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DISTRIBUTION OF ARTHROPOD PREDATORS AND THEIR RESPONSES TO *EUXESTA* SPP. (DIPTERA: ULIDIIDAE) IN THE LABORATORY AND IN CORN FIELDS IN SOUTH FLORIDA

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Abstract

Injury caused by picture-winged flies or corn silk flies (Diptera: Ulidiidae) is a major limiting factor of corn (*Zea mays* L.: Poales: Poaceae) yields in Florida. Growers have relied heavily on chemical insecticides for control of these pests. However, we are exploring other methods, such as the use of biological control by predatory arthropods, which may be factors in an integrated pest management (IPM) program. Thus, information on the distribution of the flies and their predators and responses of the predators to the flies is potentially useful. Taylor's Power Law, Iwao's Patchiness Regression, and the Index of Dispersion were used to study distribution patterns of arthropods found on sweet corn ears in 2010. Arthropod groups included eggs and larvae of *Euxesta stigmatias* (Loew), *E. eluta* (Loew), *E. annonae* F. (Diptera: Ulidiidae), nymphs and adults of *Orius insidiosus* (Say) (Hemiptera: Anthocoridae), and larvae of *Anotylus insignitus* (Gravenhorst) (Coleoptera: Staphylinidae). In each season in the R1, R2, and/or R3 corn stages, ulidiid eggs and larvae generally had aggregated distributions in corn ears (b, β, & $ID > 1$; $P \leq 0.05$). *Orius insidiosus* had aggregated distributions on most sampling dates except for random or regular distributions (b, β, & $ID \leq 1$) in the R3 stages of the spring and fall. The distribution of *A. insignitus* larvae was measured only in the summer R3 and was aggregated. In the laboratory, *O. insidiosus* had a Type III functional response to eggs of *E. stigmatias*, *E. eluta*, and *E. annonae,* and the combination of these species with similar handling times (0.43-0.45 h) and attack constants (0.03-0.05h^{-1}) for the different prey species. These results for *O. insidiosus* may be compared to those of *Zelus longipes* (L.) (Hemiptera: Reduviidae) and other predators, which may help elucidate the effectiveness of these predators in controlling ulidiid flies.

Key Words: *Orius, Anotylus,* corn silk flies, biocontrol

Resumen

Heredas causada por las moscas de alas retratados o moscas de la seda del maiz (Diptera: Ulidiidae) son un factor ilimitando en las cosechas del maiz (*Zea mays* L.: Poales: Poaceae) en Florida. Productores les han necesitada las insecticidas chemicas por el control de plagas. Pero ya los esfuerzos han empezado a examinar otros methodos, tales como usando control biologico con artrópodos depredadores, que podrian ser muy importante en el desarrollo de una programa integrada de manejar las plagas (IMP). Por consiguiente, información sobre la distribución de estas moscas y sus depredadores y las respuestas de las depredadores a sus presas es potencialmente utilo. La regla de poder Taylor, regresion discontinuo de Iwao, y el indice de dispersión fueron usados para estudiar los patrónes de distribución de los artrópodos en las espigas de maiz en 2010. Los grupos de artrópodos incluyeron huevos y larvas de *Euxesta stigmatias* (Loew), *E. eluta* (Loew), *E. annonae* F. (Diptera: Ulidiidae), nymphos y adultos de *Orius insidiosus* (Say) (Hemiptera: Anthocoridae), y larvas de *Anotylus insignitus* (Gravenhorst) (Coleoptera: Staphylinidae). En cada temporada en etapas R1, R2, y R3 del maiz, huevos y larvas de ulidiides tuvieron distribuciónes agregados (b, β, & $ID > 1$; $P \leq 0.05$) en espigas de maiz. Distribuciónes de *O. insidiosus* fueron aggregados por la mayoria de las fechas de mostreo excepte por en las etapas R3 de la primavera y otoño, quando fueron al azar o regular (b, β, & $ID \leq 1$). La distribución de larvas de *A. insignitus* fue medida solemente en el R3 de verano, quando fue aggregado. En el laboratorio, *O. insi-*

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diosus tuvo una respuesta funcional de Tipo III a los huevos de *E. stigmatias*, *E. eluta*, y *E. annonae,* y el combinación de estas especies con parecidos tiempos de manejos (0.43-0.45 h) y constantes de atacar (0.03-0.05 h^{-1}) por especies differentes de las presas. Estas resultas por *O. insidiosus* podrian ser comparado a las resultas por *Zelus longipes* (L.) (Hemiptera: Reduviidae) y otros depredadores y podrian assistir en el entendimiento de los effectividades de los depredadores en el control de moscas ulidiides.

Palabras Claves: *Orius, Anotylus*, corn silk flies, biocontrol

Since the 1960s, the United States has been the world's leading producer of sweet corn (*Zea mays* L.) (Hansen & Brester 2012). In 2009, fresh-market and processed corn (frozen and canned) were valued at $836 million and $336 million, respectively (ERS 2010). Florida led the nation from 2004 to 2009 in fresh-market sweet corn production with 19-27% of the annual national crop value (Mossler 2008, ERS 2010).

