Parasitism Rates of *Muscidifurax raptorellus* and *Nasonia vitripennis* (Hymenoptera: Pteromalidae) After Individual and Paired Releases in New York Poultry Facilities

PHILLIP E. KAUFMAN,[1] STEFAN J. LONG,[1] DONALD A. RUTZ,[1] AND J. KEITH WALDRON[2]

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ABSTRACT Commercially reared parasitoids were released into three high-rise, caged-layer poultry houses; one house received only *N. vitripennis* Walker, the second house received only *M. raptorellus* Kogan & Legner, and the third house received an equal ratio of both species. Overall, house fly parasitism by *M. raptorellus* was never higher than 7% in any house. Most parasitism in the *M. raptorellus* release house was attributed to *N. vitripennis.* Parasitism of house fly pupae by *M. raptorellus* did not significantly increase during or after the 6-wk release period in the house that received both parasitoids. However, a depression in total parasitism was not detected when releases of the two species were made in this house.

KEY WORDS *Muscidifurax raptorellus, Nasonia vitripennis,* house fly, biological control, poultry

MANAGEMENT OF HOUSE fly, *Musca domestica* L., populations in high-rise, caged-layer poultry houses is a primary concern of New York poultry producers. The potential for fly outbreaks on farms combined with a highly mobile adult insect intensifies this anxiety. Expanding urbanization and public health interest in flies associated with livestock and poultry houses often lead to neighborhood confrontation or even litigation. Producers currently have few pesticide options available for fly control because of insecticide resistance (Scott et al. 2000) and loss of insecticides due to regulatory actions. However, proven biological and cultural fly control options offer cost-effective alternatives to the use of insecticides.

Nasonia vitripennis Walker, a parasitoid commonly found in New York poultry houses (Rutz and Scoles 1989, Henderson and Rutz 1991), is commercially available for house fly control. It has three advantages over other parasitoids: (1) it is gregarious, producing from four to eight adult parasitoids from each parasitized house fly pupa; (2) it does not carry any known diseases that may decrease its effectiveness; and (3) it is inexpensive to rear commercially (Fried et al. 1990). In 1981 and 1982, *N. vitripennis* was the most abundant parasitoid species and was responsible for most house fly parasitism in New York poultry houses (Rutz and Scoles 1989). In a follow-up survey conducted 6 yr later, *S. cameroni* Perkins was the most common parasitoid in New York poultry houses (Henderson and Rutz 1991). Several possible factors including sampling time during the year, parasitoid recolonization, use of insecticides and manure management practices were offered to explain this species shift. Recently, *N. vitripennis* was reported again to be the predominant parasitoid collected in poultry houses (Long et al. 1997).

In recent years, research has focused on another parasitoid species, *Muscidifurax raptorellus* Kogan & Legner (Petersen and Cawthra 1995, Petersen and Currey 1996). This species originally was imported from Chile and has since been isolated from a beef cattle feedlot in Nebraska (Antolin et al. 1996). The gregarious nature of this parasitoid suggests excellent potential for biological control of flies (Petersen and Currey 1996). When *M. raptorellus* becomes established through augmentive releases in poultry houses, this parasitoid significantly increases overall parasitism and contributes substantially to house fly control (Long et al. 1997). Long et al. (1998) found that *M. raptorellus* on dairy facilities produced increasingly more progeny per pupa as temperatures increased. Conditions in poultry manure pits (higher, more stable temperatures) appear to offer *M. raptorellus* greater opportunities for production of multiple progeny per pupa compared with dairy facilities. Commercial insectaries are now mass-rearing *M. raptorellus* for release in livestock and poultry facilities.

