

This is to certify that the

thesis entitled

AGRICULTURAL LANDSCAPE COMPLEXITY HAS MIXED EFFECTS ON PATTERNS OF PARASITOID ABUNDANCE AND DIVERSITY

presented by

Alejandro Carlos Costamagna

has been accepted towards fulfillment of the requirements for

M.S. degree in Entomology

jor professor

Date_

MSU is an Affirmative Action/Equal Opportunity Institution

O-7639

TTSIS

d 1002



PLACE IN RETURN BOX to remove this checkout from your record. TO AVOID FINES return on or before date due. MAY BE RECALLED with earlier due date if requested.

DATE DUE	DATE DUE	DATE DUE

6/01 c:/CIRC/DateDue.p65-p.15

AGRICULTURAL LANDSCAPE COMPLEXITY HAS MIXED EFFECTS ON PATTERNS OF PARASITOID ABUNDANCE AND DIVERSITY

By

Alejandro Carlos Costamagna

A THESIS

Submitted to Michigan State University in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Department of Entomology

ABSTRACT

AGRICULTURAL LANDSCAPE COMPLEXITY HAS MIXED EFFECTS ON PATTERNS OF PARASITOID ABUNDANCE AND DIVERSITY

By

Alejandro Carlos Costamagna

The armyworm *Pseudaletia unipuncta* (Haworth) (Lepidoptera: Noctuidae) is a sporadic pest of maize and cereal grains in North America. Field experiments were conducted to determine how landscape structure and host density affect parasitism and parasitoid diversity. Sentinel host larvae were used to assess P. unipuncta parasitism in maize fields located in a complex and a simple agricultural landscape in Michigan, during 2000 and 2001. Neither landscape structure nor host density significantly influenced overall parasitism (80.2% on average) or parasitoid diversity (2.8 species per field on average). However, the two main parasitoid species reared from P. unipuncta (Hymenoptera: Braconidae) responded differently to host density. Glyptapanteles militaris (Walsh) parasitized proportionally more hosts at low host density, while Meteorus communis (Cresson) parasitized more hosts at high host densities. These results contrast with previous findings of increased parasitism in the complex landscape in 1993 and 1996, when overall parasitism was dominated by *M. communis*. However, this study corroborates that similar levels of parasitism in the complex and simple landscape are obtained in years when parasitism is dominated by G. militaris, such as 1998, 2000, and 2001. Laboratory experiments showed that carbohydrate resources significantly increase the longevity of both parasitoid species and *M. communis* fecundity.

To Marisol and Ailén, for all their love and support.

ACKNOWLEDGEMENTS

I would like to thank Dr. Doug Landis for all his support, encouragement, enthusiasm, dedication and patience. His clear perception of insect ecology and agroecosystems, as well as his meticulous approach to experimentation, contributed significantly to challenge me both intellectually and professionally. I would also like to thank Dr. Fabián Menalled for his advice and help throughout this study. I also thank Dr. Jim Miller, Dr. Mark Scriber and Dr. Carolyn Malmström for their advice pertaining to this project.

I am thankful to the Landis lab people: Chris Sebolt, Matt O'neal, Tyler Fox, and Tammy Wilkinson for their friendship and sense of humor in all weather conditions. This project was made possible by the help of many undergraduate students, both in field and lab studies: Andre Ball, Meghan Burns, Jesse Chvojka, Chris Cerveny, Sandra Clay, Alison Gould, Christy Hemming, Matt Lehnert, Allison Lewinski, Kathy McCamant, Andrea McMillian, Kevin Newhouse, Tim Schutz, and Michelle Smith.

I would also like to thank the Department of Entomology, in particular Dr. Fred Stehr, Dr. Ed Grafius and Jill Kolp, who all helped me complete the many forms and requirements necessary for this degree. Likewise, I am grateful to all the teachers that contributed to build up my entomological knowledge and ability to think critically. A special thank you goes to Dr. Jim Miller for his excitement, dedication and enthusiasm to teach in the classroom.

This study would not have been possible without the generous permission by farmers to work in their fields. Thus, I thank Raymond Covert, Lavern Eldred, Roger

iv

DeBruler, Dale Swiler, Jeffrey Wood, and Lloyd Hayhoe. I am indebted to Dr. Les Lewis and Jean Dyer for the provision of most of the armyworms used in this study, and all the support made available to help me initiate and maintain the armyworm colony. I would also like to thank Dr. John Luhman for his dedication and interest in identifying the parasitoids reared. I am very grateful to Kadir Kizilkaya, Emily Smith and Fernando Cardoso, members of the Statistical Consulting Center of the CANR Biometry group, for their advise in the statistical analyses. Personal support for this research came from an INTA-Fulbrigth fellowship.

Special thanks to my dear wife Marisol and my daughter Ailén, whose love, encouragement, understanding and continued support has made the pursuance of this degree possible.

TABLE OF CONTENTS

LIST OF TABLES	ix
LIST OF FIGURES	xi
KEY TO SIMBOLS AND ABREVIATIONS	xiv
CHAPTER 1 INTRODUCTION	1
Influence of landscape characteristics on parasitoid communities in agroecosystems	1
Effect of landscape structure on parasitism of <i>Pseudaletia unipuncta</i> in Michigan	3
Life history and biology of the principal species of the system under study	5
Pseudaletia unipuncta (Haworth)	5
Meteorus communis (Cresson)	8
Glyptapanteles militaris (Walsh)	9
Mechanisms influencing the effect of landscape structure on <i>Pseudaletia unipuncta</i> parasitism: principal hypotheses and interacting factors	12
Alternative hosts	12
Resources for adult parasitoids	13
Host density	14
Temporal and spatial variation	15
Summary and Objectives	16

CHAPTER 2 EFFECTS OF AGRICULTURAL LANDSCAPE COMPLEXITY AND HOST DENSITY ON THE PARASITOID COMMUNITY OF THE ARMYWORM.	
PSEUDALETIA UNIPUNCTA (HAWORTH), IN MICHIGAN	22
ABSTRACT	22
INTRODUCTION	23
MATERIAL AND METHODS	29
Study site	29
Field border vegetation sampling	30
Landscape structure effect on parasitism	30
Host density effect on parasitism	32
Long-term analysis	33
Data analysis	33
RESULTS	34
Field border vegetation sampling	34
Landscape structure effect on parasitism	35
Host density effect on parasitism	37
Long-term analysis	38
DISCUSSION	39

CHAPTER 3

EFFECT OF FOOD RESOURCES, HOST ACCESS, MATING STATUS,	
AND TEMPERATURE ON THE LONGEVITY AND FECUNDITY OF	
ADULT GLYPTAPANTELES MILITARIS (WALSH) AND METEORUS	
COMMUNIS (CRESSON) (HYMENOPTERA: BRACONIDAE),	
PARASITOIDS OF THE ARMYWORM, PSEUDALETIA UNIPUNCTA	
(HAWORTH) (LEPIDOPTERA: NOCTUIDAE)	66
ABSTRACT	66

INTRODUCTION	67
MATERIAL AND METHODS	71
Influence of temperature, water and carbohydrate availability on adult longevity of <i>Glyptapanteles militaris</i>	71
Longevity and fecundity of <i>Meteorus communis</i> with and without carbohydrate resources, hosts, and with different mating status	73
Maximum daily parasitism of Meteorus communis	76
Food effects on male Meteorus communis	77
Data analysis	78
RESULTS	79
Influence of temperature, water and carbohydrate availability on adult longevity of <i>Glyptapanteles militaris</i>	79
Longevity and fecundity of <i>Meteorus communis</i> with and without carbohydrate resources, hosts, and with different mating status	80
Maximum daily parasitism of <i>Meteorus communis</i>	84
Food effects on male Meteorus communis	85
DISCUSSION	85
APPENDICES	104
APPENDIX 1: Record of deposition of voucher specimens	105
APPENDIX 2: Protocol followed to make diet for <i>Pseudaletia</i> unipuncta	108
REFERENCES CITED	110

LIST OF TABLES

Table 1. Table 1. Parasitoid species previously recovered from <i>Pseudaletia</i> unipuncta larvae in Michigan	20
Table 2. Hosts and group of plants on which they feed, reported for the two most important parasitoid species of <i>Pseudaletia unipuncta</i> (Haworth) in Michigan	21
Table 3. Numbers of <i>Pseudaletia unipuncta</i> larvae released and recovered andnumbers of parasitoids reared in a simple and a complex agricultural landscape inIngham County, Michigan, USA	48
Table 4 . Frequency of tree species in hedgerows bordering maize fields in a complex and a simple agricultural landscape in Ingham County, Michigan. Frequency is the proportion of the sample (n= 360 sample points per landscape) in which a species was present in the hedgerows within each landscape type	51
Table 5. Parasitoids emerging from <i>Pseudaletia unipuncta</i> sentinel larvae releasedin a complex and a simple agricultural landscape located in Ingham County,Michigan, USA	53
Table 6. ANOVA of influence of agricultural landscape structure on the percentage parasitism of <i>Pseudaletia unipuncta</i> sentinel larvae by all species (overall parasitism), and by the two main parasitoid species separately, at low host densities, in Ingham County, Michigan, in 2000 and 2001	55
Table 7. ANOVA of the influence of agricultural landscape structure and host density on the percentage parasitism of <i>Pseudaletia unipuncta</i> sentinel larvae by all species (overall parasitism), and by the two main parasitoid species separately, in Ingham County, Michigan, USA, in 2001	59
Table 8 . ANOVA of influence of agricultural landscape structure on the percentage parasitism of <i>Pseudaletia unipuncta</i> sentinel larvae by all species (overall parasitism), and by the two main parasitoid species separately, at low host densities, in Ingham County, Michigan, over five sampling seasons (1993 – 2001).	62
Table 9. ANOVA of the effect of three food regimes (honey + distilled water, distilled water alone, and control without honey and distilled water) and two conditions of temperature (20 and 25 °C) on the longevity of <i>Glyptapanteles militaris</i> adults	90

Table 10. ANOVA of the effect of different food regimes (honey + distilled waterversus a control of distilled water alone), host access (10 hosts provided dailyversus no host provided) and mating condition (initial access to males versus noaccess) on the longevity of <i>Meteorus communis</i> females	92
Table 11. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions on the longevity of <i>Meteorus communis</i> females	95
Table 12. ANOVA of the effect of different food regimes (honey + distilled waterversus a control of distilled water alone) and reproductive conditions on thefecundity (total number of host parasitized) Meteorus communis females	96
Table 13. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated, and unmated females) on the percentage of larvae parasitized (number of host parasitized / number of host offered x 100) of <i>Meteorus communis</i> females. Fecundity was compared restricting the data to the period of life in which females provided with water were still alive	100
Table 14. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated, and unmated females) on pre-reproductive period (number of days previous to progeny production) of <i>Meteorus communis</i> females. Data was transformed (ln $[x + 1]$) before analysis to achieve homocedasticity	102

LIST OF FIGURES

Figure 1 . Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> in a complex and a simple agricultural landscape in Ingham County, Michigan (modified from Marino and Landis 1996, Menalled et al. 1999, and Menalled unpublished data). Means with asterisks are significantly different ($P < 0.05$, F test from slicing by year of sampling), ns = not significant	18
Figure 2. Percentage parasitism per day (mean + SEM) of <i>Pseudaletia unipuncta</i> by (a) <i>Meteorus communis</i> and (b) <i>Glyptapanteles militaris</i> in a complex and a simple agricultural landscape in Ingham County, Michigan (modified from Marino and Landis 1996, Menalled et al. 1999, and Menalled unpublished data). Means with asterisks are significantly different ($P < 0.05$, F test from slicing by year of sampling), ns = not significant	19
Figure 3 . Example of treatments and release site orientation in the field. Relative position of high and low density treatments during each release was random, varying from field to field. Roman numbers indicate release date, rectangles group simultaneous releases, small dots represent low density treatment release sites and larger dots represent high density treatments release sites	49
Figure 4 . The release area (inner dotted rectangle) included three rows of 20 maize plants. Each release site received at random a high or low host density treatment in 2001. Recovery was performed in the release area plus all the plants located within four rows of the release area sides and 3 m of the release area row ends (external dotted rectangle)	50
Figure 5. Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> sentinel larvae in a complex and a simple agricultural landscape in Ingham County, Michigan, USA. Means with different letters are significantly different ($P < 0.05$, Least Square Means difference, adjusted by Tukey-Kramer method)	56
Figure 6 . Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> sentinel larvae by (a) <i>Glyptapanteles militaris</i> and (b) <i>Meteorus</i> spp., in a complex and a simple agricultural landscape in Ingham County, Michigan, USA. Different letters indicate significant differences at $P < 0.05$ (LSM difference, adjusted by Tukey-Kramer method)	57
Figure 7. Number of <i>Pseudaletia unipuncta</i> sentinel larvae per 100 maize plants (mean + SEM) recovered in high host density patches and low host density patches and natural occurrence in the field in 2001, in Ingham County, Michigan, USA. Different letters are significantly different ($P < 0.05$, Least Square Means difference, adjusted by Tukey-Kramer method)	58

Figure 8 . Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> sentinel larvae at high and low density patches in a complex and a simple agricultural landscape in Ingham County, Michigan, USA, in 2001 season	60
Figure 9. Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> sentinel larvae by (a) <i>Glyptapanteles militaris</i> and (b) <i>Meteorus</i> spp., at high and low host density patches, in a simple and a complex agricultural landscape in Ingham County, Michigan, USA, in 2001 season	61
Figure 10 . Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> sentinel larvae in a complex and a simple agricultural landscape in Ingham County, Michigan, USA, including previous studies data (Marino and Landis 1996, Menalled et al. 1999, and Menalled et al. unpublished data). Means with asterisks are significantly different ($P < 0.05$, F test from slicing by year of sampling), ns = not significant	64
Figure 11. Percentage parasitism (mean + SEM) of <i>Pseudaletia unipuncta</i> sentinel larvae by (a) <i>Glyptapanteles militaris</i> and (b) <i>Meteorus</i> spp., in a complex and a simple agricultural landscape in Ingham County, Michigan, USA, including previous studies data (Marino and Landis 1996, Menalled et al. 1999, and Menalled et al. unpublished data). Means with asterisks are significantly different ($P < 0.05$, <i>F</i> test from slicing by year of sampling), ns = not significant	65
Figure 12 . Longevity (mean + SEM) of <i>Glyptapanteles militaris</i> adults under three food regimes (honey + distilled water, distilled water alone, and control without honey and distilled water) and two conditions of temperature (20 and 25 °C)	91
Figure 13. Longevity (mean + SEM) of <i>Meteorus communis</i> females under different food regimes (honey + distilled water versus a control of distilled water alone), host access condition (10 hosts provided daily versus no host provided) and mating condition (initial access to males versus no access)	94
Figure 14 . Fecundity (mean number of <i>Pseudaletia unipuncta</i> larvae parasitized / female during all life + SEM) of <i>M. communis</i> females at different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated and unmated). Columns with different letter differed significantly within food regime ($P < 0.05$, Least Square Means Differences)	98
Figure 15. Daily parasitism (mean number of host larvae parasitized / female / day) of <i>Meteorus communis</i> females at different food regimes and reproductive conditions: a) females provided with honey and b) control females provided only with distilled water	99

Figure 17. Pre-reproductive period (number of days previous to progeny production, mean + SEM) of *Meteorus communis* females at different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated and unmated) 103

KEY TO SYMBOLS AND ABBREVIATIONS

ANOVA	analysis of variance
°C	degrees Celsius
cm	centimeters
d	days
df	degrees of freedom
F	Fisher distribution
g	grams
h	hours
km	kilometers
L: D	light: dark hours
m	meters
ml	milliliters
n	number of observations
ns	not significant
Р	probability under the assumption that the null hypothesis is true
R ²	coefficient of determination
RH	relative humidity
SD	standard Deviation of the Mean
SEM	standard Error of the Mean
spp.	species
t	t-statistic

CHAPTER 1:

INTRODUCTION

Influence of landscape characteristics on parasitoid communities in agroecosystems

A major proportion of the world's biodiversity is composed of insect parasitoids, their phytophagous hosts, and their hosts' food plants (Hawkins 1994). In the past 20 years, parasitoids have been used as case-studies in the development and testing of many ecological and evolutionary theories (Hawkins and Sheehan 1994, Price 1997). In addition, the ecology of parasitoids has been widely studied for their role as biological control agents (DeBach and Rosen 1991, Van Driesche and Bellows 1996). Many studies have shown that parasitoids are the most important natural enemies of many crop pests and can act as keystone species in some ecosystems (LaSalle 1993). In other cases, where parasitoids do not regulate pest population directly, parasitism may suppress densities of host populations sufficiently to allow regulation by other mortality factors (Teder et al. 2000). However, frequent use of pesticides, lack of food and refuges for adults, absence of alternative hosts, and in general high levels of disturbance, result in agricultural habitats that are adverse environments for many parasitoid species (Altieri et al. 1993, Landis and Menalled 1998, Landis et al. 2000, Tscharntke 2000).

As ecological communities, agroecosystems have most of the characteristics of early successional habitats (annual or biennal vegetation, lower food volume, high plant vigor, lower host offer, etc) but differ from natural early successional habitats in that the plants which dominate them are highly apparent (*sensu* Feeny 1976) and generally have

poor defenses against herbivores (Price 1991). In this way they are more similar to plants of late successional habitats, which are highly apparent in space and time, resulting in easier host location (Price 1991). In contrast, natural late successional habitats are composed of more perennial, diverse (in species and structure), and abundant (i.e. in terms of biomass) vegetation, that provide resources to sustain a more diverse and permanent community of herbivores, which in turn support more parasitoid species (Price 1991, 1994; Landis and Menalled 1998). In an analysis of 1289 host-parasitoid complexes, Hawkins (1994) showed that insect herbivores feeding on trees and shrubs have significantly more parasitoid species attacking them than those that feed on monocots or herbs. Increased parasitoid species richness per host species is often associated with increased host regulation (Price 1991). However, most studies only found a correlation between higher rate of mortality and diversity of parasitoids, without demonstrating a cause-effect relationship (Price 1991, Hawkins 1994, Hochberg and Hawkins 1994).

Examining agroecosystems at the landscape level may provide a better understanding of the spatial and temporal factors that influence host-parasitoid interactions. A landscape can be defined as "a heterogeneous land area composed of a cluster of interacting ecosystems that is repeated in similar form throughout" (Forman and Godron 1986). Many parasitoids of agricultural pests find most of their resources in extra-field habitats, and only colonize crop fields to locate hosts (van Emden 1990, Altieri et al. 1993, Landis and Marino 1999, Tscharntke 2000). Therefore, studies of host-parasitoid systems performed at the within-field scale, often fail to accurately predict the interactions between these trophic levels as a result of omitting the dynamics of

populations occurring in extra-field habitats. In contrast, studies performed on a landscape scale may have greater power to explain the spatial and temporal interactions between host and parasitoids in agroecosystems (Tscharntke 2000).

Only a few empirical studies have investigated the relationships between landscape structure and parasitism. Most of them obtained reduced parasitism and parasitoid diversity with increase habitat isolation (Kruess and Tscharntke 1994, Doak 2000, Kruess and Tscharntke 2000b) or habitat fragmentation (Roland and Taylor 1997). Other authors found significantly higher rates of parasitism with increased agricultural landscape complexity, mainly considering field size and the proportion of uncultivated areas in the landscape (Marino and Landis 1996, Menalled et al. 1999, Thies and Tscharntke 1999). In addition, parasitoid species may respond to landscape structure at differing scales (Roland and Taylor 1997, Roland 2000).

Effect of landscape structure on parasitism of *Pseudaletia unipuncta* in Michigan

Marino and Landis (1996) and Menalled et al. (1999) studied the parasitoid complex of the armyworm, *Pseudaletia unipuncta* (Haworth) (Lepidoptera: Noctuidae), in commercial maize (*Zea mays*, L.) fields located in complex and simple agricultural landscapes. These authors characterized two areas of 3.2 km² according to their landscape structure. The complex landscape differed significantly from the simple landscape in that it was composed of more agricultural fields (139 versus 61, respectively) of significantly smaller size (3.4 versus 12.4 ha) embedded in a landscape of abundant hedgerows and woodlots (14.3 versus 11.2% of the area). Fields in the

complex landscape also differed significantly from those of the simple landscape in that they had significantly less perimeter (776 versus 1638 m), less maximum distance to the edge (63 versus 101 m) and more wooded perimeter per unit of field area (23.5 versus 8.7 m/ha). These authors found a significantly higher percentage of parasitism in the complex than in the simple landscape during 1993 and 1996 (Figure 1). But replication of the study in 1998 showed similar levels of parasitism in both systems (Figure 1). Parasitoid species richness was low and did not differ between landscapes (12 species in the complex versus 10 species in the simple). Moreover, individuals of only two species represented 90% of the parasitoids recovered: *Meteorus communis* (Cresson) and *Glyptapanteles militaris* (Walsh) (Hymenoptera: Braconidae). Thus, the population dynamics of these two species appear to play the key role in understanding the dynamics of *P. unipuncta* parasitism in Michigan.