Several species of picture-winged flies or corn silk flies including *Euxesta stigmatias* (Loew), *E. eluta* (Loew), *E. annonae* F., and *Chaetopsis massyla* (Walker) (Diptera: Ulidiidae) have become serious pests of sweet corn in the USA (Van Zwaluwenberg 1917; Barber 1939; Hayslip 1951; Seal & Jansson 1989; Nuessly & Hentz 2004; Goyal et al. 2010). After eclosion from pupae in the soil, the flies mate and oviposit among corn silk strands, which are bundles of flower stigmas and styles. The larvae have 3 instars that feed on corn silk, kernels, and the remainder of the cob (Seal et al. 1996, Nuessly and Capinera 2010). By severing the corn silk as they feed, the first instars disrupt pollination (Bailey 1940). Larvae then enter the kernels and feed on the cob (App 1938; Seal & Jansson 1989; Nuessly et al. 2007), and any injury to corn kernels renders the ears (and sometimes a truckload of them) unmarketable (D. R. S., personal communication).

According to Ritchie et al. (1992) and Bean (2010), each field corn plant develops about 21 leaves during its vegetative stages lasting 9 weeks. Sweet corn completes the vegetative stages about 2.5 weeks sooner than field corn. Tassels are then produced, and a few days later, the first silk marks the beginning of R1, or silking stage. In R2 or blister stage (10-14 days after first silk), the kernels are round and resemble a blister. In R3 (18-22 days after first silk), the silk becomes dry and brown, the outside of the kernel is yellow, and the inner fluid is starchy and milky white; hence, R3 is the milk stage. In R4 or dough stage (24-28 days after first silk), the fluid of the inner kernel has thickened into a paste or dough. In R5 or dent stage (35-42 days after silking), the shelled cob darkens and reddens, and most kernels are drying and becoming dented. In R6 (55-65 days after silking), physiological maturity occurs and most or all kernels have attained the maximum dry weight (Ritchie et al. 1992; Bean 2010). In R1, infestation by picture-winged flies (Diptera: Ulidiidae) begins to economically affect crops (Seal & Jansson 1993). There is maximum eclosion of ulidiid larvae from eggs in R2 (Seal & Jansson 1989). By R3, larval feeding has caused maximum damage to corn (Seal & Jansson 1989).

Chemical insecticides are presently the only effective technique for controlling ulidiid flies in sweet corn fields. Corn is often sprayed daily in the R1 through R3 stages with insecticides (Goyal 2010) that kill adult flies while leaving other life stages protected inside corn ears (eggs and larvae) or soil (pupae) (Nuessly & Capinera 2010). Biological control agents may be helpful if they would control both the adults and hidden stages, but information on biocontrol of ulidiid flies is lacking. Major predators of sweet corn pests in the northeastern United States are *Coleomegilla maculata* (DeGeer), *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae), and *Orius insidiosus* (Say) (Hemiptera: Anthocoridae) (Andow & Risch 1985; Coll & Bottrell 1992; Coderre et al. 1995; Wheeler & Stoops 1996; Hoffmann et al. 1997; Musser et al. 2004). In Mexico, the wasps, *Spalangia* spp. (Hymenoptera: Pteromalidae) and an Eurytomid (Hymenoptera: Eurytomidae) have been reported to parasitize *Euxesta* spp. pupae (Baez et al. 2010). On corn silk and husks in Germany, Eckert et al. (2006) collected predators such as lacewings (Neuroptera: Chrysopidae) and minute pirate bugs (Hemiptera: Anthocoridae). They claimed that sampling corn ears is a good method to find changes in the abundance of specific predators and other arthropod feeding groups.

Predators that had been found in a south cornfield survey preceding the present study were *O. insidiosus*, *A. insignitus*, *C. carnea*, and *Z. longipes* (Kalsi et al. 2014). Also, besides ulidiid eggs and/or larvae, the most abundant arthropod species during silking (R1) in all 3 seasons was *O. insidiosus* (Kalsi et al. 2014). Knutson & Gilstrap (1989) and Eckert et al. (2006) also found *Orius* spp. to be the most abundant predators when they surveyed the fauna of corn ears. As generalist predators, *Orius* spp. are common in different agricultural systems, where they serve as biological control agents (Isenhour & Marston 1981; Isenhour & Yeargan 1981; Isenhour et al. 1990; Reid 1991; Bush et al. 1993). *Orius insidiosus* adults are commercially available in Europe and the U.S.A. Therefore, further research into understanding relationships between these predators and the ulidiid eggs and larvae is needed to

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permit the use of the predators in integrated pest management.

