m	R_o	λ	T	Dt
Cabbage	0.28a	151a	1.32b	17.9a	2.47a
Ethiopian mustard	0.18b	25b	1.20b	18.1b	3.85b
<i>Brassica nigra</i> ¹	0.15bc	22b	1.17b	19.8bc	4.45b
<i>Brassica nigra</i> ²	0.12c	13b	1.13a	21.4c	5.68b
<i>Erucastrum arabicum</i>	0.17bc	32b	1.18a	20.9b	4.17b

1 = Accession number 231212; 2 = Accession number 229938

Means followed by the same letter in a column are not significantly different at $P = 0.05$ (Means were separated using student t-test mean comparison (Maia et al., 2000)).

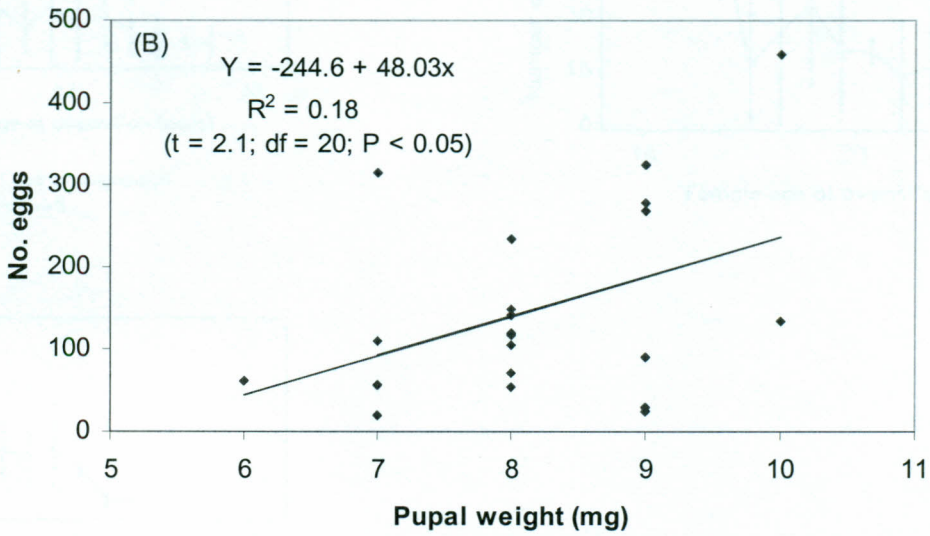
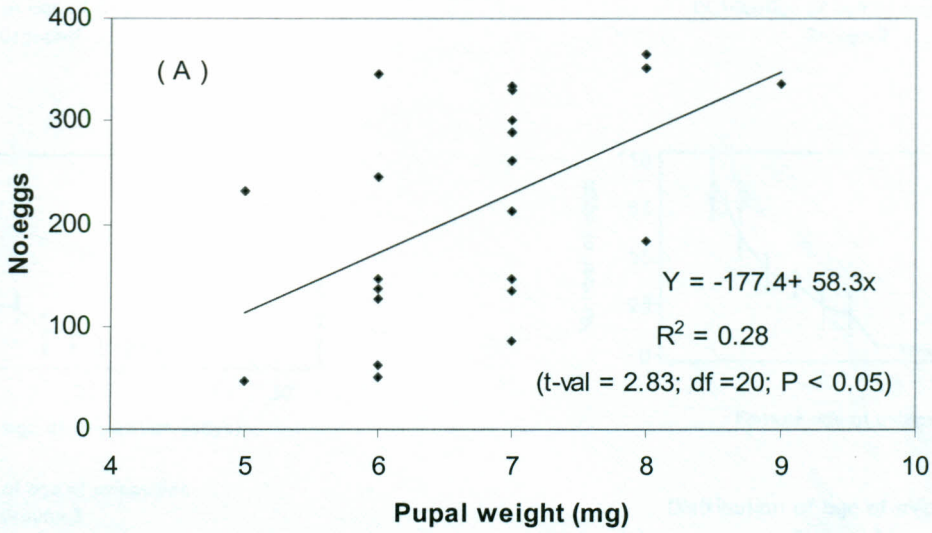
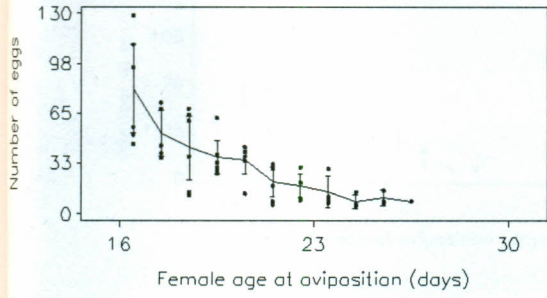
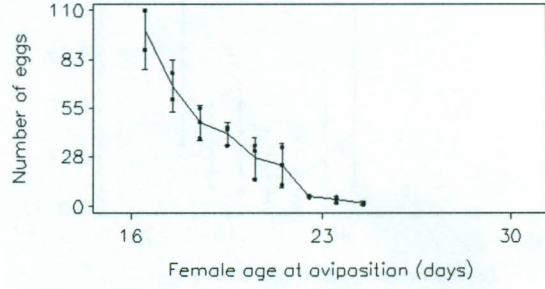