Competition among pteromalid parasitoids is expressed in several ways including location of unparasitized hosts, sex ratio of progeny, development time and host utilization, larval predation within a multiparasitized host, and interference with oviposition (Wylie 1971, 1972a, 1972b, 1976; Pawson et al. 1987; Rivers 1996). Legner (1977) reported several *Muscidifurax* species to be competitively superior to *Spalangia,* and Pawson et al. (1987) found *M. zaraptor* Kogan & Legner competitively superior to *Urolepis rufipes* (Ashmead). Wylie (1970) reported *N. vitripennis* discriminated between unparasitized house fly pupae and those previously parasitized by *Muscidifurax raptor* Girault & Sanders and *Spalangia cameroni*

[1] Department of Entomology, Cornell University, Ithaca, NY14853.
[2] New York State IPM, Cornell University, Geneva, NY 14456.

Perkins. Rivers (1996) reported that at low host densities, female *N. vitripennis* reacted aggressively and interfered with all oviposition attempts by *M. zaraptor.* However, competition studies comparing *N. vitripennis* and *M. raptorellus* have not been reported.

To evaluate the integration of these potentially complementary parasitoids into a poultry integrated pest management (IPM) program, we monitored parasitism rates following individual and mixed releases of *M. raptorellus* and *N. vitripennis* in high-rise, caged-layer poultry houses.

Materials and Methods

Muscidifurax raptorellus and *N. vitripennis* were purchased from a commercial insectary (Beneficial Insectary, Oak Run, CA) with six weekly releases being made between 29 July and 2 September 1998. The targeted parasitoid release rate was four parasitoids/bird. Releases were conducted in three conventionally ventilated high-rise, caged-layer poultry houses of similar design. In these houses, air was forced from the manure pit by pit-level exhaust fans and manure fell directly to the pit floor, resulting in wide, somewhat flat manure mounds. House 1 contained 29,000 birds and received *N. vitripennis,* house 2 contained 28,000 birds and received *M. raptorellus,* and house 3 contained 26,000 birds and received both species. Houses 1 and 2 were adjacent to each other and shared a common bird-level ventilation system; however, they had separate manure pits. No birds were present in house 1 until 1 wk before the first parasitoid release (29 July). House 2 was repopulated with birds at the beginning of June and manure was removed on 20 July. House 3 had been in continuous production since the previous fall and manure had been partially removed in May. Houses 1 and 2 were located in Ontario County, NY, and house 3 was located in Cayuga County, NY.

The number of parasitized pupae required to provide the desired number of parasitoids designated for each field release was estimated from the number of parasitoids emerging from 15 subsamples of 20 parasitized pupae each from a commercial shipment. These were determined 4 wk before the first field release. To determine the actual parasitoid emergence from each field release shipment (number of parasitoids released into each house), five subsamples, containing 20 pupae each, were collected weekly and held in the laboratory for parasitoid emergence.

Shipments of parasitized pupae for each house were divided into five mesh bags (100 × 150 cm, mesh density 5.5 squares/cm) that were placed equidistantly, suspended 1.5 m above the floor from center support beams, throughout the manure pit in each poultry house. Bags were replaced 2 wk later by the current week's parasitized pupae shipment. After removal of the release bags, five subsamples of 20 pupae were recovered from each bag to estimate the number of pupae that produced parasitoids and the number of parasitoids that had emerged.

Parasitism rates were monitored weekly using the sentinel pupal method of Rutz and Axtell (1980a). Sentinel bags (8 × 8 cm, mesh density 5.5 squares/cm), each containing 50 live house fly pupae, were placed weekly on the surface, near the base of the manure pile. After 7 d, pupae from each bag were retrieved and held in the laboratory for 8 wk to allow for fly and parasitoid emergence. During the prerelease period, 15 sentinel bags were placed in houses 2 and 3 and retrieved weekly (2 July to 16 July). During the release (29 July to 2 September) and postrelease (9 September to 14 October) periods, 10 sentinel bags were placed and retrieved weekly in all three houses. Parasitoids were identified to species, sexed, and counted. Differentiation between *M. raptorellus* and *M. raptor* was accomplished using fore wing characteristics (wing color and shape, stigma shape and presence/absence of wing fringe).