Once beneficial predators have been identified, predator and prey distributions and behavior of predatory species pursuing prey should be studied to help determine the efficacy of a predator as a natural enemy in biocontrol programs. Distribution patterns of predatory arthropods, ulidiid eggs, and larvae can be analyzed by Taylor's Power Law (Taylor 1961), Iwao's Patchiness Regression (Iwao 1968), and the Index of Dispersion (Clapham 1936; Selby 1965; Perry & Mead 1979). Also, the potential of a predatory species to control a pest species can be determined by its effect on the pest population dynamics over time (Brodeur 2006; Jafar & Goldaste 2009). Hence, another method to study the effectiveness of a predator is to determine its functional response, which Solomon (1949) defined as the number of prey attacked by a predator at a given prey density. The functional response is helpful in studying predator-prey relationships because it shows the ability of a predator to regulate prey density, or how the predator increases or decreases prey consumption with changing prey density (Hassell 1978; Livdahl & Steven 1983). The capacity to regulate prey density can be estimated by the handling time (T_h) and the attack constant [a (h^{-1})]. The attack constant is the time required by the predator to search for its prey, and handling time is the time needed to encounter and eat the prey (Ives et al. 1993). These metrics have been used in studies of protists, vertebrates (Kilpatrick & Ives 2003), and arthropods. Examples include big-eyed bugs *Geocoris* spp. (Hemiptera: Lygaeidae) feeding on eggs of cotton bollworm *Helicoverpa* sp. (Lepidoptera: Noctuidae) (Shrestha et al. 2004), and *O. insidiosus* feeding on the mites *Panonychus ulmi* (Koch) and *Tetranychus urticae* (Koch) (Acari: Trombidiformes: Tetranychidae) (Ashley 2003). However, functional responses and handling times have not been determined previously for arthropods preying on corn-infesting ulidiids.

Objectives of the present study were to 1) determine the spatial distributions of select predatory arthropods and ulidiid eggs and larvae found in corn ears and 2) determine the functional responses of selected predators to eggs, larvae, and adults of *E. stigmatias*, *E. eluta,* and *E. annonae* in the laboratory.

Materials and Methods

The study was conducted in Miami-Dade County, Florida at the University of Florida, Tropical Research and Education Center (TREC), Homestead, Florida and at Super 6 Farms, Kendall, Florida, from Feb to Dec 2010. During the 11-mo study period, temperature, humidity, and rainfall were recorded 60 cm above ground by the Florida Automated Weather Network (FAWN) station, Homestead, Florida. From Feb to Dec 2010, monthly mean temperatures ranged from 27.9 °C in Jun to 14.2 °C in Dec with a minimum of -1 °C in Dec and maximums of 35 °C in Jun, Jul, and Aug (FAWN 2010). Mean monthly relative humidities ranged from 85% in Aug and Sep to 76% in Dec, and rainfall varied from 28 cm (11.0 inches) in Aug to 2 cm (0.7 inches) in Dec (FAWN 2010).

Field Preparation

There were 3 field tests with 1 test per season in the spring, summer, and fall 2010. All field data were collected in 2 fields; there was one field per location: Kendall and Homestead. Test fields were on Krome gravelly loam soil (loamy-skeletal, carbonatic, hypothermic, lithic, udorthents) that was well drained, had a pH of 7.4-8.4, was 34-76% limestone pebbles (≥ 2 mm diam), and had low organic matter (< 2%) (Nobel et al. 1996; Li 2001). Size of each test field was 0.4 ha divided into 40 equal 0.01-ha plots (replications) that were each 2 rows wide by 27.5 m long. *Zea mays* L. 'Obsession' sweet corn (Seminis Vegetable Seeds, Oxnard, California) was planted at Homestead using a garden seeder (Model 1001-B, Earthway® Products, Bristol, Indiana) on 15 Feb, 2 June, and 1 Oct for the spring, summer, and fall tests, respectively. At Kendall, *Z. mays* 'Pioneer 3394' field corn (Pioneer Seeds, Johnston, Iowa) was planted instead of sweet corn. Planting sites were spaced 0.3 m apart in rows separated by 0.9 m. Three to 5 seeds were planted at each planting site to assure that at least 1 seed germinated, but seedlings were thinned to 1 plant per site upon emergence. In parallel bands spaced 0.1 m from the seed rows, granular fertilizer (N-P-K: 8-16-16, Diamond R fertilizers, Fort Pierce, Florida) was applied at 1,347 kg/ha at planting. In addition, a liquid foliar fertilizer spray (N-P-K: 4-0-8, Diamond R fertilizers, Fort Pierce, Florida) was applied twice at 21 and 35 days after planting (DAP) to provide 2.8 kg of N/ha/day. To control weeds, 0.09 kg/ha of the pre-emergent herbicide trifluralin (Treflan™ 4EC, Dow AgroSciences, Indianapolis, Indiana) was applied at planting, and plants were irrigated as needed by drip irrigation. Drip irrigation by a T-Tape system (John Deere Co., San Marcos, California) was provided at 0.704 kg/cm^2 by 2 parallel lines per row; each line had one hole (2 mm diam) per 15 cm of irrigation line. The system ran for 1 hr/day dispensing 151 L of well water per 30.5 m of row length.

Data Collection

Sweet corn fields were sampled once in each stage of reproductive development (R1, R2, and

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R3) (Bean 2010) per crop in each season (spring, summer, and fall). The sample in R1 was 4 days after first silk, the second sample was 5 days after the beginning of R2, and the final sample was in the first week of R3. In the spring and fall, samples in the R1, R2, and R3 stages were 49, 63, and 70 DAP, respectively, and in the summer, their respective times were 56, 70, and 77 DAP. For each sample date, 1 corn ear was collected from each of 2 randomly sampled corn plants per plot resulting in 80 ears total. Each ear was placed separately in a 17 × 22-cm, self-sealing plastic bag, and samples were stored at 26 ± 5 °C if they could not be immediately processed. Each ear was then cut in half and placed in a 100-mL beaker of 75% ethanol for 5 min to collect arthropods, then removed with forceps and discarded. Smaller pieces of corn cob, husk, and silk were also rinsed in alcohol to remove arthropods. Alcohol rinses were filtered with a 25-µm mesh (USA Standard Testing Sieve, W. S. Tyler Co., Mentor, Ohio) to collect arthropods, which were saved in 70% ethanol for identification. For field corn at Kendall, only the top 5 cm of ear sheath (including leaf tips and silks but not the cob) were collected and handled as described for sweet corn. A 10X microscope was the instrument of choice for counting arthropods, which were identified to species where possible. Unidentified arthropods were sent for identification and voucher specimens submitted to the Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Gainesville.

