PHOSPHINE RESISTANCE AND BIOLOGICAL CONTROL OF PSOCIDS (PSOCODEA: LIPOSCELIDIDAE)

By

JAMES K. DANSO

Bachelor of Science in Agriculture
Kwame Nkrumah University of Science and Technology
Kumasi, Ghana
2012

Master of Philosophy in Crop Protection (Entomology)
Kwame Nkrumah University of Science and Technology
Kumasi, Ghana
2017

Submitted to the Faculty of the
Graduate College of the
Oklahoma State University
in partial fulfillment of
the requirements for
the Degree of
DOCTOR OF PHILOSOPHY
July, 2021

PHOSPHINE RESISTANCE AND BIOLOGICAL

CONTROL OF PSOCIDS

(PSOCODEA: LIPOSCELIDIDAE)

Dissertation Approved:

Dr. George P. Opit

Dissertation Adviser

Dr. Bruce H. Noden

Dr. Kristopher L. Giles

Dr. Carla L. Goad

ACKNOWLEDGEMENTS

I would like to express my sincere gratitude to my major advisor, Dr. George P. Opit for his professional guidance, endless support, and encouragement. Dr. Opit was very influential in my graduate education achievements. Also, I would like to thank my committee members Dr. Bruce H. Noden, Dr. Kristopher L. Giles, and Dr. Carla L. Goad for their professional guidance and input into this dissertation.

I extent my sincere gratitude to Charlie Konemann and Zhaorigetu Chen for their excellent technical assistance. Also, I am grateful to the administrative board of the Department of Entomology and Plant Pathology (EPP) for providing partial financial support for my program. Again, I thank all faculty members, the staff, and my fellow graduate students in the EPP Department for providing me with a friendly environment in the course of my program.

I would like to express my heartfelt appreciation to my dear wife Mrs. Linda A. Danso for her unconditional love, support, and prayers that was the bedrock of my completion of this program. I am also grateful to my mother-in-law, Reverend Cecilia Manu for her constant prayers and encouragement. Again, I would like to thank my brothers and sisters especially, Charles Obeng, Gloria S. Danso, Christabel Danso, and Shadrack A. Danso for their prayers. Special thanks to Darren P. Jackson for his initial support. Moreover, I am indebted to my former academic advisor, Dr. Enoch A. Osekre, Department of Crop and Soil Sciences, KNUST-Ghana, for his immense contributions toward my higher education achievements. I am also grateful to Mr. James Ken-Kwofie, Newark, New Jersey, USA. Finally, I am very grateful to my parents, Mr. Frank C. Danso and Deaconess Georgina Okai for their physical, spiritual, and financial support in my entire academic adventure. Above all, I am forever grateful to God (Emmanuel).

Acknowledgements reflect the views of the author and are not endorsed by committee members or Oklahoma State University.

Name: JAMES K. DANSO

Date of Degree: JULY, 2021

Title of Study: PHOSPHINE RESISTANCE AND BIOLOGICAL CONTROL OF PSOCIDS (PSOCODEA: LIPOSCELIDIDAE)

Major Field: ENTOMOLOGY

Abstract: Phosphine (PH_3) resistance and tolerance contribute to the increased importance of psocids (Psocodea: Liposcelididae) as stored-product pests worldwide. However, there is currently no superior substitute for this fumigant to meet domestic and international export phytosanitary requirements in the United States. These studies were conducted in the context of developing PH_3 resistance monitoring strategies to manage resistance in psocids and assessing predatory behaviors of *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) to measure their potential as effective biocontrol candidates for psocid management. The first objective was to establish the levels of PH_3 tolerance in laboratory susceptible strains of psocids using a modified FAO Method No.16. To accomplish this, discriminating doses (DDs) were established for lab-cultured susceptible adults of *Liposcelis bostrychophila* (Badonnel), *L. entomophila* (Enderlein), *L. decolor* (Pearman), *L. paeta* Pearman, *L. rufa* Broadhead, *L. obscura* Broadhead, *L. fusciceps* Badonnel, and *Lepinotus reticulatus* Enderlein (Psocodea: Liposcelididae) over a 20-h and 72-h fumigation period. The established DDs showed a range of 65.6–697.3 ppm and 18.1–194.5 ppm over 20-h and 72-h fumigation periods, respectively. The higher heterogeneity levels found in the standard 20-h fumigation period indicates the potential for a significant increase in PH_3 resistance in field populations subjected to PH_3 fumigation. The second and third objectives were to assess the predatory efficiency of *C. eruditus* and *C. malaccensis* based on their foraging behaviors —functional and numerical responses of these predators to different developmental stages of *L. decolor*. Both predatory mites showed Holling Type II functional response to nymphs, adult males, or adult females of *L. decolor*. The estimated functional response variables showed that *C. eruditus* performance was preferable to *C. malaccensis*. However, numerical response parameters of predators indicated that *C. malaccensis* was more efficient than *C. eruditus.* The fourth objective was to provide quantitative data based on the ecological interactions of *C. eruditus* or *C. malaccensis* and *L. decolor* under different release ratios (predator-prey ratios), temperatures (°C), and relative humidities (RH %) over a 40-d period to determine the optimal psocids management conditions for each predatory mite. The results showed that low RH (≤63%) undermine the efficacy of both predatory mites, however, *C. eruditus* and *C. malaccensis* caused psocid population suppression of ~ 67.1–97.2% and increased their progeny by ~ 117.1–1182.6% for the 1:20–10:20 release ratios, temperatures of 20–32°C, and 75–85% RH. Future research should be aimed at using the established DDs for the detection of PH_3 resistance and estimation of resistance frequencies in field-collected populations of the psocid species investigated. Also, field evaluation of both predatory mites and their compatibility with other stored-product pest management tactics are needed to permit release for psocid management in the United States.

TABLE OF CONTENTS

Chapter Page

I. BACKGROUND AND OBJECTIVES 1

Objectives 7

II. LITERATURE REVIEW 10

Psocoptera: Etymology, Taxonomy, and Description of Psocids 10
Ecology, Biology, and Behavior of Psocids 12
Worldwide Distribution of Psocids 14
Change in Pest Status of Psocids 16
Economic Impact of Psocid Infestations 17
Health Concerns and Safety Hazards Associated with Psocids 19
Management of Psocids 20
Physical control strategies for psocid management 20
Chemical control and insecticide resistance 21
Contact insecticides 21
Fumigants 24
Phosphine resistance challenges 26
Phosphine resistance bioassay: Diagnostic methods 28
Biological control of storage arthropod pests 30
Cheyletid predatory mites in storage ecosystems 33
Ecology, biology, and behavior of *Cheyletus eruditus* and *Cheyletus malaccensis* 36
Predator-Prey Relations 38

III. ESTABLISHING DISCRIMINATING DOSES OF PHOSPHINE FOR ADULTS OF SUSCEPTIBLE LABORATORY STRAINS OF PSOCIDS (PSOCODEA: LIPOSCELIDIDAE) 45

Abstract 46
Introduction 47
Materials and Methods 51
Results 56

Chapter Page

Discussion 59
Acknowledgements 66

IV. FUNCTIONAL RESPONSES OF PREDATORY MITES, CHEYLETUS ERUDITUS (SCHRANK) AND CHEYLETUS MALACCENSIS OUDEMANS (TROMBIDIFORMES: CHEYLETIDAE) TO DIFFERENT LIFE STAGES OF LIPOSCELIS DECOLOR (PEARMAN) (PSOCODEA: LIPOSCELIDIDAE) ...71

Abstract 72
Introduction 73
Materials and Methods 76
Results 82
Discussion 86
Acknowledgements 92

V. NUMERICAL RESPONSES OF PREDATORY MITES, CHEYLETUS ERUDITUS (SCHRANK) AND CHEYLETUS MALACCENSIS OUDEMANS (TROMBIDIFORMES: CHEYLETIDAE) TO DIFFERENT LIFE STAGES OF LIPOSCELIS DECOLOR (PEARMAN) (PSOCODEA: LIPOSCELIDIDAE) .101

Abstract 102
Introduction 103
Materials and Methods 105
Results. 111
Discussion 115
Acknowledgements 121

VI. ECOLOGICAL INTERACTION OF PREDATORY MITES, CHEYLETUS ERUDITUS (SCHRANK) AND CHEYLETUS MALACCENSIS OUDEMANS (TROMBIDIFORMES: CHEYLETIDAE) AND PREY, LIPOSCELIS DECOLOR (PEARMAN) (PSOCODEA: LIPOSCELIDIDAE) UNDER DIFFERENT THERMO-HYGROMETRIC REGIMES 133

Abstract 134
Introduction 135
Materials and Methods 137
Results 142
Discussion 144
Acknowledgements 151

Chapter Page

VII. CONCLUSIONS 155

REFERENCES 162

LIST OF TABLES

Table Page

1. Probit analyses of mortality responses of susceptible adult psocids of eight species to ranges of phosphine gas concentrations over a 20-h exposure period at 25 ± 1°C and 75 ± 5% RH. Concentrations are in parts per million (ppm) 67

2. Probit analyses of mortality responses of susceptible adult psocids of eight species to ranges of phosphine gas concentrations over a 72-h exposure period at 25 ± 1°C and 75 ± 5% RH. Concentrations are in parts per million (ppm) 68

3. Comparison of lethal concentrations required to kill 50%, 95%, and 99% (LC_{50}, LC_{95}, and LC_{99}) of *L. obscura* with concentrations required to kill similar proportions of *L. bostrychophila*, *L. paeta*, *L. fusciceps*, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila* for the 20-h fumigation period. Additionally, similar comparisons for *L. reticulatus* with the other seven species for the 72-h fumigation period. Fumigations were conducted at 25 ± 1°C and 75 ± 5% RH. Lethal concentrations are in parts per million (ppm)...69

4. Comparison of discriminating doses of susceptible adult psocids of eight species based on 20-h and 72-h exposure to ranges of phosphine gas concentrations at 25 ± 1°C and 75 ± 5% RH. Phosphine concentration reduction indexes (PCRIs) were estimated for the eight species. Concentrations are in parts per million (ppm)....70

5. Maximum-likelihood estimates from logistic regressions of the proportion of *L. decolor* females, males, and nymphs consumed by *C. eruditus* or *C. malaccensis* female adults as a function of initial prey density 93

6. Parameter estimates for Holling Type II functional responses of adult female *C. eruditus* or *C. malaccensis* to *L. decolor* at different life stages and densities predicted by a linear transformation of Hollings disc equation: $1/N_a = 1/a*NT + T_h/T$...95

Table Page

7. Summary of the tests for the fixed effects of prey stage (PS) and prey density (N) *L. decolor* on numbers of prey killed (N_a) and searching efficiency (N_a/N) of predatory mites, for *Cheyletus eruditus* (CE) or *Cheyletus malaccensis* (CM).......96

8. Numbers of prey killed (N_a) (mean ± SE) per female *C. eruditus* or *C. malaccensis* per day (d^{-1}) exposed to *L. decolor* at different developmental stages and densities 97

9. Per capita searching efficiency (N_a/N) (mean ± SE) of predatory mites, *C. eruditus* or *C. malaccensis* exposed to different developmental stages and densities of *L. decolor* 98

10. Parameters (± SE) for the regression models, linear: y = a + bx, log: y = a + blnx, and polynomial: $y = a + bx + cx^2$ (Equations 1, 13, and 1003, respectively in TableCurve 2D) describing the ovipositional responses of *C. eruditus* and *C. malaccensis* on a diet of *L. decolor* females, males and nymphs at a constant temperature of 24 ± 1°C and 85 ± 5% RH..........122

11. Parameters (± SE) for the regression model, log: y = a + blnx (Equations 13 in TableCurve 2D) describing the efficiency of conversion of ingested food by *C. eruditus* and *C. malaccensis* on a diet of *L. decolor* females, males and nymphs at a constant temperature of 24 ± 1°C and 85 ± 5% RH 124

12. Summary of the tests for the fixed effects of prey stage (PS) and prey density (N) of *L. decolor* on numbers of eggs laid (N_o), oviposition efficiency (N_o/N), and the efficiency of conversion of ingested prey (ECI) of predatory mites, for *Cheyletus eruditus* (CE) or *Cheyletus malaccensis* (CM) 125

13. Numbers of eggs laid (N_o) (mean ± SE) per female *C. eruditus* or *C. malaccensis* per day (d^{-1}) exposed to *L. decolor* at different developmental stages and densities 126

14. Per capita oviposition efficiency (N_0/N) (mean ± SE) of predatory mites, *C. eruditus* or *C. malaccensis* exposed to different developmental stages and densities of *L. decolor* 127

15. Per capita efficiency of conversion of ingested food (mean ± SE) of predatory mites, *C. eruditus* or *C. malaccensis* exposed to different developmental stages and densities of *L. decolor*..........128

Table Page

16. Summary of the tests for the fixed effects of predator-prey ratio (P-P-R), temperature (T), and relative humidity (RH), and interactions for percentage prey surviving (%) and per capita progeny production (% increase in population) of predatory mites, for *Cheyletus eruditus* (CE) or *Cheyletus malaccensis* (CM) exposed to initial prey density (20 females) of *L. decolor* over a 40-day period 152

17. Mean percentage prey surviving (±SE) over a 40-day period. Predatory mite species (P) were *C. eruditus* (CE) or *C. malaccensis* (CM), initial prey density was 20 females of *L. decolor*, five levels of predator-to-prey ratio (P-P-R) (0:20, 1:20, 2:20, 4:20, and 10:20), four levels of temperature (T) (20, 24, 28, and 32°), and three levels of relative humidity (RH) (63, 75, and 85%) 153

18. Mean per capita progeny production (%) (±SE) over a 40-day period. Predatory mite species (P) were *C. eruditus* (CE) or *C. malaccensis* (CM), initial prey density was 20 females of *L. decolor*, five levels of predator-to-prey ratio (P-P-R) (1:20, 2:20, 4:20, and 10:20), four levels of temperature (T) (20, 24, 28, and 32°), and three levels of relative humidity (RH) (63, 75, and 85%) 154

LIST OF FIGURES

Figure Page

1. Functional responses of *C. eruditus* or *C. malaccensis* to different life stages (female, male, and nymph) and densities of *L. decolor*. Parameters for the fitted Holling Type II functional response curves are in Table 1 and Table 2. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* for each prey life stage 100

2. Ovipositional response of *C. eruditus* or *C. malaccensis* to different life stages (female, male, and nymph) and densities of *L. decolor*. Parameters for the fitted regression curves are in Table 1. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* for each prey life stage, and each observation was mean of seven days (7d) exposure period...........................130

3. Efficiency of conversion of ingested prey by *C. eruditus* or *C. malaccensis* when offered different life stages (female, male, and nymph) and densities of *L. decolor*. Parameters for the fitted regression curves are in Table 2. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* for each prey life stage...131

4. Predation and oviposition rate of *C. eruditus* or *C. malaccensis* exposed to different densities of *L. decolor*. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* and each observation was mean of seven days (7d) exposure period...132

CHAPTER I

BACKGROUND AND OBJECTIVES

Psocids of the genus *Liposcelis* (Psocodea: Liposcelididae; formerly order Psocoptera) have emerged as important pests of stored products worldwide over the last two-to-three decades and they cause unacceptable economic losses, mainly through costs of disinfestation to prevent damage and possible rejection of commodities (Phillips and Throne 2010, Nayak et al. 2014). Psocids belong to a relatively small order of insects with approximately 5,500 described species in 41 families and it is one of the only three insect orders known to have species that are a serious pest of stored products (Rees 2004, Ahmedani et al. 2010). The ecological significance of psocids was previously underestimated based on the available surveillance data and they were only considered as nuisance pests of minor economic importance (Stejskal et al. 2015, Athanassiou and Rumbos 2018). Psocids were often overlooked because of their small size and the existence of more damaging coleopteran and lepidopteran primary pests that influence pest management decisions (Villabobos et al. 2005). However, with the current advances in monitoring procedures and surveillance techniques coupled with effective decision support systems, severe infestations of psocids have been detected and estimated in a wide range of geographical locations, and in diverse storage systems and ecological conditions, mostly, in warm temperate d tropical regions worldwide (Dou et al. 2013,

Diaz-Montano et al. 2015, Liu et al. 2017). The economic impacts of psocids are now evidenced in grain storage, handling, and processing facilities, product warehouses, museums, and other stored commodities particularly, in warm-humid regions, including parts of the United States (Arbogast et al. 2000, Opit et al. 2009, Gautam et al. 2013). In addition to the fact that commodities infested by psocids can be rejected, severe infestations of psocids are also associated with human and animal health problems because they distribute molds and vector disease pathogens (Barker 2007, Chin et al. 2010, Nayak et al. 2014). Severe psocids infestations have been reported in West Africa and South-East Asia (Pike et al.1991), Indonesia (Kleih and Pike 1995), India (Rajendran 1994), China (Wang et al. 1998), Australia (Rees 1998, Nayak 2014), Italy (Trematerra and Fiorilli 1999), United States (Arbogast et al. 2000), Croatia (Kalinovic et al. 2006), Zimbabwe (Mashaya 1999), Czechoslovakia (Obr 1978), Canada (Smith 1985), United Kingdom (Turner 1994), Czech Republic, and Portugal (Kucerova 2006).

Currently, four genera of psocids, *Liposcelis*, *Lepinotus*, *Lachesilla*, and *Trogium* have been described as stored grain pests with *Liposcelis*, *Lepinotus*, and *Lachesilla* having a worldwide distribution, but notably *Liposcelis* has the most economically important psocid species (Ahmedani et al 2010, Liu et al. 2017). In the United States, *Liposcelis* and *Lepinotus* are considered the most economically important genera. Four psocid species, *Liposcelis bostrychophila* Badonnel (Psocodea: Liposcelididae), *Liposcelis decolor* (Pearman), *Liposcelis entomophila* (Enderlein), and *Liposcelis paeta* Pearman are considered the most economically important species with a worldwide distribution (Nayak et al. 2014). In addition to these species, *L*iposcelis *brunnea* Motschulsky, *Liposcelis obscura* Broadhead, *Liposcelis corrodens* (Heymons), *Liposcelis rufa* Broadhead, *Liposcelis fusciceps* Badonnel,

Liposcelis pearmani Lienhard, and *Lepinotus reticulatus* Enderlein (Psocodea: Trogiidae) were found infesting stored commodities in the United States (Mockford 1993, Lienhard and Smithers 2002, Opit and Throne 2008, Aminatou et al. 2011, Opit et al. 2011, Gautam et al., 2013, 2016). Also, *Trogium pulsatorium* Linnaeus (Psocodea: Trogiidae) and *Lachesilla pedicularia* Linnaeus (Psocodea: Lachesillidae) were occasionally reported (Ahmedani 2010). Nevertheless, *L. entomophila* and *L. decolor* are the most predominant species infesting stored grains in the United States (Throne et al. 2006, Opit et al. 2009).

Psocids have risen to prominence as a serious pest of stored products over the last two-to-three decades and are now considered as the most important emerging taxonomic group of pests that pose a new threat to global food safety and security (Nayak 2014, Stejskal et al. 2015, Wei et al. 2020). The pest status of psocids stems partly from the quantitative loss caused by their feeding on the endosperm and germ of grains leading to significant grain weight loss and germination failure (Kucerova 2002, Gautam et al. 2013, Athanassiou et al. 2014). Additionally, psocids have the ability to thrive on and deteriorate the quality of a variety of substrates, including stored commodities because of their presence and the resulting exuviae, feces, and cadavers (Opit and Throne 2008, Athanassiou and Rumbos 2018). Moreover, consumer concerns about commodity quality may lead to rejection of commodities destined for export that are infested with psocids thereby causing economic losses. In countries such as Australia, psocid infested grain is banned for export (Nayak 2010). Furthermore, psocid natural ecology which includes short generation times allows psocids to rapidly colonize new habitats and to quickly proliferate given conducive environmental conditions (Opit and Throne 2008, Athanassiou et al. 2014). Psocids are also

reported to be associated with human and animal health problems because they distribute molds and vector disease pathogens (Barker 2007, Chin et al. 2010).

A key factor that has led to the rise of psocids to become pests of substance is the fact that they are not successfully managed by currently available pest management strategies, particularly, chemical control tactics that have been used effectively to manage coleopteran and lepidopteran pests. This cannot be attributed to previous exposure, but to a natural phenomenon (Nayak et al. 2014, Athanassiou and Rumbos 2018). Psocids quickly develop resistance to contact insecticides that have been used successfully against other stored product insect pests. For instance, Daglish et al. (2003) found that piperonyl butoxide + chlorpyrifos-methyl was not effective against *L. entomophila*. Also, *L. entomophila*, *L. bostrychophila*, and *L. paeta* were found to be tolerant to the pyrethroid bioresmethrin + piperonyl butoxide (Nayak et al. 1998). Deltamethrin was not able to provide long-term protection against *L. paeta, L. bostrychophila*, and *L. entomophila* (Nayak et al. 2002 a, b). Pyrethroids are relatively ineffective against *Liposcelis* species (Guedes et al. 2008a). Spinosad was found to be ineffective in managing psocids, except *L. entomophila* (Lord and Howard 2004), and insect growth regulators have also been shown to be less effective against psocids (Nayak et al. 1998, Athanassiou et al. 2010).

Psocids have shown resistance to the fumigant phosphine (Hydrogen phosphide: PH_3) which is widely used in the management of insect pests in stored grains in the United States and worldwide (Opit et al. 2012, Nayak et al. 2014, Gautam et al. 2016, Afful et al. 2018). Recent studies in Australia found that adults in field populations of *L. bostrychophila* survived PH_3 gas concentrations approximately 5x greater than that which kills their susceptible counterpart strain, whereas resistant strains of *L. decolor* and *L. entomophila*

tolerated concentrations of PH_3 approximately 6x and 7x higher than those that kill the susceptible strains, respectively (Nayak et al. 2000). These authors also showed that eggs of resistant *L. bostrychophila* survived a PH_3 concentration of 2 g/m^3 whereas 0.031 g/m^3 was effective against the susceptible strain, the former concentration was 65x higher than the latter. Delay in egg development and hatching in the presence of PH_3 has also been reported (Nayak et al. 2014). The extensive and inefficient use of PH_3 against stored-product insect pests has led to control failures and selection of highly resistant populations in stored product insect pests in the United States. Notably, resistance has been found in *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) and *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) in Kansas and Oklahoma (Opit et al. 2012, Cato 2015, Chen et al. 2015, Afful et al. 2018*), Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae) in Oklahoma (Konemann et al. 2017), and *T. castaneum* and *Plodia interpunctella* (Hubner) (Lepidoptera: Pyralidae) in California (Gautam et al. 2016). There are currently no published studies on PH_3 resistance in psocids in the United States. However, it is imperative to develop PH_3 resistance monitoring strategies for managing psocids since PH_3 remains the only available effective broad-spectrum fumigant for managing stored-grain pests in the United States (Opit et al. 2012, Hagstrum et al. 2012, Gautam et al. 2016, Konemann et al. 2017). Additionally, a search for efficient biocontrol agents for management of psocids under certain scenarios where applicable and practical is prudent in order to add another tool for integrated psocid management.

Currently, management of stored product psocids primarily involves chemical and physical treatments (Nayak et al. 2014, Ocran et al. 2021). Biocontrol of psocids could be an option under certain specific scenarios such as disinfestation of pallets, empty commodities

transportation cases, animal feed packaging sites, and empty storage facilities such as storehouses and warehouses. The predatory mites in the family Cheyletidae including *Cheyletus aversor* (Rudendorf)*, Cheyletus eruditus* (Schrank)*, Cheyletus malaccensis* Oudemans, and *Cheyletus trouessarti* (Oudemans) (Trombidiformes: Cheyletidae) occur naturally in storage communities (Fain and Bochkov 2001, Eliopoulos et al. 2003, Lukas et al. 2007), and can be good biocontrol agents for managing psocids. Both *C. eruditus* and *C. malaccensis* are known to effectively prey on mite pests (Pulpán and Verner 1965). *Cheyletus eruditus* (Cheyletin®) has been approved for use against mites of the families Acaridae and Glycyphagidae (Astigmata), mainly *Acarus siro* L. (Sarcoptiformes: Acaridae), *Tyrophagus putrescentiae* (Schrank) (Sarcoptiformes: Acaridae), and *Lepidoglyphus destructor* (Schrank) (Oribatida: Glycyphagidae) (Žd'árková 1998). There is also evidence of *Cheyletus* species attacking non-mite prey. *Cheyletus malaccensis* has been observed to feed on eggs of the moths *Corcyra cephalonica* Stainton (Lepidoptera: Pyralidae) (Nangia et al. 1995) and *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) (Athanassiou and Palyvos 2006) and, on the thrip *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae) (Sengonca et al. 2004*). Cheyletus* spp. have been observed to prey on eggs of the beetles *T. confusum* Herbst (Rizk et al. 1979) and *R. dominica* (Asanov 1980), the flea *Ctenocephalides felis* Bouche (Siphonaptera: Pulicidae) (Williams and Hallas 1988), and all life stages of the psocid *L. decolor* (Kucerova 2004). Besides their broad host range, *C. eruditus* and *C. malaccensis* can naturally penetrate bulk grain, reproduce parthenogenetically, have all developmental stages being predators (do not damage stored products), are cannibalistic (can survive in the absence of prey), and have adopted to a wide range of storage physical conditions (Schöller 2006). These characteristics mean there is good potential of both

predatory mites being effective biocontrol agents against storage pests (Hubert 2006). Nevertheless, considerable knowledge about the ecological interactions of predators including their foraging behaviors under varied physical conditions is a fundamental requirement for the selection of efficient biocontrol agents (Fathipour and Maleknia 2016). To date, there is no published information on the predatory characteristics of either *C. eruditus* or *C. malaccensis* in managing stored product pests in the United States. Considering this, the current studies seek to develop PH_3 resistance monitoring strategies to manage resistance in psocids and to assess predatory behaviors of *C. eruditus* and *C. malaccensis* to gauge their potential as effective biocontrol candidates for psocid management.

Objectives:

Phosphine resistance contributes to the increased importance of psocids as stored-product pests. In addition to misuse and overuse of PH_3, interspecific differences in response to PH_3 treatments may have contributed to the development of genetically based resistance to this fumigant. This is as a result of indiscriminate exposure of different species to similar concentrations of PH_3 over time. Psocid infestations most often involve more than one species and differing ecological requirements and responses to management treatments have been found among species (Nayak 2014). Discriminating dose bioassays has been explored as a technique to monitor the development of PH_3 resistance in major storage insect pests in many countries including the United States (FAO 1975, Champ and Dyte 1976, Lorini et al. 2007, Opit et al. 2012, Nayak et al. 2013, Chen et al. 2015, Gautam et al. 2016, Konemann et

al. 2017, Cato et al. 2017, Afful et al. 2018). This approach measures the levels of tolerance in selected laboratory susceptible species using dose-mortality response data and subsequently compares the levels of tolerance to PH_3 among tested species using lethal concentration ratios (Robertson et al. 2007). So far, there is no published information on discriminating doses of PH_3 for psocid species in the United States. However, such information is required to establish resistance frequencies, that is, detect PH_3 resistance in psocid populations.

Biological control is an eco-friendly pest management tactic that provides an effective alternative or complement to pesticides and is a major tool in IPM programs (Wu et al. 2016, Fathipour and Maleknia 2016). Exploitation of predator-prey interaction through laboratory and greenhouse simulations has aimed at elucidating and forecasting the mechanisms underlying predator-prey behavior to improve the practical predictive potential of natural enemies for biocontrol (Sepulveda and Carrillo 2008, Yao et al. 2014, Patel and Zhang 2017). Generally, this helps to improve agent selection in classical and augmentative biological control and increases the chances of success in control programs. A basic understanding of the foraging behavior of predators and their prey aids predictions about predator function in its ecological niche. Foraging behavior of predators such as functional response, numerical response, mutual interference, or prey switching is usually evaluated for the selection of efficient biocontrol agents (Fathipour and Maleknia 2016). To date, there is no published information on the foraging characteristics of either *C. eruditus* or *C. malaccensis* for managing any stored product pests in the United States.

The objectives of my thesis are:

Objective 1: Establish discriminating doses of phosphine for adults of eight susceptible laboratory strains of psocids (Psocodea: Liposcelididae) at different fumigation exposure periods.

Objective 2: Determine functional responses of the predatory mites *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) to different life stages of *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae)

Objective 3: Determine numerical responses of the predatory mites *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) to different life stages of *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae)

Objective 4: Determine predator-prey dynamics of the predatory mites *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae), and prey *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae) under different thermo-hygrometric parameters

CHAPTER II

LITERATURE REVIEW

Psocoptera: Etymology, Taxonomy, and Description of Psocids

Psocoptera (= Copeognatha, Corrodentia) is derived from the Greek word, *psokhos* which means gnawed or rubbed, and *ptera,* which means wings (Meyer 2005, Ahmedani et al. 2010). It is relatively a small "suborder" of insects commonly called psocids, booklice, barklice, or barkflies that are found in a variety of natural terrestrial ecosystems (Baz 2008, Johnson et al. 2004). Fossil records indicate that psocids first originated in the Permian era, about 295–248 million years ago (Christopher 2002). Phylogenetically, psocids (Psocoptera) are related to parasitic lice (Phthiraptera) despite their greatly differing appearance. Morphological and molecular evidence have shown that parasitic lice, Phthiraptera may have evolved from within the Psocopteran suborder Troctomorpha (Johnson et al. 2004, Yoshizawa and Johnson 2006). Both insect orders were merged, and are now considered as an insect order Psocodea which was also demoted from superorder to order in the taxonomic ranking of Ectognatha (Yoshizawa and Johnson 2006). The three orders, Psocodea, Thysanoptera, and Hemiptera form a monophyletic superorder, Paraneoptera with members having characteristic mouthparts and reflecting diverse feeding habits (Grimaldi and Engel 2005). Among Paraneopterans, Psocoptera (currently Psocodea) is considered the most primitive (Christopher 2002).

More than 5500 psocid species in 41 families have been identified (Clemmons and Taylor 2016) and grouped within 3 suborders: Trogiomorpha, Troctomorpha and Psocomorpha. The genera, *Liposcelis* (Liposcelididae: Troctomorpha), *Lachesilla* (Lachesillidae: Psocomorpha), *Lepinotus* (Trogiidae: Trogiomorpha), and *Trogium* (Trogiidae: Trogiomorpha) have been described as serious stored grain pests (Ahmedani et al. 2010, Liu et al. 2017).

Psocodea are free-living exopterygote or apterygote insects ranging in body length from 0.6–25 mm, although rarely exceeding 10 mm (Baz 2008). Psocids are active, fast running insects with soft bodies, body color varies with species, but many are creamy yellow with brown abdominal bands (Mockford 1971). Psocids have large heads with slightly bulging faces, prominent black compound eyes with no ocelli, long filiform antennae with 12–50 segments which sweep back towards the abdomen, and relatively small thoraces compared to the head (Ahmedani et al. 2010). Legs are slender, and hind legs are longer with prominent femur, which allow for jumping. Abdomen consists of ten segments and the terminal region (telson), formed by the epiproct and paraprocts (Baz 2008). The external genitalia of both sexes are concealed. Adults are about 1–6 mm long, with or without wings (Gillot 1995). Nymphs of psocids are colorless to pale yellow with no abdominal bands, and their size can increase rapidly until they molt into adults (Turner and Bishop 1998). Psocids undergo simple metamorphosis with typically six instars, but this number is reduced to five in *Psyllipsocus* and four in Liposcelidiadae (Sedlacek 1996). Psocid eggs are usually simple, smooth, elongate oval, or cylindrical without a micropyle (Baz 2008). The eggs may be bare or covered with fecal material or a layer of silk webbing and can be laid in groups or singly (Ahmedani et al. 2010).

Ecology, Biology, and Behavior of Psocids

Psocids can survive and thrive in diverse habitats, and they are commonly known as booklice or barklice. Booklice are mostly indoor apterous species, often inhabit old books where they feed on mold and glue used for binding, whereas barklice are outdoor winged species that usually inhabit the bark of trees. Barklice are often considered beneficial scavengers because they consume excess accumulation of fungi, algae, and dead bark on trees. Nevertheless, most Psocopterans have omnivorous feeding habitats, e.g., *L. bostrychophila* and can survive and thrive on a variety of substrates given the favorable conditions (Sedlacek 1996, Athanassiou et al. 2010). The small body size of psocids give them the ability to exploit cracks and crevices and to remain unnoticed inflicting feeding damage on stored grains and seeds (Nayak et al. 2014). *Liposcelis bostrychophila* was reported to prey upon eggs of *P. interpunctella* and eggs of an anobiid, wood-infesting beetle *Hemicoelus gibbicolis* (LeConte) (Coleoptera: Anobiidae) (Lovitt and Soderstrom 1968) while *Liposcelis divinatorius* (Muller) (Psocodea: Liposcelididae) was reported to prey upon eggs of *Sitotroga* sp. (Lepidoptera: Gelechiidae) (Finlayson 1932). Most psocids live inside the grain bulk, within the intergranular microclimate, which they have little difficulty penetrating (Sedlacek 1996). Also, objects such as pallets can serve as both hiding places and a means of transporting psocids species among facilities (Ahmedani et al. 2010). Behavioral adaptations such as the ability to maintain body water content by actively absorbing water from the atmosphere and a surge in population when ambient humidity is above 60% have been reported (Knülle and Spadafora 1969). Large numbers of *L. decolor* have been observed leaving grain bins when substrate moisture content and relative humidity (RH) in bins

drops during the day. This was mostly observed in female psocids that need the moisture for egg production and returned when the RH increased in the morning (Rees 2003). Domestic psocids are generally more tolerant to desiccation than other psocids (Sedlacek 1996). Moreover, standard food-storage facility practices of protection and disinfestation frequently fail to control psocids (Nayak et al. 2020). Psocids are usually not successfully managed by practices that have been developed primarily to control coleopteran and lepidopteran pests, and several psocid species have shown strong resistance to phosphine (PH_3) and other commonly used grain protectants (Nayak 2006). Parthenogenesis (egg development without fertilization) coupled with obligatory thelytoky (all progeny female) allow some species, including *L. bostrychophila, L. obscura*, and *L. reticulatus* population to grow rapidly under favorable conditions (Mockford 1971, Sedlacek 1996, Nayak et al. 2014). A short life cycle, where development from eggs to adults is about 2–3 weeks at 30–35 ºC and 75% RH, along with adult longevity of 72–144 days, and the capability for surviving in adverse conditions without food make some psocids species difficult to manage (Nayak et al. 2014, Clemmons and Taylor 2016).

Psocids are paurometabolous insects, with eggs developing into adults through simple, incomplete metamorphosis involving several nymphal stages that resemble small adults. Generally, psocids can develop from egg to adult at temperatures ranging from 20–42.5 ºC, depending on the species and RH of 70–80% are optimal for growth (Opit and Throne 2008, Ahmedani et al. 2010, Opit et al. 2018, Ocran et al. 2021). In typical household conditions of approximately 20 °C and greater than 60% RH, *L. bostrychophila* can survive as long as two months without food and survival can exceed 100 days when RH is greater than 70% (Turner and Mauderoxby 1988). Psocids are

unlikely to complete development or reproduce at temperatures below 20 °C or above 42.5 °C, and they cannot survive when the RH is consistently below 50–60% (Turner 1994, Wang et al. 2009, Phillips and Throne 2010, Opit et al. 2018, Ocran et al. 2021). Cold temperature below 2°C kills adults but their eggs survive and hatch just when favorable conditions resume (Sedlacek 1996, Ahmedani et al. 2010). Also, viviparity (live birth of nymphs) has been reported in a few psocid species including, *Phallocaecilius* and *Archipsocopsis*. At maturity, females lay 50–100 eggs during their entire life. Nymphs hatch from eggs using a specialized shell breaker and nymphal instars may range from 4–6 to reach adult stage. Typically, the life cycle of psocids is completed in 21 days under favorable conditions (Ahmedani et al. 2010, Nayak et al. 2014, Opit et al. 2018, Ocran et al. 2021).

Worldwide Distribution of Psocids

Originally described based on specimens collected from the bark of trees in Africa in 1931, approximately 5,500 psocids species have been reported worldwide (Clemmons and Taylor 2016). Psocids are believed to be native to tropical and sub-tropical regions in the world. However, recent reports indicate that psocids are cosmopolitan and are continually disseminated all over the world through international trade (Kucerova 2006, Rees 2008, Ahemdani et al. 2010). The pest status of psocids began to change internationally in the late 1980s when the first reports of large infestations rapidly establishing in stored grains emerged from West Africa and Southeast Asia (Pike et al. 1991). Subsequently, heavy infestations were reported in stored grains in Indonesia

(Kleih and Pike 1995, Santoso et al. 1996), India (Rajendran 1994), China (Wang et al. 1998), and Australia (Rees 1998, Nayak 1998). Severe infestations have been reported ever since from such diverse locations as food-processing facilities and feed and flour mills in Italy (Trematerra 1999) and the United States (Arbogast et al. 2000, Opit and Throne 2008), tobacco processing houses in Zimbabwe (Mashaya 1999), museums and bakeries in Czechoslovakia (Obr 1978), and railroad cars used for transporting grain in Canada (Smith 1985). Three genera of psocids, *Liposcelis*, *Lepinotus*, and *Lachesilla* have been reported to have a worldwide distribution (Liu et al. 2017) and, members of the genus *Liposcelis* including *L. bostrychophila*, *L. decolor*, *L. entomophila*, and *L. paeta* are considered the most economically important species with a worldwide distribution (Lienhard and Smithers 2002, Nayak et al. 2014). Severe infestations of *L. bostrychophila* and *L. entomophila* have been reported from humid tropical countries, including Thailand, Malaysia, Indonesia, Singapore, China, India, and Indonesia (Wang et al. 1999). *Liposcelis decolor* were detected in the temperate regions in Australia infesting central and on-farm grain structures whereas *L. bostrychophila* and *L. entomophila* were frequently found in central storage structures and mills (Rees et al. 2003). Also, *L. entomophila* was found in tobacco-processing facilities in Zimbabwe (Mashaya 1999) while *L. bostrychophila* was reported in households in the United Kingdom (Turner 1994). In Canada, Smith (1985) found *L. entomophila* in railway vans used in conveying grains. In addition to these species, *L. brunnea*, *L. obscura*, *L. corrodens*, *L. rufa, L. pearmani*, *L. fusciceps*, and *L. reticulatus* were found infesting stored commodities in the United States (Mockford 1993, Lienhard and Smithers 2002, Opit and Throne 2008). Additionally, *T. pulsatorium* and *L. pedicularia* were

occasionally reported as stored grain pests in countries including the United States (Ahmedani 2010). However, *L. entomophila* and *L. decolor* are the most predominant species infesting stored grains in the United States (Throne et al. 2006, Opit et al. 2009, Gautam et al., 2013, 2016).

Change in Pest Status of Psocids

Psocids have long been detected in stored commodities but were considered only as fungivores, and their importance was either ignored or underestimated (Ruden 2003). Psocids were recognized as secondary colonizers and less important than stored-product coleopterans and lepidopterans pests (Nayak et al. 2014, Athanassiou and Rumbos 2018). However, over the last two-to-three decades psocids have risen to prominence as serious pests of stored products and are now considered as the most important emerging taxonomic group of pests that pose a new threat to global food safety and security (Nayak et al. 2014, Stejskal et al. 2015, Wei et al. 2020). One key reason why the pest status of psocids has risen to prominence is that they are not usually successfully controlled by strategies such as insecticides that are used effectively against coleopteran and lepidopteran pests (Nayak et al. 2014, Athanassiou and Rumbos 2018, Diaz-Montano et al. 2018). For example, their ability to delay egg hatching in a PH_3 -rich environment is a possible cause of the rise in control failures in countries in south and southeast Asia after a switch from contact insecticides to PH_3 (Nayak et al. 2003). Psocid populations recover more rapidly from poorly conducted PH_3 fumigations than beetle pests (Roesli et al. 1998). Guedes et al. (2008b) observed behavioral resistance mechanism of psocids to PH_3

where psocids disperse from grain facilities to absorb moisture during fumigation which probably reduces the efficacy of fumigants against psocids in open top silos. In bulk grains, psocids have been shown to benefit from the decrease of predation and competition especially after insecticide applications (Pranata et al. 1983). The upsurge in pest status of psocids is partly due to the quantitative losses caused by feeding on the endosperm and germ of grains leading to significant grain weight loss and germination failure (Kucerova 2002, Gautam et al. 2013, Athanassiou 2014). Psocids have evolved the ability to thrive on and deteriorate the quality of a variety of substrates, including stored commodities because of their presence and the resulting exuviae, feces, and cadavers (Opit and Throne 2008, Athanassiou et al. 2014, Athanassiou and Rumbos 2018). In addition, the increasing consumer concerns about produce quality may lead to rejection of psocid-infested commodities that are destined for export thereby causing economic losses. In countries such as Australia, psocid-infested grain is banned for export (Nayak 2010). Moreover, psocid natural ecology which includes short generation times and parthenogenetic reproduction found in some species allows psocids to rapidly colonize new habitats and to quickly proliferate given favorable conditions (Opit and Throne 2008, Athanassiou et al. 2014). Psocids are also reported to be associated with human and animal health problems because they distribute molds and vector disease pathogens (Barker 2007, Chin et al. 2010).

Economic Impact of Psocid Infestations

Psocids have become global pests of stored commodities due to considerable economic losses they inflict (Diaz-Montano et al. 2018). Psocids infest stored products, such as cereal grains and their processed products, other non-cereal-based foods, books,

records, and biological specimens; at high population densities they cause serious quantitative and qualitative losses (Kucerova 2002). Psocids can cause contamination of stored grain and grain-based commodities, and can also significantly reduce the weight and quality of stored grain (Athanassiou et al. 2010, Gautam et al. 2013). Athanassiou et al. (2014) reported that psocids can develop and persist better on mixtures of whole and cracked kernels for an extended period of time. Kucerova (1999) also found that psocids appear to have a preference for germ but will also feed on the soft endosperm of damaged or cracked kernels. However, it was recently established that psocids are able to colonize sound kernel of different commodities and may cause weight losses that can exceed 10% and 6–54% germ damage (Kucerova 1999, Stejskal et al. 2006, Athanassiou et al. 2010, Gautam et al. 2013). *Liposcelis bostrychophila* was reported to cause weight losses of up to 4–5% by feeding on rice after 6 months of storage (McFarlane 1982). Kucerova (2002) also reported 9.7% weight loss in cracked wheat kernels after 3 months of infestation by *L. bostrychophila*. Pike (1994) also reported 2.9% weight loss in lightly milled rice after 3.5 months of *L. paeta* infestation. Moreover, *L. entomophila* caused 8.5% weight loss by feeding on damaged wheat kernels for 90 days (Gautam et al. 2013). Apart from reducing the weight of stored products, psocids are associated with the presence of fungi that may cause discoloration of grain due to mold growth (Athanassiou and Rumbos 2018). Ahmedani et al. (2010) reported increase in temperature and RH in grain storage systems with large psocid infestations, which may eventually lead to grain decay. Moreover, qualitative loss due to their presence and the resulting exuviae, feces, and cadavers in stored commodities has been reported (Athanassiou et al. 2014, Athanassiou and Rumbos 2018). Gautam et al. (2013) reported that *L. paeta* can cause up to 40% germination loss

of infested intact kernels after 45 days of exposure. International trade restrictions due to presence of psocids in commodities for export have been reported (Ahmedani et al. 2010, Nayak et al. 2014).

Health Concerns and Safety Hazards Associated with Psocids

Psocids, like other insects, have been associated with instances of dust allergy in humans, (Rijckaert et al. 1981). Psocids were present in virtually all dust samples collected from houses in the Netherlands (Spieksma and Smits 1975). Association of psocids with dust allergy was later confirmed by Turner et al. (1996) who carried out investigations on antibodies produced in response to the known dominant antigen from *L. bostrychophila* and concluded that at least 5% of allergy patients showed strong positive skin reactions to the psocid antigen. Also, *L. bostrychophila* was reported to transmit onychomycosis in humans after nail attack (Lin et al. 2004). Psocids may serve as intermediate hosts by eating tapeworm eggs, harboring their larvae in their gut and eventually disseminating these parasites in the surrounding storage community (Turner 1994). Psocids may vector disease pathogens by ingesting bacteria, fungal spores or viral pathogens, nourishing them in their gut, and then disseminating these pathogens through their excreta or their physical presence in infested food commodities (Obr 1978, Turner 1996). According to Turner (1994), psocid presence in food commodities has been considered as a psychological threat to human health. Severe psocid infestations can spread to storage structures, machinery, and walkways; sometimes populations are so high that they appear like a moving carpet of brown "dust" on floors and other surfaces,

therefore, becoming a safety and/or biological hazard (Ahmedani et al. 2010). Pallets in the food industry not only serve as hiding and harboring places for psocids but also serve as a means of transport facilitating spread. A countrywide survey conducted in the UK revealed that 75% of pallets harbored psocids (Lilley 1981) where they pose a serious threat to the hygiene of food products directly and industry profit indirectly.