Relative house fly abundance was monitored weekly in the manure pit of each house throughout the release and postrelease period by placement of 10 white file cards (7.5 × 12.5 cm). The fecal and regurgitation spots on each card were counted using a standardized grid and the data averaged for each house and week. On 23 September the manure was sampled for *Carcinops pumilio* (Erichson) (Coleoptera: Histeridae), a predator of house fly eggs and larvae, using a manure core (400 cc) at 10 randomly selected sites. Individual samples were placed in Tulgren funnels to extract beetles, which were subsequently identified, counted and an average determined for each house.

The numbers of specimens of each parasitoid species collected were summed for each house and release period and species composition was determined. The percentage of fly pupae parasitized was calculated by dividing the number of pupae with emergence holes by the total number of pupae retrieved. Parasitism rates were corrected for control mortality (Abbott 1925) and percentages were arcsine transformed for statistical analysis. The percentage of pupae parasitized and the species composition was analyzed by house and release period using a multi-factorial analysis of variance (ANOVA) model (PROC GLM; SAS Institute 1996) to detect differences between release periods within houses, and between houses within release periods.

Results and Discussion

The *N. vitripennis* release level was below the target release rate for 4 of the 6 wk in houses 1 and 3; whereas, in houses 2 and 3 the estimated number of *M. raptorellus* introduced exceeded the targeted release rate throughout the release period (Fig. 1). Because manure conditions which can influence house fly population densities varied considerably between houses (moisture level, age and volume), spot card data were not analyzed statistically, but are presented to document relative adult house fly abundance. Due to water leaking into the manure pit through the building foundation, manure in house 1 was much wetter, with