Spatial Distribution of Predators and Corn-Infesting Ulidiids

Kalsi (2011) and Kalsi et al. (2014) provided results of the initial abundance and diversity survey. However, the present study also used the results to determine within-field distributions of ulidiid eggs, larvae, and 2 key predators: adults and nymphs of *O. insidiosus* and larvae of *Anotylus insignitus* (Gravenhorst) (Coleoptera: Staphylinidae: Oxytelinae) (Fig. 1). *Orius insidiosus* nymphs, adults, and ulidiid eggs and larvae in corn plants all originated from natural populations at the site where they were surveyed (Kendall or Homestead). However, *A. insignitus* larvae originated only at Homestead. We applied Taylor's Power Law (b) (Taylor 1961), Iwao's Patchiness Regression (β) (Iwao 1968), and the Index of Dispersion (ID) (Clapham 1936; Selby 1965; Perry &

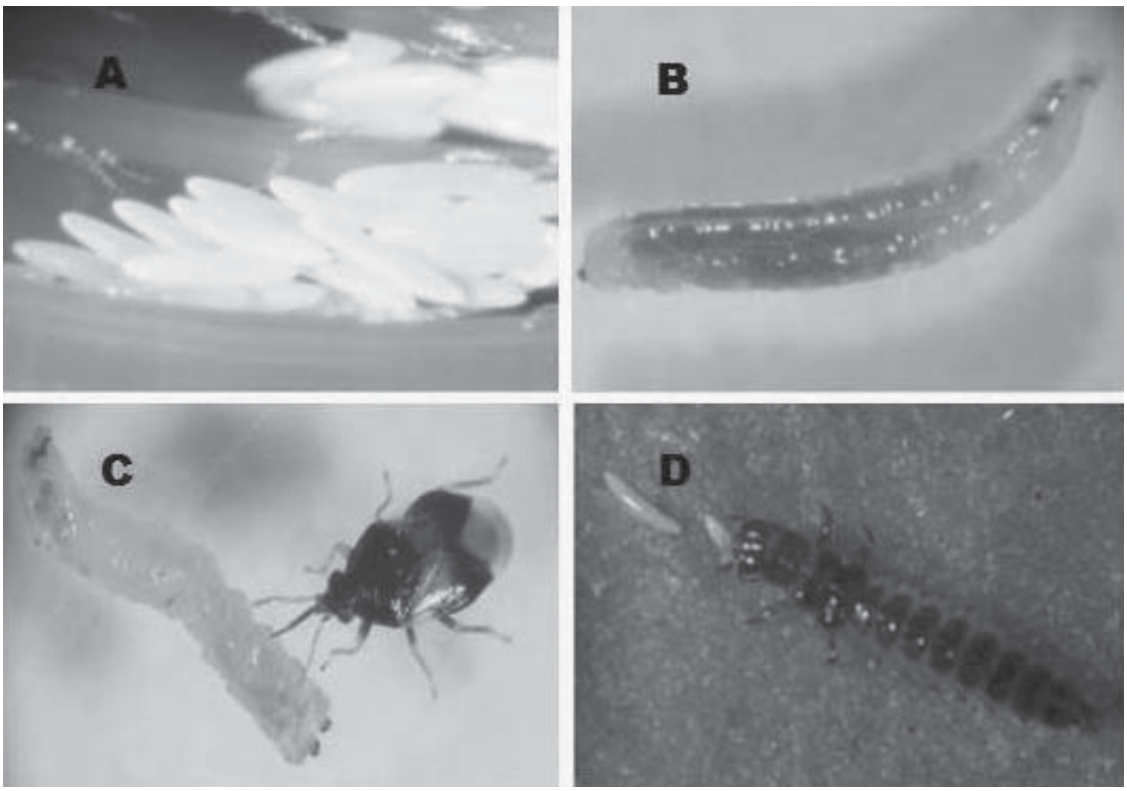


Fig. 1. *Euxesta stigmatias* and 2 of its predators. (A) Eggs of *E. stigmatias* in corn silk. (B) Third instar larva of *E. stigmatias*. (C) Adult of *O. insidiosus* feeding on a third instar larva of *E. stigmatias*. (D) *Anotylus insignitus* larva feeding on *E. stigmatias* eggs.

