1-1-2016

Effects of size and age of the host Musca domestica (Diptera: Muscidae) on production of the parasitoid wasp Spalangia endius (Hymenoptera: Pteromalidae)

S.A. Broski

B.H. King

Original Citation

Broski, S. A., and B. H. King. 2016. Effects of size and age of the host Musca domestica (Diptera: Muscidae) on production of the parasitoid wasp Spalangia endius (Hymenoptera: Pteromalidae). Journal of Economic Entomology 110:282–287

Broski S and King BH. 2017. Effects of size and age of the host *Musca domestica* (Diptera: Muscidae) on production of the parasitoid wasp *Spalangia endius* (Hymenoptera: Pteromalidae). Journal of Economic Entomology 110:282–287. doi.org/10.1093/jee/tow246
Following is the accepted manuscript. It is the same content but lacks the copyediting and typesetting of the VOR (version of record), which is at http://jee.oxfordjournals.org/content/by/year The VOR may also be obtained by email: bking@niu.edu
For other King BH reprints: http://niu.edu/biology/about/faculty/bking/bking-publications.shtml

Effects of Size and Age of the Host *Musca domestica* (Diptera: Muscidae) on Production of the Parasitoid Wasp *Spalangia endius* (Hymenoptera: Pteromalidae)

SCOTT A. BROSKI AND B. H. KING[1]
Department of Biological Sciences, Northern Illinois University, DeKalb, IL 60115-2861.
[1] Corresponding author, e-mail: bking@niu.edu

ABSTRACT One method of control of house flies, *Musca domestica* L. (Diptera: Muscidae), and other filth flies is by repeated release of large numbers of pupal parasitoids such as *Spalangia endius* Walker. Rearing these parasitoids may be facilitated by understanding how host factors affect their production. Previous studies have examined the effects of host size and host age on parasitoid production, but have not examined the interaction between host size and host age or the effects with older females, which may be less capable of drilling tough hosts. Females were given hosts of a single size-age category (small young, small old, large young, or large old) for two weeks. The effect of host size and of host age on parasitoid production depended on female age. On their first day of oviposition, females produced more offspring from large than from small hosts, but host age had no significant effect. The cumulative number of parasitoids produced in the first week was not significantly affected by host size or host age. However, the cumulative number of parasitoids produced over two weeks was affected by both host size and host age, with the greatest number of parasitoids produced from small young hosts. Thus not only are smaller hosts cheaper to produce, but these results suggest that their use may have no effect or a positive effect on the number of parasitoids that can be produced when females are ovipositing for a week or two.

KEY WORDS biological control, host size, host age, parasitoid wasp, Pteromalidae, *Spalangia*

House flies, *Musca domestica* L. (Diptera: Muscidae), and other filth flies are a problem throughout the world, particularly in association with poultry and livestock production (Meyer et al. 1991, Geden 2012). Their populations can readily reach nuisance levels, and they may transmit disease (Alam and Zurek 2004, Talley et al. 2009). The first step in the management of filth flies is to control the amount and wetness of the manure and other organic matter in which fly larvae feed. Insecticides can also be effective and are widely used, but filth flies often evolve resistance (Kaufmann et al. 2010, Wang et al. 2012, Khan et al. 2013). As distances between production facilities and urban areas shrink, complaints and lawsuits make control more important (Thomas and Skoda 1993, Clean Neighborhoods 2005), yet may also make it more difficult to apply insecticides because of environmental concerns (Blacquiere et al. 2012, Goulson 2013). Biological control agents are an additional control option. Sustained augmentative releases of pupal parasitoids have been effective in reducing fly populations in some studies (Rutz and Axtell 1979, Morgan 1980, Weinzierl and Jones 1998, Skovgård and Nachman 2004, Geden and Hogsette 2006, McKay et al. 2007), but not in all studies (Andress and Campbell 1994). Parasitoids that are commercially available include *Spalangia* species, such as *S. endius* Walker (Hymenoptera: Pteromalidae) (Leppla and Johnson 2010).

Knowing the effects of host size and age may improve planning and flexibility in insectaries that produce parasitoids. For example, if parasitoid production is equivalent over a range of host ages, hosts can be left over the weekend, and reducing weekend hours may improve convenience and reduce production costs. Information on effects of host age may also be useful in timing commercial shipments of not yet parasitized host pupae. These issues are particularly important for parasitoids that do not successfully parasitize cold-stored hosts (Floate 2002).