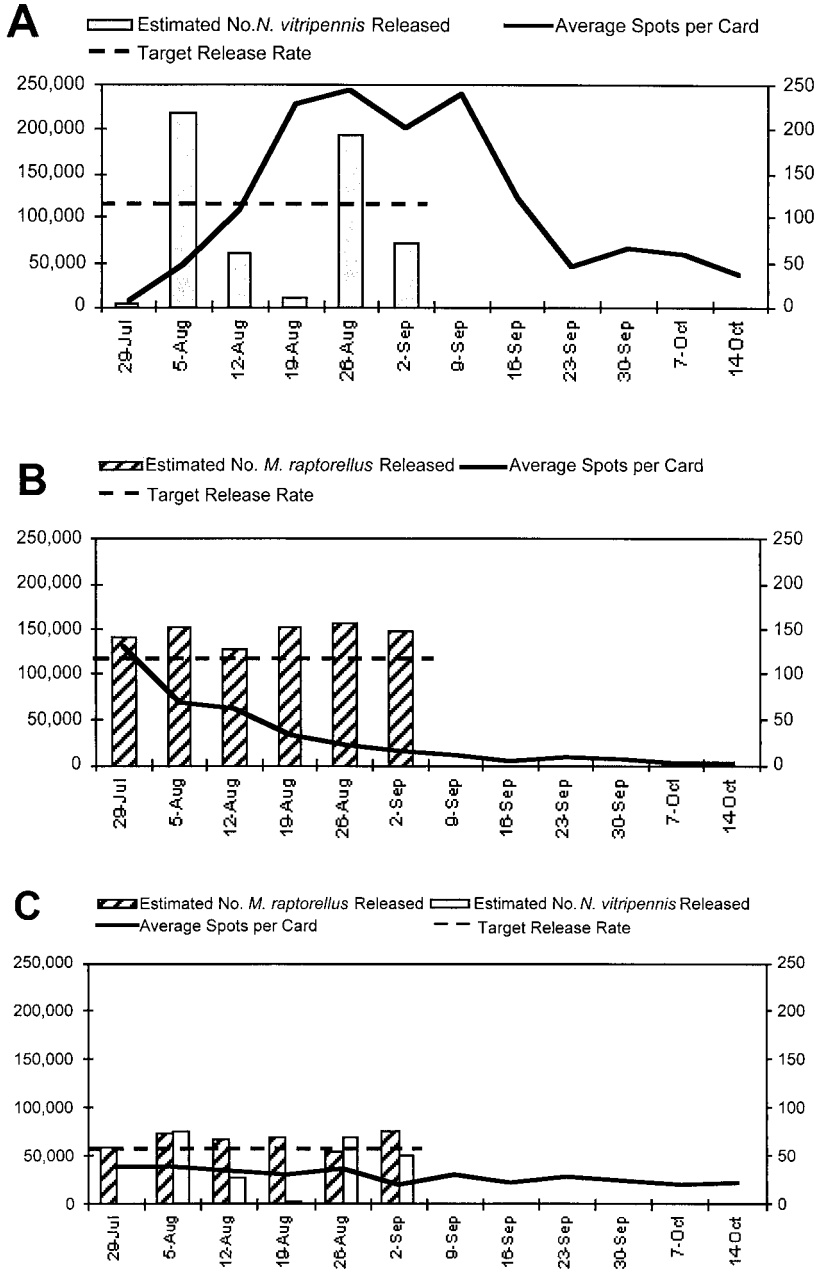

Fig. 1. Estimate of weekly parasitoid releases, targeted release levels, and house fly abundance in house 1 (A), house 2 (B), and house 3 (C)

correspondingly higher fly production, than the other houses. The mean number of spots per card in house 1 was >100 (above threshold) (Kaufman et al. 2000) for 5 wk (Fig. 1A). An average of two adult and 10 larval *C. pumilio* per manure core were recorded in house 1. In house 2, the mean number of spots per card declined sharply during the second release week and remained low for the duration of the study (Fig. 1B). This corresponded to high populations of *C. pumilio*, which were also present (23 adults and 38 larvae per core). As stated above, the manure in house 3 had been accumulating since the previous fall, with a partial cleanout 10 wk before the first release. Because only a portion of the manure was removed, the parasitoid complex in this house was not disrupted, likely resulting in the presence of a diverse parasitoid population (Table 1). The mean number of spots per card in house 3 remained low throughout the release and postrelease periods (Fig. 1C). An average of 24 adult and 20 larval *C. pumilo* per manure core were recov-

Table 1. Relative abundance of parasitoid species recovered from sentinel house fly pupae before, during and after weekly parasitoid releases in high-rise, caged-layer poultry houses

House	Species released	Species collected	No. collected, %[a]		
			Prerelease	Release	Postrelease
1[b]	*N. vitripennis*	*N. vitripennis*	—	3,211 (98.2)	8,211 (99.3)
		M. raptorellus	—	58 (1.8)	53 (0.6)
		M. raptor	—	0 (0.0)	5 (0.1)
		S. cameroni	—	1 (0.0)	0 (0.0)
		Total	—	3,270	8,269
2	*M. raptorellus*	*N. vitripennis*	1,437 (99.7)	10,504 (97.9)	5,848 (92.7)
		M. raptorellus	0 (0.0)	172 (1.6)	443 (7.0)
		M. raptor	4 (0.3)	50 (0.5)	7 (0.1)
		S. cameroni	0 (0.0)	1 (0.0)	10 (0.2)
		Total	1,441	10,727	6,308
3	*N. vitripennis* + *M. raptorellus*	*N. vitripennis*	1,733 (90.5)	2,114 (73.2)	1,179 (52.7)
		M. raptorellus	0 (0.0)	458 (15.9)	599 (26.8)
		M. raptor	0 (0.0)	257 (8.9)	194 (8.7)
		S. cameroni	183 (9.5)	55 (1.9)	265 (11.8)
		Urolepis rufipes	0 (0.0)	3 (0.1)	2 (0.0)
		Total	1,915	2,887	2,239

[a] Prerelease period (2 July to 16 July), release period (29 July to 2 September); postrelease period (9 September to 14 October).
[b] Prerelease data were not collected in this house due to lack of manure.

ered in this house. The presence of high numbers of *C. pumilio* likely contributed to the low house fly populations observed in houses 2 and 3; however, the impact of these predators in the current study is unknown. Geden et al. (1988) reported that under field conditions, adult *C. pumilio* destroyed 37 fly immatures, and larval beetles destroyed 17 fly immatures per day. Given the numbers of beetles we observed, it is likely that *C. pumilio* predation on house flies was extensive.

