by using the taxonomic keys of Wharton and Gilstrap (1983), White and Elson-Harris (1992) and Smitinand (1980), respectively. Voucher specimens of each species are kept in the Department of Biology, Faculty of Science, Mahidol University.

Chromosome preparations

Mitotic chromosomes of prepupal parasitoids were prepared following the method of Kitthawee et al. (1999). Briefly, live prepupal parasitoids were treated in a few drops of 2% colchicine for 10 min before the cerebral ganglia were dissected out in 1% sodium citrate solution. The ganglia were then transferred to fixative (5:3 absolute ethanol:glacial acetic acid) in order to make cell suspensions. Then a few drops of each cell suspension were spread on a cleaned slide placed on a warmer plate. The air-dried chromosomes were stained with 8% Giemsa for 1 hour. At least 100 cells with well spread metaphase chromosomes were scored and photographed for further analysis. Classification of mitotic chromosome shape was determined based on the centrometric position as described by Levan et al. (1964). A haploid karyogram was prepared for each species and arranged in order from large to small chromosomes.

RESULTS

In braconid Hymenoptera, a female develops from a zygotic cell (fertilized egg) containing a diploid chromosome number (2n) while a male derives from an unfertilized egg (n). The diploid chromosome numbers of the five species differed markedly ranging from 34 to 46 (Fig. 1). Haploid karyograms are presented in Fig. 2 and briefly described below.

Psyttalia fletcheri: This species showed a diploid karyotype of 2n = 34 and had 9 large (nos. 1-9) and 8 small (nos. 10-17) chromosomes (Fig. 1A). These metaphase chromosomes were classified into 7 metacentric (M) (nos. 1, 2, 3, 7, 9, 10 and 17), 7 submetacentric (SM) (nos. 4, 5, 6, 11, 12, 14 and 15) and 3 subtelocentric (ST) (nos. 8, 13 and 16) chromosomes (Fig. 2A).

Psyttalia incisi: The diploid karyotype of *P. incisi* was 2n = 34 similar to that of *P. fletcheri* described above (Fig. 1B). But the chromosomes comprises 6 M (nos. 2, 6, 9, 11, 13 and 15), 9 SM (nos. 1, 3, 4, 5, 7, 12, 14, 16 and 17) and 2 ST (nos. 8 and 10) (Fig. 2B).

Diachasmimorpha longicaudata: This species exhibited a higher chromosome number (2n = 40) (Fig. 1C) compared with the two mentioned above. The haploid karyotype consisted of 5 M (nos. 16, 17, 18, 19 and 20), 13 SM (nos. 1, 2, 3, 5, 6, 7, 8, 10, 11, 12, 13, 14 and 15) and 2 ST (nos. 4 and 9) chromosomes (Fig. 2C). Submetacentric chromosome no. 1 exhibited a distinct secondary constriction in the long arm.

Diachasmimorpha dacusii: This species showed a diploid karyotype 2n = 40 (Fig. 1D) similar to *D. longicaudata*. The metaphase chromosomes consisted of 6 M (nos. 1, 2, 8, 9, 15 and 17), 12 SM (nos. 3, 5, 7, 10, 11, 12, 13, 14, 16, 18, 19 and 20) and 2 ST (nos. 4 and 6) chromosomes (Fig. 2D). ***Fopius arisanus***: The diploid karyotype of *F. arisanus* was 2n = 46 (Fig. 1E). The metaphase chromosomes consisted of 5 M (nos. 4, 7, 12, 18 and 19), 13 SM (nos. 1, 3, 5, 6, 8, 9, 11, 13, 14, 17, 20, 21 and 22) and 5 ST (nos. 2, 10, 15, 16 and 23) chromosomes (Fig. 2E).

DISCUSSION

Morphological study has long been considered as the best method available for separation of species of fruit fly endoparasitoids. However, external morphological characters of specimens have sometimes proved inadequate for systematic studies due to small body size and variation in taxonomic characters of parasitoids (Wharton and Gilstrap 1983). In such circumstance, additional evidence is needed to confirm species separation. Of these, the study of metaphase karyotypes has served as a simple and useful tool for cytotaxonomic study in several groups of animals, particularly in clusters of closely related species and cryptic species in dipteran insects as exemplified by *Drosophila, Anopheles* and *Bactrocera* (Baimai, 1998).

Among Hymenoptera, chromosome information on the Braconidae is very limited. Makino (1951) listed chromosome numbers for only 3 species of Braconidae and only 2 species were covered in the review by Crozier (1975). Recently, Gokhman and Quicke (1995) noted the variability in chromosome numbers of 20 species of the braconid family obtained from the literature between 1927 and 1977. The haploid chromosome number (n) of the braconid family greatly varies, ranging from 4 to 17. Our chromosomal observations of five species of fruit fly parasitoids showed a higher haploid chromosome number (n = 17-23) compared with those for other species in the family Braconidae reported earlier.

The chromosome numbers of the three genera of this study, *Psyttalia* (2n = 34), *Diachasmimorpha* (2n = 40) and *Fopius* (2n = 46), clearly differed (Figs. 1 and 2). According to Crozier (1975), among parasitic Hymenoptera, the primitive chromosome number is 2n = 20. Thus, the *Psyttalia, Diachasmimorpha* and *Fopius* groups may be consecutively considered more advanced than the hypothesized primitive Hymenoptera with respect to chromosomal evolution. Our cytological evidence supports the morphological separation of these genera reported by Wharton and Gilstrap (1983). Furthermore, chromosomal differences among the five species of these solitary endoparasitoids appeared to be correlated with preferences for specific host fruit fly species. Hence, *P.*

fletcheri is specifically parasitic to *Bactrocera cucurbitae* while *P. incisi* parasitises only *B. dorsalis*. Likewise, *D. longicaudata* is an endoparasitoid of *B. correcta* while *D. dacusii* attacks only *B. cucurbitae*. Although *D. longicautata* and *D. dacusii* share the same chromosome numbers and are morphologically similar, their mitotic karyotypes are quite different in size and shape (Figs. 2C and 2D).

Our findings seem to reflect the genetic adaption and coevolution between endoparasitoids and their specific host fruit flies.

Acknowledgements: We are grateful to Dr. John Milne for valuable comments on the manuscript. This work was partly supported by the Thailand Research Fund (RSA/15/2543 and RTA/01/2542) and the TRF/BIOTEC Special Programme for Biodiversity Research and Training (BRT 540050)

REFERENCES

Baimai V. 1998. Heterochromatin accumulation in karyotypic evolution in some dipteran insects. Zool. Stud. **37:** 75-88.

Clausen CP. 1956. Biological control of fruit flies. J. Econ. Entomol. **49:** 176-178.

Crozier RH. 1975. Hymenoptera. In: B. John (ed), "Animal Cytogenetics", Vol. 3. Insecta 7. Gebrüder Borntraeger, Berlin and Stuttgart.

Gokhman VE, DLJ Quicke. 1995. The last twenty years of parasitic Hymenoptera karyology: An update and phylogenetic implications. J. Hym. Res. **4:** 41-63.

Kitthawee S, S Singhapong, V Baimai. 1999. Metaphase chromosomes of parasitic wasp, *Diachasmimorpha longicaudata* (Hymenoptera: Braconidae) in Thailand. Cytologia **64:** 111-115.

Levan A, K Fredga, AA Sandberg. 1964. Nomenclature for centrometric position on chromosomes. Hereditas **52:** 201-220.

Makino S. 1951. An Atlas of the chromosome numbers in animals. Iowa State Univ. Press, Ames.

Smitinand T. 1980. Thai plant names (botanical names - vernacular names). Royal Forest Department, Bangkhen, Bangkok, Thailand.

Wharton RA. 1987. Changes in nomenclature and classification of some opiine Braconidae (Hymenoptera). Proc. Entomol. Soc. Wash. **89:** 61-73.

Wharton RA. 1997. Generic relationships of opiine Braconidae (Hymenoptera) parasitic on fruit-infesting Tephritidae (Diptera). Contri. Amer. Entomol. Inst. **30:** 1-53.

Wharton RA. 1999. A review of the Old World genus *Fopius* Wharton (Hymenoptera: Braconidae: Opiinae), with description of two new species reared from fruit-infesting Tephritidae (Diptera). J. Hym. Res. **8(1):** 48-64.

Wharton RA, FE Gilstrap. 1983. Key to and status of opiine braconid (Hymenoptera) parasitoids used in biological control of *Ceratitis* and *Dacus* s.l. (Diptera: Tephritidae). Ann. Entomol. Soc. Am. **76:** 721-742.

Wharton RA, PM Marsh. 1978. New World Opiinae (Hymenoptera: Braconidae) parasitic on Tephritidae (Diptera). J. Wash. Acad. Sci. **68:** 147-167.

White IM, MM Elson-Harris. 1992. Fruit flies of economic significance: their identification and bionomics. Centre for Agriculture and Biosciences International, Wallingford Oxon, UK.

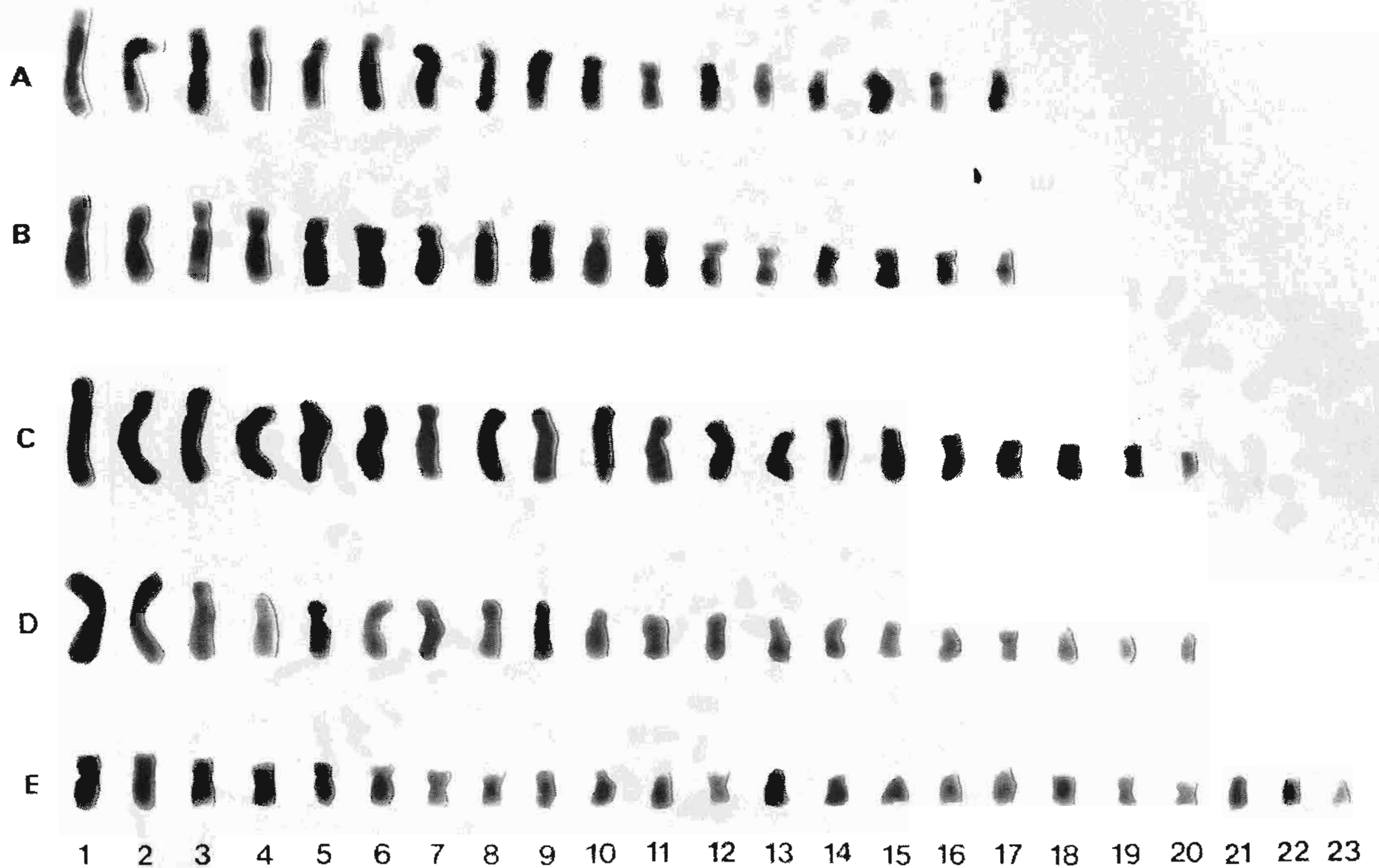
Table 1. Collection data of the five species of braconid endoparasitoids examined cytologically in this study (1 = west, 2 = northeast, 3 = central Thailand).

