

INFLUENCE OF LIQUID FOOD ON FECUNDITY AND LONGEVITY OF *MICROCTONUS HYPERODAE* LOAN

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ABSTRACT

Microctonus hyperodae Loan (Hymenoptera: Braconidae) is an introduced parasitoid of the pasture pest Argentine stem weevil, *Listronotus bonariensis* (Kuschel) (Coleoptera: Curculionidae). This paper describes the effect of providing honey solution to caged *M. hyperodae* females derived from three South American geographic populations (ecotypes) on their fecundity and longevity. Both the longevity and fecundity of parasitoids from all three ecotypes was increased when they had access to honey. There was no significant difference between ecotypes in either overall fecundity or longevity. There was evidence of an interaction between the honey and ecotype treatments in the fecundity data. The fecundity of females from the Concepcion ecotype (Chile) tended to be lower than that of the Mendoza (Argentina) and Colonia ecotypes when honey was not available, while all ecotypes had similar fecundities when they had access to honey.

Keywords: *Listronotus bonariensis*, *Microctonus hyperodae*, longevity, fecundity, biological control.

INTRODUCTION

Argentine stem weevil, *Listronotus bonariensis* (Kuschel) (Coleoptera: Curculionidae) is a major pasture pest in New Zealand (Prestidge *et al.* 1991). A parthenogenetic parasitoid, *Microctonus hyperodae* Loan (Hymenoptera: Braconidae), has been established in this country as a biological control agent of *L. bonariensis* (Goldson *et al.* 1994). The *M. hyperodae* adult oviposits in an adult *L. bonariensis*, whereupon a solitary parasitoid larva develops within the living host until maturity. When the mature larva emerges to pupate, the host dies. Parasitoids released in New Zealand were derived from eight diverse South American locations in Argentina (Ascasubi, Mendoza, General Roca and S. C. de Bariloche), Brazil (Porto Alegre), Chile (Concepcion and La Serena) and Uruguay (Colonia) (Goldson *et al.* 1990). Approximately equal numbers of *M. hyperodae* derived from each South American geographic population, termed 'ecotypes', were released to ensure the widest possible range of genetic material was liberated at each site (Goldson *et al.* 1990). Since the species' importation in 1989-90, all ecotypes have been maintained in culture to provide genetic stock for further field releases and to supply parasitoids of known ecotype for research purposes (Goldson *et al.* 1994).

Liquid food sources such as floral and extra-floral nectar, trichome exudates and homopteran honey dew often influence the fecundity and longevity of parasitic wasps (e.g. Quicke 1997). The presence of such food has been shown to influence rates of parasitism in the field (e.g. Baggen and Curr 1998). Hodgson *et al.* (1993) found that the longevity of caged *M. hyperodae* females was increased approximately threefold by provision of glucose solution, but not by provision of pollen. The glucose treatment of Hodgson *et al.* (1993) indicated that the adult stage of *M. hyperodae* may need liquid food to achieve optimal efficacy as a biological control agent. The laboratory experiment described in this contribution further examined the response of adult *M. hyperodae* to food by investigating the effect of honey on the fecundity and longevity of parasitoids derived from three widely separated South American geographic populations.

METHODS

Collection of *L. bonariensis*

Weevil adults were collected during November 1996 by sweeping pasture at night at the AgResearch Farms at Lincoln and Templeton, Canterbury. When *L. bonariensis* adult densities became very low at these locations due to natural mortality of the over-wintered population, additional weevils were obtained from Waikato. All weevils were maintained in cages for 2-3 weeks prior to use in the experiment to allow any naturally occurring immature parasitoids to emerge.

Source of parasitoids

M. hyperodae adults were obtained from the laboratory culture at AgResearch, Lincoln. The rearing procedure described by Goldson *et al.* (1993) allowed the specific lineage and generation number of each specimen to be known. The parasitoids were derived from Argentina (Mendoza; isofemale line 1), Chile (Concepcion; isofemale lines 2, 3, 8, 11) and Colonia (isofemale lines 21, 47).