Meteorus communis parasitized approximately 24% of the exposed larvae in the complex landscape during 1993 and 1996, but parasitism declined in 1998, to approximately half of its previous level, 12% parasitism (Figure 2a). Significantly less *M. communis* individuals were reared from *P. unipuncta* in the simple landscape during the whole period of study (2% on average) (Figure 2a). Parasitism by *G. militaris* was very low in both landscapes in 1993 and 1996 (< 2% on average), but reached around 27% parasitism in the simple landscape in 1998, and was significantly different from the 9% parasitism obtained in the complex landscape (Figure 2b). This indicates that *G. militaris* alone is responsible for increased parasitism in the simple landscape and contributed about half of the overall parasitism in the complex landscape in 1998. Both species showed differential responses to landscape complexity. *Glyptapanteles militaris*

parasitized *P. unipuncta* in both landscapes, but predominated in the simple landscape in years with higher parasitism rates. In contrast, *Meteorus communis* was largely restricted to the complex landscape.

After three sampling seasons (during a period of 6 years), the hypothesis of higher parasitism and species richness in the complex landscape than in the simple landscape, originally proposed and supported by Marino and Landis (1996), was supported in two of the three years of study. In the complex landscape, the presence of two major species of parasitoids appeared to reduce year-to-year variability of parasitism. In the simple landscape, on the other hand, parasitism depended in large part on the presence of G. *militaris*. Thus, the impact of parasitism may be less stable in simple versus in the complex landscapes. Parasitoid species richness was almost the same in both landscapes and remained constant from 1993 - 1998.

Life history and biology of the principal species of the system under study

Pseudaletia unipuncta (Haworth)

The armyworm, *Pseudaletia unipuncta* (Lepidoptera: Noctuidae) is a cosmopolitan pest that prefers to feed on grasses, especially maize, timothy (*Phleum pratense* L.), millet (*Panicum miliaceum* L.), and small grains such as wheat (*Triticum aestivum* L.), oat (*Avena sativa* L.), barley (*Hordeum vulgare* L.), rye (*Secale cereale* L.) and rice (*Oryza sativa* L.), but also attacks sugar cane (*Saccharum officinarum* L.) and sorghum (*Sorghum bicolor* (L.) Moench) (Breeland 1958, Metcalf and Metcalf 1993). The larvae of *P. unipuncta* also eat legumes and other plants when under stress or starved

(Breeland 1958, Metcalf and Metcalf 1993). They usually occur in small populations, attracting little attention; but at irregular intervals of 5 to 20 years, *P. unipuncta* larvae develop widespread outbreaks that produce serious injury to forage grasses and cereal crops (Guppy 1961, Untung 1978). The damage is produced by larval feeding, beginning with the skeletonization of the leaf surface in early instars and ending with the consumption of all the tissues in latter stages (Guppy 1961). The last instars produce the most damage, and since they become nocturnal feeders in the third instar, their presence is often not noticed until the crop is nearly destroyed (Breeland 1958, Metcalf and Metcalf 1993). When the available food in one field cannot support their numbers, the larvae disperse. These migrations can involve huge numbers of larvae marching to adjacent fields, resulting in the common name of "armyworm" (Metcalf and Metcalf 1993).

The life cycle of *P. unipuncta* involves several generations per year depending on the latitude, ranging to five broods in Tennessee to two in Canada (Breeland 1958, Guppy 1961). In Michigan, *P. unipuncta* may overwinter as middle instar larvae, pupae or adults, in sufficient numbers to result in subsequent seasonal build-up (Thompson 1966). However, in most years the majority of the population results from adults that migrate into Michigan from the south (Untung 1978). During 1964 and 1965, field studies in Michigan documented the presence of adults in early May and an outbreak of the population at Galien, Michigan, in late June to early July (Thompson 1966). In 1976, field studies in Southern Michigan detected two main adult population peaks, the first in mid-April, which was followed by a larval development period from mid May to the end

of June. The second adult flight occurred in July, with larvae following during mid July to mid August (Untung 1978).

Adult survival of *P. unipuncta* can be up to 24 d with and averages 10.1 ± 0.4 d for females and 9.0 ± 0.4 d for males (Breeland 1958). Females lay their eggs in masses of several rows covered with a white adhesive material, on a dry substrate such as straw, maize stubble, or dry leaves (Untung 1978). Females can lay up to 1800 eggs in several days, with a mean of 454 eggs per female (Breeland 1958). Eggs hatch in 3-14 d (averaging 6.4 ± 1.6 d), failing to hatch during hot and dry periods, probably due to infertility (Breeland 1958). Larval development usually involves six instars and is accelerated by increasing temperature up to 33.3 °C (McLaughlin 1962). Breeland (1958) reared larvae at different daily temperatures (means ranging from 17.3 to 23.6 °C) obtaining a mean larval development period of 27.9 ± 1.1 d. After consuming sufficient food, the larvae crawl into the soil where the prepupa forms the pupal cell and molts to form the pupa (Breeland 1958). Pupae are normally found beneath dead foliage, clods of dirt, or cracks in the soil, and are difficult to locate in the field (Breeland 1958). Untung 1978). The average pupation duration is 15.1 ± 1.8 d (Breeland 1958).

Sixty-nine parasitoid species and 12 associated hyperparasites have been reported to attack the larval and pupal stage of *P. unipuncta*, 35 of these species occur in the Great Lakes Region of the United States (Guppy 1967, Marino and Landis 1996). The diversity of parasitoids and the ease by which *P. unipuncta* is reared allow it to be a suitable species to test the influence of the landscape structure on larval parasitoid species richness and overall percentage of parasitism (Marino and Landis 1996). Table 1 indicates the parasitoids that have previously been reported from Michigan collections of

P. unipuncta (Untung 1978, Marino and Landis 1996, Menalled et al. 1999). The three species that dominate the parasitoid communities of P. unipuncta in North America include: *G. militaris* and *M. communis*, and *Winthemia rufopicta* (Diptera: Tachinidae) (Breeland 1958, Guppy 1967, Untung 1978, McNeil and Turgeon 1988, Marino and Landis 1996, Menalled et al. 1999). Marino and Landis (1996) and Menalled et al. (1999) did not find great numbers of *W. rufopicta* probably because they collected 3rd to 5th instar *P. unipuncta* larvae and this species prefers to parasitize the 5th and 6th instars (Breeland 1958, Guppy 1967).

Meteorus communis (Cresson)

The genus *Meteorus* Haliday (Hymenoptera: Braconidae) is comprised of solitary and gregarious parasitoids attacking Coleoptera and Lepidoptera (Krombein et al. 1979, Huddleston 1980). The mature larvae of some species spin an oblong cocoon suspended by a thread resembling a meteor, and it is from this habitat that the name of the genus is derived (Huddleston 1980). Despite the potential usefulness of this genus in biological control programs, the biology of some species is poorly studied (Huddleston 1980), West and Miller 1989).

Meteorus communis (Cresson) is a native generalist parasitoid that primarily attacks larvae of Noctuidae (Table 2) (West and Miller 1989). Its distribution includes northern U.S. states to North Carolina, Colorado and California; Canada and Alaska (Krombein et al. 1979). It is a solitary, koinobiont endoparasitoid. The first instar is mandibulate and actively eliminates supernumerary conspecifics and presumably, larvae of other parasitoid species as well (West 1988). It oviposits into second to fourth instar

hosts in the field. It typically emerges and kills the larvae in the fifth instar (Krombein et al. 1979, McNeil and Turgeon 1988, West 1988). In western Oregon, M. communis has 3-4 overlapping generations, and was found parasitizing up to 40% of the noctuid larvae sampled in peppermint and alfalfa fields (West 1988). Burbutis and Stewart (1979), using light traps as sampling method, found that *M. communis* followed 2 of the 3 flight peaks of P. unipuncta in New Jersey. The larvae overwinters inside the host larvae (West 1988) although at least some species of *Meteorus* overwinter in different host species from those which they attack in their summer generations (Huddleston 1980). The time for development depends on both temperature and the stage of host attacked, with shorter development in more advanced instars (West 1988, Miller 1996). Laboratory studies showed that the development from oviposition to adult emergence on *Peridroma saucia* (Hübner) (Lepidoptera: Noctuidae) range from 16.7 ± 0.6 d (at 31 °C) to 61.8 ± 3.4 days (at 14 °C), requiring 274 \pm 14.5 degree days above a developmental threshold of 9.5 \pm 0.5 °C to complete (Miller 1996). Prior to the studies reported here, there are no published references regarding longevity, fecundity and the food resources used by adult M. communis.

Glyptapanteles militaris (Walsh)

Glyptapanteles militaris is a native, koinobiont, generalist endoparasitoid that attacks several species of noctuids and isolated species of lymantrids and tortricids (Table 2) (Krombein et al. 1979, Oliveira et al. 1998). It has been found to be the most abundant parasitoid of *P. unipuncta* in several studies in North America (Breeland 1958, Calkins and Sutter 1976, Steinkraus et al. 1993). In addition, Whitfield and Lewis (2001) found

that this species was the most abundant braconid in a survey of six Midwestern U.S. tallgrass prairies, using Malaise traps. *Glyptapanteles militaris* was collected from May to June in Tennessee and northwest Arkansas, attacking 11.7% and 8.1% of the larvae of *P. unipuncta* recovered (Breeland 1958, Steinkraus et al. 1993). In Michigan, Untung (1978) found *G. militaris* was the most abundant parasitoid of *P. unipuncta* collected in wheat fields in 1977. He obtained two peaks of *G. militaris*: May 24-27 (48% parasitism) and June 7 (54% parasitism). However, the same author failed to collect *G. militaris* in the preceding year (1976), when the most abundant parasitoid was *W. rufopicta*. Rolim (1983) also found that *G. militaris* was the most abundant parasitoid of *P. unipuncta* in Michigan, reporting 18.4 - 54.4% of larvae parasitized during June and July of 1980. Although there are no conclusive studies, evidence suggests that *G. militaris* is capable of overwintering as cocoon in the south in the United States, and as larvae inside its host in the north in the United States (Tower 1915, Breeland 1958).

Glyptapanteles militaris is a gregarious arrhenotokous parasitoid (Tower 1915, Rolim 1983). Tower (1915) described with detail the morphology of immature stages and characterized aspects of its biology. Each female parasitoid made 1- 6 oviposition probes in different parts of the host body. Each oviposition usually lasted no longer than 1-2 seconds, and in a period of four hours with surplus of hosts available, 9 hosts could be parasitized, producing an average of 512 larvae (Calkins and Sutter 1976). Laboratory studies showed that *G. militaris* could successfully parasitize all *P. unipuncta* instars (Rolim 1983). Higher percent parasitism was obtained between L₂ and L₄ (around 60%), with lower rates at L₁ (40%) and very low in L₅ (7.5%) (Oliveira et al. 1999). Rolim (1983) found similar results but also obtained 5% parasitism on the 6th instar. The

percentage of parasitism seems to be independent of the age of females (using 1, 3, and 5 day old females, Rolim 1983). The total number of larva per host obtained significantly increased with host stage (ranging from 40.2 ± 21.4 larvae in L₁ to 82.3 ± 31.8 larvae in L₅), but not the number of cocoons obtained (ranging from 36.4 ± 19.4 cocoons in L₁ to 55.0 ± 22.7 cocoons in L₄) (Oliveira et al. 1999).

Larvae of *G. militaris* emerge as 3^{rd} instars within several minutes of each other, and almost always from the 6th host instar (Calkins and Sutter 1976). As a consequence, developmental time must decrease with increasing host age. Oliveira et al. (1999) found significant differences in the developmental time of parasitoids that develop on L₁ stage (egg-pupal development: 31.7 d) and those on L₄ (egg-pupal development: 16.3 d). Development time of *G. militaris* was significantly decreased with increasing temperatures (Calkins and Sutter 1976, Oliveira et al. 1998). Calkins and Sutter (1976) obtained egg-larvae developmental periods of 31.6 d at 21.1 °C and 12.5 d at 26.7 °C. Adult longevity was 3-4 d without food, and 6-7 d with food (fresh halved grapes), at 26.7 °C, and was prolonged to 10 days at 10 °C (Calkins and Sutter 1976). Oliveira et al. (1998) reported a shorter period of adult life but cited methodological factors that could affect it. Exposure of parasitized *P. unipuncta* to 35 °C results in the death of parasitoids without altering the normal development of the host (Kaya and Tanada 1969).

Field cage experiments showed that increasing *G. militaris* female density resulted in higher rates of parasitism of *P. unipuncta* (Rolim 1983). The same study showed an increase in the number of hosts attacked as the number of hosts exposed was increased, although not statistically significant. On the other hand, percent parasitism did not increase with increasing number of hosts exposed. Under all conditions, the third

instar was the most preferred. Calkins and Sutter (1976) concluded that given the efficiency showed in laboratory and field trials, and the easy methods involved in its rearing, *G. militaris* should be considered as a suitable biological control agent for *P. unipuncta*.

Mechanisms influencing the effect of landscape structure on *Pseudaletia unipuncta* parasitism: principal hypotheses and interacting factors.

Alternative hosts

Pseudaletia unipuncta spring populations of Michigan and Canada are most likely the product of migration from the Southern States (Thompson 1966, Untung 1978, Fields and McNeil 1984). Some evidence suggests that *M. communis* and *Glyptapanteles militaris* overwinter as larvae inside their hosts, although there are no conclusive studies (Tower 1915; Breeland 1958, West, 1988). Therefore, the typical low abundance of *P. unipuncta* populations in Michigan (i.e. except during infrequent outbreaks periods), leads to the hypothesis that alternative hosts may be involved in the life history of *G. militaris* and *M. communis*. Marino and Landis (1996) and Menalled et al. (1999) stressed the importance of the study of the alternative hosts of these parasitoid species in order to explain their spatial and temporal variations. Thompson (1966) studied the fluctuations of noctuid (Lepidoptera) pests in Michigan, using light traps, during 1964 and 1965. He found that *Feltia* sp. (one species is host of *G. militaris*) was the most abundant moth, with high populations in July and August (around 400 adults / 10 d / trap). The second species was *P. unipuncta* with fluctuations between 50 and 100 adults /

10 d / trap, in June and July. This sampling was coincident with an outbreak of *P*. *unipuncta*. Small populations (less than 20 adults / 10 d / trap) of Agrotis ipsilon (host of *M. communis*) and *Peridroma saucia* (host of both species) were found in June and July. Additionally, *Helicoverpa zea* (host of *M. communis*) was briefly recorded in August. Thus, during the period covered by this study, only *Feltia* sp. larvae occurred late in the season. There is no information available about the phenology of other hosts recorded for these parasitoid species.

Resources for adult parasitoids

Certain adult Hymenopteran parasitoids host-feed to obtain nutrients, but many require other sources of food (Godfray 1994, Quicke 1997, Gordh et al. 1999, Landis et al. 2000). The presence of non-host adult food resources in extra-field habitats is postulated as an important feature that contributes to the establishment and permanence of parasitoid populations near crop fields (Landis et al. 2000, Tscharntke 2000). Parasitoids use floral and extrafloral nectar, pollen, honey, honeydew, and other sugar solutions as natural sources of non-host food in the field (Bugg et al. 1989, Iodine and Ferro 1990, Jervis et al. 1993, Jervis and Copland 1996, England and Evans 1997, Lewis et al. 1998, Jacob and Evans 1998). Laboratory trials have shown that carbohydrate resources contribute to prolonged adult longevity and increase female fecundity (Leius 1963, Foster and Ruesink 1984, Idris and Grafius 1995, van Driesche and Bellows 1996, Dyer and Landis 1996, McDougall and Mills 1997, Jacob and Evans 2000, Olson et al. 2000, Bautista et al. 2001, Fadamiro and Heimpel 2001, Wäckers 2001, Rivero and West 2002). In addition, availability of non-host food increases the proportion of time

parasitoids spend searching for hosts instead for food, contributing to enhance field parasitism (Foster and Ruesnik 1984, Křivan and Sirot 1997, Lewis et al. 1998, Tscharntke 2000). Mating status and host availability also affect adult parasitoid longevity and fecundity, and interact with the availability of food resources (Godfray 1994, Jervis and Copland 1996). Mated females of Bathyplectes curculionis (Hymenoptera: Ichneumonidae) provided with water alone, lived significantly less days than virgin females, but had similar life spans when provided with sugar resources (Jacob and Evans 2000). In other studies, however, mating status did not affect parasitoid female longevity and fecundity (Sousa and Spence 2000, King 2001). Females of Cotesia melitaearum (Wilkinson) (Hymenoptera: Braconidae) prevented from contacting hosts lived longer than those allowed to lay eggs (Lei et al. 1997). The effects of these factors on the biology of G. militaris and M. communis have not been studied. However, knowledge about use of adult resources and the interaction of this with other factors will contribute to provide explanations and formulate predictions regarding the distribution and abundance of this parasitoid species in their habitats and in different landscapes.

Host density

Host density effects have been widely investigated in laboratory and field conditions, showing an increase (density dependent response), decrease (inverse density independent response) or no effect (density independent response) on the level of parasitism (Hassell and Waage 1984, Reeve 1987, Stiling 1987, Latto and Hassell 1988). Most of these studies were focused on specific host-parasitoid systems (population-level response) and did not show an effect of host density on the diversity of parasitoids

recovered (community-level response). Increased host density could promote the collection of more parasitoid species just by an increase of sampling size effect. However, it is also possible that some species only detect hosts at higher host densities. Therefore, the contrast between species rich and species poor parasitoid communities should be increased with increased sample size. The study of the levels of parasitism of *P. unipuncta* performed by Marino and Landis (1996) and Menalled et al. (1999) involved relatively low densities of host exposed (one larvae per maize plant). Since *P. unipuncta* is capable of producing very high host densities under outbreaks conditions, it is important to investigate the effect of host density on parasitism and on the response of parasitoid communities in structurally different landscapes.

Temporal and spatial variation

In Michigan, the pattern of landscape effect on parasitism in one study region found in 1993 by Marino and Landis (1996) was the same as found in the same region in 1996 by Menalled et al. (1999). However, the pattern failed to be replicated spatially in two additional regions (Menalled et al. 1999). Because periodic oscillations frequently occur in parasitoid species richness and abundance (Askew and Shaw 1989, Hawkins 1994), studies done at one particular time may not reveal the actual importance of extrafield habitats in conditioning parasitoid-host relationships (Menalled et al. 1999). As we previously described, replication in time also showed differences in the patterns of parasitism (Menalled et al. unpublished data). Since 1993, fluctuations in the levels of parasitism were higher in the simple landscape than in the complex one (Figure 1), which is an indication of less stability in the system. These studies, however, were performed at

intervals of two to three years and several authors have reported year-to-year oscillations in parasitoid species richness and percentage parasitism in cropping and non-cropping systems (Askew and Shaw 1989, West and Miller 1989, Onstad et al. 1991, Hawkins 1994, Cossentine and Jensen 1995, Jones and Weinzierl 1997). Therefore, to obtain an accurate estimate of the variability of parasitism at different landscapes it is necessary to incorporate a shorter interval of sampling (i.e. sampling in continuous years), in addition to long-term sampling.

Summary and Objectives

A number of studies have documented the effect of landscape structure on parasitism over several years (Kruess and Tscharntke 1994, Marino and Landis 1996, Roland and Taylor 1997, Cappuccino et al. 1998, Golden and Crist 1999, Menalled et al. 1999, Thies and Tscharntke 1999), but very few have studied both temporal and spatial scale at the same time (Teder et al. 2000). In addition, the use of natural host densities in most of these studies prevents relating host density and parasitoid diversity in a causeeffect manner. Studies that provide data for mechanistic explanations of the patterns of parasitism observed in structurally different landscapes should include parasitoid biological features that not only affect host parasitoid relationship (Godfray 1994), but also condition landscape effect on parasitism (Roland and Taylor 1997, Teder et al. 2000). The degree of dependence of parasitoids on several resources for larval (hosts), pupal (hosts or refuges) and adult (food, refuges) stages is a possible mechanism that may explain their relation with different landscapes that provide these resources.