Species	Host fruit fly species	Host plant Species	Locality (province)	Date of collection
P. fletcheri	*B. cucurbitae*	*Coccinia grandis*	Kanchanaburi[1]	Dec. 1998
P. incisi	*B. dorsalis*	*Averrhoa carambola*	Khon Kaen[2]	Nov. 1999
D. longicaudata	*B. correcta*	*Psidium guajava*	Nakhornpathom[3]	May 1998
D. dacusii	*B. cucurbitae*	*Coccinia grandis*	Kanchanaburi[1]	Jun. 1999
F. arisanus	*B. dorsalis*	*Polyathia longifolla*	Kanchanaburi[1]	Nov. 1998
		Zizyphus mauritiana	Kanchanaburi[1]	Jun. 1999

Figure Legends

Fig. 1. – Photomicrographs of diploid metaphase chromosomes (2n) of five species of braconid parasitoids: A) *Psyttalia fletcheri,* 34; B) *Psyttalia incisi,* 34; C) *Diachasmimorpha longicaudata,* 40; D) *Diachasmimorpha dacusii,* 40; E) *Fopius arisanus,* 46. The bar indicates 5 μm.

Fig. 2. – Karyogram of the haploid set (n) of metaphase chromosomes of the five species: A) *Psyttalia fletcheri,* 17; B) *Psyttalia incisi*, 17; C) *Diachasmimorpha longicaudata,* 20; D) *Diachasmimorpha dacusii*, 20; E) *Fopius arisanus*, 23.


A
B
C
D
E
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23

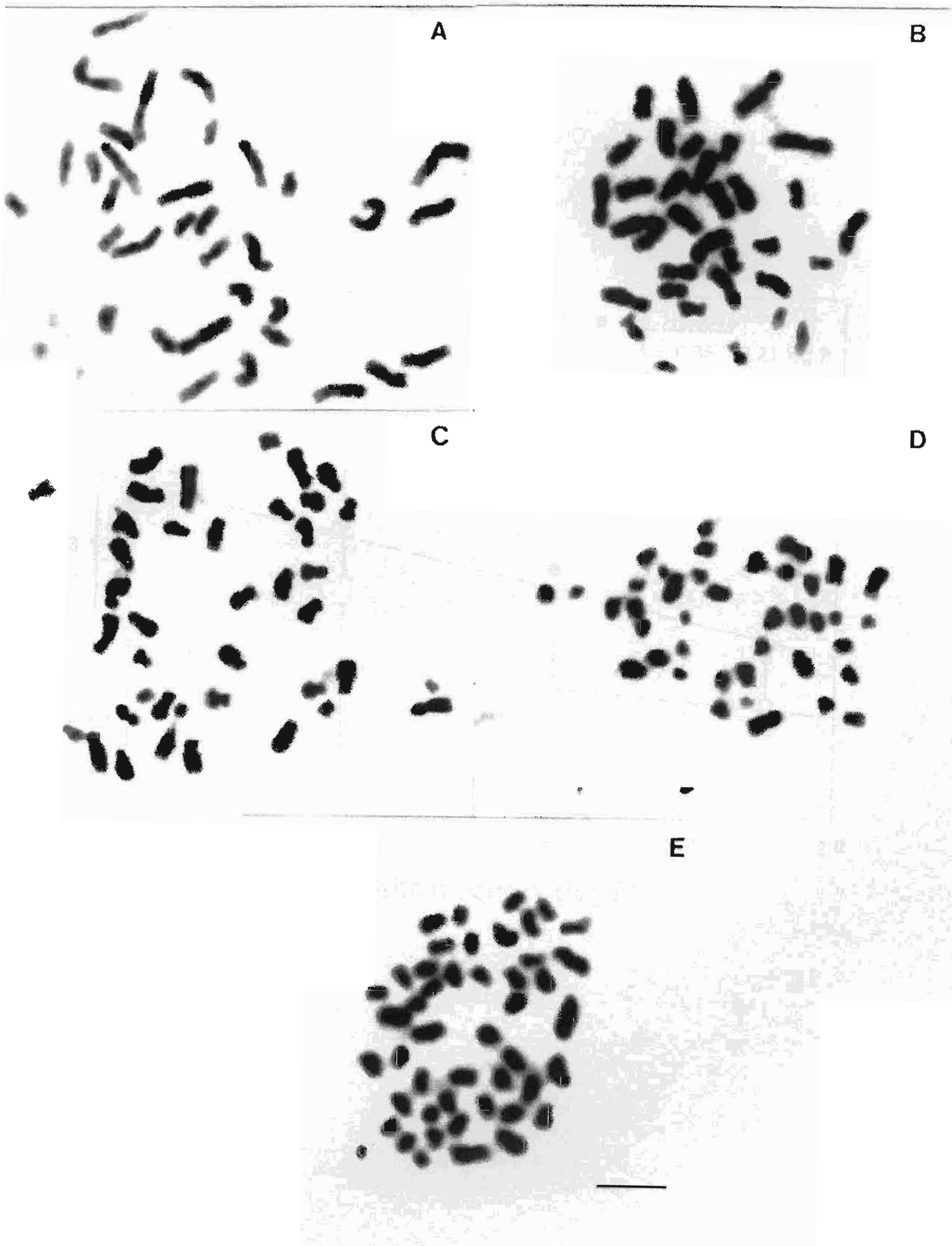
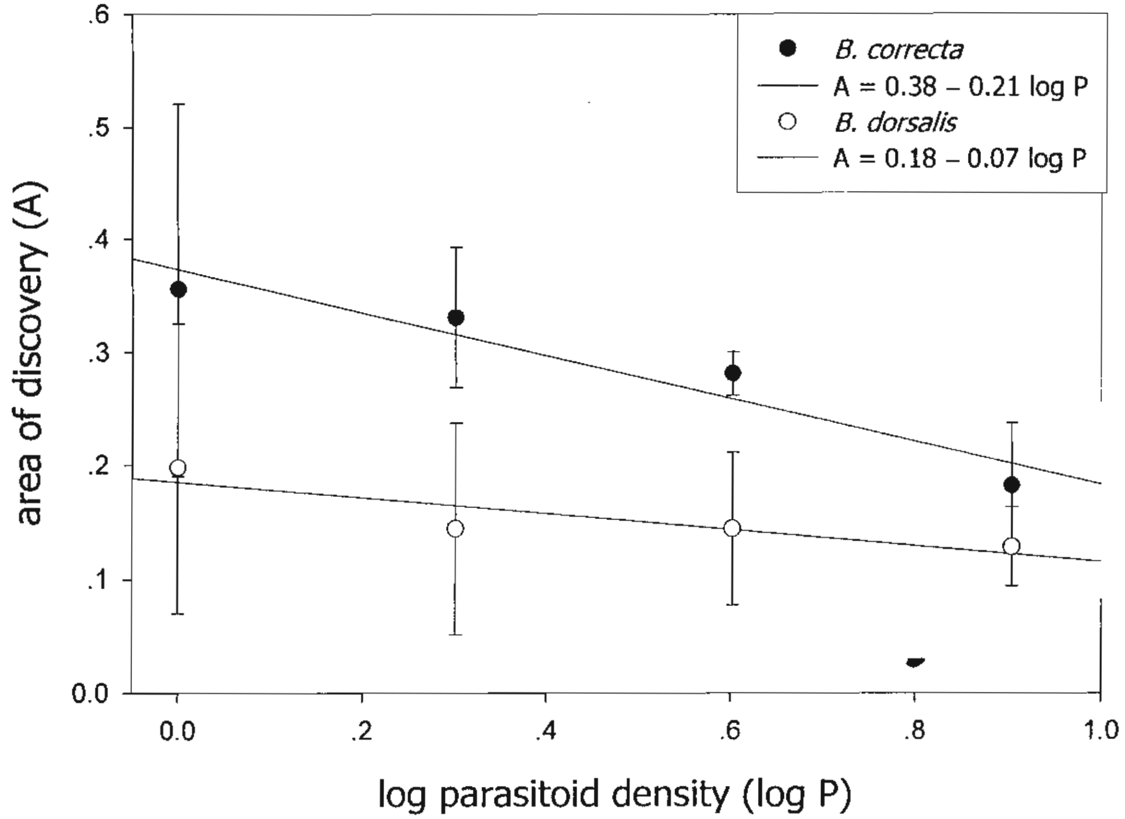


Fig. 2



B. correcta
A = 0.38 – 0.21 log P
B. dorsalis
A = 0.18 – 0.07 log P
area of discovery (A)
.6
.5
.4
.3
.2
.1
0.0
0.0
.2
.4
.6
.8
1.0
log parasitoid density (log P)

Fig 3

Abstract

Laboratory experiments were performed on the parasitoid, *Spalangia endius* Walker, attacking the fruit fly pupal hosts, *Bactrocera correcta* (Bezzi) and *B. dorsalis* (Hendel), to determine the effects of parasitoid age, pupal age, and density of both parasitoids and pupae on attack rates. *Spalangia endius* females attack at peak rates at approximately 3 days of age. The mean numbers of host pupae attacked per female parasitoid were 8.42 ± 1.19 for *B. correcta* and 7.37 ± 1.72 for *B. dorsalis*, for parasitoids aged 1–7 days. The rate of parasitism of *B. dorsalis* declined to below 50% by day 7 of pupal age, but that of *B. correcta* remained high (> 90%). The experiments on varying host density determined that the numbers of pupae parasitized increased with host density, but the percentage parasitism declined, or inversely density dependent. The results suggested that female *S. endius* exhibited Type II functional response. The ovipositional behavior of the parasitoid on the two species of pupal hosts was at random. In the experiments on variable host (or parasitoid) density, the percentage parasitism in *B. correcta* was higher than that of *B. dorsalis* at all densities (paired *t*-tests $P < 0.05$). The oviposition efficiency of *S. endius* on *B. correcta* declines with parasitoid density, and can be described by the regression: A = 0.38 – 0.21 log P (*F*

= 8.39, $df = 1{,}10$, $P < 0.05$, $r^2 = 0.46$) where A represents the area of discovery and P is number of parasitoids searching. However, searching efficiency of *S. endius* on *B. dorsalis* is lower and relatively constant with parasitoid density: A = 0.18 – 0.07 log P ($F = 1.03$, $df = 1{,}10$, $P = 0.33$, $r^2 = 0.09$). These results suggest that host and parasitoid densities play important role in the attack rate of the parasitoid, *S. endius* and that it may be more effective in biological control of tephritid fruit fly, *B. correcta* than of *B. dorsalis*.