Spalangia and other pteromalids that parasitize the pupal stage of house flies place their egg(s) on the surface of the host pupa within the host puparium. Thus the host must be a true pupa, pupariation alone having occurred is insufficient, i.e., the host must not only be ovoid shaped but also old enough that a space has formed between the puparium and the pupa (Wylie 1967). *Spalangia endius* is a solitary parasitoid, meaning that generally only one parasitoid completes development per host. Larger and younger host pupae may provide more food: they weigh more than smaller and older ones (King 1990); and as hosts age, adult body parts develop, resulting in less food for liquid feeders such as *Spalangia* larvae (Gerling and Legner 1968). On the other hand, the puparium (outer shell) of larger and older hosts may be difficult for a female to penetrate as she drills through the puparium for oviposition and host feeding. With older hosts, the first successful drill attempt takes more time (*S. cameroni* Perkins: King 1998), attempts are less likely to succeed (*S. cameroni*: King 1998, *S. endius*: King 2000), and a greater proportion of offspring fail to chew their way out as adults (*S. endius*: King 2000). Larger hosts also take longer for females to successfully drill into (*S. cameroni*: King 1994, *S. endius*: King 2002a).

In past studies of how host size and host age affect parasitoid production by *Spalangia* species, just host size or host age, not both, were varied; and females were usually given hosts for three days or less (Morgan et al. 1979; Siafacas 1980; King 1997, 1998, 2000, 2002b). Female *S. endius* live an average of 18 ± 1.07 d, n = 36, when given hosts daily (unpublished data from *Monandry versus immediate polyandry* experiment in King and Bressac 2010). The present study tested for an interaction between host size and host age, when females were given hosts for two weeks.

Materials and Methods

Spalangia endius used in this study were from a 16 year old colony originally established with wasps collected from Zephyr Hills, Florida. Vouchers were deposited at the Illinois Natural History Survey Center for Biodiversity, Insect Collection 6035 through 6054. Parasitoids were reared on *M. domestica* at 25°C, 50 – 70% relative humidity, 12:12 (L:D) h. Hosts were reared on a medium of commercial fly larva diet (Lab Diet, St. Louis, MO), fishmeal, pine shavings and water; and the density at which hosts were reared was manipulated to produce a small host size class and a large host size class (described in Broski and King 2015). In their last larval instar, the hosts crawl out of their media box into a larger empty plastic box underneath and pupate there.

When presented to *S. endius* females, the young hosts were 0 - 24 h old, timed from the initiation of tanning of the puparium, which meant that they were true pupae at least by the end of their time with the female. Pupae for the old host treatments were 3 d older. The host rearing procedure that we used resulted in average host volume that is more than twice as great for large hosts as for small hosts, with no overlap in size (mean (minimum - maximum): 33 (29 –38) mm^3 versus 14 (11–19) mm^3; whereas host age has no detectable effect on volume (Broski and King 2015). These host volumes were calculated using the equation for a prolate spheroid, $4/3\pi w^2 l$, where width (w) and length (l) were measured to the nearest 0.05 mm with an ocular micrometer on a dissecting scope.

The *S. endius* females were 0 -1 d old when first given hosts and had been obtained from a dish of parasitized hosts from which males had already begun emerging. All females mated as indicated by the subsequent production of daughters. The females were of fairly uniform size, having all developed in large hosts.

Each female was given either 10 small young hosts, 10 small old hosts, 10 large young hosts, or 10 large old hosts on each day for the first 14 d of her life. The hosts were provided to her in a glass vial (2 cm diameter, 6.8 cm height) plugged with cotton. Each day, the old hosts were poured out of the vial; then the female was allowed to walk up a test tube so that she could be tapped into a new vial with new hosts. For each of the four treatments, ten females were tested, and they all lived at least 14 d. Thus, the entire experiment involved 40 females and 5600 hosts. Hosts were transferred to females daily at room temperature (about 23°C, 50% RH), and parasitization and development were in an incubator at 25 - 26°C, 48 – 76% relative humidity, 12:12 (L:D) h. After offspring had stopped emerging and had all died (at least 2 mo), the offspring and the number of hosts with emergence holes were counted.