Fig 7.1. Relationship between fecundity and weight of pupa: First generation (A) and second generation (B)

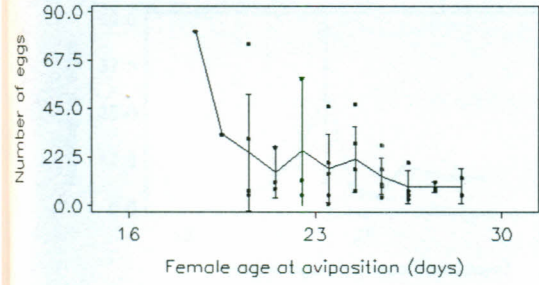
Distribution of age at oviposition
Group= 1



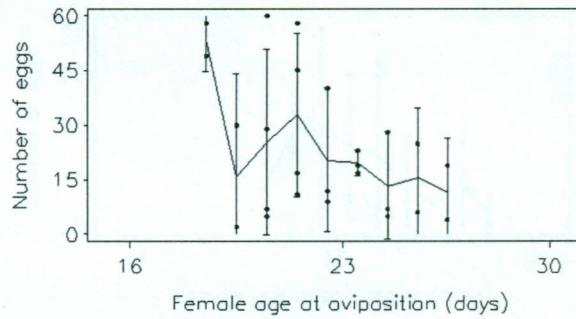
Distribution of age at oviposition
Group=2



Distribution of age at oviposition
Group=3



Distribution of age at oviposition
Group=4



Distribution of age at oviposition
Group=5

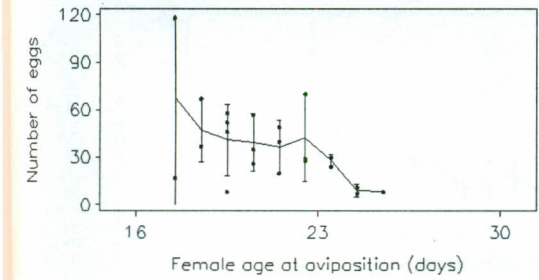
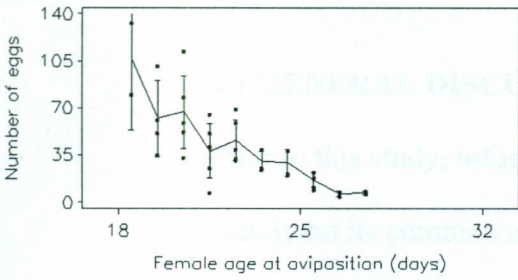
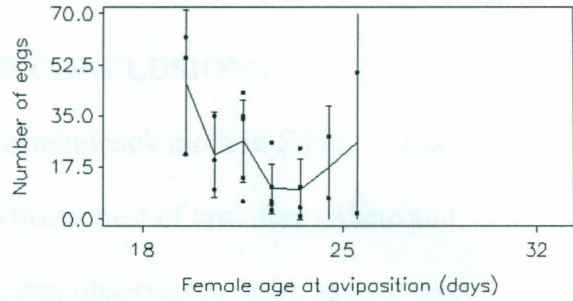