Key words: *Spalangia endius*, tephritid fruit fly, parasitism, host density, parasitoid density, searching efficiency, biological control.

INTRODUCTION

Fruit flies (Tephritidae) are major insect pests in many regions of the world (White & Harris 1992). They are widely distributed and infest a wide variety of fruits and vegetables. Early attempts to suppress infestations of fruit flies resulted in the use of exotic entomophagous species for biological control (Bess 1953; Bess *et al.* 1961).

Spalangia spp. (Pteromalidae) are pupal parasitoids of various dipteran hosts (Clancy 1950; Schmidt & Morgan 1978; Meyer *et al.* 1991; Gut & Brunner 1994; Hogsette *et al.* 1994) including tephritid fruit flies (Hardy 1973; White & Harris 1992). In Thailand, *S. endius* has been found in mixed infestations in the tephritid fruit flies, *B. correcta* and *B. dorsalis* (Kitthawee, unpublished). Thus *S. endius* may be considered a potential biological control agent against fruit flies in Thailand.

There have been reports of house fly and pteromalid parasitoid interactions (Morgan *et al.* 1975; Weidhaas *et al.* 1977). However, information on the relationships between the pteromalid parasitoid, *S. endius*, and its fruit fly hosts is still lacking. The investigation of host-parasitoid interactions may provide useful information for control of fruit flies in Thailand. In this report, we present experimental results on the relationships between *S. endius* and their fruit fly hosts, *B. correcta* and *B. dorsalis*, concerning four aspects: 1) the suitable age of hosts and parasitoids; 2) host preference of *S. endius*; 3) the effects of host density on success of the parasitoids; and 4) the effects of parasitoid density on attack rate at constant host density.

METHODS

Parasitoid and hosts

Samples of *S. endius* were collected around Bangkok and were reared in the insectary of the Department of Biology, Faculty of Science, Mahidol University. The *S. endius* colony has been maintained on pupae of the fruit flies, *B. correcta* and *B. dorsalis*, which have been reared on bananas under laboratory conditions at 27 ± 2 °C and 70 ± 10 %RH.

Age suitability of fruit fly hosts

Seven different aged groups (1–7 days old) of pupal fruit flies were marked by different colors of luminous paint (BioQuip Product, Inc.). Ten fruit fly pupae in each age group were placed in a plastic cage (10 x 10 x 6 cm). Each cage of pupae was exposed to 7 adult female parasitoids for 24 h. Then the parasitoids were removed and the fruit fly pupae were isolated by age group in different containers for about 5 days. The fruit fly pupae were dissected under a stereomicroscope to investigate their parasitism pupae. The number of parasitized pupae of both *B. correcta* and *B. dorsalis* was analyzed to determine host suitability. This procedure was replicated 3 times for each species of fruit fly.

Age suitability of parasitoid *S. endius*

Four emerging *S. endius* were placed in each plastic cage (7 x 9.5 x 5 cm) and 40 pupal fruit flies 1 day old were also placed in the same cage. After 24 h, the pupal fruit flies were removed and replaced by a new set of host pupae aged 1 day. This procedure was performed for 7 consecutive days. Upon removal, the pupae were dissected for parasitoids. The data were analyzed for daily parasitism per parasitoid female in relation to parasitoid age. Three replications were performed for each species of fruit fly.

Effects of host density on parasitoid success

Groups of 1-day-old fruit fly pupae were set up in plastic cages (10 x 10 x 6 cm) at densities at 10, 20, 40 and 80 pupae per cage. Four female parasitoids aged 3 days were introduced into each cage and kept under controlled laboratory conditions. After 24 h, the parasitoids were removed from each cage, and the number of fruit fly pupae parasitized was counted and recorded. There were 3 replicates for each fruit fly species and density, for a total of 24 experimental cages.

Effects of parasitoid density on host searching

Parasitoids were set up in plastic cages at 4 densities, 1, 2, 4 and 8 parasitoids per cage, with each cage containing 40 pupal fruit fly hosts 1 day old. After 24 h, the parasitoids were removed from each cage and the number of surviving fruit flies was recorded. This procedure was replicated 3 times for each fruit fly species.

Data analysis

Descriptive statistics [mean ($\overline{X}$) and standard deviation (SD)] were used to compare the number (or the percentage) of fruit flies parasitized. Mean numbers or percentages of parasitized fruit fly pupae among pupal age groups and parasitoid age groups were compared using one-way analysis of variance (ANOVA). Comparisons of means were also made using the least significant differences (LSD) test. The age suitability of fruit fly hosts and parasitoids was determined. The two-sample *t*-test was used to compare the differences between the overall mean number (or percentage) of parsitized pupae of *B. correcta* and *B. dorsalis*, to determine the host preference.

For each density of fruit flies (or parasitoids), the average number (or percentage) of pupae parasitized was calculated. The mean number (or percentage) of parasitized pupae of *B. correcta* and *B. dorsalis* was compared using the paired *t*-test. The relationships between the number (or the percentage) of hosts parasitized and host (or parasitoid) density were evaluated by regression analysis. Variances were compared by Bartlett's test. For unequal variances, weighted regression was performed by the inverse of variance of the number (or the percentage) of hosts parasitized. A log transformation was performed on data to linearize the relationships. Normality was verified by the Shapiro-Wilk test.

The random oviposition of parasitoids was predicted by using the "random attack model" (Rogers 1972). The model is

$$N_{par} = N(1 - e^{-Enc/N})$$

where N_{par} = the number of hosts parasitized, N = the total number of hosts, and Enc = the total number of encounters that the parasitoids make with the hosts. In order to determine whether the data fit the random oviposition of parasitoid or not, we used the chi-square test for goodness of fit.

Percentage parasitism was defined as the number of fruit fly pupae parasitized divided by the number fruit fly pupae per cage. We investigated the relationship between the percentage parasitism and host density. If it is positive, the relationship is density dependent. If the percentage parasitism declines with increasing host density, the relationship is inversely density dependent.

The ability of parasitoids to find or attack the hosts may depend not only on host density but also on their ability to find or attack hosts (or the parasitoid's "area of discovery"). The "area of discovery" for each density of parasitoids ovipositing at a constant density of fruit flies was calculated from the formula (Varley et. al. 1973):

$$A = (1/P)*(\log_e N/S)$$

where A = area of discovery, P = the number of parasitoids searching, N = the number of hosts exposed, and S = the number of hosts not parasitized. A simple linear regression model was tested for the relationship between the area of discovery of *S. endius* and its density. All analyses were performed with Statistix® (NH Analytical Software).

RESULTS

Age suitability of fruit fly pupae

The mean percentage parasitism of *S. endius* varied with age of fruit fly pupae and species. Pupae of *B. correcta* of all ages were attacked by *S. endius* about equally ($F = 0.88$, $df = 6,14$, $P > 0.05$) while pupae of *B. dorsalis* were attacked by *S. endius* at a rate that tended to decline with pupal age ($F = 2.83$, $df = 6,14$, $P = 0.05$) (Table 1). The highest mean percentage parasitism of *B. dorsalis* was on the first day puparium (93.3 ± 11.5%); hence, fruit fly pupae aged 1 day were used later in our experiments. However, the overall mean percentages of parasitism for *B. correcta* and *B. dorsalis* were significantly different in a two-sample *t*-test under unequal variances ($t = 4.17$, $df = 26.70$, $P < 0.001$). The overall mean percentage parasitism for *B. correcta* (93.8 ± 8.1%) was significantly higher than for *B. dorsalis* (74.6 ± 19.4%).

Age suitability of parasitoid, *S. endius*

The mean parasitism of pupal hosts per parasitoid for different parasitoid age groups was compared by using one-way ANOVA. The results showed that the attack rate of *S. endius* did not vary significantly with age group for either *B. correcta* ($F = 1.38$, $df = 6,14$, $P > 0.05$) or *B. dorsalis* ($F = 0.38$, $df = 6,14$, $P > 0.05$). However, *S. endius* 3 days old attacked pupae at the highest rate: *B. correcta* with an average of 9.42 ± 0.80 pupae per female parasitoid and *B. dorsalis* with 8.33 ± 1.28 pupae (Table 2). The overall mean numbers of pupae parasitized per female parasitoid for *B. correcta* and *B. dorsalis* were 8.42 ± 1.19 and 7.37 ± 1.72, respectively. These means were significantly different in a two-sample *t*-test ($t = 2.29$, $df = 40$, $P < 0.05$). These results showed that *S. endius* preferably attacked *B. correcta* more than *B. dorsalis*.

Effects of pupal host density

There were significant effects of pupal host density on the rate of parasitism with *S. endius* density held constant at 4 females. Increase in the density of pupal hosts caused an increase in the number parasitized, but a decrease in percentage parasitism for both *B. correcta* and *B. dorsalis* (Table 3). The range of pupae parasitized per female parasitoid for each host density group (10–80 pupae) was 2.00 ± 0.00 to 10.75 ± 2.41 (*B. correcta*) and 1.42 ± 0.14 to 7.00 ± 0.66 (*B. dorsalis*). Regression analysis suggested that the relationships between pupal host density (N) and the number hosts parasitized (N_{par}) were significant for both *B. correcta* ($F = 84.71$, $df = 1,10$, $P < 0.001$, $r^2 = 0.89$) and *B. dorsalis* ($F = 69.63$, $df = 1,10$, $P < 0.001$, $r^2 = 0.87$) (Figs. 1a, 1b). At low pupal host density, the number parasitized was low, but it increased with increasing host density. Apparently, *S. endius* attacked more pupal hosts when available, suggesting a Type II functional response (Table 3). The ovipositional behavior of *S. endius* at different pupal host densities for *B. correcta* and *B. dorsalis* apparently occurred at random ($\chi^2 = 4.16$, $df = 11$, $P > 0.05$ and $\chi^2 = 4.13$, $df = 11$, $P > 0.05$, respectively) (Table 4).

