

Development time of *Urolepis rufipes* (Hymenoptera: Pteromalidae) and effect of female density on offspring sex ratio and reproductive output

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ABSTRACT

Two experiments studying i) offspring developmental time and ii) female density-effects on offspring sex ratio and female reproductive output were conducted on a Manitoba population of *Urolepis rufipes*, a solitary pupal parasitoid of flies, including *Musca domestica* and *Stomoxys calcitrans* (Diptera: Muscidae). Mean development time (\pm SEM) was 12.90 ± 0.03 d for males and 14.43 ± 0.03 d for females at 23°C. A significant correlation ($P = 0.005$) was observed between offspring sex ratio and log female density as the number of females in the arena increased from 1 to 6, although the data were highly variable ($r^2 = 0.18$). *Urolepis rufipes* offspring sex ratios were male-biased when 4 or 6 females were present within the arena. Number of emerging offspring per female was not significantly ($P > 0.05$) affected by female density. Although the

development time of *U. rufipes* offspring is short relative to other pteromalid species, offspring sex ratios become male-biased at even modest female densities, perhaps making mass rearing of *U. rufipes* inefficient for inundative biological control.

INTRODUCTION

Urolepis rufipes (Ashmead) (Hymenoptera: Pteromalidae) is distributed throughout much of the United States and Canada, including Manitoba, and parts of Denmark and Germany, mainly as a solitary pupal parasitoid of flies, including *Musca domestica* L. and *Stomoxys calcitrans* (L.) (Diptera: Muscidae) (McKay and Galloway 1999; Gibson 2000; Gibson and Floate 2004). It has been suggested that *U. rufipes* may be an effective biocontrol agent of filth flies in northern regions because of its short developmental time relative to other pteromalids (Smith and Rutz 1986, Matthews and Petersen 1989). Rearing conditions balancing efficiency costs and productivity are required to mass-rear parasitoids economically. A short development time would allow for rapid turnover of parasitoid generations, possibly reducing rearing costs associated with incubating host puparia and potentially allowing for subsequent generations of the parasitoid to reduce the pest population. In addition, a large reproductive output per ovipositing female and a female-biased offspring sex ratio are desired for parasitoid release (Heinz 1998). Female density has a significant effect on reproductive decisions made by females of other pteromalid species (Werren 1983; King 1989; King and Seidl 1993), but the importance of *U. rufipes* female density in determining offspring sex ratio and reproductive output is not known. We conducted a study to evaluate two aspects of parasitoid reproduction, offspring developmental time and female density-effects on offspring sex ratio and female reproductive output, in a Manitoba population of *U. rufipes* in the context of evaluating it as a potential inundative biocontrol agent.

MATERIALS AND METHODS

The developmental time in the Manitoba population was measured using a laboratory colony initially started from *U. rufipes* collected from 7 June, 1998 to 2 August, 1998 at the Glenlea Research Station, Faculty of Agricultural and Food Sciences (Glenlea, MB). Each of 255 females was offered 50 house fly pupae in a 10 cm x 10 cm x 3 cm, covered, transparent dish for 24 h at 23°C (L18:D6). Up to six females were allowed to oviposit at any one time within each arena. Pupae were then placed in 96-well tissue culture plates (one pupa/well), incubated at 23°C (L18:D6), and monitored daily for parasitoid emergence. Number of days from oviposition to emergence was recorded as development time. Development time data from the subsequent experiment was consistent with those from this experiment and was included in the analysis.

An experiment was conducted to observe *U. rufipes* oviposition behaviour under conditions of changing female density. The experiment was conducted over a period of four consecutive days. Females emerging on each day were randomly assigned to one of the four treatments. One 2-3 day-old male was mated to 1, 2, 4, or 6 newly emerged

(<24 h) females in a 10 cm x 10 cm x 3 cm, covered, transparent dish. Mating was assumed to occur after a male mounted a female for ≥ 10 s. Males were then removed and fifty 2-3 day-old house fly pupae were added per female, resulting in the host: parasitoid ratio being held constant across the different treatments, and the oviposition arena was incubated for 24 h at 23°C (L18:D6). After 24 h, pupae were treated as described above. Sex and number of emerging adults were recorded. Each treatment was replicated 10 or 11 times.

RESULTS AND DISCUSSION

Males developed significantly faster than females ($t_{2734} = 36.3$; $P < 0.001$). Mean development time (\pm SEM) was 12.90 ± 0.03 d for males ($n = 1605$ males, range: 11-21 days) and 14.43 ± 0.03 d for females ($n = 1131$ females, range: 12-19 days). This is shorter than the development time of males and females from Denmark (21.7 and 24.1 d, respectively, at 20°C and 14.2 and 15.5 d, respectively, at 25°C; Stenseng *et al.* 2003) and slightly longer than that for males and females from Nebraska (12.3 and 13.5 d, respectively, at 20°C; Smith and Rutz 1986), suggesting that development time varies among populations of *U. rufipes*. The development time observed here is comparable to that observed for *Nasonia vitripennis* (Walker) and shorter than that observed for other pteromalid species (Rueda and Axtell 1985; Lysyk 2001).