Fig. 7.2. Distribution of eggs per female at different ages in the first generation of diamondback moth on common cultivated and wild crucifers grown in Ethiopia (Group 1 = Cabbage; Group 2 = Ethiopian mustard; Group 3 = *Brassica nigra* accession number 231212; Group 4 = *Brassica nigra* accession number 229938; Group 5 = *Erucastrum arabicum*)

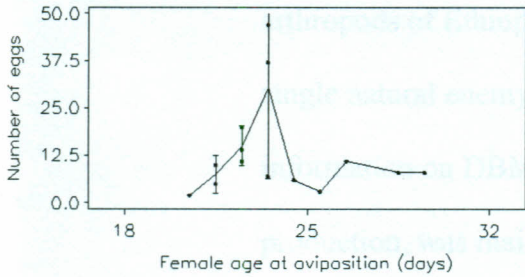
Distribution of age at oviposition
Group=1



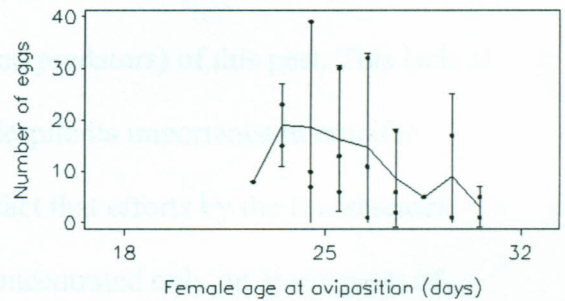
Distribution of age at oviposition
Group=2



Distribution of age at oviposition
Group=3



Distribution of age at oviposition
Group=4



Distribution of age at oviposition
Group=5

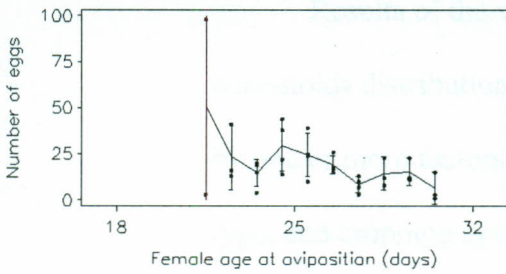


Fig. 7.3. Distribution of eggs per female at different ages in the second generation of diamondback moth on common cultivated and wild crucifers grown in Ethiopia (Group 1 = Cabbage; Group 2 = Ethiopian mustard; Group 3 = *Brassica nigra* accession number 231212; Group 4 = *Brassica nigra* accession number 229938; Group 5 = *Erucastrum arabicum*)

CHAPTER 8

8.0 GENERAL DISCUSSION, CONCLUSIONS AND SUGGESTIONS

8.1 GENERAL DISCUSSION AND CONCLUSIONS

Prior to this study, information on Diamondback moth in Ethiopia was mainly on its common occurrence as insect pest of crucifers (Abate and Ayalew, 1994). The insect, however, was observed to result to total crop failure in seasons of heavy infestation especially in the main crucifer producing areas of the central Rift Valley regions. Entomophagous arthropods of Ethiopia, compiled by Abate (1991) does not contain a single natural enemy (parasitoids and predators) of this pest. This lack of information on DBM in Ethiopia, despite its importance in crucifer production, was mainly due to the fact that efforts by the few research entomologists in the country had concentrated only on insect pests of major food crops.

Results of the survey of Diamondback moth and the indigenous parasitoids distribution and severity showed that pest density is influenced by one or more factors including level of insecticide use, parasitism, crop type, and cropping system. In general, higher DBM density is associated with heavy pesticide use areas and low level of parasitism. High level of DBM density in pesticide use areas as opposed to minimal or no pesticide use areas can be explained by the development of resistance by the pest to the pesticides being used as observed in several countries (Tabashnik *et*

al., 1987; Cheng *et al.*, 1992; Shelton and Wyman, 1992; Syed, 1992; Kibata, 1996) and low level of parasitism because of the pesticide effect on the parasitoids (Sastrosiswojo and Sastrodihardjo, 1986; Ooi, 1992; Talekar and Shelton, 1993).