The percentage parasitism of fruit fly pupae was inversely density dependent. There was a significant decrease in percentage parasitism with increasing pupal host density for both *B. correcta* ($F = 18.41$, $df = 1,10$, $P < 0.005$, $r^2 = 0.65$) and *B. dorsalis* ($F = 11.44$, $df = 1,10$, $P < 0.01$, $r^2 = 0.53$) (Figs. 1a, 1b). The mean percentage parasitism differed significantly between *B. correcta* and *B. dorsalis* (paired *t*-test, $t = 8.18$, $df = 11$, $P < 0.001$) (Table 3).

Effects of parasitoid density

The percentage parasitism ($\%N_{par}$) increased with parasitoid density (P) in both *B. correcta* and *B. dorsalis* (Table 5). The relationship was highly significant in *B. correcta* ($F = 51.16$, $df = 1,10$, $P < 0.001$, $r^2 = 0.84$) and in *B. dorsalis* ($F = 34.92$, $df = 1,10$, $P < 0.001$, $r^2 = 0.78$) (Figs. 2a, 2b). The mean percentage parasitism in *B. correcta* was significantly higher than that of *B. dorsalis* (paired *t*-test, $t = 5.25$, $df = 11$, $P < 0.001$). The number of fruit fly pupae parasitized per parasitoid, however, was higher at low parasitoid density (Table 5). There was a significant decrease in number of pupae parasitized but increase in pupae parasitized per parasitoid female with increasing parasitoid density for *B. correcta* ($F = 87.17$, $df = 1,10$, $P < 0.001$, $r^2 = 0.89$) but a weaker relationship for *B. dorsalis* ($F = 5.17$, $df = 1,10$, $P < 0.05$, $r^2 = 0.34$) (Figs. 2a, 2b).

The ovipositional efficiency of *S. endius* females can be described by the area of discovery: $A = (1/P)*(\log_e N/S)$. The effect of *S. endius* density on searching efficiency for *B. correcta* can be presented as:

$$A = 0.38 - 0.21 \log P$$

($F = 8.39$, $df = 1{,}10$, $P < 0.05$, $r^2 = 0.46$). When the parasitoid density was increased at constant host density, the area of discovery tended to decrease (Fig. 3).

The relationship between the area of discovery and parasitoid density for *B. dorsalis*, however, is weak (A = 0.18 – 0.07 log P) and not significant ($F = 1.03$, $df = 1{,}10$, $P = 0.33$, $r^2 = 0.09$) (Fig. 3). The slope of the relationship is approximately 0, and the area of discovery (A) is approximately constant (A = 0.18).

DISCUSSION

Spalangia endius has been shown to be a solitary pupal parasitoid useful for biological control of several species of flies (Legner & Brydon 1966; Morgan *et al.* 1981). Based on our results, *S. endius* is a solitary parasitoid of fruit fly (*Bactrocera*) pupae. The host suitability test showed that *S. endius* females positively responded to both *B. correcta* and *B. dorsalis* pupae. They attacked pupae of *B. correcta* of all ages effectively, but attacked younger (aged 1– 3 days) pupae of *B. dorsalis* more than older pupae. A high percentage parasitism was also reported in various parasitoid host pupae aged 2–3 days (Wylie 1963; Chabora & Pimentel 1966; Morgan *et al.* 1979). When they attack old pupal hosts, the host may emerge before the parasitoid eggs hatch. As pupal age had no effect on parasitism of *B. correcta*, *S. endius* may be more effective in controlling natural populations of this species.

Female *S. endius* laid only one egg per pupa in these fruit flies; no superparasitism was observed in this study. On the day of emergence adult females started to oviposit and continued at a constant rate; parasitoid age had no significant effect on the rate of parasitism, although 3-day-old females produced the highest rate of parasitism. The overall mean rate of parasitism in *B. correcta* (8.42 pupae per day) was significantly higher than that of *B. dorsalis* (7.37 pupae per day) suggesting that *S. endius* may be a more effective control agent for the former species.

Host density and parasitoid density are important factors affecting fly mortality and parasitism (Hassell 1978; Mann *et al.* 1990a,b). In our experiments, at low host density, pupal hosts were attacked at a high attack rate which was somewhat inversely density dependent. There was strong competition for hosts and a high percentage of parasitism (Figs. 1a, 1b). At higher pupal host densities, the number of hosts parasitized increased but the percentage parasitized declined. Legner (1967) also showed that *S endius* had a higher attack at higher host density as a functional response. The rates of parasitism observed support the random attack model (Table 4) (Rogers 1972), with a type II functional response

(Table 3). Since the functional response resulted in inversely density-dependent mortality, the parasitoid may be incapable of controlling or regulating the host population by itself (Hassell & May 1973; Hassell 1978). However, most biological control programs are intended to inundate the host population, so that a numerical response should result. It may also be possible to obtain a type III functional response under some environmental conditions which would achieve biological control of the fruit fly. Our results call for field trial experiments, and several other factors should be taken into consideration before a conclusion can be made.

In the experiments on variable parasitoid density, the number of pupae parasitized per female parasitoid and the percentage parasitism were similar to those with varying host density. (Tables 3, 5). Increasing parasitoid density did not result in greater numbers of pupae parasitized per female parasitoid. It therefore may be assumed that increased parasitoid density will reduce the response in the parasitoid, *S. endius*. The efficiency of finding or attacking host pupae can be measured by the "area of discovery". Increases in parasitoid density caused increases the percentage parasitism but decreases in the area of discovery in *B. correcta*. Our results agree with those described by Hassell & Varley (1969) with regard to increase in parasitism being related to decrease in the area of discovery. On the other hand, the area of discovery in *B. dorsalis* was nearly constant at approximately A = 0.18, which agrees more closely with Nicholson's assumption that parasitoids would search for their hosts at a constant area of discovery (Nicholson 1933; Nicholson & Bailey 1935).

The data on *B. correcta* imply that the parasitoid did not lay eggs on already parasitized pupae, and that *S. endius* is a solitary parasitoid of fruit fly hosts. If it can discriminate between parasitized and unparasitized host pupae, it should be more useful for biological control against *B. correcta.* Nonetheless, detailed studies under field conditions are necessary before releasing *S. endius* for control the fruit fly in population.

ACKNOELEDGMENTS

We thank Miss Chonlawan Tong-Aum for assisting in rearing insects. This work was financially supported by the Thailand Research Fund, RSA/15/2543 and RTA/01/2542.

REFERENCES

Analytical Software. (2000) *Statistix*, version 7, Analytical Software, Tallahassee, Florida.

Bess H. A. (1953) Status of *Ceratitis capitata* in Hawaii following the introduction of *Dacus dorsalis* and its parasites. *Proc. Hawaii. Entomol. Soc.* **15**, 221–34.

Bess H. A., van den Bosch R. & Haramoto F. H. (1961) Fruit fly parasites and their activities in Hawaii. *Proc. Hawaii. Entomol. Soc.* **17**, 367–78.

Chabora P. C. & Pimentel D. (1966) Effect of host (*Musca domestica* Linnaeus) age on the pteromalid parasite *Nasonia vitripennis* (Walker). *Can. Entomol.* **98**, 1226–31.

Clancy D. W. (1950) Notes on parasites of tephritid flies. *Proc. Hawaii. Entomol. Soc.* **14**, 25–6.

Gut L. J. & Brunner J. F. (1994) Parasitism of the apple maggot, *Rhagoletis pomonella*, infesting hawthorns in Washington. *Entomophaga* **39**, 41–9.

Hardy D. E. (1973) The fruit flies (Tephritidae: Diptera) of Thailand and bordering countries. *Pac. Insects Monogr.* **31**, 1–353.

Hassell M. P. (1978) *The Dynamics of Arthropod Predator-Prey Systems,* Princeton University Press, Princeton, New Jersey.

Hassell M. P. & May R. M. (1973) Stability in insect host-parasite models. *J. Anim. Ecol.* **42**, 693–726.

Hassell M. P. & Varley G. C. (1969) New inductive population model for insect parasites and its bearing on biological control. *Nature, Lond.* **223**, 1133–7.

Hogsette J. A., Farkas R. & Coler R. R. (1994) Hymenopteran pupal parasites recovered from house fly and stable fly (Diptera: Muscidae) pupae collected on livestock and poultry facilities in northern and central Hungary. *Environ. Entomol.* **23**, 778–81.

Legner E. F. (1967) Behavior changes the reproduction of *Spalangia cameroni*, *S. endius*, *Muscidifurax raptor*, and *Nasonia vitripennis* (Hymenoptera: Pteromalidae) at increasing fly host densities. *Ann. Entomol. Soc. Am.* **60**, 819–26.

Legner E. F. & Brydon H. W. (1966) Suppression of dung-inhabiting fly populations by pupal parasites. *Ann. Entomol. Soc. Am.* **59**, 638–51.

Mann J. A., Axtell R. C. & Stinner R. E. (1990) Temperature-dependent development and parasitism rates of four species of Pteromalidae (Hymenoptera) parasitoids of house fly (*Musca domestica*) pupae. *Med. Vet. Entomol.* **4**, 245–53.

Mann J. A., Stinner R. E. & Axtell R. C. (1990) Parasitism of house fly (*Musca domestica*) pupae by four species of Pteromalidae

(Hymenoptera): effects of host-parasitoid densities and host distribution. *Med. Vet. Entomol.* **4**, 235–43.

Meyer J. A., Shultz T. A., Collar C. & Mullens B. A. (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**, 915–21.

Morgan P. B., LaBrecque G. C., Weidhaas D. E. & Patterson R. S. (1979) Interrelationship between two species of muscoid flies and the pupal parasite *Spalangia endius* (Hymenoptera: Pteromalidae). *J. Med. Entomol.* **16**, 331–4.

Morgan P. B., Patterson R. S., LaBrecque G. C., Weidhaas D. E. & Benton A. (1975) Suppression of a field population of houseflies with *Spalangia endius*. *Science* **189**, 388–9.

Morgan P. B., Weidhaas D. E. & Patterson R. S. (1981) Programmed releases of *Spalangia endius* and *Muscidifurax raptor* (Hymenoptera: Pteromalidae) against estimated populations of *Musca domestica* (Diptera: Muscidae). *J. Med. Entomol.* **18**, 158–66.

Nicholson A. J. (1933) The balance of animal populations. *J. Anim. Ecol.* **2**, 132–78.

Nicholson A. J. & Bailey V. A. (1935) The balance of animal population. Part I. *Proc. Zool. Soc. Lond.* pp. 551–98.

Rogers D. J. (1972) Random search and insect population models. *J. Anim. Ecol.* **41**, 369–83.

Schmidt C. D. & Morgan P. B. (1978) Parasitism of pupae of the horn fly, *Haematobia irritans* (L.) by *Spalangia endius* Walker. *Southwest-Entomol.* **3**, 69-72.

Varley C. G., Gradwell G. R. & Hassell M. P. (1973) *Insect Population Ecology: An Analytical Approach,* University of California Press, Berkeley and Los Angeles, California.

Weidhaas D. E., Haile D. G., Morgan P. B. & LaBrecque G. C. (1977) A model to simulate control of house flies with a pupal parasite, *Spalangia endius*. *Environ. Entomol.* **6**, 489-500.

White I. M. & Elson-Harris M. M. (1992) *Fruit Flies of Economic Significance: Their Identification and Bionomic,* Centre for Agriculture and Biosciences International Wallingford Oxon, UK.