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Mead 1979) to quantify aggregation patterns. *ID, b,* and β are indices of dispersion (aggregation) for a species (Southwood 1978), and each indicates if the distribution pattern is aggregated, random, or regular (uniform), which occurs when *ID, b,* or β are either > 1, ~ 1, or < 1, respectively. Using the sample mean ($\bar{x}$) and sample variance (s^2), index of dispersion (*ID*) values were determined (equation 1). Taylor's power law (equation 2) and Iwao's patchiness regression parameters (equation 3) were calculated using general linear regression models (Southwood 1978; SAS Institute 2003). Taylor power law determines relationships between log $\bar{x}$, log s^2, and log a (sampling factor) (equation 2). Iwao patchiness regression relates Lloyd (1967) mean crowding index [($s^2/\bar{x}$) -1], the sample mean ($\bar{x}$), and the index of contagion or tendency toward crowding (α) in equation (3). To determine within-field distributions for the predators and ulidiids using the Taylor (b) and Iwao (β) indices, we first determined the goodness of fit to each linear model using regression coefficients (r^2). Then, a student's t-test helped to determine if b and β were significantly different from 1.0: if r^2 approximately equals 1, the model provided the best fit.

$$ID = s^2/\bar{x} \quad (1)$$

$$b = (\log s^2 - \log a) \,/\, \log \bar{x} \quad (2)$$

$$\beta = [(s^2/\bar{x}) - 1] + \bar{x} - \alpha \quad (3)$$

Functional Response of *O. insidiosus* to Eggs of *Euxesta* Species

Ulidiid Colonies to Supply Predators. Each laboratory colony was established with 100 adults in the summer of 2010 with separate colonies for *E. stigmatias*, *E. eluta,* and *E. annonae* collected from corn fields. Each *Euxesta* sp. was reared in a separate 31 × 31 × 31-cm cage maintained at 27 ± 5 °C and 75 ± 10% RH with rearing methods the same for each fly species. Adult flies were supplied with 1% honey solution and fresh water, and colonies were fed a beet armyworm (*Spodoptera exigua* (Hübner); Noctuidae) artificial diet (BAW, Southland Co., Lake Village, Arkansas) using the methods of Seal & Jansson (1989). Diet mixture was attached to the bottoms of plastic 28-g vials (BioServe™, Beltsville, Maryland, USA) that were placed upside-down on the cage ceilings with the tops facing down to allow oviposition. Eggs were collected at 24-h intervals and then transferred to fresh BAW diet for larval emergence in the same environmental conditions as adults to develop a uniform colony of each fly species. Newly eclosed first-instars were removed from egg containers every 24 h, transferred to different 28-g vials, maintained on BAW diet, and allowed to pupate. Every 4 h, diet vials were checked to remove pupae, which were washed gently with tap water to remove dietary residue and to avoid fungal infection. Pupae were then air-dried and placed into Petri dishes each with a moist, disk-shaped, 5-cm-diam paper to prevent desiccation. Petri dishes with pupae were placed into 31 × 31 × 31-cm cages and checked every 2 h to collect newly emerged adults.

Functional responses were calculated for *O. insidiosus* to eggs of *E. stigmatias, E. eluta,* and *E. annonae* in the laboratory. The tests were conducted in a growth chamber at 27 ± 3.5 °C and 75 ± 10% humidity. Eggs of *Euxesta* spp. were obtained from the laboratory colonies and *O. insidiosus* adults were obtained from Koppert Biological Systems (Romulus, Michigan). Each experimental unit was the same kind of 28-g plastic vial used to maintain *Euxesta* spp. colonies, but the vials remained empty. However, a 3.5-cm-diam parafilm layer was placed over and depressed 2 cm into each vial and provided with *Euxesta* spp. eggs at specific densities, hence, an area was formed where predator and prey were introduced. Prey *Euxesta* spp. eggs were killed by cooling for 30 min at 2 °C, then 1-day-old eggs were added to the vials at 5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55,and 60 eggs per vial with 1 *Euxesta* sp. per vial. One *O. insidiosus* adult was then added to each vial after being starved for 24 h. Two or 3 water drops were added to the paraffin layer of each vial as a moisture source, then the vial was covered with a Petri dish lid. Eggs preyed upon by *O. insidiosus* became deflated with an air vacuole that did not appear in control vials without the predators, thus, deflated eggs were assumed to have been attacked by *O. insidiosus*. After 24 h, the number of deflated eggs in each arena was counted, and if the predator died or larvae emerged from eggs, the container was excluded from the test and discarded. Each treatment with a given number of eggs per arena was replicated 8 times.

The predation data allowed us to determine the shape of the resulting curves and the type of functional responses (Type I, II or III), handling times, and attack constants. First, the type and shape of each functional response curve was determined with the polynomial regression model, equation (4) (PROC CATMOD, SAS Institute 2003) in which N_e is the number of prey eaten, N_0 is the initial number of prey, and P_0, P_1, P_2, and P_3 are the estimated parameters. For each initial prey density and proportion of eggs eaten, a polynomial equation was determined. If the first parameter (P_1) was negative, the functional response was type II, and if positive, it was a type III. Once the type of functional response was determined, data were fit to the mechanistic model and the random predator equation, and handling times and attack constants were estimated (PROC NLIN, SAS Institute 2003). A non-linear, least-square regression of the number of flies of-

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fered versus number eaten was used to estimate and compare the functional response parameters. The Hassell equation (5) was used to estimate the search rate and attack constant parameters using a constant (b), handling time (T_h), time taken by a predator to search for its prey (a), and the total time available for *O. insidiosus* to search for and attack its prey (T).