Management of Psocids

Physical control strategies for psocid management

Psocid ecological studies over the past few decades primarily focused on predicting favorable environmental conditions required for optimal population growth and development (Opit and Throne 2008, Gautam et al. 2010, Opit et al. 2018). For example, *L. brunnea, L. fusciceps, L. pearmani, L. rufa*, *L. reticulatus*, and *L. obscura* were reported to survive at all temperatures within the 22.5–42.5 ºC range at 75% RH. Also, optimal population growth conditions for *L. reticulatus, L. brunnea, L. rufa, L. fusciceps*, *L. pearmani*, and *L.obscura* were: 32.5 °C and 75% RH, 32.5 °C and 63% RH, 35 °C and 75% RH, 30 ºC and 75%, 32.5 °C and 75% RH, and 40 °C and 75% RH, respectively (Opit and Throne 2008, Gautam et al. 2010, Aminatou et al. 2011, Gautam et al. 2015, Opit et al. 2018). Highest intrinsic rates of increase for most species including *L. badia, L. bostrychophila*, *L. decolor*, *L. paeta*, and *L. tricolor* occurred at 27.5–30 °C (Wang et al. 2000, Dong et al. 2007, Jiang et al. 2008, Tang et al. 2008, Wang et al. 2009). Physical control measures for management of storage insect pests are an ecological-based approach that mainly exploits extreme temperatures and RHs beyond

the optimal conditions required for growth and development by insects. Back (1939) observed that applying dry heat with temperatures of 50–60 °C controlled *L. bostrychophila* populations within 1 h. Adults of *L. bostrychophila*, *L. decolor*, and *L. paeta* were more susceptible to heat treatment when subjected to elevated temperatures in the range of 45–55 °C than their egg stages (Beckett and Morton 2003). Turner (1988) suggested that freezing rapidly kills all stages of *L. bostrychophila*, and limited disinfestation can be achieved by subjecting commodities to sub-zero temperatures for a few days. Arthur et al. (2017) also reported that cold treatment can be used to manage all life stages of *L. bostrychophila*, *L. entomophila*, *L. paeta*, and *L. decolor* at -18 ºC for 24 hours. Similarly, dehumidification (low RH) has been used as a physical control tool to manage psocids (Opit and Throne 2009, Gautam et al. 2010, Aminatou et al. 2011, Gautam et al. 2015, Ocran et al. 2021). Psocids multiply in environments with RH above 63%, but do not survive below consistent 55% RH (Opit and Throne 2008, Opit and Throne 2009, Gautam et al. 2010, Gautam et al. 2015). Recently, Ocran et al. (2021) found that different species and life stages of *L. entomophila*, *L. decolor*, *L. bostrychophila*, and *L. paeta* responded differently to 43 and 50% RH, as maximum survivorship times under these dehumidification conditions were estimated as 8–12 and 10–16 days, respectively.

Chemical control and insecticide resistance

Contact insecticides

Chemical control is the strategy of choice among other management tactics against psocids in stored products, but the list of pesticides is limited because of the strict safety requirement imposed on use of synthetic insecticides on or near food (Ahmedani et al. 2010). Also, continuous use of insecticides to control stored-grain pests has resulted in insecticide tolerance or resistance (Pacheco et al. 1990, Nayak et al. 2014). Another problem associated with chemical control is that the efficacy of some insecticides varies greatly in multi-species infestations (Suchita et al. 1989, Pinto et al. 1997). Recent studies have confirmed that psocids are tolerant to many grain protectants used to successfully manage beetles and moths; this is not due to previous exposure, it is a natural phenomenon (Collins et al. 2001, Daglish et al. 2003, Nayak et al. 2014, Athanassiou and Rumbos 2018). For example, *L. entomophila*, *L. bostrychophila*, and *L. paeta* were found to be tolerant to the pyrethroid bioresmethrin + piperonyl butoxide (Nayak et al. 1998). In similar studies, piperonyl butoxide + chlorpyrifos-methyl did not control *L. entomophila* (Daglish et al. 2003). Deltamethrin was not able to provide long-term protection against *L. bostrychophila*, *L. paeta*, and *L. entomophila* (Nayak et al. 2002a, b). Regarding resistance to insecticides, psocids have shown significant resistance to organophosphates and pyrethroids due to mixed-function oxidases, which presumably play a significant role in the detoxification of insecticides (Ahmedani et al. 2010). Also, the efficacies of azamethiphos, fenitrothion, chlorpyrifos-methyl, and pirimiphos-methyl were tested as surface treatments on porous and nonporous surface panels against adults of *L. bostrychophila*, *L. entomophila*, and *L. paeta* and the results showed that *L. bostrychophila* was the most susceptible species to the organophosphates tested, however, none of the four organophosphates provided long-term protection against *L.*

paeta and *L. entomophila* (Collins et al. 2001). Pyrethroids were less effective against *Liposcelis* species (Guedes et al. 2008a). Insect growth regulators (IGRs) including fenoxycarb seem to be effective against *L. bostrychophila* when applied to their diet, however, pyriproxyfen, applied as a surface treatment on concrete was not effective against *L. bostrychophila*, *L. decolor*, and *L. paeta* (Bucci 1994, Nayak et al. 1998, Athanassiou et al. 2010). Chlorfenapyr, which belongs to a group of microbial-produced compounds called halogenated pyrroles, is effective as a surface treatment against *Liposcelis* (Guedes et al. 2008b). Guedes et al. (2008a) reported that spinosad, a bacteria derived bio-pesticide also seems ineffective in managing psocids but, a mixture of spinosad and natural pyrethrum provide up to 3 months of protection against infestations of *L. bostrychophila*, *L. decolor*, *L. entomophila*, and *L. paeta.* The admixture comprising spinosad 1 mg kg-1 with chlorpyrifos-methyl 10 mg kg-1 can control all the four *Liposcelis* species, but the high application rate of 10 mg kg-1 of chlorpyrifos methyl may restrict its use to seed treatments only (Nayak and Daglish 2007). Synergistic effect of controlled atmosphere (35% CO_2 and 1% O_2) and organophosphate insecticides (for example dichlorvos) slows the development of resistance by psocids, and also provides a significant increase in mortality compared with using these methods separately (Wei et al. 2002). Nevertheless, psocids are capable of developing resistance to controlled atmosphere (Wang et al.1999). The use of diatomaceous earth (DE) is another way of controlling stored-product insect pests, and it is effective when used in systems with lower RH (Korunic et al. 1996). Diatomaceous earth, when used alone, does not reduce progeny production of *L. entomophila*, *L. reticulatus*, and *L. decolor* (Athanassiou et al. 2009).

Fumigants

Fumigation has been used routinely over the past several decades as a primary component in the management of stored grain pests (Afful et al. 2018, Nayak et al. 2020). Over reliance on fumigants to meet domestic and international market demand for high-quality grain, free of insects is a routine practice globally (Chaudhry 2000, Cao et al. 2003, Nayak et al. 2012, Opit et al. 2012). The use of PH_3 fumigant has increased markedly over the last 2–3 decades mostly due to market concerns over protectant chemical residues and resistance in target pests to contact insecticides (Emery et al. 2011, Nayak et al. 2015). The first global survey of PH_3 resistance was conducted during the 1970s by Champ and Dyte (1976) who reported the occurrence of PH_3 resistance in several key stored grain pest species in different parts of the world. Several studies have revealed the existence of psocid populations with strong resistance (Cao et al. 2003, Athanassiou 2010, Nayak et al. 2014). According to Athanassiou and Rumbos (2018), PH_3 resistance contributes to the increased importance of psocids as stored-product pests. Nayak et al. (2000) found that resistant adults of *L. bostrychophila* were able to survive PH_3 concentrations approximately 5x greater than that which kills the susceptible strain, whereas resistant strains of *L. decolor* and *L. entomophila* tolerated concentrations of PH_3 approximately 6x and 7x higher than those that kill the susceptible strains, respectively. Similarly, eggs of resistant *L. bostrychophila* survived a PH_3 concentration of 2 g/m^3 (1438 ppm) whereas 0.031 g/m^3 (22 ppm) was effective against the susceptible strain; the concentration required to kill the resistant population was 65x higher than that required to kill the susceptible strain. Cao et al. (2003) and Pike (1994) found strains of *L. entomophila* with strong resistance in China and Indonesia, respectively. Pinniger (1985)

reported that the eggs of *L. bostrychophila* are tolerant to phosphine and that the required doses for controlling psocids were similar to those of stored-product moths. Eggs of psocids were found to be much more tolerant than the adults. Pinniger (1985) noticed PH_3 tolerance in eggs of *L. bostrychophila* compared with the adult stage and reported lethal doses (LD_{99}) of 0.5 mg/L (360 ppm) and 0.02 mg/L (14 ppm), respectively, after 24 hours of exposure. Nayak et al. (2003) also reported that PH_3 delays the development of *L. bostrychophila* eggs and suggested that this delay was a mechanism of resistance to PH_3. Pike (1994) found that PH_3 concentration as high as 1.7 mg/L (1223 ppm) over 5 days was required to control eggs of *L. entomophila*. Also, 2 mg/L (1438 ppm) concentration of PH_3 over 6 days was needed to control eggs of *L. bostrychophila* (Nayak et al 2002 b). These rates were higher than the recommended label rates for controlling resistant beetle pests in Australia and Indonesia and subsequently, it was established that the most effective strategy to control resistant psocids is by applying relatively low concentrations of PH_3 for extended exposure periods, for example 0.05 mg/L (36 ppm) for 16 days, which allows all eggs to hatch and develop to the much less tolerant nymphal stage (Nayak 2005). Again, it was observed that at fixed concentration, the efficacy of PH_3 increases with rise in temperature from 20–35 °C and under humidity regimes of 55–70% (Nayak et al. 2014). Nayak and Collins (2008) reported that at constant PH_3 concentration, high temperature and low RH provided the shortest effective fumigation period to control resistant strains of *L. bostrychophila*.

With respect to the other fumigants like methyl bromide (MeBr), Pike (1994) tested a strain of *L. entomophila*, which exhibited strong resistance to PH_3 and found that all life stages of the tested species were effectively controlled at 50 mg/L of MeBr over a

4-h exposure period. At 1.3 mg/L, eggs of a field strain of *L. bostrychophila* were controlled with a 24-h exposure of methyl bromide (Rajendran 1994). Athanassiou et al. (2015) found that MeBr was more effective in controlling all life stages of *L. bostrychophila*, *L. decolor*, *L. entomophila*, and *L. paeta* compared with sulfuryl fluoride (SF), however, MeBr has phased out of use worldwide. Sulfuryl fluoride is a potential replacement for MeBr or PH_3 (Nayak et al. 2014). Athanassiou et al. (2012) showed that dosages well below the maximum recommended dose of 31.25 g/m^3 for 48 h controlled adults and nymphs of all major pest species except *L. decolor*. The eggs of *L. decolor* were completely controlled with 72 g/m^3 whereas complete mortality of *L. paeta* was not achieved using 96 g/m^3 for 48 h, dosages that are 2x and 3x the label rate, respectively (Nayak et al. 2014).

Phosphine resistance challenges

Leaky storage facilities and the practice of overdosing to compensate for the lack of airtight storage structures lead to fumigation failures because of under-dosing and facilitate resistance development. This results in higher frequency of applications leading to a higher selection pressure for PH_3 resistance. Consequently, over time, presence of resistant pest insects combined with the selection pressure results in higher resistance frequencies in pest populations and ultimately loss of efficacy of PH_3 against resistant populations (Benhalima et al. 2004). In addition, the occurrence of multi-species infestations which is frequently observed in psocid infestations in stored product communities have been documented to undermine the efficacy of PH_3 treatments (Rees

1998, Athanassiou et al. 2010). According to Nayak et al. (2014) interspecific differences in response to PH_3 may have contributed to the development of genetically based resistance due to indiscriminate exposure of different species to similar concentrations of PH_3 in the same habitat over an extended period of time. More recent work has confirmed high frequencies of PH_3 resistance in key pest species in India (Kaur et al. 2015), Australia (Nayak et al. 2013), Brazil (Lorini et al. 2007), and the United States (Opit et al. 2012, Chen et al. 2015, Konemann et al. 2017, Afful et al. 2018).

In the late 1980s and 1990s, Zettler and others reported PH_3 resistance in the United States in *T. castaneum*, *Cadra cautella* (Walker) (Lepidoptera: Pyralidae), and *P. interpunctella* from peanut storage facilities (Zettler et al. 1989); in *T. castaneum* and *R. dominica* from wheat stored in farms (Zettler and Cuperus 1990); in *T. castaneum* and *T. confusum* from flour mills (Zettler, 1991); and in *Lasioderma serricorne* (F.) (Coleoptera: Anobiidae) from tobacco storage facilities (Zettler and Keever 1994). Twenty years later, substantial increase in PH_3 resistance was reported in *R. dominica* and *T. castaneum* populations in Oklahoma (Opit et al. 2012). Recently, PH_3 resistance has also been reported in *C. ferrugineus* in Oklahoma (Konemann et al. 2014, 2015). The levels of resistance of the most resistant strains of *R. dominica*, *T. castaneum*, and *C. ferrugineus* in these studies were about 1519x, 119x, and 134x, respectively, more resistant than their susceptible counterparts (Opit et al. 2012, Konemann et al. 2017). Recent studies have gone beyond simply documenting the presence or absence of PH_3 resistance in a pest population. They have revealed additional information on the occurrence of two general phenotypes for either 'strong' PH_3 resistance in individuals that can tolerate extremely high concentrations of phosphine and also a 'weak' resistance phenotype for insects that

could be killed at PH_3 concentrations just a few fold greater than concentrations needed to kill susceptible insects (Lorini et al. 2007, Opit et al. 2012, Nayak et al. 2013). Significant progress has also been made in bioassay methods that can classify PH_3 resistance as either weak or strong phenotypes in some species (Nayak et al. 2013). The genetic locus responsible for strong PH_3 resistance, a phenotype that requires resistance alleles for weak resistance to be fixed at another locus, was recently reported by Schlipalius et al. (2012) and that information then allowed use of molecular diagnostics to enable researchers to determine the presence of strongly-resistant genes occurring in populations with resistant insects (Chen et al. 2015).

Phosphine resistance bioassay: Diagnostic methods

Phosphine resistance monitoring assays most often involved the use of standardized Food and Agriculture Organization of the United Nations (FAO) discriminating doses technique (FAO 1975). Discriminating doses (DDs) bioassay has been accepted over the decades as a technique for monitoring and managing development of PH_3 resistance in major storage insect pests in many parts of the world including the United States (FAO 1975, Champ and Dyte 1976, Lorini et al. 2007, Opit et al. 2012, Nayak et al. 2013, Chen et al. 2015, Gautam et al. 2016, Konemann et al. 2017, Cato et al. 2017, Afful et al. 2018). This approach measures the levels of tolerance in laboratory susceptible strains using dose-mortality response data and subsequently, comparing the levels of tolerance among tested species using lethal concentration ratios (Robertson et al. 2007). A discriminating dose for tested species makes it possible to detect the presence of

PH_3 resistance in samples of related insects and permits the estimation of resistance levels and frequencies (Konemann et al. 2017). Two DDs are used; a lower DD which discriminates between susceptible and resistant insects and a higher DD which is designed to detect resistances higher than the common 'weak' resistance (Daglish and Collins 1999). Insects believed to be homozygous for PH_3 susceptibility are used to determine the lower DD, while strains homozygous for weak resistance are used to determine the upper DD. Also, the 'rapid knockdown test', originally developed by Reichmuth (1991) and Bell et al. (1994) is used to give a quick yes-no answer with field-collected insects. The diagnostic test allows determination of whether a tested population sample is resistant or not in order for immediate management action (control, eradication, and quarantine) to be taken where appropriate. The drawback with this method is that it is difficult to determine the strength of resistance in some species (Daglish and Collins 1999).

A third method is the flow-through technique that exposes mixed-age cultures of insects to a continuous flow of phosphine at a constant concentration (Winks and Hyne 1997, Daglish et al. 2002). This method is very laborious and lengthy, but it gives an accurate prediction of the time required for complete extinction of an insect population at a nominated PH_3 concentration (Daglish and Collins 1999). It is used to characterize the resistance and predict concentrations and exposure periods needed to manage insects in the field. The fourth diagnostic method is by the use of the molecular-based technique. The identification of the strong resistance gene, *rph2*, made the development of diagnostic molecular assays possible (Kaur et al. 2013, Chen et al. 2015, Kaur et al. 2015, Kocak et al. 2015, Schlipalius et al. 2018). Strong resistance from *rph2* requires the

simultaneous presence of homozygous resistance variants of both *rph1* and *rph2* genes, but in developing the molecular resistance assay, the *rph1* is ignored and we assumed that resistance variants of *rph1* were always present. This assumption was made because resistance variants of *rph1* were already very common throughout Australia and the world at the time that the *rph2* gene was identified (Schlipalius et al. 2002). The enzyme encoded by the *rph2* gene is critical to aerobic energy metabolism (Schlipalius 2012) that only seven changes to the amino acid sequence of dihydrolipoamide dehydrogenase (DLD) (Kaur et al. 2015) have been found that can confer resistance to PH_3 while preserving function of the enzyme sufficiently to allow the insects to survive in the field. The strategy developed for screening large numbers of field-collected insects for all resistance variants relies on polymerase chain reaction (PCR) amplification of each exon of the *rph2* gene using tagged primers (Nayak et al. 2020). The amplified DNA from all of the insects is then pooled prior to identification of resistance variants using high-throughput DNA sequencing. The tags on the primers allow each sequence variant to be attributed to the insect from which it originated for thousands of insects at one time (Schlipalius et al. 2018).

Biological control of storage arthropod pests

Biological control is an eco-friendly pest management tactic that provides a feasible alternative or complement to pesticides in the food industry and stored grain IPM programs (Flinn and Hagstrum 1996, Wu et al. 2016, Fathipour and Maleknia 2016). Insect and acarine pests of stored products interact with numerous natural enemies

including predatory insects and mites, hymenopterous parasitoids, other invertebrates, and pathogenic microorganisms (Eliopoulos et al. 2003). In the United States, bioagents such as *Anisopteromalus calandrae (Howard) (Hymenoptera: Pteromalidae) , Bracon hebetor Say (Hymenoptera: Braconidae), Cephalonomia waterstoni (Gahan) (Hymenoptera: Bethylidae), Dibrachys sp. (Hymenoptera: Pteromalidae) Dufuriellus ater (Dufour) (Hemiptera: Antocoridae), Habrocytus thyridopterigis Howard (Hymenoptera: Pteromalidae), Holepyris sp. (Hymenoptera: Bethylidae), Laelius sp. Ashmead (Hymenoptera: Bethylidae), Lariophagus distinguendus (Forster) (Hymenoptera: Pteromalidae), Mesostenus sp. Gravenhorst (Hymenoptera: Ichneumonidae), Pteromalus sp. Swederus (Hymenoptera: Pteromalidae), Theocolax elegans (Westwood) (Hymenoptera: Pteromalidae), Trichogramma evanescens Westwood (Hymenoptera: Trichogrammatidae), Venturia canescens (Gravenhorst) (Hymenoptera: Ichneumonidae)*, and *Xylocoris flavipes (Reuter) (Hemiptera: Anthocoridae) have been approved for use against stored-product insect pests (*Hagstrum and Subramanyam 2006).

Some spiders, daddy long legs, and several types of small birds are known natural predators of nymphs and adult stages of psocids (Ahmedani et. al. 2010). Also, some hymenopterans, such as mymarid wasps (*Alaptus*) and braconid wasps (*Euphoriella*) have been found to parasitize egg and nymphal stages of psocids (Mockford 1993). Entomopathogenic fungi including *Beauveria bassiana* (Bals.-Criv.) Vuill (Hypocreales: Cordycipitaceae)*, Aspergillus parasiticus* Speare (Eurotiomycetes: Trichocomaceae)*, Isaria fumosoroseus* Wize (Hypocreales: Cordycipitaceae), or *Metarhizium anisopliae* (Metchnikoff) (Hypocreales: Clavicipitaceae) are commercially available and have been

reported to cause about 16% mortality of *L. bostrychophila* (Lord and Howard 2004). Zoophagous acarids are specialist, selective, or generalist predatory mites that kill and utilize living arthropod hosts to complete their developmental and reproductive processes, and represent a broad group of bioagents, including the predatory mites that prey on all life stages of insects and other phytophagous mites for their physiological functions and survival (Rosomer and Stoffolano 1997, Castañé et al. 2011).

There are numerous predatory mites mostly in the family Phytoseiidae, including *Phytoseiulus persimilis* Athias-Henriot, *Neoseiulus californicus* (McGregor), *Amblyseius swirskii* (Athias-Henriot), and *Iphiseius degenerans* (Berlese) (Mesostigmata: Phytoseiidae) that are commercially available and have successfully been used to manage arthropod pests in glasshouse, orchard, or field IPM systems (Escudero and Ferragut 2005, McMurtry et al 2013, Nguyen et al. 2015). In contrast, the use of mite predators to manage pests in storage is highly limited (Lukas et al. 2007). Haines (1998) reviewed the past 30 years of stored-product pest management research and reported that arthropod natural enemies, mostly mite predators in stored products are overlooked and underexploited. The only commercialized predatory mite approved for use in storage communities against stored-product pests is *Cheyletus eruditus* (Schrank) (Trombidiformes: Cheyletidae) which has been identified and tested in practice against mites of the families Acaridae and Glycyphagidae (Astigmata), mainly *Acarus siro* L. (Sarcoptiformes: Acaridae), *Tyrophagus putrescentiae* (Schrank) (Sarcoptiformes: Acaridae), and *Lepidoglyphus destructor* (Schrank) (Oribatida: Glycyphagidae) (Žd'árková 1998). However, during the past two decades, as biological control has become more important in stored-product IPM research programs, predatory mites have

received increasing attention, particularly, in the field of acarology and entomology, due to their efficiency as bioagents under laboratory simulated conditions with reported success of 70–100% (Eliopoulos et al. 2003, Hagstrum and Subramanyam 2006).

Cheyletid predatory mites in storage ecosystems

Cheyletidae is one of the central families of trombidiform mites and comprises about 150 species (Volgin 1987). Cheyletid mites are mainly free-living predators that feed on various micro-arthropods, mostly on herbivorous, fungivorous, and saprophagous acaroid mites (Žd'árková 1979, Mullen and OConnor 2019). Cheyletids penetrate insects and mites with their chelicerae and immediately inject saliva that paralyzes the prey (Yoshikawa 1985). Currently, 29 species of the genus *Cheyletus* have been characterized and all these species are found either in vertebrate nests or in grain stores and are considered to be acarophagous predators (Cebolla et al 2009). *Cheyletus aversor* Rohdendorf, *Cheyletus eruditus*, *Cheyletus hendersoni* Baker, *Cheyletus malaccensis*, *Cheyletus trouessarti* Oudemans, and *Cheyletus trux* Rohdendorf (Trombidiformes: Cheyletidae) were reported from stored products (Žd'árková 1979, Eliopoulos et al. 2003, Lukas et al. 2007, Valbuza et al.2020) and are often found in granaries, warehouses, and barns (Hughes 1976). *Cheyletus malaccensis* and *Cheyletus eruditus* are the most common predatory mite species in grain stores (van Hage-Hamsten and Johansson 1992). The two cheyletid species are widely distributed in storages, often in large numbers. They feed on various pest phytophagous mites such *as A. siro*, small insects such as book lice (*Liposcelis*), and stored grain insect eggs and larvae (Kucerova 2004, Palyvos et al. 2008,

Cebolla et al. 2009, Silva et al. 2013), and are considered as potential natural enemies for biocontrol of storage arthropod pests (Lukas et al. 2007, Horn et al. 2018, Valbuza et al. 2020).

Cheyletus malaccensis is the dominant cheyletid in tropical stores and the second most frequently occurring cheyletid in former Czechoslovakia (Žd'árková 1979, Horn et al. 2018); it is also the most common predatory mite in stored products in Greece (Palyvos and Emmanouel 2009, Palyvos et al. 2008). In Southern Europe (Athanassiou et al. 2002), Asia (Sun et al 2020), and South America (Valbuza et al 2020), *C. malaccensis* was reported as a dominant predatory mite species in stored commodities. *Cheyletus malaccensis* is a generalist predator that preys on stored product pests, such as *A. siro*, *Aleuroglyphus ovatus* Troupeau (Sarcoptiformes: Acaridae), *Caloglyphus redickorzevi* Zachvatkin (Sarcoptiformes: Acaridae), *L. destructor*, *T. putrescentiae*, and *Caloglyphus rodriguezi* Samsinak (Sarcoptiformes: Acaridae) (Cebolla et al. 2009, Al-Shammery 2014, Zhu et al. 2019). It is also known to prey on non-mites, especially, eggs of *Ephestia cautella* (Walker) (Lepidoptera: Pyralidae), *C. cephalonica*, and *T. confusum*, and it attacks young larvae or nymphs of other insects (Yousef et al. 1982, Nangia et al. 1994, Horn et al. 2018). *Cheyletus malaccensis* is associated with both bulk grain and grain residues (Hubert et al. 2007). The natural ability of *C. malaccensis* to penetrate bulk grain indicates the potential of this species for biological control of pest mites (Hubert et al. 2007). Also, the high fertility of *C. malaccensis*, its ability to multiply fast, tolerance to a wide temperature range, and the fact that it is parthenogenetic indicates it has beneficial characteristics that facilitate its production for biological control (Cebolla et al. 2009, Hubert et al. 2016, Zhu et al. 2019).

Lukas et al. (2007) obtained grain samples from 147 geographically isolated grain stores in the Czech Republic (Central Europe) and reported that microarthropod pests in grain stores are associated with four species of predatory mites of the genus *Cheyletus*, and these are *C. eruditus*, *C. malaccensis*, *C. aversor*, and *C. trouessarti* of which *C. eruditus* was the most abundant and frequently encountered species. A similar natural composition of predatory mites has been documented from various regions of Europe (Franz et al. 1997, Stejskal et al. 2003) and North America (Mullen and OConnor 2019). The predatory mite *Cheyletus eruditus* is more common in grain residues, while *Cheyletus malaccensis* is mostly found in bulk grain. Although both predators feed on pest mites and non-acarid pests (Hughes 1976), only *C. eruditus* is used as a biological control agent (Cheyletin ®) (Ž d'árková 1990, 1998). *Cheyletus* spp. have high potential to effectively control storage microarthropod pests (Hagen et al. 1999), however, this control tactics can also fail. The success of these bioagents depends on several factors including predator to prey ratio, temperature, humidity, the required duration of the control period, and grain moisture content (Pulpán and Verner 1965, Pekar and Ž d'árková 2004, Lukas et al. 2007). Solomon (1969) found that control was unsuccessful when the grain was stored at low temperatures. Žd'árková and Horák (1999) reported that the control fails when the temperature is lower than 15 °C, and when prey density is too high—more than 1000 individuals of *A. siro* per kg of grain (Žd'árková 1986, 1998); with such a threshold, *Cheyletus* was unable to contend with the fast population increase of *A. siro*. Nevertheless, there are no remarkable differences in life-history parameters, prey specificity or nutritional requirements among *Cheyletus* spp. and thus, one can

expect similar bio-control characteristics (Wharton and Arlian 1972, Barker 1991, Thind and Ford 2006, Pekar and Hubert 2008).

Ecology, biology, and behavior of *Cheyletus eruditus* and *Cheyletus malaccensis*

Cheyletus eruditus are white to pale yellow in color. The female is about 0.5 mm long, but males are slightly shorter. *Cheyletus eruditus* have five developmental stages: egg, larvae, protonymph, deutonymph, and adult. Like other species of the family Cheyletidae, the mouthparts are large, prominent, and join the body at its anterior limit and have two large pedipalps in front of the body. The movable digit of each of the two chelicerae is needle-like and used to pierce the prey's exoskeleton. Predatory cheyletids are thought to inject a paralyzing saliva immediately after piercing. The body fluids of the immobilized prey can then be sucked out. It can reproduce parthenogenetically, that is without males. Females lay their eggs in clusters and guard them until the young hatch. All developmental stages are predators and thus do not damage stored products (Schöller and Žd'árková 2006). All active life stages of *C. eruditus* are cannibalistic in the absence of other prey. Clutches of eggs often include a few that have collapsed, their contents having been eaten by the mother and/or her newborn daughters. Starvation decreases egg-laying and initiates a nomadic period (Solomon 1969, Hughes 1976; Yoshikawa 1987, Kucerova 2004). The predator is resistant to low temperature and does not develop at temperatures below 12 °C. It is also resistant to organophosphate insecticides (Schöller and Žd'árková 2006). *Cheyletus eruditus* prefers slowly moving prey species to fast-moving ones and has a voracious appetite. The predatory mites can complete their life cycle at temperatures and humidities ranging from 12–35 °C and 60–90% RH,

respectively (Schöller and Žd'árková 2006). The higher the temperature and humidity, the faster their development. Their life cycle lasts from 18–164 days, depending on temperature (Žd'árková and Horák 1999). Žd'árková and Pulpán (1973) tested the survival of these predators at low temperatures ranging from -1.7–2.0 °C and relative humidities of 80–90%. The best results were obtained at 2.0 °C. About 50% of the population of the predatory mites survived for 62 days, and 8% for 200 days, without losing their ability to reproduce when transferred back to favorable conditions (Schöller and Žd'árková 2006). The developmental stage of the prey does not affect the development of *C. eruditus*. Fifty larvae of *C. eruditus* were fed either eggs, larvae, protonymphs, tritonymphs, or adults of *A. siro* from hatching to the adult stage. The type of food did not affect the length of development and individual stages of the mites preferred prey sizes similar to their own size (Schöller and Žd'árková 2006). All stages of mites preferred to consume larvae, while adults were consumed least frequently. The eggs were also eaten despite the fact that the predators preferred moving prey. Females that fed on adults and tritonymphs laid the most eggs, whereas females that fed on prey eggs did not lay any eggs at all. Females that fed on larvae laid eggs approximately 3.7x lesser than when adult or tritonymphs stage of *A. siro* was consumed (Žd'árková and Horák 2000).

Cheyletus malaccensis is translucent white to pale creamy with similar body size and behavioral characteristics as *C. eruditus* and have five developmental stages: egg, larvae, protonymph, deutonymph, and adult, however, the deutonymph is absent in males (Liu 2018). *Cheyletus malaccensis* also exhibits cannibalism and Arrhenotokous parthenogenesis (haplodiploidy) where virgin females always produce males, whereas

fertilized females give rise to offspring of both sexes (Palyvos and Emmanouel 2009). The idea temperature range for reproduction and growth of *C. malaccensis* was 24–28 ºC (Sun et al. 2020). The optimum temperature for the growth and development of *C. malaccensis* was 28 ºC. Populations could increase rapidly with the highest net reproductive rate (R^0 = 290.25) and highest fecundity (544.52) occurring at 28 ºC (Sun et al. 2020). Toldi et al. (2017) found that fecundity was highest at 25 ºC with 415.62 ± 24.78 eggs/female, and lowest at 20 ºC. Female developmental time from egg to adult is longer (20–23 days) than that of the males (15–17 day) at 25 ºC. The duration of immature stages of *C. malaccensis* varied from 11.3–13.8 days. The generation time of *C. malaccensis* ranged from 11.10–27.50 days. Increasing temperature shortened the development time. Lower and upper developmental thresholds were 11.6 and 37.8 ºC, respectively. *Cheyletus malaccensis* showed positive preference for larvae and nymphs, and negative preference for adults and eggs of *A. ovatus* (Zhu et al. 2019).

Predator-Prey Relations

Predators and their prey are linked through their trophic relationship and fluctuation in their population is dependent on densities of each of them (Leeuwen et al. 2007). The mechanism in which predation varies with predator and prey population dynamics is core to understanding of predator-prey relations. Exploitation of predator-prey interaction through laboratory and greenhouse simulations has aimed at elucidating and forecasting the mechanisms underlying predator-prey behavior to improve the practical predictive potential of natural enemies for biocontrol (Sepulveda and Carrillo 2008, Yao et al. 2014, Patel and Zhang 2017). The use of high-quality biocontrol agents

for release is a fundamental step in the successful implementation of biocontrol programs. Before using a biocontrol agent in an IPM program, it is essential to know about the efficiency of the bioagent (Fathipour et al. 2006). Knowledge about the characteristics of predators helps to understand their influence on the population dynamics of prey and their influence on the structure of the storage communities in which they exist (Jervis and Kidd 1996). Thus, prior information about predatory potential of biocontrol agents is a necessary prerequisite for the selection of mite predators for biocontrol programs and for the evaluation of their performance after release. The main methods that are used in the evaluation of the efficiency of predators include life table parameters, foraging behavior, and multitrophic interactions (Rahman et al. 2009ab, Rahman et al. 2012, Fathipour and Maleknia 2016, Jiadong et al. 2019, Zhu et al. 2019, Sun et al. 2020). Among life table parameters, the intrinsic rate of increase (r) is a key parameter in the prediction of population growth potential and has been widely used to evaluate efficiency of predators (Rahman et al. 2009a, Kianpour et al. 2011). Also, studies on multiple interactions like intraguild predation (predator–predator), olfactory response (plant–predator), tritrophic interaction (plant–prey–predator), cannibalism, and competition (predator–predator) have been used in evaluation of predatory mites (Maleknia et al. 2012, Maleknia et al. 2013, Khanamani et al. 2014, Farazmand et al. 2015, Maleknia et al. 2015). Foraging behaviors of predators including functional response, numerical response, mutual interference, preference, and switching are useful tools and have been used frequently to evaluate the efficiency of predators as bioagents (Holling 1959a, Opit 1986, Opit et al. 1997, Leeuwen et al. 2007, Rahman et al. 2009b,

Yao et al. 2014, Fathipour and Maleknia 2016, Patel and Zhang 2017, Souza-Pimentel et al. 2018, Jiadong et al. 2019, Zhu et al. 2019, Sun et al. 2020).

Among foraging behaviors, functional and numerical responses are key components in selection of natural enemies for biological control and these give information on practical suppression ability and colonization capacity of predators, respectively, in their habitat (Lester and Harmsen 2002). The functional response of predators describes the relationship between the numbers of prey attacked and prey densities whereas numerical response is defined as the change in a predator's reproductive output at varying prey densities (Holling 1959a, 1959b, Rahman et al. 2012). Functional response is an important aspect in the dynamics of a predator-prey relationship and a major component of population modelling, and it has been used to predict the mechanisms underlying predator-prey behavior to improve the practical predictive potential of predator candidates for biocontrol (Sepúlveda and Carrillo 2008). The major factors that affect the functional response are time of exposure of predator to prey, search rate, identification, capture, and prey consumption (Holling 1959b). Holling (1959a, 1959b, 1961) established three types of functional response models: Type I, II, and III.

For the Holling Type I, the per predator predation rate increases linearly with increase in prey density until it reaches a maximum consumption rate, implying a constant rate of capture per prey at a random search pattern, with a plateau of the curve occurring at the point of satiation. Generally, Type I response is found in filter feeders such as crustacean predators which prey on plankton in direct proportion to their availability in the niche (Hassel 1978). Holling (1959a, 1959b) Type I functional

response model per predator predation is represented by the equation (1): $N_a = aTN$. In this model, N_a is the number of preys killed, N is the initial density of prey, T is the time available for searching (experimental duration), a is the instantaneous rate of successful attacks, and N_a/N is the searching efficiency. Searching efficiency is constant for Type I predators since prey are killed at a constant rate till satiation.

A logistic regression of the proportion of prey consumed as a function of initial prey density can be used to determine the shape of the functional response curve of predators to different types or stages of prey: $N_a/N = exp\ (P_o + P_1 N + P_2 N^2 + P_3 N^3) / 1 + exp\ (P_o + P_1 N + P_2 N^2 + P_3 N^3)$, where N_a is the number of prey consumed, N is the initial prey density, (Na/N) is the probability of prey consumption and P_0, P_1, P_2, and P_3 are the maximum likelihood estimates of the intercept, linear, quadratic, and cubic coefficients, respectively (Juliano 2001). To determine the type of functional response, the signs of P_1 and P_2 are used. The predator displays a Type II response when the linear coefficient is significantly negative ($P_1 < 0$), which indicates that the proportion of prey consumed declines monotonically with the initial prey density. When the linear coefficient is positive ($P_1 > 0$), and the quadratic coefficient is negative ($P_2 < 0$), the predator has a Type III functional response (Juliano 2001).

Holling Type II has a cyrtoid curve with the rate of prey consumption per predator rising as prey density increases but does so at a decelerating rate until it reaches asymptote at which the consumption rate remains stable with increasing prey density. The searching efficiency (N_a/N) of Type II predators decreases with increase in prey density. Murdoch and Oaten (1975) and Murdoch (1973) explained that the initial rise in response curve is as a result of greater chance of contacts with prey with increasing prey

density, but, because some time is spent in handling prey with rise in prey density, the amount of time available for searching decreases, and the asymptote is as a result of decline in predator's average hunting rate due to satiation, thus, predator is likely to be more satiated often at high prey densities relative to low prey densities. Type II functional response incorporate predator handling time, which refers to the time required for predator to subdue, kill, and eat a prey and then perhaps clean and rest before moving on to search for more prey. This is thought to occur mainly in specialist predators (Leeuwen et al. 2007). A Type II functional response model per predator is described by the Holling (1959a, 1959b) disc equation (2): $N_a = aTN/ (1+aT_hN)$. In this model, N_a is the number of preys killed, N is the initial density of prey, T is the time available for searching during the experiment, a and T_h are the rate of successful attacks and the time required to handle prey item, respectively. The parameters a and T_h have been estimated using a linear regression technique where $1/Na$ is regressed on $1/N$; a and T_h are the reciprocal of its slope and intercept, respectively (Rahman et al. 2009b, Rahman et al. 2012, Yao et al. 2014). The a/Th value indicates the effectiveness of predation and the maximum predation rate (K) can also be estimated from T/T_h.

Holling Type III is described by a sigmoid curve with a positive accelerating rate up to an inflection and thereafter diminishing rate up to the plateau. According to O'Neil (1989) the upper asymptote of this response curve is due to the effects of handling time since at high prey densities, predators spend all available time handling prey. In a Type III functional response, the rate at which prey are killed increases with increasing prey density, however, at a given limit of prey density (O'Neil 1989). Unlike Type I and Type II responses which are not density dependent, Type III functional response results in

partial density-dependent prey mortality. A Type III functional response is associated with generalist predators (Andersson and Erlinge 1977, Hansson and Henttonen 1985). Similar to Type I and Type II functional responses, Type III functional response is unable to stabilize predator-prey dynamics, despite being partly density-dependent at a given threshold of prey density. This is due to the time-lag needed for numerical response to prey consumption (Hassel 1978). However, ignoring this time lag may result in stabilized predator-prey models of Type III functional response which is an indicator of efficient biocontrol agents (Hassel 1978). According to Murdoch (1973) and Murdoch and Oaten (1975), the Type III response may arise for two reasons: the predator may not receive enough stimuli from the prey at low prey densities to make it hunt intensively, and also, there may be refuges providing security for prey, however, as prey density increased with limited hidden places available, a greater proportion of prey become exposed and vulnerable to predation. Type III functional response predators were reported to have the ability to learn (Holling 1959a). Learning improves a predator's searching and handling efficiencies, which means the rate of successful search increases while handling time decreases. Also, the ability of predators to discern prey density and subsequently adjusting searching effort has been suggested as the basis of Type III response (O'Neil 1989). Hassel et al (1977) equation (3): $Na = N [1- exp \{-a (T-T_h Na)\}]$ was used to model Type III functional response curves with parameter description and estimation similar to Type II response. Predatory arthropods exhibit both Type II and Type III functional responses, but Type II is mostly observed in many predators that have been released as biocontrol agents (Cedola et al. 2001).

Numerical response of a predator is a progressive change in the number of its progeny in relation to prey density (Solomon 1949). It may be considered as a strategy of female predators to augment their offspring at different prey densities (Cédola et al., 2001). Omkar and Pervez (2004) estimated the efficiency of conversion of ingested food (ECI) into eggs biomass at different prey densities. The ECI was modelled by the equation (4): *ECI = [(Number of eggs laid / number of prey consumed) x 100]*. The data on oviposition and ECI at different prey densities are fitted using regression analysis to determine the relationship between (1) oviposition and prey density and (2) ECI of female predators and prey density. The ECI reveals the relationship between conversion of prey biomass and prey density in which the ECI is more at low prey density and subsequently decrease at higher prey densities. This probably indicates that female predators at low prey density probably invest most of their energy in egg production and, in the process, invest less in maintenance and metabolic activities. The decreased ECI at higher prey densities possibly suggests that well-fed females laid large numbers of eggs, besides investing much in maintenance and metabolic costs (Omkar and Pervez, 2004).

CHAPTER III

ESTABLISHING DISCRIMINATING DOSES OF PHOSPHINE FOR ADULTS OF SUSCEPTIBLE LABORATORY STRAINS OF PSOCIDS (PSOCODEA: LIPOSCELIDIDAE)

(To be submitted to Journal of Economic Entomology)

Abstract. Phosphine (PH_3) resistance and tolerance contribute to the increased importance of psocids (Psocodea: Liposcelididae) as stored-product pests. This study established the discriminating doses (DDs) of phosphine for laboratory susceptible adults of *Liposcelis bostrychophila* (Badonnel), *L. entomophila* (Enderlein), *L. decolor* (Pearman), *L. paeta* Pearman, *L. rufa* Broadhead, *L. obscura* Broadhead, *L. fusciceps* Badonnel, and *Lepinotus reticulatus* Enderlein. Discriminating dose is defined as the upper limit of the 95% confidence interval (95% CI) of the concentration that kills 99% of the individuals in a test sample (LC_{99}). Psocids were exposed to incremental phosphine concentrations of 0–300.0 ppm and 5.0–150.0 ppm over a 20-h and 72-h fumigation period, respectively, at 25°C using a modified FAO Method No.16. The established DDs had a range of 65.6–697.3 ppm and 18.1–194.5 ppm over a 20-h and 72-h fumigation period, respectively. For the 20-h fumigation period, the tested psocid species had higher DDs than those established for all stored-product insect pests in the FAO Method No.16 publication. Also, *L. entomophila* and *L. decolor* were the most tolerant psocid species over a 20-h and 72-h fumigation period, respectively. Moreover, *L. entomophila* required approximately 9.3 and 6.7x higher phosphine concentration compared to the most susceptible *L. obscura* and *L. reticulatus* over a 20-h and 72-h fumigation period, respectively, to achieve similar levels of LC_{50} mortality. The higher heterogeneity values in the standard 20-h fumigation period indicate the potential for increase in phosphine resistance in field populations subjected to phosphine fumigation. Discriminating doses estimated in this study can be used for detection of phosphine resistance and estimation of resistance frequencies in field-collected populations of the psocid species investigated.

Key words: booklouse, insecticide resistance, fumigation, tolerance, stored-product pest

Introduction

Fumigants are major tools for stored-product pest management and have been used for over a century to meet domestic and international export phytosanitary requirements worldwide (Chaudhry 2000, Cao et al. 2003, Opit et al. 2012, Nayak et al. 2020). The fumigant, phosphine (hydrogen phosphide: PH_3) is a primary component of stored-grain IPM systems for managing stored-grain pests in the United States (Opit et al. 2012, Nayak et al. 2014, Gautam et al. 2016, Afful et al. 2018). Phosphine usage has increased considerably over the last three decades due to methyl bromide phase-out and restricted use, its efficacy against a broad spectrum of pestiferous arthropods, market reluctance to accept protectant insecticide residuals, and resistance in target pests to contact insecticides (Emery et al. 2011, Nayak et al. 2015). Leaky storage facilities, lower fumigation temperatures, and the practice of overdosing to compensate for the lack of airtight storage structures result in phosphine fumigation failures because of sub-lethal concentrations against targeted pest population (Opit et al. 2012, Gautam et al. 2016, Nayak et al. 2020). In effect, a higher frequency of application above recommended lethal dose to control surviving population results in higher selection pressure for phosphine resistance in the presence of resistance genotypes (Nayak et al. 2020). Over time, the presence of resistant insect pests coupled with selection pressure leads to higher resistance frequencies in pest populations, and ultimately loss of efficacy of phosphine against insect populations (Benhalima et al. 2004). In addition, the occurrence of multi-species infestations by pestiferous insects in storage communities have been shown to undermine the efficacy of phosphine treatments (Rees 1998, Athanassiou et al. 2010). Interspecific differences in response to phosphine may have contributed to the

development of genetically based resistance due to indiscriminate exposure of different species to similar doses or sub-lethal concentrations of phosphine in storage communities (Nayak et al. 2014).