Wylie H. G. (1963) Some effects of host age on parasitism by *Nasonia vitripennis* (Walk.) (Hymenoptera: Pteromalidae). *Can. Entomol.* **95**, 881–6.

Table 1. Mean (± SD) percentage pupae parasitized at different pupal ages.

Day*	Mean** % pupae parasitized	
	B. correcta	*B. dorsalis*
1	96.7 ± 5.8 [a]	93.3 ± 11.5 [a]
2	93.3 ± 5.8 [a]	81.7 ± 16.8 [a]
3	100.0 ± 0.0 [a]	80.3 ± 18.6 [a]
4	90.0 ± 10.0 [a]	73.0 ± 10.9 [ab]
5	96.3 ± 6.4 [a]	81.7 ± 16.8 [a]
6	86.7 ± 15.3 [a]	66.7 ± 14.4 [ab]
7	93.3 ± 5.8 [a]	45.8 ± 18.2 [b]
Overall mean	93.8 ± 8.1	74.6 ± 19.4

* = Day, age of fruit fly hosts after puparium.
** = Mean within a column followed by the same letter are not significantly different (comparison method's LSD test, $P < 0.05$)

Table 2. Mean (± SD) number pupae parasitized per *S. endius* female for each parasitoid age.

Day*	Mean no. pupae parasitized	
	B. correcta	*B. dorsalis*
1	6.92 ± 1.70	7.58 ± 1.26
2	8.42 ± 0.38	8.17 ± 0.95
3	9.42 ± 0.80	8.33 ± 1.28
4	8.33 ± 0.29	7.08 ± 1.53
5	8.92 ± 0.72	6.92 ± 2.47
6	8.58 ± 0.72	7.00 ± 2.05
7	8.33 ± 2.04	6.50 ± 2.95
Overall mean	8.42 ± 1.19	7.37 ± 1.72

* = Day, age of *S. endius* after emergence.

Table 3. Mean (± SD) number of pupae parasitized after exposure for 24 h to constant *S. endius* density (4 females) at different pupal densities.

Host density	Parasitoid: host ratio	Mean no. pupae parasitized per *S. endius*		Mean % pupae parasitized	
		B. correcta	*B. dorsalis*	*B. correcta*	*B. dorsalis*
10	1:2.5	2.00 ± 0.00	1.42 ± 0.14	80.0 ± 0.0	56.7 ± 5.8
20	1:5	3.58 ± 0.29	2.42 ± 0.63	71.7 ± 5.8	48.3 ± 12.6
40	1:10	7.08 ± 0.29	4.08 ± 1.18	70.8 ± 2.9	40.3 ± 11.8
80	1:20	10.75 ± 2.41	7.00 ± 0.66	53.7 ± 12.0	35.0 ± 3.3

Table 4. Mean (± SD) number of pupae parasitized per *S. endius* female after fruit fly pupae were exposed for 24 h to constant *S. endius* density (4 females) for different pupal densities compared to the predicted mean (Roger, 1972).

Host density	Parasitoid: host ratio	*B. correcta*		*B. dorsalis*	
		Observed	Predicted*	Observed	Predicted*
10	1:2.5	2.00 ± 0.00	1.63 ± 0.00	1.42 ± 0.14	1.89 ± 0.06
20	1:5	3.58 ± 0.29	3.44 ± 0.12	2.42 ± 0.63	3.95 ± 0.27
40	1:10	7.08 ± 0.29	6.92 ± 0.12	4.08 ± 1.18	8.23 ± 0.51
80	1:20	10.75 ± 2.41	15.33 ± 1.05	7.00 ± 0.66	16.96 ± 0.29

* = random oviposition of parasitoid was calculated from $N_{par} = N(1 - e^{-Enc/N})$
$\chi^2 = 4.16$; $df = 11$; $P > 0.05$ (random oviposition of parasitoid on *B. correcta*)
$\chi^2 = 4.13$; $df = 11$; $P > 0.05$ (random oviposition of parasitoid on *B. dorsalis*)

Table 5. Mean (± SD) number of pupae parasitized after 40 fruit fly pupae were exposed to varying densities of *S. endius* females for 24 h.

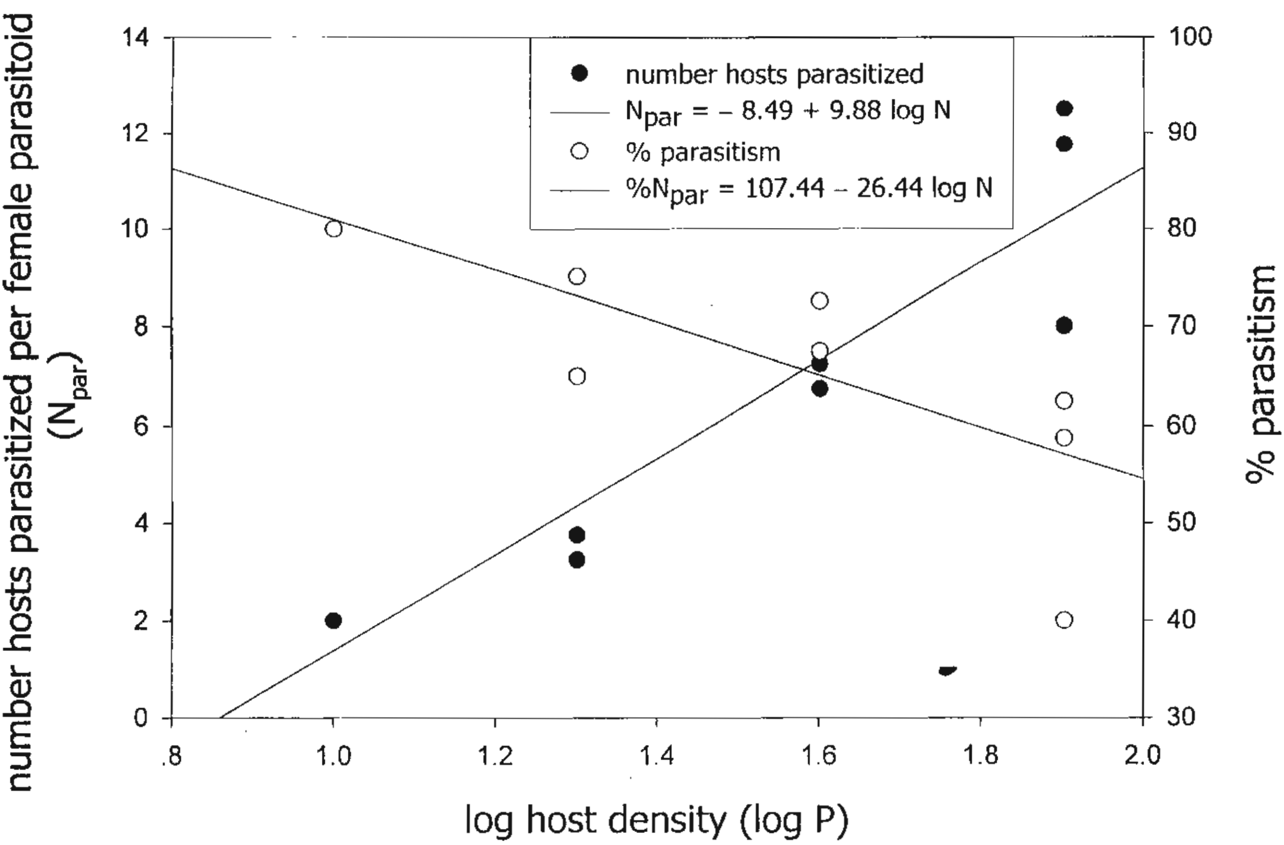
Parasitoid density	Parasitoid: host ratio	Mean no. pupae parasitized per *S. endius*		Mean % pupae parasitized	
		B. correcta	*B. dorsalis*	*B. correcta*	*B. dorsalis*
8	1:5	3.92 ± 0.44	3.17 ± 0.50	78.3 ± 8.8	63.3 ± 10.1
4	1:10	6.17 ± 0.29	4.25 ± 1.50	61.7 ± 2.9	42.5 ± 15.0
2	1:20	9.67 ± 1.26	4.83 ± 2.93	48.3 ± 6.3	24.2 ± 14.6
1	1:40	12.33 ± 4.73	7.00 ± 4.36	30.8 ± 11.8	17.5 ± 10.9

Figure Legends

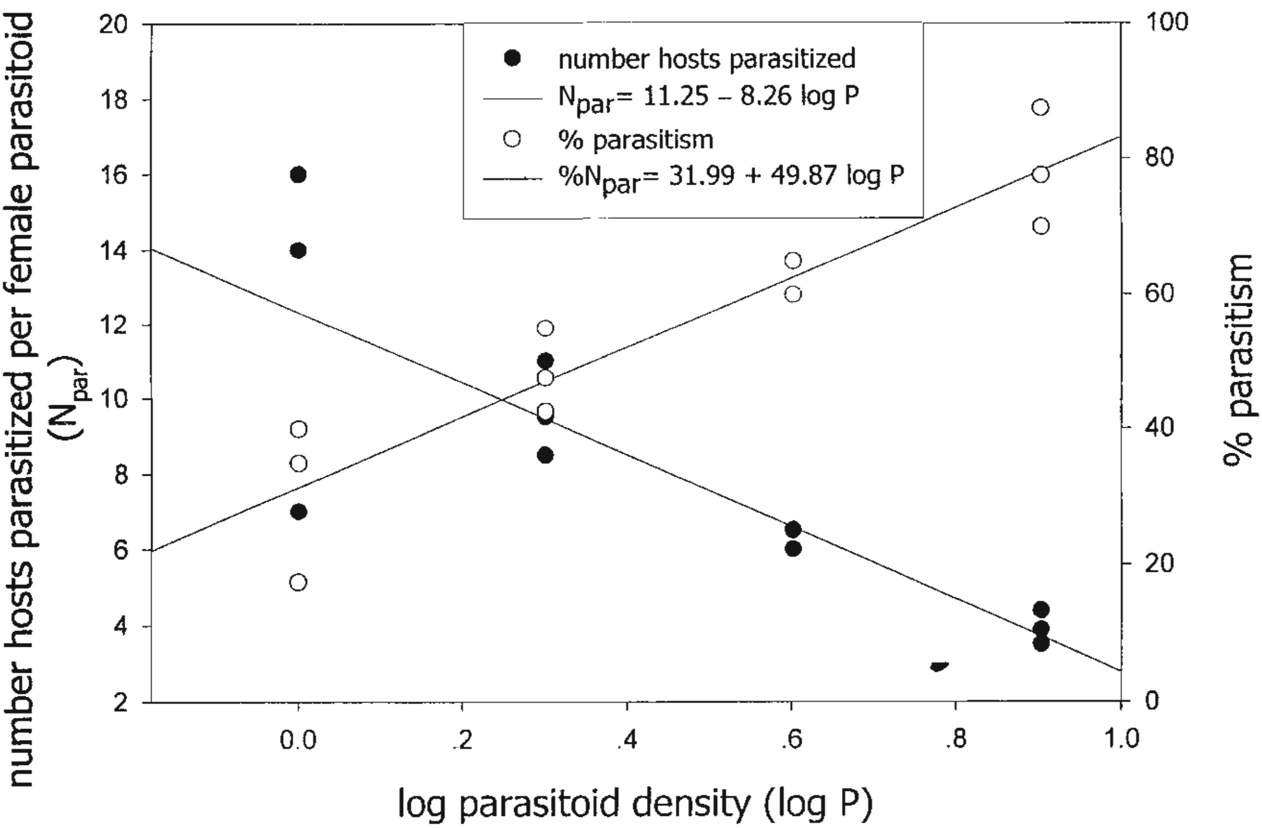
Fig. 1. Relationship between number of hosts parasitized per female parasitoid (left) or percentage parasitism (right), and pupal host density of (a) *B. correcta*, and (b) *B. dorsalis*.