$$N_e / N_0 = \{exp\,(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3) / [1 + exp\,(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3)]\} \quad (4)$$

$$N_e = N_0\{1 - \exp[-aTN_0/(1 + bN_0 + aT_hN_0^2)]\} \quad (5)$$

Results

Spatial Distribution of Predators and Ulidiids

Spring 2010. Based on the Taylor (b), Iwao (β), and Index of Dispersion (ID) variables, distributions of ulidiid eggs in all 3 corn reproductive development stages and ulidiid larvae in the R2 and R3 stages were aggregated (b, β, & $ID > 1$; $P \leq 0.05$; Table 1). Numbers of ulidiid larvae in the R1 stage were not sufficiently large to estimate distribution patterns. Similarly, *O.*

Table 1. Dispersion indices for ulidiid eggs and larvae, nymphs and adults of orius insidiosus, and larvae of anotylus insignitus in sweet corn ears at silking (R1), blister (R2), and milk (R3) stages in 2010[1].

		Taylor Power Law[4]		Iwao Patchiness Regression[4]		Index of Dispersion[4]
Insect group[2]	Corn stage[3]	b	r^2	β	r^2	ID
Spring						
UE	R1	4.1 AGG	0.99	3.39 AGG	0.97	1.2 AGG
UE	R2	2.59 AGG	0.98	2.76 AGG	0.99	23.7 AGG
UE	R3	1.5 AGG	0.99	2.0 AGG	0.98	13.7 AGG
UL[5]	R2	20.1 AGG	0.98	14.7 AGG	0.99	18.0 AGG
UL	R3	4.12 AGG	0.99	2.48 AGG	0.98	7.71 AGG
OI	R1	2.32 AGG	0.99	7.29 AGG	0.97	1.20 AGG
OI	R2	2.03 AGG	0.98	3.34 AGG	0.99	2.96 AGG
OI	R3	0.86 REG	0.99	0.72 REG	0.98	0.91 RAN
Summer						
UE	R1	3.56 AGG	0.98	3.98 AGG	0.99	5.43 AGG
UE	R2	4.32 AGG	0.89	3.76 AGG	0.81	5.01 AGG
UE	R3	2.09 AGG	0.73	2.13 AGG	0.69	2.19 AGG
UL[5]	R2	6.12 AGG	0.99	5.98 AGG	0.97	7.59 AGG
UL	R3	3.09 AGG	0.97	4.04 AGG	0.98	9.65 AGG
OI	R1	2.35 AGG	0.97	2.67 AGG	0.99	4.42 AGG
OI	R2	1.92 AGG	0.83	2.18 AGG	0.89	3.7 AGG
OI	R3	1.95 AGG	0.84	1.99 AGG	0.91	1.2 AGG
SL[6]	R3	3.21 AGG	0.98	3.09 AGG	0.99	5.6 AGG
Fall						
UE	R1	1.86 AGG	0.99	1.02 RAN	0.98	13.6 AGG
UE[7]	R2	2.26 AGG	0.98	1.75 AGG	0.99	21.9 AGG
UL[5]	R2	3.14 AGG	0.98	2.26 AGG	0.99	23.7 AGG
UL	R3	4.12 AGG	0.99	2.48 AGG	0.98	7.71 AGG
OI	R1	1.82 AGG	0.99	1.75 AGG	0.97	9.4 AGG
OI	R2	1.62 AGG	0.99	2.28 AGG	0.99	1.12 AGG
OI	R3	0.96 RAN	0.99	0.94 RAN	0.99	0.87 REG

[1]All values for dispersion indices were based on a 0.4-ha-field.
[2]UE (ulidiid eggs), UL (ulidiid larvae), OI (*Orius insidiosus* nymphs and adults), and SL (*Anotylus insignitus* larvae).
[3]R1 (silking), R2 (blister), and R3 (milk) stages of corn growth. For each insect group, absence of a corn stage indicates no data for that stage.
[4]Insect distributions in the field. AGG (aggregated): *b, β, or ID* significantly >1, RAN (random): *b, β, or ID* not significantly different from 1, and REG (regular or uniform): *b, β, or ID* significantly < 1.
[5]Numbers of ulidiid larvae in R1 stages of all 3 seasons were not high enough to estimate distribution patterns.
[6]Larvae of *A. insignitus* were not present in the spring, summer R1 or R2, or the fall in sufficient numbers to determine dispersion indices.
[7]There were too few ulidiid eggs to determine their fall R3 distributions.

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insidiosus had aggregated distributions with *b*, *β,* and *ID* all significantly > 1 during R1 and R2 (Table 1). Also, for each stage and dispersion index, the coefficient of determination (r^2) was near 1 indicating a good fit. However, in the R3 stage, *O. insidiosus* was distributed randomly (*ID* ~ 1) or regularly (*b* and $\beta < 1$) (Table 1).