Muscidifurax raptorellus were not recovered during the prerelease period in any of the houses (Table 1). When considered as the percentage of the total number of parasitoids collected, significantly more *N. vitripennis* were recovered from house 2 (99.7%) than house 3 (90.5%) during the prerelease period ($F = 9.92$, df = 5, $P \leq 0.0345$). Manure in house 2 was ≈10 wk old when the prerelease period began; whereas, house 3, with a diverse parasitoid complex, had received only a partial manure cleanout 5 wk before the prerelease period. Rutz and Scoles (1989) in New York documented *N. vitripennis* presence throughout the year with the greatest abundance in the summer, whereas *N. vitripennis* was collected only during the summer in North Carolina (Rutz and Axtell 1980b). The current study was conducted during the summer months when all parasitoid species were (most) active. Our results suggest that *N. vitripennis* may be an early colonizer of poultry houses because it predominated in a recently cleaned manure pit, but did not maintain high densities in a house with a diverse parasitoid complex.

During the prerelease period, the percentage of house fly pupae parasitized by *N. vitripennis* accounted for 97 and 55% of the overall parasitism in house 2 and house 3, respectively (Table 2). The dominance of this species in the absence of commercial parasitoid releases in New York poultry facilities was also reported by Rutz and Scoles (1989). In house 1, *N. vitripennis* accounted for >94% of the overall

Table 2. Parasitism of sentinel house fly pupae in three New York high-rise, caged-layer poultry houses before, during and after releases of *N. vitripennis* and *M. raptorellus*

House[a]	Sample time[b]	House fly pupae parasitized (%) mean (SE)[c]			% of total parasitism by	
		N. vitripennis	*M. raptorellus*	Total[d]	*N. vitripennis*	*M. raptorellus*
1	Pre-release	—	—	—	—	—
	Release	10.4 (3.10)a	0.6 (0.53)a	11.0 (3.13)a	94.5	<0.1
	Post-release	30.3 (4.65)b	0.9 (0.63)a	31.4 (4.69)b	96.5	<0.1
2	Pre-release	6.8 (2.55)a	0.0 (0.00)a	7.0 (2.64)a	97.1	0.0
	Release	37.1 (4.05)c	1.4 (0.93)ab	40.2 (4.24)b	92.3	<0.1
	Post-release	21.2 (3.88)b	6.5 (2.40)b	28.3 (4.28)b	74.9	23.0
3	Pre-release	9.9 (3.10)a	0.0 (0.00)a	18.0 (3.30)a	55.0	0.0
	Release	9.7 (2.18)a	5.5 (1.49)ab	26.0 (3.91)a	37.3	21.2
	Post-release	6.3 (1.72)a	6.9 (2.22)b	29.5 (4.05)a	21.4	23.4

—, Prerelease data were not collected in this house due to lack of manure.
[a] *N. vitripennis* released in house 1; *M. raptorellus* released in house 2; *N. vitripennis* and *M. raptorellus* released in house 3.
[b] Prerelease period (2 July to 16 July); release period (29 July to 2 September); post-release period (9 September to 14 October).
[c] Within a column and house, means followed by the same lower case letter are not significantly different (alpha = 0.05; Tukey's multiple range test; house 1 df = 110; house 2 and 3 df = 161).
[d] Sum of released and indigenous *(Muscidifurax raptor, Spalangia cameroni,* and *Urolepis rufipes)* parasitism.