Previous studies showed that parasitism of *P. unipuncta* was higher in a complex than in a simple landscape in two of three seasons sampled, revealing greater variability of parasitism in the simple landscape (Marino and Landis 1996, Menalled et al. 1999). The use of controlled host densities could provide a better contrast between landscapes and allow to a more realistic estimation of parasitoid diversity. The two main parasitoid species of the system have differences in their reproductive strategies and alternative host species, but their use of food or host resources and their influence in the biology of the adult wasps requires more study.

Thus, the objectives of my research were to:

- Determine if there are changes in the percentage and diversity of parasitism of *P. unipuncta* in consecutive years as a function of agricultural landscape complexity,
- 2) Determine if the percentage parasitism and parasitoid species diversity respond to host density, and
- 3) Determine the influence of food, hosts, temperature, and mating status on the longevity and fecundity of *G. militaris* and *M. communis*.

These objectives will also contribute to a long-term study of *P. unipuncta* parasitism in contrasting landscapes and provide a better understanding of the effect of landscape complexity on parasitism and parasitoid diversity of generalist herbivores. Knowledge of the impact of landscape structure on parasitism of agricultural pests is necessary for the development of conservation biological control strategies by habitat management.



Figure 1. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* in a complex and a simple agricultural landscape in Ingham County, Michigan (modified from Marino and Landis 1996, Menalled et al. 1999, and Menalled unpublished data). Means with asterisks are significantly different (P < 0.05, F test from slicing by year of sampling), ns = not significant.



Figure 2. Percentage parasitism per day (mean + SEM) of *Pseudaletia unipuncta* by (a) *Meteorus communis* and (b) *Glyptapanteles militaris* in a complex and a simple agricultural landscape in Ingham County, Michigan (modified from Marino and Landis 1996, Menalled et al. 1999, and Menalled unpublished data). Means with asterisks are significantly different (P < 0.05, F test from slicing by year of sampling), ns = not significant.
Order	Family	Species
Hymenoptera	Braconidae	Meteorus communis (Cresson)
		Glyptapanteles militaris (Walsh)
		Diclogaster auripes (Provancher)
		Rogas terminalis (Cresson)
	Ichneumonidae	Therion sassacus Viereck
		Nepiera marginata (Provancher)
		Enicospilus purgatus (Say)
		Netelia (Netelia) sayi (Cushman)
		Euplectrus mellipes (Prov.)
		Hyposoter sp.
		Campoletis spp.
		Ichneumon spp.
Diptera	Tachinidae	Winthemia rufopicta (Bigot)
		Periscepsia helymus (Walker)
		P. laegivata (Wulp)
		Archytas apicifer Wlk.
		Chaetoplagia atripennis Coquilett

Table 1. Parasitoid species previously recovered from *Pseudaletia unipuncta* larvae inMichigan¹.

¹From Untung (1978), Marino and Landis (1996), Menalled et al. (1999), and Menalled et al. (unpublished data).

Table 2. Hosts¹ and group of plants on which they feed, reported for the two most important parasitoid species of *Pseudaletia unipuncta* (Haworth) in Michigan.

Glyptapanteles militaris ²	Host	ts ³		_	Meteorus communis ⁴	Hos	ts ³		
Noctuidae					Noctuidae				
-Pseudaletia unipuncta	G*	F			-Pseudaletia unipuncta	G*	F		
-Peridroma saucia (Hübner)	G*	F	S	Т	-Peridroma saucia	G*	F	S	Т
-Epiglaea apiata (Grote)			S		- Agrotis ipsilon (Hufnagel)	G*	F		
-Euxoa auxiliaries (Grote)	G*	F			-Dargida procincta (Grote)		F		
-Feltia ducens Wlkr.	G*				- Autographa californica (Speyer)		F		
-Helicoverpa zea (Boddie)	G*	F			-Amphipyra pyramidoides Guenée			S	Т
-Leucaenia latiustula H. –S.	G				-Xylena nupara (Lintner)				Т
-L. phragmatidicola Guenée	G				-Eupsilia sidus (Guenée)				Т
-Spodoptera exugia (Hübner)	G*	F			-Lithophane laticinerea Grote				Т
-S. frugiperda (J. E. Smith)	G*	F			-L. bethunei (Grote and Robinson)				Т
-S. ornithogalli (Guenée)	G*	F			-Orthosia hibisci (Guenée)				Т
					-O. revicta (Morrison)			S	Т
					<u>Lymantriidae</u>				
					-Malacosoma americanum (Fabricius)				Т
					Tortricidae				
					-Grapholitha sp				Т
Total	10	7	2	1		3	5	3	10

¹ only North American species included

² from Krombein et al. (1979)

³ from Covell (1984), Stehr (1987) and Metcalf and Metcalf (1993); abbreviations: G = grasses, F = forbs, S

= shrubs, T = trees, * present in maize fields

⁴ from Marino and Landis (1996)

CHAPTER 2:

EFFECTS OF AGRICULTURAL LANDSCAPE COMPLEXITY AND HOST DENSITY ON THE PARASITOID COMMUNITY OF THE ARMYWORM, *PSEUDALETIA UNIPUNCTA* (HAWORTH), IN MICHIGAN.

ABSTRACT

The armyworm, *Pseudaletia unipuncta* (Haworth) (Lepidoptera: Noctuidae), is a sporadic pest of maize and cereal grains in the United States. Previous studies have shown that under conditions of constant host density, parasitism of P. unipuncta can be influenced by landscape complexity. Experiments were conducted to determine how landscape structure and host density affect parasitism and parasitoid diversity of P. unipuncta in 2000 and 2001. The study site consisted of a complex and a simple agricultural landscape separated by a transition zone. The complex landscape was composed of a highly heterogeneous mixture of crop and non-crop habitats, while the simple landscape contained few non-crop habitats. Pseudaletia unipuncta sentinel larvae were used to assess parasitism in commercial maize fields. Neither landscape structure nor host density significantly influenced overall parasitism (80.2% on average) or parasitoid diversity (2.8 species per field on average). Two braconid wasp species accounted for 98.4% of the observed parasitism. *Glyptapanteles militaris* (Walsh) parasitized a significantly greater proportion of hosts at low than at high host density, while the opposite occurred with Meteorus communis (Cresson). These offsetting responses of the major parasitoid species resulted in a lack of host density effect on overall parasitism. These results contrast with previous findings in this system of

increased parasitism in the complex landscape in 1993 and 1996. Possible reasons for these changes in the pattern of parasitism include: 1) the presence of an outbreak population of *P. unipuncta* during 2001, that might have allowed *M. communis* to colonize the simple landscape, and 2) the higher population of *G. militaris* than in previous seasons, that could allow this species to invade the complex landscape. These results demonstrated that studies directed to address landscape-scale effects on insect communities need also to incorporate a sufficiently wide temporal-scale to obtain accurate descriptions of the system.

KEY WORDS: agroecosystems; landscape structure; host patch size; parasitism; parasitoid diversity; Noctuidae; Braconidae; *Glyptapanteles militaris; Meteorus communis*

INTRODUCTION

Lack of food, refuges, and alternative hosts as well as frequent use of pesticides and other disturbances due to management practices can reduce parasitoid diversity and abundance in annual crop fields (Altieri et al. 1993, Landis and Menalled 1998, Landis et al. 2000, Tscharntke 2000). Consequently, many parasitoid species find most of their resources in extra-field habitats and enter crop fields only to locate hosts (van Emden 1990, Altieri et al. 1993, Corbett and Rosenheim 1996, Marino and Landis 1996, Landis and Menalled 1998, Thies and Tscharntke 1999, Tscharntke 2000). Extra-field habitats can provide alternative food sources, shelter and favorable microclimates, alternative

hosts, or a combination of these resources (Altieri et al. 1993, van Driesche and Bellows 1996, Dyer and Landis 1996, Barbosa 1998, Landis et al. 2000). Consequently, the abundance and diversity of natural enemies within agricultural landscapes may be influenced by the presence of non-crop habitats (Elliot et al. 1998, Thies and Tscharntke 1999, Nicholls et al. 2001). Price (1991, 1994) has proposed that late successional habitats dominated by trees and shrubs may harbor a more abundant and diverse community of herbivores than herbaceous habitats, and concomitantly, a higher number of generalist parasitoids. In addition, empirical evidence has shown that insect species that feed on trees, including external feeders, support a richer community of parasitoids than those that feed on monocots and herbs (Askew 1980, Hawkins and Lawton 1987, Hawkins 1994). However, parasitoid interactions with habitat structure have traditionally been studied in annual field crops at a within-field scale level of analysis (Landis and Haas 1992, Pavuk and Barret 1993, Williams et al. 1995, Ogol et al. 1998, Golden and Crist 1999).

Few empirical studies have examined the impacts of landscape structure on insect parasitism. Kruess and Tscharntke (1994) found that parasitism of the communities of stem borers and seed feeders of red clover (*Trifolium pratense*) was reduced from 60 to 19% with increased habitat fragmentation. In the same study these authors demonstrated increased species loss (of both phytophagous and parasitoid species) with increase habitat fragmentation. They postulated that habitat fragmentation acted to reduce parasitism in their system by a reduction of the connectivity (measured as distance) of more isolated red clover patches with areas of natural populations of red clover. A similar study showed similar result for the parasitism of the community of insect seed feeders of *Vicia*

sepium L. (Kruess and Tscharntke 2000b). Roland and Taylor (1997) showed that parasitism of Malacosoma disstria Hübner (Lepidoptera: Lasiocampidae) in aspen forests was also significantly affected by landscape structure (defined as the level of deforestation of the area). The response, however, varied among the four parasitoid species collected (three Tachinidae and one Sarcophagidae: Diptera). Three of the parasitoid species caused higher parasitism in continuous forests. By contrast, the smallest parasitoid species attacked more hosts in fragmented forest. In addition, parasitism by different parasitoid species responded to forest structure at different scales. They proposed that forest structure affected parasitism rates by affecting the movement of the parasitoid species of the system. Another study performed by Thies and Tscharntke (1999), showed that parasitism of *Meligethes aeneus* (Coleoptera: Nitidulidae) on oilseed rape (Brassica napus) was significantly increase in proximity to fallow fields and in structurally complex landscapes with up to 50% uncultivated habitats. The parasitoid species collected in this study overwinter as pupa in the soil and are negatively affected by agricultural practices. Therefore, they suggested that the increased proportion of undisturbed areas in the complex landscape allowed parasitoid populations to build up over years and to enhance parasitism in the crop fields. In addition, Doak (2000) reported significantly more overall parasitism of Itame andersoni (Lepidoptera: Geometridae) in small ($< 0.30 \text{ m}^2$) and medium ($0.30 - 1.56 \text{ m}^2$) size, isolated and surrounded patches of its host plant, Dryas drummondii, than in large (2.73 – 20 m^2) and extra large (> 150 m²) size, surrounded patches. However, parasitism by the three more abundant parasitoid species responded to different patches sizes and level of isolation.

Marino and Landis (1996) and Menalled et al. (1999) obtained significantly higher parasitism of *Pseudaletia unipuncta* Haworth (Lepidoptera: Noctuidae) in a complex than in a simple agricultural landscape in Michigan, during 1993 and 1996. These authors characterized two agricultural landscapes based on structural and compositional diversity. The complex landscape contained an increase proportion of non-cultivated areas and was composed of fields with significantly less area, and taller and wider field borders. In contrast, the simple landscape consisted of fewer but bigger areas, most of them cultivated, and with few non-cultivated areas, and narrower and shorter field borders. They obtained significantly higher P. unipuncta parasitism in maize fields located in the structurally complex landscape than in the simple landscape during 1993 (24.1 versus 6.9%, respectively) and 1996 (30.0 versus 5.8%, respectively) (Marino and Landis 1996, Menalled et al. 1999). However, no significant difference in overall parasitism was obtained in 1998 (24.4 versus 31.2%, respectively) (Menalled et al. unpublished data). In these studies, two braconid wasps, Meteorus communis (Cresson) and Glyptapanteles militaris (Walsh), represented more than 90% of the parasitoids recovered from P. unipuncta. Agricultural landscape structure influenced both parasitoid species assemblages and the temporal stability of armyworm parasitism. Meteorus communis was significantly more abundant in the complex landscape during the three sampling seasons. Glyptapanteles militaris was present in very low numbers in 1993 and 1996, with no significant differences between landscapes. In contrast to M. communis, G. militaris was significantly more abundant in the simple than in the complex landscape during 1998, when higher numbers were collected. These results allowed these authors to propose a significant effect of landscape structure on this two

parasitoid species, with higher parasitism by *M. communis* in the complex landscape and higher parasitism by *G. militaris* in the simple landscape.

Parasitoid host density also strongly influences host-parasitoid interactions. The consequences of host density on parasitism level have been widely investigated in laboratory and field conditions. Studies on different parasitoid species have revealed an increase (density dependent response), decrease (inverse density dependent response) or no difference in parasitism (density independent response) as a result of host density (Hassell and Waage 1984, Lessells 1985, Reeve 1987, Stiling 1987, Latto and Hassell 1988). However, most of these studies focused on specific host-parasitoid systems (population-level response) and did not measure the effects of host density on the diversity of parasitoids (community-level response).

The effect of an interaction between host density and landscape structure on parasitism has not been addressed by host density-manipulation studies. Observational studies indicate that parasitism by some parasitoid species could be independent (Thies and Tscharntke 1999, Doak 2000, Kruess and Tscharntke 2000a), positively, or negatively affected by host density (Roland and Taylor 1997, Doak 2000), depending on the level of habitat fragmentation. Furthermore, the response of some parasitoid species to host density interacted with landscape structure, while others showed an independent response (Roland and Taylor 1997). In addition, the relation between rates of parasitism and host density may vary at different spatial scales (Teder et al. 2000).

The armyworm, *Pseudaletia unipuncta* (Haworth) (Lepidoptera: Noctuidae), is a cosmopolitan pest that feeds on small grain crops, maize (*Zea mays* L.), sugar cane (*Saccharum officinarum L.*), millet (*Panicum miliaceum* L.), and sorghum (*Sorghum*

bicolor (L.) Moench) (Breeland 1958, Metcalf and Metcalf 1993). Outbreaks of *P. unipuncta* take place at intervals ranging from 5 to 20 years, and can produce serious injury to forage grasses and cereal crops (Guppy 1961, Untung 1978). Many parasitoid (69) and hyperparasitoid (12) species attack *P. unipuncta* larvae and pupae, and 35 of these species are reported to occur in the Great Lakes region of the United States (Breeland 1958, Guppy 1967, Untung 1978, McNeil and Turgeon 1988).

The purpose of this study was to gain further insights into the effect of landscape structure on the parasitism and parasitoid diversity of *P. unipuncta* in Michigan. The increase parasitism by G. militaris obtained in 1998 disrupted the previous pattern of higher parasitism in the complex landscape found in 1993 and 1996. Therefore, the question of whether 1998 was an odd year and that usually G. militaris is not abundant in the system, or whether this species is more permanently present in the system and contributes to reduce differences between landscapes, promoted further study of this system to search for a clear pattern. In addition, only a relatively low host density was used to assess the parasitism level in the previous studies, and this could contribute to decrease the contrast between landscape structures. Specifically, the objectives of this study were to determine 1) if there are changes in the percentage and diversity of parasitism of *P. unipuncta* through a narrow scale of time (i.e. consecutive years) as a function of agricultural landscape complexity, 2) if percent parasitism and parasitoid species diversity respond to host density, and 3) if there is an interaction between the effects of landscape structure and host density in the percentage and diversity of parasitism of *P. unipuncta*.

MATERIAL AND METHODS

Study site

The study site consisted of a highly heterogeneous agricultural area (hereafter termed "complex landscape") and a more homogeneous agricultural area ("simple landscape") in Leslie and Onondaga Townships, Ingham County, Michigan (42°25'30") N, 84°29'00'' W). Both landscapes were areas of 3.2 x 3.2 km, and were separated by a transition zone of 6.5 km, with the simple landscape north of the complex landscape. These areas represent points along a continuous gradient of agricultural landscapes typical of southern Michigan (Marino and Landis 1996). Using black and white aerial photographs (1: 2000) and digital land-use data from the Michigan Resource Inventory System (Michigan Department of Natural Resources), Marino and Landis (1996) showed significant differences in the landscape structure of the study areas. Sixty-one fields were present in the simple landscape in contrast to 139 present in the complex landscape. The complex landscape contained less percentage of total cover (area) with crops (59.4%) and more with deciduous forest vegetation (14.3%) than the simple landscape (71.4%) and 11.2%, respectively). The comparison between 30 fields selected at random per landscape showed significant structural differences between both landscapes. Fields in the complex landscape were of significantly smaller size, had less proportion of crop border to uncultivated border, wider edges, shorter perimeter and maximum distance from field edges to centers, and more wooded perimeters per unit of field area than those in the simple landscape. The number of edges per field, the number of edge types per field (characterized as: woodland, shrubland, wide hedgerows [> 10 m], narrow

hedgerows [5 – 10 m], herbaceous old field, herbaceous roadside, crop and residential), and the area-to-perimeter index did not differ significantly between landscapes. In the complex landscape, the field perimeters had significantly increased percentage of wide hedges but significantly less percentage of herbaceous roadside vegetation and crops, than in the simple landscape. The other categories of field edges did not differ between landscapes. Further details on the landscape structure characterization of these areas are found in Marino and Landis (1996) and Menalled et al. (1999).

Field border vegetation sampling

In 2000 and 2001, 100 m transects were established at three randomly selected edges in three commercial first year maize fields in each landscape. Field border vegetation height was measured at 20 m intervals along transects using a clinometer. At the same points, width of the field border vegetation was measured and plant species composition was characterized by the point intercept method (Bonham 1989). Plant species intercepted at 5 m intervals by 20 m perpendicular transects originated in each of the five points of the 100 m transect were identified to species. Field border width was measured to a maximum of 20 m.

Landscape structure effect on parasitism

Parasitism was evaluated by releasing sentinel *P. unipuncta* larvae in the same fields where border vegetation was sampled. Second to fourth instars (primarily second to third) were released at three times between mid June and early July (four times in one field at the complex landscape and one field at the simple landscape in 2000, to compensate for previous low recovery) (Table 3). These are the favored larval instars for parasitism by the two dominant species of larval parasitoids in the region (Calkins and Sutter 1976, West and Miller 1989, Oliveira et al. 1999). In addition, use of early *P*. *unipuncta* instars reduces larval movement between plants, increasing the chance of recovery. The *P. unipuncta* larvae used for these studies were obtained from a laboratory colony maintained by the USDA-ARS Corn Insects and Crop Genetics Research Unit (Ames, Iowa).

In each field, releases were performed in the middle of three sides chosen at random (Figure 3). On each side, release sites were 27 m from the border and 25 m apart from previous release sites. The use of these distances allows comparisons with previous studies where the smallest field studied had a 27 m border-center distance (Marino and Landis 1996, Menalled et al. 1999). Each release site consisted of 60 healthy maize plants distributed evenly in three adjacent rows (Figure 4). Three consecutive releases were performed between mid June to early July (four in one field in 2000). Prior to each release, plants in each patch were inspected to assure the absence of naturally occurring P. unipuncta. Individual P. unipuncta larvae were put in the upper part of consecutive maize plants and after 6 d (7 d for the first release of 2000) were recovered by carefully inspecting the plants. The larvae of *P. unipuncta* recovered were reared in plastic 30 ml cups with paper lids (Bio-serv, Frenchtown NJ), containing approximately 10 ml of an pinto bean diet (appendix 2, at 26 °C, 60% relative humidity, and a photoperiod of 16:8 (L: D) h. Diet was replaced as needed. Larval status was periodically checked until a parasitoid or adult moth emerged or the larvae died. The parasitoids obtained were sent to specialists for identification. Voucher specimens are deposited in the collection of the Department of Entomology of Michigan State University.