M. hyperodae oviposition

Adult parasitoids which had emerged less than 48 hours earlier were placed for 20 minutes in 55 mm x 20 mm diameter glass vials containing filter paper (10 mm x 10 mm) soaked in either 50:50 honey:water solution, or water. Parasitoid oviposition was then measured in gauze-covered cages (220 mm x 135 mm x 75 mm). Each cage contained one parasitoid, a variable number of weevils (described below) and a bouquet of *Lolium multiflorum* (cultivar 'Tama'). Cages were held at 24°C and 12:12 L:D day length. Every 24 hours, parasitoids were removed from cages and placed in vials containing paper soaked in either honey solution or water. After 20 minutes, parasitoids were transferred to new cages containing fresh, unparasitised *L. bonariensis*. This procedure was repeated until the parasitoids died.

The number of *L. bonariensis* exposed to parasitoids each day was calculated so that it was 20-50% greater than the maximum daily oviposition of *M. hyperodae* as measured in earlier experiments (Phillips *et al.* 1996); this minimised the number of weevils that had to be dissected after the experiment. Thirty weevils were exposed to each parasitoid on the first two days of the experiment, 20 weevils on days 3-5, 15 weevils on days 6-8, 12 weevils on days 9-10, seven weevils on days 11-14, and five weevils from day 15 until the parasitoid died.

At the end of each 24 hour exposure, the *L. bonariensis* were removed from the cages, put into Petri dishes containing artificial diet and ryegrass, then placed in another controlled environment room at 20°C. After 7-10 days, the *L. bonariensis* were stored at -40°C pending dissection. Fecundity on each day of each parasitoid's life-span was estimated by dissecting all exposed weevils and counting the number of immature parasitoids.

Eggs remaining in parasitoid ovaries

M. hyperodae females were recovered as quickly as possible from the cages after they had died and the eggs remaining in their ovaries were counted before they became desiccated. Egg counts were conducted using the method of Phillips (unpublished) which involved removing the parasitoids' ovaries, staining them, separating the eggs and counting them at approximately 100 X magnification.

Statistical analyses

Egg count data were fitted to a generalised linear model with Poisson errors and compared by analysis of deviance using the Chi-square distribution for probability estimates (Green and Silverman 1994). The longevity data were compared by analysis of variance.

RESULTS

Longevity

Overall, *M. hyperodae* had a mean longevity (\pm SE) of 12 ± 1.3 days. There was no significant difference in longevity between ecotypes (means Mendoza 14 ± 4.4 , $n=3$; Concepcion 12 ± 2.4 , $n=9$; Colonia 11 ± 1.8 , $n=14$). Parasitoids that were provided with honey solution lived significantly longer than those provided with water ($P=0.011$;

means 17 ± 1.3 , $n=12$ and 5 ± 0.6 , $n=14$ respectively). There was no evidence of interaction between the ecotype and honey treatments in the longevity data.

Fecundity

Overall, *M. hyperodae* had a mean fecundity of 28 ± 0.2 eggs. There was no significant difference in overall mean fecundity between ecotypes (Mendoza 28 ± 0.7 ; Concepcion 25 ± 0.3 ; Colonia 29 ± 0.4). Honey-fed parasitoids laid significantly more eggs than *M. hyperodae* with access to water only (Table 1; $P < 0.001$). There was evidence of interaction between the ecotype and honey treatments ($P = 0.065$). The fecundity of the Mendoza and Colonia ecotypes was increased by 12% and 27% respectively by the provision of honey, while the fecundity of the Concepcion ecotype was increased by 88% (Table 1).

TABLE 1: Mean fecundity per parasitoid, mean eggs remaining in ovaries per dead parasitoid, and mean total eggs per parasitoid, for each ecotype and feeding treatment. Number of parasitoids is shown in brackets.