Statistical analyses were with SPSS (IBM Corp 2013). Alpha was 0.05, except where noted. Parasitoid production was examined by repeated measures ANOVA (analysis of variance), with host size and host age as between subject factors and day of oviposition as a within subject factor. The Greenhouse-Geisser correction (Hinton et al. 2014) was used to correct for the lack of sphericity in the data, hence the noninteger degrees of freedom, although multivariate analysis of variance, which does not assume sphericity, led to the same conclusions. Then total production across all 14 d that females were given hosts, across their first 7 d, and across their first day were each analyzed using a two-way ANOVA, with host size and host age as independent variables. ANOVA is robust to assumptions of normality and homogeneity, particularly when sample sizes are equal, which they were (Kikvidze and Moya-Laraño 2008, Zar 2010). Assumptions of normality and homogeneity were met at alpha of 0.001. Log transformation was not used because doing so greatly increased kurtosis.

Results and Discussion

The effect of host size and host age on parasitoid production changed over days of oviposition (Fig. 1).

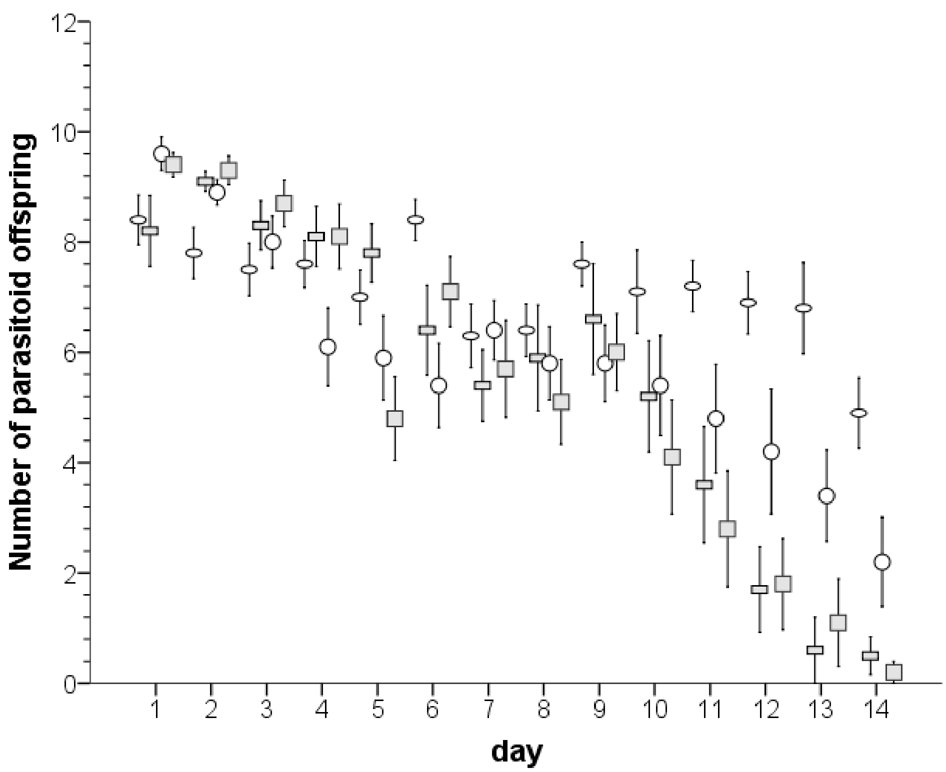

Fig. 1. Mean ± SE number of *Spalangia endius* offspring produced daily across two weeks when each mother received one type of hosts, hosts that were small young (small oval), small old (small rectangle), large young (large circle), or large old (large square).

Repeated Measures ANOVA:
day: $F_{5.99, 215.71} = 45.55$, $P < 0.001$;
day * host size: $F_{5.99, 215.71} = 2.52$, $P = 0.02$;
day * host age: $F_{5.99, 215.71} = 8.15$, $P < 0.001$;
day * host size * host age: $F_{5.99, 215.71} = 1.89$, $P = 0.08$

Table 1. **Total number of offspring produced [mean ± SE (minimum - maximum)] in a female's first day of life, added across her first seven days, and added across her first fourteen days of life. Each female was given a single category of host size and age throughout her life, n = 10 females for each of the four host treatments**