Host density effect on parasitism

In 2001, two sets of 60 maize plants, 25 m apart, were selected and randomly assigned with low or high P. unipuncta density. The low-density treatment (one larva per plant) corresponds to the level used in previous studies (Marino and Landis 1996, Menalled et al. 1999). In the high density treatment three larvae were used per maize plant. A preliminary experiment conducted under greenhouse conditions showed that 50% of P. unipuncta larvae at low density and more than 80% at high density abandoned the maize plant after 72 h. Therefore, to secure recovery of larvae, plants were searched in a 7.7 \pm 0.4 m x 10.5 \pm 1.4 m area (mean \pm SD), which included four additional maize rows on either side of the release rows, and all plants located within 3 m of the release area (Figure 4). Additional plants outside the recovery area were also inspected in the first release, but contained less than 1% of the P. unipuncta larvae recovered and were excluded from the analysis. High field mortality of P. unipuncta occurred in the third release (only 25 larvae were recovered), probably due to a disease, preventing the use of this data in the analysis. An average of 527 ± 136 maize plants (mean \pm SD) were inspected in each release site, for a total of 37,917 plants in the first two releases. Because during 2001 a natural outbreak of P. unipuncta occurred in Michigan (DiFonzo 2001), the presence of naturally occurring P. unipuncta in the fields used was assessed 2 d prior to each release by inspecting plants for larvae in nine transects of 100 maize plants per field. Therefore, additional 10,800 maize plants were surveyed for the presence of wild *P. unipuncta* larvae in the fields used in this study.

Long-term analysis

The data of the present two year study was statistically compared with the results previously obtained by Marino and Landis (1996), Menalled et al. (1999) and Menalled et. al. (unpublished data). Therefore, the effect of landscape structure on overall percentage parasitism and parasitism by *M. communis* and by *G. militaris* individually were compared within and among years for the sampling seasons 1993, 1996, and 1999 (prior studies), and 2000 and 2001.

Data analysis

Vegetation height, width, proportion of sampling points with trees and tree species richness of the field border, were compared between simple and complex landscapes using ANOVA factorial models (Proc Mixed, SAS Institute Inc. 1996). Data from *P. unipuncta* natural population surveys were modeled as number of larvae recovered / number of maize plants inspected x 100, using the logit link function and a binomial distribution, and compared by date of sampling (SAS Gimmlix macro, Li Hell et al. 1996). Parasitism data was combined across releases for each year and analyzed with factorial ANOVA models (Kuehl 2000). Percent overall parasitism, parasitism by *G. militaris* and parasitism by *Meteorus spp.* were modeled as the number of *P. unipuncta* larvae parasitized / number of *P. unipuncta* larvae recovered, using also the logit link function and a binomial distribution. To evaluate the landscape effect, landscape structure and year were modeled as fixed effects, and field (nested in landscape x year) and side (nested in field) as random effects (Proc Mixed, SAS, Li Hell et al. 1996). A similar model was used to determine the effect of host density and landscape

structure for 2001 data. Number of species and Shannon-Weaver diversity index (Price 1997) were obtained per field and compared using a factorial ANOVA similar to the preceding, but assuming a normal distribution and without field side included in the model. The effect of landscape structure on parasitism over a long-term temporal scale was assessed by comparing 2000 - 2001 results with three previous data sets. The same Factorial ANOVA model as described for the two year experiment was used, but without field side effects, and assuming a normal distribution of the residuals. Therefore, percentage parasitism was arc sine transformed previous to the analysis to meet the normality requirement. Slicing by main effects was performed when significant interactions were present. Means were compared using the method of Least Square Means adjusted by Tukey-Kramer (SAS Institute Inc. 1996).

RESULTS

Field border vegetation sampling

Field borders in the complex landscape had more late succession vegetation than those of the simple landscape (Table 4). Field edges were wider in the complex than in the simple landscape (mean \pm SEM; 14.1 \pm 0.8 m in the complex landscape, 5.9 \pm 0.7 m in the simple landscape, df = 1, 10; F = 9.66; P = 0.01). Significantly taller vegetation was present at the edges of fields located in the complex than in the simple landscape (mean \pm SEM; 9.4 \pm 0.7 m in the complex landscape, 3.3 \pm 0.6 m in the simple landscape, df = 1, 10; F = 11.40; P = 0.01). The complex landscape had more tree species and a higher proportion of trees and shrubs at the edges of fields than the simple landscape (mean \pm SEM; tree species number: 6.2 \pm 1.1 in the complex landscape, 1.7 \pm 0.7 in the simple landscape; df = 1, 16; F = 9.64; P = 0.04; proportion of trees and shrubs in field edge: 0.60 \pm 0.09 in the complex landscape, 0.17 \pm 0.06 in the simple landscape; df = 1, 16; F = 13.44; P = 0.02). The parameters of the field border vegetation measured were consistent between both sampling years, as indicated by non-significant year and year x landscape structure terms in all the analysis performed.

Landscape structure effect on parasitism

Three parasitoid species were collected during 2000 and 2001 in low host density patches (Table 5). *Glyptapanteles militaris* accounted for 89.3% of the parasitism observed in both years while *M. communis*, the second most abundant species, accounted for 9.5% of the parasitism in 2001, being almost absent in 2000 (Table 5). In previous studies only *M. communis* was recovered from *P. unipuncta* (Marino and Landis 1996, Menalled et al. 1999, Menalled et al. unpublished data). However, in 2001 a subsample (n = 95) of *M. communis* sent to a specialist to confirm their identity revealed the presence of *M. dimidiatus* (Cresson) and *M. hyphantriae* Riley, representing 5.3 and 3.2%, respectively. Because individual specimens of all wasps were not saved for identification, hereafter, individuals of these three species will be referred to as *Meteorus* spp. Unidentified individuals were used to start a lab colony, which has proved to be 100% *M. communis*.

During both years in most fields only one additional species was collected besides G. militaris and Meteorus spp. $(2.2 \pm 0.7 \text{ species per field, mean} \pm \text{SD, n} = 12)$. Therefore, the number of parasitoid species collected was not affected by landscape

structure, year of sampling or the interaction of landscape x year (all P > 0.10). Similarly, parasitoid diversity (Shannon-Weaver index) was not affected by landscape structure (df = 1, 6; F = 1.57; P = 0.79). However, diversity was significantly higher in 2001 (Shannon-Weaver index= 0.51 ± 0.07 , mean \pm SEM, n = 6) than in 2000 (Shannon-Weaver index= 0.05 ± 0.02 , mean \pm SEM, n = 6) (df = 1, 6; F = 41.85; P < 0.01), probably due to the increase in the presence of *Meteorus spp*. in both landscapes during 2001. No significant interaction between landscape structure and year of sampling was detected (df = 1, 6; F = 0.44; P = 0.53). These results corroborate the findings of previous studies in these study areas, in which a total of 10 parasitoid species was collected in 3 sample years, with most individuals representing *M. communis* (Marino and Landis 1996, Menalled et al. 1999) or *G. militaris* (Menalled et al. unpublished data).

Parasitism was not significantly affected by landscape structure in 2000 and 2001 (Table 6, Figure 5). Higher levels of parasitism were obtained in 2001 (combined across landscapes: $81.4\% \pm 2.9$, mean \pm SEM) than in 2000 (combined across landscapes: $66.0\% \pm 4.1$, mean \pm SEM). A significant landscape structure by year of sampling interaction was found (Table 6, Figure 5). The comparison of simple effects showed significant differences between parasitism only in the complex landscape in 2000 and 2001 (least squares means adjusted by Tukey-Kramer, P < 0.01) (Figure 5).

The analysis of the parasitism by parasitoid species did not show a significant landscape structure effect on either G. militaris or Meteorus spp. in 2000 and 2001 (Table 6, Figure 6). However, a significant interaction of landscape structure x year of sampling was found for G. militaris, with higher parasitism obtained in the complex landscape in 2001 than in 2000 (least squares means adjusted by Tukey-Kramer, P < 0.01) (Table 6,

Figure 6a). Only three individuals were parasitized by *Meteorus spp.* in 2000, contrasting significantly with the 14.8% parasitism (averaged across landscapes) obtained in 2001 (Table 6, Figure 6b).

Host density effect on parasitism

Two times more *P. unipuncta* larvae were recovered in high host density patches than in low host density patches, and the recovery in the artificial host patches were in turn between 17 and 61 times greater than the natural occurrence of larvae in the field (df = 2, 32; F = 26.93, P < 0.01, and F = 94.64, P < 0.01; for first and second release, respectively) (Figure 7). As expected, landscape structure did not affect this relation among density treatments and the natural population of *P. unipuncta* present in the field (landscape structure effect: df = 1, 4; F = 0.45; P = 0.53, and F = 0.07, P = 0.81; *P. unipuncta* density and landscape structure interaction: df = 2, 32; F = 1.01; P = 0.37, and F = 0.72; P = 0.49; for first and second release, respectively) (Figure 7). Therefore, treatments were successful in generating two different host densities and a significant increase of *P. unipuncta* density over natural occurrence field populations was achieved.

Seven species of parasitoids were collected in high host density patches and only three in low host density patches (Table 5). Despite this, the average number of parasitoid species per field did not differ significantly between host density treatments $(2.8 \pm 0.7 \text{ species per field, mean } \pm \text{SD}, n = 12; \text{ df} = 1, 8; F = 4.00; P = 0.08)$. Landscape structure also did not significantly affect parasitoid species richness (df = 1, 8; F = 1.00; P = 0.35). In addition, the Shannon-Weaver index did not differ significantly between host density treatments, but showed a tendency to be higher at high host density patches

(df = 1, 4.11, F = 5.77; P = 0.07), and in the simple landscape, probably due to a more equitable distribution of the two main parasitoid species in that landscape (df = 1, 5.57; F = 5.24; P = 0.07). In both analyses interaction terms were not significant.

Neither host density nor landscape structure significantly affected the overall parasitism rate, and there was no significant host density x landscape structure interaction (Table 7, Figure 8). However, both parasitism by *G. militaris* and *Meteorus spp.* showed significant responses to host density (Table 7, Figure 9). *Glyptapanteles militaris* parasitized more hosts at low host density (Table 7, Figure 9a), while *Meteorus spp.* parasitized significantly more hosts at high host density (Table 7, Figure 9b). These opposite responses to host density compensated each other, resulting in no host density effect on overall parasitism.

Long-term analysis

The overall percentage parasitism and parasitism by *M. communis* was significantly affected by landscape structure, year of sampling and the interaction between these two factors (Table 8a, Figures 10 and 11). Parasitism by *G. militaris* showed the same trend although the effect of landscape structure was not significant (p = 0.09) (Table 8a, Figure 11). The comparison of effects of year of sampling showed significant effects for overall parasitism and parasitism by both parasitoid species (Table 8b, Figures 10 and 11). Overall parasitism was significantly higher in the complex landscape in 1993 and 1996, but was significantly higher in the simple landscape in 2000 (Table 8c, Figure 10). This analysis differs from the previous one (considering only 2000 and 2001) in the significant difference between landscapes found for 2000. Two reasons account for that difference. First it is performed under normality assumptions with transformed data, and second and more important, has more degrees of freedom due to the inclusion of more years of sampling, and therefore has more power.

Parasitism by G. militaris was significantly higher in the simple landscape in 1998, but showed the opposite trend in 2001, although it was not significant (p = 0.06) (Table 8c, Figure 11a). Parasitism by M. communis was significantly higher in the complex landscape during the first three sampling seasons, but did not differ significantly in 2000 and 2001 (Table 8c, Figure 11b). This long-term analysis showed that overall parasitism and parasitism by species varied from year-to-year, and interacted with landscape structure.

DISCUSSION

The capture of *M. dimidiatus* and *M. hyphantriae* specimens in the last year of the study did not affect the general pattern obtained for *M. communis* due to their low proportion in the sample. This is the first record of *M. dimidiatus* and *M. hyphantriae* parasitizing *P. unipuncta*. Moreover, *P. unipuncta* would be the only host in common for these three species. The other 6 and 17 hosts reported for *M. dimidiatus* and *M. hyphantriae*, respectively, are not reported to be attacked by *M. communis* (Krombein et al. 1979). One possible reason of their occurrence on *P. unipuncta* in 2001 may be a result of *M. dimidiatus* and *M. hyphantriae* host switching to this abundant host during the outbreak year.

It might be argued that complete independence between high and low host density treatments cannot be guaranteed by the methodology used, as parasitoids may move

between treatments, as there was no barrier between them. In addition, no attempt was made to control the potential interference between patches by the attraction of volatiles released by damaged maize plants of neighbor patches. Thus, our comparison of treatment effects may involve some degree of choice since parasitoids could theoretically move between plots in response to host density. However, 2000 data showed no consistent pattern of increase or decrease in parasitism in low host density patches when consecutive releases were separated by 25 m, providing evidence that this distance is sufficient to reduce the interference between host patches (relation between percentage parasitism and consecutive release sites in 2000: $R^2 = 0.03$; df = 1, 52; F = 2.54; P = 0.12).

The overall parasitism obtained in 2000 and 2001 was dramatically higher (73% in average, range: 58 - 87%) than in the previous studies in the same area (20% on average, range: 5 - 31%; Marino and Landis 1996, Menalled et al. 1999, Menalled et al. unpublished data) (Figure 10). These results also differ from previous studies in the occurrence of significantly higher parasitism in the simple landscape (2000). In previous studies parasitism was significantly higher in the complex than in the simple landscape during 1993 and 1996 seasons (Marino and Landis 1996, Menalled et al. 1999), with no significant effect of landscape structure in 1998 (Menalled et al. unpublished data). This long-term analysis shows that parasitism was significantly affected by landscape structure in three out of five years studied, but with trends in opposite directions depending on which parasitoid species predominated in a particular year. In 2000 and 2001, the overall pattern of parasitism was driven by *G. militaris* and not by *M. communis* as occurred in previous sampling years (Figure 11a). *Glyptapanteles militaris* was collected in much

higher numbers than in previous sampling seasons and, more important, without being affected by landscape structure. Moreover, this species showed opposite trends between years, parasitizing more hosts in the simple landscape in 2000 and more in the complex landscape in 2001 (Figure 11a). *Meteorus communis* was almost absent in 2000, but in 2001 the parasitism produced by this species was as high as in previous years (Figure 11b). However, unlike previous sampling seasons, it was not significantly affected by landscape structure, and showed a trend of slightly higher parasitism in the simple landscape. This data shows for the first time that both parasitoid species are not strictly limited by landscape structure and can parasitize significant numbers of hosts in either landscape.

These results demonstrated that the simple interactions proposed between the parasitism by *G. militaris* and *M. communis* and landscape structure were not constant in the system. Hence the patterns of parasitism by these species were influenced by factors that operate either independently of landscape structure or at a different scale than the one used in these studies. However, despite the different outcomes obtained in different sampling years, the consideration of all sampling seasons together provides some new insights about possible interactions between these parasitoid species and landscape structure. Parasitism differed significantly between landscapes in years of low overall parasitism that were dominated by *M. communis* (1993 and 1996, Figure 10). In contrast, the increase of parasitism by *G. militaris* since 1998 (Figure 11a), initially in the simple landscape, but in years of high overall parasitism in both landscapes, resulted in landscapes with the same level of parasitism (Figure 10).

Several factors could be proposed as possible causes of this pattern of parasitism. Among them, the fluctuation of alternative host populations remained as the most important one. *Pseudaletia unipuncta* does not overwinter in Michigan in great numbers, and damaging populations are derived from adults that migrate northward in spring (Thompson 1966, Untung 1978). A similar situation occurs with *P. unipuncta* in Québec (Canada), where migration from Southern United States, such as Tennessee, was proposed as the main source of spring adults (Fields and McNeil 1984). In addition, studies on the flight capacity of this species showed that is capable to flight up to 1,600 km in its migration from the South to Iowa and Missouri (Hendrix and Showers 1992). *Glyptapanteles militaris* and *M. communis* are generalist parasitoids and, although there are no conclusive studies, evidence suggests both species overwinter as larvae inside their hosts (Tower 1915; Breeland 1958, West, 1988). Therefore, alternative host species could be important as winter hosts and their abundance and distribution may limit these parasitoid populations.

Marino and Landis (1996) and Menalled et al. (1999) proposed a link between the relative abundance of the vegetation that supports alternative hosts for these parasitoid species and the level of parasitism observed in each landscape. While alternative hosts for *M. communis* feed mostly on trees and shrubs that are proportionally more abundant in the complex landscape, alternative hosts of *G. militaris* feed primary on grasses and forbs that are abundant in both landscapes (Krombein et al. 1979, Covell 1984, Stehr 1987, West and Miller 1989, Metcalf and Metcalf 1993). According to this hypothesis, parasitism by *M. communis* should be significantly higher in the complex landscape. The absence of differences in parasitism by *M. communis* between landscapes obtained in

2001 does not support this hypothesis. However, at least two different factors might account for this disagreement. First, during 2001 an outbreak of *P. unipuncta* occurred early in the season, and therefore it is possible that unusually high numbers of this host allowed *M. communis* to extend its distribution to areas in the simple landscape with fewer alternative hosts. However, there is no direct data available to support this hypothesis. A second possibility involves the temporal fluctuation of alternative host populations. Generalist parasitoids are likely to respond to the overall host density rather than to the abundance of a single host species (Teder et al. 2000). There is no information available regarding the population of any of the alternative hosts of *M. communis* feed on trees and shrubs, few are grass feeders (Marino and Landis 1996). Therefore, it is possible that a year of higher abundance of a grass-feeding host of *M. communis* allowed this parasitoid to move to the simple area.

Another possible factor that could mediate the effect of landscape structure on parasitism is a differential distribution of food resources between landscape types (Marino and Landis 1996). Non-crop areas are proposed to harbor more sources of food (particularly carbohydrates and proteins) than crop fields, especially early in the season when this study was conducted (Landis et al. 2000, Tscharntke 2000). Laboratory studies (Chapter 3) demonstrated that provision of carbohydrate resources significantly increased the longevity of both *G. militaris* and *M. communis* and the fecundity of *M. communis*. No attempt was made to quantify the distribution of these resources between landscapes. However, since these resources are used by both parasitoid species, and similar parasitism rates were obtained in both landscapes in 1998 and 2000, and opposite trends

were found between the first two sampling seasons and 2001, carbohydrate resource distribution is not likely to be the limiting factor in these landscapes.

Habitat connectivity and fragmentation (measured as distance from the source areas of parasitoids) have been proposed as landscape structure features that affected parasitism by several studies (Kruess and Tscharntke 1994, Roland and Taylor 1997, Thies and Tscharntke 1997, Doak 2000, Kruess and Tscharntke 2000b). The mechanism by which this operates remains unclear. The scale at which landscape structure is perceived by each parasitoid species could be different, even for parasitoids that attack the same host in the same area. Roland and Taylor (1997) showed that different parasitoid species respond to forest structure at different scales and this was related to the body sizes of the parasitoids studied. The movement of tenebrionid beetles has also been shown as significantly affected by habitat structure at a microlandscape level (Wiens and Milne 1989). Therefore, it is clear that parasitoid flight capacity and movement through different environments could play a key role at the landscape scale. Marino and Landis (1996) showed that the parasitism of P. unipuncta obtained near field borders (at 10 m from the field edge) did not differ from the parasitism obtained near the center of the field (at 90 m from the field edge). Therefore, parasitism was not affected at this small scale. However, P. unipuncta parasitism might be influenced by landscape structure at a much bigger scale than the one used in these studies (3.5 km^2) . Unfortunately, there is no published information on either the dispersal capacities of G. militaris and M. communis, nor the possible barriers to such movement.

Weather has also been found correlated with the abundance of insects and interacting with landscape structure (Powers et al. 1999). However, there are no studies

that specifically relate weather effects with parasitism at a landscape scale. Another possible factor that may affect the outcome of my study are competitive interactions between *G. militaris* and *M. communis*. The first larval instar of *M. communis* is mandibulate and can actively eliminate other parasitoid larvae present in the host, including conspecifics (West 1988). However, I did not dissect *P. unipuncta* larvae to look for evidence of competition. Finally, the results of my study, such as some other authors explicitly state for theirs (Doak 2000), are not independent of factors that may be strongly correlated with landscape structure.