Summer 2010. Each dispersion index for ulidiid eggs in the R1, R2, and R3 stages yielded an aggregated distribution (*b, β,* & *ID* > 1) (Table 1). During the R1 stage of corn development, numbers of ulidiid larvae were too sparse for distributions to be determined, but in R2 and R3, they had aggregated distributions based on all 3 dispersion indices. The indices *b*, *β*, and *ID* also showed aggregated distributions for *O. insidiosus* during the R1, R2, and R3 stages (Table 1). Larvae of *A. insignitus* were not present in sufficient numbers in R1 or R2 to determine dispersion indices, but in R3, they had aggregated distributions based on all 3 indices.

Fall 2010. The indices all yielded aggregated distributions (*b, β,* & *ID* > 1) for ulidiid eggs during the R1 and R2 stages except for β in R1, which was random (β ~ 1). However, there were too few eggs to determine distributions in R3 (Table 1). Ulidiid larvae were too sparse in R1 to determine distributions, but in R2 and R3, larval numbers were sufficient to reveal aggregated distributions based on all 3 indices. All 3 indices showed that *O. insidiosus* distributions were aggregated during R1 and R2, but they were random (*b* & β ~ 1) or regular (*ID* < 1) during R3.

Functional Response of *O. insidiosus* to Eggs of *Euxesta* spp.

Linear regression equations (Hassell 1978) were calculated for the proportions of eggs consumed per initial number of eggs of *E. stigmatias*, *E. eluta, E. annonae,* and their cumulative means (Fig. 2). The first terms of the equations were positive: hence, *O. insidiosus* yielded Type III functional responses to different egg densities of *Euxesta* spp. The Hassell equation was therefore used to estimate the search rate and attack constant parameters. Also, the number of prey eggs declined as they were consumed because they were not replaced with fresh eggs, thus, Juliano's (1993) method was used to fit the data to the Rogers (1972) random predator equation. At the greatest prey density (60 eggs per arena), *O. insidiosus* consumed an average of about 40 *Euxesta* spp. eggs. For each initial number of eggs offered, there was no significant difference in predation rate for *O. insidious* among eggs of the 3 *Euxesta* spp., which also yielded similar handling times (0.43-0.45 h) and attack constants (0.03-0.05) (Table 2).

Discussion

Insect populations have 3 types of distributions: random, aggregated, and uniform (Taylor 1961, Southwood 1978). According to Southwood (1978), an insect's distribution pattern is greatly influenced by its within-field density. A low capture rate during sampling will often lower the

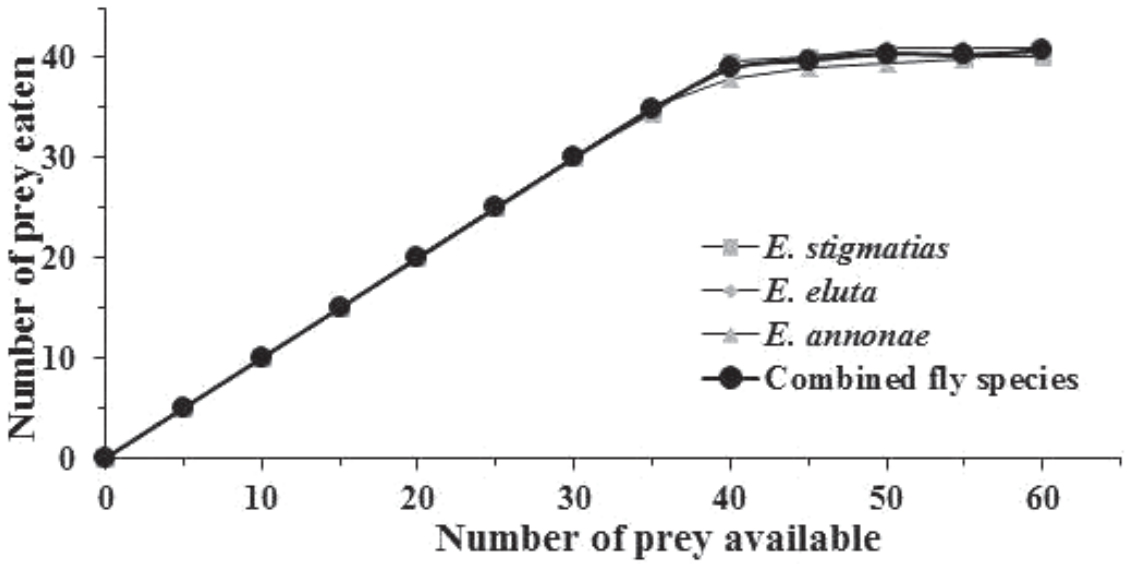


Fig. 2. Type III functional responses of *Orius insidiosus* to eggs of *Euxesta stigmatias*, *E. eluta, E. annonae,* and the combination (mean) of these prey species. Total exposure time was 24 h.