Host size and age	**Number of offspring produced in a female's 1st d**	**Number of offspring produced in a female's 1st 7 d**	**Number of offspring produced in a female's 1st 14 d**
Small Young	8.40 ± 0.45 (5 – 10)	53.00 ± 1.85 (43 – 63)	99.90 ± 3.63 (83 – 118)
Small Old	8.20 ± 0.65 (4 – 10)	53.30 ± 2.85 (32 – 63)	77.40 ± 6.07 (33 – 106)
Large Young	9.60 ± 0.31 (8 – 11)	50.30 ± 2.37 (36 – 61)	81.90 ± 4.22 (64 – 102)
Large Old	9.40 ± 0.22 (8 – 10)	53.10 ± 1.97 (44 – 63)	74.20 ± 4.38 (53 – 96)
host size:	$F_{1, 36} = 7.54$, $P = 0.009$	$F_{1, 36} = 0.40$, $P = 0.53$	$F_{1, 36} = 5.16$, $P = 0.029$
host age:	$F_{1, 36} = 0.21$, $P = 0.65$	$F_{1, 36} = 0.46$, $P = 0.50$	$F_{1, 36} = 10.48$, $P = 0.003$
host size * host age:	$F_{1, 36} < 0.001$, $P = 1.00$	$F_{1, 36} = 0.30$, $P = 0.59$	$F_{1, 36} = 2.52$, $P = 0.12$

Looking at cumulative production, there was no significant interaction between host size and host age, regardless of whether production was examined across the first day of oviposition, the first week, or the whole two weeks (Table 1). The effect of host size on cumulative production was significant for the first day and across the whole two weeks. However, on the first day more parasitoids were produced from larger hosts (Fig. 1), whereas across the whole two weeks more were produced from smaller hosts (Table 1). The effect of host age also depended on the time period examined. Across the whole two weeks of oviposition, production was greater from the younger hosts; whereas on the first day and across the first week of oviposition, host age had no significant effect.

Although *S. endius* is considered a solitary species, occasionally two offspring develop on a single host (in the present study about 3% of hosts with an emergence hole (n = 3334)). However, larger hosts increasing the chances of that happening do not explain more offspring being produced from large hosts than from small hosts on the first day of oviposition in the present study: conclusions were the same when the analysis was on the number of hosts with emergence holes. Perhaps the greater production from large hosts on the first day is because females can both host feed and oviposit on a single large host more readily than on a small host.

That small young hosts resulted in more offspring only later in life in the present study may be related to large old hosts being difficult to drill into (King 2000, Broski and King 2015). Ovipositor wear, as measured by the depth of a female's ovipositor serrations, does not decrease with repeated drilling of hosts, even for large old hosts (Broski and King 2015). However, as a female senesces she may become unable to muster the extra force needed to penetrate tough hosts.

Host size effects and host age effects have been variable among studies of solitary parasitoids of house fly pupae. Here we look just at studies in which females received a single host size or age category as in the present study. *Spalangia cameroni* produced more parasitoids from large hosts on their second day, more from small hosts on their third day and no difference on their first day (King 1994). *Muscidifurax zaraptor* produced more offspring from small hosts than from large hosts when tested on their second day of hosts (McKay and Broce 2008). Evidence to date from the present study and earlier studies of *Spalangia* species suggests that, provided hosts are not buried in wet media, host pupae being older either reduces the number of *Spalangia* offspring that are produced or has no effect (Table 2). In contrast to these studies of host age effects on offspring production in *Spalangia* species, there are relatively few such studies for other solitary species of pupal parasitoids of filth flies.

Table 2. Effect of host age on offspring production by solitary pupal parasitoids of filth flies

