Separation of three common hover fly predators of woolly apple aphid based on the exochorionic sculpturing of eggs

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The woolly apple aphid, *Eriosoma lanigerum* (Hausmann, 1802) (Hemiptera: Aphididae), is a cosmopolitan, indirect pest of cultivated apple, *Malus domestica* (Borkh.) (Rosaceae), (Baker 1915) that can colonize both the roots and arboreal portions of trees and adversely affect the growth, vigor, and productivity of the host (Weber and Brown 1988; Brown and Schmitt 1990; Brown *et al*. 1991, 1995). Historically, the solitary endoparasitoid *Aphelinus mali* (Haldeman, 1851) (Hymenoptera: Aphelinidae) has been considered the most important biological control agent for *E. lanigerum* in many, but not all, apple-growing regions (Yothers 1953; Nicholas 2000). The role of arthropod predators in regulating woolly apple aphid populations, particularly early in the growing season, has long been recognized (Dumbleton and Jeffreys 1938; Bodenheimer 1947; Nicholas 2000; Mols and Boers 2001) but has not been adequately addressed (Asante 1997). A review of the world literature revealed that 73 species of insect predators from five orders and seven families have been reported preying on woolly apple aphid (Asante 1997). Coccinellidae species were most abundant (48% of species), followed by species of Syrphidae (21%), Chrysopidae (14%), and Cecidomyidae, Forficulidae, and Lygaeidae (17% combined). In the United States of America, five coleopteran species and two species of Diptera have been recorded (Asante 1997).

During a severe outbreak of woolly apple aphid in the mid-Atlantic region in 2000, Bergh and Louque (2000) found that hover fly larvae were abundant in arboreal colonies in Virginian apple orchards, but predation by other insects was not observed. Brown and Schmitt (1990) reported that *A. mali* and syrphid larvae were the only natural enemies of woolly apple aphid in West Virginian apple orchards. Larvae of the three most common hover fly species found during the 2000 outbreak were reared to adulthood on woolly apple aphid and identified as *Heringia calcarata* (Loew, 1866), *Eupeodes americanus* (Wiedemann, 1830), and *Syrphus rectus* Osten Sacken, 1875 (Diptera: Syrphidae) by F.C. Thompson, USDA Systematic Entomology Laboratory, Washington, District of Columbia (Bergh and Louque 2000). *Eupeodes americanus* and *S. rectus* are generalist aphidophagous species, also preying on rosy apple aphid, *Dysaphis plantaginea* (Passerini, 1860), and spirea aphid, *Aphis spiraecola* Patch, 1914, (Hemiptera: Aphididae) in the apple ecosystem, whereas *H. calcarata* is a specialized predator of woolly apple aphid (Short and Bergh 2004). *Eupeodes americanus* is a wide-ranging species occurring across Canada and south to California, Mexico, Texas, and Florida; *S. rectus* occurs from central and eastern Canada south to Colorado, Texas, Mississippi, and North Carolina (Vockeroth 1992). The range of *H. calcarata* (*Neocnemodon calcaratus* in Wirth *et al*. (1965)) extends from Quebec to British Columbia and south to Kansas and Virginia (Thompson 2005).

Klein-Krautheim (1936) was the first to show that hover fly species can be identified by differences in the patterning and sculpturing of the egg exochorion. Chandler (1968) used the terms patterning and sculpturing, respectively, to describe the appearance of the surface of intact eggs at low magnification and the appearance of the tubular outgrowths, elevated ribs, and surface indentations of the exochorion viewed at higher magnification. Several authors have

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described these differences for numerous species of Syrphidae (Chandler 1968; Kuznetsov 1988; Chu 1992; Kula 1993), including species in apple orchards in the USSR (Karelin 1980). The sculpturing of eggs of *H. calcarata*, *E. americanus*, and *S. rectus* has not been described previously.