Teder et al. (2000) reported a similar response of parasitism to host density as the one obtained in this study. These authors reported an absence of density dependence at a big scale (*Betula pubescens* and *B. pendula* trees separated by distances of 2 - 10 km) of the parasitism of natural populations of *Epirrita autumnata* (Bkh.) (Lepidoptera: Geometridae). However, at a small scale (individual trees), the two main parasitoid species responded significantly to artificially increased host densities of *E. automnata*. The braconid *Protapanteles immunis* (Haliday) produced higher rates of parasitism at low host densities, while the ichneumonid Phobocampe bicingulata (Grav.) parasitized more hosts at higher host densities. As in my study, these differences compensate each other and no response of overall parasitism to host density was obtained. They postulated that differences in parasitoids size, among other biological features, might account for the different response obtained. Similarly, the different responses of the G. militaris and M. communis to host density may be related to their different life history characteristics. Glyptapanteles militaris is a gregarious parasitoid that deposits an average of 49 eggs per host larvae (range 3 to 226) and has been observed to parasitize up to nine hosts in 4 h in

laboratory conditions (Tower 1915, Breeland 1958, Calkins and Sutter 1976, Rolim 1983, Oliveira et al. 1999). It completes its life cycle (from egg to 50% adult death) between 19.0 d (at 27 °C) and 22.1 d (at 23 °C) (Calkins and Sutter 1976, Rolim 1983). Meteorus. communis, on the contrary, is a solitary species and very little is known about its biology (West 1988, Miller 1996). The development from egg to adult takes 16.7 ± 0.6 d (at 31 $^{\circ}$ C) to 61.8 ± 3.4 d (at 14 $^{\circ}$ C) (mean ± SD) (Miller 1996). Preliminary studies that I conducted in the laboratory showed that the adult longevity at 24 $^{\circ}$ C could be 54.8 ± 6.6 d (mean \pm SD) if sugar resources are made available, and that such females parasitize a mean of 114.8 \pm 14.7 hosts (\pm SD) (Chapter 3). Due to its gregarious oviposition pattern, G. militaris is likely to be temporally egg-limited, and may leave a host patch after parasitizing relatively few hosts. *Meteorus communis*, in contrast, should be able to parasitize more hosts per patch because it lays only one egg per host, thus being able to expend more time per host patch. Stiling (1987) proposed that solitary parasitoids should be more likely to show host density dependence than gregarious parasitoids, although he did not find evidence to support that hypothesis in the studies he reviewed. These possible behavioral patterns need further testing for G. militaris and M. communis.

In summary, the present study demonstrates that the initial report of a positive effect of landscape complexity on parasitism based on 1993 and 1996 results (Marino and Landis 1996, Menalled et al. 1999) is not always true. Multiple variables could operate at landscape level studies and are difficult to control and replicate (Menalled et al. 1999). Among them, a change in the distribution of alternative hosts and the outbreak of *P*. *unipuncta* that occurred during the last year of study, seem to be the most likely causes of the different pattern of parasitism obtained in 2000 and 2001 with respect to previous

seasons. In addition, *G. militaris* and *M. communis* have the potential to respond differently to host density, independent of the landscape structure. In the system under study, further years of sampling could demonstrate if the parasitism obtained by *M. communis* in the simple landscape during 2001 was an isolated and unlikely event, or is part of a change in the distribution of this parasitoid mediated by other factors. In addition, more years of low parasitism with *G. militaris* restricted to the simple landscape are also necessary to confirm the initial pattern obtained. Alternatively, additional years of sampling may show that parasitism by this species is affected by phenomena operating at a different scale. This work suggests that studies directed to address landscape scale effects on insect communities need also to incorporate a sufficiently wide temporal scale to obtain accurate descriptions of the system and capture the variability of the factors that affects their dynamics.

	ds reared		High Host	Density		ł	393	527
	Parasitoi		Low Host	Density	200	248	186	265
		ered	High Host	Density		1	510	652
JSA.	<i>uncta</i> larvae	recov	Low Host	Density	265	426	245	303
unty, Michigan, I	Pseudaletia unip	ed	High Host	Density		1	3240	3240
ape in Ingham Co		releas	Low Host	Density	1682	1706	1080	1080
gricultural landsc:		Landscape	I		Simple	Complex	Simple	Complex
complex a{		Year			2000		2001	

Table 3. Numbers of *Pseudaletia unipuncta* larvae released and recovered and numbers of parasitoids reared in a simple and a



Figure 3. Example of treatments and release site orientation in the field. Relative position of high and low density treatments during each release was random, varying from field to field. Roman numbers indicate release date, rectangles group simultaneous releases, small dots represent low density treatment release sites and larger dots represent high density treatments release sites.



Figure 4. The release area (inner dotted rectangle) included three rows of 20 maize plants. Each release site received at random a high or low host density treatment in 2001. Recovery was performed in the release area plus all the plants located within four rows of the release area sides and 3 m of the release area row ends (external dotted rectangle).

Table 4. Frequency of tree species in hedgerows bordering maize fields in a complex and a simple agricultural landscape in Ingham County, Michigan. Frequency is the proportion of the sample (n= 360 sample points per landscape) in which a species was present in the hedgerows within each landscape type.

	Landsca	pe type
Tree species ¹	Complex	Simple
	Frequ	ency
Prunus serotina Ehrh.	0.192	0.031
Quercus rubra L.	0.103	0.017
Fraxinus americana L.	0.078	0.014
Tilia americana L.	0.072	•••
Quercus alba L.	0.050	
Rhus typhina Torn.	0.019	0.033
Ulmus americana L.	0.031	0.019
Catalpa spp.	0.028	
Ulmus spp.	0.011	0.028
Acer rubrum L.	0.025	0.022
Ulmus rubra Muhl.	0.003	0.022
Juglans nigra L.	0.022	
Acer saccharum Marsh.	0.019	0.014
Populus deltoides Bartr.	0.017	0.006
Rhus copallina L.	0.017	0.006

Table 4 (cont'd)

0.017	
0.017	
0.014	
0.014	
0.011	
0.011	
0.008	
0.006	0.006
0.006	
0.006	
0.006	
0.006	
0.006	
	0.017 0.017 0.014 0.014 0.011 0.011 0.008 0.006 0.006 0.006 0.006 0.006 0.006 0.006

¹ Species represented in at least two samples.

Table 5. Parasitoids emerging from Pseudaletia unipuncta sentinel larvae released in a complex and a simple agricultural

landscape located in Ingham County, Michigan, USA.

	Year 2	0000		Year	2001	
	Low Host	Density	Low Host	Density	High Host	Density
	Complex	Simple	Complex	Simple	Complex	Simple
Glyptapanteles militaris (Walsh)	244	196	224	146	369	241
Meteorus spp. ¹	1	7	38	45	149	156
Enicospilus purgatus (Say)	÷	÷	÷	÷	7	1
Campoletis oxylus (Cresson)	÷	÷	7	7	÷	5
<i>Campoletis</i> sp.	1	1	÷	÷	÷	÷
Therion circumflexum (L.)	÷	÷	÷	÷	÷	ю
Sinophorus sp.	:	÷	÷	:	1	:

Table 5 (cont'd)		:	:	:	0	1
Tachinidae						
Unidentified ²	7	1	7	1	Ś	4

¹ In 2001, a subsample identified by a specialist (n = 95) revealed that M. dimidiatus (Cresson) represented 5.3%, and M. hyphantriae Riley represented 3.2% of these specimens recorded as M. communis (Cresson). In previous studies only M. communis has been identified from P. unipuncta in Michigan.

² Unidentified parasitoid larvae that failed to spin the cocoon.

Table 6.	6. ANOVA of influence of agricultural landscape structure on	le percentage parasitism of Pseudaletia unipuncte
sentinel la	el larvae by all species (overall parasitism), and by the two main p	asitoid species separately, at low host densities, in
Ingham C	m County, Michigan, in 2000 and 2001.	

		Overall P	arasitism	G. mil	litaris	Meteor	.dds sn.
Source of variation	df	F	Р	F	Р	F	Ρ
Landscape Structure	1, 4	0.00	0.96	0.00	0.98	0.30	0.61
Year of sampling	1, 16	15.45	< 0.01	0.00	0.99	46.27	< 0.01
Landscape Structure × Year of sampling	1, 16	11.91	< 0.01	18.18	< 0.01	0.03	0.87


Figure 5. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* sentinel larvae in a complex and a simple agricultural landscape in Ingham County, Michigan, USA. Means with different letters are significantly different (P < 0.05, Least Square Means difference, adjusted by Tukey-Kramer method).



Figure 6. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* sentinel larvae by (a) *Glyptapanteles militaris* and (b) *Meteorus* spp., in a complex and a simple agricultural landscape in Ingham County, Michigan, USA. Different letters indicate significant differences at P < 0.05 (LSM difference, adjusted by Tukey-Kramer method).



Figure 7. Number of *Pseudaletia unipuncta* sentinel larvae per 100 maize plants (mean + SEM) recovered in high host density patches and low host density patches and natural occurrence in the field in 2001, in Ingham County, Michigan, USA. Different letters are significantly different (P < 0.05, Least Square Means difference, adjusted by Tukey-Kramer method).

Pseudaletia unipuncta sentinel larvae t	y all specie	s (overall pa	rasitism), and	by the two n	ain parasitoid	l species sep	arately, in
Ingham County, Michigan, USA, in 20	01.	I					
		Overall P	arasitism	G. mi	litaris	Meteori	es spp.
Source of variation	df	F	Р	F	b	F	Р
Landscape Structure	1,4	4.12	0.11	4.12	0.11	1.01	0.37
Host Density	1, 16	1.64	0.22	27.86	< 0.01	15.30	< 0.01
Landscape Structure \times Host Density	1, 16	2.21	0.16	0.98	0.34	0.00	0.95

Table 7. ANOVA of the influence of agricultural landscape structure and host density on the percentage parasitism of



Figure 8. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* sentinel larvae at high and low density patches in a complex and a simple agricultural landscape in Ingham County, Michigan, USA, in 2001 season.



Figure 9. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* sentinel larvae by (a) *Glyptapanteles militaris* and (b) *Meteorus* spp., at high and low host density patches, in a simple and a complex agricultural landscape in Ingham County, Michigan, USA, in 2001 season.

Table 8. ANOVA of influence of agricultural landscape structure on the percentage parasitism of <i>Pseudaletia unipunc</i> sentinel larvae by all species (overall parasitism), and by the two main parasitoid species separately, at low host densities,
Ingham County, Michigan, over five sampling seasons (1993 – 2001).

a) Main effects model

		Overall P	arasitism	G. mil	litaris	Meteo	rus spp.
Source of variation	df	F	Р	F	Р	F	Р
Landscape Structure	1	13.87	< 0.01	3.04	0.09	30.13	< 0.01
Year of sampling	4	117.19	< 0.01	140.21	< 0.01	56.83	< 0.01
Landscape Structure \times Year of sampling	4	11.91	< 0.01	6.03	< 0.01	14.14	< 0.01

b) Effect of year of sampling: model sliced by landscape.

		Overall P	arasitism	G. mil	itaris	Meteor	.dds sn.
Effect of Year	df	F	Р	F	Р	F	Р
Complex	4	46.64	< 0.01	79.62	< 0.01	58.84	< 0.01
Simple	4	82.46	< 0.01	66.61	< 0.01	12.13	< 0.01

Table 8 (cont'd)

sampling.
5
year
N
<u> </u>
sliced
e]
Ð,
ğ
-
structure:
ð
al
SC
g
aı
fl
0
) Effect
်ပ်)

		Overall Pa	arasitism	G. mil	itaris	Meteoi	rus spp.
Landscape structure effect	df _	F	Р	F	Р	F	Р
1993	-	12.42	< 0.01	0.38	0.57	17.59	0.01
1996	1	30.04	< 0.01	0.59	0.45	32.57	< 0.01
1998	1	2.21	0.16	17.03	< 0.01	5.84	0.03
2000	1	5.55	0.04	3.08	0.10	0.41	0.53
2001	1	5.14	0.08	4.63	0.06	0.14	0.72



Figure 10. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* sentinel larvae in a complex and a simple agricultural landscape in Ingham County, Michigan, USA, including previous studies data (Marino and Landis 1996, Menalled et al. 1999, and Menalled et al. unpublished data). Means with asterisks are significantly different (P< 0.05, F test from slicing by year of sampling), ns = not significant.



Figure 11. Percentage parasitism (mean + SEM) of *Pseudaletia unipuncta* sentinel larvae by (a) *Glyptapanteles militaris* and (b) *Meteorus* spp., in a complex and a simple agricultural landscape in Ingham County, Michigan, USA, including previous studies data (Marino and Landis 1996, Menalled et al. 1999, and Menalled et al. unpublished data). Means with asterisks are significantly different (P < 0.05, F test from slicing by year of sampling), ns = not significant.

CHAPTER 3:

EFFECT OF FOOD RESOURCES, HOST ACCESS, MATING STATUS, AND TEMPERATURE ON THE LONGEVITY AND FECUNDITY OF ADULT *GLYPTAPANTELES MILITARIS* (WALSH) AND *METEORUS COMMUNIS* (CRESSON) (HYMENOPTERA: BRACONIDAE), PARASITOIDS OF THE ARMYWORM, *PSEUDALETIA UNIPUNCTA* (HAWORTH) (LEPIDOPTERA: NOCTUIDAE).

ABSTRACT

The presence and abundance of extra-host resources for adult parasitoids, such as food sources and refuges, can limit the level of parasitism they inflict in their hosts. Structurally complex landscapes are postulated to contain more suitable habitats for adult parasitoid needs, than more highly simplified landscapes. Two braconid wasps are the most important parasitoid species that attacks the armyworm *Pseudaletia unipuncta* (Haworth) (Lepidoptera: Noctuidae) in Michigan. In laboratory studies I investigated the effect of carbohydrate food sources and temperature on the longevity and fecundity of these two parasitoids species. *Glyptapanteles militaris* (Walsh) adults had increased longevity when provided with honey diluted in water and when reared at 20 °C than at 25 °C. *Meteorus communis* (Cresson) adults also lived significantly longer when provided with honey diluted in waters. Host access but not mating status significantly reduced honey-fed female longevity. Honey-fed females parasitized significantly more hosts, as a

consequence of their increased longevity, but did not differ from control females fed with water in the daily rate of oviposition. Mated females provided with honey had a significantly higher rate of parasitization than unmated females. These results indicate that in the field the presence of carbohydrates resources, such as floral and extrafloral nectar and aphid honeydew, may significantly increase the life span and parasitism produced by these parasitoid species. Structurally complex landscapes that provide more heterogeneity of habitats and potentially more food sources and refuges are predicted to support more effective populations of these parasitoids.

KEY WORDS: *Meteorus communis, Glyptapanteles militaris, Pseudaletia unipuncta* parasitoids, longevity, fecundity, carbohydrate resources, mating status, host access

INTRODUCTION

Most parasitoids rely mainly on the adult stage to find and select adequate resources to complete their life cycle. These resources include food, refuges from abiotic and biotic mortality factors for adults and host for their immature instars. However, the spatial distribution of the resources that adults need to survive could limit the scope of host search. Although some adult parasitoids obtain some needed resources by host feeding, many hymenopteran parasitoids use other food sources for maintenance and to support egg production (Foster and Ruesnik 1984, Vinson and Barbosa 1989, Quicke 1997, Jacob and Evans 1998, Gordh et al. 1999, Landis et al. 2000, Tscharntke 2000).

Potential non-host food sources include floral and extrafloral nectar, pollen, and aphid honeydew (Idoine and Ferro 1990, England and Evans 1997, Jacob and Evans 1998). Flowering plants are frequently visited by parasitoids (Bugg et al. 1989, Jervis et al. 1993), and consumption of floral nectar can enhance adult longevity (Leius 1963, Foster and Ruesink 1984, Idris and Grafius 1995, van Driesche and Bellows 1996). Parasitoids fed with other carbohydrates resources, such as honey, sucrose, glucose, fructose and other sugars in solution, showed increased longevity in laboratory conditions (Dyer and Landis 1996, McDougall and Mills 1997, Jacob and Evans 2000, Bautista et al. 2001, Fadamiro and Heimpel 2001, Wäckers 2001, Rivero and West 2002). Accessibility of non-host food is not only related to an increased life span, but also with increased fecundity, number of hosts parasitized, and proportion of time searching for hosts instead of food (Foster and Ruesnik 1984, Tscharntke 2000). Adult parasitoid longevity is also affected by two additional factors; mating status and number of hosts available (Godfray 1994, Lei et al. 1997, Jacob and Evans 2000). Finally, microclimatic conditions, in particular temperature and relative humidity, can have a strong influence on insect longevity. Increased temperatures tend to reduce insect longevity, particularly under high relative humidity conditions where evaporative cooling potential is reduced (Chapman 1998). Similarly, parasitoid longevity is often reduced with increased temperature within the suitable range (Dyer and Landis 1996, Jervis and Copland 1996, McDougall and Mills 1997).

In crop monocultures, the presence of non-host adult food resources is limited in time and space, and may not be in synchrony with the presence of the appropriate stage of the host (Jacob and Evans 1998). Therefore, many parasitoid species that attack crop

pest are benefited by the presence of undisturbed extra-field habitats that contain these resources and could serve as refuges and contribute to the establishment and retention of parasitoid populations near crop fields (Landis et al. 2000, Tscharntke 2000).

The armyworm, *Pseudaletia unipuncta* (Haworth) (Lepidoptera: Noctuidae) is cosmopolitan generalist feeder that is inflicting of causing important economic damage to grass crops in North America (Breeland 1958, Guppy 1961, Metcalf and Metcalf 1993). Economically important injury is typically confined to outbreak years, which are generally unpredictable in time and space (Guppy 1961, Untung 1978). *Pseudaletia unipuncta* cause damage by larval feeding, with latter instars responsible for most damage (Guppy 1961, Breeland 1958, Metcalf and Metcalf 1993). In Michigan, recent *P. unipuncta* outbreaks involving several counties have occurred in 1964, 1976, 1978, and recently in 2001, causing losses in oat, wheat and maize (Thompson 1966, Untung 1978, DiFonzo 2001). A complex of 69 parasitoids is known to attack *P. unipuncta* in North America, with 35 of these species recorded from the Great Lakes region (Breeland 1958, Guppy 1967, Untung 1978, McNeil and Turgeon 1988, Marino and Landis 1996, Menalled et al. 1999).

Two braconid parasitoids dominate the parasitoid community of *P. unipuncta* in southern Michigan: *Meteorus communis* (Cresson), and *Glyptapanteles militaris* (Walsh) (Marino and Landis 1996, Menalled et al. 1999, Menalled et al. unpublished data, Chapter 2). *Meteorus communis* is a native generalist, koinobiont parasitoid that primarily attacks larvae of various Noctuidae (Krombein et al. 1979). In addition to maize and small grains, it was also recorded from peppermint and alfalfa fields in Oregon (West 1988). Using light traps, Burbutis and Stewart (1979) found that *M. communis*

follows the first and third of the three flight peaks of *P. unipuncta* in New Jersey. It is a solitary, arrhenotokous endoparasitoid and the first instar is mandibulate and actively eliminates supernumerary conspecifics and perhaps, larvae of other parasitoid species as well (West 1988). It oviposits into second to fourth instar hosts in the field, and typically emerges and kills the larvae in the fifth instar (McNeil and Turgeon 1988, West 1988, West and Miller 1989). Immature developmental time depends on both temperature and the stage of host attacked, with shorter development in more advanced instars at up to 28 °C (West 1988, Miller 1996). At the present, there are no studies reporting longevity or the food resources used by adult *M. communis*.

Glyptapanteles militaris has been reported as the most important parasitoid of *P*. unipuncta in several studies in North America (Breeland 1958, Calkins and Sutter 1976, Steinkraus et al. 1993), and in previous studies in Michigan (Untung 1978, Rolim 1983). It is a native, koinobiont, generalist endoparasitoid that attacks several species of noctuids. Most of the studies performed on this species are in reference to its potential as a biological control agent (Calkins and Sutter 1976, Oliveira et al. 1998, 1999), and relatively little is known regarding its basic biology. *Glyptapanteles militaris* is a gregarious arrhenotokous parasitoid (Tower 1915, Rolim, 1983). Previous studies showed that provision of carbohydrates contributes to prolonged adult longevity in *G*. *militaris* (Calvin and Sutter 1976, Oliveira et al. 1998). High temperature (35 °C) has been reported to kill *G. militaris* larvae inside its host and to reduce female fecundity (Kaya and Tanada 1969).