In the United States, early studies on phosphine resistance by Zettler et al. (1989) reported resistance in *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae), *Cadra cautella* (Walker) (Lepidoptera: Pyralidae), and *Plodia interpunctella* (Hubner) (Lepidoptera: Pyralidae) from peanut storage facilities. Zettler and Cuperus (1990) reported phosphine resistance in *T. castaneum* and *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) from wheat stored in farms; Zettler (1991) again reported resistance in *T. castaneum* and *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) from flour mills; Zettler and Keever (1994) in *Lasioderma serricorne* (F.) (Coleoptera: Anobiidae) from tobacco storage facilities. More recent reports of phosphine resistance in the United States involved *R. dominica* and *T. castaneum* in the states of Kansas and Oklahoma (Opit et al. 2012, Cato 2015, Chen et al. 2015, Afful et al. 2018), *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae) in Oklahoma (Konemann et al. 2017), and *T. castaneum* and *P. interpunctella* in California State (Gautam et al. 2016). However, there is a lack of information on phosphine resistance of psocids (Psocodea: Liposcelididae) in the United States.

Phosphine resistance and tolerance contribute to the increased importance of psocids as stored-product pests and this has occurred in many parts of the world (Ahmedani et al. 2010, Nayak et al. 2014). Recent studies in Australia found that a population of *L*iposcelis *bostrychophila* (Badomel) (Psocodea: Liposcelididae) adult insects survived a phosphine concentration approximately 5x greater than required to kill

all individuals of the susceptible strain in a sample, whereas resistant populations of *L. decolor* (Pearman) and *L. entomophila* (Enderlein) required concentrations of phosphine approximately 6x and 7x higher, respectively (Nayak et al. 2000). Also, to kill resistant *L. bostrychophila* eggs needed a phosphine concentration (2 g/m^3) 65 times greater than that required for the susceptible strain (0.031 g/m^3). Cao et al. (2003) and Pike (1994) have also found resistant strains of *L. entomophila* in China and Indonesia, respectively. In India, *L. bostrychophila* was found to be resistant to phosphine (Rajendran 1994). Given the resistance to phosphine found in other parts of the world, it is important to conduct resistance studies for psocids collected in the United States as well. Discriminating dose is a diagnostic bioassay technique mostly used for detection of phosphine resistance in major storage insect pests (FAO 1975, Champ and Dyte 1976, Opit et al. 2012, Gautam et al. 2016, Konemann et al. 2017, Cato et al. 2017, Afful et al. 2018). Discriminating dose of phosphine, for a given species, is estimated by determining the concentration of fumigant that kills 99% (LC_{99}) of susceptible laboratory-reared insects in a fumigation that lasts a specified period of time and temperature (20 h fumigation period and 25°C, respectively, in the standard protocol) (FAO 1975). The discriminating dose is then estimated as the upper limit of the 95% confidence interval of the LC_{99} value (lethal concentration required to kill 99% of the population or individuals in a sample) (Konemann et al. 2014, Gautam et al. 2016). The discriminating dose bioassay is important because it permits the estimation of resistance frequency— resistance frequency is the percentage of insects in each sample exposed to the discriminating dose of a fumigant that survives.

The discriminating dose of phosphine for insect pests may be affected by factors such as insect biotype and developmental stages, fumigation period, and the geographic location where the laboratory-reared susceptible insects were originally collected. For example, 0.05 m/L (~ 35.7 ppm), 0.06 m/L (~ 42.9 ppm), and 0.08 m/L (~ 56.2 ppm) are three different estimated discriminating doses that have been established for the detection of resistant individuals of *C. ferrugineus* under 20-h period at 25°C from previous studies by Sartori et al. (1990), Nayak et al. (2012), and Konemann et al. (2017) in Brazil, Australia, and the United States, respectively. Also, in the United States, Gautam et al. (2016) investigated the discriminating doses of phosphine for eggs and larvae of *P. interpunctella* and eggs of *T. castaneum* using laboratory susceptible strains of the two species. Their results showed that for *T. castaneum* and *P. interpunctella* eggs, discriminating doses were 62.4 and 107.8 ppm, respectively, over a 3-d fumigation period, and for *P. interpunctella* larvae, the discriminating dose was 98.7 ppm over a 20-h fumigation period. The fact that discriminating dose varies in insect species and developmental stages, fumigation temperatures and exposure time as well as geographical locations means it is prudent to accordingly determine species-specific discriminating dose for any given stored-product insect species. Therefore, this study was conducted to determine the discriminating doses of *Liposcelis bostrychophila* (Badonnel), *L. entomophila* (Enderlein), *L. decolor* (Pearman), *L. paeta* Pearman, *L. rufa* Broadhead, *L. obscura* Broadhead, *L. fusciceps* Badonnel, and *Lepinotus reticulatus* Enderlein using laboratory susceptible strains collected within the United States. The fumigation conditions were 20-h and 72-h exposure periods at a constant temperature of 25°C and 75 ± 5% RH. Phosphine gas application under different fumigation periods may result in

different lethal concentrations of phosphine required to manage specific species of insect pests. Therefore, it was expected that the longer fumigation period would produce lower discriminating doses of phosphine that are economical, environmentally friendly, and suitable for detection of phosphine resistance and estimation of resistance frequencies in field-collected populations of psocids under laboratory conditions in the United States.

Materials and Methods

Insects. Laboratory susceptible strains of eight psocid species, *L. bostrychophila*, *L. decolor*, *L. entomophila*, *L. fusciceps*, *L. obscura*, *L. paeta, L. reticulatus*, and *L. rufa* were taken from laboratory cultures maintained at the Stored Product Entomology Laboratory in the Department of Entomology and Plant Pathology, Oklahoma State University, Stillwater, OK. Cultures of *L. bostrychophila* and *L. paeta* used in this study were started from insects originally collected from a grain elevator at the USDA-ARS Center for Grain and Animal Health Research (CGHAR), Manhattan, KS whereas insects for starting cultures of *L. decolor*, *L. entomophila*, and *L. reticulatus* were collected from steel bins containing wheat also at CGAHR. *Liposcelis fusciceps* was collected from an animal feed warehouse in West Lafayette, IN and *L. obscura* from a peanut warehouse in Anadarko, OK. *Liposcelis rufa* was collected from steel bins containing wheat in Stillwater, OK. Adult female of the selected psocid species were used for this study. Voucher specimens of 100 female *L. bostrychophila, L. decolor*, *L. entomophila, L. paeta*, *L. fusciceps*, *L. obscura*, *L. reticulatus*, and *L. rufa* preserved in 95% ethyl alcohol that were used in this study were deposited at the K. C. Emerson Entomology Museum at

Oklahoma State University (OSU) under lot numbers 106, 107, 110, 111, 115, 119, 120, and 101, respectively. Psocids were reared in a laboratory at OSU on a mixture 93% cracked wheat (Duster variety), 5% Rice Krispies (Kellogg Company, Battle Creek, MI), and 2% wheat germ (wt/wt; referred to as psocid diet below) in 360-ml glass canning jars with mite-proof lids (Opit and Throne 2008), and the top 3 cm of the inner surface of each jar was coated with Fluon (polytetrafluoroethylene; Northern Products, Woonsocket, RI) to prevent psocids accessing and gathering on the inside of the lid. Cultures were maintained in a laboratory chamber set at 30.0 ± 1°C and 75 ± 5% relative humidity (RH). The eight species of psocids used have been kept under laboratory conditions for over 10–15 years.

Dose-Response Bioassay. Discriminating doses of phosphine for adults of *L. bostrychophila*, *L. decolor*, *L. entomophila*, *L. fusciceps*, *L. obscura*, *L. paeta, L. reticulatus*, and *L. rufa* were determined in laboratory dose-response experiments where susceptible laboratory strains of the psocid species were exposed to concentrations of phosphine ranging from 5.0–300.0 ppm and 5.0–150.0 ppm for 20-h and 72-h fumigation periods, respectively. The dose-response test for each species was based on a modified FAO Method No. 16 protocol (Food and Agriculture Organization 1975). These concentrations were attained by injecting pre-calculated volumes of 10,000 ppm of phosphine gas, in 3.92-liter fumigation jars (S-12758M, Uline, Waukegan, IL) (referred to as fumigation jars hereafter). The volume of 10,000 ppm of phosphine to be injected in each fumigation jar was calculated using a formula, $C_1V_1 = C_2V_2$ — where C_1 is the starting concentration of 10,000 ppm, C_2 is the target concentration, V_2 is the 3,920 ml volume of the fumigation jar, and V_1 is that volume of 10,000 ppm phosphine gas that is

injected in each fumigation jar. The volume was then adjusted by 10% to account for any loss during the process of injection. Each concentration was replicated three times. Fumigations were conducted at 25°C.

Preparation of Insects. For each of the eight species, 50 live female adults were placed in individual well labeled 3.5-cm diameter Petri dishes with three pieces of cracked wheat and red-colored psocid diet (Opit and Throne 2008). Petri dishes with psocids were accordingly placed in the fumigation jars, including the control jars. Female adults of each species that were 1- to 2-week-old were used, and were obtained using the method of Opit and Throne (2008). The current study used only adult females of the different psocid species because they are robust, aggressive, and mostly, tolerant motile stage to manage. There were three replicates for each concentration, i.e. three jars were assigned to each phosphine concentration. Each Petri dish was labeled with information indicating the species, phosphine concentration, and replication. The labeled vials were then placed inside respective fumigation jars assigned to different concentrations of phosphine.

Fumigation Jars. A fumigation jar consisted of a 3.92-liter glass jar (S-12758M, Uline, Waukegan, IL) along with a plastisol® lined metal lid (S-18023, Uline, Waukegan, IL). The lid was equipped with a port in the center, which was fitted with a rubber injection septum for introduction and sampling of the fumigant. A double layer of Teflon tape was applied to the outside of the lid after the lid was screwed on and to the outside edge of the rubber septum to prevent gas leakage. Prior to placing Petri dishes containing psocids inside jars, two drops of water were added to each jar to maintain 65 ± 5% RH. After placing Petri dishes containing psocids in fumigation jars and prior to

injection of phosphine, a volume of air 1.5-times the amount of gas to be added was removed using a gas-tight syringe (100 ml, Hamilton 1100 SL SYR, Hamilton Inc., Reno, NV). Pre-calculated volumes of 10,000 ppm phosphine were then added (injected) into fumigation jars to give the desired concentrations in fumigation jars. Phosphine was injected through the rubber septum placed on each port.

Phosphine Quantification. Laboratory fumigation methods and gas chromatographic-flame photometric detector (GC-FPD) quantification of average applied concentration of phosphine in fumigation jars were by the methods described by Sekhon et al. (2010). The gas chromatograph that was used is an SRI instruments 8610-C with a flame photometric detector (FPD) equipped with a phosphorous filter. The gas was passed through an Rt™ – QS-Bond 30-meter, 0.53 mm ID Silica Column. The carrier gas used was ultra-high purity helium (Linweld, Lincoln, NE).

The GC-FPD was calibrated using 200 ppm phosphine gas (Matheson Tri-gas) before taking samples from the jars. The concentrations were established using a standard curve based on 50, 40, 30, 20, and 10 µl of 200 ppm phosphine. For establishing the standard curve, gas samples corresponding to each of the above volumes was withdrawn from the Tedlar® bag containing 200 ppm of phosphine using a calibrated gas-tight syringe (50 µl, Hamilton 1705 TLL SYR, Hamilton Inc., Reno, NV) and injected into the on-column injector of the GC-FPD. The areas under the peak in microvolts (µV) counts were recorded along with the volume of phosphine injected. Phosphine volumes were regressed against measured peak areas to generate a straight-line regression equation that had a coefficient of determination (R^2) value between 0.96 and 0.99 in all cases. Thirty microliter gas samples from each fumigation jar were analyzed using the GC-FPD and

quantified using the regression equation generated from the standard curve. The average between the estimated initial and the final phosphine concentration during the 20-h or 72-h fumigation period was considered as the concentration for the sampled jar.

Post-Fumigation Protocol. After the sampling of end concentrations, jars were aerated by removing lids inside a certified fume hood. The Petri dishes in each jar were removed and kept in a plastic box (42.9 cm × 29.2 cm × 23.5 cm) in an incubator maintained at 25 ± 1°C and 70 ± 5% RH for 14 d. Final mortality assessments were conducted after 14 d. Psocids that did not move after prodding with a damp horsetail painting brush were counted as dead.

Probit Analysis. The experimental designs for determining discriminating doses were completely randomized designs with three replications. Phosphine dose-response mortality data for each species was subjected to probit analysis using PoloPlus Software (LeOra Software 2005) to estimate lethal concentrations to kill 50, 95, and 99% individuals in samples, i.e., LC_{50}, LC_{95}, and LC_{99} values, and their 95% confidence intervals (CIs). The discriminating dose is expected to kill all susceptible insects (FAO 1975). The objective of this study was to establish doses to control 99% of the susceptible individuals in samples tested. Therefore, depending on the species, the discriminating dose is the upper limit of the 95% CI of the LC_{99} value at a given exposure period at 25°C. In order to evaluate that the value of the mean is within the limit at 95% probability, we calculated G-factor using the equation, $t^2V(b)/b^2$, where t = student's *t*-test with error degrees of freedom, V(b) is the slope variance estimate given in the variance-covariance matrix, and b is the slope estimate. If G-values are less than 0.5, it

suggests that the value of the mean is within the limit at 95% probability (Gautam et al. 2020).

Results

Discriminating Doses. Based on dose-response studies using susceptible adults of *L. obscura, L. bostrychophila*, *L. paeta*, *L. fusciceps*, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila*, phosphine discriminating doses for these species were 65.6, 77.5, 124.7, 149.4, 159.9, 205.0, 249.8, and 697.3 ppm over a 20-h fumigation exposure period, respectively, at 25°C and 75% RH (Table 1). For the 72-h fumigation exposure period, the discriminating doses for adults of *L. reticulatus*, *L. fusciceps*, *L. obscura*, *L. rufa*, *L. paeta*, *L. bostrychophila*, *L. entomophila*, and *L. decolor* were 18.1, 19.2, 23.2, 25.5, 39.6, 49.1, 157.1, and 194.5 ppm, respectively (Table 2). PoloPlus uses the heterogeneity factor (describes a function of ratio of estimated chi-square value to its degree of freedom, and it gives an indication of the differences in response to phosphine in the specific psocid species tested) as a correction factor when the value of Pearson's chi-square statistic (χ^2) is significant at P = 0.05 (LeOra software 2005). According to Finney (1952), a significantly large χ^2 value indicates that all weights have been overestimated by a factor χ^2/ (k-2), where k is the number of dosages tested. Therefore, all variances should be multiplied by this heterogeneity factor as compensation for the overestimation. Despite heterogeneity values greater than one, the respective fitted lines tracked the experimental data. Heterogeneity values greater than one in our data for both the 20-h and 72-h fumigation exposure periods are evidence of considerable heterogeneity in response to phosphine of the tested species (Table 1 and Table 2). For

the 20-h exposure period, heterogeneity values greater than one were found in all the species tested except for *L. obscura*. In the 72-h exposure period, tested species were more homogenous in their response to phosphine except *L. bostrychophila*, *L. entomophila*, and *L. decolor* that showed considerable heterogeneity to phosphine with heterogeneity values greater than one (Table 2). The index of significance of potency estimation g-factor indicates that the value of the mean is within the limits at all probability levels as it is less than 0.5. When values of heterogeneity factor are less than one they denote that in the replicate tests of random samples the concentration response lines would fall within 95% confidence limits, and that the model fits the data adequately. Thus, the estimated g-factor estimates indicates that LC_{99} value is still good in cases where the heterogeneity factor estimates exceeded one (Table 1 and Table 2). A narrower range of slopes was estimated for the 20-h exposure period (2.5–4.3) compared to the 72-h exposure period (1.7–4.2) indicating a higher heterogeneity level for shorter fumigation time (Table 1 and Table 2).

Lethal concentration ratios. Comparison of proportion of lethal concentrations required to kill 50%, 95%, and 99% of individuals in samples (LC_{50}, LC_{95}, and LC_{99}) of the eight laboratory-susceptible strains (*Lab-S*) of psocids were established for the 20-h and 72-h fumigation periods (Table 3). For the 20-h exposure period, *L. obscura* was the most susceptible species to phosphine and had the lowest LC_{50}, LC_{95}, and LC_{99} values. In the case of the 72-h exposure period, *L. reticulatus* had the lowest concentrations required to kill similar proportions of individuals in samples. Therefore, lethal concentration ratios (LCRs) were established by comparing LC_{50}, LC_{95}, and LC_{99} values of *L. obscura* with corresponding values of each of the other seven species under the 20-h fumigation period.

Similarly, LC_{50}, LC_{95}, and LC_{99} values of *L. reticulatus* were compared with the other seven species for the 72-h fumigation period. Cases where the comparisons resulted in 95% CIs that did not include one indicate that the LC values of the species being compared in each case were significantly different from each other for a specified fumigation period (Table 3). Based on the LC_{50} comparisons for 20-h exposure period, the concentrations of phosphine required for *L. bostrychophila*, *L. paeta*, *L.* fusciceps, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila* were 1.3, 1.7, 3.4, 3.8, 2.1, 2.2, and 9.3x, respectively, higher than dose required to kill 50% of *L. obscura* samples (Table 3). Based on LC_{50} values for the 72-h exposure period, the concentrations of phosphine required for *L. fusciceps, L. obscura, L. rufa, L. paeta, L. bostrychophila, L. entomophila*, and *L. decolor* were 2.0, 2.0, 3.7, 2.1, 1.9, 6.7, and 2.8x, respectively, more than dose required to kill 50% of *L. reticulatus* samples. Therefore, in the case of both the 20-h and 72-h exposure periods, *L. entomophila* had the highest level of phosphine tolerance, and was 9.3x and 6.7x, respectively, more tolerant than *L. obscura* and *L. reticulatus* (Table 3).

Phosphine concentration reduction indexes (PCRIs). The estimates of PCRIs are proportions of discriminating dose values of 20-h to 72-h fumigation periods of each species. The PCRI for *L. decolor*, *L. bostrychophila*, *L. obscura*, *L. paeta*, *L. entomophila*, *L. rufa*, *L. fusciceps*, and *L. reticulatus* were 1.28, 1.58, 2.83, 3.15, 4.44, 6.27, 7.78, and 11.36, respectively (Table 4). Species with higher PCRIs such as *L. reticulatus, L. fusciceps*, and *L. rufa* rapidly responded to phosphine concentrations with extended fumigation exposure period and may be effectively managed by increasing fumigation time at a constant temperature of 25°C. However, species with lower indexes

including *L. decolor, L. bostrychophila*, and *L. obscura* may not be effectively managed by increasing fumigation time, but instead require other factors such as increase in phosphine concentrations, temperatures, or decrease in fumigation RH to increase phosphine toxicity against such fumigation time non-responsive species.

Discussion

Discriminating doses for laboratory-susceptible strains (*Lab-S*) of eight psocid species (adults of *L. obscura, L. bostrychophila*, *L. paeta*, *L. fusciceps*, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila*) over a 20-h and 72-h fumigation period showed considerable variations in levels of dose-response mortality to incremental concentrations of phosphine gas under a constant temperature of 25°C. The adults of *Lab-S* used for establishing phosphine dose-response mortality levels were sampled from the United States and kept under laboratory conditions for over 10–15 years without any exposure to phosphine fumigation prior to this experiment. However, the range of discriminating dose over a 20-h fumigation period (65.6–697.3 ppm) was substantially high across all psocid species tested compared with that of the 72-h exposure period (18.1–194.5 ppm). Again, the discriminating doses under the current 20-h fumigation period for psocids were by far the highest for stored-product insects compared to those listed by FAO (1975) for adults of major stored-product pests; — a range of discriminating doses of ~21.6 – 50.3 ppm for eight laboratory susceptible strains of stored-product beetles were established at $LC_{99.9}$. FAO Protocol #16 established discriminating doses at $LC_{99.9}$ over a 20 h fumigation period and 25°C, however, because

it is difficult to estimate discriminating doses at the specified lethal concentration ($LC_{99.9}$), most published studies have estimated discriminating doses using the upper limit of the 95% CI of LC_{99} (Opit et al. 2012, Gautam et al. 2016, Konemann et al. 2017, Cato et al. 2017, Afful et al. 2018). The present discriminating doses for psocids are remarkably high when compared with available data for other insect pests (FAO 1975). Based on the present LC_{50} estimates, the range of phosphine concentrations were 9.2–71.0 ppm and 2.3–10.3 ppm, for psocids in 20-h and 72-h fumigation periods, respectively, but ~5.0– 9.4 ppm in the FAO (1975) list of major beetle pests of stored products. The FAO (1975) tentative method No. 16 required adults of the tested sample to be exposed for a 20-h fumigation period at 25°C and 70% RH, and end-point dose mortality responses established 14 days after the end of the exposure period.

Previous publications besides FAO (1975) have reported on discriminating doses for different developmental stages of beetles and moths which were lower relative to the most phosphine tolerant adult psocid species in the present study: *L. entomophila* and *L. decolor*— 697.3 and 249.8 ppm or 157.1 and 194.5 ppm over a 20-h or 72-h fumigation period, respectively. For example, discriminating doses for adult *C. ferrugineus* using laboratory susceptible insects from Australia, Brazil, and the United States were 35.7 ppm, 42.9 ppm, and 56.2 ppm, respectively over a 20-h fumigation period at 25°C (Sartori et al. 1990, Nayak et al. 2012, Konemann et al. 2017). According to Gautam et al. (2016) phosphine discriminating doses for eggs of *T. castaneum* and *P. interpunctella* were 62.4 and 107.8 ppm over a 72-h fumigation period, respectively, at 25°C. For larvae of *P. interpunctella* and adult *T. castaneum*, the discriminating doses were 98.7 ppm and 8.2 ppm over a 20-h and 72-h fumigation period, respectively. Bell et al. (1977) also,

established discriminating dose for *R. dominica* eggs as 114.3 ppm over a 24-h fumigation period.

The discriminating doses for psocids in this study confirm findings from other studies (Nayak et al. 1998, Collins et al. 2001, Daglish et al. 2003, Athanassiou and Rumbos 2018) that showed that psocids exhibit natural tolerance to many insecticides including grain protectants that are effective against coleopteran and lepidopteran pests of stored products. According to Athanassiou and Rumbos (2018) natural tolerance in psocids is a phenomenon that should not be considered as an effect of previous exposure, but more as a genetic-related issue. Although the phosphine resistance levels and frequencies of psocids (particularly, *L. entomophila* and *L. decolor*) have not yet been established in the United States, it can be expected to be as high as or higher than the most phosphine resistant stored-product insect pests such as *C. ferrugineus*. Moreover, adult susceptible strains of psocids that were more tolerant to phosphine would predictably have higher phosphine tolerance levels in their egg stages than their respective adult developmental stages. Correlation between egg and adult stages for phosphine resistance frequencies in stored-product insect pests have been reported (Bell 1977, Gautam et al. 2016). Nevertheless, interspecific variations in dose-response mortality were found among psocids species over a 20-h or 72-h fumigation period.

As indicated before, phosphine tolerance levels vary considerably with fumigation time among psocid species at constant fumigation temperature. *Liposcelis entomophila* and *L. decolor* were the most phosphine tolerant psocid species over a 20-h and 72-h fumigation period, respectively. However, phosphine dose-response mortality levels were mostly significant and higher in *L. obscurus* and *L. reticulatus* over these

periods. According to Nayak et al. (2020) the two important non-biotic factors affecting phosphine efficacy are concentration and exposure time, both of which can be manipulated to maximize phosphine toxicity against insect pests. Daglish et al. (2002), Collins et al. (2005) and Kaur et al. (2015) found that increasing either concentration or time will increase efficacy against resistant insects, with fumigation time being more important than concentration. While increasing fumigation exposure time would substantially reduce discriminating doses of *L. paeta*, *L. entomophila*, *L. rufa*, *L. fusciceps,* and *L. reticulatus*, the discriminating doses of *L. decolor, L. bostrychophila*, and *L. obscura* were not substantially affected. However, the latter group of psocids that are more stable to increasing fumigation exposure time may be further evaluated by exposing them to other variables such as higher phosphine concentrations, elevated temperatures, and lower fumigation relative humidities (Daglish et al. 2002, Kaur et al. 2015, Nayak et al. 2020).

The phosphine concentration reduction indexes (PCRI) established in this study could serve as a quick management decision tool for deciding on psocid species that can or cannot be controlled by varying fumigation exposure time. This would be necessary when working with highly tolerant psocid species with unrealistically higher lethal concentrations over a 20-h fumigation period. The occurrence of multiple-species infestations which are frequently found among psocids population in stored product commodities has been reported to undermine the efficacy of phosphine treatments (Rees 1998, Athanassiou et al. 2010). Therefore, information from the current study provides species-specific diagnostic doses which serve as base-line information to evaluate phosphine resistance levels and frequencies in the field-collected populations of the

psocid species that were investigated to monitor phosphine resistance. Subsequently, this would enhance the development of rapid detection methods of phosphine resistance including molecular-based diagnostic techniques.

The current study revealed higher heterogeneity levels among psocid species when exposed to a shorter fumigation period compared with a longer exposure time. All tested species were heterogeneous in response to phosphine under a shorter fumigation duration, except *L. obscura* that showed homogeneity in response to phosphine over a 20-h fumigation time. Similarly, *L. bostrychophila, L. entomophila,* and *L. decolor* were heterogonous over a 72-h fumigation period. This confirms the previous report of other studies that indicated that the response of psocids to phosphine was heterogeneous (Ho and Winks 1995, Nayak et al. 1998, Konemann et al. 2015). Storage communities with such high heterogeneity insect species in response to phosphine concentrations suggests that for effective control of all species including the tolerant strains, it is important that concentration that controls the most fumigant tolerant species should be used (Gautam et al. 2016). In addition, the low slopes of *L. decolor* and *L. entomophila* over a 20-h and 72-h exposure period indicated the potential for a significant increase in phosphine resistance in these populations (Cao et al. 2003).

Given the recommended dosage of phosphine for stored grain (wheat) in Oklahoma State of 200 ppm over a 4-d or longer exposure at 20–30 °C, and the fact that commercial grain storage facilities fumigate on an average 3 times per year (Cuperus et al. 1990, Leesch et al. 1995, Phillips et al. 2012), we can expect widespread phosphine resistance in field populations of psocids in Oklahoma particularly, in *L. entomophila* and *L. decolor* which are the most predominant species infesting stored grains in the United

States (Throne et al. 2006, Opit et al. 2009). Most of the current psocid species were collected 10–15 years ago in similar storage facilities to those in Oklahoma where the most resistant *T. castaneum* and *R. dominica* were sampled almost a decade ago (Opit et al. 2012).

In conclusion, this study has confirmed that compared to adults of coleopteran stored-product insect pests, the phosphine discriminating doses are relatively higher in *L. decolor, L. bostrychophila, L. obscura, L. paeta, L. entomophila, L. rufa, L. fusciceps,* and *L. reticulatus* and is the first report of discriminating dose data for psocids collected in the United States. Based on the FAO (1975) recommended 20-h fumigation period at 25°C, the present established discriminating doses for adults of eight psocids species are by far the highest estimated diagnostic values among all studied stored-product insect pests. Also, *L. entomophila* and *L. decolor* were the most tolerant psocid species over a 20-h and 72-h fumigation period, respectively. Again, to achieve similar levels of phosphine dose-response mortality at LC_{50}, *L. entomophila* required approximately 9.3 and 6.7 times higher phosphine concentrations than the most susceptible strains *L. obscura* and *L. reticulatus* over a 20-h and 72-h fumigation period, respectively.

This study has confirmed heterogeneity in response to phosphine in psocids, particularly over a shorter phosphine fumigation time, a mechanism that facilitates the development of genetic-based resistance in both inter-and con-specifics storage insect pests. The phosphine concentration reduction index (PCRI) provides managerial reference resources for phosphine resistance management in psocids. This gives an indication of the importance of extending fumigation period to reduce heterogeneity in psocids in response to phosphine, and provides information on discriminating doses

estimates which are economical, environmentally friendly, and practically suitable for use to determine phosphine resistance frequencies of the field-collected population under laboratory conditions. However, psocid species such as *L. decolor* and *L. bostrychophila* may be less affected by extending fumigation time by 3.6 folds from the standard 20-h fumigation period. Future research should be aimed at establishing the discriminating doses over similar fumigation periods for eggs stage of laboratory susceptible strains of *L. decolor*, *L. bostrychophila*, *L. obscura*, *L. paeta*, *L. entomophila*, *L. rufa*, *L. fusciceps,* and *L. reticulatus*. Thereafter, data from the current study (adult stage) and the subsequent research on egg stage could be used to evaluate phosphine resistance level and frequencies in field-sampled populations of the aforementioned species from Oklahoma and other key grain growing areas in the United States.

Acknowledgments

We thank Charlie Konemann and Zhaorigetu Chen for excellent technical assistance. This research was funded by Oklahoma Agricultural Experiment Station (Project number OKL2949). Trade names or commercial products mentioned in this paper are solely for the purpose of providing specific information and does not imply recommendation or endorsement by Oklahoma State University.

Table 1. Probit analyses of mortality responses of susceptible adult psocids of eight species to ranges of phosphine gas concentrations over a 20-h exposure period at 25 ± 1°C and 75 ± 5% RH. Concentrations are in parts per million (ppm).

Species	*N*	Slope ± SE	LC_{50} (95% CI)	LC_{95} (95% CI)	LC_{99} (95% CI)[a]	χ^2 (df)[H[b]]	G-factor[c]
L. obscura	1200	3.1 ± 0.2	9.2 (8.2–10.0)	31.0 (27.5–35.9)	51.2 (42.3–65.6)	21.7 (22) [0.98]	0.021
L. bostrychophila	1450	3.4 ± 0.2	10.8 (9.2–12.5)	33.3 (26.9–44.3)	53.1 (40.5–77.5)	76.8 (27) [2.8]	0.012
L. paeta	1350	3.2 ± 0.2	13.6 (9.9–17.2)	43.6 (33.4–65.2)	70.8 (50.2–124.7)	134.2 (25) [5.4]	0.016
L. fusciceps	1200	3.7 ± 0.2	27.8 (25.1–30.7)	76.9 (66.6–92.3)	117.2 (97.1–149.4)	41.7 (22) [1.9]	0.011
L. rufa	1200	4.3 ± 0.3	31.0 (26.3–35.3)	74.1 (61.7–98.7)	106.3 (85.1–159.9)	76.1 (22) [3.46]	0.021
L. reticulatus	1200	2.6 ± 0.2	16.2 (13.1–19.2)	70.4 (55.7–97.4)	129.6 (94.3–205.0)	53.5 (22) [2.43]	0.017
L. decolor	1200	2.5 ± 0.2	17.4 (13.0–21.8)	78.1 (59.9–113.8)	145.6 (102.1–249.8)	87.0 (24) [3.6]	0.015
L. entomophila	900	2.9 ± 0.3	71.0 (55.3–84.0)	265.2 (223.5–343.3)	457.7 (351.7–697.3)	25.4 (16) [1.6)	0.038

[a]None overlapping 95% confidence interval (CI) in any comparison suggests that the two lethal concentrations (LCs) are significantly different.
[b]Heterogeneity factor [H], chi-square value (χ^2), degrees of freedom (df). Chi-square is significant, ($P<0.05$).
[c]G-factor, $t^2V(b)/b^2$, where t=student's t with error degrees of freedom, V(b) is the slope variance estimate given in the variance–covariance matrix, and b is the slope estimate. G-values that are less than 0.5 suggest that the value of the mean is within the limit at 95% probability.

Table 2. Probit analyses of mortality responses of susceptible adult psocids of eight species to ranges of phosphine gas concentrations over a 72-h exposure period at 25 ± 1°C and 75 ± 5% RH. Concentrations are in parts per million (ppm)

Species	*N*	Slope ±SE	LC_{50} (95% CI)	LC_{95} (95% CI)	LC_{99} (95% CI)[b]	χ^2 (df)[H^a]	G-factor[c]
L. reticulatus	1050	3.4 ± 0.7	2.3 (1.3–2.9)	6.8 (5.8–8.8)	10.7 (8.4–18.1)	3.3 (19) [0.2]	0.176
L. fusciceps	1050	3.6 ± 0.5	3.0 (2.6–3.6)	8.6 (7.4–10.8)	13.2 (10.6–19.2)	1.3 (19) [0.1]	0.088
L. obscura	1050	3.2 ± 0.4	3.0 (2.3–3.6)	9.8 (8.4–12.2)	16.1(12.8–23.2)	6.0 (19) [0.3]	0.072
L. rufa	1050	4.2 ± 0.3	5.8 (5.3–6.3)	14.2 (12.5–16.6)	20.5 (17.4–25.5)	8.1 (19) [0.4]	0.028
L. paeta	1050	2.5 ± 0.3	3.0 (2.0–5.5)	14.1 (12.0–17.5)	26.7 (20.7–39.6)	17.5 (19) [0.9]	0.069
L. bostrychophila	1050	2.4 ± 0.3	2.5 (1.1–3.8)	12.5 (9.9–17.9)	24.1 (17.1–49.1)	34.8 (19) [1.8]	0.082
L. entomophila	1200	2.3 ± 0.1	10.3 (8.7–12.0)	54.9 (45.1–70.6)	109.7 (83.5–157.1)	36.21 (22) [1.7]	0.014
L. decolor	1050	1.7 ± 0.2	4.0 (2.5–5.5)	38.0 (28.5–57.7)	96.2 (62.3–194.5)	30.68 (19) [1.6]	0.040

[a]None overlapping 95% confidence interval (CI) in any comparison suggests that the two lethal concentrations (LCs) are significantly different.
[b]Heterogeneity factor [H], chi-square value (χ^2), degrees of freedom (df). Chi-square is significant, (P<0.05).
[c]G-factor, $t^2V(b)/b^2$, where t=student's t with error degrees of freedom, V(b) is the slope variance estimate given in the variance–covariance matrix, and b is the slope estimate. G-values that are less than 0.5 suggest that the value of the mean is within the limit at 95% probability.

Table 3. Comparison of lethal concentrations required to kill 50%, 95%, and 99% (LC_{50}, LC_{95}, and LC_{99}) of *L. obscura* with concentrations required to kill similar proportions of *L. bostrychophila*, *L. paeta*, *L. fusciceps*, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila* for the 20-h fumigation period. Additionally, similar comparisons for *L. reticulatus* with the other seven species for the 72-h fumigation period. Fumigations were conducted at 25 ± 1°C and 75 ± 5% RH. Lethal concentrations are in parts per million (ppm).

Fumigation period	Lethal concentration ratios		
(Species compared)	LC_{50} (95% CI)	LC_{95} (95% CI)	LC_{99} (95% CI) [a]
20-h (*L. obscura*)			
L. bostrychophila	1.2 (1.0–1.3)	1.1 (0.7–1.3)	1.0 (0.8–1.4)
L. paeta	1.5 (1.3–1.7)	1.4 (1.1–1.7)	1.4 (1.0–1.8)
L. fusciceps	3.0 (2.7–3.4)	2.5 (2.0–3.0)	2.3 (1.8–3.0)
L. rufa	3.4 (3.0–3.8)	2.4 (2.0–2.9)	2.1 (1.6–2.7)
L. reticulatus	1.6 (1.5–2.1)	2.3 (1.8–2.9)	2.5 (1.8–3.5)
L. decolor	1.9 (1.6–2.2)	2.5 (2.0–3.2)	2.8 (2.1–3.9)
L. entomophila	7.7 (6.4–9.3)	8.6 (6.8–10.6)	8.9 (6.5–12.4)
72-h (*L. reticulatus*)			
L. fusciceps	1.4 (0.9–2.0)	1.3 (1.0–1.6)	1.2 (0.8–1.9)
L. obscura	1.3 (0.9–2.0)	1.5 (1.1–1.9)	1.5 (1.0–2.3)
L. rufa	2.6 (1.8–3.7)	2.1 (1.7–2.6)	1.9 (1.3–2.8)
L. paeta	1.3 (0.8–2.1)	2.1 (1.6–2.7)	2.5 (1.6–3.9)
L. bostrychophila	1.1 (0.7–1.9)	1.8 (1.4–2.4)	2.3 (1.4–3.6)
L. entomophila	4.6 (3.1–6.7)	8.1 (6.3–10.4)	10.2 (6.3–15.4)
L. decolor	1.8 (1.1–2.8)	5.6 (4.1–7.6)	8.9 (5.3–15.2)

[a]A 95% confidence interval (CI) that includes one in any comparison means that the two lethal concentrations are not significantly different from each other at a specified fumigation period

Table 4. Comparison of discriminating doses of susceptible adult psocids of eight species based on 20-h and 72-h exposure to ranges of phosphine gas concentrations at 25 ± 1°C and 75 ± 5% RH. Phosphine concentration reduction indexes (PCRIs) were estimated for the eight species. Concentrations are in parts per million (ppm).

Species	Exposure period		PCRI[a]
	20-h	72-h	
L. decolor	249.76	194.50	1.28
L. bostrychophila	77.45	49.09	1.58
L. obscura	65.59	23.20	2.83
L. paeta	124.71	39.64	3.15
L. entomophila	697.29	157.14	4.44
L. rufa	159.90	25.49	6.27
L. fusciceps	149.36	19.19	7.78
L. reticulatus	204.98	18.05	11.36

[a] Phosphine concentration reduction indexes (PCRIs) were proportions of discriminating doses (upper limit of 95% CI of LC_{99} value in parts per million) of 20-h to 72-h exposure period of the related tested species.

CHAPTER IV

FUNCTIONAL RESPONSES OF PREDATORY MITES, *CHEYLETUS ERUDITUS* (SCHRANK) AND *CHEYLETUS MALACCENSIS* OUDEMANS (TROMBIDIFORMES: CHEYLETIDAE) TO DIFFERENT LIFE STAGES OF *LIPOSCELIS DECOLOR* (PEARMAN) (PSOCODEA: LIPOSCELIDIDAE)

(To be submitted to Journal of Economic Entomology)

Abstract: Psocids (Psocodea: Liposcelididae) are stored-product pests of substance that are not easily managed by standard management tools and practices, such as insecticides. Biological control can be an effective alternative or complement to pesticides in the management of psocids. This study was carried out to evaluate the efficiency of two predatory mite species, *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) to manage *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae), a key stored product psocid pest species that is phosphine tolerant. The functional responses of these two cheyletid mites to nymphs and adult males and adult females of *L. decolor* was determined under laboratory conditions at 24 ± 1°C and 0: 24 (L: D) photoperiod. Based on the results of maximum likelihood estimates (MLE) of a logistic regression analysis, the functional responses of the two predatory mites to nymphs, adult males or adult females of *L. decolor* were Holling Type II. Subsequent estimations of attack rate (a), handling time (T_h), maximum predation (K), and predation efficiency (η) per day (d) using a linearly transformed Hollings Type II model for predators against prey life stages revealed that *C. eruditus* performance was preferable compared to that of *C. malaccensis* based on the aforementioned parameters. Similarly, the per capita consumption rate and searching efficiency of *C. eruditus* were considerably higher compared to *C. malaccensis* for all life stages of *L. decolor*. Nevertheless, further studies are required to validate the efficiency of both predatory mites based on other evaluation criteria including field trials, and their compatibility with other management strategies targeted at stored product pests in order to facilitate the incorporation of these mite species into existing IPM systems for managing psocids.

Key words: psocid, biological control, stored product, pest management, *Cheyletus*

Introduction

Psocids (Psocodea: Liposcelididae) are stored-product pests of substance that are difficult to manage even with the most potent pesticides, including phosphine (hydrogen phosphide: PH_3) which is a key component in integrated pest management (IPM) of insect pests in the US stored grain system (Opit et al. 2012, Nayak et al. 2014, Gautam et al. 2016). Biological control is a tool in IPM programs worldwide and can be an effective alternative or complement to pesticides (Wu et al. 2016, Fathipour and Maleknia 2016). Bioagents such as parasitoids (Hymenoptera), endoparasites (cestodes, fungi, gregarines, and nematodes), and ectoparasites (Acari) are known to be important natural regulators of stored-grain insect pest populations, including psocids (Mockford 1993, Lord and Howard 2004, Nayak et al. 2005, Villalobos et al. 2005). Acarine mites in the family Cheyletidae, such as *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae), occur naturally in stored grain (Fain and Bochkov 2001, Eliopoulos et al. 2003, Lukas et al. 2007). Studies have showed that they occur naturally in various commodities and in different types of storage facilities, feeding on both mites and moth eggs (Athanassiou et al. 2003, 2011). *Cheyletus eruditus* and *C. malaccensis* are generalist predators that prey on stored product mites of the families Acaridae and Glycyphagidae (Astigmata) including *Acarus siro* Linnaeus (Sarcoptiformes: Acaridae), *Aleuroglyphus ovatus* Troupeau (Sarcoptiformes: Acaridae), *Lepidoglyphus destructor* Schrank (Oribatida: Glycyphagidae), *Tyrophagus putrescentiae* Schrank (Sarcoptiformes: Acaridae), *Caloglyphus rodriguezi* Samsinak (Sarcoptiformes: Acaridae), and *Rhizoglyphus echinopus* Fumouze and Robin (Sarcoptiformes: Acaridae) (Yousef et al. 1982, Cebolla et al. 2009, Al-Shammery 2014, Zhu et al. 2019). There is evidence of

Cheyletus spp. attacking non-mite prey. *Cheyletus malaccensis* was reported to feed on eggs of the moths *Corcyra cephalonica* Stainton (Lepidoptera: Pyralidae) (Nangia et al. 1995) and *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) (Athanassiou and Palyvos 2006) and, on the thrip *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae) (Sengonca et al. 2004*). Cheyletus eruditus* and related species have been found to consume eggs of the beetles *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) (Rizk et al. 1979) and *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae) (Asanov 1980), and prey on all life stages of the psocid *Liposcelis decolor* (Psocodea: Liposcelididae) (Kucerova 2004). In addition to their broad nutrition ecology, *C. eruditus* and *C. malaccensis* can naturally penetrate bulk grain, reproduce parthenogenetically, have all developmental stages being predators (do not damage stored products), are cannibalistic (can survive in the absence of prey), and have adopted to a wide range of storage physical conditions (Schöller 2006). These characteristics provide potential for both predatory mites to be used as biocontrol agents against storage pests (Hubert et al. 2006).

In the United States, bioagents such as *Anisopteromalus calandrae (Howard) (Hymenoptera: Pteromalidae) , Bracon hebetor Say (Hymenoptera: Braconidae), Cephalonomia waterstoni (Gahan) (Hymenoptera: Bethylidae), Dibrachys sp. (Hymenoptera: Pteromalidae) Dufuriellus ater (Dufour) (Hemiptera: Antocoridae), Habrocytus thyridopterigis Howard (Hymenoptera: Pteromalidae), Holepyris sp. (Hymenoptera: Bethylidae), Laelius sp. Ashmead (Hymenoptera: Bethylidae), Lariophagus distinguendus (Forster) (Hymenoptera: Pteromalidae), Mesostenus sp. Gravenhorst (Hymenoptera: Ichneumonidae), Pteromalus sp. Swederus (Hymenoptera:*

Pteromalidae), Theocolax elegans (Westwood) (Hymenoptera: Pteromalidae), Trichogramma evanescens Westwood (Hymenoptera: Trichogrammatidae), Venturia canescens (Gravenhorst) (Hymenoptera: Ichneumonidae), and *Xylocoris flavipes (Reuter) (Hemiptera: Anthocoridae)* have been approved for use against stored-product insect pests (Hagstrum and Subramanyam 2006). The release of efficient bioagents is a fundamental step in the successful implementation of biocontrol programs. Before using a biocontrol agent in an IPM system, it is essential to know about the efficiency of the bioagent (Fathipour et al. 2006). Adequate knowledge about predatory characteristics helps to understand the influence of predators on the population dynamics of prey and their influence on the structure of the storage communities as a whole (Jervis and Kidd 1996).

Foraging behavior of predators such as functional and numerical responses are the primary components for the selection of predatory mites for biocontrol programs and for the evaluation of their performance after release (Fathipour and Maleknia 2016). Among foraging behaviors, functional response which describes the relationship between the numbers of prey attacked and prey densities remains a key component in the selection of natural enemies for biological control (Holling 1959a, 1959b). Functional response gives information on the practical suppression ability of a predator in its niche (Lester and Harmsen 2002). Thus, functional response is a major component of population modeling, and it has been used to predict the mechanisms underlying predator-prey behavior to improve the practical predictive potential of predator candidates in biological control programs (Sepúlveda and Carrillo 2008). To date, there is no published study on the predatory characteristics of mites in the genus *Cheyletus* in managing stored product

pests in the United States. However, adequate knowledge about a predator's functional response such as its attack rate, handling time, searching efficiency, effective predation, and resource preference is necessary for the selection of high-quality biocontrol agents for IPM. Considering this, the current study was conducted to assess the functional responses of two cheyletid mites *C. eruditus* and *C. malaccensis* to different life stages and densities of the psocid *L. decolor* in order to facilitate selection of efficient biocontrol candidates for IPM of psocids.