During field studies of the predator–prey associations of *H. calcarata*, three distinct types of syrphid eggs were most commonly observed in colonies of *E. lanigerum*. Unhatched eggs were collected and separated based on marked differences in the patterning of the exochorion. Larvae that eclosed from those eggs were reared individually on woolly apple aphid and the resultant adult flies were identified by F.C. Thompson as *H. calcarata*, *E. americanus*, or *S. rectus*. At low magnification (15×), the patterning of the dorsal surface of *H. calcarata* eggs consisted of parallel, longitudinal ribs. Unhatched eggs of *H. calcarata* were pearly white. The dorsal surface of *E. americanus* eggs possessed short, longitudinal ribs, and eggs of *S. rectus* were covered with tubular outgrowths. Eggs of *E. americanus* were greyish white, whereas those of *S. rectus* were white with a yellow tinge. Length and width measurements (mm ± SE) of eggs made with a calibrated ocular micrometer in a stereomicroscope were, respectively, 0.65 ± 0.004 and 0.24 ± 0.002 for *H. calcarata*, 0.96 ± 0.002 and 0.26 ± 0.002 for *E. americanus*, and 1.19 ± 0.007 and 0.45 ± 0.009 for *S. rectus*. Exochorionic sculpturing was examined using scanning electron microscopy. Preparation of eggs followed protocols recommended by J.M. Ehrman, Mount Allison University, New Brunswick, Canada. Eggs were fixed in a 2.5% glutaraldehyde and 0.1 mol/L phosphate buffer solution for 1 h at 4 °C, then rinsed in this solution three times for 10 min per rinse. Dehydration for 10 min in each of 20, 50, 70, 85, and 95% ethanol was followed by four 10-min rinses in 100% anhydrous ethanol. Eggs were placed in a Tousimis Samdri®-780A critical-point drier (Tousimis Research Corp., Rockville, Maryland) and then sputter-coated with gold (Hummer® VI, Anatech Ltd., Springfield, Virginia). Micrographs revealing the exochorionic sculpturing confirmed our descriptions of the patterning of eggs of the three species (Figs. 1A–F).

Descriptions of the patterning and (or) sculpturing of eggs of other hover fly species have revealed similarities among species within genera (Chandler 1968; Kula 1993), including similarities to the three species described here. However, Short (2003) reared many individuals collected as eggs from colonies of woolly apple aphid, rosy apple aphid, and spirea aphid infesting apple in Virginia and found that the morphological characteristics described are accurate and consistent for identifying the three major species that prey on aphid pests in the apple ecosystem. Given that the geographical ranges of these three species overlap broadly in regions of North America where apples are grown and where woolly apple aphid is a pest of apple, the use of egg morphology to separate them may prove to have broad applicability.