The objective of this study was to establish biological characteristics of adult *M*. communis and *G. militaris* that could be related to the pattern of habitat use by these

parasitoid species. The effects of temperature and access to food resources on adult parasitoid longevity were evaluated for *G. militaris*. The effects of food resources, hosts and mating status, on the longevity and fecundity were investigated for *M. communis*.

MATERIAL AND METHODS

Influence of temperature, water and carbohydrate availability on adult longevity of *Glyptapanteles militaris*

A colony of G. militaris was initiated from P. unipuncta larvae released and recaptured in maize fields in Ingham County, Michigan during the 2000 growing season. The colony was maintained in a rearing room at 24 °C and 60% RH, and a photoperiod of 16:8 (L: D) h. To supply G. militaris with hosts, a colony of P. unipuncta initiated from larvae provided by the USDA-ARS Corn Insects and Crop Genetics Research Unit (Iowa State University Ames, IA) was also maintained. Newly emerged adult G. militaris were placed in 500 ml plastic cups provided with a hole covered with a fine mesh in their tops. Adults were arranged in pairs of one female and one male. Containers with solitary females were included as a control for negative effects of males on female longevity (Jervis and Copland 1996). Three food regimes were tested: honey-water (50% honey in distilled water), distilled water exclusively, and control without food or water. Two 30 ml cups with a cotton wick through their lids were placed inside all the 500 ml containers. Honey-water treatment consisted in one cup filled with 50% honey-water and one with distilled water, both cups were filled with distilled water for the treatment with water, and finally two empty cups served as no-food controls. Honey-water and distilled

water cups were replaced as needed, typically every 7 days, two avoid the development of fungi. Early season temperatures in southern Michigan maize fields typically range between 20 to 30 °C and seldom reach 35 °C (Dyer and Landis 1996). We selected two temperature treatments within that range that were compared in different growth chambers: 20.1 ± 0.5 °C and, and 25.0 ± 0.2 °C (mean \pm SD). The relative humidity differed slightly between temperature regimes at: 62.8 ± 4.0 % and 77.8 ± 4.8 % (mean \pm SD), respectively. The same photoperiod of 16:8 (L: D) h was used in both growth chambers. Temperature and humidity inside containers with different treatments were measured and compared with general growth chamber conditions to control for any micro-environmental difference between treatments. Ten containers with parasitoid pairs and 15 containers with females alone were tested in each growth chamber for each treatment. Wasp survivorship was assessed twice a day, in the morning and evening. Insects that showed no movement in reaction to touch were considered dead. A priori predictions were that adults would live for fewer days at 25 °C than at 20 °C, that honey provision would increase the longevity of adults under both temperature conditions, and that water would enhance adult survivorship with respect to the control treatment, especially at the highest temperature. In addition, it was also predicted that females would live longer than males, and that the presence of males would reduce female longevity in comparison with those maintained individually in treatments provided with honey, and that these effects would be less important in no-honey treatments.

Micro-environmental conditions among treatments were compare using a oneway ANOVA (SAS Institute Inc. 1996). Adult longevity was compared between males and females in the same cage, and between females with males and control solitary

females. A three factor (temperature, food and sex or male presence) ANOVA was performed (PROC MIXED, SAS Institute Inc. 1996). To meet the assumption of normality, data were transformed (Ln [x]) prior to analysis (Sokal and Rohlf 1995). Independent variances were estimated for each food treatment to avoid heterogeneity of variances effects, and the suitability of this model was assessed using the Ratio Likelihood Test (P < 0.05, SAS Institute Inc. 1996).

Longevity and fecundity of *Meteorus communis* with and without carbohydrate resources, hosts, and with different mating status

Meteorus communis adults from P. unipuncta larvae placed in maize fields in Ingham County, Michigan during 2001 were used to start a laboratory colony. A colony of P. unipuncta initiated from larvae provided by the USDA-ARS Corn Insects and Crop Genetics Research Unit (Iowa State University Ames, IA) was maintained and used as host for M. communis. Parasitoid cocoons from the laboratory colony were isolated in 30 ml plastic cups and checked daily for adult emergence. All adults were manipulated by carefully allowing them to enter 5 ml plastic tubes. Newly emerged females and males were randomly paired and were placed in 3.8-liter cylindrical plastic cups with three openings closed by a thin mesh to allow air interchange. Two plastic cups with distilled water (the same as described in the G. militaris longevity experiment) were put in each container on open plastic petri dishes (9 cm diameter) to contain potential spills or wicking from the cups. Males typically attempted to mate with females immediately after introduction and copulation was observed on several occasions. Because mating in this species is reported to occur in the first half hour of male contact with virgin females

(West 1988), all females were assumed to be mated after this time. Males were taken out of the container after a 24 h mating period, similar to that used in previous studies with other parasitoid species (Calkins and Sutter 1976, Hu et al. 1986, Oliveira et al. 1999).

After male removal, females were randomly assigned to one of four conditions representing combinations of food and host resources: 1) females with honey-water and hosts, 2) females with honey-water but without hosts, 3) females without honey-water but with hosts, and 4) control females without honey-water and hosts. Honey-water treatment consisted in replacing one of the original distilled water cups with one cup filled with a 50% solution of honey in distilled water. Treatments with no honey access (food controls) retained both cups with distilled water provided. A treatment without water access was not used since in normal conditions parasitoids are not water deprived in the field (Dyer and Landis 1996) and to reduce the number of treatment combinations. To prevent the development of fungi, cups with honey-water and distilled water were replaced as needed. Host treatment consisted of a daily provision of ten P. unipuncta larvae of second to fourth instar, but primarily third, obtained from the laboratory colony. Although there are no studies that explore the instar suitability of *P. unipuncta* for *M.* communis, data from other noctuids indicate higher rates of parasitism from second to fourth instar larvae (West and Miller 1989). In addition, I observed M. communis to readily accept these host stages in preliminary trials. Host larvae were carefully taken from the rearing containers using soft forceps and placed in groups of five in plastic 60 ml containers. These groups were randomly assigned to treatments in each block of replicates. *Pseudaletia unipuncta* larvae were placed in an open plastic petri dish, on a rectangular piece $(7 \times 5 \times 1 \text{ cm})$ of artificial diet based in pinto beans (appendix 1) to

provide food for the larvae. In addition, a piece of maize leaf (9 x 5 cm) was included to ensure the presence of host plant cues for female wasp host location. The diet was replaced as needed, usually once a week and the maize leaf renewed daily. Each 24 h larvae were replaced and those exposed to parasitoids were placed individually in 30 ml plastic cups with paper lids (Bio-serve, Frenchtown, New Jersey), containing approximately 10 ml of artificial diet. Larvae were checked periodically until death, pupation or emergence of the parasitoids. Diet was replaced as needed, typically once a week. No data was available concerning daily fecundity of *M. communis*. Therefore, I attempted to provide a surplus of hosts in order to correctly estimate the effect of food regimes on fertility (Jervis and Copland 1996), based on the experiments of Miller (1996) in which 20 hosts were provided for 2-4 females during 30 h, and obtained ca. 70%parasitism, and West (1988) in which a ratio of 8.5 hosts per female during 24 h was used and a parasitism between 35 to 67 % obtained. The initial four blocks were provided with 20 host larvae per female per day until females were 3 to 7 days of age (varying among blocks), then 15 larvae per day until females were 22 to 27 days of age, and finally 10 larvae per day were provided until female death. Inspection of data revealed that female of *M. communis* seldom attacked more than 10 hosts per day (1.5 % of cases), and therefore in following blocks ten larvae were provided daily to each female through the test. Due to sporadic P. unipuncta rearing problems, on some days less than ten hosts were provided in certain blocks, resulting in an overall 8.8 % reduction of the number of hosts offered with respect to the original experimental design. However, no less than 2.5 larvae per day were made available to each female. To avoid bias, parasitism obtained in days when less than ten host larvae were provided is not included in the daily fecundity

results, however, it is included in the total parasitism obtained. Larvae exposed to females were reared initially (blocks 1-4) in groups of 3 or 2 larvae in 30 ml cup, but due to observation of cannibalism, individual rearing was adopted in subsequent replicates.

In addition to honey and host provision effects on female longevity and fecundity, the effect of mating was assessed in blocks 1-4. Females were randomly assigned to stay their first day with males (as previously described), or were placed individually in the same conditions as mated females, but without a male. Therefore, a counterpart of four treatments with virgin females was included in the first four blocks. Mating effect was excluded in further blocks. A total of 5,948 *P. unipuncta* larvae were exposed to 48 *M. communis* females and reared.

Female longevity was expected to increase with honey provision, absence of hosts and absence of mating (Roff 1992). In addition, I predicted that host access would decrease female longevity, under any other conditions. It was also predicted that mating would decrease female longevity in conditions of food absence and host availability. Female total fecundity and rate of oviposition would be increased by honey provision. On the contrary, absence of carbohydrates resources and preventing mating was predicted to delay reproduction.

Maximum daily parasitism of Meteorus communis

A separate, shorter experiment was conducted to assess maximum daily parasitism of *M. communis* females and to corroborate findings of the previous study. Newly emerged females were placed in containers as previously described, but provided with honey in addition to water and two older males (> 1 day of age). After 3 to 6 days,

males were removed and 20 second to third instar host larvae were provided in the same conditions described before, but without the artificial diet, to minimize the potential refuges of the larvae. To estimate the maximum daily parasitism, *M. communis* females received hosts during three consecutive days, in order to avoid underestimation that can occur in 24 h experiments due to insufficient time for acclimation or learning of host cues. Host larvae were replaced daily during the three days, and reared in individual cups and in the same conditions as previously described. Females were arranged in blocks according to day of emergence in order to compare the mortality of the host larvae exposed to females with a control set of host larvae exposed to the same conditions, but without a female wasp in the container. In addition, parasitism level of dead larvae was assessed by dissection. Two outcomes were predicted; the first being that daily parasitism would be less than 10 host per day per female, and the second being that parasitism would increase with time.

Food effects on male Meteorus communis

The effect of food access was investigated for mated males of *M. communis*. Newly emerged males were placed in the same containers and conditions as described for females, and allowed to stay with one older (> 1 day old) female. After 24 h females were removed and males randomly assigned to honey-water or water (control) treatment, following the same protocol as for females, until completing 20 replicates per treatment. As for females, I predicted that honey access would extend male longevity.

Adult *M. communis* were reared in a growth chamber and *P. unipuncta* larvae exposed to parasitism were reared in a rearing room, both at 24 °C, 60 % RH and 16:8 (L:D) photoperiod. Survivorship of adults was checked daily.

Data analysis

The experiments were conducted in a Complete Randomized Block design, using starting day as the blocking factor. The following responses were measured: longevity of females and males, number of *P. unipuncta* larvae parasitized, number of progeny produced daily and total female fecundity. Effect of food resource, host access and mating status on female longevity and fecundity (number of host larvae successfully parasitized per female) were compared using a three-factor factorial ANOVA with fixed effects (Proc Mixed, SAS Institute Inc. 1996). Significant interactions were investigated by slicing by main effects in order to detect significant specific effects of interest (Kuehl 2000). Block effect was removed from models when was not significant. To avoid heterocedasticity, independent variances were estimated for each food treatment, and the suitability of this model was assessed using the Ratio Likelihood Test (P < 0.05, SAS Institute Inc. 1996), except for data of pre-reproductive female period that was transformed $(\ln [x + 1])$ to meet this assumption previous to the analysis (Sokal and Rohlf 1995). Proportion of larvae parasitized was modeled as number of P. unipuncta larvae parasitized / number of P. unipuncta larvae offered / day, using the logit link function and a binomial distribution, and using a factorial ANOVA similar to the previous (SAS Gimmlix macro, Li Hell et al. 1996). The same model was used to estimate the effects of treatments on the proportion of P. unipuncta larvae missing and mortality, in order to

establish the presence of bias in the data. The effect of different numbers of host larvae offered to be parasitized was assessed using simple linear regression analysis.

RESULTS

Influence of temperature, water and carbohydrate availability on adult longevity of *Glyptapanteles militaris*.

Measurements inside the containers with different treatments showed no significant difference among treatments and with growth chambers in temperature and humidity conditions (20 °C growth chamber: Temperature: df = 3,12; F = 0.28; P = 0.84; RH: df = 3,12; F = 0.34; P = 0.80; and 25 °C growth chamber: Temperature: df = 3,11; F = 2.01; P = 0.17; RH: df = 3,12; F = 2.19; P = 0.14).

Temperature and food resources significantly affected the longevity of male and female (with or without male) *G. militaris* (Table 9). Adults lived significantly longer at 20 °C than at 25 °C (Table 9, Figure 12). Provision of honey significantly increased the longevity of male and female *G. militaris* over water alone or no food, at both temperature regimes. Access to honey increased mean adult longevity by ca. three fold at 20 °C (20.3 ± 4.0 d, mean ± SEM, range: 0.5 to 32.5 d) and two fold at 25 °C (7.7 ± 2.7, mean ± SEM, range of 0.5 to 21 d) (Figure 12). Survivorship of adults provided with water alone or in the absence of any resource did not differ and was less than 2.6 d (1.7 ± 0.8 d, mean ± SD, n = 69). Females lived significantly longer than males, and female longevity was approximately twice that of males when honey was available (Table 9a,

Figure 12). The presence of males did not affect female longevity under any food treatments or temperature conditions (Table 9b, Figure 12).

Longevity and fecundity of *Meteorus communis* with and without carbohydrate resources, hosts, and with different mating status.

Pseudaletia unipuncta larvae showed cannibalistic behavior when reared in groups of two or three individuals per cup (blocks 1-4), that resulted in the loss of larvae (10.3 % of larvae offered) before their parasitism status were established. However, a comparison of the proportion of *P. unipuncta* larvae lost / number of larvae offered, revealed that there was no significant bias among treatments that could affect the results (comparing the first 6 days, when females of all treatments were alive, food treatment: df = 1,12; F = 0.05; P = 0.83; mating treatment: df = 1,12; F = 0.04; P = 0.84; and food x mating interaction: df = 1.12; F = 0.05; P = 0.82). The absolute value of parasitism (i.e. not the comparison among treatment effects), however, was perhaps affected by cannibalism, resulting in a possible underestimation of parasitism. Nevertheless, the level of parasitism obtained in larvae reared in groups of 2 or 3 individuals is consistent with the level obtained from those reared individually, indicating that cannibalism was independent of larval parasitism. *Pseudaletia unipuncta* mortality was low (3.3% of larvae offered) and the proportion of dead larvae obtained was not affected by treatments (comparing the first 6 days, when females of all treatments were present, food treatment: df = 1,18; F = 0.06; P = 0.81; mating treatment: df = 1,18; F = 0.00; P = 0.96; and food x mating interaction: df = 1,18; F = 0.00; P = 0.97). Finally, the influence of different number of host larvae offered in blocks 1 to 4 on the number of parasitized larvae

obtained revealed that the increased number of host provided initially only explained 6 % of the parasitism obtained (regression between number of larvae parasitized and number of larvae offered, excluding initial and final days of female life without offspring production: $R^2 = 0.06$, df = 1, 228; F = 13.85; P < 0.01).

Honey provision significantly increased female life between four to nine times, either in females provided or not provided with hosts (Table 10 a and b, Figure 13). Access to host larvae, on the contrary, significantly reduced female longevity when honey was provided, but had no effect in females provided only with water (Table 10 a and c, Figure 13). Females that were paired with males during their first day of life did not differ in their longevity from those that were kept solitary throughout their life and this condition did not interact with the other treatments. Therefore, mating status was removed from the model used to compare the two previous effects by slicing (Table 10 b and c). However, pairing with males did not guaranteed a successful mating and, moreover, eight females that were paired produced only male progeny, indicating either that mating did not occurred or was insufficient to allow the female to produce fertilized eggs. Based in this post hoc information, three reproductive conditions were established for analysis with females that had initial access to males but produced only male offspring grouped into a third category ("unsuccessfully mated females") and compared against truly unmated females (i.e. those that never contacted males, "unmated females) and truly mated females (i.e. those that were with males the first day of life and produced both male and female offspring, "successfully mated females"). Since it was not possible to establish this category among the females that did not received hosts, only treatments that had access to hosts were included in this analysis. In concordance with the previous

result, there was no significant influence of the mating status of females in their longevity, and again females provided with honey-water lived significantly longer than those provided only with water, under any mating condition (Table 11). Finally, the same result was obtained by grouping all the females that produced only male offspring in one category and comparing them with successfully mated females.

Female fecundity was not significantly affected by female contact with males, and this condition did not interact significantly with any other treatment. However, mating condition effect analyzed for females re-grouped according to their offspring sex in the three categories described before, had a significant effect on female fecundity and interacted with the effect of food provision (Table 12). Slicing by food regime revealed that mating condition only affected the fecundity of females provided with honey-water (Table 12a). Successfully mated females produced significantly more offspring than unmated females, while females that had initial contact with males but only produced unfertilized eggs were intermediate in fecundity (Table 12b, Figure 14). Honey provision significantly increased fecundity up to 6 times over the control for all mating conditions except for unmated females. The unmated females showed the same trend but the effect was only marginally significant (Table 12, Figure 14).

The increased number of progeny produced during the female life span due to access to carbohydrate resources or different mating conditions could be the result of two separate effects: 1) an increase in female longevity (significant for honey-water treatment), and 2) an increase in female fecundity *per se*, i. e. a higher rate of oviposition. The distribution of reproductive output on female life span showed an initial increase in parasitism until females reached approximately 20 days of age, and then a decline (Figure

15). To test a possible increase in the rate of oviposition excluding the effect of longevity, the proportion of *P. unipuncta* larvae parasitized (number of larvae parasitized / number of larvae offered) was compared among treatments. Due to a decline in the rate of parasitism with female age (Figure 15), the comparison was performed using the parasitism obtained during the lifespan of the females that had access to water only (the maximum number of days that a female survived with water was used for each block to select the number of days that were enter in the analysis for females of treatments that had access to honey within that block). Honey access did not increase the rate of oviposition significantly, but significant differences in the rate of parasitism were found among reproductive conditions (Table 13, Figure 16). Successfully mated females parasitized an average of $32.9 \pm 4.4\%$ of hosts (mean \pm SEM, n = 8), and differed significantly from unmated females that only parasitized $17.0 \pm 3.4\%$ of hosts (mean \pm SEM, n = 8) (Least Square Means Difference, t = 2.64, P = 0.02), while unsuccessfully mated females parasitized $22.1 \pm 8.1\%$ of hosts (mean \pm SEM, n = 8) and did not differ significantly from the others. A trend of delayed reproduction in females fed with honey was obtained, although it was only marginally significant (Table 14, Figure 17). Mating condition had no significant effect on the length of the pre-reproductive period (Table 14, Figure 17). To explore the effect of mating conditions on the distribution of fecundity of honey-fed females, the number of days required to achieve 50% of total fecundity was compared. Half of reproductive output was achieved around half the life for all the mating conditions: successfully mated females required $18.3 \pm 2.2 d$ (mean \pm SEM, representing 0.45 of life span), unsuccessfully mated females required 19.8 ± 1.7 d (mean \pm SEM, 0.46 of life span), and unmated females required 14.5 \pm 2.7 d (mean \pm SEM, 0.41 of life span), not being significantly different (df = 2,9; F = 0.18; P = 0.84).

Females fed exclusively with water died before reaching a post-reproductive stage, in contrast, females fed with honey-water lived 5.5 ± 4.8 days (mean \pm SD) without producing viable offspring before death (Figure 15). The proportion of males produced by mated females (n=8) did not differed significantly between females fed with honey-water (0.66 \pm 0.18 males) and females fed only with water (0.51 \pm 0.09 males) (df = 1,6; F = 0.68; P = 0.44).

Maximum daily parasitism of Meteorus communis

Only seven of the 15 females used to estimate maximum fecundity produced offspring during the 72 h of the test, therefore the results presented are restricted to those females. Mortality of *P. unipuncta* exposed to the parasitoid did not differ significantly from control larvae mortality (df = 1, 7.91; F = 3.33; P = 0.11). A dissection of 39% of the larvae dead (n = 98) revealed that 11.2% of the larvae were parasitized and died before parasitoids emerged. Female daily fecundity was highly variable and did not increase with time (df = 2, 18; F = 0.45; P = 0.64). An average of 2.7 ± 3.3 larvae were parasitized per day (± SD), with a maximum of 11 larvae parasitized per day (4.5% of the cases). These results confirmed that daily parasitism is on average less than 10 hosts per day and that *P. unipuncta* larvae mortality is independent of female attempts to oviposit.