Materials and Methods

Predatory Mites. Laboratory stock cultures of *Acarus siro, Cheyletus eruditus*, and *Cheyletus malaccensis* were obtained from the Academy of State Administration of Grain, Beijing, China. *Acarus siro* was reared on a culture media consisting of a mixture of a wheat meal, oat flakes, and dried yeast (5:5:1) (wt/wt) and this mixture was milled (sieved) to a thickness of ~0.5 mm (hereafter referred to as mite diet) by sieving using a sieve with Tyler equivalent 35 mesh size (U.S. Standard #40 sieve (0.419-mm openings; Central Scientific Company., Chicago, IL). The *A. siro* cultures were started using ~500–1000 individuals in glass canning jars (360-ml) that had mite-proof lids and contained 200 g of mite diet in each. Jars were kept in plastic boxes (42 x 29 x 24 cm high) painted black with saturated KCl solution (Potassium Chloride, anhydrous, free-flowing, Redi-Dri^{Tm}, ACS reagent, ≥99%, 746436-2.5KG, Sigma-Aldrich, Inc.) beneath perforated false floors to maintain an RH of 85 ± 5% (RH was monitored by HOBO data loggers (Onset Computers, Bourne, MA, USA) to facilitate population increase within 2–3 weeks and

these mites were used as diet in starting and maintaining colonies of *C. eruditus* and *C. malaccensis*.

Laboratory cultures of *C. eruditus* and *C. malaccensis* originally obtained from Jiangxi and Harbin Provinces, China, respectively, were used for this study. Rearing methods for the two predatory mites were similar. Therefore, both species were cultured on *A. siro* but as pure cultures in 500-ml paper bags (lunch bags, chromated copper arsenate (cca), Albertson's Inc., Boise, ID 83726) which contained 200 g of lettuce seeds. The lettuce seeds were cleaned by sieving using a sieve of Tyler equivalent 20 mesh size (U.S. Standard #20 sieve (0.841-mm openings; Fisher Scientific Company, Pittsburgh, PA) and dried for two hours at 80 ± 1°C to prevent possible contamination by micro-arthropods and other unwanted organisms. Paper bags with lettuce seeds were placed on plates in 250-mm diameter desiccators (Thermo Fisher Scientific™ Nalgene™ Polypropylene Desiccator with Stopcock, 53100250, Pittsburgh, PA 15275) which had saturated KCl solution beneath perforated plates to maintain 85 ± 5% RH within the paper bags. After a week, ~1000–2000 individuals of *A. siro* were added, bags gently shaken, and the paper bags folded. The desiccators with paper bags were incubated at 24 ± 1°C and 0: 24 (L: D) photoperiod. After 24 hours, 25–50 individuals of *C. eruditus* or *C. malaccensis* were added to the contents in each paper bag and maintained for 3-4 weeks in order for the populations of predatory mites to increase. Adult female predatory mites were selected and used for this study. Given that both predatory mites are cannibalistic, cultures were frequently monitored and *A. siro* were added biweekly to prevent decline in predatory mite populations because of starvation or conspecific predation in paper bags.

Liposcelis decolor. *Liposcelis decolor*, used as prey, was reared on a mixture of 93% cracked wheat (*Triticum aestivum* L.) (Duster variety), 5% Rice Krispies (Kellogg North America Company, Battle Creek, MI), and 2% wheat germ (The Quaker Oats Company, Chicago, IL) (wt/wt; referred to as psocid diet hereafter) in 360-ml glass canning jars with mite-proof lids (Opit and Throne 2008). The top one-third of the inner surface of each jar was coated with Fluon (polytetrafluoroethylene; Northern Products, Woonsocket, RI) to prevent psocids from accessing and gathering on the inside of the lid. Cultures of *L. decolor* were placed inside a growth chamber maintained at 30 ± 1°C and 0: 24 (L: D) photoperiod. Culture jars were placed in plastic boxes (42 x 29 x 24 cm high) painted black which had saturated NaCl solution (Sodium Chloride, anhydrous, free-flowing, Redi-Dri^{Tm}, ACS reagent, ≥ 99%, 746398-2.5KG, Sigma-Aldrich, Inc.) beneath perforated false floors to maintain a 75 ± 5% RH. Subsequently, nymphs, adult males, and adult females from established cultures were selected and used for this study.

Experimental Arenas. Experimental arenas consisted of two 3.5-cm diameter Petri dishes (forming a total cylindrical surface area of 41.23 cm^2; a total migration area for a predator in a closed cylinder) (35 x 10 mm Style Polystyrene, Falcon®, Becton Dickinson and Company, Franklin Lakes, NJ, USA) with one serving as a base and the other as a lid were used for this study. The top one-third of the inner surface of each basal Petri dish was coated with Fluon to confine prey within the bottom portion of the arena (thus, prey were located within a 2/3 portion of the basal cylindrical arena with an area of 16.88 cm^2; a total migration area of individual prey), and a second Petri dish (a lid) was inverted over the basal Petri dish to prevent the escape of predatory mites (by observation, *Cheyletus* spp. generally prefer a refuge to hide and feed on prey, but may

escape from arenas without lids). The top and bottom Petri dishes were held together with sticky tape along 90% of the circumference of the meeting point of the basal and the upper Petri dishes. The 10% of the circumference not taped permitted air movement in and out of the basal Petri dishes. Each basal Petri dish contained a mixture of 0.2 g of red colored diet (Opit and Throne 2008) and cracked wheat (9:1 wt/wt). The colored diet enhanced easily identification of predators and prey whereas cracked wheat was food for *L. decolor* (nymphs, adult males, or adult females).

Functional Responses of *C. eruditus* and *C. malaccensis*. Adult females that had freshly molted from tritonymph (3-7-day-old) were selected from pure cultures of *C. eruditus* or *C. malaccensis* and these were starved for 24 h prior to being placed in arenas containing their prey, nymphs, adult males, or adult females of *L. decolor* at varying densities. Starvation decreases oviposition, standardizes their level of hunger, and initiates a nomadic period (Opit et al. 1997, Gerson et al. 2003, Kucerova 2004). Experimental arenas containing 0.2 g of mixed colored diet and cracked wheat and a female of either *C. eruditus* or *C. malaccensis* were exposed to different densities of the specific prey items. Prey densities of 3, 6, 12, 18, 24, 30, 40, or 50 were transferred into arenas using a moist camel brush under a stereomicroscope. Each prey density was replicated six times for the different prey stages used in the determination of functional response of *C. eruditus* or *C. malaccensis*. The six replications for each prey stage were run in two batches of 3 replicates. For each three replicates both predatory mites were simultaneously evaluated to reduce temporal variations between the functional responses of predatory mites to prey stages and densities. Thus, 2-predators x 3-prey stages x 8-prey densities x 3-replications were run concurrently in 2-batches to achieve a total of 6-

replications for each prey density. For each replication set consisting of 144 experimental arenas, arenas were arranged randomly in plastic boxes (42 x 29 x 24 cm high) painted black, which has saturated KCl solution beneath perforated false floors to maintain 85 ± 5% RH and these were kept inside a growth chamber maintained at 24 ± 1°C and 0: 24 (L: D) photoperiod. Arenas were assessed at 24 h intervals to count for the number of prey attacked and killed by predatory mites under a stereomicroscope, and cadavers were removed and replaced with equal numbers of prey killed. Eggs of predators and prey (in cases where female *L. decolor* was used as prey) were also removed along with cadavers to prevent predators using the eggs as a diet. For each prey stage, a total of 48 females of *C. eruditus* or *C. malaccensis* were assessed. Each experiment spanned 168 h and seven-data sets per observation (single female *C. eruditus* or *C. malaccensis*) were made by 24-h intervals. Thus, for each predator and prey stage, a total of 336 observations were recorded after the experimental duration (168 h). It was assumed that the average number of cadavers at the end of the seventh consecutive data represented the daily prey consumption rate of each female *C. eruditus* or *C. malaccensis* in their confined arenas at a specified prey density.

Statistical Analysis. The data were analyzed using SAS software Version 9.4 (SAS Institute, Cary, NC, USA) in two steps. First, the logistic procedure (PROC LOGISTIC) in SAS was used to modeled a logistic regression of the proportion of prey consumed (N_a/N) as a function of initial prey density (N) in order to determine the shape of the functional response curves of predators (*C. eruditus* and *C malaccensis*) to different prey stages (female, male, and nymph of *L. decolor*). That is, $N_a/N= exp\ (P_o + P_1 N + P_2 N^2 + P_3 N^3) / 1 + exp\ (P_o + P_1 N + P_2 N^2 + P_3 N^3)$, where N_a is the number of

prey consumed (events) and N is the initial prey density (trials); both were the response variables. The model (Na/N) (the probability of prey consumption) distribution was binary with logit link function, and the optimization techniques was Fisher's scoring. The P_0, P_1, P_2, and P_3 are the maximum likelihood estimates (MLE) of the intercept, linear, quadratic, and cubic coefficients, respectively. The signs of P_1 and P_2 were used to determine the type of functional response (Juliano 2001). The predator displays a Type II functional response when the linear coefficient is significantly negative ($P_1 < 0$), which indicates that the proportion of prey consumed declines monotonically with the initial prey density. When the linear coefficient is positive ($P_1 > 0$), and the quadratic coefficient is negative ($P_2 < 0$), the predator has a Type III functional response (Juliano 2001). The significantly negative coefficients of P_1 in MLE confirmed a Type II functional response of predatory mites to the subjected prey life stages of *L. decolor* (Table 5 in results section).

Subsequently, a Type II functional response curves were fitted for the observed number of prey killed per capita per day using the Holling (1959a, 1959b) disc equation: $N_a = aTN/(1+aT_hN)$. In this model, N_a is the number of preys killed, N is the initial density of prey, T is the time available for searching during the experiment, a and T_h are the rate of successful attacks and the time required to handle prey items, respectively. A linear transformation of Hollings disc equation ($1/N_a = 1/a*NT + T_h/T$) (Livdahl and Stiven 1883) was used to estimate the parameters a and T_h using a non-linear regression technique (The Reg. Procedure, SAS Institute, Cary, NC, USA) where $1/N_a$ was regressed on $1/N$. The reciprocal of the slope of the fitted line by least squares, and the time of exposure (T = 1 day) multiplied by the y-intercept was the attack rate and the

handling time, respectively (Livdahl and Stiven 1883, Rahman et al. 2012, Yao et al. 2014).

The mean prey killed, N_a, and the searching efficiency, N_a/N, were compared across the three prey stages (nymph, male, and female) and eight prey densities (3, 6, 12, 18, 24, 30, 40, and 50) using generalized linear mixed models methods for each of the predator species (*C. eruditus* and *C. malaccensis*). PROC GLIMMIX modeled the fixed effects of prey stage and prey density and their interaction for each of the response variables with the specified response distribution (N_a ~ Poisson and N_a/N ~ exponential) in SAS. Least squares means were compared for the appropriate significant effects. All tests were conducted at the nominal 0.05 level of significance.

Results

Functional response curves. The estimated parameters by logistic regression of the proportion of prey consumed (N_a/N) as a function of initial prey density (N) per day is shown in Table 5. The significantly negative coefficients of P_1 in MLE confirmed a Type II functional response of predatory mites to the subjected prey life stages of *L. decolor* (Table 5). Results of *Cheyletus eruditus* showed a significantly ($p < 0.05$) negative linear coefficient ($P_1 < 0$) and positive quadratic coefficient ($P_2 > 0$) for all prey stages, indicating that the percentage of prey consumed by *C. eruditus* for each prey stage declined monotonically as prey density increased (Table 5). Thus, the predator *C. eruditus* showed a Type II functional response to all motile life stages of *L. decolor* (Table 5, Fig. 1A, 1B, and 1C). Likewise, *Cheyletus malaccensis* showed a significant (*p*

< 0.05) negative linear coefficient ($P_1 < 0$) and positive quadratic coefficient ($P_2 > 0$) for all prey stages tested, signifying that the proportion of prey consumed for each prey stage decreased predictably as prey density increased (Table 5). This indicated that the fitted curves for *C. malaccensis* (Fig. 1D, 1E, and 1F) configured a Type II functional response to nymphs, adult females, and adult males of *L. decolor*.

Holling Type II functional response models for fitted curves. A Type II functional response model (a cyrtoid curve rising at a decreasing rate to a plateau with increasing prey density) presented a good fit to observed data for all prey stages and predators as indicated by the *p*-values of *Chi-square* (χ^2) and the coefficient of determination estimates R^2 (Table 6 and Figure 1). Strong R^2 values were noted in *C. eruditus* compared to the weak estimates of *C. malaccensis* across different prey stages. This can be attributed to prolonged culture of predatory mites which can genetically change the effectiveness of predation of *C. malaccensis*. The *p*-value ($p < 0.05$) of χ^2 suggests that the observed and predicted number of prey killed by a predator was significantly different, and that predator's predation rate was substantially influenced by increased in prey density (Figure 1). However, differences in the rate of response by *C. eruditus* and *C. malaccensis* to individual life stages of *L. decolor* were noted (Table 6, Table 7, and Fig. 1). Attack rate coefficients (*a*) (the predator's rate of successful search or the probability of capture for each prey while the predator is searching) varied significantly ($p < 0.05$) between predators and life stages of *L. decolor*. Interestingly, the highest attack rate of *C. eruditus* (0.696 d^{-1}) was found on adult females, and this was 1.16 and 1.05x more than when presented with nymphs and adult males of *L. decolor*, respectively. However, the highest attack rate of *C malaccensis* (0.643 d^{-1}) was found on

nymphs and this was 3.85 and 1.51x greater than when offered with adult female and male prey, respectively. In general, the attack rate coefficient (mean of combined prey stages) for *C. eruditus* (0.64 d^{-1}) was considerably higher than *C. malaccensis* (0.41 d^{-1}). In addition, the attack rate of *C. eruditus* increased with an increase in prey size, however, for *C. malaccensis*, the attack rate was inversely proportional to prey size (Table 6 and Fig. 1). Handling time (T_h) increased significantly ($p < 0.05$) as prey size increased for both predators (Table 6). The estimated handling time for *C. eruditus* was much shorter on nymphal stage (0.07 d) than adult female and male stages of *L. decolor* (0.16 and 0.08 d, respectively). Similarly, *C. malaccensis* spent the highest amount of time (0.27 d) on adult female prey, approximately, 1.83x more than time required to handle the nymphal stage of prey (0.146 d) (Table 6). Generally, *C. eruditus* had a shorter handling time than *C. malaccensis* when offered *L. decolor* as prey, 0.104 d and 0.196 d, respectively (mean of combined prey stages) (Table 6).

Cheyletus eruditus consumed higher numbers of *L. decolor* daily than *C. malaccensis* (Table 6, Table 8, and Fig. 1). The maximum predation rate (K) on adult females, adult males, and nymphal stage of *L. decolor* was 6.21, 11.91, and 14.71 prey.d^{-1} for *C. eruditus*, and 3.75, 5.68, and 6.849 prey.d^{-1} for *C. malaccensis*, respectively (Table 6). Similar to maximum predation rate (K), predation efficiency (η) increased significantly with a decrease in prey size. Predation efficiency of *C. eruditus* was significantly higher on nymphs (8.79 prey.d^{-1}) and this was approximately 2.03 and 1.18x higher than when adult female and male prey, respectively, were offered. For *C. malaccensis*, predation efficiency on nymphs (4.40 prey.d^{-1}) was computed as 7.04 and 1.82x greater than that for adult females and males of *L. decolor*, respectively (Table 6).

In general, maximum predation rate and predation efficiency were approximately, 2.02 and 2.78x higher, respectively, for *C. eruditus* than *C. malaccensis* when preying on *L. decolor* (mean of combined prey stages) (Table 6).

Per capita predation of *C. eruditus* and *C. malaccensis*. The results of the per capita prey consumption rate of *C. eruditus* and *C. malaccensis* showed no significant interactions between prey stage and prey density ($p > 0.05$) However, the main effects of prey stage and prey density were significant ($p < 0.05$) for both predatory mites, indicating significant differences in functional responses of predators to different life stages of *L. decolor* (Table 8). Thus, an incremental prey consumption rate was noted, however, at a decreased trend to the asymptote where it levels off (Table 8 and Fig. 1). For a combined data of *C. eruditus* and *C. malaccensis*, there was no significant interaction between predator species and prey stage for per capita predation rate (F= 2.92, DF= 2, 281, p =0.0555). The main effects of predator species (F= 35.18, DF= 1, 281, p = 0.0001) and prey stage (F= 37.76, DF= 2, 281, p = 0.0001) were significant. The consumption rate of predatory mites was considerably higher on nymphs (4.71 prey.d^{-1}) and adult males (4.22 prey.d^{-1}) but, lowest on adult females (2.51 prey.d^{-1}). Typically, per capita predation rate of *C. eruditus* was substantially higher (4.37 prey.d^{-1}) and approximately 1.41x more than that of *C. malaccensis* (3.10 prey.d^{-1}) when *L. decolor* was consumed (mean across prey densities and prey stages) (Table 8).

Per capita searching efficiency of *C. eruditus* and *C. malaccensis*. Distinct differences in searching efficiency were noted as a result of different prey stage, prey density, and predator species. The interaction between prey stage and prey density in relation to the per capita searching efficiency of *C. eruditus* was not significant ($p > 0.05$)

(Table 7). Also, the main effect of prey stage was not significant *($p > 0.05$)*, however, the main effects of prey density was significantly different ($p < 0.05$) (Table 7). Searching efficiency was higher at lower prey densities for both predatory mites (Table 9). In relation to *C. malaccensis*, there was no significant interaction between prey stage and prey density ($p > 0.05$) (Table 7). However, the main effects of prey stage and prey density were significant ($p < 0.05$) (Table 9). Searching efficiency was highest on nymphal and adult male stages of prey, and at the lowest prey density (Table 9). For a combined data of *C. eruditus* and *C. malaccensis*, the interaction between predator species and prey stage was not significantly different (F= 1.51, DF= 2, 282, $p = 0.2221$) for per capita searching efficiency. However, the main effects of predator species (F= 9.98, DF= 1, 282, $p = 0.0018$) and prey stage (F= 10.74, DF= 2, 282, $p = 0.0001$) were significant. The searching efficiency of predatory mites at a given prey density was considerably higher on nymphs (0.33) and adult males (0.30) but, lowest on adult females (0.18). This suggest that searching efficiency related directly to prey density, and inversely related to prey size given that the largest prey size was an adult female and the smallest, the nymphal stage of *L. decolor*. Generally, searching efficiency of *C. eruditus* was significantly higher (0.31) and approximately 1.48x greater than *C. malaccensis* (0.21) when *L. decolor* was consumed (mean across prey densities and prey stages) (Table 9).

Discussion

Cheyletus species are frequently found to be associated with pestiferous arthropods in storage ecosystems (Athanassiou and Palyvos 2015). The natural

occurrence of *Cheyletus* spp. and their apparent interactions with several insect and mite species in stored commodities indicates that these predatory mites are potential natural enemies with a broad prey range (Cebolla et al. 2009, Athanassiou et al. 2011). However, little quantitative information was available on cheyletid predatory mites and their capacity for managing insect pests in stored products until the present study (Haines 1998, Kucerova 2004, Athanassiou and Palyvos 2015). Based on this study, *C. eruditus* and *C. malaccensis* have been showed to prey on all mobile life stages (nymphs, adult males, and adult females) of *L. decolor*. Earlier findings by Hughes (1976), Roesli et al. (1999), Kucerova (2004), Pascual-Villalobos (2005), and Athanassiou et al. (2011) had showed that cheyletids were predatory mites with good potential for managing micro-arthropods in various commodities and in different types of storage facilities under tropical and temperate conditions.

Functional response facilitates the identification of the density at which a targeted pest would escape control. Thus, it signifies the influence of a predator on the dynamics of a pest population (O'Neil 1990). Type II functional response of *C. eruditus* and *C. malaccensis* to nymphs, and adult males, and adult females of *L. decolor* indicates that these predators increase their prey consumption with increasing prey availability to a maximum (K), after which prey consumption decreases monotonically (Holling 1959 b). The maximum predation rate is limited by total available time for searching and a predator's handling time (Hassell 1978), however, the latter varies substantially across prey stages and influences the nature of the response of predators to different stages of *L. decolor*. Xia et al. (2007) and Zhu et al. (2019) reported similar predatory responses (Type II functional response) for *C. eruditus* and *C. malaccensis* to *Aleuroglyphus*

ovatus. Again, these findings were consistent with the type of functional response displayed by many other predatory mites, such as *Neoseiulus californicus* on *Tetranychus urticae* Koch and *N. barkeri* Hughes preying on *A. ovatus* (Holling 1959 b, Li et al. 2008, Ahn et al. 2010). Predatory arthropods exhibit both Type II and Type III functional responses, but Type II is commonly observed in many predators that have been released as a biocontrol agents (Cedola et al. 2001). The reason for an increase in predation with increase prey densities may be associated with the disturbance of predators while feeding that results in the wasteful or defensive killing of prey. The wasteful killing of prey increased with higher densities of prey because predators may only partially consume each attacked prey. This is an ecological mechanism by which predators may suppress pest population below certain thresholds of pest known to cause economic loss (Sandness and McMurtry 1970, Metz et al 1988, Zhu et al. 2019).

Attack rate and handling time regulate the magnitude of functional responses of predators (Pervez and Omkar 2006). These are important indicators of prey consumption rate and predator efficiency (Bazgir et al. 2020), and this was apparent in the present study for *C. eruditus* and *C. malaccensis*. The attack rate of *C. eruditus* increased with an increase in prey size, however, for *C. malaccensis*, this was inversely related to prey size. The attack rate coefficient for *C. eruditus* was considerably higher than for *C. malaccensis*. The handling time for both predators was negatively correlated with prey size. Overall, *C. eruditus* had a shorter handling time than *C. malaccensis* when offered *L. decolor* as prey. These trends indicate that *C. eruditus* is a better predator than *C. malaccensis*, and implies that *C. eruditus* would be more efficient in managing psocids in storage communities than *C. malaccensis*. Nonetheless, different factors were reported to

affect attack rate and handling time of biocontrol agents, and these include prey movement, predator speed, predator species and prey stages, temperature, relative humidity, and the time spent subduing individual prey, which could be related to behavioral and structural defense mechanisms of prey (Hassell 1978, Ali et al. 2011, Bazgir et al. 2020). However, Zhu et al. (2019) suggested that the estimates for an attack rate and handling time in laboratory stimulation of predator-prey interactions should be used only for comparison purposes because of the assumption that the individuals in the trials were always searching, attacking, and consuming prey. Such values may be overestimated and may not represent the predatory capacity of predators in their natural environment. Nevertheless, the maximum predation rate and predation efficiency were still higher for *C. eruditus* than *C. malaccensis* and the former could be inundatively released to manage psocids in storage environment.

Cheyletus eruditus and *C. malaccensis* attacked and killed more nymphs than adult males and adult females of *L. decolor*, however, the consumption rate was consistently higher for *C. eruditus* for all prey stages tested. Based on this study, *C. eruditus* seems to have superior predatory features for searching, identification, capturing, and utilization of stored-product pest psocids. Adaptive predatory characteristics of mite predators decrease handling time and improve predation efficiency (Bazgir et al. 2020). Similar to the mechanism used by other stored-product predatory mites, *C. eruditus* and *C. malaccensis* insert the chelicerae in the prey cuticle, immediately inject saliva that paralyzes it, and then suck body fluids of the immobilized prey (Yoshikawa 1985, Athanassiou and Palyvos 2006). The preference of *C. eruditus* and *C. malaccensis* for immature stages of prey has previously been reported (Kucerova

2004, Zhu et al. 2019), and this is an important characteristic of effective predatory mites for preventing the proliferation of pest populations (Rahman et al. 2012).

Our results revealed that prey size correlated strongly with the consumption rate and resource preference of the predators. The consumption rate of predators was inversely related to the size of the prey life stages. *Cheyletus malaccensis* has been showed to have positive preference for larvae and nymphs but negative preference for adults and eggs of *A. ovatus* (Zhu et al. 2019). The estimated mean size of the eggs, males, and females of *L. decolor* were 0.37, 0.9, and 1.25 mm, respectively (Kucerova 2002, Gunther 1974). Judging by these estimates, predators may require more time and energy to capture and subdue female prey relative to nymphs and adult males of *L. decolor*. Despite the report that the developmental stage of the prey does not affect the development of *Cheyletus* spp. (Schöller and Žd'árková 2006), the female predators may need more nymphs (a nutritionally poor stage of prey) to balance energy cost and enhance oviposition (Ganjisaffar and Perring 2015). Similarly, the per capita searching efficiency of predators decreased with increasing prey size, however, the decline was steeper in *C. eruditus* than in *C. malaccensis* which had a gradual but a sporadic trend, implying that *C. malaccensis* may be consistent in keeping the dynamics of the pest population compared to *C. eruditus*. The observed higher searching efficiency of *C. eruditus* indicates that this predator would be more efficient at regulating increasing densities of all developmental stages of *L. decolor* than *C. malaccensis*. However, Thind and Ford (2006) and Pekar and Hubert (2008) suggested that since there are no remarkable differences in prey specificity or nutritional requirements among *Cheyletus*

spp. one can expect similar biocontrol characteristic when these are deployed as biocontrol agents solely or in IPM systems in storage communities.

In conclusion, to our knowledge, this is the first study to provide quantitative data demonstrating the biological control potential of *C. eruditus* and *C. malaccensis* for managing psocids in laboratory simulated storage conditions. Based on the results of this study, *C. eruditus* performed better than *C. malaccensis* in relation to most of the parameters assessed. Although functional response studies under laboratory conditions provide some insight into predator-prey interaction, this has been criticized for ignoring the environmental complexities and multispecies predator and prey systems that occur in the field. Therefore, further studies are required to validate the effectiveness of *C. eruditus* and *C. malaccensis* based on other foraging behaviors such as numerical response, interference, prey preference, and intraguild predation. Other data that could facilitate evaluation of the effectiveness of these two predators includes life table parameters, multitrophic interactions, thermo-hydro parameters, and their compatibility with other stored-product pest management strategies. Field trials would also be required to enhance the incorporation of these mite species into existing IPM systems for managing psocids and other stored-product arthropod pests.

Acknowledgments

We thank Dr. Yi Wu, the Academy of State Administration of Grain, Beijing, China for providing the colonies of *Cheyletus eruditus*, *Cheyletus malaccensis*, and *Acarus siro* for this work. This research was funded by Oklahoma Agricultural Experiment Station (Project number OKL2949). Trade names or commercial products mentioned in this paper are solely for the purpose of providing specific information and do not imply recommendation or endorsement by Oklahoma State University.

Table 5. Maximum-likelihood estimates from logistic regressions of the proportion of *L. decolor* females, males, and nymphs consumed by *C. eruditus* or *C. malaccensis* female adults as a function of initial prey density.

Predator	Prey Stage	Parameter	Estimate	Standard Error	Chi-Square (χ^2)	*P* value	Number of observation (*n*)
C. eruditus	Female	Intercept (P_0)	-0.0945	0.1910	0.2449	0.6207	336
		Linear (P_1)	-0.0652	0.0272	5.7500	0.0165	
		Quadratic (P_2)	0.0003	0.00111	0.0731	0.7869	
		Cubic (P_3)	0.000004	0.000013	0.1121	0.7377	
	Male	Intercept (P_0)	0.5402	0.1837	8.6491	0.0033	336
		Linear (P_1)	-0.0882	0.0252	12.2119	0.0005	
		Quadratic (P_2)	0.00129	0.00101	1.6307	0.2016	
		Cubic (P_3)	-0.000008	0.000012	0.4696	0.4932	
	Nymph	Intercept (P_0)	0.4515	0.1822	6.1373	0.0137	336
		Linear (P_1)	-0.0611	0.0248	6.0758	0.0137	
		Quadratic (P_2)	0.000207	0.000983	0.0443	0.8334	
		Cubic (P_3)	0.000006	0.000011	0.2450	0.6206	
C. malaccensis	Female	Intercept (P_0)	-1.0488	0.2368	19.6238	< 0.0001	336
		Linear (P_1)	-0.0958	0.0339	7.9802	0.0047	
		Quadratic (P_2)	0.00251	0.00137	3.3442	0.0674	
		Cubic (P_3)	-0.00002	0.000016	2.0250	0.1547	
	Male	Intercept (P_0)	0.1325	0.1882	0.4954	0.4815	336
		Linear (P_1)	-0.1215	0.0264	21.2034	< 0.0001	
		Quadratic (P_2)	0.00316	0.00106	8.8839	0.0029	
		Cubic (P_3)	-0.00003	0.000012	5.7927	0.0161	
	Nymph	Intercept (P_0)	0.4367	0.1848	5.5849	0.0181	336
		Linear (P_1)	-0.1400	0.0257	29.7700	< 0.0001	

Quadratic (P_2)	0.00391	0.00103	14.4847	0.0001
Cubic (P_3)	-0.00004	0.000012	10.7294	0.0011

Table 6. Parameter estimates for Holling Type II functional responses of adult female *C. eruditus* or *C. malaccensis* to *L. decolor* at different life stages and densities predicted by a linear transformation of Hollings disc equation: $1/N_a = 1/a*NT + T_h/T$.

Predator	Prey Stage	a (d^{-1}) (95% CI)*	T_h (d) (95% CI)	K (prey.d^{-1}) (95% CI)	η (prey.d^{-1}) (95% CI)	P value	R^2 (Adjusted R^2) [n]
C. eruditus	Female	0.696a (0.638 – 0.765)	0.161a (0.145 – 0.177)	6.211b (5.650 – 6.897)	4.323b (4.322 – 4.400)	< 0.0001	0.919 (0.915) [46]
	Male	0.627a (0.572 – 0.694)	0.084b (0.063 – 0.105)	11.905a (9.524 – 15.873)	7.464a (6.610 – 9.078)	< 0.0001	0.904 (0.902) [48]
	Nymph	0.598a (0.531 – 0.685)	0.068b (0.038 – 0.097)	14.706a (10.309 – 26.316)	8.794a (7.062 – 13.973)	< 0.0001	0.845 (0.841) [48]
C. malaccensis	Female	0.167b (0.111 – 0.338)	0.267a (0.127 – 0.662)	3.745b (1.511 – 7.874)	0.626c (0.511 – 0.874)	0.0003	0.265 (0.248) [46]
	Male	0.427ab (0.305 – 0.709)	0.176ab (0.047 – 0.305)	5.682ab (3.279 – 21.277)	2.426b (2.325 – 6.489)	< 0.0001	0.357 (0.343) [48]
	Nymph	0.643a (0.501 – 0.895)	0.146ab (0.085 – 0.206)	6.849ab (4.854 – 11.765)	4.404bc (0.589 – 4.345)	< 0.0001	0.526 (0.515) [48]

Instantaneous attack rate (***a***); Handling time (***T**h*); Total time available for searching (***T***) = 1 d; Maximum predation rate (***K***) = (T/T_h); Predation efficiency (***η***) = ***a/T**h*; * None overlapping 95% confidence intervals (CI) in a column are significantly different for mean estimates of parameters.

Table 7. Summary of the tests for the fixed effects of prey stage (PS) and prey density (N) of *L. decolor* on numbers of prey killed (N_a) and searching efficiency (N_a/N) of predatory mites, for *Cheyletus eruditus* (CE) or *Cheyletus malaccensis* (CM).

Parameter	Variable	Source	Df	*F*	*P* value
CE	N_a	PS	2, 119	8.86	0.0003
		N	7, 119	13.43	< 0.0001
		PS*N	14, 119	0.10	1.0000
	N_a/N	PS	2, 120	2.48	0.0878
		N	7, 120	3.65	0.0013
		PS*N	14, 120	0.02	1.0000
CM	N_a	PS	2, 119	19.77	< 0.0001
		N	7, 119	12.79	< 0.0001
		PS*N	14, 119	0.09	1.0000
	N_a/N	PS	2, 120	9.44	0.0002
		N	7, 120	2.89	0.0079
		PS*N	14, 120	0.03	1.0000

Table 8. Numbers of prey killed (N_a) (mean ± SE) per female *C. eruditus* or *C. malaccensis* per day (d^{-1}) exposed to *L. decolor* at different developmental stages and densities.

Predator	Prey density (N)	Prey Killed per Predator (N_a) d^{-1} (95% CI)		
		Prey Stage		
		Female	Male	Nymph
C. eruditus	3	1.26 ± 0.45 aC (0.61 – 2.63)	1.64 ± 0.52 aD (0.86 – 3.13)	1.67 ± 0.53 aE (0.88 – 3.16)
	6	2.31 ± 0.62 aBC (1.34 – 3.98)	3.24 ± 0.74 aCD (2.05 – 5.12)	3.29 ± 0.74 aDE (2.08 – 5.18)
	12	3.76 ± 0.79 bAB (2.46 – 5.76)	4.83 ± 0.90 aBC (3.32 – 7.04)	5.12 ± 0.92 aCD (3.55 – 7.37)
	18	4.26 ± 0.84 bAB (2.86 – 6.36)	6.07 ± 1.00 aAB (4.34 – 8.49)	6.50 ± 1.04 aBC (4.70 – 8.99)
	24	4.43 ± 0.86 bAB (2.99 – 6.56)	6.81 ± 1.07 aAB (4.96 – 9.34)	7.41 ± 1.11 aBC (5.47 – 10.03)
	30	5.10 ± 0.92 bA (3.53 – 7.35)	7.05 ± 1.08 aAB (5.16 – 9.62)	8.05 ± 1.16 aBC (6.02 – 10.77)
	40	4.88 ± 0.90 bA (3.36 – 7.09)	7.64 ± 1.13 aAB (5.67 – 10.30)	8.33 ± 1.18 aAB (6.26 – 11.09)
	50	5.69 ± 0.97 cA (4.03 – 8.04)	7.98 ± 1.15 bA (5.95 – 10.69)	10.10 ± 1.30 aA (7.78 – 13.09)
C. malaccensis	3	0.56 ± 0.29 bE (0.20 – 1.61)	0.89 ± 0.33 aE (0.43 – 1.87)	1.42 ± 0.46 aD (0.74 – 2.73)
	6	0.80 ± 0.36 bDE (0.33 – 1.97)	1.36 ± 0.42 aE (0.73 – 2.55)	1.63 ± 0.50 aD (0.88 – 3.03)
	12	1.33 ± 0.47 bDE (0.64 – 2.74)	1.77 ± 0.50 aDE (1.00 – 3.12)	3.75 ± 0.69 aCD (1.66 – 4.56)
	18	1.58 ± 0.53 bDE (0.80 –3.12)	2.51 ± 0.63 aCD (1.51 – 4.16)	3.44 ± 0.79 aBC (2.16 – 5.49)
	24	2.07 ± 0.63 bCD (1.12 – 3.83)	3.00 ± 0.71 aBC (1.86 – 4.85)	4.35 ± 0.93 aBC (2.82 – 6.70)
	30	2.17 ± 0.65 bBC (1.19 – 3.97)	3.51 ± 0.80 aAB (2.22 – 5.56)	4.88 ± 1.01 aAB (3.22 – 7.41)
	40	2.95 ± 0.79 bAB (1.71 – 5.09)	3.79 ± 0.85 aAB (2.42 – 5.95)	5.49 ± 1.09 aAB (3.67 – 8.21)
	50	3.34 ± 0.87 bA (1.98 – 5.64)	4.40 ± 0.94 aA (2.85 – 6.79)	5.84 ± 1.14 aA (3.93 – 8.68)

Significant differences among prey stages for each prey density are denoted with different lower-case letters (within the same row) for each predator, and differences among prey densities for each prey stage are denoted by different upper-case letters (within a column) for each predator ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

Table 9. Per capita searching efficiency (N_a/N) (mean ± SE) of predatory mites, *C. eruditus* or *C. malaccensis* exposed to different developmental stages and densities of *L. decolor*.

Predator	Prey density (N)	Searching Efficiency (N_a/N) (95% CI)		
		Prey Stage		
		Female	Male	Nymph
C. eruditus	3	0.42 ± 0.17 bA (0.19 – 0.94)	0.55 ± 0.22 aA (0.24 – 1.23)	0.56 ± 0.23 aA (0.25 – 1.25)
	6	0.39 ± 0.16 bAB (0.17 – 0.86)	0.54 ± 0.22 aA (0.24 – 1.21)	0.55 ± 0.22 aA (0.24 – 1.22)
	12	0.31 ± 0.13 bBC (0.14 – 0.70)	0.40 ± 0.16 aAB (0.18 – 0.90)	0.43 ± 0.17 aA (0.19 – 0.96)
	18	0.24 ± 0.10 bBC (0.11 – 0.53)	0.34 ± 0.14 aAB (0.15 – 0.75)	0.36 ± 0.15 aA (0.16 – 0.81)
	24	0.19 ± 0.08 bBC (0.08 – 0.41)	0.28 ± 0.11 aAB (0.13 – 0.64)	0.31 ± 0.13 aA (0.14 – 0.69)
	30	0.17 ± 0.07 aBC (0.08 – 0.40)	0.24 ± 0.10 aAB (0.11 – 0.53)	0.27 ± 0.02 aA (0.12 – 0.60)
	40	0.12 ± 0.05 bBC (0.05 – 0.27)	0.19 ± 0.07 aAB (0.09 – 0.43)	0.21 ± 0.09 aA (0.09 – 0.47)
	50	0.11 ± 0.05 cC (0.05 – 0.25)	0.16 ± 0.06 bB (0.07 – 0.36)	0.20 ± 0.08 aA (0.09 – 0.45)
C. malaccensis	3	0.23 ± 0.09 cA (0.10 – 0.52)	0.50 ± 0.20 bA (0.22 – 1.12)	0.64 ± 0.25 aA (0.28 – 1.43)
	6	0.16 ± 0.07 bA (0.07 – 0.36)	0.38 ± 0.16 aAB (0.17 – 0.86)	0.37 ± 0.14 aAB (0.16 – 0.82)
	12	0.14 ± 0.06 bA (0.06 – 0.30)	0.25 ± 0.10 aAB (0.11 – 0.55)	0.31 ± 0.13 aAB (0.14 – 0.69)
	18	0.11 ± 0.04 bA (0.05 – 0.24)	0.23 ± 0.10 aAB (0.10 – 0.53)	0.26 ± 0.10 aAB (0.11 – 0.58)
	24	0.11 ± 0.04 bA (0.05 – 0.24)	0.21 ± 0.08 aAB (0.09 – 0.47)	0.24 ± 0.10 aAB (0.11 – 0.55)
	30	0.09 ± 0.04 bA (0.04 – 0.20)	0.20 ± 0.08 aAB (0.09 – 0.44)	0.22 ± 0.09 aAB (0.10 – 0.49)
	40	0.09 ± 0.04 bA (0.04 – 0.20)	0.16 ± 0.07 aAB (0.07 – 0.36)	0.18 ± 0.07 aB (0.08 – 0.41)
	50	0.08 ± 0.03 aB (0.04 – 0.18)	0.15 ± 0.06 aB (0.06 – 0.33)	0.16 ± 0.06 aB (0.07 – 0.35)

Significant differences among prey stages for each prey density are denoted with different lower-case letters (within the same row) for each predator, and differences among prey densities for each prey stage are denoted by different upper-case letters (within a column) for each predator ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

Figure Caption

Fig. 1. Functional responses of *C. eruditus* or *C. malaccensis* to different life stages (female, male, and nymph) and densities of *L. decolor*. Parameters for the fitted Holling Type II functional response curves are in Table 1 and Table 2. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* for each prey life stage.

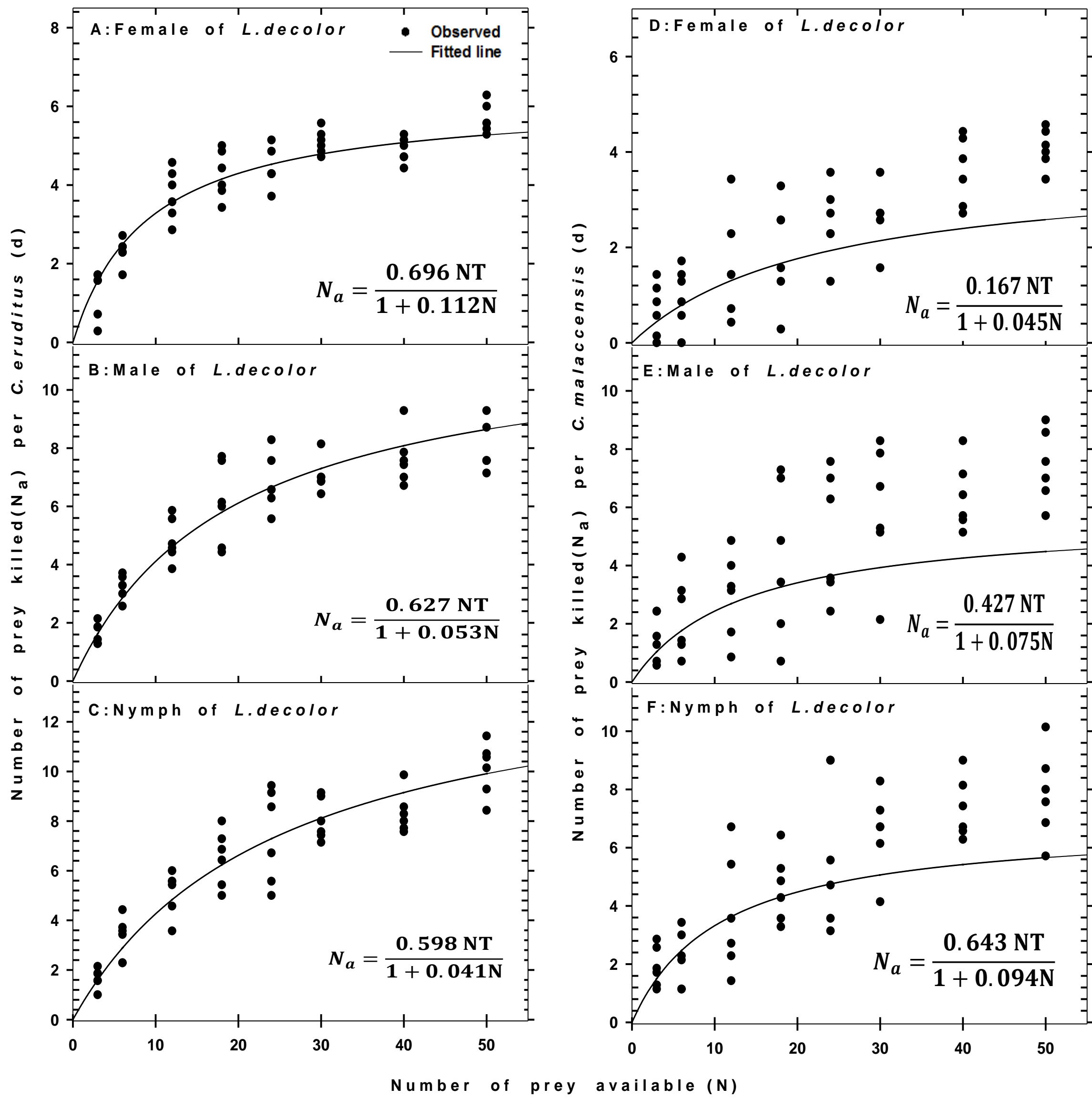

Fig. 1

CHAPTER V

NUMERICAL RESPONSES OF THE PREDATORY MITES, *CHEYLETUS ERUDITUS* (SCHRANK) AND *CHEYLETUS MALACCENSIS* OUDEMANS (TROMBIDIFORMES: CHEYLETIDAE) TO DIFFERENT LIFE STAGES OF *LIPOSCELIS DECOLOR* (PEARMAN) (PSOCODEA: LIPOSCELIDIDAE)

(To be submitted to Journal of Economic Entomology)

Abstract: Predatory mites display diverse ecological mechanisms to suppress pest population density below certain thresholds of pest known to cause economic loss. The current study explored the numerical response of the predatory mites, *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) when on a diet of *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae). The numerical responses of these two cheyletid mites to nymphs, adult males, and adult females of *L. decolor* were determined under laboratory conditions at 24 ± 1°C, 85 ± 5 RH, and 0: 24 (L: D) photoperiod. Oviposition rate, oviposition efficiency, and efficiency of conversion of ingested (ECI) food resource were the key numerical response parameters assessed. The present study revealed a general trend of a strong negative and positive correlation between oviposition rates and increase in prey densities of *C. eruditus* and *C. malaccensis*, respectively. The oviposition efficiency was mostly similar for both predatory mites and was inversely related to prey density. Moreover, the results showed that *C. eruditus* would only be more efficient and lay a substantial number of eggs (~4 eggs per predator per day) at a prey density of ~12 prey/16.88 cm^2 or less, and above which oviposition rate monotonically changed with increased in prey density. However, the oviposition rate of *C. malaccensis* increased with prey density and peaked at a predicted ~ 9 eggs per predator per day at a prey density of ~ 55 prey/16.88 cm^2. Generally, *C. malaccensis* was more efficient than *C. eruditus* in utilizing prey biomass (ECI). Given the complex nature of predator-prey interactions, we recommended further assessment of these predatory mites before being used for managing stored-product insect pests in the United States.