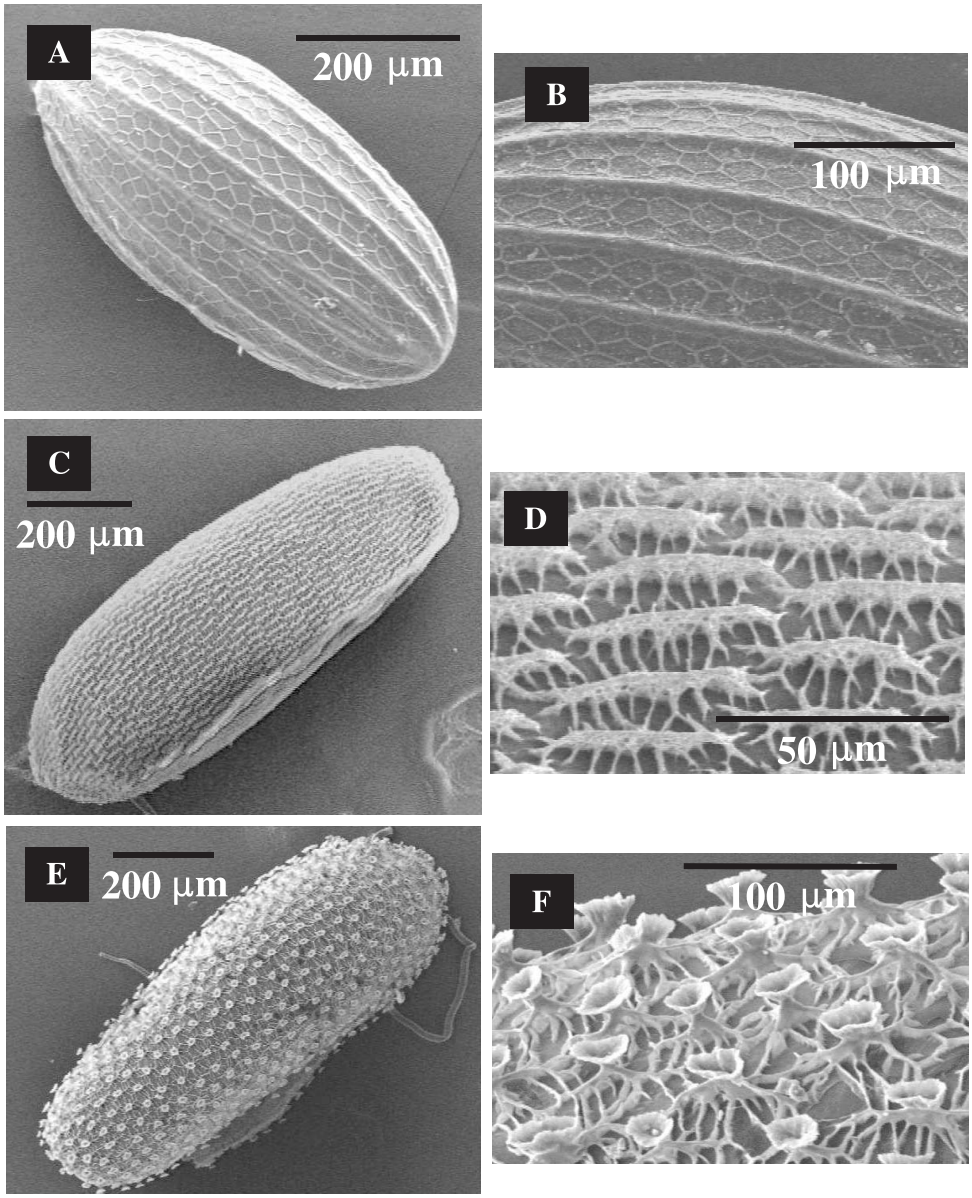
Chandler (1968) discussed the utility of positively identifying species of aphidophagous Syrphidae based on egg morphology. Syrphids are often difficult to mate in captivity (Schneider 1948), and in the absence of a rearing system we have used larvae from field-collected eggs for life-history studies (Short 2003) and examination of prey preference and prey suitability (Short and Bergh 2004). Young, potted apple trees artificially infested with arboreal colonies of woolly apple aphid and placed in apple orchards have proven highly effective for collecting eggs of *H. calcarata* and *E. americanus* (Short 2003). We believe that eggs are the most efficient sampling unit for field studies of the ecology of these species. Identification of species based on egg morphology enabled measurement of the distribution of hover flies among colonies of woolly apple aphid, rosy apple aphid, and spirea aphid. Females of *E. americanus* and *S. rectus* oviposited near colonies of all three species, whereas *H. calcarata* eggs were found only in woolly apple aphid colonies, indicating specialization of *H. calcarata* on woolly apple aphid in the apple ecosystem (Short and Bergh 2004). Unhatched eggs deposited on sentinel trees are a very current indication of adult activity in orchards because they are stationary, deposited in close proximity to aphid colonies, and have a 3-d developmental period (Short 2003). Using eggs deposited on sentinel trees, we are currently examining the seasonal phenology, relative abundance, and voltinism of the three hover fly species.

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Fig. 1. Scanning electron micrographs of (A) *Heringia calcarata* egg, (B) surface of *H. calcarata* egg, (C) *Eupeodes americanus* egg, (D) surface of *E. americanus* egg, (E) *Syrphus rectus* egg, and (F) surface of *S. rectus* egg.



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