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TABLE 2. HANDLING TIMES AND ATTACK CONSTANTS OF *ORIUS INSIDIOSUS* WITH RESPECT TO EGGS OF *EUXESTA* SPP.[1]

Euxesta sp.	Handling time[2] T_h (h)	Attack constant[2] a (h^{-1})
E. stigmatias	0.45 ± 0.02	0.03 ± 0.05
E. eluta	0.43 ± 0.04	0.03 ± 0.02
E. annonae	0.44 ± 0.05	0.05 ± 0.01

[1]*Orius insidiosus* had a Type III functional response to eggs of each *Euxesta* sp.
[2]Mean ± S.E.

density and indices of dispersal, such as from aggregated distribution to random. However, if the arthropods are caught more often during sampling, density may be higher and hence aggregated instead of random or uniform. Taylor Power Law, Iwao Patchiness Regression, and the Index of Dispersion helped to clarify distribution patterns of ulidiid eggs and larvae and of their predators found on corn ears. Except for the present study and the investigation of ulidiids and *Z. longipes* adults by Kalsi (2011), these techniques have not been used previously to find spatial distributions of corn-infesting ulidiids or their natural enemies. However, Goyal (2010) used Morisita's index to study distribution patterns of adult *E. stigmatias*, *E. eluta,* and *C. massyla* caught in sticky traps in corn fields from the first appearance of corn silk until harvest. He found that the flies had aggregated distributions on most sampling dates from the first appearance of silk until harvest in corn fields of 1.8 ha and 4-16 ha. In the present study, the distribution pattern of ulidiid larvae was not detected in R1 of any season because they were absent or had low populations. However, similar to Goyal (2010) for adult ulidiids, we found that distributions of ulidiid eggs and larvae on the other sample dates were mostly aggregated.

The abundance of *O. insidiosus* increased during summer 2010, when abundances of ulidiid eggs and larvae were also greatest (Kalsi et al. 2014). Other than ulidiid eggs and/or larvae, *O. insidiosus* was among the most abundant arthropod species during R2 of the summer and fall and statistically the most abundant within R1 of the spring, summer and fall (Kalsi et al. 2014). However, *O. insidiosus* populations were numerically lowest during R3 of each season (Kalsi et al. 2014). Knutson & Gilstrap (1989) found that mean numbers of *O. insidiosus* were greatest during anthesis but declined as corn silk became dried and brown during R3. This may have resulted from a lack of fresh corn silk causing unfavorable breeding conditions for the predators during R2 and R3 (Barber 1936). Based on all 3 indices, *O. insidiosus* exhibited regular or random distributions during R3 of the spring and fall possibly because of low predator density caused by poor breeding conditions or low food supply, but the distributions were otherwise aggregated.

In corn fields of Ohio, Everly (1938) found 6 species of staphylinids including *Atheta* spp., *Barydoma* spp., *Leptolinus rubripennis* Lec., *Mycetoporus* spp., *Philonthus* spp., and *Coproporus* spp., which were collected from corn plants or on nearby ground (Everly 1938). Several staphylinid species are predaceous and possible biocontrol agents of agricultural pests (Cividanes et al. 2009). Larvae of *A. insignitus*, another predator of ulidiids, were found usually within dry corn silk in corn ears during spring R2 and summer R2 and R3. They were statistically among the most abundant taxa other than ulidiids in the summer R2 and R3 stages, and were numerically highest in the summer R3 (Kalsi et al. 2014). *Anotylus insignitus* larvae had aggregated distributions in the summer R3, the only season and stage when they were evaluated; hence, they were distributed like the majority of other insect species per corn stage considered.

There were no significant differences in rates of predation by *O. insidious* among the 3 *Euxesta* spp. as was also suggested by the similar attack constants and handling times for eggs of the fly species. In the laboratory, *O. insidiosus* had type III functional responses to eggs of *Euxesta* spp. A Type III, known as an accelerating functional response, is density dependent and typically exhibited by generalist predators (Murdoch & Oaten 1975). When plotted, a type III functional response has a sigmoid curve and is more characteristic of vertebrates than invertebrate predators or parasitoids (Hassell et al. 1977). However, many invertebrates also have a type III functional response, such as big-eyed bugs feeding on eggs of the cotton bollworm (Shrestha et al. 2004) and *O. insidiosus* feeding on the mites *P. ulmi* and *T. urticae* (Ashley 2003). Possible reasons affecting the occurrence of a type III functional response include selective foraging at high prey density, a predator's ability to switch prey species, and the effects of learning and experience on prey searching (Holling 1959; Murdoch 1969; Schauber et al. 2004). *Orius insidiosus* is facultatively phytophagous and can feed on plant material and thus maintain a high population density when prey is scarce (Wiedenmann & O'Neil 1991). Hence, when ulidiid eggs are in clusters that are randomly distributed in tightly packed corn silk, a Type III functional response may also occur because of the availability of plant material and possibly other prey species such as mites. Ashley (2003) reported that hemipteran predators such as *O. insidiosus* limit their search area by selective foraging, i.e., high frequency turning movements followed by feeding until they reach a threshold time. Also, when prey are scarce, predators may straighten their paths to reach and exploit areas with high

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prey density. *Orius insidiosus* may be able to easily feed on eggs in clusters without making many movements, but when the eggs are depleted, it can switch to other prey such as thrips.

The present study found indices of dispersal indicating spatial distributions for ulidiid eggs, larvae, and 2 key predators, *O. insidious* and to a lesser extent *A. insignitus.* In addition, handling times, attack constants, and functional responses of *O. insidiosus* to ulidiid eggs were found. These findings may be compared to those of other predatory arthropod species, such as *Z. longipes*, and can help in understanding the effectiveness of *O. insidiosus* and other predators in controlling ulidiid flies. This would permit the further identification and evaluation of potential for predatory arthropods to biologically control all ulidiid life stages.

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