Key words: psocid, biological control, stored product, *Cheyletus*, pest management

Introduction

Cheyletidae are the most common and abundant family of Prostigmata found in stored products (Athanassiou and Palyvos 2015). Cheyletids are usually found in barns, granaries, and warehouses, but can also be detected in nests of birds and rodents, in association with Acari mites of the cohort Astigmatina (Hughes 1976, Athanassiou and Palyvos 2015). High fertility, fast multiplication, and parthenogenesis are desirable physiological traits exhibited by Cheyletidae, and these mites have the potentials for use as biocontrol agents against pestiferous insects and mites of stored products (Palyvos and Emmanouel 2011, Hubert et al. 2016, Zhu et al. 2019). In addition, the genus *Cheyletus* show vital adaptive characteristics such as easy penetration through intergranular spaces, broad prey preference, tolerance to wide temperature range, and are natural enemies of many economically important insect pests of stored commodities (Yousef et al. 1982, Cebolla et al. 2009, Hubert et al. 2016). *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) remain the most dominant and widely distributed cheyletid species in storage and processing facilities in temperate and tropical regions (Zdarkova 1998, Palyvos et al. 2008, Athanassious and Palyvos 2015). Both predatory mites are natural enemies of multiple arthropods and forage on mite and non-mite pests such as *Acarus siro* Linnaeus (Sarcoptiformes: Acaridae), *Aleuroglyphus ovatus* Troupeau (Sarcoptiformes: Acaridae), *Caloglyphus redickorzevi* Zachvatkin (Sarcoptiformes: Acaridae), *Lepidoglyphus destructor* Schrank (Oribatida: Glycyphagidae), *Tyrophagus putrescentiae* Schrank (Sarcoptiformes: Acaridae), *Rhizoglyphus echinopus* Fumouze and Robin (Sarcoptiformes: Acaridae) (Yousef et al. 1982, Zdarkova 1998, Al-Shammery 2014, Zhu et al. 2019), *Corcyra cephalonica*

(Stainton) (Lepidoptera: Pyralidae), *Ephestia cautella* Walker (Lepidoptera: Pyralidae), *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae), *Rhyzopertha dominica* (Fabricius) (Coleoptera: Bostrichidae), *Tribolium confusum* Jacquelin Du Val (Coleoptera: Tenebrionidae), *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae), and psocids in the genera, *Liposcelis* and *Lepinotus* (Psocodea: Liposcelididae) (Rizk et al. 1979, Asanov 1980, Nangia et al. 1995, Kucerova 2004, Athanassiou and Palyvos 2006, Cebolla et al. 2009).

Numerical response is a key component for the selection of mite predators for biocontrol programs and it gives information on the effectiveness of resource utilization, reproductive output, and colonization potentials of predators (Lester and Harmsen 2002, Rahman et al. 2012). Numerical response describes a progressive change in the number of a predator's progeny in relation to change in prey density (Solomon 1949, Fathipour and Maleknia 2016). It may be considered as a strategy by female predators to augment their offspring at different prey densities (Cédola et al. 2001). Although the prey range of *C. eruditus* and *C. malaccensis* is well established and includes pestiferous mites, and coleopteran and lepidopteran insect pests of stored products, there is little information on their influence on the population dynamics of psocids (Psocodea: Liposcelididae). Over the last three to four decades, psocids have risen to become stored product pests of substance (Phillips and Throne 2010) and are difficult to manage even with the most potent pesticides. Foraging behaviors of predators are usually affected by biotic and abiotic factors (Fathipour and Maleknia 2016). Prey related biotic factors such as prey stage, prey type, prey quality, and prey quantity can impact the ability of predators to augment their ecological niche to effectively manage pests (Xiao and Fadamiro 2010,

Seiedy et al. 2012, Zhu et al. 2019). Palyvos and Emmanuel (2011) indicated that prey type affected the life table characteristics of cheyletid mites, including their fecundity and the number of individuals that developed from eggs to adult. Considering this, the current study aimed to assess the numerical responses of the predatory mites *C. eruditus* and *C. malaccensis* to different developmental stages of *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae). Additionally, this study evaluated the efficiency of conversion of ingested food (ECI) by these two predators. The ECI pertains to the relationship between the conversion of prey biomass and prey density (Fathipour and Maleknia 2016). This study will provide baseline information for further evaluation of both predatory mites for use as biocontrol agents in stored grain IPM systems against psocid species and other related stored-product arthropod pests in the United States.

Materials and Methods

Predatory Mites. Laboratory stock cultures of *Acarus siro, Cheyletus eruditus*, and *Cheyletus malaccensis* were obtained from the Academy of State Administration of Grain, Beijing, China. *Acarus siro* was reared on a culture media consisting of a mixture of a wheat meal, oat flakes, and dried yeast (5:5:1) (wt/wt) and this mixture was milled (sieved) to a thickness of ~0.5 mm (hereafter referred to as mite diet) by sieving using a sieve with Tyler equivalent 35 mesh size (U.S. Standard #40 sieve (0.419-mm openings; Central Scientific Company., Chicago, IL). The *A. siro* cultures were started using ~500–1000 individuals in glass canning jars (360-ml) that had mite-proof lids and contained 200 g of mite diet in each. Jars were kept in plastic boxes (42 x 29 x 24 cm high) painted

black with saturated KCl solution (Potassium Chloride, anhydrous, free-flowing, Redi-Dri^{Tm}, ACS reagent, ≥ 99%, 746436-2.5KG, Sigma-Aldrich, Inc.) beneath perforated false floors to maintain an RH of 85 ± 5% (RH was monitored by HOBO data loggers (Onset Computers, Bourne, MA, USA) to facilitate population increase within 2–3 weeks and these mites were used as diet in starting and maintaining colonies of *C. eruditus* and *C. malaccensis*.

Laboratory cultures of *C. eruditus* and *C. malaccensis* originally obtained from Jiangxi and Harbin Provinces, China, respectively, were used for this study. Rearing methods for the two predatory mites were similar. Therefore, both species were cultured on *A. siro* but as pure cultures in 500-ml paper bags (lunch bags, chromated copper arsenate (cca), Albertson's Inc., Boise, ID 83726) which contained 200 g of lettuce seeds. The lettuce seeds were cleaned by sieving using a sieve of Tyler equivalent 20 mesh size (U.S. Standard #20 sieve (0.841-mm openings); Fisher Scientific Company, Pittsburgh, PA) and dried for two hours at 80 ± 1°C to prevent possible contamination by micro-arthropods and other unwanted organisms. Paper bags with lettuce seeds were placed on plates in 250-mm diameter desiccators (Thermo Fisher Scientific™ Nalgene™ Polypropylene Desiccator with Stopcock, 53100250, Pittsburgh, PA 15275) which had saturated KCl solution beneath perforated plates to maintain 85 ± 5% RH within the paper bags. After a week, ~1000–2000 individuals of *A. siro* were added and gently shaken and the paper bags folded. The desiccators with paper bags were incubated at 24 ± 1°C and 0: 24 (L: D) photoperiod. After 24 hours, 25–50 individuals of *C. eruditus* or *C. malaccensis* were added to the contents in each paper bag and maintained for 3-4 weeks in order for the populations of predatory mites to increase. Adult female predatory mites

were selected and used for this study. Given that both predatory mites are cannibalistic, cultures were frequently monitored and *A. siro* were added biweekly to prevent decline in predatory mite populations because of starvation or conspecific predation in paper bags.

Liposcelis decolor. *Liposcelis decolor*, used as prey, was reared on a mixture of 93% cracked wheat (*Triticum aestivum* L.) (Duster variety), 5% Rice Krispies (Kellogg North America Company, Battle Creek, MI), and 2% wheat germ (The Quaker Oats Company, Chicago, IL) (wt/wt; referred to as psocid diet hereafter) in 360-ml glass canning jars with mite-proof lids (Opit and Throne 2008). The top one-third of the inner surface of each jar was coated with Fluon (polytetrafluoroethylene; Northern Products, Woonsocket, RI) to prevent psocids from accessing and gathering on the inside of the lid. Cultures of *L. decolor* were placed inside a growth chamber maintained at 30 ± 1°C and 0: 24 (L: D) photoperiod. Culture jars were placed in plastic boxes (42 x 29 x 24 cm high) painted black which had saturated NaCl solution (Sodium Chloride, anhydrous, free-flowing, Redi-Dri^{Tm}, ACS reagent, ≥ 99%, 746398-2.5KG, Sigma-Aldrich, Inc.) beneath perforated false floors to maintain a 75 ± 5% RH. Subsequently, nymphs, adult males, and adult females from established cultures were selected and used for this study.

Experimental Arenas. Experimental arenas consisted of two 3.5-cm diameter Petri dishes (forming a total cylindrical surface area of 41.23 cm^2; a total migration area for a predator in a closed cylinder) (35 x 10 mm Style Polystyrene, Falcon®, Becton Dickinson and Company, Franklin Lakes, NJ, USA) with one serving as a base and the other as a lid were used for this study. The top one-third of the inner surface of each basal Petri dish was coated with Fluon to confine prey within the bottom portion of the arena (thus, prey were located within a 2/3 portion of the basal cylindrical arena with an area of

16.88 cm^2; a total migration area of prey), and a second Petri dish (a lid) was inverted over the basal Petri dish to prevent the escape of predatory mites (by observation, *Cheyletus* spp. generally prefer a refuge to hide and feed on prey, but may escape from arenas without lids). The top and bottom Petri dishes were held together with sticky tape along 90% of the circumference of the meeting point of the basal and the upper Petri dishes. The 10% of the circumference not taped permitted air movement in and out of the basal Petri dishes. Each basal Petri dish contained a mixture of 0.2 g of red colored diet (Opit and Throne 2008) and cracked wheat (9:1 wt/wt). The colored diet enhanced easily identification of predators and prey whereas cracked wheat was food for *L. decolor* (nymphs, adult males, or adult females).

Numerical Responses of *C. eruditus* and *C. malaccensis*. Adult females freshly molted from tritonymphs (3-7-day-old) were selected from pure colonies of *C. eruditus* or *C. malaccensis* and these were starved for 24 h prior to introduction to their prey (nymphs, adult males, and adult females) of *L. decolor* at varying densities. Starvation decreases initial variation in oviposition, standardizes the level of hunger, and initiates a nomadic period (Opit et al. 1997, Gerson et al. 2003, Kucerova 2004). Experimental arenas containing 0.2 g of mixed colored diet and cracked wheat, had a female of either *C. eruditus* or *C. malaccensis* with different densities of the specific prey items. Prey densities of 3, 6, 12, 18, 24, 30, 40, or 50 were set up in arenas by transferring the required numbers of the three stages of *L. decolor* using a partially wet camel brush under a stereomicroscope. There were six replications for each prey density and for the different prey items used in establishing the numerical response of either *C. eruditus* or *C. malaccensis*. The six replications of each prey stage were run in two batches of 3

replicates. For each three replicates, both predatory mites were simultaneously evaluated to reduce temporal variations between the numerical response parameters of predatory mites to prey stages and densities. Thus, 2 predator types x 3 prey stages x 8 prey densities x 3 replications were run concurrently in 2 batches to achieve a total of 6 replications for each prey density. For each replication set consisting of 144 experimental arenas, arenas were arranged randomly in plastic boxes (42 x 29 x 24 cm high) painted black, which has saturated KCl solution beneath perforated false floors to maintain 85 ± 5% RH and these were kept inside a growth chamber maintained at 24 ± 1°C and 0: 24 (L: D) photoperiod. Using a stereomicroscope, arenas were assessed at 24 h intervals to count the number of eggs laid/female/day and the number of prey consumed/female/day by predators. Cadavers of prey killed were removed and replaced with equal numbers killed. Eggs of predators and prey (in cases where female *L. decolor* was used as prey) were also removed along with cadavers to prevent predators using eggs as a diet. For each prey stage, a total of 48 females of *C. eruditus* or *C. malaccensis* were assessed. Each experiment spanned 168 h and seven data sets per observation (single female *C. eruditus* or *C. malaccensis*) were made at 24-h intervals. Thus, for each predator and prey stage, a total of 336 observations were recorded after the experimental duration (168 h). It was assumed that the average number of eggs laid (N_o) or number of prey consumed (N_a) at the end of the seventh consecutive data taken represented the daily prey oviposition rate or predation rate, respectively, of each female *C. eruditus* or *C. malaccensis* in their confined arenas at a specified prey density (N). The collected data were used to model the numerical responses and efficiency of conversion of ingested (ECI) prey of *C. eruditus*

and *C. malaccensis*. Also, per capita oviposition rate (eggs/ day), per capita oviposition efficiency (N_0/N), and per capita ECI (%) were estimated for each predatory mite.

Statistical Analysis. The numerical responses of *C. eruditus* or *C. malaccensis* to nymphs, adult females, and adult males of *L. decolor* were evaluated based on regression models to determine the relationship between oviposition and prey density. Regression models, *linear:* $y = a + bx$, *log:* $y = a + b\ln x$, and *polynomial:* $y = a + bx + cx^2$ have been used to describe the ovipositional response of mite predators to changing prey densities (Rahman et al. 2012, Yao et al. 2014). In order to fit the data, the model showing significant regressors ($p < 0.05$), lack of fit tests ($p > 0.05$), and with a better regression coefficient R^2 was selected to fit all data to allow standard comparison among predators and different life stages of the prey. Parameters of the model were estimated by equation 1 (linear), equation 13 (log), and equation 1003 (polynomial: quadratic) using 'EVALUATE' and 'NUMERIC' procedures in TableCurve 2D software (TableCurve 2D; Systat Software Inc., 2007). Note the equation numbers are in reference to the numbers of these equations in TableCurve 2D software. The efficiency of conversion of ingested food (ECI) pertains to the relationship between the conversion of prey biomass and prey density (Fathipour and Maleknia 2016). The ECI was modeled by the equation: $ECI = [(N_o/N_a) \times 100]$ at different prey densities (N); N_o is eggs laid and N_a is number of prey consumed (Omkar and Pervez 2004). The data on ECI at different prey densities were fitted using equation 13 (*log:* $y = a + b\ln x$) (TableCurve 2D; Systat Software Inc., 2007) based on aforementioned criteria to determine the relationship between ECI of predator and prey density for different prey life stages.

The mean eggs laid, (N_o), the oviposition efficiency, N_o/N, and the efficiency of conversion of ingested food (ECI) were compared across the three prey stages (nymph, male, and female) and eight prey densities (3, 6, 12, 18, 24, 30, 40, and 50) using generalized linear mixed models methods for each of the predator species (*C. eruditus* and *C. malaccensis*). PROC GLIMMIX modeled the fixed effects of prey stage and prey density and their interaction for each of the response variables with the specified response distribution (N_o ~ Poisson, N_o/N ~ exponential, and ECI ~ exponential) in SAS. Least squares means were compared for the appropriate significant effects. All tests were conducted at the nominal 0.05 level of significance.

Results

Numerical response curves of *C. eruditus* and *C. malaccensis* to *L. decolor*.

The ovipositional rate of *C. eruditus* and *C. malaccensis* per day varied considerably with an increase in prey density for different life stages (adult females, adult males, and nymphs) of *L. decolor* (Fig. 2 and Table10). Regression equations described the relationship between oviposition rate and prey density well ($p < 0.05$) for both predators and all prey life stages studied (Table 10). Among the fitted regression models (linear, log, and quadratic models), linear equations described the relationship between oviposition and prey density well, and the estimated regressors were also significantly different ($p < 0.05$) (Fig. 2 and Table 10). For *C. eruditus*, the rate of oviposition decreased significantly with an increase in female and male prey densities, however, these variables showed a significant positive correlation when nymphal stage of *L.*

decolor was consumed. The relationships can be expressed by the equations $y = 5.70 - 0.07x$ ($R^2 = 0.19$), $y = 5.40 - 0.06x$ ($R^2 = 0.20$), and $y = 1.64 + 0.07x$ ($R^2 = 0.39$) for females, males, and nymphs of *L. decolor*, respectively. In these equations, y is the per capita rate of oviposition per day, and x is the prey density (Figs. 2A, 2B, 2C, and Table 10). In the case of *C. malaccensis*, the oviposition rate significantly increased with increases in prey density for all prey life stages assessed (Table 10). The relationship between the numbers of eggs laid by *C. malaccensis* and the densities of females, males, and nymphs of *L. decolor* are described by linear equations: $y = 1.01 + 0.12x$ ($R^2 = 0.23$), $y = 2.68 + 0.13x$ ($R^2 = 0.18$), and $y = 2.40 + 0.16x$ ($R^2 = 0.25$), respectively (Fig. 2D, 2E, 2F, and Table 10). When all prey stages of *L. decolor* were combined, the relationship between oviposition rate and prey density for *C. malaccensis* was described well by the cubic polynomial model, that is, $y = -3.6 - 0.15x - 0.01x^2 - 0.0001x^3$ (F=14.56, P = 0.0128, $R^2 = 0.92$, Fig. 4B). In the case of *C. eruditus* the cubic polynomial model did not describe the relationship between oviposition rate and prey stage, but fitted line tracked data ($y = 3.71 + 0.08x - 0.004x^2 + 0.00005x^3$, F=1.65, P = 0.3122, $R^2 = 0.55$, Fig. 4A).

Efficiency of conversion of ingested food (ECI) by *C. eruditus* and *C. malaccensis*. The relationship between ECI (%) and prey density was best described by log equations. For *C. eruditus*, the relationships between ECI and prey density of females, males, and nymphs of *L. decolor* were described by log equations ($p < 0.05$), that is: $y = 423.31 – 102.12\ln(x)$ ($R^2 = 0.49$), $y= 337.59 – 85.03\ln(x)$ ($R^2 = 0.69$), and $y = 107.96 – 18.04\ln(x)$ ($R^2 =0.21$); the percent ECI (y) decreased substantially with prey density (x) (Fig 3A, 3B, 3C, and Table 11). However, the log equations did not describe the relationships between ECI and prey density for the various prey items in the case of

C. malaccensis, but their respective lines tracked data (lack of fit: *p*-values > 0.05). These were expressed in non-linear regression log equations: $y = 2132.45 - 22.39\ln(x)$ ($R^2 = 0.014$), $y = 140.70 - 8.84\ln(x)$ ($R^2 = 0.007$), and $y = 156.26 - 15.35\ln(x)$ ($R^2 = 0.019$) for females, males, and nymphs of *L. decolor*, respectively (Fig. 3D, 3E, 3F, and Table 11). The cubic polynomial model described the relationship between predation rate and prey density for *C. eruditus*, that is, $y = 0.18 + 0.52x - 0.01x^2 + 0.0001x^3$ ($F = 829.51$, $p < 0.0001$, $R^2 = 0.998$). In the case of *C. malaccensis*, the hyperbolic model described the relationship between predation rate and prey density, that is, $y = 0.34x / 1 + 0.03x$ ($F = 39.78$, $p < 0.001$, $R^2 = 0.74$). In these equations, *y* is the per capita mean number of prey killed and oviposition per day, and *x* is the prey density (Fig. 4).

Per capita oviposition rate of *C. eruditus* and *C. malaccensis*. The results of oviposition rate showed significant ($p < 0.05$) interaction between prey stage and prey density for *C. eruditus* and *C. malaccensis* when fed on *L. decolor* (Table 12), indicating that oviposition rate of predator species was substantially influenced by the type of prey stage consumed and with varied prey density; *Cheyletus eruditus* had the highest reproductive rate when fed on female *L. decolor*, and laid more eggs at lower prey densities (Table 13). Conversely, the oviposition rate was highest when the nymphal stage of prey was consumed by *C. malaccensis*, and more eggs were laid at higher prey densities. Similarly, the interaction between predator species and prey stage was significant ($F = 13.86$, DF= 2, 281, $p = 0.0001$). The rate of oviposition was markedly higher for *C. malaccensis* (3.18 eggs/day) when fed on nymphal stage of *L. decolor* but this was lowest in *C. eruditus* (1.68 eggs/day) when fed on adult female of *L. decolor*.

Per capita oviposition efficiency of *C. eruditus* and *C. malaccensis*. The interactions between prey stage and prey density for per capita oviposition efficiency was not significantly different ($p > 0.05$) for both *C. eruditus* and *C. malaccensis* (Table 12). The main effects of prey stage and prey density were significant for *C. malaccensis*, however, only the main effect of prey density was significant for *C. eruditus* (Table 12). The oviposition efficiency of *C. malaccensis* was highest when prey on nymphs (0.33) and at the lowest prey density (3 prey/ 16.88 cm^2) (Table 14). Similarly, *C. eruditus* oviposition efficiency was considerably higher at the lowest prey densities (3 – 6 prey/16.88 cm^2) (Table 14). The interaction between predator species and prey stage was significantly different (F= 8.44, DF= 2, 282, $p = 0.0003$). The oviposition efficiency was substantially higher (0.44) in *C. eruditus* and when adult females of *L. decolor* were present, but this was lowest (0.41) in the same predator species when nymphal stage of prey was consumed.

Per capita efficiency of conversion of ingested prey by *C. eruditus* and *C. malaccensis*. There was no significant ($p > 0.05$) interaction between prey stage and density for either *C. eruditus* or *C. malaccensis* (Table 12). For *C. eruditus*, the main effects of prey stage and prey density were significant ($p < 0.05$). The ECI was considerably higher (317.0%) at the lowest density of adult female *L. decolor* (3 prey/ 16.88 cm^2) and lowest (29.3%) at the highest density (50 prey/ 16.88 cm^2) of adult male prey (Table 15). In the case of *C. malaccensis*, the main effects of prey stage and prey density were also not significant ($p > 0.05$). The ECI was statistically similar across all the developmental stages and densities of *L. decolor* (ranged: 47.3 – 243.5%) (Table 15). The predators species and prey stage interactions were not significant (F= 2.47, DF= 2,

282, *p*= 0.0862), however, the main effects of predator species (*F*= 6.98, DF= 1, 282, *p*= 0.0087) and prey stage (*F*= 8.01, DF= 2, 282, *p*= 0.0004) were significant. Generally, *C. malaccensis* was more efficient (125.5%) in converting a substantial quantity of ingested prey to oviposition compared with *C. eruditus* (91.9%). For predator species, ECI was much higher when fed on adult female *L. decolor* (143.5%) than when nymphs (80.6%) and adult male (107.1%) of prey were consumed.

Discussion

Predatory mites display diverse ecological mechanisms to suppress pest population density below certain thresholds of pest known to cause economic loss (Metz et al 1988). These include functional response and defensive predation behaviors, and these are mostly a result of a surge in prey population density and simulation associated with the disturbance of predators at the ecological threshold (Sandness and McMurtry 1970, Zhu et al. 2019). The current study explored an alternative prey suppression mechanism, numerical response, and used the predatory mites *Cheyletus eruditus* and *Cheyletus malaccensis*. According to Cédola et al. (2001), the numerical response may be considered as a strategy of female predators to augment their offspring at different prey densities in order to manage pest populations. The present study revealed that the rate of oviposition and oviposition efficiency of *C. eruditus* and *C. malaccensis* may correlate positively or negatively with *L. decolor* population density and developmental stages. Other studies have reported similar findings that foraging behavior of predatory mites is not fixed but can vary depending on several factors, including the developmental stage of

the prey item, the system for interaction, physical conditions, and the type and spatial distribution of the predators' resources (Santos 1975, Ryoo 1986, Skirvin and Fenlon 2001).

For *C. eruditus*, the rate of oviposition decreased significantly with an increase in female and male prey densities, however, a significant positive correlation was observed when the nymphal stage of *L. decolor* was consumed. This is interesting given that the immature stage of *L. decolor* is considered a nutritionally poor food resource compared with the mature male and female development stages (Kucerova 2004). The oviposition rate in *C. eruditus* may be influenced by prey quantity rather than prey quality. In previous work by Danso et al. (unpublished), the predation rate of *C. eruditus* was significantly higher with the nymphal stage diet than with adult male and female *L. decolor* as prey. It is possible that larger prey size (Kucerova 2002), thicker cuticular layers, and greater anti-predation behaviors of prey including fast movement of adult male and female *L. decolor* may cause *C. eruditus* to invest more energy in predation, and in nest protection than when the nymphs are prey. Nevertheless, the oviposition rate significantly increased with increasing prey density across all life stages of prey for *C. malaccensis*. Furthermore, there was a general trend of a strong negative correlation between oviposition rates and increased in prey densities for *C. eruditus.* However, for *C. malaccensis* a strong positive correlation was found between both variates. The oviposition efficiency was in most cases similar for both predatory mites, and this was inversely related to prey density. The inverse relationship between oviposition and prey density for *C. eruditus* could have occurred due to the interference-stimulation phenomenon (Sandness and McMurtry 1970) — interference increases with increasing

prey density and this decreases the total acquired nutrition (energy) from prey by a predator. Consequently, this decreases the oviposition rate of a predator at a higher prey densities (Souza-Pimentel et al. 2018). However, the increase in oviposition rate in response to increasing prey density found in *C. malaccensis* is an important foraging characteristic of an efficient biocontrol agent. Thus, in a case of increase in psocid population, *C. malaccensis* population is expected to increase in an inoculative release which can facilitate managing the pest population below economic threshold (Rahman et al. 2012, Yao et al. 2014). On the other hand, *C. eruditus* predation rate suggests that this mite can be used effectively for inundative augmentative release to provide rapid psocid suppression in empty storage structures, grain residues, in stacked pallets, and other similar scenarios in the storage environment.

The results showed that *C. eruditus* would only be efficient and lay a substantial number of eggs (~4 eggs per predator per day) at a prey density of ~12 prey/16.88 cm^2 or less, but above this density oviposition rate monotonically changed. In the case of *C. malaccensis*, oviposition rate increased considerably with prey density (even surpassing its predation rate), and peaked at ~ 9 eggs per predator per day at a predicted prey density of ~ 55 prey/16.88 cm^2. These fecundity levels indicate that *C. malaccensis* would be better than *C. eruditus* in terms of establishing and colonizing storage environments for the suppression of *L. decolor*. In contrast, the implications of the ovipositional response of *C. eruditus* is that this predatory mite species would be unable to keep up with rapid growth of *L. decolor* populations. However, high predation rates of *C. eruditus* tend to reduce *L. decolor* populations more rapidly than *C. malaccensis* (Danso et al. unpublished). It would be logical to have alternative rich resources including other stored

product arthropods pests that enhance *C. eruditus* oviposition rate, however, as stated before, augmentation by inundative release of this cheyletid would be essential to increase its effectiveness for use in psocid management. It would be prudent to implement inoculative releases of *C. eruditus* that target the early stage of pest infestation where oviposition per predation rate is substantially high to facilitate their survival and establishment in storage environments (Xu and Zhang 2015, Patel and Zhang 2017).

In order to validate findings of this study, information on female life table parameters of these cheyletids would be necessary to ascertain whether oviposition positively correlates with the number of individuals that develop from eggs to female adults, and are consistently higher for *C. malaccensis* than *C. eruditus*. In most cases efficient predatory mites exhibit a higher intrinsic rate of increase than their prey (Fathipour and Maleknia 2016), and this also needs to be compared.

In most cases, the efficiency of conversion of ingested (ECI) prey decreased considerably with increasing prey density across different prey types for *C. eruditus*. However, for *C. malaccensis*, ECI was not affected by prey stage and density of *L. decolor*. Fathipour and Maleknia (2016) reported that the ECI is higher at low prey density and subsequently decreases at higher prey densities. The female *C. eruditus* at low prey density probably invest more energy in egg production and, in the process, invest less in maintenance and metabolic activities (Omkar and Pervez 2004). Besides, the lower ECI at higher prey densities possibly suggests that female predators expend considerable energy defending their nests through the wasteful killing of prey (Zhu et al. 2019) and may not necessarily utilize most of the attacked prey for physiological activities such as oviposition. This can be validated from earlier work by Danso et al.

(unpublished) on functional responses of both predatory mites to *L. decolor* that indicated that the predation rate of *C. eruditus* was significantly greater than that for *C. malaccensis* especially at higher prey densities. The estimated ECI indicated that *C. malaccensis* would be a more efficient predator than *C. eruditus* due to the higher oviposition to predation ratio observed in the former. Predation on different developmental life stages of *L. decolor* by predatory mites showed that *C. eruditus* was more effective in converting the biomass of adult female *L. decolor* to produce eggs than when fed on males and nymphs. Interestingly, there were no considerable differences in ECI for *C. malaccensis* when different prey developmental stages were consumed. Therefore, prey quality rather than quantity may determine the success of *C. eruditus* when released for biological control programs.

Cheyletid mites, *C eruditus* and *C malaccensis* can prey on *L. decolor* to survive, establish, and produce significant numbers of offspring to suppress and stabilize *L. decolor* populations. The current study has established that *C. malaccensis* is far better than *C. eruditus* in relation to numerical response variates assessed. Information from our previous work on functional response parameters supports the superior effectiveness of *C. eruditus* when compared to *C. malaccensis* under similar experimental conditions. Therefore, it can be concluded that the foraging behavior of these cheyletids alone is limited in scope for determining the effectiveness of these predatory mites for use as biocontrol agents in psocid management. Combined application of *C. malaccensis* and *C. eruditus* to complement each other functionally and numerically would most probably produce synergistic suppression effect on psocid populations within the storage environment. Otherwise, inundative and inoculative releases of *C. eruditus* and *C.*

malaccensis, respectively, would be recommended given their predation and reproductive capacities. Nevertheless, further research on mutual inference and interspecific competition between *C. eruditus* and *C. malaccensis* is required. Because predator-prey interactions are complex, laboratory environmental conditions do not allow accurate predictions of the effectiveness of predators as biological control agents under the field conditions. Therefore, further assessment studies other than foraging behaviors must be explored in order to make more accurate decisions on selection and application of these predatory mites solely or in IPM systems for managing stored-product insect pests in the United States.

Acknowledgments

We thank Dr. Yi Wu, the Academy of State Administration of Grain, Beijing, China for providing the colonies of *Cheyletus eruditus*, *Cheyletus malaccensis*, and *Acarus siro* for this work. This research was funded by Oklahoma Agricultural Experiment Station (Project number OKL2949). Trade names or commercial products mentioned in this paper are solely for the purpose of providing specific information and do not imply recommendation or endorsement by Oklahoma State University.

Table 10. Parameters (± SE) for the regression models, linear: y = a + bx, log: y = a + blnx, and polynomial: $y = a + bx + cx^2$ (Equations 1, 13, and 1003, respectively in TableCurve 2D) describing the ovipositional responses of *C. eruditus* and *C. malaccensis* on a diet of *L. decolor* females, males and nymphs at a constant temperature of 24 ± 1°C and 85 ± 5% RH

Predator	Prey Stage	Regression model	Regression *P-value* (F-stat)	Lack of fit *P-value* (F-stat)	Maximum R^2 (Adj. R^2) [R^2]	A (95% CI)	b (95% CI)	c (95% CI)
C. eruditus	Female	Linear	0.0018 (11.0026)	0.6284 (0.7298)	0.27 (0.16) [0.19]	5.70 ± 0.57* (4.57 – 6.84)	-0.07 ± 0.02* (-0.11 – -0.03)	–
		Log	0.0145 (6.4617)	0.2502 (1.3700)	0.27 (0.08) [0.12]	6.74 ± 1.07* (4.59 – 8.90)	-0.93 ± 0.36* (-1.66 – - 0.19)	–
		Polynomial	0.0075 (5.4615)	0.5225 (0.8504)	0.27 (0.14) [0.20]	5.48 ± 0.85* (3.76 – 7.19)	-0.04 ± 0.08 (-0.20 – 0.12)	-0.001 ± 0.002 (-0.004 – 0.003)
	Male	Linear	0.0016 (11.2480)	0.5386 (0.8513)	0.29 (0.16) [0.20]	5.40 ± 0.51* (4.37 – 6.43)	-0.06 ± 0.02* (-0.1 – -0.03)	–
		Log	0.0049 (8.7431)	0.3286 (1.1953)	0.29 (0.12) [0.16]	6.66 ± 0.95* (4.74 – 8.58)	-0.96 ± 0.32* (-1.61 – -0.31)	–
		Polynomial	0.0069 (5.5791)	0.4321 (0.9967)	0.29 (0.14) [20]	5.60 ± 0.77* (4.05 – 7.15)	-0.09 ± 0.07 (-0.23 – 0.06)	0.0005 ± 0.001 (-0.002 – 0.003)
	Nymph	Linear	< 0.0001 (28.9299)	0.1778 (1.5812)	0.50 (0.36) [0.39]	1.64 ± 0.34* (0.96 – 2.32)	0.07 ± 0.01* (0.04 – 0.09)	–
		Log	< 0.0001 (26.9244)	0.1221 (1.8081)	0.50 (0.34) [0.37]	0.07 ± 0.62 (-1.18 –1.33)	1.10 ± 0.21* (0.67 – 1.52)	–
		Polynomial	< 0.0001 (16.0821)	0.2444 (1.4020)	0.50 (0.38) [0.42]	1.08 ± 0.50* (0.08 – 2.08)	0.14 ± 0.05* (0.04 – 0.23)	-0.001 ± 0.0001 (-0.003 – 0.004)
C. malaccensis	Female	Linear	0.0005 (13.8595)	0.1600 (1.6453)	0.38 (0.20) [0.23]	1.01 ± 0.87 (-0.75 –2.76)	0.12 ± 0.03* (0.05 – 0.18)	–
		Log	0.0079 (7.7118)	0.0321 (2.5967)	0.38 (0.11) [0.14]	-0.73 ± 1.68 (-4.11 –2.64)	1.58 ± 0.57* (0.43 – 2.72)	–

	Polynomial	0.0011	0.1825	0.38 (0.21)	2.28 ± 1.29	-0.04 ± 0.12	0.003 ± 0.002
		(7.9231)	(1.5993)	[0.26]	(-0.32 –4.88)	(-0.28 – 0.21)	(-0.002 – 0.008)
Male	Linear	0.0026	0.6168	0.26 (0.14)	2.68 ± 1.15*	0.13 ± 0.04*	
		(10.1316)	(0.7450)	[0.18]	(0.36 – 4.99)	(0.05 – 0.21)	–
	Log	0.0062	0.4374	0.26 (0.11)	-0.11 ± 2.13	2.08 ± 0.72*	
		(8.2493)	(1.0022)	[0.15]	(-4.40– 4.18)	(0.62 – 3.53)	–
	Polynomial	0.0112	0.4985	0.26 (0.13)	2.91 ± 1.73	0.10 ± 0.16	0.001 ± 0.003
		(4.9756)	(0.8876)	[0.18]	(-0.58– 6.40)	(-0.22 – 0.43)	(-0.006 – 0.007)
Nymph	Linear	0.0003	0.8135	0.30 (0.21)	2.40 ± 1.10*	0.16 ± 0.04*	
		(15.025)	(0.4878)	[0.25]	(0.18 – 4.62)	(0.08 – 0.24)	–
	Log	0.0024	0.3874	0.30 (0.15)	-0.45 ± 2.09	2.28 ± 0.71*	
		(10.318)	(1.0857)	[0.18]	(-4.65– 3.76)	(0.85 – 3.71)	–
	Polynomial	0.0013	0.78481	0.30 (0.20)	3.29 ± 1.65	0.05 ± 0.16	0.002 ± 0.003
		(7.6995)	(0.4858)	[0.26]	(-0.04– 6.62)	(-0.27 – 0.36)	(-0.004 – 0.008)

* Regression model parameter estimate was significantly different at $P < 0.05$; regression DF = 1, 46; lack of fit DF = 6, 40

Table 11. Parameters (± SE) for the regression model, log: y = a + blnx (Equations 13 in TableCurve 2D) describing the efficiency of conversion of ingested food by *C. eruditus* and *C. malaccensis* on a diet of *L. decolor* females, males and nymphs at a constant temperature of 24 ± 1°C and 85 ± 5% RH

Predator	Prey Stage	Regression *P-value* (F-stat)	Lack of fit *P-value* (F-stat)	Maximum R^2 (Adj. R^2) [R^2]	A (95% CI)	b (95% CI)
C. eruditus	Female	< 0.0001 (43.824)	0.9961 (0.0993)	0.50 (0.47) [0.49)	423.31 ± 45.48* (331.77 – 514.85)	-102.12 ± 15.43* (-133.17 – -71.07)
	Male	< 0.0001 (100.694)	0.4610 (0.9652)	0.73 (0.67) [0.69)	337.59 ± 24.98* (287.31 – 387.88)	-85.03 ± 8.47* (-102.09 – -67.97)
	Nymph	0.0012 (11.8594)	0.1589 (1.6497)	0.36 (0.17) [0.21]	107.96 ± 15.45* (76.87 – 139.05)	-18.04 ± 5.24* (-28.59 – -7.50)
C. malaccensis	Female	0.4321 (0.6282)	0.4849 (0.9288)	0.13 (0.0) [0.014]	213.45 ± 83.29* (45.79 – 381.11)	-22.39 ± 28.25 (-79.27 – 34.48)
	Male	0.5669 (0.3327)	0.4215 (1.0284)	0.14 (0.0) [0.007]	140.70 ± 45.19* (49.75 – 231.65)	- 8.84 ± 15.33 (-39.69 – 22.01)
	Nymph	0.3450 (0.9110)	0.3898 (1.0816)	0.16 (0.0) [0.019]	156.26 ± 47.41* (60.82 – 251.69)	-15.35 ± 16.08 (-47.72 – 17.03)

* Regression model parameter estimate was significantly different at $P < 0.05$; regression DF = 1, 46; lack of fit DF = 6, 40

Table 12. Summary of the tests for the fixed effects of prey stage (PS) and prey density (N) of *L. decolor* on numbers of eggs laid (N_o), oviposition efficiency (N_o/N), and the efficiency of conversion of ingested prey (ECI) of predatory mites, for *Cheyletus eruditus* (CE) or *Cheyletus malaccensis* (CM).

Parameter	Variable	Source	Df	*F*	*P* value
CE	N_o	PS	2, 119	3.84	0.0242
		N	7, 119	0.79	0.5996
		PS*N	14, 119	3.06	0.0004
	N_o/N	PS	2, 120	1.25	0.2898
		N	7, 120	18.44	< 0.0001
		PS*N	14, 120	0.99	0.4687
	ECI	PS	2, 120	5.92	0.0035
		N	7, 120	6.09	< 0.0001
		PS*N	14, 120	0.75	0.7239
CM	N_o	PS	2, 119	15.18	< 0.0001
		N	7, 119	19.93	< 0.0001
		PS*N	14, 119	2.86	0.0010
	N_o/N	PS	2, 120	5.10	0.0075
		N	7, 120	7.59	< 0.0001
		PS*N	14, 120	0.86	0.6021
	ECI	PS	2, 120	0.72	0.4878
		N	7, 120	2.00	0.0601
		PS*N	14, 120	0.77	0.6971

Table 13. Numbers of eggs laid (N_o) (mean ± SE) per female *C. eruditus* or *C. malaccensis* per day (d^{-1}) exposed to *L. decolor* at different developmental stages and densities.

Predator	Prey density (*N*)	Eggs laid per Predator (N_o) d^{-1} (95% CI)		
		Prey Stage		
		Female	Male	Nymph
C. eruditus	3	2.89 ± 0.63 aBC (1.87 – 4.46)	2.89 ± 0.63 aBC (1.87 – 4.46)	1.64 ± 0.46 C (0.63 – 2.25)
	6	3.86 ± 0.75 aAB (2.61 – 5.68)	4.29 ± 0.81 aA (2.96 – 6.22)	1.19 ± 0.38 bC (0.78 – 2.54)
	12	4.05 ± 0.78 aA (2.77 – 5.92)	3.32 ± 0.69 aAB (2.21 – 5.01)	1.40 ± 0.42 bBC (0.80 – 2.58)
	18	3.37 ± 0.69 aAB (2.24 – 5.07)	2.65 ± 0.60 aBC (1.69 – 4.15)	1.52 ± 0.44 bBC (0.86 – 2.69)
	24	2.63 ± 0.60 aBC (1.67 – 4.13)	2.66 ± 0.60 aBC (1.70 – 4.17)	2.37 ± 0.56 aBC (1.48 – 3.80)
	30	2.28 ± 0.55 aBC (1.41 – 3.67)	1.92 ± 0.50 aBC (1.14 – 3.21)	3.24 ± 0.68 aA (2.14 – 4.91)
	40	1.66 ± 0.46 bC (0.96 – 2.88)	2.18 ± 0.54 aBC (1.34 – 3.55)	3.06 ± 0.66 aA (2.00 – 4.68)
	50	1.76 ± 0.48 bC (1.03 – 3.01)	1.64 ± 0.46 bC (0.95 – 2.85)	2.84 ± 0.63 aAB (1.83 – 4.39)
C. malaccensis	3	1.30 ± 0.33 bBC (0.78 – 2.16)	1.06 ± 0.30 bD (0.61 – 1.85)	1.77 ± 0.40 aCD (1.13 – 2.77)
	6	0.62 ± 0.22 bC (0.31 – 1.27)	2.31 ± 0.47 aC (1.55 – 3.46)	1.13 ± 0.31 bD (0.66 – 1.94)
	12	1.14 ± 0.31 bBC (0.66 – 1.95)	1.77 ± 0.40 aCD (1.13 – 2.77)	2.40 ±0.48bcBC (1.62 – 3.56)
	18	0.68 ± 0.23 bC (0.34– 1.34)	2.21 ± 0.46 aC (1.47 – 3.32)	2.31 ± 0.47 aBC (1.55 – 3.44)
	24	2.32 ± 0.47 aAB (1.56 – 3.47)	2.40 ± 0.48 aBC (1.62 – 3.56)	1.65 ± 0.38 aCD (1.04 – 2.61)
	30	0.75 ± 0.25bBC (0.39 – 1.44)	2.20 ± 0.45 aC (1.46– 3.31)	3.39 ± 0.59 aAB (2.40 – 4.79)
	40	3.21 ± 0.57 aA (2.25 – 4.57)	4.84 ± 0.74 aA (3.56 – 6.57)	3.99 ± 0.66 aA (2.88 – 5.53)
	50	3.32 ± 0.58 aA (2.34 – 4.70)	3.78 ± 0.64 aAB (2.72 – 5.28)	4.80 ± 0.74 aA (3.53 – 6.52)

Significant differences among prey stages for each prey density are denoted with different lower-case letters for each predator and differences among prey densities for each prey stage are denoted by different upper-case letters for each predator ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

Table 14. Per capita oviposition efficiency (N_0/N) (mean ± SE) of predatory mites, *C. eruditus* or *C. malaccensis* exposed to different developmental stages and densities of *L. decolor.*

Predator	Prey density (N)	Oviposition Efficiency (N_o/N) (95% CI)		
		Prey Stage		
		Female	Male	Nymph
C. eruditus	3	1.42 ± 0.58 aA (0.63 – 3.19)	1.42 ± 0.57 aA (0.63 – 3.18)	0.59 ± 0.24 bA (0.26 – 1.32)
	6	0.95 ± 0.39 aA (0.42 – 2.12)	1.06 ± 0.17 aAB (0.47 – 2.37)	0.35 ± 0.14 bAB (0.15 – 0.77)
	12	0.50 ± 0.20 aAB (0.22 – 1.12)	0.41 ± 0.17 aBC (0.18 – 0.92)	0.18 ± 0.07 aBC (0.09 – 0.40)
	18	0.28 ± 0.11 aBC (0.12 – 0.62)	0.22 ± 0.09 aCD (0.10 – 0.48)	0.13 ± 0.05 aBC (0.06 – 0.28)
	24	0.16 ± 0.07 aCD (0.07 – 0.36)	0.17 ± 0.08 aCD (0.07 – 0.37)	0.15 ± 0.06 aBC (0.07 – 0.33)
	30	0.11 ± 0.05 aCD (0.05 – 0.25)	0.10 ± 0.04 aDE (0.04 – 0.21)	0.16 ± 0.07 aBC (0.07 – 0.36)
	40	0.06 ± 0.03 aD (0.03 – 0.13)	0.08 ± 0.03 aDE (0.05 – 0.17)	0.12 ± 0.05 aBC (0.05 – 0.25)
	50	0.05 ± 0.02 aD (0.02 – 0.12)	0.05 ± 0.02 aE (0.02 – 0.11)	0.08 ± 0.03 aC (0.04 – 0.19)
C. malaccensis	3	0.96 ± 0.39 abA (0.42 – 2.16)	0.79 ± 0.32 aA (0.35 – 1.77)	1.31 ± 0.53 aA (0.58 – 2.94)
	6	0.23 ± 0.09 bB (0.10 – 0.52)	0.86 ± 0.35 aA (0.38 – 1.92)	0.42 ± 0.17 aAB (0.19 – 0.94)
	12	0.21 ± 0.09 bB (0.09 – 0.47)	0.33 ± 0.13 aAB (0.15 – 0.74)	0.44 ± 0.18 aAB (0.20 – 1.00)
	18	0.08 ± 0.03 aBC (0.04 – 0.19)	0.27 ± 0.11 aAB (0.12 – 0.61)	0.28 ± 0.12 aB (0.13 – 0.64)
	24	0.22 ± 0.09 aB (0.10 – 0.48)	0.22 ± 0.09 aB (0.10 – 0.50)	0.15 ± 0.06 aB (0.07– 0.34)
	30	0.06 ± 0.02 aC (0.02 – 0.12)	0.16 ± 0.07 aB (0.07 – 0.37)	0.25 ± 0.10 aB (0.11 – 0.57)
	40	0.18 ± 0.07 aB (0.08 – 0.40)	0.27 ± 0.10 aAB (0.12 – 0.60)	0.22 ± 0.09 aB (0.10 – 0.50)
	50	0.15 ± 0.06 aBC (0.06 – 0.33)	0.17 ± 0.07 aB (0.08 – 0.38)	0.21 ± 0.08 aB (0.09 – 0.48)

Significant differences among prey stages for each prey density are denoted with different lower-case letters (within the same row) for each predator, and differences among prey densities for each prey stage are denoted by different upper-case letters (within a column) for each predator ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

Table 15. Per capita efficiency of conversion of ingested food (mean ± SE) of predatory mites, *C. eruditus* or *C. malaccensis* exposed to different developmental stages and densities of *L. decolor*

Predator	Prey density (*N*)	Efficiency of Conversion of Ingested Food (ECI) % (95% CI)		
		Prey Stage		
		Female	Male	Nymph
C. eruditus	3	317.0 ± 129.4 aA (141.3– 711.4)	257.4 ± 105.1 aA (114.7–577.5)	108.1 ± 44.1 bA (48.2 – 242.7)
	6	249.5 ±101.8 aAB (111.1–559.8)	200.1 ± 81.7 aAB (89.2–449.1)	68.7 ± 28.0 bB (30.6 – 154.1)
	12	159.7 ± 65.2 aBC (71.2–358.4)	104.4 ± 42.6 bC (46.5 – 234.3)	43.2 ± 17.6 cBC (19.3 – 96.9)
	18	120.5 ± 49.2 aCD (53.7–270.4)	63.7 ± 25.9 bCD (28.4 –142.8)	35.1 ± 14.3 cC (15.6 – 78.7)
	24	86.0 ± 35.1 aDE (38.3 –192.9)	55.9 ± 22.8 bCD (24.9 – 125.4)	48.2 ± 19.7 cBC (21.5 – 108.1)
	30	65.1 ± 26.6 aEF (29.0 –145.2)	40.7 ± 16.6 bD (18.1 – 91.3)	60.5 ± 24.7 abBC (26.9 – 135.7)
	40	50.9 ± 20.8 aF (22.7 –114.3)	40.8 ± 16.7 aDE (18.2 – 91.6)	53.8 ± 21.9 aBC (23.9 – 120.7)
	50	45.7 ± 18.6 aF (20.4 – 102.5)	29.3 ± 11.9 bE (13.0 – 65.7)	41.2 ± 16.8 aBC (18.3 – 92.3)
C. malaccensis	3	243.5 ± 99.3 aA (108.5 – 546.4)	119.3 ± 48.7 aA (53.1 – 267.8)	192.3 ± 78.5 aA (85.6 –431.5)
	6	179.4 ± 73.2 aA (79.9 – 402.5)	170.7 ±69.7 aA (76.1– 383.1)	90.0 ± 36.7 aA (40.1 – 202.0)
	12	108.3 ± 44.2 aA (48.2 – 243.1)	113.2 ± 46.2 aA (50.4 – 254.0)	107.6 ± 43.9 aA (47.9 –241.4)
	18	54.9 ± 22.4 aA (24.4 – 123.1)	89.7 ± 36.5 aA (39.9 –201.1)	99.7 ± 40.7 aA (44.4 –223.76)
	24	174.4 ± 71.2 aA (77.7– 391.4)	82.0 ± 33.5 aA (36.5 –184.1)	47.3 ± 19.3 aA (21.0 – 106.2)
	30	60.4 ± 24.6 aA (26.9 – 135.6)	68.1 ± 27.7 aA (30.3 – 152.7)	105.2 ± 42.9 aA (46.8 – 236.1)
	40	201.1 ± 82.1 aA (89.6– 451.3)	169.0 ± 69.0 aA (75.3– 379.3)	119.0 ± 48.5 aA (53.0 – 267.1)
	50	182.9 ± 74.6 aA (81.5– 410.5)	115.1 ± 46.9 aA (51.2 – 258.1)	144.4 ± 58.9 aA (64.3–324.0)

Significant differences among prey stages for each prey density are denoted with different lower-case letters (within the same row) for each predator, and differences among prey densities for each prey stage are denoted by different upper-case letters (within a column) for each predator ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

Figure Captions

Fig. 2. Ovipositional response of *C. eruditus* or *C. malaccensis* to different life stages (female, male, and nymph) and densities of *L. decolor*. Parameters for the fitted regression curves are in Table 1. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* for each prey life stage, and each observation was mean of seven days (7d) exposure period.