Parasitoid	Parasitoid production	Host age definition	# ♀s: hosts	Hosts	Oviposition	Reference	
S. endius	fewer on old hosts (3-d than 0-d)	0-day-old hosts were 0–24 h old, timed from the initiation of puparium tanning	1:10	unburied	1st 2 wk	present study	21-25ºC
S. endius	NS[a]	as above	1:10	unburied	1st d, 1st wk	present study	21-25ºC
S. endius	fewer on 1- or 3-d than 2-d old	1-, 2-, 3-d old	10:5 to 10:250	unburied	1st d	Morgan et al. 1979	27.8ºC
S. nigroaenea	fewer on old hosts	0-, 1-, 2-, 3-, 4-d old	1:20	unburied	1st 5 d	Siafacas 1980	26.7ºC
S. cameroni	fewer on old hosts	0-, 1-, 2-, 3-, 4-, 5-d old; 0-day-old hosts were 0-24 h old, timed from the puparium being red	1:30	unburied	1st d	King 1998	24-26ºC
S. cameroni	NS[a]	0-day-old hosts were 0-24 h old, timed from the initiation of puparium tanning	2:50	unburied	1st d	King 1997	23-28ºC
S. cameroni	more on old hosts (3-d than 0-d)	as above	2:50	buried	1st d	King 1997	23-28ºC
M. zaraptor	fewer on old hosts	1-, 2-, 3-, 4-, 5-d old; 1-day-old hosts were 24 h old puparia with head and appendages having begun eversion and a space between puparium and pupa	1:20	unburied	1st d	Coats 1976	28: 24ºC 14:10 h L:D
M. zaraptor	very few from 0-d	1-h, 1-d, 2-, 3-, 4--d old; timed from pupariation	5:50, 10:50	unburied	1st d	Petersen and Matthews 1984	26ºC
U. rufipes	varied with temperature and host density	0-, 1-, 2-, 3-, 4-, 5-d old; 0-day-old hosts were 1-24 h old, timed from pupariation	10:50 or 25:50	unburied	1st d	Matthews and Petersen 1990	25 or 30ºC

NS: not statistically significant at alpha = 0.05

Given that small hosts do not appear to reduce parasitoid production in *S. endius* when females are used for a week, and may even increase production by two weeks, one might expect small hosts to provide maximum value for insectaries rearing *S. endius*. Small hosts are obtained by rearing fly larvae at higher densities, which decreases the financial investment in fly media and saves space. The tradeoff is effects on the quality and timing of the parasitoids that are produced. Both male and female *S. endius* are larger when they develop on larger hosts and on younger hosts (Napoleon and King 1999, King 2000, 2002b; King and Napoleon 2006). Larger females produce more offspring and are better at burrowing (King and Napoleon 2006). Females that develop on larger hosts take longer to develop and live longer, although neither of these two effects is mediated through the female's size (Napoleon and King 1999; King and Napoleon 2006). For males, both being larger and developing on a larger host do not affect development duration or mating success. When production of *S. endius* is on a large enough scale and the goal is large numbers of parasitoids for release, the quicker development of females (Napoleon and King 1999) and greater number of total parasitoids over two weeks (Table 1) may mean that small hosts are best. In contrast, for someone purchasing parasitoids to release, parasitoids raised on large young hosts may be more effective.

Acknowledgments

Thanks to E. Burgess IV for assistance with the colony; to C. Hackman, H. Hildebrand, and E. Stauffer for their assistance in data collection; and to N. Blackstone and N. Barber for feedback on the writing and experimental design.

References Cited

Alam, M. J., and L. Zurek. 2004. Association of *Escherichia coli* O157:H7 with houseflies on a cattle farm. Appl. Environ. Microbiol. 70: 7578–7580.

Andress, E. R., and J. B. Campbell. 1994. Inundative releases of pteromalid parasitoids (Hymenoptera: Pteromalidae) for the control of stable flies, *Stomoxys calcitrans* (L.) (Diptera: Muscidae) at confined cattle installations in west central Nebraska. J. Econ. Entomol. 87: 714–722.

Blacquiere, T., G. Smagghe, C. A. M. van Gestel, and V. Mommaerts. 2012. Neonicotinoids in bees: a review on concentrations, side-effects and risk assessment. Ecotoxicology 21: 973–992.

Broski, S. A., and B. H. King. 2015. Drilling-in and chewing-out of hosts by the parasitoid wasp *Spalangia endius* (Hymenoptera: Pteromalidae) when parasitizing *Musca domestica* (Diptera: Muscidae). Environ. Entomol. 44: 1116–1124

Clean Neighbourhoods. 2005. Clean Neighbourhoods and Environment Act, HMSO, London, UK.

Coats, S. A. 1976. Life cycle and behavior of *Muscidifurax zaraptor* (Hymenoptera: Pteromalidae). Ann. Entomol. Soc. Am. 69: 772–780.

Floate, K. D. 2002. Production of filth fly parasitoids (Hymenoptera: Pteromalidae) on fresh and on freeze-killed and stored house fly pupae. Biocontrol Sci. Techn. 12: 595-603.