Fig. 2. Relationship between number of hosts parasitized per female parasitoid (left) or percentage parasitism (right), and parasitoid density of (a) *B. correcta* and (b) *B. dorsalis*.

Fig. 3. Relationship between area of discovery and parasitoid density.

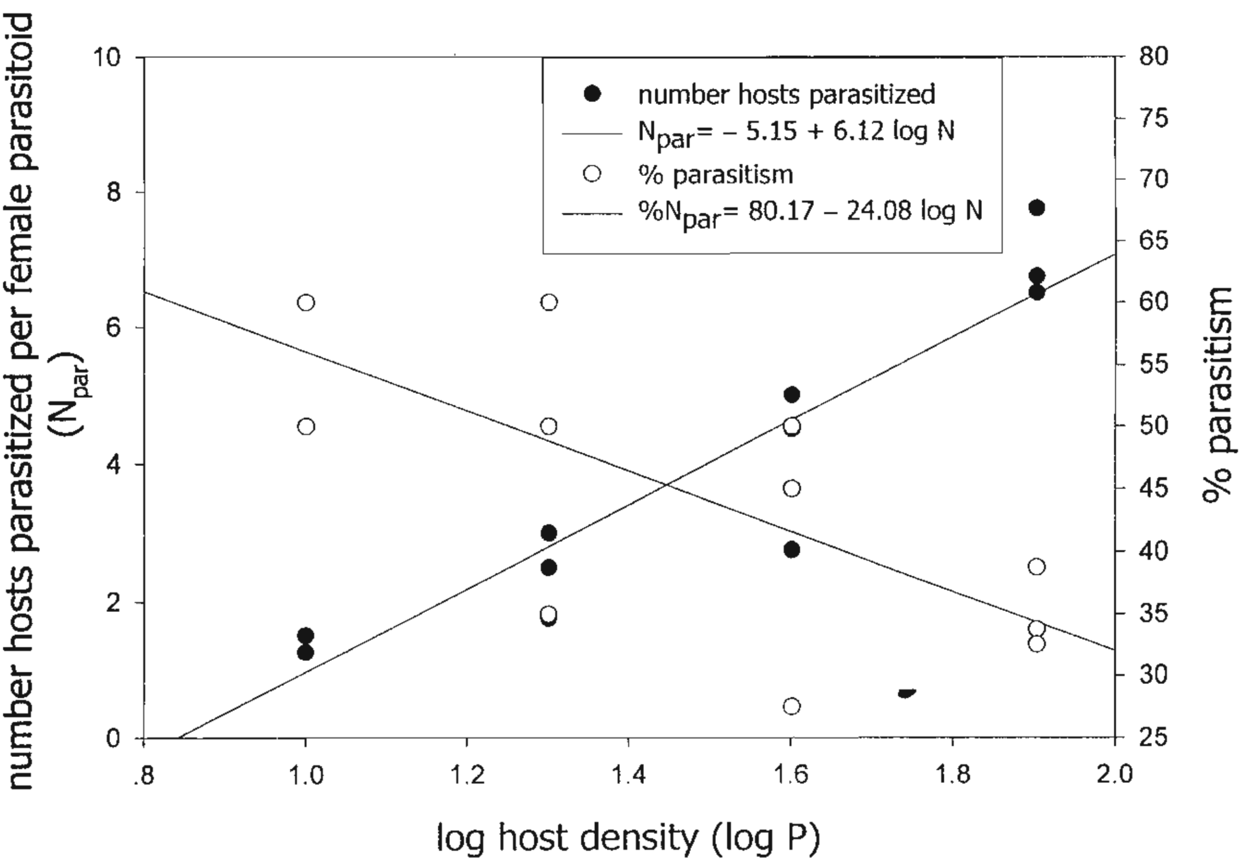


number hosts parasitized per female parasitoid (N_{par})
number hosts parasitized
N_{par} = – 8.49 + 9.88 log N
% parasitism
$\%N_{par}$ = 107.44 – 26.44 log N
log host density (log P)
% parasitism
0
2
4
6
8
10
12
14
30
40
50
60
70
80
90
100
.8
1.0
1.2
1.4
1.6
1.8
2.0



number hosts parasitized
N_par= 11.25 – 8.26 log P
% parasitism
%N_par= 31.99 + 49.87 log P
number hosts parasitized per female parasitoid (N_par)
% parasitism
log parasitoid density (log P)
20
18
16
14
12
10
8
6
4
2
100
80
60
40
20
0
0.0
.2
.4
.6
.8
1.0

Fig. 2



number hosts parasitized
Npar= – 5.15 + 6.12 log N
% parasitism
%Npar= 80.17 – 24.08 log N
number hosts parasitized per female parasitoid (Npar)
% parasitism
log host density (log P)
10
8
6
4
2
0
80
75
70
65
60
55
50
45
40
35
30
25
.8
1.0
1.2
1.4
1.6
1.8
2.0

Fig. 1b

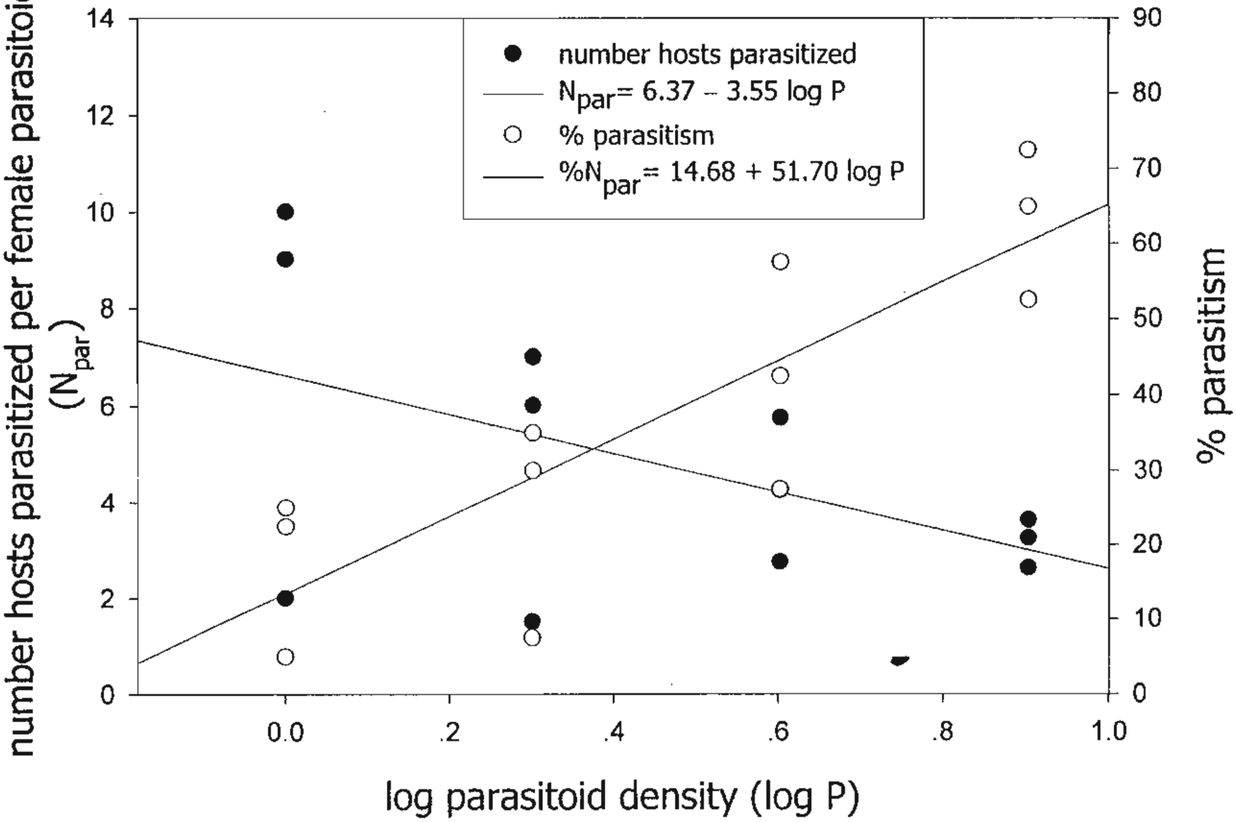

number hosts parasitized
N_{par}= 6.37 – 3.55 log P
% parasitism
$\%N_{par}$= 14.68 + 51.70 log P
number hosts parasitized per female parasitoid (N_{par})
% parasitism
log parasitoid density (log P)
0
2
4
6
8
10
12
14
0
10
20
30
40
50
60
70
80
90
0.0
.2
.4
.6
.8
1.0

Fig 2b

Laboratory evaluation in density relationships of the parasitoid, *Spalangia endius* (Hymenoptera: Pteromalidae), with two species of tephritid fruit fly pupal hosts in Thailand

Sangvorn Kitthawee*, Kamolwan Sriplang, Warren Y. Brockelman and Visut Baimai

Current address: Department of Biology, Faculty of Science, Mahidol University, Rama VI Road, Bangkok 10400, Thailand

*Corresponding author: Sangvorn Kitthawee, Department of Biology, Faculty of Science, Mahidol University, Rama VI Road, Bangkok 10400, Thailand. Tel: + 662 201 5276; Fax: + 662 274 0079.

e-mail: grskt@mahidol.ac.th

Presenter
Miss Kamolwan Sriplang
Biology Department, Faculty of Science, Mahidol University

Density relationships of the parasitoid, *Spalangia endius* (Hymenoptera: Pteromalidae) with their fruit fly hosts

Kamolwan Sriplang , Sangvorn Kitthawee[1]

One of the biological methods for controlling fruit flies is by using parasitoids. For successful control, an understanding of the relationships between fruit fly hosts and their parasitoids is necessary. In this experiment, *Spalangia endius* is used as a parasitoid for attacking pupae of the fruit flies, *Bactrocera dorsalis* and *Bactrocera correcta,* to study the effect of their density.

This study was conducted in the laboratory and consisted of two experiments. Firstly, *S. endius* parasitoids at a constant density were allowed to search for 1 day for puparia held at different densities of fruit fly hosts. Secondly, parasitoids at different densities were allowed to search for fruit fly hosts held at constant density.

The results showed that at constant parasitoid density, an increase in number of hosts result in an increase in the number of parasitized hosts but the percentage of parasitized hosts declines at high host density. These results suggest that the mortality factor is inversely density dependent. Further, searching at constant host density tends to increase as parasitoid density is reduced. These results suggest that host-parasitoid densities are an important consideration in biological control program that uses parasitoids.