Seven hymenopteran parasitoid species from five families have been recorded in this study. Of these *O. sokolowskii*, *Diadegma* sp. and *Apanteles* sp. are the dominant ones accounting for over 90 percent of the parasitoid complex. Parasitism tended to be higher in southwestern Ethiopia, where *Apanteles* sp. and *Diadegma* sp. are the dominant parasitoid species. *O. sokolowskii* was recorded only from two fields in that area and accounted for less than 1 percent of the parasitoid complex. On the other hand, *O. sokolowskii* was the major parasitoid in central Rift Valley areas and northern Ethiopia. The low number of *O. sokolowskii* in southwestern Ethiopia could be due to competition (Liu *et al.*, 2000) and host-parasitoid relationship. The dominant brassica species sampled in south western Ethiopia was the Ethiopian mustard, *Brassica carinata* Braun grown in with intercropping system that might have contributed to the observed variation in host parasitoid relationship. Plant species influence on host parasitoid relationship has been reported (Verkerk, *et al.*, 1998; Liu *et al.*, 2000). However, estimation of their influence is difficult without a controlled experiment (Liu *et al.*, 2000).

The *Diadegma* sp. recorded in this study appeared to be different from the species of *Diadegma* known to attack DBM in Africa. Until

reports of Azidah *et al.* (2000), *Diadegma* spp. of Africa parasitizing DBM were regarded to be *Diadegma semiclausum* Hellén. Azidah *et al.* (2000) suggested that the African *Diadegma* attacking DBM belongs to one species, *D. mollipla*, a parasitoid first described from the potato tuber moth, *Phthorimaea operculella* (Zeller) (Lepidoptera: Gelechiidae) in South Africa (Broodryk, 1971). Molecular studies conducted recently to characterize the African *Diadegma* suggested that *Diadegma* sp. attacking DBM in Ethiopia is a new species, which needs to be described (Wagner *et al.*, 2002). However, *D. mollipla* was earlier reported (Negasi *et al.*, 1986) from a potato, *Solanum tuberosum* L., field in Ethiopia. *Diadegma* sp. reared from PTM attacking potato recently also appeared to be *D. mollipla*. Considering the fact that brassicas and potatoes are produced side by side, the total absence of *D. mollipla* from DBM in Ethiopia is not clear. One possible reason could be replacement by the new yet to be described species. This requires further detailed studies.

Yield loss due to DBM on head cabbage ranged between 36.1 and 85.2 percent corresponding to 12 to 27.6 tons per hectare for head cabbage. One of the difficulties in yield loss assessment under natural infestation is the occurrence of more than one insect pest, making assessment due to one species alone difficult. In this study, although DBM and the cabbage aphid, *Brevicoryne brassicae* L., occurred together and affected yield of cabbage, comparison of yield using *Bt* treatment alone enabled the estimation of yield loss caused by DBM due to its specificity.

The pre-heading stage appeared to be the critical period of DBM infestation. This finding agrees with reports of Lumaban and Raros (1975) and disagrees with reports of Palis (1983). This variation could be due to differences in DBM density in relation to crop growth stage which can vary from season to season and field to field. If population buildup starts slowly as observed in this study (Chapter 4) in areas of low DBM density (Chapter 5), the pre-heading stage could be the critical infestation period.

Results of the temporal dynamics study showed occurrence of two to three generations in the highland site and three to five generations in the lowland site per head cabbage growing season depending on the production season. *Diadegma* sp. was the dominant parasitoid species at the highland site and *O. sokolowskii* at the lowland site. Relatively higher level of parasitism was observed at the highland site in rainy season and was attributed mainly to *Diadegma* sp. This higher level of parasitism by *Diadegma* sp. suggests a potential of conservation practices in the highland rain-fed brassica production areas

The wild crucifer, *Erucastrum arabicum* was found to be as suitable as some of the tested cultivated brassicas for DBM development under laboratory condition. The abundance of DBM and its parasitoids on wild crucifers under field condition is not known. A large number of crucifers both wild and cultivated belonging to 21 genera are known to occur in the country (Jonsell, 2000). Their influence in the spatio-temporal

dynamics of DBM and their parasitoids is expected to be high as observed in several brassica production regions (Talekar and Shelton, 1993).