Geden, C. J. 2012. Status of biopesticides for control of house flies. J. Biopesticides 5(Supplementary): 1–11.

Geden, C. J., and J. A. Hogsette. 2006. Suppression of house flies (Diptera: Muscidae) in Florida poultry houses by sustained releases of *Muscidifurax raptorellus* and *Spalangia cameroni* (Hymenoptera: Pteromalidae). Environ. Entomol. 35: 75–82.

Gerling, D., and E. F. Legner. 1968. Developmental history and reproduction of *Spalangia cameroni*, parasite of synanthropic flies. Ann. Entomol. Soc. Am. 61: 1436–1443.

Goulson, D. 2013. An overview of the environmental risks posed by neonicotinoid insecticides. J. Appl. Ecol.: 977–987.

Hinton, P. R., I. McMurray, and C. Brownlow. 2014. SPSS explained, 2nd ed. Routledge, New York.

IBM Corp. 2013. IBM SPSS Statistics for Windows, Version 22.0, IBM Corp, Armonk, NY.

Kaufman, P. E., S. C. Nunez, R. S. Mann, C. J. Geden, and M. E. Scharf. 2010. Nicotinoid and pyrethroid insecticide resistance in houseflies (Diptera: Muscidae) collected from Florida dairies. Pest Manag. Sci. 66: 290–294.

Khan, H. A. A., S. A. Shad, and W. Akram. 2013. Resistance to new chemical insecticides in the house fly, *Musca domestica* L., from dairies in Punjab, Pakistan. Parasitol. Res. 112: 2049–2054.

Kikvidze, Z., and J. Moya-Laraño. 2008. Unexpected failures of recommended tests in basic statistical analyses of ecological data. Web Ecology 8: 67–73.

King, B. H. 1990. Sex ratio manipulation by the parasitoid wasp *Spalangia cameroni* in response to host age: a test of the host-size model. Evol. Ecol. 4: 149–156.

King, B. H. 1994. How do female parasitoid wasps assess host size during sex-ratio manipulation. Anim. Behav. 48: 511–518.

King, B. H. 1997. Effects of age and burial of house fly pupae (Diptera: Muscidae) on parasitism by *Spalangia cameroni* and *Muscidifurax raptor* (Hymenoptera: Pteromalidae). Environ. Entomol. 26: 410–415.

King, B. H. 1998. Host age response in the parasitoid wasp *Spalangia cameroni* (Hymenoptera: Pteromalidae). J. Insect Behav. 11: 103–117.

King, B. H. 2000. Sex ratio and oviposition responses to host age and the fitness consequences to mother and offspring in the parasitoid wasp *Spalangia endius*. Behav. Ecol. Sociobiol. 48: 316–320.

King, B. H. 2002a. Breeding strategies in females of the parasitoid wasp *Spalangia endius*: effects of mating status and body size. J. Insect Behav. 15: 181–193.

King, B. H. 2002b. Offspring number and sex ratio response to proportion of host sizes and ages in the parasitoid wasp *Spalangia cameroni* (Hymenoptera: Pteromalidae). Environ. Entomol. 31: 505–508.

King, B. H., and C. Bressac. 2010. No fitness consequence of experimentally induced polyandry in a monandrous wasp. Behavior 147: 85–102.

King, B. H., and M. E. Napoleon. 2006. Using effects of parasitoid size on fitness to test a host quality model assumption with the parasitoid wasp *Spalangia endius*. Can. J. Zool. 84: 1–5.

Leppla, N.C., and K. Johnson. 2010. Guidelines for purchasing and using commercial natural enemies and biopesticides in Florida and other states. IPM-146, University of Florida Institute of Food and Agricultural Sciences. http://ipm.ifas.ufl.edu/pdfs/Natural_Enemy_Guidelines.pdf

Matthews, J. R., and J. J. Petersen. 1990. Effects of host age, host density and parent age on reproduction of the filth fly parasite *Urolepis rufipes* (Hymenoptera: Pteromalidae). Med. Vet. Entomol. 4: 255–260.

McKay, T., and A. B. Broce. 2008. Host size and oviposition site effects on sex ratio of pupal parasitoid *Muscidifurax zaraptor* (Hymenoptera: Pteromalidae). Ann. Entomol. Soc. Am. 101: 650–655.