ผู้อธิบายโครงงานวิจัย
นางสาว กมลวรรณ ศรีปลั่ง
ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

ความสัมพันธ์ทางความหนาแน่นระหว่างแมลงวันผลไม้และแตนเบียน *Spalangia endius* (Hymenoptera:Pteromalidae)

กมลวรรณ ศรีปลั่ง และ สังวรณ์ กิจทวี[1]

แตนเบียน(parasitoid)เป็นศัตรูธรรมชาติชนิดหนึ่งสามารถใช้กำจัดแมลงวันผลไม้ได้ การควบคุมแมลงวันผลไม้ให้มีประสิทธิภาพจำเป็นต้องเข้าใจถึงความสัมพันธ์ระหว่างความหนาแน่นของแมลงวันผลไม้และแตนเบียน ในการทดลองนี้ใช้แตนเบียนชนิด *Spalangia endius* โจมตีแมลงวันผลไม้ชนิด *Bactrocera dorsalis* และ *Bactrocera correcta* ในระยะดักแด้ ศึกษาผลการเปลี่ยนแปลงความหนาแน่นของ *S. endius* และความหนาแน่นของดักแด้แมลงวันผลไม้ เพื่อนำไปวิเคราะห์และประเมินหาจำนวน *S. endius* ที่เหมาะสมต่อการควบคุมแมลงวันผลไม้

การทดลองนี้ได้ศึกษาในห้องปฏิบัติการ แบ่งเป็น 2 ชุดการทดลอง การทดลองแรก ศึกษาการเปลี่ยนแปลงความหนาแน่นของ *S. endius* การทดลองที่ 2 ศึกษาการเปลี่ยนแปลงความหนาแน่นของดักแด้แมลงวันผลไม้ โดยให้เวลาในการโจมตี 1 วัน

พบว่าเมื่อจำนวนดักแด้แมลงวันผลไม้เพิ่มขึ้น จำนวนดักแด้แมลงวันผลไม้ที่ถูกโจมตีจะเพิ่มขึ้น แต่เปอร์เซ็นต์ของดักแด้แมลงวันผลไม้ที่ถูกโจมตีเมื่อเทียบกับความหนาแน่นของดักแด้แมลงวันผลไม้ที่เพิ่มขึ้นกลับลดลง เมื่อจำนวนดักแด้แมลงวันผลไม้คงที่และเพิ่มจำนวน *S. endius* พบว่าความสามารถในการหาดักแด้แมลงวันผลไม้ของ *S. endius* ลดลง จากผลการทดลองนี้บอกถึงความสัมพันธ์ระหว่างความหนาแน่นของแมลงวันผลไม้กับแตนเบียน และความสามารถในการค้นหาแมลงวันผลไม้ของแตนเบียนด้วย ซึ่งสามารถนำไปใช้ในการวางแผนควบคุมการจำกัดแมลงวันผลไม้โดยชีววิธีได้

---

[1]อาจารย์ที่ปรึกษา : รองศาสตราจารย์ ดร.สังวรณ์ กิจทวี ภาควิชาชีววิทยา
คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

4005170SCBI : MAJOR BIOLOGY

KEY WORDS : *SPALANGIA ENDIUS, BACTROCERA CORRECTA, DIACHASMIMORPH LONGICAUDATA*, HYPERPARASITOID, PARASITISM

WANTHIP YIMLAMAI : SUITABILITY OF A FRUIT FLY AND ITS PARASITOID AS HOSTS OF THE HYPERPARASITOID, *SPALANGIA ENDIUS* (HYMENOPTERA : PTEROMALIDAE)

PROJECT ADVISOR : ASSOCIATE PROFESSOR
SANGVORN KITTHAWEE, PH.D

Spalangia endius is a pteromalid pupal parasitoid which is found in various dipteran hosts. From a former study, *S. endius* was found to act as a primary parasitoid of fruit flies and as a hyperparasitoid of a braconid parasitoid of the same fruit flies.

This experiment was conducted in the laboratory to first determine the most suitable age of S. endius female to reproduce. Then, the ability of *S. endius* to attack fruit flies, especially *Bactrocera correcta*, was compared with its ability to attack the fruit fly parasitoid, *Diachasmimorpha longicaudata*.

The results from this study showed that three day old *S. endius* are most suited for parasitism and also showed biostatistic analysis with 95% confidence by Two sample T -test ($P=0.0001$)which *S. endius* could attack *B. correcta* at a greater rate than *D. longicaudata*. These results indicate that *S. endius* prefers to attack the fruit fly host. This study suggests that *S. endius* is a useful insect as a potential biological control agent against *B. correcta*

4005170SCBI : สาขาวิชาเอก : ชีววิทยา

คำสำคัญ : *Spalangia endius, Bactrocera correcta, Diachasmimorpha longicaudata,* hyperparasitoid, parasitism

วันทิพย์ ยิ้มละมัย : Suitability of a fruit fly and its parasitoid as hosts of the hyperparasitoid, *Spalangia endius* (Hymenoptera : Pteromalidae)

อาจารย์ที่ปรึกษา : รองศาสตราจารย์ ดร. สังวรณ์ กิจทวี

Spalangia endius (Hymenoptera : Pteromalidae) เป็นแตนเบียน(parasitoid)ของแมลงวัน(fly)ในอันดับDipteraในระยะดักแด้ มีข้อมูลที่บ่งชี้ว่า *S. endius* เป็น hyperparasitoid สามารถทำลายแตนเบียนของแมลงวันผลไม้ (fruit fly parasitoid)ได้

การทดลองนี้จึงทำการเปรียบเทียบความสามารถของ *S. endius* ต่อการทำลายแมลงวันผลไม้ (ชนิด*Bactrocera correcta)*กับแตนเบียนของแมลงวันผลไม้(ชนิด*Diachasmimorpha longicaudata)* เพื่อเป็นการยืนยันข้อบ่งชี้ดังกล่าว จึงต้องทำการตรวจสอบอายุของ *S. endius* ที่ให้ประสิทธิภาพการทำลายสูงสุดด้วย

ผลการทดลองพบว่า*S. endius*อายุ3วันมีประสิทธิภาพการทำลายสูงสุดและเมื่อวิเคราะห์ทางชีวสถิติด้วยTwo Sample T- test ก็พบว่า *S. endius* สามารถโจมตีระยะดักแด้ของแมลงวันผลไม้ได้มากกว่าโจมตีระยะดักแด้ของแตนเบียนชนิด *D. longicaudata*โดยมีค่าP=0.0001 ที่ระดับความเชื่อมั่น95% ผลการทดลองนี้ชี้ให้เห็นว่า *S. endius* ชอบโจมตีแมลงวันผลไม้ชนิด *B. correcta* มากกว่าแตนเบียนชนิด *D. longicaudata* ข้อมูลนี้ยังแสดงว่า *S. endius*เป็นแมลงที่เป็นประโยชน์สามารถนำมาใช้ในการควบคุมและกำจัดแมลงวันผลไม้ได้ซึ่งอาจใช้เป็นทางเลือกที่ดีอีกทางหนึ่ง

Presenter
Miss Siriya Seneewongse Na Ayudthaya
Biology Department, Faculty of Science, Mahidol University

Fecundity of virgin females and mated females of the parasitoid *Diachasmimorpha longicaudata*

Siriya Seneewongse Na Ayudthaya, Sangvorn Kitthawee[1] and Sukathida Ubol[2]

Diachasmimorpha longicaudata (Hymenoptera: Braconidae) is a larval endoparasitoid that is used for biological control of *Bactrocera correcta* (Diptera: Tephritidae). *Diachasmimorpha longicaudata* females lay their eggs in larvae of *B. correcta*. Larvae of *D. longicaudata* develop inside the host and cause death of the host.

In this study, a comparison of fecundity was made between virgin females and mated females. *Diachasmimorpha longicaudata* were permitted to lay their eggs in larvae of *B. correcta*. Then the numbers of *D. longicaudata* that emerged were determined. DNA Isolation and Electrophoresis techniques were used to compare DNA between haploid males and diploid females.

The results showed that females (2n=diploid) arise from fertilized eggs, whereas males (n=haploid) develop from unfertilized eggs of virgin females and mated females. During their life span, virgin females lay their eggs in 2 cycles, whereas mated females lay their eggs in 3 cycles. Haploid males and diploid females have the same size of DNA but diploid females have a greater quantity of DNA than haploid males.

References :

1. Ramadan MM, Wong TTY and Beardsley JW. (1989) Survivorship, potential, and realized fecundity of *Biosteres tryoni* (Hymenoptera: Braconidae), a larval parasitoid of *Ceratitus capitata* (Diptera: Tephritidae). Entomophaga 34: 291-297.
2. Ramadan MM, Wong TTY and Beardsley JW. (1989) Insectary production of *Biosteres tryoni* [Cameron] (Hymenoptera: Braconidae), a larval parasitoid of *Ceratitus capitata* [Wiedemann] (Diptera: Tephritidae). Proc Hawaii Entomol Soc 29: 41-48.

INDEX KEY WORD : *Diachasmimorpha longicaudata, Bactrocera correcta,* fecundity, DNA Isolation, haploid male

ผู้อธิบายโครงงานวิจัย
นางสาวสิริยา เสนีวงศ์ ณ อยุธยา
ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

ความสามารถในการผลิตลูกหลานของเพศเมียที่ไม่ได้รับการผสมพันธุ์และเพศเมียที่ได้รับการผสมพันธุ์ของ *Diachasmimorpha longicaudata*

สิริยา เสนีวงศ์ ณ อยุธยา สังวรณ์ กิจทวี[1] และ ศุขธิดา อุบล[2]

Diachasmimorpha longicaudata (Hymenoptera: Braconidae) เป็นแตนเบียน (parasitoid) ที่ใช้ในการควบคุมแมลงวันผลไม้ (fruit flies) ชนิด *Bactrocera correcta* (Diptera: Tephritidae)โดย *D. longicaudata* จะวางไข่ในตัวอ่อน (larvae) ของ *B. correcta* ไข่ของ *D. longicaudata* จะพัฒนาอยู่ภายในตัวอ่อนของ *B. correcta* พร้อมกับทำลาย *B. correcta* ไปด้วย

ในการศึกษาเปรียบเทียบระบบสืบพันธุ์และความสามารถในการผลิตลูกหลานระหว่าง *D. longicaudata* เพศเมียที่ได้รับการผสมพันธุ์และที่ไม่ได้รับการผสมพันธุ์ ทำการศึกษาโดยให้ *D. longicaudata* เพศเมียวางไข่ในตัวอ่อน (larvae) ของ *B. correcta* เมื่อไข่พัฒนาเป็นตัวเต็มวัยแล้วจึงนับจำนวน *D. longicaudata* ที่เกิด ส่วนการศึกษาเปรียบเทียบ DNA ของเพศผู้ (haploid) และเพศเมีย (diploid)ใช้เทคนิค DNA Isolation & Electrophoresis

จากผลการทดลองพบว่า *D. longicaudata* เพศเมียที่ไม่ได้รับการผสมพันธุ์จะผลิตลูก (progeny) ออกมาเป็นเพศผู้ (haploid) ทั้งหมด โดยสามารถผลิตลูกได้มากถึง 2 ช่วง ส่วนเพศเมียที่ได้รับการผสมพันธุ์จะผลิตลูกได้ทั้งเพศผู้ (haploid?) และเพศเมีย (diploid) โดยสามารถผลิตลูกได้มากถึง 3 ช่วง และพบว่า DNA ของทั้ง 2 เพศ มีขนาดเท่ากันแต่ในเพศเมียจะมีปริมาณมากกว่าเพศผู้

---

[1]อาจารย์ที่ปรึกษา : รองศาสตราจารย์ดร. สังวรณ์ กิจทวี ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

[2]อาจารย์ที่ปรึกษาร่วม : ดร. ศุขธิดา อุบล ภาควิชาจุลชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

Mr. Wanwiwat Lovichit
Biology Department, Faculty of Science, Mahidol University

Morphometric Analysis of Selected Strains in the *Diachasmimorpha longicaudata* complex (Hymenoptera: Braconidae) by Using Statistical Analysis and Computer Techniques

Wanwiwat Lovichit, Suang Udomvaraphunt [1] and Sangvorn Kitthawee[2]

Morphometric analysis was conducted in order to distinguish members of the *Diachasmimorpha longicaudata* complex, which were fruit fly parasitoids. This species has been widely employed as a biological control agent. Four strains from this species complex and *Psytalia incisi,* another species of parasitoid, were used in this study based on their wing patterns.