Food effects on male Meteorus communis

Male longevity was significantly improved by honey provision. Males provided with honey-water lived 26.2 ± 2.6 days in contrast to water-fed males that only lived 4.6 ± 0.6 days (mean \pm SEM, n = 20 per treatment) (df = 1, 21; F = 67.48; P < 0.01).

DISCUSSION

The data obtained in this study support the prediction of a reduction in *G*. *militaris* adult longevity at the higher temperature. Insect metabolism functions between a range of temperatures, and typically with an optimum in which survivorship and fecundity are maximized (Chapman 1998). *Glyptapanteles militaris* adults exposed to higher temperatures (25 °C) lived less than half as long as those at the lower temperature (20 °C), suggesting that the optimal temperature for this species is less than 25 °C. This result is consistent with the findings of Calkins and Sutter (1976) that longevity of adults is greater at 10 °C (10 days) than at 27 °C (6 -7 d). In other study, Oliveira et al. (1998) reported an adult longevity with similar pattern of response to temperature, but significantly reduced in comparison to this and Calkins and Sutter (1976) studies, although they claimed that methodological problems might have reduced it in their experiment.

The prediction of an increased longevity for *G. militaris* adults provided with honey was also supported by this experiment. A similar result was reported by Calkins and Sutter (1976), females provided with fresh halved grapes as carbohydrate source lived on average 3 more days (6 - 7 d) than without food resources (3 - 4 d). As predicted, longevity of males was shorter than female longevity when honey was

provided. Frequent mating has been shown to reduce life span in male and female insects (van den Assem 1986). Although the experiment was not designed to verify the occurrence of multiple mating, *G. militaris* was reported to mate several times in contiguous days (Tower 1915). In the present study, however, male presence did not affect female longevity.

Honey access also dramatically increased the longevity and fecundity of female *M. communis* and the longevity of males. Thireau and Régnière (1995) showed similar values of longevity (22.0 and 23.8 d for males and females, respectively) and higher fecundity (194.2 eggs per unmated female) for *Meteorus trachynotus* (Vier.), provided a 10% honey-water solution. Potential fecundity of the braconid parasitoid Fopius arisanus (Sonan) was reduced in the absence of honey as food source (Bautista et al. 2001). However, in the present study, the proportion of hosts parasitized was not larger in honey-fed females, indicating that the increase in fecundity due to honey provision is mainly a consequence of the increase in female longevity, without detectable effects in the rate of oviposition. Similar results were found for the ichneumonid *Bathyplectes* curculionis (Thomson) (Jacob and Evans 2000), and the pteromalid Nasonia vitripennis (Walker) (Rivero and West 2002). Contrary to expectations, *M. communis* showed a trend of delayed reproduction in honey-fed females. Increased oviposition as a response to stress conditions has been documented for *Leptopilina heterotoma* (Hymenoptera: Eucoilidae) in conditions of low barometric pressure and short photoperiod (Roitberg et al. 1992, 1993). Our result could be interpreted as a faster reproduction of food-deprived females contrasting to a more selective behavior of honey-fed females that resulted in less host parasitized initially.

The trade off between female reproduction and longevity predicts a reduction in the life span of females that allocate resources to reproduction (Roff 1992). Females of the braconid *Cotesia melitaearum* (Wilkinson) provided with fresh flowers but prevented from contacting hosts lived longer than those allowed to parasitize hosts (Lei et al. 1997). Provision of hosts significantly reduced *M. communis* female longevity, confirming this pattern.

Mating stimulates physiological changes that result in increased allocation of resources for maintenance into oogenesis and egg maturation (Jacob and Evans 2000). This could lead to a reduction in female longevity under conditions of low food quality, however it may not have any effect when food quality is high, as shown for the ichneumonid *B. curculionis* (Jacob and Evans 2000). Mating did not affect *M. communis* longevity. However, total fecundity and the rate of oviposition obtained was greater for mated than unmated *M. communis* females. The same result was obtained with the braconid *Cotesia glomerata* (L.); furthermore, dissections of ovaries proved that it was a behavioral change rather than a physiological alteration, since mated and unmated females had the same egg load (Tagawa 1987, in Jervis and Copland 1996). Studies in other parasitoids, however, have not revealed differences between fecundity of mated and unmated females (Jervis and Copland 1996, Sousa and Spence 2000).

The sex of the offspring obtained determined the establishment of a third category regarding mating status: females that initially had access to males but produced only male offspring. In arrhenotokous parasitoids, like both *G. militaris* and *M. communis*, virgin females produce only males. However, mated parasitoid females that receive inviable sperm or that mate several times and could have the spermathecal duct blocked, could

also produce only male progeny (Godfray 1994). In my study, these "unsuccessfully mated" *M. communis* produced an intermediate level of total fecundity and rate of parasitization. While the experiment was not design to provide evidence supporting this mechanistic explanation, one possible explanation of that response is that females received inviable sperm and used it as a source of energy or materials to increase the reproductive output over unmated females.

The importance of food availability in determining the field levels of parasitism are conditioned by direct effects of food on fecundity and life span, and by interactions with female mating status and availability of hosts. Food resources increased dramatically the longevity of G. militaris and the longevity and fecundity of M. communis, under different conditions of temperature, host access and mating status. Habitats with available food resources have shown a significant increase in parasitoid populations than habitats without these resources (Leius 1967). However, in the maizefields where these parasitoids were collected, sugar resources are scarce and if available at all, occur only during limited periods of time (i.e. weed flowers and aphid honeydew) (Dyer and Landis 1996). In addition, my results showed a deleterious effect of high temperature on G. militaris and the same is true for other species of Meteorus (Caballero et al. 1992), indicating that prevailing micro-environmental conditions in the maize fields are probably not favorable for these parasitoids. Milder conditions and more abundant food resources, in addition to alternative hosts, are present in field borders that have late successional vegetation and receive less stress factors (Dyer and Landis 1996, Landis et al. 2000). The existence of resources in extra-field habitats could influence the presence and abundance of parasitoids in crop fields (van Emden 1990, Altieri et al. 1993, Landis

and Marino 1999, Menalled et al. 1999, Tscharntke 2000). Dyer and Landis (1997) showed that the ichneumonid *Eriborus terebrans* (Gravenhorst), with similar requirements of food resources and micro-environmental conditions (Dyer and Landis 1996) as the species studied here, was more abundant near the wooded field edges than at the center of the field despite the distribution of its host. The result of this study provides new information about the biology of the adults of *G. militaris* and *M. communis* that contribute to a better understanding of the abundance of these parasitoids in different landscapes and their potential as biological control agents.

Table 9. ANOVA of the effect of three food regimes (honey + distilled water, distilled water alone, and control without honey and distilled water) and two conditions of temperature (20 and 25 °C) on the longevity of *Glyptapanteles militaris* adults.

Source of Variation	df	F	Р
Temperature	1, 25	9.29	0.01
Food regime	2, 29	37.51	< 0.01
Sex	1, 23	5.70	0.03
Temperature x Food	2, 29	1.84	0.18
Temperature x Sex	1, 23	0.27	0.61
Food x Sex	2, 27	1.64	0.21
Temperature x Food x Sex	2, 27	0.76	0.48

a) Females versus males

b) Females with males versus solitary females

Source of Variation	df	F	Р
Temperature	1, 66	15.97	< 0.01
Food regime	2, 83	100.49	< 0.01
Male presence	1, 66	1.51	0.22
Temperature x Food	2, 83	3.06	0.05
Temperature x Male presence	1,66	0.09	0.77
Food x Male presence	2, 83	1.39	0.25
Temperature x Food x Male presence	2, 83	0.88	0.42



Figure 12. Longevity (mean + SEM) of *Glyptapanteles militaris* adults under three food regimes (honey + distilled water, distilled water alone, and control without honey and distilled water) and two conditions of temperature (20 and 25 °C).
Table 10. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone), host access (10 hosts provided daily versus no host provided) and mating condition (initial access to males versus no access) on the longevity of *Meteorus communis* females.

Source of variation	df	F	Р
Food regime	1, 20.3	238.71	< 0.01
Host access	1, 20.3	6.34	0.02
Food regime x Host	1, 20.3	6.47	0.02
Mating condition	1, 21.4	1.13	0.30
Food regime x Mating condition	1, 20.3	2.95	0.10
Host access x Mating condition	1, 20.3	0.00	0.99
Food regime x Host access x Mating condition	1, 20.3	0.03	0.87

a) Main effects model

b) Effect of Food regime: model sliced by Host access status and without mating effect.

Source of variation	df	F	Р
With hosts	1, 23.7	103.38	< 0.01
Without hosts	1, 23.7	193.43	< 0.01

c) Effect of Host provision: model sliced by Food regime and without mating effect.

Source of variation	df	F	Р

Table 10 (cont'd).

Honey-water	1, 22	7.36	0.01
Control (water alone)	1,22	0.01	0.93



Figure 13. Longevity (mean + SEM) of *Meteorus communis* females under different food regimes (honey + distilled water versus a control of distilled water alone), host access condition (10 hosts provided daily versus no host provided) and mating condition (initial access to males versus no access).

Table 11. ANOVA of the effect of different food regimes (honey + distilled water versus

 a control of distilled water alone) and reproductive conditions ⁽¹⁾ on the longevity of

 Meteorus communis females.

Source of variation	df	F	Р
Food regime	1, 10.4	178.88	< 0.01
Reproductive condition	2, 10.5	0.66	0.54
Food regime x Reproductive condition	2, 10.5	1.76	0.22

⁽¹⁾ Based on offspring production three reproductive conditions were compared: females never paired with males, females paired with males that produce female and male offspring, and females paired with males that only produced male offspring. **Table 12**. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions ⁽¹⁾ on the fecundity (total number of host parasitized) *Meteorus communis* females.

a) Main effects model

Source of variation	df	F	Р
Food	1, 10.4	52.15	<0.01
Mating	2, 10.5	3.91	0.05
Food x Mating	2, 10.5	4.70	0.03

b) Effect of female reproductive conditions: model sliced by food regime.

Source of variation	df	F	Р
Honey-water	2, 9	4.56	0.04
Control (water alone)	2, 9	0.21	0.81

c) Effect of food regime: model sliced by reproductive conditions.

Source of variation	df	F	Р
successfully mated	1, 9.87	34.85	< 0.01
unsuccessfully mated	1, 3.84	19.02	< 0.01
Unmated	1, 10.4	4.29	0.06

⁽¹⁾ Based on offspring production, three reproductive conditions were compared: females never paired with males (that produced only male offspring, unmated), females paired with males that

Table 12 (cont'd)

produced female and male offspring (successfully mated) and females paired with males that only produced male offspring (unsuccessfully mated).



Figure 14. Fecundity (mean number of *Pseudaletia unipuncta* larvae parasitized / female during all life + SEM) of *M. communis* females at different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated and unmated). Columns with different letter differed significantly within food regime (P < 0.05, Least Square Means Differences).



Figure 15. Daily parasitism (mean number of host larvae parasitized / female / day) of *Meteorus communis* females at different food regimes and reproductive conditions: a) females provided with honey-water and b) control females provided only with distilled water.

Table 13. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated, and unmated females) on the percentage of larvae parasitized (number of host parasitized / number of host offered x 100) of *Meteorus communis* females. Fecundity was compared restricting the data to the period of life in which females provided with water were still alive.

Source of variation	df	F	Р
Food regime	1, 18	1.19	0.29
Reproductive condition	2, 18	3.50	0.05
Food regime x Reproductive condition	2, 18	1.98	0.17



Figure 16. Percentage of *Pseudaletia unipuncta* larvae parasitized per day, per female of *Meteorus communis* (mean + SEM), at different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated and unmated). The data was restricted to the parasitism obtained during the period of life of control females.

Table 14. ANOVA of the effect of different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated, and unmated females) on pre-reproductive period (number of days previous to progeny production) of *Meteorus communis* females. Data was transformed (ln [x + 1]) before analysis to achieve homocedasticity.

Source of variation	df	F	Р
Food regime	1, 18	3.93	0.60
Reproductive condition	2, 18	0.18	0.84
Food regime x Reproductive condition	2, 18	0.29	0.75



Figure 17. Pre-reproductive period (number of days previous to progeny production, mean + SEM) of *Meteorus communis* females at different food regimes (honey + distilled water versus a control of distilled water alone) and reproductive conditions (successfully mated, unsuccessfully mated and unmated).

APPENDICES

Appendix 1

Record of Deposition of Voucher Specimens*

The specimens listed on the following sheet(s) have been deposited in the named museum(s) as samples of those species or other taxa, which were used in this research. Voucher recognition labels bearing the Voucher No. have been attached or included in fluid-preserved specimens.

Voucher No.: 2002 - 07

Title of thesis or dissertation (or other research projects):

AGRICULTURAL LANDSCAPE COMPLEXITY HAS MIXED EFFECTS ON PATTERNS OF PARASITOID ABUNDANCE AND DIVERSITY

Museum(s) where deposited and abbreviations for table on following sheets:

Entomology Museum, Michigan State University (MSU)

Other Museums:

Investigator's Name(s) (typed) Alejandro Carlos Costamagna

Date _____8/22/2002

*Reference: Yoshimoto, C. M. 1978. Voucher Specimens for Entomology in North America. Bull. Entomol. Soc. Amer. 24: 141-42.

Deposit as follows:

Original: Include as Appendix 1 in ribbon copy of thesis or dissertation.

Copies: Include as Appendix 1 in copies of thesis or dissertation. Museum(s) files. Research project files.

This form is available from and the Voucher No. is assigned by the Curator, Michigan State University Entomology Museum.

Voucher Specimen Data

Page_1_of _2_Pages

Museum where deposited Other Number of: Adults ဖ 2 ი Adults -Received the above listed specimens for deposit in the Michigan State University Pupae Nymphs Date Larvae Eggs Entomology Museum. Voucher No. 2002-07 USA MI Ingham Co., 2 mi. NW/SW Leslie Label data for specimens collected or Curator used and deposited VI 2001 VI 2001 VI 2001 VI 2001 8/22/02 Investigator's Name(s) (typed) **Alejandro Carlos Costamagna** (Use additional sheets if necessary) Campoletis oxylus (Cresson) Therion circumflexum L. Species or other taxon Campoletis spp. Sinophorus sp. Date

Voucher Specimen Data Page<u>2</u> of <u>2</u> Pages

APPENDIX 2

Protocol followed to make diet for Pseudaletia unipuncta¹

<u>A - List of ingredients</u> :	
Water	2869 ml
Agar	75 g
Wheat germ oil	15 ml
Phosphoric acid diluted in Propionic acid (8.4%)	4.5 ml
Pre-mixed dry diet:	
Alphacel	18 g
Ascorbic acid	15 g
Aureomycin	3 g
Casien (vitamin free)	42 g
Cerophyll	6 g
Cholesterol	0.25 g
D-sucrose	42 g
Methyl-Paraben	6 g
Pinto beans (finely grinded)	420 g
Sorbic acid	4.5 g
Torula yeast	97.5 g
Vanderzant vitamin mixture	6 g
Wesson salt	12 g

<u>B</u> - Directions:

1. Mix 1350 ml of tap water and 75 g of agar. Heat to 90 °C in microwave.

2. Place the wheat germ oil, the phosphoric/propionic acid solution, 525 ml water, and half of the pre-mixed dry diet in a blender. Set blender to high until the mixture is smooth.

3. Agar and water should be heated to 90 °C. Check the degrees periodically with a thermometer. At 90 °C the agar will emulsify. Cool to 74 °C with 234 ml of refrigerated water, then add diet from blender. Turn up stirrer, as fast as possible, without throwing contents out. Mix the other half of dry ingredients and water in blender. When mixed well, pour into cups and containers.

4. Diet should be completely cooled before placing in plastic bags to prevent excessive moisture build up and to let gases escape before storage. Shelf life is about 2 weeks at 4.4 °C. Diet shouldn't be used for 24 hours.

¹ modified from a protocol designed to make Black Cutworm diet, from Jeanette Dyer (personal communication)

REFERENCES CITED

- Altieri, M. A., J. R. Cure and M. A. Garcia. 1993. The role and enhancement of parasitic Hymenoptera biodiversity in agroecosystems, pp. 257–275. *In*: J. LaSalle and I. D. Gauld [eds], Hymenoptera and Biodiversity, C.A.B. International, Oxon, UK.
- Askew, R.R. 1980. The diversity of insect communities in leaf-mines and plant galls. J. Anim. Ecol. 49, 817-829.
- Askew, R. R., and M. R. Shaw. 1989. Parasitoid communities: their size, structure and development, pp. 225-259. In: J. Waage and D. Greathead [eds.]. Insect parasitoids. 13th symposium of the Royal Entomological Society of London. Academic Press, California.
- van den Assem, J. 1986. Mating behavior in parasitic wasps, pp. 137 167. In: J. Waage and D. Greathead [eds.], Insect Parasitoids. Academic Press, NY.
- Barbosa, P. 1998. Agroecosystems and conservational biological control, pp. 39 54.In: P. Barbosa [ed.] Conservational Biological Control. Academic Press.
- Bautista, R. C., E. J. Harris, and R. I. Vargas. 2001. The fruit fly parasitoid Fopius arisanus: reproductive attributes of pre-released females and the use of added sugar as a potential food supplement in the field. Entomol. Exp. Applic. 101: 247 - 255.
- Bonham, C.D., 1989. Measurements for terrestrial vegetation. John Wiley & Sons, New York, NY.
- Breeland, S.G. 1958. Biological studies on the armyworm, *Pseudaletia unipuncta* (Haworth), in Tennessee (Lepidoptera: Noctuidae). J. Tenn. Acad. Sci. 18: 263 347.
- Bugg, R. L., R. T. Ellis, and R. W. Carlson. 1989. Ichneumonidae (Hymenoptera) using extrafloral nectar of Faba Bean (*Vicia faba L.*, Fabaceae) in Massachusetts. Biological Agriculture and Horticulture 6: 107 – 114.
- Burbutis, P. P., and J. A. Stewart. 1979. Blacklight trap collecting of parasitic Hymenoptera. Ent. News 90: 17 22.
- Caballero, P., E. Vargas-Osuna, and C. Santiago-Alvarez. 1992. Biología de Meteorus rubens (Hym.: Braconidae), parasitoide primario de Agrotis ipsilon (Lep.: Noctuidae). Entomophaga 37: 301 – 309.

- Calkins, C. O., and G. R. Sutter. 1976. Apanteles militiraris and its host Pseudaletia unipuncta: biology and rearing. Environ. Entomol. 5: 147-150.
- Cappuccino, N., D. Lavertu, Y. Bergeron, and J. Régnière. 1998. Spruce budworm impact, abundance and parasitism rate in a patchy landscape. Oecologia 114: 236-242.
- Chapman, R. F. 1998. The insects, structure and function. Cambridge University Press.
- Corbett, A., and J. A. Rosenheim. 1996. Impact of natural enemy overwintering refuge and its interaction with the surrounding landscape. Ecol. Entomol. 21: 155 – 164.
- Cossentine, J. E., and L. B. Jensen. 1995. Orthosia hibisci Guenée (Lepidoptera: Noctuidae): indigenous parasitoids and the impact of Earinus limitarus (Say) (Hymenoptera: Ichneumonidae) on its host feeding activity. Can. Entomol. 127: 473-477.
- Covell, C. V. 1984. A field guide to the moth of eastern North America. Houghton Mifflin, Massachusetts.
- DeBach, P., and D. Rosen. 1991. Biological control by natural enemies. Cambridge University Press, Cambridge. New York.
- DiFonzo, C. 2001. Armyworm in wheat. Field Crop Advisory Team Alert 16 (9): 1-2.
- Doak, P. 2000. The effects of plant dispersion and prey density on parasitism rates in a naturally patchy habitat. Oecologia 122: 556 -567.
- van Driesche, R. G., and T. S. Bellows, Jr. 1996. Biological Control. Chapman & Hall, New York.
- Dyer, L. E., and D. A. Landis. 1996. Effects of habitat, temperature, and sugar availability on longevity of *Eriborus terebrans* (Hymenoptera: Ichneumonidae). Environ. Entomol. 25: 1192 1201.
- Dyer, L. E., and D. A. Landis. 1997. Influence of noncrop habitats on the distribution of *Eriborus terebrans* (Hymenoptera: Ichneumonidae) in cornfields. Environ. Entomol. 26: 924 – 932.
- Elliot, N. C., R. W. Kieckhefer, J.-H. Lee, and B. W. French. 1998. Influence of withinfield and landscape factors on aphid predator populations in wheat. Landscape Ecology 14: 239 – 252.
- van Emden, H. F. 1990. Plant diversity and natural enemy efficiency in agroecosystems, pp. 63 80. *In*: M. Mackauer, L. Ehler, and J. Roland [eds.], Critical Issues in Biological Control. Intercept Ltd, Andover, UK.