Fig. 3. Efficiency of conversion of ingested prey by *C. eruditus* or *C. malaccensis* when offered different life stages (female, male, and nymph) and densities of *L. decolor*. Parameters for the fitted regression curves are in Table 2. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* for each prey life stage.

Fig. 4. Predation and oviposition rate of *C. eruditus* or *C. malaccensis* exposed to different densities of *L. decolor*. The number of observations (*n*) used was 48 adult females of *C. eruditus* or *C. malaccensis* and each observation was mean of seven days (7d) exposure period.

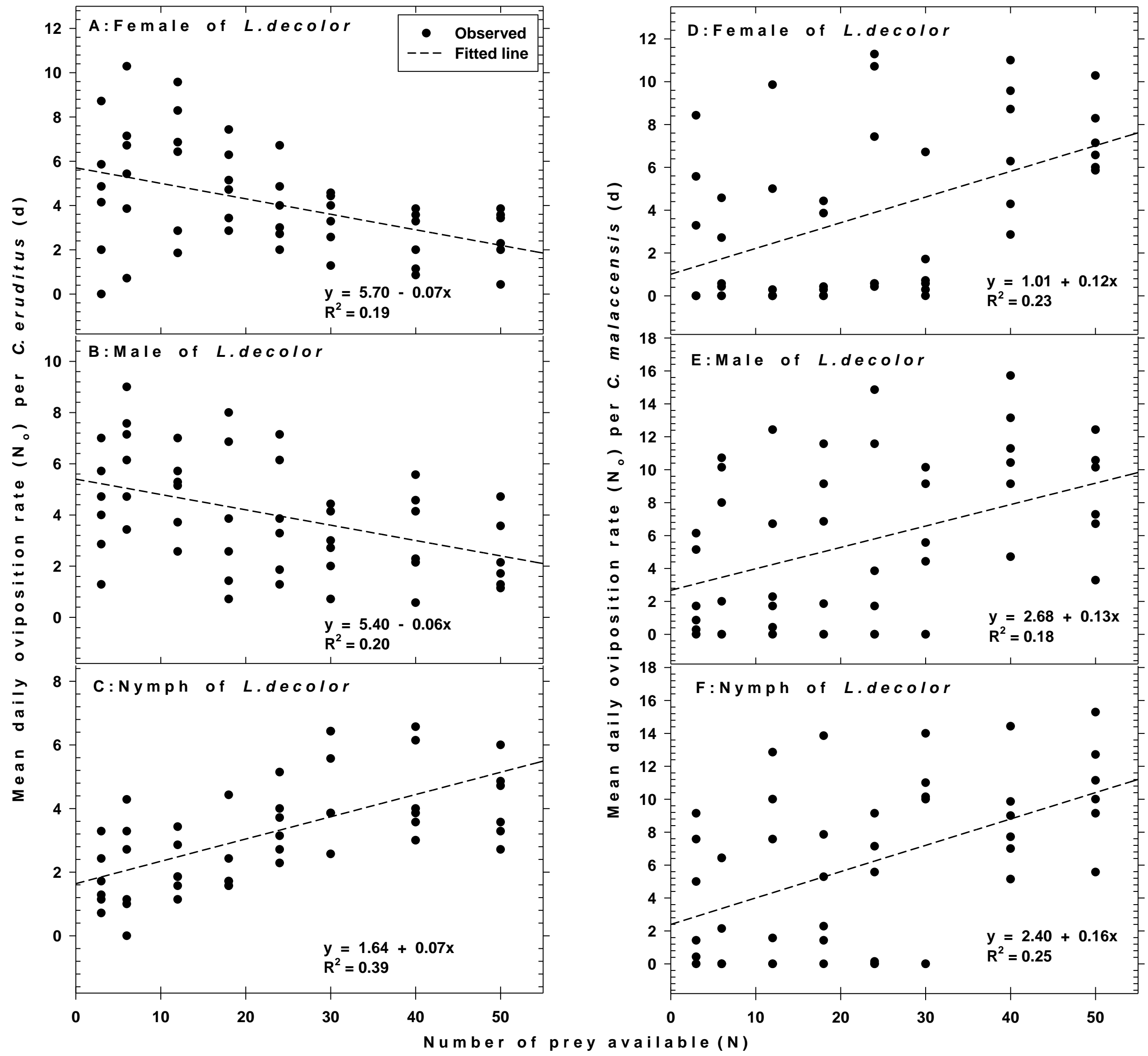

Fig. 2

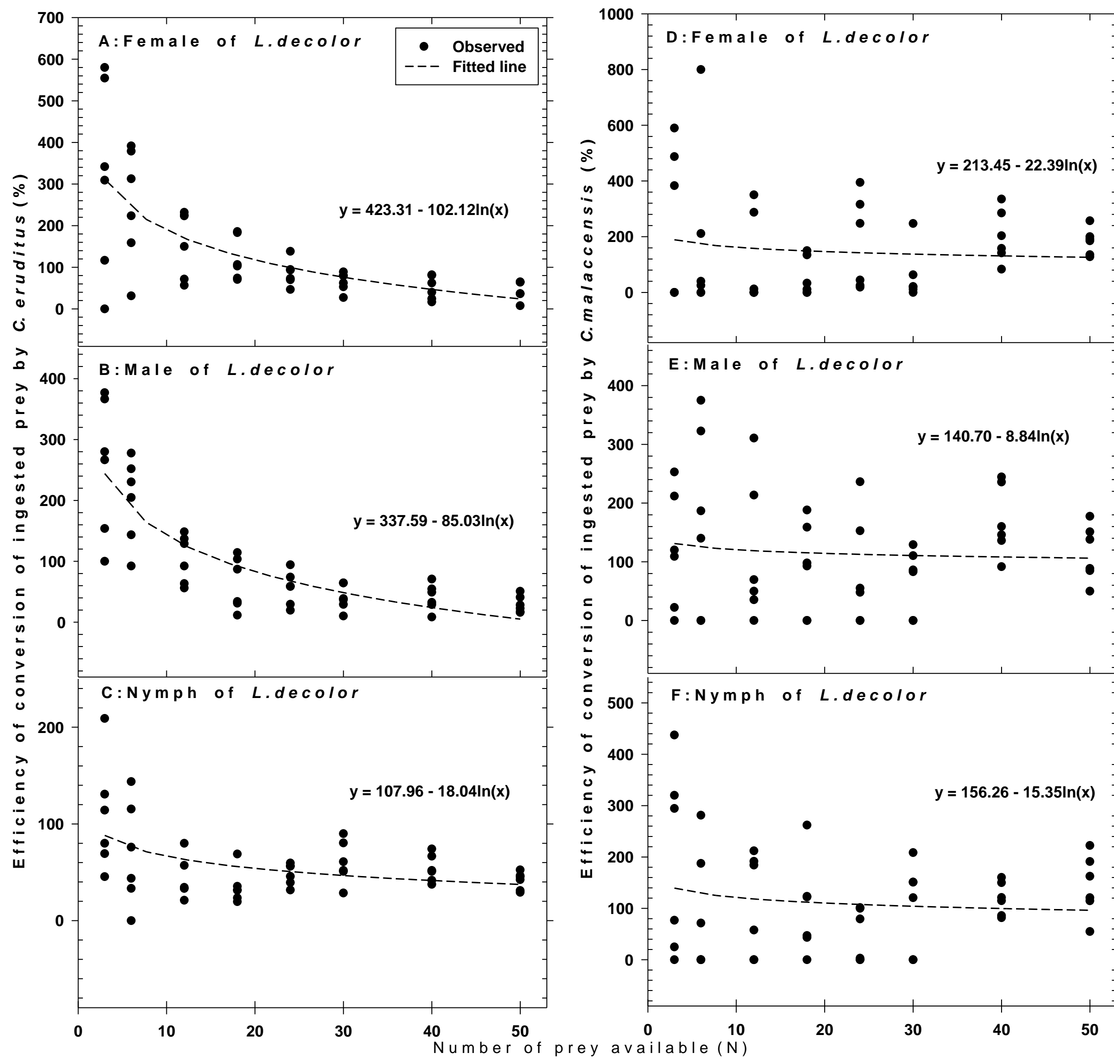

Fig. 3

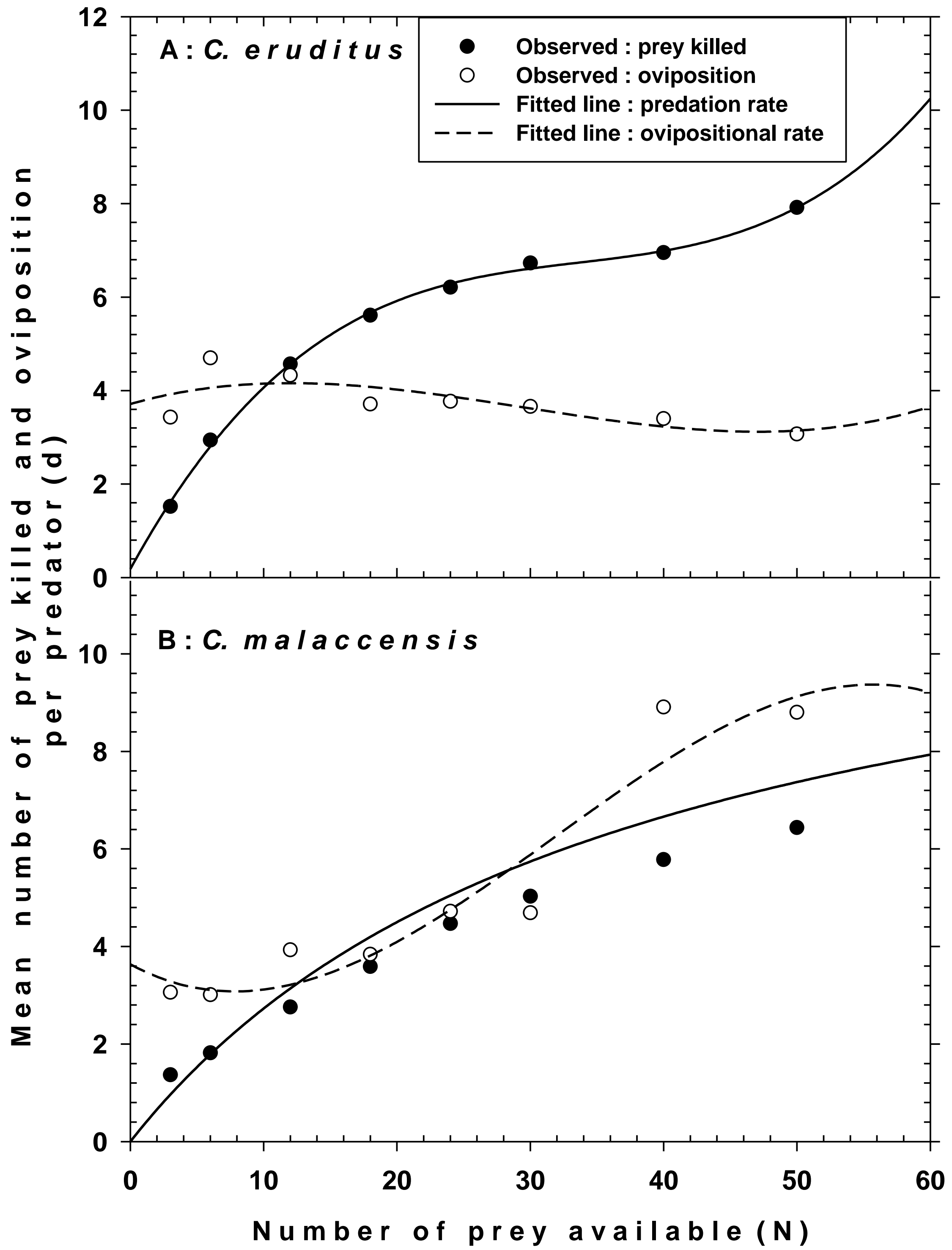

Fig. 4

CHAPTER VI

ECOLOGICAL INTERACTION OF THE PREDATORY MITES, *CHEYLETUS ERUDITUS* (SCHRANK) AND *CHEYLETUS MALACCENSIS* OUDEMANS (TROMBIDIFORMES: CHEYLETIDAE) AND PREY, *LIPOSCELIS DECOLOR* (PEARMAN) (PSOCODEA: LIPOSCELIDIDAE) UNDER DIFFERENT THERMO-HYGROMETRIC REGIMES

(To be submitted to Journal of Economic Entomology)

Abstract. Predator-prey interactions are linked through trophic relationships, and individual population dynamics are a function of multiple interactions among many ecological factors. The present study considered the efficacy of the predatory mites *Cheyletus eruditus* (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) to manage *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae). Prey population suppression and progeny replacement efficiency of the predators were assessed under different predator-prey ratios (10:20, 4:20, 2:20, 1:20, and 0:20), temperatures (20, 24, 28, and 32 °C), and relative humidities (RH) (63, 75, and 85%) over 40 days under laboratory condition of 0:24 (L: D) photoperiod. Suppression of *L. decolor* population when *C. eruditus*-related predator-to-prey ratios of 10:20, 4:20, 2:20, and 1:20 were used was ~ 96.3%, 94.5%, 88.4%, and 67.1%, respectively, relative to the Control (0:20). In the case of *C. malaccensis*, suppression of ~ 97.2%, 95.0%, 83.9%, and 70.4%, respectively, was achieved. Although the low 63% RH limited efficacy of these cheyletid mites, both predatory mites caused pest population suppression of ~ 67.1–97.2% and increased their progeny by ~117.1–1182.6% for the 1:20–10:20 release ratios, temperatures of 20–32°C, and 75–85% RH. These temperature and RH ranges represent physical conditions that permit survival of psocids. The levels of psocid population suppression achieved indicate the potential of both predatory mites for psocid management. Field trials need to be conducted to validate results of this study.

Key words: psocid, biological control, stored product, Cheyletidae, pest management

Introduction

Cheyletus eruditus (Schrank) and *Cheyletus malaccensis* Oudemans (Trombidiformes: Cheyletidae) are the most dominant and widely distributed cheyletid species in post-harvest agricultural systems in temperate and tropical regions (Zdarkova 1998, Palyvos et al. 2008, Athanassiou and Palyvos 2015). Both predatory mites are natural enemies of multiple arthropods and prey on mite and non-mite pests (Mullen and OConnor 2019), including psocids (Psocodea: Liposcelididae) (Kucerova 2004). Predator-prey interactions are linked through trophic relationships, and individual population dynamics are a function of multiple interactions among many biotic and abiotic variables (Leeuwen et al. 2007, Fathipour and Maleknia 2016). For example, natural enemy complex may exist in the storage community due to the presence of different prey species and leads to trophic interactions such as competition, interference, cannibalism, and intraguild predation even among conspecifics with a profound effect on biocontrol agents in pest management (Rosenheim et al. 1995, Pakyari and Fathipour 2009, Farazmand et al. 2015, Zhu et al. 2019). Also, prey biotic related factors such as prey characteristics, prey type and composition, seasonal occurrence, and spatial abundance and distribution are major determinants for the establishment, colonization, predation, survivorship, and reproduction of predators to allow them effectively manage pest populations (Santos 1975, Ryoo 1986, Xiao and Fadamiro 2010, Kalinoski and DeLong 2016, Uiterwaal et al. 2017). Similarly, physical conditions within storage environments including temperature and relative humidity (RH) are key variables that influence the overall outcome of ecological interactions; predators and prey can survive and thrive under different temperature and RH ranges favorable for their growth and

development (Zhu et al. 2019). For instance, *C. malaccensis* can reproduce at temperatures between 17.5–35 °C, and develop in the range between 11.6–37.8 °C (Zhu et al. 2019). *Cheyletus eruditus* populations multiply rapidly during summer and autumn when temperatures are high, whereas its prey *Acarus siro* L. (Sarcoptiformes: Acaridae) population levels decline substantially as a result of extreme temperatures and RH (Solomon 1949). *Cheyletus eruditus* can complete its life cycle at temperatures and RH ranging from 12–35°C and 60–90%, respectively (Schöller et al. 2006).

Release ratio (predator-to-prey ratio) is a prerequisite variable to establish prior to the release of biocontrol agents, and this is critical for successful biological control programs (Fathipour and Maleknia 2016). The degree of control would be influenced by how many predators are released at a given pest infestation level. Thus, pest control would be poor if a limited number of predators were released, and on the other hand the release of predators at a higher density would likely lead to underutilization of their control potential through density-dependent negative feedbacks (Zhu et al. 2019). To date, little is known about the potential of the cheyletid mites, *C. eruditus* and *C. malaccensis* to manage psocid populations under physical conditions that exist in storage environments. Currently, only Kucerova (2004) has documented the behavioral and ecological aspects of *C. eruditus* and how these relate to its potential to suppress *Liposcelis decolor* (Pearman) (Psocodea: Liposcelididae) populations in stored grain. Previous data (Danso et al. unpublished) on the foraging behaviors of *C. eruditus* and *C. malaccensis* showed that these cheyletids had the potential to suppress *L. decolor* population under the same temperature (24 ± 1°C) and RH (85 ± 5%) conditions. Given that the general performance of an effective biocontrol agent is a function of multiple

ecological interactions among many biotic and abiotic variables, the evaluation of predator-prey interaction based on other variates such as predatory-to-prey ratio and thermo-hygrometric factors are necessary to investigate. The current study aimed to assess the ecological interaction between the cheyletid mites *C. eruditus* and *C. malaccensis* and the psocid species *L. decolor*. How *L. decolor* population dynamics is influenced by different predator-to-prey ratios at varying temperatures and RH regimes was investigated. Specifically, percentage prey suppression efficiencies of *C. eruditus* and *C. malaccensis* were estimated and compared under different predator-to-prey ratios and thermo-hygrometric conditions (temperature and RH).

Materials and Methods

Predatory Mites. Laboratory stock cultures of *Acarus siro, Cheyletus eruditus*, and *Cheyletus malaccensis* were obtained from the Academy of State Administration of Grain, Beijing, China. *Acarus siro* was reared on a culture media consisting of a mixture of a wheat meal, oat flakes, and dried yeast (5:5:1) (wt/wt) and this mixture was milled (sieved) to a thickness of ~0.5 mm (hereafter referred to as mite diet) by sieving using a sieve with Tyler equivalent 35 mesh size (U.S. Standard #40 sieve (0.419-mm openings; Central Scientific Company., Chicago, IL). The *A. siro* cultures were started using ~500–1000 individuals in glass canning jars (360-ml) that had mite-proof lids and contained 200 g of mite diet in each. Jars were kept in plastic boxes (42 x 29 x 24 cm high) painted black with saturated KCl solution (Potassium Chloride, anhydrous, free-flowing, Redi-Dri^{Tm}, ACS reagent, ≥ 99%, 746436-2.5KG, Sigma-Aldrich, Inc.) beneath perforated false floors to maintain an RH of 85 ± 5% (RH was monitored by HOBO data loggers

(Onset Computers, Bourne, MA, USA) to facilitate population increase within 2–3 weeks and these mites were used as diet in starting and maintaining colonies of *C. eruditus* and *C. malaccensis*.

Laboratory cultures of *C. eruditus* and *C. malaccensis* originally obtained from Jiangxi and Harbin Provinces, China, respectively, were used for this study. Rearing methods for the two predatory mites were similar. Therefore, both species were cultured on *A. siro* but as pure cultures in 500-ml paper bags (lunch bags, chromated copper arsenate (cca), Albertson's Inc., Boise, ID 83726) which contained 200 g of lettuce seeds. The lettuce seeds were cleaned by sieving using a sieve of Tyler equivalent 20 mesh size (U.S. Standard #20 sieve (0.841-mm openings; Fisher Scientific Company, Pittsburgh, PA) and dried for two hours at 80 ± 1°C to prevent possible contamination by micro-arthropods and other unwanted organisms. Paper bags with lettuce seeds were placed on plates in 250-mm diameter desiccators (Thermo Fisher Scientific™ Nalgene™ Polypropylene Desiccator with Stopcock, 53100250, Pittsburgh, PA 15275) which had saturated KCl solution beneath perforated plates to maintain 85 ± 5% RH within the paper bags. After a week, ~1000–2000 individuals of *A. siro* were added, bags gently shaken, and the paper bags folded. The desiccators with paper bags were incubated at 24 ± 1°C and 0: 24 (L: D) photoperiod. After 24 hours, 25–50 individuals of *C. eruditus* or *C. malaccensis* were added to the contents in each paper bag and maintained for 3-4 weeks in order for the populations of predatory mites to increase. Adult female predatory mites were selected and used for this study. Given that both predatory mites are cannibalistic, cultures were frequently monitored and *A. siro* were added biweekly to

prevent decline in predatory mite populations because of starvation or conspecific predation in paper bags.

Liposcelis decolor. *Liposcelis decolor*, used as prey, was reared on a mixture of 93% cracked wheat (*Triticum aestivum* L.) (Duster variety), 5% Rice Krispies (Kellogg North America Company, Battle Creek, MI), and 2% wheat germ (The Quaker Oats Company, Chicago, IL) (wt/wt; referred to as psocid diet hereafter) in 360-ml glass canning jars with mite-proof lids (Opit and Throne 2008). The top one-third of the inner surface of each jar was coated with Fluon (polytetrafluoroethylene; Northern Products, Woonsocket, RI) to prevent psocids from accessing and gathering on the inside of the lid. Cultures of *L. decolor* were placed inside a growth chamber maintained at 30 ± 1°C and 0: 24 (L: D) photoperiod. Culture jars were placed in plastic boxes (42 x 29 x 24 cm high) painted black which had saturated NaCl solution (Sodium Chloride, anhydrous, free-flowing, Redi-Dri^{Tm}, ACS reagent, ≥ 99%, 746398-2.5KG, Sigma-Aldrich, Inc.) beneath perforated false floors to maintain a 75 ± 5% RH. Subsequently, adult females from established cultures were selected and used for this study.

Experimental Arenas. Experimental arenas consisted of two 6.0-cm diameter Petri dishes (forming a total cylindrical surface area of 84.82 cm^2; a total migration area of individual predator in a closed cylinder) (60 x 15 mm, Style Polystyrene, Becton Dickinson and Company, Franklin Lakes, NJ. USA) with one serving as a base and the other as a lid were used for this study. The top one-third of the inner surface of each basal Petri dish was coated with Fluon to confine prey within the bottom portion of the arena (thus prey were located within a 2/3 portion of the basal cylindrical arena with an area of 47.12 cm^2; a total migration area of individual prey), and a second Petri dish (a lid) was

inverted over the basal Petri dish to prevent the escape of predatory mites (by observation, *Cheyletus* spp. generally prefer a refuge to hide and feed on prey, but may escape from arenas without lids). The top and bottom Petri dishes were held together with sticky tape along 90% of the circumference of the meeting point of the basal and the upper Petri dishes. The 10% of the circumference not taped permitted air movement in and out of the basal Petri dishes. The basal Petri dishes contained 5.0 g of cracked wheat covering the entire bottom portion of each arena and was food for *L. decolor* (adult females).

Prey Population Suppression. Prey (adult female *L. decolor*) population suppression (levels by *C. eruditus* or *C. malaccensis* in 5.0 g grain samples per 84.83-experimental arenas were considered at different predator-to-prey ratios under varying temperatures and RH over a 40-day experimental period. Predator-to-prey ratios of 0:20, 1:20, 2:20, 4:20 and 10:20 were assigned to well-labeled experimental arenas. Adult females freshly molted from tritonymph (3-7-day-old) were selected from pure cultures of *C. eruditus* or *C. malaccensis* and were assigned separately to the experimental arenas. Twenty (20) female adults of *L. decolor* were introduced into each arena, however, with different predatory mite densities or numbers (0, 1, 2, 4, or 10), with zero (0) or a predator-to-prey ratio of 0:20 being the Control treatment. Predatory mites were starved for 24 h prior to introduction to their prey as this decreases initial variability in oviposition, standardizes hunger level, and initiates a nomadic period (Opit et al. 1997, Gerson et al. 2003, Kucerova 2004). The inoculated experimental arenas together with the Control treatments were randomly arranged in plastic boxes (42 x 29 x 24 cm) painted black, which had either $NaNO_2$ (Sodium nitrite, anhydrous, free-flowing, Redi-

Dri ™, ACS reagent, ≥ 97%, 799416-2.5KG, Sigma-Aldrich, Inc.), NaCl, or KCl saturated solution beneath perforated false floors to maintain 63, 75, or 85% RH, respectively, and these were placed inside growth chambers maintained at 20, 24, 28, or 32 °C for 40 days. The experimental design was a three-factor factorial Completely Randomized Design. Factors were predatory-prey ratio with five levels (0:20, 1:20, 2:20, 4:20, and 10:20), temperature with four levels (20, 24, 28, and 32 °C), and RH with three levels (63, 75, or 85%). However, analysis was performed as a two-factor factorial-CRD (a 5 x 4 or 5 x 3 factorial-CRD factorial design) and, each treatment was replicated three (3) times, except the Control treatment that was replicated six (6) times. Altogether there were 70 treatments (factor level combinations) for both *C. eruditus* and *C. malaccensis*. After 40 days, the number of surviving adults and nymphs of *L. decolor* in each treatment were counted to assess and compare prey suppression efficiency of *C. eruditus* or *C. malaccensis* under the different predator-to-prey ratios, temperatures, and RH. This was determined by comparing treatments with predators (1–10 density of a predatory mite species) against the Control treatment (only prey; no predator) under different temperatures (20, 24, 28, and 32 °C) and RH (63, 75, and 85%) after 40 days. Additionally, the reproductive responses of each predatory mite species were estimated by counting all the mobile stages of each predator species and using the data for estimating the per capita progeny production of *C. eruditus* or *C. malaccensis* under the different predator-to-prey ratios, temperatures, and RH.

Statistical Analysis. Statistical analyses were performed with SAS Version 9.4 (SAS Institute, Cary, NC). The mean percentage prey survival (%) and per capita progeny (%) were compared across the five predator-prey ratios (0:20, 1:20, 2:20, 4:20,

and 10:20), four temperatures (20, 24, 28, and 32°C), and three RH (63, 75, and 85%) using generalized linear mixed models methods for each of the predator species (*C. eruditus* and *C. malaccensis*). PROC GLIMMIX modeled the fixed effects of predator-prey ratio, temperature, and RH, and interactions for each of the response variables with the specified response distribution (~ Gaussian) in SAS. Data were analyzed using a square root transformation and a heterogeneous variances model since the response variables exhibited heterogeneity of variances. Least squares means were compared for the appropriate significant effects. All tests were conducted at the nominal 0.05 level of significance.

Results

Effects of predator-to-prey ratio, temperature, and RH on percentage survival of *L. decolor*. The results of percentage prey survival after 40 days of exposure of *L. decolor* to predatory mites showed that the three-way interaction of predator-to-prey ratio (hereafter referred to as release ratio), temperature, and relative humidity (RH), was significant ($p < 0.05$) for *C. eruditus* (Table 16). Percentage *L. decolor* survival increased with increasing temperature and RH along with decreasing release ratio, and was considerably higher at 28 and 32°C, 75% RH, and 0:20 release ratio. However, a significantly higher prey suppression level was mostly noticed under a combined effects of a higher release ratios (≥ 4:20), 20 – 24°C, and 63% RH (Table 17). Relative to the Control treatment (0:20), *C. eruditus* substantially suppressed *L. decolor* population size by~ 96.3%, 94.5%, 88.4%, and 67.1% in the 10:20, 4:20, 2:20, and 1:20 release ratios,

respectively, however, the estimated value for the 1:20 release ratio was significantly different from the 10:20, 4:20, and 2:20 release ratios under the combined temperature and RH effects (Table 17).

Similarly, the three-way interaction of release ratio, temperature, and relative humidity (RH), was significant ($p < 0.05$) for *C. malaccensis* (Table 16). *Liposcelis decolor* survival increased with increased in RH and with decreasing temperature and release ratio, and was considerably higher at 20 and 24°C, 85% RH, and 0:20 release ratio. However, a significantly higher prey suppression level was noticed under a combined effects of a higher release ratios (≥ 4:20), 28 – 32°C, and 63% RH (Table 17). At 10:20 release ratio, a complete prey suppression (~100%) was achieved by *C. malaccensis* under the interaction effects of 28 or 32°C and 63% RH. Relative to the Control treatment, *C. malaccensis* substantially suppressed *L. decolor* population size by ~ 97.2%, 95.0%, 83.9%, and 70.4% in the 10:20, 4:20, 2:20, and 1:20 release ratios, respectively, under the combined temperature and RH effects (Table 17).

In the absent of *C. eruditus* or *C. malaccensis* (thus, in the Control treatment; 0:20), prey (*L. decolor*) population growth was markedly higher in 32°C and 75% RH, and lowest in 20°C and 63% RH (Table 17). Thus, prey survival rate was significantly higher at 32°C and 75% RH which represents the optimal physical conditions for the growth and development of *L. decolor* (Table 17). The rapid growth rate of prey at 32°C and 75% RH may have contributed to the higher survival rate in the present of either *C. eruditus* or *C. malaccensis* at varying predator densities (Table 17).

Effect of predator-to-prey ratio, temperature, and RH on *C. eruditus* and *C. malaccensis* progeny production. In relation to per capita predator progeny production (percentage increased in predator's population after 40 days), the interaction of release ratio, temperature, and RH, was significant ($p < 0.05$) for both predatory mites, *C. eruditus* and *C. malaccensis* (Table 16). For *C. eruditus*, the maximum offspring production (excluding the egg stage) was observed at 24°C and 75 – 85% RH across all the tested release ratios (~179.8 – 2674.5% increase in population per predator) (Table 18), however, this was substantially lower at a combined interactions of 28°C and 63% RH (~ 0.0% offspring per predator) or 32°C and 75% RH (≤ 5.6% offspring per predator) across all the release ratios. Thus, a general trend of increasing progeny production with decreasing release ratio (≤ 1:10) and temperature (20 –24°C), and increasing RH (≥75%) was observed in *C. eruditus* (Table 18).

Similarly, for *C. malaccensis*, the highest progeny production was observed at 24°C and 75 – 85% RH across all the tested release ratios (~195.1 – 1444.4% increase in population per predator) (Table 18). However, no offspring of *C. malaccensis* was produced under a combined interactions of 28°C and 63% RH or 32°C and 63% RH across all the tested release ratios. Mostly, a trend of increasing progeny production with increase in RH (≥ 75%) and decreased in temperature (20 – 24°C) and release ratio was found in *C. malaccensis* (Table 18).

Discussion

The most significant result of this study was that *C. eruditus* and *C. malaccensis* were found to effectively prey on *L. decolor* and suppress its population in a wide range of ecological conditions. Our data has confirmed observations by several reports that both predatory mites are good potential natural enemies of most pestiferous insects in stored products including psocids in the genera, *Liposcelis* and *Lepinotus* (Rizk et al. 1979, Asanov 1980, Nangia et al. 1995, Kucerova 2004, Athanassiou and Palyvos 2006, Cebolla et al. 2009). Besides, *C. eruditus* (Cheyletin®) is the only biological agent used to control mite pests in food storage systems such as stored-grain mass and in grain residues, debris in empty stores, or seed stores (Zdarkova and Horak 1990, Zdarkova 1998). Therefore, these predatory mites can be used for disinfesting empty storehouses, pallets, and transportation containers of psocids through augmentation by inundative release to minimize pesticide used. This study has also showed that *C. eruditus* and *C. malaccensis* require a small number of *L. decolor* to complete their development, and parthenogenetically augment their offspring in grain under a wide range of release ratios (1:20 –10:20 predator-to-prey ratio), and use cannibalism to survive when *L. decolor* are absent (as observed in some of the replications). Schöller et al. (2006) reported similar biological features in the warehouse pirate bug *Xylocoris flavipes* (Reuter) (Hemiptera: Anthocoridae), one of the most-studied and efficient stored-grain predatory insect registered by the Environmental Protection Agency (EPA) for use against stored-product insect pests in the United States (Hagstrum and Subramanyam 2006).

The results also showed that *C. eruditus* and *C. malaccensis* can reproduce considerably under different release ratios in a diverse range of temperatures and relative humidities when fed on *L. decolor*. However, their optimal prey suppression capacities

and progeny production was mostly found in dissimilar biotic and abiotic conditions. For instance, previous research by Kucerova (2004) showed that *C. eruditus* prey on all developmental stages of *L. decolor* and significantly suppressed the prey population size in grain samples under laboratory conditions of 25°C and 85% RH, and in 40 days exposure duration. The author found that in 1:2 release ratio, the number of individual *L. decolor* decreased substantially from ~ 100 individuals in Control treatment to ~ 20 individuals in the predator treatments, representing ~80.0% population suppression; in the 1:5 released ratio, the population size decreased considerably from ~ 190 to ~ 30 individuals of *L. decolor*, representing ~84.2% population suppression. This range of *L. decolor* population suppression was consistent with the results of the current study, although a trend of increasing prey mortality with an increasing release ratio was found in the current study. Thus, *C. eruditus* substantially suppressed *L. decolor* population by 67.1–96.3% in 1:20–10:20 release ratios, whereas that of *C. malaccensis* was mostly higher with the estimated values of ~ 70.4– 97.2% in the range of 1:20–10:20 release ratios when compared with the Control population. A similar range of prey mortalities was reported when 13 natural enemies of 19 stored-products insect pest were assessed; a range of 70.0–100% prey suppression efficiency was reported (Hagstrum and Subramanyam 2006).

Based on the present study, it can be deduced that *L. decolor* mortality by *C. eruditus* and *C. malaccensis* would be low if a limited number of predators are released for biological control. However, predators released at a higher density would under-perform due to density-dependent factors such as competition, mutual interference, and cannibalism (Zhu et al. 2019). Therefore, establishing predator-prey balance either

spatially or temporally through accurate estimation of release ratio would be important for a successful biological control program. Again, this information is critical for commercial production of these cheyletids where lower release ratio is recommended for mass rearing and release of C. *eruditus* and *C. malaccensis* for psocids management. The exact critical *L. decolor* density limiting successful prey suppression by both predators was not established in the present study due to significant prey mortality by predators observed in all the release ratios (1:20–1:2 predator-prey ratio). However, it is expected that *C. eruditus* and *C. malaccensis* would effectively manage *L. decolor* populations even when the release ratio is as low as 1:20, and in a temperature range of 20–28°C, and 75% RH (an optimum growth and developmental RH for *L. decolor*). Both predators increased their progeny production with decreasing release ratio. Therefore, for inoculative release of *C. eruditus*, lower release ratios (≤ 1:10 predator-prey ratios) should be targeted where the predator's population can increase considerably to ~1174.2 – 2674.5% under a temperature of 24°C and 75 – 85% RH after 40 days of release. Likewise, with lower release ratios (≤ 1:10 predator-prey ratios) and temperature and RH range of 24°C and 75 – 85%, respectively, *C. malaccensis* population is expected to increase substantially to ~354.2 – 1444.4% after 40 days when managing psocid infestations. Nevertheless, ≤ 63% RH in the storage environment or rearing facilities can hinder the growth, development, and proliferation of both predatory mites.

Storage ecological factors influence the overall performance of biological control agents (Schöller et al. 2006). The contradictory trend between the current results and the work by Kucerova (2004) and the higher prey mortality in the present findings can probably be attributed to the influence of a wider range of temperature (20–32°C) and RH

(63–85%) used in the present work. Athanassiou et al. (2011) noted that the storage ecology of *C. malaccensis* in various commodities and in different types of storage facilities was mostly influenced by abiotic factors, such as temperature and moisture, and not by predation. Likewise, the variation in *C. eruditus* efficiency can be related to the existence of various biotypes of this *Cheyletus* spp. (Athanassiou and Palyvos 2015). The marginal differences in performances of *C. malaccensis* over *C. eruditus* in most of the release ratios can be explained by the texture of the medium used in the experimental arenas (coarse-wheat grain) which is mostly preferred by *C. malaccensis* over *C. eruditus*. Hubert et al. (2006) indicated that *C. eruditus* is more common in grain residues, while *C. malaccensis* is mostly found in grain mass because of its ability to penetrate bulk grain. Despite their co-existence in the storage environment (Zdarkova and Fejt 1999, Athanassiou et al. 2011), it is generally considered that *C. eruditus* is more adapted in tropical conditions, while *C. malaccensis* is more abundant in temperate regions (Athanassiou and Palyvos 2015). Again, the present study revealed a general trend of increasing progeny production size with decreased in release ratio and temperature, and increased in RH for both predatory mites. *Cheyletus eruditus* and *C. malaccensis* increased their population size considerably in the release ratio of 1:20, temperature range of 20–24 °C, and 75–85% RH. However, 63% RH proved to be a serious detrimental physical condition against predator survival and population growth across all the release ratios tested. This implies that RH would be the main limiting factor that can influence the level of control achieved by *C. eruditus* and/or *C. malaccensis* in any biological control program against psocid species especially, species such as *L obscura* that can survive in RH conditions below 63% (Opit et al. 2018). Therefore,

effective inundative release of these cheyletid mites should target the early part of the winter season when ambient or storage RH begins to rise and enables the proliferation of *C. eruditus* and *C. malaccensis* thereby enhancing their prey suppression efficiency (Solomon 1949, Schöller et al. 2006, Zhu et al. 2019).

The current study has provided information, for the first time, on the efficacy of cheyletid mites *C. eruditus* and *C. malaccensis* to manage psocids (*L. decolor*) in diverse thermo-hygrometric regimes under laboratory conditions. The capacity of both predatory mites to effectively suppress *L. decolor* populations was established while the progeny production by predators was significant under the tested biotic and abiotic conditions. Although the low 63% RH limited efficacy of these cheyletid mites, both predatory mites caused population suppression of ~ 67.1–97.2% and increased their progeny by ~117.1–1182.6% for the 1:20–10:20 release ratios, temperatures of 20 – 32°C, and 75 – 85% RH after 40 days of exposure to psocids infested grain. These temperature and RH ranges represent physical conditions that permit survival of psocids. The levels of psocid population suppression achieved indicate the good potential of both predatory mites for psocid management. Whereas laboratory assessment is a critical step along a continuum of screening and evaluation procedures for the selection of efficient biocontrol agents, the laboratory stimulations alone do not allow predictions of the success of predatory mites under the field conditions. Therefore, further assessment under storage ecological conditions (field trials) is needed. This should include wider release ratios, simultaneous interactions of predators, psocid species specificity, and applicability. Also, research on the compatibility of these cheyletids mites with other stored-product pest management strategies (especially, the effects of residual pesticides on survival of these mites) should

be conducted to enable integration of these cheyletids in storage IPM systems for psocid species management in the United States.

Acknowledgments

We thank Dr. Yi Wu, the Academy of State Administration of Grain, Beijing, China for providing the colonies of *Cheyletus eruditus*, *Cheyletus malaccensis*, and *Acarus siro* for this work. This research was funded by Oklahoma Agricultural Experiment Station (Project number OKL2949). Trade names or commercial products mentioned in this paper are solely for the purpose of providing specific information and do not imply recommendation or endorsement by Oklahoma State University.

Table 16. Summary of the tests for the fixed effects of predator-prey ratio (P-P-R), temperature (T), and relative humidity (RH), and interactions for percentage prey surviving (%) and per capita progeny production (% increase in population) of predatory mites, for *Cheyletus eruditus* (CE) or *Cheyletus malaccensis* (CM) exposed to initial prey density (20 females) of *L. decolor* over a 40-day period.

Variable	Predator sp.	Source	DF	*F*	*P* value
Prey survival	CE	T	3, 91.64	6.10	0.0008
		RH	2, 91.64	30.90	< 0.0001
		T*RH	6, 91.64	5.41	< 0.0001
		P-P-R	4, 57.25	249.09	< 0.0001
		T*P-P-R	12, 80.62	7.07	< 0.0001
		RH*P-P-R	8, 73.59	6.68	< 0.0001
		T*RH*P-P-R	24, 87.52	1.81	0.0250
	CM	T	3, 84.59	0.42	0.7383
		RH	2, 84.59	25.37	< 0.0001
		T*RH	6, 84.59	1.61	0.1539
		P-P-R	4, 60.7	469.66	< 0.0001
		T*P-P-R	12, 80.7	12.13	< 0.0001
		RH*P-P-R	8, 73.89	7.29	< 0.0001
		T*RH*P-P-R	24, 87.18	1.62	0.0539
Predator progeny	CE	T	3, 66.28	39.08	< 0.0001
		RH	2, 66.28	247.13	< 0.0001
		T*RH	6, 66.28	8.65	< 0.0001
		P-P-R	3, 49.45	43.95	< 0.0001
		T*P-P-R	9, 62.3	6.02	< 0.0001
		RH*P-P-R	6, 57.99	13.76	< 0.0001
		T*RH*P-P-R	18, 66.04	3.98	< 0.0001
	CM	T	3, 58.69	63.85	< 0.0001
		RH	2, 58.69	137.73	< 0.0001
		T*RH	6, 58.69	8.20	< 0.0001
		P-P-R	3, 46.72	10.35	< 0.0001
		T*P-P-R	9, 58.59	3.02	0.0050
		RH*P-P-R	6, 54.64	17.91	< 0.0001
		T*RH*P-P-R	18, 61.77	4.53	< 0.0001

Table 17. Mean percentage prey surviving (±SE) over a 40-day period. Predatory mite species (P) were *C. eruditus* (CE) or *C. malaccensis* (CM), initial prey density was 20 females of *L. decolor*, five levels of predator-to-prey ratio (P-P-R) (0:20, 1:20, 2:20, 4:20, and 10:20), four levels of temperature (T) (20, 24, 28, and 32°), and three levels of relative humidity (RH) (63, 75, and 85%).