parasitism during both the release and postrelease periods. This suggests that *N. vitripennis* can remain the predominant parasitoid species when released into recently cleaned poultry houses. In house 2, which received only *M. raptorellus* releases, *N. vitripennis* remained the predominant species (>74%) throughout the release and postrelease periods. In house 3, parasitism attributed to *N. vitripennis* was highest during the prerelease period (55%) and declined during both the release (37%) and postrelease (21%) periods. Parasitism attributed to *M. raptorellus* in house 3, which received releases of *N. vitripennis* and *M. raptorellus*, was similar during the release and postrelease period (≈22%). These results suggest that when these two species are released into poultry houses with a diverse parasitoid species complex parasitism attributed to *N. vitripennis* will decline, whereas *M. raptorellus* will become established.

In house 1, significantly more parasitism was attributed to *N. vitripennis* during the postrelease period than during the release period (Table 2), indicating that this species became established in the relative absence of competition (Table 1). The increase in *N. vitripennis* parasitism resulted in significantly more overall parasitism during the postrelease period. During the release and postrelease period, parasitism attributed to *M. raptorellus* within each house did not differ significantly throughout the study (Table 2). This was unexpected given that houses 2 and 3 received weekly *M. raptorellus* releases, and suggests that higher release rates of *M. raptorellus* may be needed in high-rise, caged-layer poultry houses to achieve acceptable fly control. Total parasitism did not increase significantly in the dual release house (house 3), during either the release or postrelease period.

Parasitism by *M. raptorellus* significantly increased in house 2 following the 6-wk release period (Table 2). Overall, parasitism by *M. raptorellus* was never higher than 7% in any house. Most parasitism in the *M. raptorellus* release house (house 2) was attributed to *N. vitripennis*, indicating that despite large weekly releases of *M. raptorellus*, this species was either unable to compete with established *N. vitripennis* populations or unable to establish at the given release level. Additional studies are needed to examine the effectiveness of *M. raptorellus* releases in northeastern poultry houses.

A depression in total parasitism was not observed when releases of *N. vitripennis* and *M. raptorellus* were made in the same poultry house (house 3) (Table 2). The presence of other indigenous parasitoids, predominantly *M. raptor* and *S. cameroni*, may have obscured detection of interspecific competition between *N. vitripennis* and *M. raptorellus.* In several studies (Merchant et al. 1987, Henderson and Rutz 1991, Long et al. 1997), including the current one, when *S. cameroni* parasitism was observed, parasitism attributed to *N. vitripennis* was curtailed. Wylie (1970) observed that *N. vitripennis* preferred to oviposit in unparasitized house fly pupae, however, when parasitized hosts were presented, oviposition restraint was most pronounced toward hosts previously attacked by *M. raptor* and less toward those attacked by *S. cameroni.* Larval *M. zaraptor* prey on allospecific eggs and larvae, whereas *S. cameroni* only prey on larval competitors (Wylie 1972b). The selective oviposition behavior exhibited by *N. vitripennis* may be the result of prior encounters with *Muscidifurax* and *Spalangia* species. Many of the differences in species predominance in poultry facilities is believed due to the degree of manure dryness (Axtell and Rutz 1986) and this potential interaction or confounding factor deserves further study.

In this study, releases of *N. vitripennis* were successful in establishing this species in a recently cleaned poultry house; however, *N. vitripennis* was not successful in increasing parasitism in a poultry house with an established and diverse parasitoid complex. Because of this reduced impact, the potential competitive interaction between *N. vitripennis* and *S. cameroni* must be examined. Although *M. raptorellus* parasitism and establishment was low, this species was recovered during the postrelease period suggesting that with further testing it may become a suitable candidate for use in a biologically based IPM program. However, at this point in time, we still recommend the use of only *M. raptor* and *N. vitripennis* for release in New York poultry houses.

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