The analysis consisted mainly of two parts. The first one was to separate strains within the complex (strains A, B, C and D). The second was to segregate among strains A, B and *P. incisi*. Seventy characters were used and they were derived from vein lengths, cell perimeters and cell areas.

Stepwise discriminant analysis showed that 19 wing characters were selected by the model, indicated by highly significant differences ($P<0.01$) among strains, *D. longicaudata* A, B, C and D. Also, 15 wing characters showed highly significant differences ($P<0.01$) among *D. longicaudata* A, B and *P. incisi*. A total of 150 specimens were assigned to species by cross-validation and this method was shown to be accurate about 100%, 100%, 93.3%, 90.0% of specimens for each strain (*D. longicaudata* A, B, C and D respectively) and 100%, 100% 100% of *D. longicaudata* A, B and *P. incisi* respectively. Therefore, this method is beneficial, and can be applied to discriminating species in other complexes, which have distinctive wings or characters. Moreover, the method is not only convenient and inexpensive but suitable for developing countries as well.

INDEX KEY WORDS: *Diachasmimorpha longicaudata*, morphometric analysis, stepwise discriminant analysis

นายวันวิวัฒน์ หล่อวิจิตร
ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

การวิเคราะห์ทางสัณฐานวิทยาของแมลงเบียนกลุ่มซับซ้อน โดยใช้วิธีการทางสถิติและคอมพิวเตอร์

วันวิวัฒน์ หล่อวิจิตร สรวง อุดมวรภัณฑ์ [1] สังวรณ์ กิจทวี [2]

การวิเคราะห์ทางสัณฐานวิทยา (Morphometric Analysis) ได้ถูกนำมาใช้ในการจำแนกชนิดแมลงเบียน ซึ่ง เป็นแมลงที่ใช้กันมากในการควบคุมแมลงวันผลไม้ (fruit flies) โดยที่นำแมลงเบียนมาใช้ในการทดลองทั้งหมด 2 กลุ่มคือ แมลงเบียนกลุ่มซับซ้อน (Diachasmimorpha *longicaudata* complex) ซึ่งกลุ่มนี้มีทั้งหมด 4 สายพันธุ์ (strains) และ *Psytalia incisi* ซึ่งเป็นแตนเบียน (parasitoid) อีกชนิดหนึ่งถูกนำมาใช้ในการทดลอง

โดยจะทำการทดลองแบ่งออกเป็น 2 ลักษณะ การทดลองแรกจะทำการจำแนกภายในกลุ่มแมลงเบียนซับซ้อน (strains A,B,C,D) การทดลองต่อมาจำแนกระหว่าง strains A,B และ *P. incisi* ลักษณะของปีกทั้งหมด 70 ลักษณะ ซึ่งมาจาก ความยาว (vein length) เส้นรอบวง (cell perimeter) และพื้นที่ (cell area) ของเส้นปีก จากจำนวนตัวอย่างทั้งหมด 150 ตัวอย่าง จะถูกการจำแนกด้วยการวิเคราะห์ทางสถิติ คือ การวิเคราะห์จำแนกประเภท (Stepwise Discriminant Analysis)

พบว่าการวิเคราะห์ทางสัณฐานวิทยานั้นจะได้สมการการจำแนกประเภทที่มีความแม่นยำ คือ 100%, 100%, 93.3%, 90.0% ในกลุ่มของ strains A,B,C และ D และ 100%, 100%,100% ในกลุ่มของ strain A,B และ *P. incisi* ดังนั้นวิธีการนี้จะสามารถนำไปเป็นอีกทางเลือกหนึ่งและประยุกต์ใช้กับการจำแนกแมลงชนิดอื่นที่มีลักษณะของเส้นปีกที่ชัดเจนหรือลักษณะอื่นๆได้ นอกจากนั้นความสะดวกเร็วและค่าใช้จ่ายที่ไม่สูง จึงเหมาะอย่างยิ่งสำหรับประเทศที่กำลังพัฒนาและมีความหลากหลายชีวภาพสูงอย่างประเทศไทย

---

[1] อาจารย์ที่ปรึกษา: อาจารย์ สรวง อุดมวรภัณฑ์ สถาบันวิจัยและพัฒนาวิทยาศาสตร์และเทคโนโลยี มหาวิทยาลัยมหิดล

[2] อาจารย์ที่ปรึกษา: รองศาสตราจารย์ ดร.สังวรณ์ กิจทวี ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

Presenter
Mr. Thatri Iampornsin
Department of Biology, Faculty of Science, Mahidol University

---

Biology of the fruit fly parasitoid, *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae), for the control of fruit flies

Thatri Iampornsin and Sangvorn Kitthawee[1]

Fruit flies are serious pests of various commercially important fruits. Current interest in fruit fly parasitoids as biological control agents is increasing. *Diachasmimorpha longicaudata* (Ashmead) is a larval parasitoid that parasitizes the fruit fly, *Bactrocera correcta*. It kills fruit fly hosts by the act of ovipositing into the larval hosts and developing to adults. Adult parasitoids emerge from the fruit fly puparia. This parasitoid is considered to be a beneficial wasp and useful in biological control programs. However, its biology needs to be investigated to optimize its biocontrol potential.

In this experiment, 10 naive pairs of *Diachasmimorpha longicaudata* were allowed to parasitize fruit fly larvae of *Bactrocera correcta* in the banana every day. Numbers of parasitoid's progenies were recorded when the larvae developed to adult. During the experiment, the deaths of 10 pairs of parasitoids were recorded and collected to dissect wings to evaluate the relationship between body size and survivorship.

The results demonstrated that female parasitoids layed a maximum mean number of eggs at age 6 days. Female parasitoids that were allowed to lay eggs had a fewer survivorship than females not allowed to lay eggs. There was no relationship between body size and survivorship ($P>0.05$) in both male and female parasitoids. These biological studies will give basic knowledge for biological control in the future.

References:

1. Messing, R.H., and Jang, E.B. 1992. Response of the fruit fly parasitoid *Diachasmimorpha longicaudata* (Hymenoptera: Braconidae) to host-fruit stimuli. Environ. Entomol. 21:1189-95.
2. Montoya, P., Liedo, P., Benrey, B., Cancino, J., Barrera, J.F., Sivinski, J., and Aluja, M. 2000. Biological control of *Anastrepha* spp. (Diptera: Tephritidae) in mango orchards through augmentative releases of *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae). Biol. Control 18:216-24.
3. Ramadan, M.M., Wong, T.T.Y., and Beardsley, J.W. 1989. Survivorship, potential, and realized fecundity of *Biosteres tryoni* (Hymenoptera: Braconidae), a larval parasitoid of *Ceratitis capitata* (Diptera: Tephritidae). Entomophaga 34:291-7.

INDEX KEY WORDS: ***Diachasmimorpha longicaudata*, *Bactrocera correcta*, biological control**

ผู้อธิบายโครงงานวิจัย
นาย ธาตรี เอี่ยมพรสิน
ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล

การศึกษาชีววิทยาของแตนเบียนชนิด *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae) เพื่อการควบคุมแมลงวันผลไม้

ธาตรี เอี่ยมพรสิน และ สังวรณ์ กิจทวี [1]

แมลงวันผลไม้ เป็นศัตรูสำคัญที่ทำให้เกิดความเสียหายกับผลผลิตทางการเกษตร การควบคุมโดยชีววิธีเป็นอีกวิธีการหนึ่งที่ได้มีการศึกษามากขึ้นในปัจจุบัน แตนเบียนชนิด *Diachasmimorpha longicaudata* เป็นศัตรูทางธรรมชาติ (natural enemies) ที่สามารถเข้าทำลายแมลงวันผลไม้ได้ โดยการโจมตีตัวหนอนแมลงวันผลไม้เพื่อวางไข่ ตัวอ่อนของแตนเบียนจะเจริญเติบโตอยู่ภายในพร้อมกับค่อยๆ ทำลายตัวหนอนแมลงวันผลไม้ แล้วพัฒนาออกมาเป็นตัวเต็มวัยแทน ดังนั้น จึงเป็นที่น่าสนใจในการศึกษาเพื่อนำมาใช้ควบคุมแมลงวันผลไม้

การทดลอง ให้แตนเบียน *Diachasmimorpha longicaudata* จำนวน 10 คู่ โจมตีตัวหนอนแมลงวันผลไม้ *Bactrocera correcta* ในกล้วยทุกๆวัน เมื่อตัวหนอนเจริญเป็นตัวเต็มวัย ทำการนับจำนวนแมลงที่เกิดทั้งหมด ในระหว่างทดลอง บันทึกจำนวนการตายของแตนเบียนทั้ง 10 คู่ และวัดความยาวปีกเพื่อศึกษาถึงความสัมพันธ์ระหว่างขนาดตัวกับอัตราการอยู่รอด

ผลการทดลองพบว่า แตนเบียนจะมีช่วงการวางไข่มากที่สุดเมื่อมีอายุได้ 6 วัน แตนเบียนเพศเมียที่ให้วางไข่มีอัตราการอยู่รอดน้อยกว่าแตนเบียนที่ไม่ได้ให้วางไข่ และไม่พบความสัมพันธ์ของขนาดตัวกับอัตราการอยู่รอด ($P>0.05$) ทั้งในแตนเบียนเพศผู้และเพศเมีย การศึกษาชีววิทยาของแตนเบียนนี้จะเป็นข้อมูลที่สามารถนำไปพัฒนาวิธีการควบคุมแมลงวันผลไม้ให้มีประสิทธิภาพต่อไป

---

[1] อาจารย์ที่ปรึกษา: รองศาสตราจารย์ ดร.สังวรณ์ กิจทวี ภาควิชาชีววิทยา คณะวิทยาศาสตร์ มหาวิทยาลัยมหิดล