Urolepis rufipes offspring sex ratios were highly variable; however, a positive correlation was observed between offspring sex ratio ($\sin^{-1} \sqrt{\text{proportion male}}$) and log female density (Fig. 1). Offspring sex ratios were significantly male-biased when 4 or 6 females were present within the arena; backtransformed mean sex ratios (95% confidence interval) for the 1, 2, 4, and 6 female treatments were 0.39 (0.21 - 0.58), 0.60 (0.43 - 0.76), 0.68 (0.60 - 0.75), and 0.64 (0.58 - 0.70), respectively. As one male was used to mate all females in an oviposition arena, it is possible that males became sperm-depleted and some females were constrained by the amount of sperm received. However, if this were the case, offspring sex ratios should continue to increase as the number of constrained females in the oviposition arena increased. Instead, offspring sex ratio did not differ among the three highest female density treatments ($P > 0.05$, Tukey-Kramer HSD), indicating that the increase in offspring sex ratio was not due to sperm depletion. No significant difference in the mean number of offspring per female (following square root transformation) was observed among the different treatments ($F_{3,39} = 0.63$; $P = 0.6$). Mean numbers of offspring per female were 8.9 (4.9 - 14.1), 10.2 (6.6 - 14.6), 12.0 (8.6 - 16.1), and 12.7 (8.9 - 17.2) for the 1, 2, 4, and 6 female treatments, respectively. No more than one parasitoid emerged from a single host.

During oviposition at low host density, increasing host density can affect offspring sex ratio and/or number of offspring (King *et al.* 1995, Kumar *et al.* 2000, Sagarra *et al.* 2000). When host density is maintained at a constant level, the presence of additional female parasitoids in an experimental arena results in a reduction in the number of hosts present per female, which could also influence parasitoid behaviour. For this study, the host:parasitoid ratio was held constant to isolate the effect on parasitoid behaviour of increasing the number of females present in the arena. Since no effect of host density

was observed for any of the measured behaviours in the above studies at the host: parasitoid ratio (50:1), this ratio was chosen for the current study, in which none of the treatments resulted in 100 % parasitism as adult house fly emergence was observed in all replicates. This confirms that the number of hosts present in the arenas was not limiting and the results observed here were likely due to changing female density and not the confounding variable, changing host density.

Urolepis rufipes has attributes beneficial for its potential use as a biological control agent of muscid flies in Manitoba and possibly elsewhere in Canada and the northern United States, including its rapid rate of development and its tolerance to being cultured in laboratory colonies. However, *U. rufipes* offspring sex ratios become male-biased at even modest female densities, perhaps making mass rearing of *U. rufipes* inefficient.

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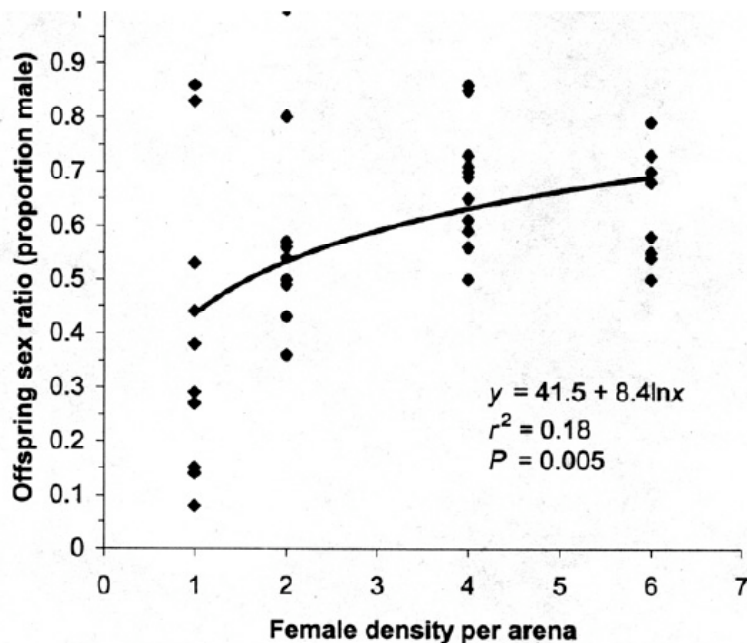


Figure 1. Logarithmic regression of *Urolepis rufipes* offspring sex ratio by the density of mated females in oviposition arenas. One, 2, 4, or 6 mated females were confined to an oviposition arena containing fifty *Musca domestica* pupae per female and allowed to oviposit for 24 h.