P	T	RH	P-P-R				
			0:20	1:20	2: 20	4:20	10:20
CE	20	63	34.9 ± 0.25 aH	21.2 ± 0.17 bDE	6.4 ± 0.85 bC	9.4 ± 0.98 bAB	0.5 ± 0.57 cD
	20	75	58.7 ± 0.25 aDE	33.0 ± 0.17 bB	11.0 ± 0.85 cBC	5.4 ± 0.98 dBC	3.2 ± 0.57 eBC
	20	85	72.9 ± 0.25 aCD	21.2 ± 0.17 bDE	12.3 ± 0.85 bBC	5.4 ± 0.98 cBC	0.5 ± 0.57 dD
	24	63	42.4 ± 0.25 aGH	20.5 ± 0.17 bDE	6.6 ± 0.85 cC	4.1 ± 0.98 dCD	6.4 ± 0.57 dBC
	24	75	61.9 ± 0.25 aDE	24.7 ± 0.17 bCD	13.1 ± 0.85 cBC	6.4 ± 0.98 cB	2.2 ± 0.57 dCD
	24	85	64.1 ± 0.25 aDE	19.8 ± 0.17 bD	11.9 ± 0.85 bcBC	1.5 ± 0.98 dD	6.4 ± 0.57 cB
	28	63	44.6 ± 0.25 aFG	22.9 ± 0.17 bD	13.1 ± 0.85 bBC	3.2 ± 0.98 cD	1.6 ± 0.57 cBC
	28	75	204.7 ± 0.25 aA	50.9 ± 0.17 bA	11.4 ± 0.85 cBC	8.0 ± 0.98 cBC	8.0 ± 0.57 cAB
	28	85	95.0 ± 0.25 aBC	32.6 ± 0.17 bB	16.4 ± 0.85 cA	1.6 ± 0.98 dBC	1.1 ± 0.57 eCD
	32	63	69.0 ± 0.25 aDE	16. 4 ± 0.17 bE	2.2 ± 0.85 cD	0.5 ± 0.98 dD	0.5 ± 0.57 dD
	32	75	231.7 ± 0.25 aA	57.2 ± 0.17 bA	16.0 ± 0.85 cA	12.3 ± 0.98 cA	9.9 ± 0.57 cA
	32	85	110.0 ± 0.25 aB	37.8 ± 0.17 bB	6.6 ± 0.85 cC	1.6 ± 0.98 dD	0.5 ± 0.57 dD
CM	20	63	43.2 ± 0.18 aG	18.1 ± 0.24 bC	7.7 ± 0.94 cC	4.4 ± 1.02 cBC	7.7 ± 0.36 cAB
	20	75	63.5 ± 0.18 aF	34.6 ± 0.24 bA	21.4 ± 0.94 bA	9.6 ± 1.02 cA	3.2 ± 0.36 dBC
	20	85	72.7 ± 0.18 aEF	33.3 ± 0.24 bA	26.2 ± 0.94 bA	7.7 ± 1.02 cAB	6.5 ± 0.36 cAB
	24	63	57.9 ± 0.18 aFG	28.1 ± 0.24 bAB	14.7 ± 0.94 cBC	6.5 ± 1.02 dAB	1.7 ± 0.36 dC
	24	75	94.2 ± 0.18 aDE	27.7 ± 0.24 bAB	22.1 ± 0.94 bA	6.5 ± 1.02 cAB	0.6 ± 0.36 dC
	24	85	90.1 ± 0.18 aDE	36.5 ± 0.24 bA	28.0 ± 0.94 bA	6.5 ± 1.02 cAB	1.6 ± 0.36 dC
	28	63	59.6 ± 0.18 aFG	16.1 ± 0.24 bC	1.7 ± 0.94 cD	0.6 ± 1.02 cD	0.0 ± 0.36 dD
	28	75	164.7 ± 0.18 aB	37.9 ± 0.24 bA	19.3 ± 0.94 bAB	1.7 ± 1.02 cC	0.0 ± 0.36 dD
	28	85	113.4 ± 0.18 aCD	37.3 ± 0.24 bA	26.3 ± 0.94 bA	7.7 ± 1.02 cAB	0.6 ± 0.36 dC
	32	63	75.5 ± 0.18 aEF	16.1 ± 0.24 bC	9.6 ± 0.94 bC	1.7 ± 1.02 cC	0.0 ± 0.36 dD
	32	75	219.5 ± 0.18 aA	38.2 ± 0.24 bA	7.4 ± 0.94 cC	1.7 ± 1.02 dC	2.2 ± 0.36 dBC
	32	85	133.3 ± 0.18 aC	28.7 ± 0.24 bAB	6.5 ± 0.94 cC	4.4 ± 1.02 dBC	9.6 ± 0.36 cdA

Significant differences among P-P-R for each T*RH are denoted with different lower-case letters (within the same row) for each predator, and differences among T*RH for each P-P-R are denoted by different upper-case letters for each predator (within a column) under a given variable ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

Table 18. Mean per capita progeny production (%) (±SE) over a 40-day period. Predatory mite species (P) were *C. eruditus* (CE) or *C. malaccensis* (CM), initial prey density was 20 females of *L. decolor*, five levels of predator-to-prey ratio (P-P-R) (1:20, 2:20, 4:20, and 10:20), four levels of temperature (T) (20, 24, 28, and 32°), and three levels of relative humidity (RH) (63, 75, and 85%).

P	T	RH	P-P-R			
			1:20	2: 20	4:20	10:20
CE	20	63	240.3 ± 17.53 aE	64.8 ± 7.48 bE	16.2 ± 5.78 bD	51.3 ± 2.59 bD
	20	75	1310.3 ± 17.53 aBC	805.0 ± 7.48 bB	332.6 ± 5.78 cBC	323.3 ± 2.59 cAB
	20	85	1157.1 ± 17.53 aBC	761.3 ± 7.48 bB	307.9 ± 5.78 cBC	132.6 ± 2.59 dCD
	24	63	11.1 ± 17.53 aF	5.6 ± 7.48 aE	32.4 ± 5.78 aD	35.5 ± 2.59 aD
	24	75	1174.2 ± 17.53 aBC	1369.9 ± 7.48 aA	1059.0 ± 5.78 aA	179.8 ± 2.59 bBC
	24	85	2674.5 ± 17.53 aA	1697.8 ± 7.48 bA	750.6 ± 5.78 cA	307.6 ± 2.59 dAB
	28	63	0.0 ± 17.53 aF	0.0 ± 7.48 aE	0.0 ± 5.78 aD	0.0 ± 2.59 aD
	28	75	1068.9 ± 17.53 aBC	429.3 ± 7.48 bC	202.1 ± 5.78 cC	142.3 ± 2.59 cCD
	28	85	361.0 ± 17.53 aDE	359.5 ± 7.48 aCD	245.4 ± 5.78 aBC	115.4 ± 2.59 bCD
	32	63	0.0 ± 17.53 aF	0.0 ± 7.48 aE	0.0 ± 5.78 aD	5.6 ± 2.59 aD
	32	75	466.4 ± 17.53 aCD	74.0 ± 7.48 bD	58.9 ± 5.78 bCD	371.6 ± 2.59 aA
	32	85	1248.9 ± 17.53 aBC	503.4 ± 7.48 bB	364.7 ± 5.78 bB	139.7 ± 2.59 cCD
CM	20	63	11.1 ± 14.00 cE	154.3 ± 8.18 aC	5.6 ± 2.78 cCD	115.7 ± 1.15 bBC
	20	75	513.3 ± 14.00 aBC	281.5 ± 8.18 abB	237.7 ± 2.78 bcB	179.2 ± 1.15 cAB
	20	85	707.2 ± 14.00 aB	647.1 ± 8.18 abAB	335.2 ± 2.78 bAB	145.9 ± 1.15 cAB
	24	63	0.0 ± 14.00 bF	0.0 ± 8.18 bE	0.0 ± 2.78 bD	80.4 ± 1.15 aC
	24	75	354.2 ± 14.00 aBC	477.5 ± 8.18 aAB	411.4 ± 2.78 aA	195.3 ± 1.15 bAB
	24	85	1444.4 ± 14.00 aA	776.1 ± 8.18 bA	568.4 ± 2.78 bA	195.1 ± 1.15 cA
	28	63	0.0 ± 14.00 aF	0.0 ± 8.18 aE	0.0 ± 2.78 aD	0.0 ± 1.15 aD
	28	75	11.1 ± 14.00 bE	11.1 ± 8.18 bD	33.3 ± 2.78 bC	118.7 ± 1.15 aBC
	28	85	470.8 ± 14.00 aBC	162.9 ± 8.18 bC	162.9 ± 2.78 bBC	71.2 ± 1.15 cCD
	32	63	0.0 ± 14.00 aF	0.0 ± 8.18 aE	0.0 ± 2.78 aD	0.0 ± 1.15 aD
	32	75	129.5 ± 14.00 aCD	0.0 ± 8.18 cE	5.6 ± 2.78 bC	21.7 ± 1.15 bCD
	32	85	649.5 ± 14.00 aBC	5.6 ± 8.18 bE	0.0 ± 2.78 cD	10.0 ± 1.15 bCD

Significant differences among P-P-R for each T*RH are denoted with different lower-case letters (within the same row) for each predator, and differences among T*RH for each P-P-R are denoted by different upper-case letters for each predator (within a column) under a given variable ($P < 0.05$, LSMeans under Proc GLIMMIX in SAS).

CHAPTER VII

CONCLUSIONS

Psocids have become global pests of stored commodities due to considerable economic losses they inflict and the fact that they are not successfully managed by available pest management strategies. As a result of the regulatory phase-out of methyl bromide (CH_3Br), the ovicidal deficiencies of sulfuryl fluoride (SO_2F_2), and limited use of chloropicrin (CCl_3NO_2) (used for empty bin treatment only), phosphine (PH_3) remains the only effective pesticide among registered fumigants for use against stored-grain pests in the United States. Phosphine resistance and tolerance contribute to the increased importance of psocids as stored-product pests globally, however, there is currently no better substitute for this fumigant to meet domestic and international export phytosanitary requirements. Therefore, this work was conducted in the context of first, establishing phosphine resistance diagnostic doses (discriminating doses: DDs) for monitoring and managing the development of phosphine resistance in psocids. Secondly, to assess the effectiveness of natural enemies of psocids (cheyletid predatory mites) to provide a feasible alternative or to complement phosphine usage for managing psocids infestations.

The PH_3 diagnostic test allows determination of whether a tested population sample has resistant individuals or not in order for immediate management action (control, eradication, or quarantine) to be taken where appropriate. The current study establishes the levels of phosphine tolerance in laboratory susceptible strains of psocids

using laboratory fumigation methods. The psocid species, *L. bostrychophila*, *L. entomophila*, *L. decolor, L. paeta, L. rufa*, *L. obscura*, *L. fusciceps*, and *L. reticulatus* were exposed to incremental phosphine concentrations of 0–300.0 ppm and 5.0–150.0 ppm over a 20-h and 72-h fumigation period, respectively, at 25°C using a modified FAO Method No.16. Gas chromatographic-flame photometric detector (GC-FPD) quantification of average applied concentration of phosphine was used to achieve dose-mortality response data. Subsequently, interspecific differences in the levels of phosphine tolerance among tested species were established using lethal concentration ratios (LCRs) required to kill 50%, 95%, and 99% of individuals in test samples (LC_{50}, LC_{95}, and LC_{99}) over a 20-h and 72-h fumigation period. Moreover, phosphine concentration reduction indexes (PCRIs) were established as the proportions of discriminating dose estimates of 20-h to 72-h fumigation period of each species. This was to investigate the impact of extending fumigation exposure time on the level of discriminating doses for individual psocid species.

The biological control aspect of the current studies aims to provide quantitative data demonstrating the biological control potential of *C. eruditus* and *C. malaccensis* for managing psocids under simulated storage conditions. Knowledge about predatory characteristics of these cheyletid mites was required to understand their influence on the population dynamics of psocid infestations in storage communities. *Cheyletus eruditus* and *C. malaccensis* were assessed based on their foraging behaviors. Thus, functional and numerical responses of each predatory mite to varied densities (3, 6, 12, 18, 24, 40, and 50) of nymphs, adult males, and adult females of *L. decolor* were investigated under laboratory conditions at 24 ± 1°C and 0: 24 (L: D) photoperiod. Functional response data

were fitted by Holling (1959 a, 1959b) functional response curves whereas numerical response data were described by regression models to practically predict the predatory potential of each selected biocontrol candidate. In addition, the interactions between each predatory mite species and adult females of *L. decolor* were evaluated under different predator-to-prey release ratios (0:20, 1:20, 2:20, 4:20, or 10:20 predator-prey ratio), temperatures (20, 24, 28, or 32 °C), and relative humidities (RH) (63, 75, or 85%) over a 40-d exposure period under laboratory conditions of 0: 24 (L: D) photoperiod. This was to determine the optimal range of release ratios, temperatures, and r.h at which *C. eruditus* or *C. malaccensis* can successfully suppress psocid populations at different infestation levels by inundative release of predatory mites.

Based on dose-response studies using susceptible adults of *L. obscura, L. bostrychophila*, *L. paeta*, *L. fusciceps*, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila*, phosphine DDs for these species were 65.6, 77.5, 124.7, 149.4, 159.9, 205.0, 249.8, and 697.3 ppm over a 20-h fumigation exposure period. For the 72-h fumigation exposure period, the DDs for adults of *L. reticulatus, L. fusciceps, L. obscura*, *L. rufa*, *L. paeta*, *L. bostrychophila*, *L. entomophila*, and *L. decolor* were 18.1, 19.2, 23.2, 25.5, 39.6, 49.1, 157.1, and 194.5 ppm, respectively. Interspecific variations in dose-response mortality were found among psocid species over a 20-h or 72-h fumigation period. Based on the LC_{50} comparisons for 20-h exposure period, the concentrations of phosphine required for *L. bostrychophila*, *L. paeta*, *L.* fusciceps, *L. rufa*, *L. reticulatus*, *L. decolor*, and *L. entomophila* were 1.3, 1.7, 3.4, 3.8, 2.1, 2.2, and 9.3x, respectively, higher than those required to kill 50% of *L. obscura*. Similarly, at 72-h fumigation period, the concentrations of phosphine required for *L. fusciceps, L. obscura, L. rufa, L. paeta, L.*

bostrychophila, L. entomophila, and *L. decolor* were 2.0, 2.0, 3.7, 2.1, 1.9, 6.7, and 2.8x, respectively, more than dose required to kill 50% of *L. reticulatus*. The PCRI for *L. decolor*, *L. bostrychophila*, *L. obscura*, *L. paeta*, *L. entomophila*, *L. rufa*, *L. fusciceps*, and *L. reticulatus* were 1.28, 1.58, 2.83, 3.15, 4.44, 6.27, 7.78, and 11.36, respectively. Based on the FAO (1975) recommended 20-h fumigation period at 25°C, the present established DDs for adults of eight psocid species are by far the highest estimated diagnostic values among all studied stored-product insect pests. Also, *L. entomophila* and *L. decolor* were the most tolerant psocid species over a 20-h and 72-h fumigation period, respectively. In relation to LC_{50} mortality, *L. entomophila* required approximately 9.3 and 6.7x higher phosphine concentration compared to the most susceptible *L. obscura* and *L. reticulatus* over a 20-h and 72-h fumigation period, respectively. Moreover, this study has revealed heterogeneity in response to phosphine in psocids, particularly over a shorter phosphine fumigation period, a mechanism that facilitates the development of genetic-based resistance in both inter-and con-specifics storage insect pests. The higher heterogeneity levels in the standard 20-h fumigation period indicate the potential for a significant increase in phosphine resistance in field populations subjected to phosphine fumigation. Therefore, for effective control of all species in storage communities, it is most important that a concentration that controls the most phosphine tolerant species *L. entomophila* and *L. decolor* should be used. Discriminating doses estimated in this study can be used for detection of phosphine resistance and estimation of resistance frequencies in field-collected populations of the psocid species investigated.

Studies to assess the predatory efficiency of *C. eruditus* and *C. malaccensis* based on their functional responses to different developmental stages of *L. decolor* showed

that the functional responses of the two predatory mites to nymphs, adult males or adult females of *L. decolor* were Holling (1959a, 1959b) Type II. Subsequent estimations of attack rate (*a*), handling time (T_h), maximum predation (*K*), and predation efficiency (η) per day (d) for predators against prey life stages revealed that *C. eruditus* performance was preferable to *C. malaccensis* based on the aforementioned parameters. Similarly, the per capita consumption rate and searching efficiency of *C. eruditus* were considerably higher than *C. malaccensis* for all life stages of *L. decolor*. These predatory characteristics of *C. eruditus* suggest that this mite could be used effectively by inundative release to provide rapid psocid suppression in empty storage structures, grain residues, in stacked pallets, and other similar scenarios in storage environments. However, since there are no remarkable differences in prey specificity or nutritional requirements between these two predatory mites one can expect similar biocontrol characteristic when these mites are deployed as biocontrol agents in storage communities. Nevertheless, field trials would be required to validate these conclusions. Likewise, information on compatibility of predators with other management strategies targeted against stored-product pests is required in order to facilitate their incorporation into existing IPM systems for managing psocids.

The study on numerical responses of *C. eruditus* and *C. malaccensis* to nymphs, adult males, and adult females of *L. decolor* showed that both predatory mites can prey on psocids and produce significant numbers of offspring to suppress *L. decolor* populations. The present study revealed a general trend of a strong negative or positive correlation between oviposition rate and prey density for *C. eruditus* and *C. malaccensis*, respectively. Also, the oviposition efficiency was mostly similar for both predatory mites

and was inversely related to prey density. Moreover, the efficiency of conversion of ingested food (*L. decolor*) decreased considerably with increasing prey density across different prey types for both predators. The estimated numerical response variables (oviposition rate, oviposition efficiency, and efficiency of conversion of ingested food) indicate that *C. malaccensis* would be more efficient than *C. eruditus* in suppressing psocid infestations when long-term pest management (inoculative release) is the goal of a biocontrol program. For mass rearing of both predatory mites for inundative release, *L. decolor* or other psocids species can be used as a diet. The current study on foraging behaviors of the two predatory mites suggests that functional and numerical responses data are not sufficient for determining the effectiveness of predatory mites for use as biocontrol agents in psocid management. Therefore, further assessment procedures other than foraging behaviors must be explored in order to make more accurate decisions on selection and application of one or both of these predatory mites for managing stored-product psocid pests.

Based on results of ecological interaction of *C. eruditus* and *C. malaccensis* with *L. decolor* under different thermo-hygrometric regimes, suppression of *L. decolor* population when *C. eruditus*-related predator-to-prey ratios of 10:20, 4:20, 2:20, and 1:20 were used was ~ 96.3%, 94.5%, 88.4%, and 67.1%, respectively, relative to the Control (0:20). In the case of *C. malaccensis*, suppression of ~ 97.2%, 95.0%, 83.9%, and 70.4% for the 10:20, 4:20, 2:20, and 1:20 release ratios, respectively, under the combined temperature and RH effects. Although the low 63% RH limited the efficacy of these cheyletid mites, both predatory mites caused psocid mortalities of ~ 67.1–97.2% and increased their progeny by ~117.1–1182.6% for the 1:20–1:2 release ratios, temperatures

of 20–32°C, and 75–85% RH. High effective predation and reproductive rates of predators coincided with the optimal physical conditions for growth and development of psocids. These together with the levels of prey suppression capacities of predators indicate the good potential of these cheyletid mites for use against psocid infestations.

In conclusion, this research has added novel knowledge on diagnostic doses required to monitor and develop phosphine resistance management strategies for key psocid species infesting stored products in the United States. Data on biocontrol control—foraging behaviors of *C. eruditus* and *C. malaccensis* and their ecological interactions— provides pioneering quantitative information on potential for use of predatory mites as an alternative or to complement pesticide usage to manage psocids in the United States. Future research should be aimed at establishing discriminating doses for eggs of the eight studied psocid species to enhance effective phosphine resistance management of psocids. Also, field evaluation of both predatory mites is critical to permit their use against stored-product psocid pests in the United States.

REFERENCES

Afful, E., B. Elliott M. K., Nayak, and T. W. Phillips. 2018. Phosphine resistance in North American field populations of the lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae). J. Econ. Entomol. 111: 463–69.

Ahmedani, M. S., N. Shagufta, M. Aslam, and S. A. Hussnain. 2010. Psocid: A new risk for global food security and safety. App. Entomol. Zool. 45: 89 –100.

Ahn J., K. Kim, and J. H. Lee. 2010. Functional response of *Neoseiulus californicus* (Acari: Phytoseiidae) to *Tetranychus urticae* (Acari: Tetranychidae) on strawberry leaves. J. Appl. Entomol., 134: 98-104.

Ali M., A. A. Naif, and D. Huang. 2011. Prey consumption and functional response of a phytoseiid predator, *Neoseiulus womersleyi*, feeding on spider mite, *Tetranychus macfarlanei*. J. Insect Sci., 11: 167.

Al-Shammery, K. A. 2014. Influence of feeding on three stored product pests on rearing of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) in Hail, Saudi Arabia. Life Sci. J. 11 (5), 260–266.

Aminatou, B., S. Gautam, G. Opit, J. Talley, and K. Shakya. 2011. Population growth and development of *Liposcelis pearmani* (Psocoptera: Liposcelididae) at constant temperatures and relative humidities. Environ. Entomol. 40: 788–796.

Andersson, M., and S. Erlinge. 1977. Influence of predation on rodent populations. Oikos 29:591–597.

Arbogast, R. T., P. E. Kendra, D. K. Weaver, and D. Shuman. 2000. Insect infestation of stored oats in Florida and field evaluation of a device for counting insects electronically. J. Econ. Entomol. 93: 1035–1044.

Arthur, F. H., K. L. Hartzer, J. E. Throne, and P. W. Flinn. 2017. Freezing for control of stored-product psocids. J. Stored Prod. Res. 72: 166-177.

Asanov, K. 1980. Predators and parasites of the lesser grain borer *Rhyzopertha dominica*. Zashchita Rastenii 5, 23–24.

Athanassiou, C. G., and C. I. Rumbos. 2018. Emerging Pests in Durable Stored Products. In; C. G. Athanassiou and F. H. Arthur (eds.), *Recent Advances in Stored Product Protection*, https://doi.org/10.1007/978-3-662-56125-6_10.

Athanassiou, C. G., and N. E. Palyvos. 2015. The Cheyletoidea (Prostigmata), with special reference to potential of *Cheyletus malaccensis* Oudemans as biological control agent of post-harvest pests. In D. Carrillo et al. (eds.), Prospects for biological control of plant feeding mites and otter harmful organisms, Prog. Biol. Control 19, pp. 241.

Athanassiou, C. G, M. M. Hasan, T. W. Phillips, M. J, Aikins, and J. E. Throne. 2015. Efficacy of Methyl Bromide for Control of Different Life Stages of Stored-Product Psocids, *J. Econ. Entomol.* 108 (3): 1422–1428

Athanassiou, C. G., N. G. Kavallieratos, J. E. Throne, and C. T. Nakas. 2014. Competition among species of stored product psocids in stored grains (Psocoptera). PlosONE 9:e102867.

Athanassiou, C. G., N. G. Kavallieratos, A. Sciarretta, N. Palyvos, P. Trematerra. 2011. Spatial associations of insects and mites in stored wheat. J. Econ. Entomol. 104:1752–1764.

Athanassiou, C. G., F. H. Arthur, J. E. Throne, G. P. Opit, M. M. Hasan, M. J. Aikins, T. W. Phillips, and N. G. Kavallieratos. 2010. Efficacy of insecticides for control of stored-product psocid, pp: 805-814. *In*: Carvalho, O.M, P. G. Fields, C. S. Adler, F. H. Arthur, C. G. Athanassiou, J. F. Campbell, F. Fleurat-Lessard, P. W. Flinn, R. J. Hodges, A. A. Isikber, S. Navarro, R. T. Noyes, J. Riudavets, K. K. Sinha, G. R. Thorpe, B. H. Timlick, P. Trematerra, N. D. G. White (Eds), Proceedings of the 10th International Working Conference on Stored Product Protection, 27 June-2 July 2010, Estoril, Portugal, Julius-Kuhn-Archiv, Berlin, Germany, DOI: 10.5073/jka.2010.425.257

Athanassiou, C. G., F. H. Arthur, G. P. Opit, and J. E. Throne. 2009. Insecticidal effect of diatomaceous earth against three species of stored-product psocids on maize, rice, and wheat. J. Econ. Entomol. 102: 1673-1680.

Athanassiou, C. G., and N. E. Palyvos. 2006. Laboratory evaluation of two diatomaceous earth formulations against *Blattisocius keegani* Fox (Mesostigmata, Ascidae) and *Cheyletus malaccensis* Oudemans (Prostigmata, Cheyletidae). Biol. Control 38:350–355.

Athanassiou, C. G, N. G. Kavallieratos, N. E. Palyvos, and C. T. Buchelos. 2003. Three-dimensional distribution and sampling indices of insects and mites in horizontally-stored wheat. Appl. Entomol. Zool. 38:413–426.

Athanassiou, C. G., N. E., Palyvos, P. A., Eliopoulos, and G. T., Papadoulis. 2002. Mites associated with stored seed cotton and related products in Greece. Phytoparasitica 30(4):387–394.

Back, E. A. 1939. Psocids in dwellings. J. Econ. Entomol. 32: 419–423.

Baker, D. G. 2007. Flynn's parasites of laboratory animals. Second edition. American college of laboratory animal medicine. Blackwell Publishing, USA.

Baz, A. 2008. Bark-Lice, Book-Lice or Psocids (Psocoptera). Encyclopedia of Entomology. Springer Netherlands. 381-399.

Bazgir, F., J. Shakaram, S. Jafari. 2020. Functional response of the predatory mite *Amblyseius sweirskii* (Acari: Phytoseiidae) to *Eotetranychus frosti* (Tetranychidae) and *Cenopalpus irani* (Tenuipalpidae). Acarol. 60(1): 30 – 39.

Bell, C. H. 1976. The tolerance of developmental stages of four stored product moths to phosphine. J. Stored Prod. Res. 12: 77–86.

Bell C. H., N. Savvidou, K. A. Mills, S. Bradberry, and M. L. Barlow. 1994. A same day test for detecting resistance to phosphine. In *Proceedings of the 6th International Working Conference on Stored Product Protection*, ed. E Highley, E. J. Wright, HJ Banks, BR Champ, pp. 41–44.Wallingford, UK: CAB Int.

Benhalima, H., M. Q. Chaudhry, K. A. Mills, and N. R. Price. 2004. Phosphine resistance in stored-product insects collected from various grain storage facilities in Morocco. J. Stored Prod. Res. 40: 241–249.

Bucci, R. 1994. Effects of two insect growth regulators on the booklouse *Liposcelis bostrychophila*. J. Stored Prod. Res. 30: 157–161.

Cao, Y., Y. Son, and G. Y. Sun. 2003. A survey of psocid species infesting stored grain in China and resistance to phosphine in field populations of *Liposcelis entomophila* (Enderlein) (Psocoptera: Liposcelididae), pp. 662–667. In P. F. Credland, D. M. Armitage, C. H. Bell, P. M. Cogan, and E. Highley (eds.), Proceedings of the 8th International Working Conference on Stored Product Protection, 22–26 July 2002, New York, NY. CAB International, Wallingford, United Kingdom (ISBN 0851996914).

Castane, C., J. Amo, R. Gabarra, and O. Alomar. 2011. Plant damage to vegetable crops by zoophytophagous mirid predators. Biol. Cont. 59: 22-29.

Cato, A. 2015. Phosphine resistance in North American *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). M.S. thesis. Kansas State University, KS. (http://krex.k-state.edu/dspace/handle/2097/20337).

Cato, A. J., B. Elliott, M. K. Nayak, T. W. Phillips. 2017. Geographic variation in phosphine resistance among North American populations of the Red flour beetle (Coleoptera: Tenebrionidae). J. Econ. Entomol. 110 (3): 1359 – 1365.

Cebolla, R., S. Pekar, and J. Hubert. 2009. Prey range of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) and its efficacy in the control of seven stored-products pests — Biol. Cont., 50:1-6.

Cedola, C. V., N. L. Sanchez, and G. Lijesthrom. 2001. Effect of Tomato Leaf Hairiness on Functional and Numerical Response of *Neoseiulus californicus* (Acari: Phytoseiidae). Exp. Appl. Acarol. 25: 819-831.

Champ, B. R., and C. E. Dyte. 1976. FAO global survey of pesticide susceptibility of stored grain pests. FAO Plant Production and Protection Series 5, FAO, Rome, Italy.

Chaudhry, M. Q. 2000. Phosphine resistance. Pesticide Outlook 11: 88–91.

Chen, Z., D. Schlipalius, G. Opit, B. Subramanyam, and T. W. Phillips. 2015. Diagnostic molecular markers for phosphine resistance in U.S. populations of *Tribolium castaneum* and *Rhyzopertha dominica*. PLoS ONE 10: e0121343.

Chin, H. C., J. Jeffery, N. W. Ahmad, H. S. Kiang, B. Omar, H. Othman, and L. H. Lim. 2010. First Report of *Liposcelis bostrychophila* Badonnel (Psocoptera: Liposcelidae) as a Museum Insect Pest in Malaysia. Sains Malaysiana 39: 329-331.

Christopher, O. T. 2002. Firefly Encyclopedia of Insects and Spiders, pp-240. Firefly books, Willow dale, ON.

Clemmons, E. A., and D. K. Taylor. 2016. Booklice (*Liposcelis* spp.), Grain Mites (*Acarus siro*), and Flour Beetles (*Tribolium* spp.): 'Other Pests' Occasionally Found in Laboratory Animal Facilities. J. American Assoc. Lab. Ani. Sci. 55 (6): 737-743.

Collins, P. J., G. J. Daglish, M. K. Nayak, P. R. Ebert, D. Schlipalius, W. Chen, H. Pavic, T. M. Lambkin, R. Kopittke, and B. W. Bridgeman. 2001. Combating resistance to phosphine in Australia, pp. 593–607. In E. J. Donahaye, S. Navarro, and J. G. Leesch (eds.), International Conference Controlled Atmosphere and Fumigation in Stored Products, 29 October–3 November 2000, Fresno, CA. Executive Printing Services, Clovis, CA.

Collins, P. J., G. J. Daglish, H. Pavic, and K. A., Kopittke. 2005. Response of mixed-age cultures of phosphine resistant and susceptible strains of the lesser grain borer, *Rhyzopertha dominica*, to phosphine at a range of concentrations and exposure periods. J. Stored Prod. Res. 41:373–85.

Cuperus, G. W., R. T. Noyes, W. S. Fargo, B. L. Clary, D. C. Arnold, and K. Anderson. 1990. Management practices in a high-risk stored-wheat system in Oklahoma. Am. Entomol. 36: 129–134.

Daglish, G. J., P. J. Collins, H. Pavic, and K.A. Kopittke. 2002. Effects of time and concentration on mortality of phosphine-resistant *Sitophilus oryzae* (L.) fumigated with phosphine. Pest Manag. Sci. 58:1015–21.

Daglish, G. J., B. E Wallbank, and M. K. Nayak. 2003. Synergized bifenthrin plus chlorpyrifos-methyl for control of beetles and psocids in sorghum in Australia. l 96:525–32

Daglish, G.J. and P. J. Collins. 1999. Improving the relevance of assays for phosphine resistance. In *Proceedings of the 7th InternationalWorking Conference on Stored Product Protection*, ed. Z Jin,Q Liang, Y Liang, X Tan, L Guan, pp. 584–93.Wallingford, UK: CAB Int.

Diaz-Montano, J., J. F. Campbell, T. W. Phillips, and J. E. Throne. **2015**. Evaluation of potential attractants for six species of stored-product Psocids (Psocoptera: Liposcelididae, Trogiidae). J. Econ. Entomol. 108: 1398–1407.

Diaz-Montano, J., J. F. Campbell, T. W. Phillips, and J. E. Throne. 2018. Evaluation of Light Attraction for the Stored-Product Psocids, *Liposcelis entomophila*, *Liposcelis paeta,* and *Liposcelis brunnea.* J. Econ. Entomol. 111(3), 1476–1480.

Dong, P., J. J. Wang, F. X. Jia, and F. Hu. 2007. Development and reproduction of the psocid *Liposcelis tricolor* (Psocoptera: Liposcelididae) as a function of temperature. Ann. Entomol. Soc. Am. 100: 228-235.

Dou, W., G. M. Shen, J. Z. Niu, T. B. Ding, D. D. Wei, and J. J. Wang. 2013. Mining Genes Involved in Insecticide Resistance of *Liposcelis bostrychophila* Badonnel by Transcriptome and Expression Profile Analysis. PLoS ONE 8(11): e79878.

Emery, R. N., M. J. Nayak, and J. Holloway. 2011. Lessons learned from phosphine resistance monitoring in Australia. Stewart Postharvest Review. 7(3): 1 – 8.

Eliopoulos, P. A., C. G. Athanassiou, N. E. Palyvos., G. J. Stathas, and C. T. Buchelos. 2003. Dominance and frequency of predatory mites in stored products in Greece. In: Credland, P.F., Armitage, D.M., Bell, C.H., Cogan, P.M., Highley, E. (Eds.), Advances in Stored Product Protection. Proceedings of the Eighth International Working Conference on Stored Product Protection, York, UK, 22– 26 July 2002. CABI Publishing, Wallingford, UK, pp. 258–262.

Escudero, L.A., and F. Ferragut. 2005. Life-history of predatory mites *Neoseiulus californicus* and *Phytoseiulus persimilis* (Acari: Phytoseiidae) on four spider mite species as prey, with special reference to *Tetranychus evansi* (Acari: Tetranychidae). Biological Control 32, 378–384.

Fain, A. and A. V. Bochkov. 2001. A review of the genus *Cheyletus* Latreille, 1776 (Acari: Cheyletidae). Bulletin de l'Institut Royal des Sciences Naturelles de Belgique Entomologie 71, 83–113.

Farazmand, A., Y. Fathipour, K. Kamali, K. 2015. Intraguild predation among *Scolothrips longicornis* (Thysanoptera: Thripidae), *Neoseiulus californicus* and *Typhlodromus bagdasarjani* (Acari: Phytoseiidae) under laboratory conditions. Insect Sci. 22, 263–272.

Fathipour, Y. and B. Maleknia. 2016. Mite Predators. In: Omkar (Ed.), Ecofriendly Pest Management for Food Security. Elsevier, San Diego, USA, pp. 329-366.

Fathipour, Y., A. Hosseini, A. A. Talebi, S. Moharramipour. 2006. Functional response and mutual interference of *Diaeretiella rapae* (Hymenoptera: Aphidiidae) on *Brevicoryne brassicae* (Homoptera: Aphididae). Entomologica Fennica 17, 90–97.

Flinn, P. W. and D. W. Hagstrum. 1996. Suppression of beetles in stored wheat by augmentative releases of parasitic wasps. Environ. Entomol. **25**, 505 – 511.

Food and Agriculture Organization. 1975. Tentative method for adults of some major pest species of stored cereals with methyl bromide and phosphine, FAO method no. 16. FAO Plant Prot. Bull. 23: 12–25.

Finlayson, L. R. 1932. Some notes on the biology and life history of psocids. Entomol. Soc. Ontario Ann. Rpt. 63: 56–58.

Finney, D. J. 1952. Probit analysis: a statistical treatment of the sigmoid response curve, p. 318. Cambridge University Press. London, United Kingdom.

Franz, J. T., G. Masuch, H. Musken, and K. C. Bergmann. 1997. Mite fauna of German farms. Allergy 52(12):1233–1237.

Ganjisaffar F. and T. M. Perring. 2015. Prey stage preference and functional response of the predatory mite *Galendromus flumenis* to *Oligonychus pratensis*. Biol. Control, 82: 40-45.

Gautam, S. G., G. P. Opit, K. Shakya, and E. Hosoda. 2020. Phosphine resistance in saw-toothed grain beetle, *Oryzaephilus surinamensis* in the United States. J. Stored Prod. Res. 89: 101 – 690.

Gautam, S. G., G. P. Opit, and E. Hosoda. 2016. Phosphine resistance in adult and immature life stages of *Tribolium castaneum* (Coleoptera: Tenebrionidae) and *Plodia interpunctella* (Lepidoptera: Pyralidae) populations in California. J. Econ. Entomol. 109: 2525–2533. doi: 10.1093/jee/ tow221

Gautam, S. G., and G. P. Opit. 2015. Phosphine resistance in eggs of *Tribolium castaneum* and *Plodia interpunctella* from almond storage facilities in the Central Valley of California. The 10th Conference of the IOBC/ WPRS (OILB/SROP) Working Group on "Integrated Protection of Stored Products". Zagreb, Croatia. 28 June–1 July, 2015.

Gautam, S. G., G. P. Opit, K. L. Giles, and B. Adam. 2013. Weight loss and germination failure caused by psocids in different wheat varieties. J. Econ. Entomol. 106: 491-498.

Gautam, S. G., G. P. Opit, and K. L. Giles. 2010. Population Growth and Development of the Psocid *Liposcelis rufa* (Psocoptera: Liposcelididae) at Constant Temperatures and Relative Humidities. J. Econ. Entomol. 103: 1920-1928.

Gerson, U., R. Ochoa, and R. L. Smiley. 2003. Mites (acari) for pest control. 2nd ed. Oxford: Blackwell Science.

Gillot, C. 1995. *Entomology*. 2nd ed. Plenum Press, New York. 798 pp.

Grimaldi, D. and M. Engel. 2005. Evolution of the insects. European J. Entomol. 103 (1): 273 – 275.

Guedes, R. N. C., K. Y. Zhu, G. Opit, and J. Throne. 2008a. Differential heat shock tolerance and expression of heat-inducible proteins in two stored-product psocids. J. Econ. Entomol. 101: 1974-1982.

Guedes, R. N. C., J. F. Campbell, F. H. Arthur, K. Y. Zhu, G. P. Opit, and J. E. Throne. 2008b. Acute lethal and behavioral sublethal responses of two stored-product psocids to surface insecticides. Pest Manag. Sci. 64: 1314-1322.

Gunther K. **1974**. Staublause, Psocoptera. – Die Tierwelt Deutschlands, 61 Teil, VEB Gustav Fischer Verlag Jena: 314 pp.

Hagen, K.S., N. J. Mills, G. Gordh, and J. A. McMurtry. 1999. Terrestrial arthropod predators of insect and mite pests. In: Bellows, T.S., Fisher, T.W. (Eds.), Handbook of Biological Control. Academic Press, San Diego, pp. 383–503.

Hagstrum D. W., T. W. Phillips, and G. Cuperus. 2012. Introduction. In *Stored Product Protection*, ed. DW Hagstrum, T. W. Phillips, G Cuperus, pp. 1–5. Manhattan, KS: Kansas State Univ.

Hagstrum D. W and B. Subramanyam. 2006. *Fundamentals of Stored-Product Entomology*. St. Paul,MN: AACC International.

Haines, C. P. 1998. Arthropod natural enemies in stored product- overlooked and under-exploited. Proc. 7th Int. Work. Conf. Stored-product Prot., Beijing, 2:1205 – 1226.

Hansson, L., and H. Henttonen. 1985. Gradients in density variations of small rodents: the importance of latitude and snow cover. Oecologia 67:394–402.

Hassel, M. P. 1978. Dynamics of arthropod predator-prey systems. Princeton University Press. 237 pp.

Hassel, M. P J. H. Lawton and J. R. Beddigton. 1977. Sigmoid functional responses by invertebrate predators and parasitoids. J. Anim. Ecol. 46: 249–262.

Ho, S. H. and R. G. I. Winks 1995. The response of *Liposcelis bostrychophila* Badonnel and *L. entomophila* Enderlein (Psocoptera) to phosphine. J. Stored Prod.Res. 31: 191–197.

Holling, C. S. 1959a. The components of predation as revealed by a study of small-mammal predation of the European pine sawfly. Can. Entomol. 91: 293-320.

Holling, C. S. 1959b. Some characteristics of simple types of predation and parasitism. Can. Entomol. 91: 385-398.

Holling, C. S. 1961. Principles of insect predation. Ann. Rev. Entomol. 6: 163-182

Horn, T.B., J. Granich, J. H. Korbes, G. L., Da Silva and N. J. Ferla 2018. Mite fauna (Acari) associated with the poultry industry in different laying hen management systems in Southern Brazil: a species key. Acarologia 58(1): 140-158.

Hubert, J., Z. Mûnzbergova, Z. Kučerová, and V. Stejskal. 2006. Comparison of communities of stored product mites in grain mass and grain residues in the Czech Republic. Exp. Appl. Acarol. 39:149–158.

Hubert, J., J. Hyblova, Z. Munzbergova, S. Pekar, I. Krizova-Kudlı´kova, L. Doleckova- Maresova, V. Stejskal and M. Mares. 2007. Combined effect of an antifeedant a-amylase inhibitor and a predator *Cheyletus malaccensis* in controlling the stored-product mite Acarus siro. Physiological Entomology 32, 41–49.

Hubert, J., V. Stejskal, M. Nesvorna, R. Aulicky, J. Kopecky, and T. Erban. 2016. Differences in the bacterial community of laboratory and wild populations of the predatory mite *Cheyletus eruditus* (Acarina: Cheyletidae) and bacteria transmission from its prey Acarus siro (Acari: Acaridae). J. Econ. Entomol. 109 (3), 1450-1457.

Hughes, A. M. 1976. The mites of stored food and houses. Tech Bull Minist Agric Fish Food 9:1–400.

Jervis, M., N. and Kidd. 1996. Insect Natural Enemies. Practical Approaches to Their Study and Evaluation. Chapman & Hall, p. 491.

Jiadong, Y., L. Yanhui, X. Li, W. Mo, Z. Cong, W. Jing, T. Xin, Z. Zou, and B. Xia. 2019. Predation of *Cheyletus malaccensis* (Acari: Cheyletidae) on *Megoura japonica* (Hemiptera: Aphididae) under five different temperatures. Int. J. acarol. vol. 45 (3) 176–180.

Jiang, H., J. Liu, Z. I. Wang, and J. Wang. 2008. Temperature-dependent development and reproduction of a novel stored product psocid, *Liposcelis badia* (Psocoptera: Liposcelididae). Environ. Entomol. 37: 1105-1112.

Johnson K. P, K. Yoshizawa, and V. S. Smith. 2004. Multiple origins of parasitism in lice. Proc Biol Sci 271:1771–1776.

Juliano, S. 2001. Nonlinear curve fitting: predation and functional response curves. In: Scheiner, S.M., Gurevitch, J.(Eds.), Design and Analysis of Ecological Experiments. Oxford University Press, NewYork, pp. 178–196.

Kalinovic I., V. Rozman, and A. Liska. 2006. Significance and feeding of psocids (Liposcelididae, Psocoptera) with microorganisms, pp. 1087-1094. *In* I. Lorini, B. Bacaltchuk, H. Beckel, D. Deckers, E. Sundfeld, J. P. dos Santos, J. D. Biagi, J. C. Celaro, L. R. D‟A. Faroni, L de O. F. Bortolini, M. R. Sartori, M. C. Elias, R.N.C. Guedes, R. G. da Fonseca, and V. M. Scussel (Eds.). Proceedings of the 9th International Working Conference on Stored Product Protection, October 15-18, 2006, Campinas, São Paulo, Brazil. Brazilian Post-harvest Association – ABRAPOS,

Campinas, Brazil. Brazilian Post-harvest Association – Associação Brasileira de Pós-Colheita (ABRAPOS), Campinas, São Paulo, Brazil.

Kalinoski, R. M., and J. P., DeLong. 2016. Beyond body mass: how prey traits improve predictions of functional response parameters. Oecol. 180 (2), 543 – 550.

Kaur, R., M. Subbarayalu, R. Jagadeesan, G. J. Daglish, M. K. Nayak, H. R.Naik, S. Ramasamy, C. Subramanian, P. R. Elbert, and D. I. Schlipalius. 2015. Phosphine resistance in India is characterized by a dihydrolipoamide dehydrogenase variant that is otherwise unobserved in eukaryotes. Heredity 115: 188–194.