Within-field distribution of larvae and pupae of DBM was contagious for all larval and pupal stages in head cabbage and kale. In Ethiopian mustard, however, only young stages showed contagious distribution. This difference might have been brought by influence of the cropping system and natural enemies in area of the production of these crops as data were gathered from a wider geographic area.

Spatial analysis showed that DBM is aggregated in the fields of its host plants. This aggregation could be due to limited dispersal range of the adult insect (Mo *et al.*, 2003). Newly established crucifers therefore suffer little DBM damage until the time the population develops to a level causing economic damage. This can give opportunity for the management of pesticide resistant DBM populations (Tabashnik *et al.*, 1987, Caprio and Tabashnik, 1992). The restriction of the large proportion of DBM in its host field may also contribute positively to the management of the pest through mating disruption. Success in mating disruption using pheromones has been reported in different countries (Talekar and Shelton, 1993). Further, monitoring over large areas may not be economically feasible. In this case, spatial analysis allows to circumscribe areas with high DBM infestations, where monitoring efforts should be focused.

8.2 SUGGESTED FUTURE WORK

1. The DBM problem in Ethiopia is currently serious in cabbage fields of the Rift Valley regions and Arsi highland. The contribution of the indigenous natural enemies to DBM mortality in these areas seems to be low. Pesticide use to manage the DBM problem has been observed to be ineffective, especially in the central Rift Valley region, where pesticide use is very high. Classical biological control, i.e. the introduction of effective parasitoids with a good success history should be a priority in DBM IPM in these areas. Successful DBM biocontrol projects in south east Asian countries were implemented using the larval parasitoid, *Diadegma semiclausum* (Talekar *et al.*, 1992). Introduction of the same parasitoid to Kenya and Tanzania under similar growing environment (highland brassica fields) indicated a good establishment in all release sites with higher level of parasitism. *Cotesia plutellae*, a larval parasitoid which is well adapted in hotter regions is reported to give effective control in South Africa and Asian countries.

The following works are suggested in line with this.

- Introduction of *D. semiclausum* in the highland brassica fields and *C. plutellae* in the low land brassica production areas and assessment of their impact on DBM mortality..
- Field life table studies at representative established sites need to be conducted to assess the impact of the locally occurring parasitoids and

other mortality factors including pathogens. This will help to determine the feasibility of promoting biocontrol of DBM through conservation practices.

- Bio-control effort in areas with history of high pesticide use should be augmented with the use of biocontrol compatible products such as *Bacillus thuringiensis*. Effective utilization entails determination of economic threshold
- Creating awareness among farmers through training on how biological control works will facilitate implementation of biological control

2. The cabbage aphid, *Brevicoryne brassicae*, has been observed to be equally important as DBM in brassica fields. Promotion of DBM bio-control will be difficult unless biocontrol compatible options are developed against *B. brassicae*. Hence studies towards the control of *B. brassicae* is needed as part of Brassica IPM in Ethiopia.

3. The role of wild crucifers in spatio-temporal dynamics of DBM and its parasitoids be assessed through a concerted survey work.

4. A sampling plan for decision making in DBM management be developed that considers all factors affecting thresholds. This entails studies on the determination of economic threshold.

5. Results of spatial dynamics reported were from limited observations in small cabbage growing areas. Aggregation and spatial distribution can vary depending on seasons and geographic locations. Detailed studies on factors that affect the spatial and temporal distribution of DBM should be conducted to determine the influence of several factors such as wild host plant, season, cropping system and landscape in DBM population movement and buildup in the different brassica growing areas through the deployment of sufficient monitoring system. Also, correlation between spatial distributions of adults, larvae and damages on plants, can improve our understanding of DBM population spatial dynamics and can better direct control interventions of larvae, using pheromone trap monitoring.

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