Ecotype	water			honey solution		
	fecundity	eggs left in ovaries	total eggs	fecundity	eggs left in ovaries	total eggs
Mendoza	26 (1)	28 (1)	54	29 (2)	9 (1)	46
Concepcion	17 (4)	23 (3)	36	32 (5)	11 (4)	45
Colonia	26 (7)	6 (1)	42	33 (7)	7 (6)	42
Overall mean	23	21	41	32	9	43

Eggs remaining in the ovaries of dead parasitoids

Significantly fewer eggs remained in the ovaries of honey-fed parasitoids than in those of water-provisioned *M. hyperodae* ($P < 0.001$; Table 1). The numbers of eggs remaining in parasitoids were negatively correlated with fecundity ($R^2 = 0.34$, $P = 0.018$). The number of eggs left in dead parasitoids did not differ according to ecotype.

Total eggs

The total egg load of each parasitoid was calculated as eggs laid in weevils (i.e. fecundity) plus eggs remaining in the ovaries after death. Total egg loads did not differ according to either ecotype or feeding treatment (Table 1).

DISCUSSION

M. hyperodae fed with honey solution lived 17 days, while those provided with water lived five days. This supports the results of earlier studies. Hodgson *et al.* (1993) found that, when *M. hyperodae* was confined without hosts at 20°C, it lived 17 days when fed with 50% glucose solution, and four days when provided with water. *M. hyperodae*, when provided with honey solution and confined with weevils at 19°C, lived 21 days (Goldson *et al.* 1995).

In this experiment, *M. hyperodae* fed with honey laid a mean of 32 eggs per parasitoid. This measurement is lower than those recorded in earlier studies which also provided *M. hyperodae* with honey solution and documented means of >40 eggs (Goldson *et al.* 1995; Phillips *et al.* 1996). Although the fecundity of the Mendoza and Concepcion ecotypes has not previously been measured, the Colonia ecotype was shown to have a fecundity >60 eggs (Goldson *et al.* 1995; Phillips *et al.* 1996), compared to the mean of 33 measured in this experiment (Table 1). The reason for this difference is not clear, but could be due to the smaller number of weevils exposed to parasitoids on most days of the experiment compared with earlier studies. This experiment's estimate for each parasitoid's total egg load was comparable with estimates of *M. hyperodae* fecundity from earlier experiments (Goldson *et al.* 1995; Phillips *et al.* 1996).

Honey-fed *M. hyperodae* parasitised 39% more weevils than those provided with water. This gain in fecundity could have occurred because: a) honey increased the rate of parasitoid oviposition; b) honey increased the number of parasitoid eggs available for oviposition; and/or c) honey increased the time available for oviposition. The results of this experiment, however, indicated that during the first five days of the experiment (when survival of both honey-fed and water-provisioned parasitoids was high), the rate of *M. hyperodae* oviposition was not influenced by the feeding treatment. Similarly, honey did not influence the total number of eggs produced by *M. hyperodae* females (Table 1); this is consistent with the observation of Goldson *et al.* (1995) that *M. hyperodae* is pro-ovigenic. The increase in fecundity associated with honey, therefore, occurred only because the greater parasitoid longevity associated with the provision of honey increased the time available for females to lay their eggs.

The 39% increase in fecundity associated with honey resulted from a 240% increase in longevity. This non-linear response of fecundity to longevity occurred because, under the conditions of relatively high host availability used in this experiment, *M. hyperodae* laid most of its eggs in the first five days before the positive effect of honey on longevity became realised. In the field, where hosts are presumably more difficult to find and the rate of parasitoid oviposition may, therefore, be lower, it seems likely that longevity would have a stronger influence on parasitoid fitness.

Although, over all ecotypes, honey did not influence the rate of oviposition during the first five days of the experiment, there was evidence of an interaction between the ecotype and honey treatments. During the first five days of the experiment, the realised fecundity of the Concepcion ecotype was higher when given honey rather than water ($P=0.015$). In contrast, parasitoids from Colonia and Mendoza tended to be more fecund when provided with water rather than honey. This possible interaction requires further work to verify it and explore its implications.

It is clear that liquid food is important in determining the longevity of *M. hyperodae* adults and that liquid food could be an important determinant of *M. hyperodae*'s fecundity and its efficacy as a biological control agent. Work is now underway to determine if *M. hyperodae* has access to food in the field and, if not, to find means of providing it.

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