Kaur, R., and M. K. Nayak. 2015. Developing effective fumigation protocols to manage strongly phosphine-resistant *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae). Pest. Manag. Sci. 71: 1297–1302.

Kaur R, E. Daniels, M. K. Nayak, P. R. Ebert, and D. L. Schlipalius. 2013. Determining changes in the distribution and abundance of a *Rhyzopertha dominica* phosphine resistance allele in farm grain storages using a DNA marker. *Pest Manag. Sci.* 69:685–88

Koçak E., D. Schlipalius, R. Kaur, A. Tuck, P. Ebert. 2015. Determining phosphine resistance in rust red flour beetle, *Tribolium castaneum* (Herbst.) (Coleoptera: Tenebrionidae) populations from Turkey. *Türk. Entomol. Derg.* 39:129–36

Khanamani, M., Y. Fathipour, H. Hajiqanbar, and A. Sedaratian. 2014. Two-spotted spider mite reared on resistant eggplant affects consumption rate and life table parameters of its predator, *Typhlodromus bagdasarjani* (Acari: Phytoseiidae). Experimental and Applied Acarology 63, 241–252.

Kianpour, R., Y. Fathipour, K. Kamali, K., Omkar, 2011. Effects of mixed prey on the development and demographic attributes of a generalist predator, *Coccinella septempunctata* (Coleoptera: Coccinellidae). Biocontrol Science and Technology 21, 435–447.

Kleih, U., and V. Pike. 1995. Economic assessment of psocid infestations in rice storage. Trop. Sci. 35: 280-289.

Knülle, W., and R. R. Spadafora. 1969. Water vapor sorption and humidity relationships in *Liposcelis* (Insecta: Psocoptera). J. Stored Prod. Res. 5: 49-55.

Konemann C. E., Z. Hubhachen, G. P. Opit, S. Gautam, N. S. Bajracharya. 2017. Phosphine resistance in *Cryptolestes ferrugineus* (Coleoptera: Laemophloeidae) collected from grain storage facilities in Oklahoma, USA. *J. Econ. Entomol.* 110:1377–83.

Konemann, C. E., G. P. Opit, and S. G. Gautam. 2015. Establishing discriminating doses of phosphine for adults of three psocids (Psocoptera: Liposcelididae) species. Inte. Prot. Stored pro. IOBC-WPRS Bull. 3: 61-65

Konemann, C. E, G. P. Opit, K. Shakya, and N. S. Bajracharya. 2014. Phosphine resistance in rusty grain beetles, *Cryptolestes ferrugineus* (Stephens), (Coleoptera: Laemophloeidae) from stored wheat in Oklahoma, pp. 363-366. Proceedings of the International Organization for Biological and Integrated Control - West Palaearctic Region Section Working Group on Integrated Protection of Stored Products, July, Bordeaux, France. IOBC/WPRS Bulletin 82

Korunic, Z., P. Fields, M. Kovacs, J. Noll, O. Lukow, C. Demianyk, and K. Shibley. 1996. The effect of diatomaceous earth on grain quality. Post. Bio. Tech. 9: 373-387.

Kucerova, Z., 2004. Stored product psocids as one of the prey of the predatory mite Cheyletus eruditus (Schrank). IOBC/WPRS Bulletin 27, 191–195.

Kucerova, Z. 2002. Weight loss of wheat grains caused by psocid infestation (*Liposcelis bostrychophila*: Liposcelididae: Psocoptera). Plt. Proct. Sci. 38 (3): 103–107.

Kucerova, Z. 1999. Vulnerability of wheat varieties to stored-product psocids. pp 1251-1254. *In* Z. Jin, Q. Liang, Y. Liang, X. Tan, and L. Guan (Eds.), Proceedings of the 7th 24 International Working Conference on Stored Product Protection Beijing, China . Sichuan Publishing House of Science and Technology, Chengdu, China.

Kucerova, Z., M. Carvalho, and V. Stejskal. 2006. Faunistic records of new stored product psocids (Psocoptera: Liposcelididae) for Portugal. *In* I. Lorini, B. Bacaltchuk, H. Beckel, D. Deckers, E. Sundfeld, J. P. d. Santos, J. D. Biagi, J. C. Celaro, L. d. O. F. Bortolini, M. R. Sartori, M. C. Elias, R. N. C. Guedes, R. G. d. Fonseca and V. M. Scussel (eds.), Proceedings of the 9th International Working Conference on Stored-Product Protection. October 15-18, 2006, Campinas, São Paulo, Brazil. Brazilian Post-harvest Association – ABRAPOS, Campinas, Brazil. Brazilian Post-harvest Association – Associação Brasileira de Pós-Colheita (ABRAPOS), Campinas, São Paulo, Brazil.

Leesch, J. G., G. Cuperus, J. Criswell, J. Sargent, and J. Mueller. 1995. Practical fumigation considerations, chapter 19, pp. 139–152. In Stored product management. Oklahoma State University Circular No. E-912.

Leeuwen, E. V., V. A. A. Jansen, and P. W. Bright. 2007. How population dynamics shape the functional response in a one-predator–two-prey system. Ecology, 88(6), pp. 1571–1581.

LeOra Software. 2005. PoloPlus user's manual, version 2.0. LeOra Software, Petaluma, CA.

Lester, P. J., and R. Harmsen. 2002. Functional and numerical responses do not always indicate the most effective predator for biological control: an analysis of two predators in a two-prey system. J. Appl. Ecol. 39: 455-468.

Li, P. X., B. Xia, C. Shu, L. Zhong, A. H. Li. 2008. Predation of Amblyseius barkeri on *Aleuroglyphus ovatus*. Plant Prot. 34 (3), 65 – 68.

Lilley, D. E. 1981. Psocids—the food manufacturers problems. In *Psocids—A New Pest Problem in the Food Industry*, Symp. Soc. Food Hygiene Technol. pp. 11–17.

Lin, Y. C., M. L. Chan, C. W. Ko, and M. Y. Hsieh 2004. Nail infestation by *Liposcelis bostrychophila* Badonnel. Clin. Exp. Dermatol. 29:620-621.

Lienhard, C., and N. Smithers. 2002. Psocoptera (Insecta) world catalogue and bibliography. Instrumenta Biodiversitatis V, Museum d'histoire naturelle, Genève, Switzerland.

Liu, L. J., A. H. Pang, S. Q. Feng, B. Y. Cui, Z. H. Zhao, Z. Kucerova, V. Stejskal, G. Opit, R. Aulicky, Y. Cao, F. J. Li, Y. Wu, T. Zhang, and Z. H. Li 2017. Molecular Identification of ten species of stored-product psocids through microarray method based on ITS2 rDNA. Scientific reports. 7:16694. DOI: 10.1038/s41598-017-16888-z

Liu, L. 2018. A preliminary study on the growing development and predatory ability of *Cheyletus malaccensis* (Oudemans) [master's thesis]. Henan University of Technology.

Lividahl, J. P. and A. E. Stiven. 1983. Statistical difficulties in the analysis of predation functional response data. Can. Entomol, 115: 1365 – 1370.

Lord, J. and R.W. Howard. 2004. A proposed role for the cuticular fatty amides of *Liposcelis bostrychophila* (Psocoptera: Liposcelidae) in preventing adhesion of entomopathogenic fungi with dry-conidia. *Mycopathologia* 158: 211–217.

Lorini, I., P. J. Collins, G. J. Daglish, M. K. Nayak, and H. Pavic. 2007. Detection and characterization of strong resistance to phosphine in Brazilian *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae). Pest Manag. Sci. 63: 358–364.

Lovitt A. E and E. L. Soderstrom. 1968. Predation on Indian meal moth eggs by *Liposcelis bostrychophilus*. J. Econ. Entomol. 61:1444–45.

Lukas, J., V. Stejskal, V. Jarosik, and J.Hubert, J., E. Zdarkova, E. 2007. Differential natural performance of four *Cheyletus* predatory mite species in Czech grain stores. Journal of Stored Products Research 43, 97–102.

Maleknia, B., A. Z. Golpayegani. F. Farhoudi, S. R. Mirkhalilzadeh, and A. Allahyari. 2012. Effect of a heterospecific predator on the oviposition behavior of *Phytoseiulus persimilis*. Persian Journal of Acarology 1, 17–24.

Maleknia, B., A. Z. Golpayegani, A., Saboori,, and S. Magalhaes. 2013. Olfactory responses of *Phytoseiulus persimilis* to rose plants with or without prey or competitors. Acarologia 53, 273–284.

Maleknia, B., Y. Fathipour, and M. Soufbaf. 2015. Tritrophic Interactions in a System of Cucumber, Two-Spotted Spider Mite and Phytoseiid Mites (Ph.D. thesis). Tarbiat Modares University, p. 165.

Mashaya, N. 1999. Population dynamics of *Liposcelis entomophila* (Enderlein) (Psocoptera: Liposcelidae) in farm tobacco processing buildings. J. Stored Prod. Res. 35: 355-368.

Metz, J. A. J, M, W. Sabelis, J. H. Kuchlein. 1988. Sources of variation in predation rate at high prey densities: an analytic model and a mite example. Exptl. Appl. Acarol. 5: 187-205.

Meyer, J. R. 2005. “Psocoptera”. North Carolina State University. Archived from the original on 5 February 2007.

Mcfarlane, J. A. 1982. Damage to milled rice by psocids. Trop. Stored Prod. Inf. 44: 3-10.

McMurtry, J. A., G. J. D. Moraes, N. F. Sourassou. 2013. Revision of the lifestyles of phytoseiid mites (Acari: Phytoseiidae) and implications for biological control strategies. Systematic and Applied Acarology 18, 297–320.

Mockford, E. L. 1993. North American Psocoptera (Insecta), Sandhill Crane Press, Inc.

Mockford E., 1971. A new species of *Liposcelis* (Psocoptera, Liposcelidae) from North America with records of another species. J. Nat. Hist, 5: 263-270.

Murdoch, W. W. 1973. The functional response of predators. J. Appl. Ecol. 10: 335–341.

Mullen, G, R. and B. M. OConnor. 2019. Mites (Acari). In Medical and Veterinary Entomology (Third Edition).

Murdoch, W. W., and A. Oaten. 1975. Predation and population stability. Adv. Ecol. Res. 9:1–131.

Nangia, N., G. P. ChannaBasavanna, and P. S. Jagadish. 1995. The biology of *Cheyletus malaccensis* (Acari: Prostigmata) – a predator of primary acariforms in storage. Current Research University of Agricultural Science Bangalore 24, 13–15.

Nangia N, G. Channabasavanna, and P. Jagadish. 1994. A predator of primary acariforms in storage and its biology. Curr Res Univ Agric Sci Bangalore India 23(10):116–118

Nayak, M. K., G. J. Daglish, T. W. Phillip and P. R. Ebert. 2020. Resistance to the Fumigant Phosphine and Its Management in Insect Pests of Stored Products: A Global Perspective. Annu. Rev. Entomol. 2020. 65:333 –50.

Nayak, M. K., G. J. Daglish, and T. W. Phillips. 2015. Managing resistance to chemical treatments on stored product pests. Stewart Postharvest Rev. 1: 2

Nayak M. K., P. J. Collins, J. E. Throne, J. J. Wang. 2014. Biology and management of psocids infesting stored products. Annu. Rev. Entomol. 59:279–97.

Nayak, M. K., J. C. Holloway, R. N. Emery, H. Pavic, J. Bartlet, and P. J. Collins. 2013. Strong resistance to phosphine in the rusty grain beetle, *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae): its characterization, a rapid assay for diagnosis and its distribution in Australia. Pest Manag. Sci. 69: 48–53

Nayak, M. K., J. C. Holloway, R. N. Emery, H. Pavic, J. Bartlet, and P. J. Collins. 2012. Strong resistance to phosphine in the rusty grain beetle *Cryptolestes ferrugineus* (Stephens) (Coleoptera: Laemophloeidae): Its characterization, a rapid assay for diagnosis and its distribution in Australia. Pest Manag. Sci. 69: 48–53.

Nayak, M. K. 2010. Potential of piperonyl butoxide-synergised pyrethrins against psocids Psocoptera: Liposcelididae) for stored-grain protection. (www.interscience.wiley.com) DOI 10.1002/ps.1873

Nayak M. K., and P. J. Collins. 2008. Influence of temperature and humidity on toxicity of phosphine against strongly resistant *Liposcelis bostrychophila* Badonnel (Psocoptera: Liposcelididae), a cosmopolitan pest of stored commodities. Pest Manag. *Sci.* 64:971–76.

Nayak, M. K., and G. J. Daglish. 2007. Combined treatments of spinosad and chlorpyrifos-methyl for management of resistant psocid pests (Psocoptera: Liposcelididae) of stored grain. Pest Manag. Sci. 63:104-109.

Nayak, M. K. 2006. Psocid and mite pests of stored commodities: small but formidable enemies, pp. 1061-1073. *In* I. Lorini, B. Bacaltchuk, H. Beckel, D. Deckers, E. Sundfeld, J. P. dos Santos, J. D. Biagi, J. C. Celaro, L. R. D‟A. Faroni, L de O. F. Bortolini, M. R. Sartori, M. C. Elias, R.N.C. Guedes, R. G. da Fonseca, and V. M. Scussel (Eds.), Proceedings of the 9th International Working Conference on Stored Product Protection, October 15-18, 2006, Campinas, São Paulo, Brazil. Brazilian Post-harvest Association – Associação Brasileira de Pós-Colheita (ABRAPOS), Campinas, São Paulo, Brazil.

Nayak, M. K., G. J. Daglish and V. S. Byrne, 2005. Effectiveness of spinosad as a grain protectant against resistant beetle and psocid pests of stored grain in Australia. J. Stored Prod. Res. 41: 455–467.

Nayak, M. K., P. J. Collins, H. Pavic, and R. A. Kopittke. 2003. Inhibition of egg development by phosphine in the cosmopolitan pest of stored products *Liposcelis bostrychophila* (Psocoptera: Liposcelididae). Pest Manag. Sci. 59: 1191-1196.

Nayak, M. K., P. J. Collins, and H. Pavic. 2002a. Resistance to phosphine in psocids: Challenges ahead!, pp. 113-118. In E. J. Wright, H. J. Banks, and E. Highley (Eds.), Proceedings of the 2nd Australian Postharvest Technical Conference, August 1-4, 2000, Adelaide, Australia. Commonwealth Scientific and Industrial Research Organization - CSIRO, Adelaide, Australia.

Nayak, M. K., P. J. Collins, and H. Pavic. 2002b. Long-term effectiveness of grain protectants and structural treatments against *Liposcelis decolor* (Pearman) (Psocoptera: Liposcelididae), a pest of stored products. Pest Manag. Sci. 58: 1223-1228.

Nayak, M. K., P. J. Collins and H. Pavic 2000. Resistance to phosphine in psocids: challenge ahead! In *2nd Australian Postharvest Technical Conference* (E. J. Wright, H. J. Banks and E. Highley, eds.). CSIRO-SGRL, Adelaide, Australia, pp. 113–118.

Nayak, M. K., P. J. Collins, and S. R. Reid. 1998. Efficacy of grain protectants and phosphine against *Liposcelis bostrychophila, L. entomophila, and L. paeta* (Psocoptera: Liposcelididae). J. Econ. Entomol. 91: 1208-1212.

Nguyen, D.T., V. Bouguet, T. Spranghers, D. Vangansbeke, and P. De Clercq. 2015. Beneficial effect of supplementing an artificial diet for *Amblyseius swirskii* with *Hermetia illucens* haemolymph. J. Appl. Ento. 139, 342–351.

Obr, S. 1978. Psocoptera of food-processing plants and storages, dwellings, and collections of natural objects in Czechoslovakia. Acta Entomol. Bohemoslov. 75: 226-242.

Ocran, A. F., G. P. Opit, B. H. Noden, F. H. Arthur, and B. M. Kard. 2021. Effects of Dehumidification on the Survivorship of Four Psocid Species, *J. Econ. Entomol*. Pp. 1 – 9.

Omkar and A. Pervez. 2004. Functional and numerical responses of *Propylea dissecta* (Col., Coccinellidae). J. Appl. Ento. 128, 140–146.

O'Neil, R. J. 1990. Functional response of arthropod predators and its role in the biological control of insect pests in agricultural systems. In R.R. Baker and P. E. Dunn, eds. New directions in biological control: alternative for suppressing agricultural pests and diseases: proceedings of a UCLA colloquium, held at Frisco, Colorado, January 20 – 27, 1989. Pp. 83 – 96

O'Neil, R. J. 1989. Functional response of arthropod predators and its role in the biological control of insect pests in agricultural systems. In R.R. Baker and P. E. Dunn, eds. New directions in biological control: alternative for suppressing agricultural pests and diseases: proceedings of a UCLA colloquium, held at Frisco, Colorado, January 20 – 27, 1989. Pp. 83 – 96

Opit, G. P. 1986. Predation of two-spotted spider Mites, *Tetranychus urticae* Koch (Acari: Tetranychidae) by the predatory midge*, Feltiella minuta* Felt (Diptera: Cecidomyiidae). Ms. Thesis submitted to the department of Biological Sciences, Simon Fraser University. Pp. 1–122.

Opit, G. P., A. Ocran, and K. Shakya. 2018. Population growth and development of *Liposcelisobscura* Broadhead (Psocodea: Liposcelididae) at constant temperatures and relative humidities, pp. 151–159. In C.S. Adler, G. Opit, B. Fürstenau, C. Müller-Blenkle, P. Kern, F.H. Arthur, C.G. Athanassiou, R. Bartosik, J. Campbell, M.O. Carvalho, W. Chayaprasert, P. Fields, Z. Li, D. Maier, M. Nayak, E. Nukenine, D. Obeng-Ofori, T. Phillips, J. Riudavets, J. Throne, M. Schöller, V. Stejskal, H. Talwana, B. Timlick, P. Trematerra (eds.) Proceedings of the 12th International Working Conference on Stored Product Protection (IWCSPP). Berlin, Germany, 7–11 October 2018. Julius Kuhn Institute, Berlin, Germany.

Opit, G. P., T. W. Phillips, M. J. Aikins, and M. M. Hasan. 2012. Phosphine resistance in *Tribolium castaneum* and *Rhyzopertha dominica* from stored wheat in Oklahoma. J. Econ. Entomol. 105: 1107–1114.

Opit, G. P., J. E. Throne, and P. W. Flinn. 2009. Evaluation of Five Sampling Methods for the Psocids *Liposcelis entomophila* and *L. decolor* (Psocoptera: Liposcelididae) in Steel Bins Containing Wheat. J. Econ. Entomol. 102: 1377-1382.

Opit, G. P. and J. E. Throne. 2008. Population growth and development of the psocid *Lepinotus reticulatus* at constant temperatures and relative humidities. J. Econ. Entomol. 101: 605–615.

Opit, G. P., B. Roitberg, and D. R. Gillespie. 1997. The functional response and prey preference of *Feltiella acarisuga* (Vollot) (Diptera: Cecidomiidae) for two of its prey: male and female two-spotted spider mites, *Tetranychus urticae* Koch (Acari: Tetranychiidae). Can. Entomol. 129: 221-227.

Pacheco, I. A., M. R. Sartori and R. W. D. Taylor. 1990. Levantamento de resiste ncia de insetos-pragas de graos armazenados a fosna no Estado de Sao Paulo. *Coletanea de Institute de Tecnologia de Alimentos* 20: 144–154.

Pakyari, H., Y. Fathipour, M. Rezapanah, K. Kamali. 2009. Temperature-dependent functional response of *Scolothrips longicornis* (Thysanoptera: Thripidae) preying on *Tetranychus urticae*. J. Asia-Pacific Entomol. 12, 23–26.

Palyvos, N. E. and N. G. Emmanouel. 2011. Reproduction, survival, and life table parameters of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) at various constant temperatures. Exp Appl Acarol 54:139–150.

Palyvos N, E., N. G. Emmanouel, and C. J. Saitanis. 2008. Mites associated with stored products in Greece. Exp Appl. Acarol 44(3):213–226.

Palyvos, N. E. and N. G. Emmanouel. 2009. Temperature-dependent development of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae). Exp. Appl. Acarol. 47:147–158.

Pascual-Villalobos, M. J. 2005. Occurrence of psocids and natural predators in organic rice in Callaspara (Murcia, Spain). J. Stored Prod. Res. 41:231–235.

Patel, K. and Z. Q. Zhang. 2017. Prey preference and reproduction of predatory mites, *Amblybromalus limonicus* and *Neoseiulus cucumeris,* on eggs of and 1st instar nymphs of the Tomato/Potato Psyllid. *International Journal of Acarology*, 43(6), 468–474.

Pekar, S. and J. Hubert. 2008. Assessing biological control of Acarus siro by *Cheyletus malaccensis* under laboratory conditions: effect of temperatures and prey density. Journal of Stored Product Research 44, 335–340.

Pekar, S. and E. Zˇ d'árková´. 2004. A model of the biological control of Acarus siro by *Cheyletus eruditus* (Acari: Acaridae, Cheyletidae) on grain. Journal of Pest Science 77, 1–10.

Pervez A. and Omkar. 2006. Ecology and biological control application of multicoloured Asian ladybird, *Harmonia axyridis*: A review. Biocontrol Sci. Technol., 16: 111-128.

Phillips, T., E. Thoms, J. DeMark, and S. Walse. 2012. Fumigation, pp. 157–178. In D. W. Hagstrum, T. W. Phillips, and G. Cuperus, Stored-product protection. KState Research and Extension publication S156.

Phillips, T.W., and J. E. Throne. 2010. Biorational approaches to managing stored-product insects. Annu. Rev. Entomol. 55: 375-397.

Pike, V. 1994. Laboratory assessment of the efficacy of phosphine and methyl bromide fumigation against all life stages of *Liposcelis entomophila* (Enderlein). *Crop Protect.* 13:141-45

Pike, V., D. Rees, and R. Hatch. 1991. Bionomics of *Liposcelis paetus* in stored grain (Psocoptera: Liposcelidae). In *Proc. 5th Int. Working Conf. Stored Prod. Prot.*, ed. F Fleurat-Lessard, P Ducom, pp. 175–79. Bordeaux, Fr.: Impr. M´edoc

Pinto Jr., A. R., R. S. Furiatti, P. R. V. S. Pereira and F. A. Lazzari. 1997. Avaliaao de Inseticidas no Controle de *Sitophilus oryzae* L. (Coleoptera: Curculionidae) e *Rhyzopertha dominica* Fab. (Coleoptera: Bostrichidae) em Arroz Armazenado. Anais da Sociedade Entomologica do Brasil. 26: 285–290.

Pinniger, D. 1985. Recent research on psocids. In: Aspects of pest control. Symp. Soc. Food Hygiene Technol. pp. 57-65.

Pranata, R., C. Haines, R. Roesli, and D. Sunjaya. 1983. Dust admixture treatment with permethrin for the protection of rough rice and milled rice, pp. 132-136. *In*, N. C. Teter, R. N. Sample and A. S. Frio (eds.), Maintaining Good Grain Quality, Proceedings of the 6th Annual Workshop on Grain Post-harvest Technology. Bogor, Indonesia.

Pulpan, J. and P. H., Verner. 1965. Control of tyroglyphoid mites in stored grain by the predatory mite *Cheyletus eruditus* (Schrank). Can. J. Zool. 43:417–432.

Rahman, V. J., A. Babu, A. Roobakkumar, and K. Perumalsamy. 2012. Functional and numerical responses of the predatory mite, *Neoseiulus longispinosus* to the red spider mites, *Oligonychus coffeae*, infesting tea. J. Insect Sci. 12: 125.

Rahman, H., Y. Fathipour, and K. Kamali. 2009a. Life history and population growth parameters of (Acari: Phytoseiidae) fed on *Thrips tabaci* (Thysanoptera: Thripidae) in laboratory conditions. Syst. Appl. Acarol. 14, 91–100.

Rahman, M. M., W. Islam and K. N. Ahmed. 2009b. Functional Response of the Predator *Xylocoris flavipes* to three Stored Product Pests. Int. J. Agric. Biol., 11: 316–320

Rajendran, S. 1994. Psocids in food commodities and their control. Pestology 28:14–19

Rees, D. P. 1998. Psocids as pests of Australian grain storages, pp. 46–51. *In* H. J. Banks, E. J. Wright, and K. A. Damcevski (eds.), Stored Grain in Australia. Proceedings of the Australian Postharvest Technical Conference, May 26–29, 1998. Canberra, Australia

Rees, D. P. 1998. Psocids as pests of Australian grain storages, pp. 46–51. *In* H. J. Banks, E. J. Wright, and K. A. Damcevski (eds.), Stored Grain in Australia.

Rees, D., 2004. Insects of Stored Products. CSIRO Publishing, Collingwood, Victoria, Australia.

Rees, D. P. 2003. Psocoptera (psocids) as pests of bulk grain storage in Australia: a cautionary tale to industry and researchers, pp. 59-64. *In* P. F. Credland, D. M. Armitage, C. H. Bell, P. M. Cogan, and E. Highley (eds.), Advances in stored product protection, Proceedings of the 8th International Working Conference on Stored Product Protection, 22-26 July 2002, York, UK. CAB International, Wallingford, UK.

Reichmuth, C. 1991. A quick test to determine phosphine resistance in stored products research. GASGA Newsl. 15:14–15.

Rijckaert, G., J. Van Bronswijk, and H. Linskens. 1981. House-dust community (Fungi, Mites) in different climatic regions. Oecol. 48: 183-185.

Rizk, G.N., E. El-Badry, S. M. Hafez. 1979. The effectiveness of predacious and parasitic mites in controlling *Tribolium confusum* Duv. Mesopotamia Journal of Agriculture 14, 167–182.

Robertson, J. L., R. M. Russell, H. K. Preisler, and N. E. Savin. 2007. Bioassays with arthropods, p. 199, 2nd ed. CRC, Boca Raton, FL.

Roesli, R., R. Jones, and D. Rees. 1998. Factors affecting outbreaks of *Liposcelis* (Psocoptera: Liposcelidae) populations in grain storage, pp. 14-19. *In* J. Zuxun, L. Quan, L. Yongsheng, T. Xianchang and G. Lianghua (eds.), Proceedings of the 7th international working conference on stored-product protection. 14–19 October 1998, Chengdu, China. Sichuan Publ. House Sci. Technol.

Rosenheim, J. A. 2005. Intraguild predation of *Orius tristicolor* by *Geocoris* spp. and the paradox of irruptive spider mite dynamics in California cotton. Biol. Control 32, 172–179.

Rosomer, W. S., and J. G. Stoffolana. 1997. The Science of Entomology. Dubuque, IA: William C. Brown Pub.

Ruden, B. 2003. *Pest Management Strategic Plan for Northern Wheat South Dakota Project*. North Central IPM Center, S. Dakota State Univ., USA. 83 pp.

Ryoo, M. 1986. Studies on the basic component of the predation of Phytoseiulus persimilis (Acari: Phytoseiidae). Res. Pop. Ecol. 28:17–26.

Sandness J. N. and J. A. McMurtry. 1970. Functional response of three species of Phytoseiidae (Acarina) to prey density. Can. Entomol. 102: 692-704.

Santos, M. A. 1975. Functional and numerical responses of the predatory mite, *Amblyseius fallacis*, to prey density. Environ Entomol 4:989–992.

Santoso, T., Sunjaya, O. Dharmaputra, H. Halid, and R. Hodges. 1996. Pest management of psocids in milled rice stores in the humid tropics. Int. J. Pest Manag. 42: 189-197.

Sartori, M. R., I. Pacheco, and R.M.G. Vilar. 1990. Phosphine resistance in stored-grain insects in Brazil, pp. 1041–1050. In: F. Fleurrat-Lessard and P. Ducom (eds.), Proceedings of the 5th International Working Conference on Stored-Product Protection, September 9–14, 1990, Bordeaux, France. Imprimerie du Me´doc, Bordeaux, France, 1991.

Schlipalius, D. I, A. G. Tuck, H. Pavic, G. J. Daglish, M. K. Nayak, and P. R. Ebert. 2018. A high-throughput system used to determine frequency and distribution of phosphine resistance across large geographical regions. Pest Manag. Sci. 75:1091–98.

Schlipalius, D. I, N. Valmas, A. G. Tuck, R. Jagadeesan, L. Ma. 2012. A core metabolic enzyme mediates resistance to phosphine gas. *Science* 338:807–10.

Schlipalius, D. I., Q. Cheng, P. E. B. Reilly, P. J. Collins, P. R. Ebert. 2002. Genetic linkage analysis of the lesser grain borer *Rhyzopertha dominica* identifies two loci that confer high-level resistance to the fumigant phosphine. Genetics 161:773–82.

Schöller, M. E., P. W. Flinn, M. J. Grieshop, E. Zd'árková. 2006. Biological control of stored product pests. In: Heaps, J.W. (Ed), Insect Management for Food Storage and Processing Second Edition. American Association of Cereal Chemistry International, St Paul, MN, USA, pp. 67-87.

Sedlacek, J. D., P. A. Weston and R. J. Barney 1996. Lepidoptera and Psocoptera. In *Integrated Management of Insects in Stored Products* (B. Subramanyam and D. W. Hagstrum, eds.). Marcel Dekker Inc., New York, pp. 63–66.

Seiedy, M., A. Saboori, H. Allahyari, R. Talaei-Hassanloui, M. Tork. 2012. Functional response of *Phytoseiulus persimilis* (Acari: Phytoseiidae) on untreated and *Beauveria bassiana*-treated adults of *Tetranychus urticae* (Acari: Tetranychidae).Journal of Insect Behavior 25, 543–553.

Sekhon, R. K., M. W. Schilling, T. W. Phillips, M. J. Aikins, M. M. Hasan, A. Corzo, and W. B. Mikel. 2010. Effects of phosphine and methyl bromide fumigation on the volatile flavor profile and sensory quality of dry cured ham. Meat Sci. 86: 411-417.

Silva, G.L., N. J. Ferla, M. Toldi.,and D, C, C. Faleiro. 2013. Mite fauna (Acari) associated to commercial laying hens and bird nests in Vale do Taquari, Rio Grande do Sul, Brazil. Rev. Biotemas 26, 255e264.

Sengonca, C., T. Zegula, P. Blaeser. 2004. The suitability of twelve different predatory mite species for the biological control of *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae). Zeitschrift fur Pflanzenkrankheiten und Pflanzenschutz 4, 388–399.

Sepulveda, F. and R. Carrillo. 2008. Functional response of the predatory mite *Chileseius camposi* (Acarina: Phytoseiidae) on densities of it prey, *Panonychus ulmi* (Acarina: Tetranychidae). Revista de Biología Tropical 56, 1255–1260.

Skirvin, D. J. and J. S. Fenlon. 2001. Plant species modifies the functional response of *Phytoseiulu*s *persimilis* (Acari: Phytoseiidae) to *Tetranychus urticae* (Acari: Tetranychidae): implications for biological control. Bull Entomol Res 91:61–67.

Smith, L. B. 1985. Insect infestation in grain loaded in railroad cars at primary elevators in southern Manitoba, Canada. J. Econ. Entomol. 78: 531-534.

Solomon, M .E. 1949. The natural control of animal populations. J. Ani. Ecol. 18, 1–35.

Solomon, M. E. 1969. Experiments on predatory–prey interactions of storage mites. Acarol. 11, 484–503.

Souza-Pimentel, G. C., P. R. Reis, G. R. Liska and M. A. Cirillo. 2018. Predatory Potential of Phytoseiulus macropilis (Banks) Preying on Tetranychus urticae Koch (Acari: Phytoseiidae, Tetranychidae). Adv. Entomol. 6, 134-147.

Spieksma, F. Th. M. and C. Smits. 1975. Some ecological and biological aspects of the booklouse *Liposcelis bostrychophila* Badonnel 1931 (Psocoptera). Netherlands J. Zool. 25: 219–230.

Stejskal, V., J. Hubert, R. Aulicky, and Z. Kučerová. 2015. Overview of present and past and pest-associated risks in stored food and feed products: European perspective. J. Stored Prod Res. 64:122–132

Stejskal, V., J. Hubert, Z. Kucerova, Z. Munzbergova, J. Lukas, and E. Markova. 2003. The influence of the type of storage on pest infestation of stored grain in the Czech Republic. Plant Soil Environ. 49: 55-62.

Stejskal, V., O. Douda, M. Zouhar, Manasova J. E. Throne, G. P. Opit, P. W. Flinn. 2006. Seasonal distribution of psocids in stored wheat. In: Lorini I, Bacaltchuk B, Beckel H, Deckers E, Sundfeld E, Dos Santos JP, Biagi JD, Celaro JC, Faroni LRDA, Bortolini L et al.(eds) Proceedings of the 9th International Working Conference on Stored-Product Protection, Campinas. ABRAPOS, Rodovia, Brazil, 15–18 Oct 2006

S**uchita, M. G., G. P. U. Reddy and M. M. K. Murthy. 1989**. Relative efficacy of pyrethroids against rice weevil (*Sitophilus oryzae* L.) infesting stored wheat. *Ind. J. Plant Protec*. 17: 243–246.

Systat Software, Inc., 2007. TableCurve 2D, version 5.01. Systat Software, Inc., San Jose, CA.

Sun, W., M. Cui, L. Xia, Q. Yu, Y. Cao, and Y. Wu. 2020. Age-Stage, Two-Sex Life Tables of the Predatory Mite *Cheyletus malaccensis* Oudemans at Different Temperatures. Insects. 11 (181), 1 – 13.

Tang, P. A., J. J. Wang, Y. He, H. B. Jiang, and Z. Y. Wang. 2008. Development, Survival, and Reproduction of the Psocid *Liposcelis decolor* (Psocoptera: Liposcelididae) at Constant Temperatures. Ann. Entomol. Soc. Am. 101: 1017-1025.

Thind, B. B., and H. L., Ford. 2006. Laboratory studies on the use of two new arenas to evaluate the impact of the predatory mites *Blattisocius tarsalis* and *Cheyletus eruditus* on residual populations of the stored product mite *Acarus siro*. Experimental and Applied Acarol. 38, 167–180.

Throne J. E., G. P. Opit, and P. W. Flinn. 2006. Seasonal distribution of psocids in stored wheat, pp. 1095-1103. *In* I. Lorini, B. Bacaltchuk, H. Beckel, D. Deckers, E. Sundfeld, J. P. dos Santos, J. D. Biagi, J. C. Celaro, L. R. D"A. Faroni, L de O. F. Bortolini, M. R. Sartori, M. C. Elias, R.N.C. Guedes, R. G. da Fonseca, and V. M. Scussel (Eds.), Proceedings of the 9th International Working Conference on Stored Product Protection, October 15-18, 2006, Campinas, São Paulo, Brazil. Brazilian Post-harvest Association – Associação Brasileira de Pós-Colheita (ABRAPOS), Campinas, São Paulo, Brazil.

Toldi, M., D. Cristina., C. Faleiro, Liberato, G. Da Silva, and N. J. Ferla. 2017. Life cycle of the predatory mite *Cheyletus malaccensis* (Acari: Cheyletidae) fed on poultry red mite *Dermanyssus gallinae* (Acari: Dermanyssidae). Syst. Appl. Acarol. 22:1422-1430

Trematerra, P., and F. Fiorilli. 1999. Occurrence of Arthropods in a Central Italy feed-mill. J. Pest Sci. 72: 158-163.

Turner, B. D. 1994. *Liposcelis bostrychophila* (Psocoptera: Liposcelididae), a stored food pest in the UK. Int. J. Pest. Manag. 40: 179-190.

Turner, B. D., and H. Maude-Roxby. 1988. Starvation survival of the stored product pest *Liposcelis bostrychophilus* Badonnel (Psocoptera: Liposcelidae). J. Stored Prod. Res. 24: 23-28.

Turner, B., and J. Bishop. 1998. What's bugging your kitchen? Environ. Health J. 106: 310-314.

Turner, B. D., N. Staines, J. Brostoff, C. A. Howe, K. Cooper. 1996. Allergy to psocids. In *Proc. 2nd Int. Conf. Insect Pests Urban Environ.*, ed. KB Wildey, p. 609. Edinburgh: BCP Wheatons

Uiterwaal, S. F., C. Mares, J. P. DeLong. 2017. Body size, body size ratio, and prey Type Influence the functional response of damselfly nymphs. Oecologia 185 (3), 339 – 346.

Valbuza, M. F., A. L. Matioli, M. E. Sato, M. R. Potenza, and A. E. C. Campos. 2020. Mites in spice and medicinal dehydrated plants stored in bulk in the metropolitan area of Sao Paulo. J. Stored Prod. Res. 85 101-540

van Hage-Hamsten, M., and S. G. Johansson 1992. Storage mites. Exp Appl Acarol. 16 (1-2):117-128.

Villalobos, M. J. P., A. Baz and P. D. Estal .2005. Occurrence of psocids and natural predators on organic rice in Calasparra (Murcia, Spain). *J. Stored Prod. Res*. 41: 231–235.

Volgin, V. I. 1987. Acarina of the family Cheyletidae of the world. Akad Nauk SSSR Zool Inst Keys to the fauna of the USSR 101:1–532.

Wang, J. J., Y. Ren, X. Q. Wei, and W. Dou. 2009. Development, Survival, and Reproduction of the Psocid *Liposcelis paeta* (Psocoptera: Liposcelididae) as a Function of Temperature. J. Econ. Entomol. 102: 1705-1713.

Wang, J. J., J. H. Tsai, Z. M. Zhao, and L. S. Li. 2000. Development and reproduction of the psocid *Liposcelis bostrychophila* (Psocoptera: Liposcelididae) as a function of temperature. Ann. Entomol. Soc. Am. 93: 261-270.

Wang, J. J., Z. M. Zhao, and L. S. Li 1999. Induced tolerance of the psocid, *Liposcelis bostrychophila* Badonnel (Psocoptera: Liposcelididae), to controlled atmosphere. Int. J. Pest Manag. 45: 75–79.

Wang, J., Z. Zhao, and L. Li. 1998. Studies on Bionomics of *Liposcelis Entomophila* (Psocoptera: Liposcelididae) Infesting Stored Products. Insect Sci. 5: 149-158.

Wei, D. D., W. He, Z. Q. Miao, T. Q. Tu, L.Wang, W. Dou and J. J. Wang. 2020. Characterization of Esterase Genes Involving Malathion Detoxification and Establishment of an RNA Interference Method in *Liposcelis bostrychophila*. Front. Physiol. 11:274.

Wei, D., J. J. Wang, Z. M. Zhao, and J. H. Tsai. 2002. Effects of controlled atmosphere and DDVP on population growth and resistance development by the psocid, *Liposcelis bostrychophila* Badonnel (Psocoptera: Liposcelididae). J. Stored Prod. Res. 38: 229-237.

Williams, B., and T. E. Hallas. 1988. *Cheyletus eruditus* (Schrank) (Acari) feeding on eggs of the cat flea *Ctenocephalides* felis (Bouche) (Siphonaptera). Entomologist’s Monthly Magazine 124, 144.

Winks, R. G, and E. A. Hyne 1997. The use of mixed-age cultures in the measurement of response to phosphine. International Conference on Controlled atmospheres and Fumigation in Stored Products, (Edited by Donahaye E, Navarro S and Varnava, A) Nicosia, Pp. 3–16.

Wharton, G.W., and L. G. Arlian. 1972. Predatory behavior of the mite *Cheyletus aversor*. Animal Behavior 20, 719–723.

Wu, Y. F. Li, Z. Li, V. Stejskal, R. Aulicky, Z. Kucerova, T. Zhang, P. He, and Y. Cao. 2016. Rapid diagnosis of two common stored-product predatory mite species based on species-specific PCR. J. Stored. Prod. Res.69: 213–216.

Xia, B., D. M. Luo, Z. W. Zou, R. Xu, Z. M. Zhu. 2007. Predation of *Cheyletus eruditus* on *Aleuroglyphus ovatus*. Chin. Bull. Entomol. 44, 549 – 552.

Xiao, Y. and H. Y. Fadamiro. 2010. Functional responses and prey-stage preferences of three species of predacious mites (Acari: Phytoseiidae) on citrus red mite, *Panonychus citri* (Acari: Tetranychidae). Biol. Control. 53, 345–352.

Xu, Y. and Z. Q. Zhang. 2015. *Amblydromalus limonicus*: a "new association" predatory mite against an invasive psyllid (*Bactericera cockerelli*) in New Zealand. Syst. and Appl. Acarol., 20, 375–382.

Yao, H., W. Zheng, K. Tario, and H. Zhang. 2014. Functional and Numerical Responses of Three Species of Predatory Phytoseiid Mites (Acari: Phytoseiidae) to Thrips flavidulus (Thysanoptera: Thripidae). Neotrop. Entomol. 43:437–445.

Yoshikawa, M. 1985. Ecology and control of mites in Japanese houses. J. Environ. Cont. Technol. 3:4–11.

Yoshizawa, K., and K. P. Johnson. 2006. "Morphology of male genitalia in lice and their relatives and phylogenetic implications". Syst. Entomol. 31 (2): 350–361.

Yousef, A.A., M. A. Zaher, and M. M. Kandil. 1982. Effect of prey and temperature on the development and biology of *Cheyletus malaccensis* Oudemans (Acari: Cheyletidae). Zeitschrift fur Angewandte Entomologie 93, 39–42.

Žd'árková, E. 1979. Cheyletid fauna associated with stored products in Czechoslovakia. J. Stored Prod. Res. 15, 11–16.

Žd'árková, E. 1986. Mass rearing of the predator *Cheyletus eruditus* (Schrank) (Acarina: Cheyletidae) for biological control of acarid mites infesting stored products. Crop Prot. Sci. 5, 122–124.

Žd'árková, E. 1998. Biological control of storage mites by *Cheyletus eruditus*. Integrated Pest Manag. Review 3, 111–116.

Žd'árková E., and J. Pulpan 1973. Low temperature storage of the predatory mite *Cheyletus eruditus* (Schrank) for future use in biological control. J. Stored Prod. Res. 9 (4): 217 – 220.

Žd'árková E., and E. Horák. 1990. Preventive biological control of stored food mites in empty stores using *Cheyletus eruditus* (Schrank). Crop Prot. 9, 378–382.

Žd'árková, E., and P. Horák. 1999. Development of *Cheyletus eruditus* (Schrank) (Acarina: Cheyletidae) at low temperature. Plant Prot. Sci. 35, 14–16.

Žďárková, E and P. Horák. 2000. Number of prey necessary for completing development of *Cheyletus eruditus* (Acarina: Cheyletidae). In: 21st International Congress of Entomology, Abstract. Book I, August 2000, Iquassu Falls, Embrapa Soja, Londrina, p 19

Zettler, J. L.1991. Pesticide resistance in *Tribolium castaneum* and *T. confusum* (Coleoptera: Tenebrionidae) from Flour Mills in the United States. J. Econ. Entomol. 84: 763–767.

Zettler, J. L., and G. W. Cuperus. 1990. Pesticide resistance in *Tribolium castaneum* (Coleoptera: Tenebrionidae) and *Rhyzopertha dominica* (Coleoptera: Bostrichidae) in wheat. J. Econ. Entomol. 83: 1677–1681.

Zettler, J. L., and D. W. Keever. 1994. Phosphine resistance in cigarette beetle (Coleoptera: Anobiidae) associated with tobacco storage in the Southeastern United States. J. Econ. Entomol. 87: 546–550.

Zettler, J. L., W. R. Halliday, and F. H. Arthur. 1989. Phosphine resistance in insects infesting stored peanuts in the Southeastern United States. J. Econ. Entomol. 82: 1508–1511

Zhu, P., Y. Fan, W. Mo, T. Xin, B. Xia, and Z, Zou. 2019. Functional response of adult *Cheyletus malaccensis* (Acari: Cheyletidae) to different developmental stages of *Aleuroglyphus ovatus* (Acari: Acaridae). J. Stored. Prod. 84: 101–525.

VITA

James K. Danso

Candidate for the Degree of

Doctor of Philosophy

Dissertation: PHOSPHINE RESISTANCE AND BIOLOGICAL CONTROL OF PSOCIDS (PSOCODEA: LIPOSCELIDIDAE)

Major Field: Entomology

Biographical:

Education:

Completed the requirements for the Doctor of Philosophy in Entomology at Oklahoma State University, Stillwater, Oklahoma in July, 2021.

Completed the requirements for the Master of Philosophy in Crop Protection (Entomology) at Kwame Nkrumah University of Science and Technology, Kumasi, Ghana in 2017.

Completed the requirements for the Bachelor of Science in Agriculture at Kwame Nkrumah of Science and Technology, Kumasi, Ghana in 2012.

Experience:

Graduate Associate (Teaching and Research Assistant) at the Department of Entomology and Plant Pathology, Oklahoma State University while pursuing a Ph.D., 2018–2021.

Interim Laboratory Technician at the Department of Entomology and Plant Pathology, Oklahoma State University while pursuing a Ph.D., in summer 2019 and spring 2020.

Professional Memberships:

Entomological Society of America.