McKay, T., C. D. Steelman, S. M. Brazil, and A. L. Szalanski. **2007**. Sustained mass release of pupal parasitoids (Hymenoptera: Pteromalidae) for control of *Hydrotaea aenescens* and *Musca domestica* (Diptera: Muscidae) in broiler-breeder poultry houses in Arkansas. J. Agr. Urban Entomol. 24: 67–85.

Meyer, J. A., T. A. Schultz, C. Collar, and B. A. Mullens. 1991. Relative abundance of stable fly and house fly (Diptera: Muscidae) pupal parasites (Hymenoptera: Pteromalidae; Coleoptera: Staphylinidae) on confinement dairies in California. Environ. Entomol. 20: 915–921.

Morgan, P. B. 1980. Sustained releases of *Spalangia endius* (Hymenoptera: Pteromalidae) for the control of *Musca domestica* (L.) and *Stomoxys calcitrans* (L.) (Diptera: Muscidae). J. Kansas Entomol. Soc. 53: 367–372.

Morgan, P. B., G. C. LaBrecque, D. E. Weidhaas, and R. S. Patterson. 1979. Interrelationship between two species of muscoid flies and the pupal parasite *Spalangia endius* (Hymenoptera: Pteromalidae). J. Med. Entomol. 16: 331–334.

Napoleon, M. E., and B. H. King. 1999. Offspring sex ratio response to host size in the parasitoid wasp *Spalangia endius*. Behav. Ecol. Sociobiol. 46: 325–332.

Petersen, J. J., and J. R. Matthews. 1984. Effects of freezing of host pupae on the production of progeny by the filth fly parasite *Muscidifurax zaraptor* (Hymenoptera: Pteromalidae). J. Kansas Entomol. Soc. 57: 387-393

Rutz, D. A., and R. C. Axtell. 1979. Sustained releases of *Muscidifurax raptor* (Hymenoptera: Pteromalidae) for house fly (*Musca domestica*) control in two types of caged-layer poultry houses. J. Econ. Entomol. 8: 1105–1110.

Siafacas, V. 1980. Biology of *Spalangia nigroaenea* Curtis (Hymenoptera: Pteromalidae), a pupal parasite of the house fly, *Musca domestica* L. M.S. thesis, North Carolina State University, Raleigh.

Skovgård, H., and G. Nachman. 2004. Biological control of house flies *Musca domestica* and stable flies *Stomoxys calcitrans* (Diptera: Muscidae) by means of inundative releases of *Spalangia cameroni* (Hymenoptera: Pteromalidae). Bull. Entomol. Res. 94: 555–567.

Talley, J. L., A. C. Wayadande, L. P. Wasala, A. C. Gerry, J. Fletcher, U. DeSilva, and S. E. Gilliland. 2009. Association of *Escherichia coli* O157:H7 with filth flies (Muscidae and Calliphoridae) captured in leafy greens fields and experimental transmission of *E. coli* O157:H7 to spinach leaves by house flies (Diptera: Muscidae). J. Food Protection 72: 1547–1552.

Thomas, G. D., and S. R. Skoda. 1993. Rural flies in the urban environment? North Central Regional Research Bulletin No. 335. Institute of Agriculture and Natural Resources Research Bulletin No. 317, University of Nebraska Agricultural Research Division, Lincoln, NE.

Wang, Q. M., M. Li, J. Pan, M. C. Di, Q. Y. Liu, F. X. Meng, J. G. Scott, and X. H. Qiu. 2012. Diversity and frequencies of genetic mutations involved in insecticide resistance in field populations of the house fly (*Musca domestica* L.) from China. Pest. Biochem. Physiol. 102: 153–159.

Weinzierl, R. A., and C. J. Jones. 1998. Releases of *Spalangia nigroaenea* and *Muscidifurax zaraptor* (Hymenoptera, Pteromalidae) increase rates of parasitism and total mortality of stable fly and house fly (Diptera, Muscidae) pupae in Illinois cattle feedlots. J. Econ. Entomol. 91: 1114–1121.

Wylie, H. G. 1967. Some effects of host size on *Nasonia vitripennis* and *Muscidifurax raptor* (Hymenoptera: Pteromalidae). Can. Entomol. 99: 742-748.

Zar, J. H. 2010. Biostatistical Analysis, 5th ed. Prentice Hall-Pearson, Upper Saddle River, NJ.