- England, S. and E. W. Evans. 1997. Effects of pea aphid (Homoptera: Aphididae) honeydew on longevity and fecundity of the alfalfa weevil (Coleoptera: Curculionidae) Parasitoid *Bathyplectes curculionis* (Hymenoptera: Ichneumonidae). Environ. Entomol. 26: 1437 - 1411.
- Fadamiro, H. Y., and G. E. Heimpel. 2001. Effects of partial sugar deprivation on lifespan and carbohydrate mobilization in the parasitoid *Macrocentrus grandii* (Hymenoptera: Braconidae). Ann. Entomol. Soc. Am. 94: 909 – 916.
- Feeny, P. 1976. Plant apparency and chemical defense. Recent Adv. Phytochem. 10: 1-40.
- Fields, P. G., and J. N. McNeil. 1984. The overwintering potential of true armyworm, *Pseudaletia unipuncta* (Lepidoptera: Noctuidae), population in Québec. Can. Entomol. 116: 1647 – 1652.
- Forman, R. T. T., and M. Godron. 1986. Landscape Ecology. Wiley and Sons, New York.
- Foster, M. A., and W. G. Ruesink. 1984. Influence of flowering weeds associated with reduced tillage in corn on a Black Cutworm (Lepidoptera: Noctuidae) parasitoid, *Meteorus rubens* (Nees von Esenbeck). Environ. Entomol. 13: 664 – 668.
- Godfray, H. C. J. 1994. Parasitoids: Behavioral and Evolutionary Ecology. Princeton University Press, New Jersey.
- Golden, D. M., and T. O. Crist. 1999. Experimental effects of habitat fragmentation on old-field canopy insects: community, guild and species responses. Oecologia 118: 371-380.
- Gordh, G., E. F. Legner and L. E. Caltagirone. 1999. Biology of Parasitic Hymenoptera, pp. 355 – 382. In: T. S. Bellows and T. W. Fisher [eds.], Handbook of Biological Control. Principles and Applications of Biological Control, Academic Press, London, UK.
- Guppy, J. C. 1961. Life history and behavior of the armyworm *Pseudaletia unipuncta* (Haw.) (Lepidoptera:Noctuidae), in eastern Ontario. Can. Entomol. 93: 1141-1153.
- Guppy, J. C. 1967. Insect parasites of the armyworm *Pseudaletia unipuncta* (Lepidoptera: Noctuidae), with notes on species observed in Ontario. Can. Entomol. 99: 94-106.
- Hassell, M.P., and J. K. Waage. 1984. Host-parasitoid population interactions. Ann. Rev. Entomol. 29: 89 – 114.

- Hawkins, B. A. 1994. Pattern and process in host-parasitoid interactions. Cambridge University Press, Cambridge.
- Hawkins, B.A., and J.H. Lawton. 1987. Species richness for parasitoids of British phytophagous insects. Nature 326, 788-790.
- Hawkins, B. A., and W. Sheehan [eds.]. 1994. Parasitoid Community Ecology. Oxford Science Publications, Oxford, UK.
- Hendrix III, W. H., and W. B. Showers. 1992. Tracing black cutworm and armyworm (Lepidoptera: Noctuidae) northward migration using *Pithecellobium* and *Calliandra* pollen. Environ. Entomol. 21: 1092 1096.
- Hochberg, M. E. and B. A. Hawkins. 1994. The implications of population dynamics theory to parasitoid diversity and biological control, pp. 472 491. In: B. A. Hawkins and W. Sheehan [eds.], Parasitoid Community Ecology, Oxford Science Publications, Oxford, UK.
- Hu, C., P. Barbosa, and P. Martinat. 1986. Reproductive biology and related hostparasitoid interactions between the gypsy moth, Lymantria dispar and Glyptapanteles flavicoxis, a gregarious endoparasitoid. Ann. Appl. Biol. 109: 485-490.
- Huddleston, T. 1980. A revision of the western Paleartic species of the genus *Meteorus* (Hymenoptera: Braconidae). Bull. Br. Mus. Nat. Hist. 41: 1 58.
- Idris, A. B., and E. Grafius. 1995. Wildflowers as Nectar Sources for *Diadegma insulare* (Hymenoptera: Ichneumonidae), a Parasitoid of Diamondback Moth (Lepidoptera: Yponomeutidae). Environ. Entomol. 24: 1726 – 35.
- Iodine, K., and D. N. Ferro. 1990. Persistence of Edovum puttleri (Hymenoptera: Eulophidae) on potato plants and parasitism of Leptinotarsa decemlineata (Coleoptera: Chrysomelidae): effects of resource availability and water. Environ. Entomol. 19: 1732 – 1737.
- Jacob, H. S., and E. W. Evans. 1998. Effects of sugar spray and aphid honeydew on field populations of the parasitoid *Bathyplectes curculionis* (Hymenoptera: Ichneumonidae). Environ. Entomol. 27: 1563 1568.
- Jacob, H. S., and E. W. Evans. 2000. Influence of Carbohydrate Foods and Mating on Longevity of the Parasitoid Bathyplectes curculionis (Hymenoptera: Ichneumonidae). Environ. Entomol. 29: 1088 – 95.
- Jervis, M. A., and M. J. M. Copland. 1996. The life cycle, pp. 63 161. In M. Jervis and N. Kidd [eds.], Insect Natural Enemies, practical approaches to their study and evaluation. Chapman & Hall, London.

- Jervis, M. A., N. A. C. Kidd, M. G. Fitton, T. Huddleston, and H. A. Dawah. 1993. Flower-visiting by hymenopteran parasitoids. J. Nat. Hist. 27: 67 – 105.
- Jones, C. J., and R. A. Weinzierl. 1997. Geographical and temporal variation in Pteromalid (Hymenoptera: Pteromalidae) parasitism of stable fly and house fly (Diptera: Muscidae) pupae collected from Illinois cattle feedlots. Environ. Entomol. 26: 421-432.
- Kaya, H. K., and Y. Tanada. 1969. Responses to high temperature of the parasite *Apanteles militaris* and of its host, the armyworm, *Pseudaletia unipuncta*. Ann. Entomol. Soc. Am. 62: 1303 1036.
- King, B. H. 2001. Breeding strategies in females of the parasitoid wasp *Spalangia* endius: Effects of mating status and size. J. Ins. Behav. 15: 181 – 193.
- Křivan, V., and E. Sirot. 1997. Searching for food or hosts: the influence of parasitoids behavior on host-parasitoid dynamics. Theoretical Population Biology 51: 201 209.
- Krombein, K. V., P. D. Hurd, D. R. Smith, and B. D. Burks. 1979. Catalog of Hymenoptera in America north of Mexico. Smithsonian Institution, Washington, D.C.
- Kruess, A., and T. Tscharntke. 1994. Habitat fragmentation, species loss, and biological control. Science 264, 1581-1584.
- Kruess, A., and T. Tscharntke. 2000a. Effects of habitat fragmentation on plant-insect communities, pp.53 – 70. In: B. Ekbom, M. Irwin, and Y. Robert [eds.], Interchanges of insects between agricultural and surrounding landscapes. Kluwer Academic Publishers, Dordrecht.
- Kruess, A., and T. Tscharntke. 2000b. Species richness and parasitism in a fragmented landscape: experiments and field studies with insects on *Vicia sepium*. Oecologia 122: 129 137.
- Kuehl, R. O. 2000. Design of Experiments, Statistical Principles of Research Design and Analysis, second edition. Duxbury, Pacific Grove, California.
- Landis, D. A., and M. J. Hass. 1992. Influence of landscape structure on abundance and within-field distribution of European Corn Borer (Lepidoptera: Pyralidae) larval parasitoids in Michigan. Environ. Entomol. 21: 409 416.
- Landis, D. A., and P. C. Marino. 1999. Landscape Structure and Extra-Field Processes: Impact on Management of Pests and Beneficials, pp. 79 – 104. *In*: J. R. Ruberson [ed.], Handbook of Pest Management, Marcel Dekker, New York.

- Landis, D. A., and F. D. Menalled. 1998. Ecological considerations in conservation of parasitoids in agricultural landscapes, pp. 101-121. *In*: P. Barbosa [ed.], Conservation Biological Control, Academic Press, California.
- Landis, D. A., S. T. Wratten, and G. M. Gurr. 2000. Habitat Management to Conserve Natural Enemies of Arthropod Pests in Agriculture. Annu. Rev. Entomol. 45: 175-201.
- LaSalle, J. 1993. Parasitic Hymenoptera, Biological Control and Biodiversity, pp. 197 216. In: J. LaSalle and I. D. Gauld [eds.], Hymenoptera and Biodiversity, C.A.B. International, Oxon.
- Latto, J., and M. P. Hassell. 1988. Generalist predators and the importance of spatial density dependence. Oecologia 77: 375 377.
- Lei, G. C., V. Vikberg, M. Nieminen, and M. Kuussaari. 1997. The parasitoid complex attacking Finnish populations of the Glanville fritillary *Melitaea cinxia* (Lepidoptera: Nymphalidae), an endangered butterfly. J. Nat. Hist. 31: 635 648.
- Leius, K. 1963. Effects of pollens on fecundity and longevity of adult *Scambus* buolianae (Htg.) (Hymenoptera: Ichneumonidae). Can. Entomol. 95: 202 207.
- Leius, K. 1967. Influence of wild flowers on parasitism of tent caterpillar and codling moth. Can. Entomol. 99: 444 446.
- Lessells, C. M. 1985. Parasitoid foraging: should parasitism be density dependent? J. Anim. Ecol. 54: 27 41.
- Lewis, W. J., J. O. Stapel, A. M. Cortesero, and K. Takasu. 1998. Understanding how parasitoids balance food and host needs: importance to biological control. Biological Control 11: 175 183.
- Li Hell, R. C., G. A. Milliken, W. W. Stroup, and R. D. Wolfinger. 1996. SAS System for Mixed Models. SAS Institute Inc., Cary NC.
- Marino P. C. and D. A. Landis. 1996. Effect of landscape structure on parasitoid diversity in agroecosystems. Ecol. Appl. 6: 276-284.
- McDougall, S. J., and N. J. Mills. 1997. The influence of hosts, temperature and food sources on the longevity of *Trichogramma platneri*. Entomol. Exp. Appl. 83: 195 - 203.
- McLaughlin, R. E. 1962. The effect of temperature upon larval mortality of the armyworm *Pseudaletia unipuncta* (Harworth). J. Ins. Pathol. 4: 279-284.

- McNeil, J. N., and J. J. Turgeon. 1988. Primary parasitoids of the true armyworm, *Pseudaletia unipuncta* (Lepidoptera: Noctuidae) in Québec. Revue d'entomologie du Québec 32: 1 – 8.
- Menalled, F. D., P. C. Marino, S. Gage, and D. A. Landis. 1999. Does agricultural landscape structure affect parasitism and parasitoid diversity? Ecol. Appl. 9: 634-641.
- Metcalf, R. L., and R. A. Metcalf. 1993. Destructive and useful insects: their habits and control, 5th ed. McGraw-Hill, New York.
- Miller, J. C. 1996. Temperature-Dependent Development of *Meteorus communis* (Hymenoptera: Braconidae), a Parasitoid of the Variegated Cutworm (Lepidoptera: Noctuidae). J. Econ. Entomol., 89: 877 – 880.
- Nicholls, C. I., M. Parrella, and M. A. Altieri. 2001. The effects of a vegetational corridor on the abundance and dispersal of insect biodiversity within a northern California organic vineyard. Landscape Ecology 16: 133 146.
- Ogol, C. K. P. O., J. R. Spence, and A. Keddie. 1998. Natural enemy abundance and activity in maize-leucaena agroforestry system in Kenya. Environ. Entomol. 27: 1444 1451.
- Oliveira, L., R. Melo, and J. Tavares. 1998. Response of *Glyptapanteles militaris* (Walsh) (Hymenoptera: Braconidae), a larval parasitoid of the armyworm, *Mythimna unipuncta* (Haworth) (Lepidoptera: Noctuidae), to different temperatures. J. Hym. Res. 7: 268 – 273.
- Oliveira, L., R. Melo, and J. Tavares. 1999. Host age of *Pseudaletia unipuncta* (Lepidoptera: Noctuidae) and parasitic capacity of *Glyptapanteles militaris* (Hymenoptera: Braconidae). Environ. Entomol. 28: 513 – 517.
- Olson, D. N., H. Fadamiro, J. G. Lundgren, and G. E. Heimpel. 2000. Effects of sugar feeding on carbohydrate and lipid metabolism in a parasitoid wasp. Phisiol. Entomol. 25: 17 26.
- Onstad, D. W., J. P. Siegel, and J. V. Maddox. 1991. Distribution of parasitism by Macrocentrus grandii (Hymenoptera: Braconidae) in maize infested by Ostrinia nubilalis (Lepidoptera: Pyralidae). Environ. Entomol. 20: 156-159.
- Pavuk, D. M., and G. W. Barrett. 1993. Influence of successional and grassy corridors on parasitism of *Plathypena scabra* (F.) (Lepidoptera: Noctuidae) larvae in soybean agroecosystems. Environ. Entomol. 22: 541 – 546.

- Powers, J. S., P. Sollins, M. E. Harmon, and J. A. Jones. 1999. Plant-pests interactions in time and space: A Douglas-fir bark beetle outbreak as a case study. Landscape Ecology 14: 105 – 120.
- Price, P. W. 1991. Evolutionary theory of host and parasitoid interactions. Biol. Control 1: 83-93.
- Price, P. W. 1994. Evolution of parasitoid communities, pp. 472 491. In: B. A. Hawkins and W. Sheehan [eds.], Parasitoid Community Ecology, Oxford Science Publications, Oxford, UK.
- Price, P. W. 1997. Insect Ecology. Wiley and Sons, New York.
- Quicke, D. L. J. 1997. Parasitic Wasps. Chapman & Hall, New York.
- Reeve, J. D. 1987. Foraging behavior of *Aphytis melinus*: effects of patch density and host size. Ecology, 68: 530 538.
- Rivero, A., and S. A. West. 2002. The physiological costs of being small in a parasitic wasp. Evolutionary Ecology Research 4: 407 420.
- Roff, D. A. 1992. The evolution of life histories: theory and analysis. Chapman and Hall, NY.
- Roitberg, B. D., M. Mangel, R. G. Lalonde, C. A. Roitberg, J. J. M. van Alphen, and L. Vet. 1992. Seasonal dynamic shifts in patch exploitation by parasitic wasps. Behavioral Ecology 3: 156 – 165.
- Roitberg, B. D., J. Sircom, C. A. Roitberg, J. J. M. van Alphen, and M. Mangel. 1993. Life expectancy and reproduction. Nature 364: 108.
- Roland, J. 2000. Landscape Ecology of Parasitism, pp. 235-253. In: M. E. Hochberg and A. R. Ives [eds.], Parasitoid population biology. Princeton University Press, New Jersey.
- Roland, J., and P. D. Taylor. 1997. Insect parasitoid species respond to forest structure at different spatial scales. Nature 386: 710-713.
- Rolim, A. E. 1983. Laboratory comparison of the potential of Apanteles ruficrus Haliday and Apanteles militaris Walsh (Hymenoptera: Braconidae) for control the armyworm, Pseudaletia unipuncta (Haworth) (Lepidoptera: Noctuidae) in Michigan. Ph. D. Thesis, Michigan State University, East Lansing.
- Ryszkowski, L., J. Karg, G. Margarit, M. G. Paoletti, and R. Zlotin. 1993. Aboveground insect biomass in agricultural landscapes of Europe, pp. 71 – 82. *In*: R. G.

H. Bunce, L. Ryszkowski, and M. G. Paoletti [eds.], Landscape ecology and agroecosystems. CRC Press, Boca Raton, FL.

- SAS Institute. 1996. SAS/STAT User's guide for personal computers. 6th ed., vol 2, SAS Institute, Cary, North Carolina.
- Sokal R. R., and F. J. Rohlf. 1995. Biometry, 3rd ed. W. H. Freeman, New York.
- Sousa, J. M., and J. R. Spence. 2000. Effects of mating status and parasitoid density on superparasitism and offspring fitness in *Tiphodytes gerriphagus* (Hymenoptera: Scelionidae). Ann. Entomol. Soc. Am. 93: 548 553.
- Stehr, F. W. [ed.]. 1987. Immature insects. Volume 1. Kendall/Hunt, Iowa.
- Steinkraus, D. C., A. J. Mueller, and R. A. Humber. 1993. Furia virescens (Thaxter) Humber (Zygomycetes: Entomophthoraceae) Infections in the Armyworm, Pseudaletia unipuncta (Haworth) (Lepidoptera: Noctuidae) in Arkansas with Notes on Other Natural Enemies. J. Entomol. Sci. 28: 376 - 386.
- Stiling, P. D. 1987. The frequency of density dependence in insect host-parasitoid systems. Ecology 68: 844 856.
- Teder, T., M. Tanhuanpää, K. Ruohomäki, P. Kaitaniemi, and J. Henriksson. 2000. Temporal and spatial variation of larval parasitism in non-outbreaking populations of a folivorous moth. Oecologia 123: 516-524.
- Thies, C., and T. Tscharntke. 1999. Landscape structure and biological control in agroecosystems. Science 285: 893-895.
- Thireau, J. C., and J. Régnière. 1995. Development, reproduction, voltinism and host synchrony of *Meteorus trachynotus* with it hosts *Choristoneura fumiferana* and *C. rosaceana*. Entomol. Exp. Applic. 76: 67 82.
- Thompson, R. E. 1966. Seasonal appearance of selected species of Noctuidae in Michigan. M. S. Thesis, Michigan State University, East Lansing.
- Tower, D. G. 1915. Biology of Apanteles militaris. J. Agric. Res. 5: 495-507.
- Tscharntke, T. 2000. Parasitoid populations in the agricultural landscape, pp. 235-253. In: M. E. Hochberg and A. R. Ives [eds.], Parasitoid population biology. Princeton University Press, New Jersey.
- Untung, K. 1978. The distribution and bionomics of the armyworm, *Pseudaletia* unipuncta (Haw.) in Michigan. M. S. Thesis, Michigan State University, East Lansing.

- Vinson, S. B., and P. Barbosa. 1989. Interrelatioships of nutritional ecology of parasitoids, pp. 673 – 695. In: F. Slansky Jr., and J. G. Rodriguez [eds.], Nutritional ecology of insects, mites, spiders, and related invertebrates. John Wiley and Sons, NY.
- Wäckers, F. L. 2001. A comparison of nectar- and honeydew sugars with respect to their utilization by the hymenopteran parasitoid *Cotesia glomerata*. J. Insect Physiology 47: 1077 – 1084.
- West, K.J. 1988. Biology and Host Relations of *Meteorus communis* (Cresson). M. S. Thesis, Oregon State University, Corvallis.
- West, K. J. & J. C. Miller. 1989. Patterns of host exploitation by *Meteorus communis* (Hymenoptera:Braconidae). Environ. Entomol. 18: 537-540.
- Whitfield, J. B., and C. N. Lewis. 2001. Analytical survey of the braconid wasp fauna (Hymenoptera: Braconidae) on six Midwestern U. S. tallgrass prairies. Ann. Entomol. Soc. Am. 94: 230 238.
- Wiens, J. A., and B. T. Milne. 1989. Scaling of 'landscapes' in landscape ecology, or, landscape ecology from a beetle's perspective. Landscape Ecology 3: 87 96.
- Williams, C. E., D. M. Pavuk, D. H. Taylor, and T. H. Martin. 1995. Parasitism and disease incidence in the Green Cloverworm (Lepidoptera: Noctuidae) in stripintercropped soybean agroecosystems. Environ. Entomol. 24: 253 – 260.

