**FIELD PARASITISM OF *RHINOCYLLUS CONICUS*
FROELICH (COLEOPTERA: CURCULIONIDAE) BY
MICROCTONUS AETHIOPOIDES LOAN (HYMENOPTERA:
BRACONIDAE) IN OTAGO AND SOUTH CANTERBURY**

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ABSTRACT

Microctonus aethiopoides was first recorded parasitising the weed biocontrol agent *Rhinocyllus conicus* in 1994. Subsequent studies found parasitism at several sites in Otago and South Canterbury. In this study a further 15 sites in Otago and South Canterbury were sampled to determine the extent and level of parasitism of *R. conicus* by *M. aethiopoides*. *Rhinocyllus conicus* adults were collected from nodding thistles and were either reared or dissected to determine parasitism levels. For dissected weevils the stages of parasitoid development, presence of teratocytes, weevil size and female weevil reproductive status were recorded. Parasitism was recorded at 12 sites. Lucerne, the host plant of the weevil *Sitona discoideus*, for which *M. aethiopoides* was originally introduced into New Zealand, was absent from most of these sites. Levels of parasitism were similar to those recorded in previous studies, reaching up to 17% in the Hakataramea Valley. Results relating to weevil size and female weevil reproductive state are discussed.

Keywords: Non-target host parasitism, *Rhinocyllus conicus*, *Microctonus aethiopoides*, nodding thistle.

INTRODUCTION

Under the HSNO Act 1996, ERMA is charged with determining the potential environmental impacts of new organisms before field release in New Zealand. This includes laboratory host specificity testing of foreign parasitoids to determine their host range. Studying target and non-target host parasitism by species already released in New Zealand may improve the predictive power of host-specificity tests (e.g. Barratt et al. 1997), the importance of which is now well accepted.

The parasitoid *Microctonus aethiopoides* Loan (Hymenoptera :Braconidae) was introduced in 1982 against the lucerne pest *Sitona discoideus* Gyllenhal (Coleoptera: Curculionidae) (Stufkens et al. 1987). Despite limited pre-release tests the wasp is now known to parasitise at least 17 non-target weevil species (Barratt et al. 1997; B.I.P. Barratt, unpubl. data), including the introduced nodding thistle (*Carduus nutans* L.) biocontrol agent *Rhinocyllus conicus* Froelich. Parasitism of *R. conicus* was first recorded in the Hakataramea Valley (Ferguson et al. 1994) and subsequently in the Waitaki Valley and Strath Taieri (Ferguson et al. 1998, 1999) and on Ashley Dene farm in Canterbury (M. McNeill, pers. com). The host weevil is killed when the pre-pupal parasitoid emerges but while in the host the parasitoid feeds on teratocytes. These are cells originating from the parasitoid egg (Dahlmann 1990) which absorb nutrients from the weevil haemolymph, allowing the parasitoid to feed without damaging host tissue (Okuda & Kadono-Okuda 1995). A circumstantial case of parasitism by the tachinid *Hyalomyodes triangulifer* (Loew) in Virginia is the only other record of parasitism in the adult stage of *R. conicus* (Dowd & Kok 1982). This study aimed to conduct a more intensive survey of parasitism levels, host acceptance and host response in *R. conicus* in Otago and South Canterbury.

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METHODS

Fifteen lucerne, pasture or road verge sites were sampled in eastern Otago and South Canterbury (Table 1). Adult weevils were collected in December 2001 by gathering nodding thistle flower-heads and extracting the weevils in Tullgren funnels. Up to 100 weevils per site were then held in double tiered clear plastic cages (160x180x75 mm deep) at 20°C ± 2°C with a 14:10 h light:dark photoperiod. The upper compartment housed up to 25 weevils, food (Californian thistle, *Cirsium arvense* (L.) Scop.) and moistened dental wicks, while the lower, separated by a mesh partition, was lined with paper towel providing a substrate for pupation of emergent pre-pupal parasitoids. Every three days wicks were re-wetted, food replaced and dead weevils removed. Parasitoid pupae were collected, placed in covered Petri-dishes and their identity confirmed as *M. aethiopoides* when the adult wasp emerged. Weevils collected in excess of the 100 caged were preserved in ethanol, and their abdominal and thoracic cavities were dissected to search for parasitoid larvae. The sub-basal elytral width, gender, the presence and developmental stage of parasitoid larvae, presence of teratocytes and the number of mature eggs in the calyces of female weevils were recorded.

Generalised Linear Models (GLM) (Nedler & Wedderburn 1972) were used to determine the effects of gender and site on size, the effect of size on parasitism and the effect of parasitism on egg number. The effect of gender on parasitism was analysed using GLM with binomial distribution and logit transformation.

RESULTS

Parasitism was recorded from 11 of the 15 survey sites with 63 parasitoids obtained from caged weevils and a further 45, of which 90% were 1st instar larvae, from dissected weevils (Table 1).

The highest level of parasitism of *R. conicus* was 16.8% at Kenilworth followed by Middlemarch (site A), Sutton Stream and Hakatarama Downs (Table 1). The proportion of male *R. conicus* parasitised (12%) was higher ($P < 0.001$) than that of females (3.7%) despite their lower abundance in samples, and this was consistent between sites. The sub-basal elytral width of males and females was similar in distribution ranging from 1.62–2.70 and 1.56–2.88 mm respectively. GLM detected no size difference between genders, both having a mean width of 2.23 mm, or any effect of site on size. Parasitism was concentrated in medium sized weevils and absent from those under 1.80 or over 2.64 mm in width (Fig. 1).

A higher proportion ($P < 0.001$) of parasitised females (73%) contained no mature eggs than unparasitised females (6%). The mean number of mature eggs was lower in parasitised females (1.72 ± 0.90) than those that were not parasitised (5.55 ± 0.15). Teratocytes were present in all dissected parasitised weevils except the females that still contained eggs, a superparasitised female and two males.

DISCUSSION

Field parasitism of *R. conicus* by *M. aethiopoides* has previously been found in both lucerne and pasture sites in South Canterbury and Otago (Ferguson et al. 1998, 1999). The current results support these findings and further confirm, as suggested by Barratt et al. (1997), that such parasitism is not restricted to the habitat of *S. discoideus*. In this study lucerne was absent from the majority of sites where parasitism of *R. conicus* by *M. aethiopoides* was recorded.

The higher ratio of female to male weevils collected in the survey may have resulted from sampling bias as females may be more common on thistle flowers during summer while ovipositing, whereas males may be more evenly distributed on the plants. In agreement with Barratt & Johnstone (2001) male and female weevils did not differ in elytral width, yet parasitism was significantly higher in males. Host selection by *M. aethiopoides* might not therefore be influenced by weevil elytral width but by host activity, which is required for successful ovipositor penetration (Goldson et al. 1990). Gender-dependent variability in

TABLE 1: Number of cage-reared and dissected weevils, sex ratio of dissected weevils, number parasitised (superparasitised) and total percent parasitised for each site.

Site	Weevils			Parasitoids		Total parasitism (%)
	Caged	Dissected	♀/♂	Caged	Dissected	
Hakataramea						
Upper Hakataramea ^{1,2}	64	0	-	4	0	6.3
Kennilworth Station ¹	100	90	1.5	17	17 (2)	16.8
Hakataramea Downs ¹	49	6	1	5	0	9.1
Cloverbank ¹	98	77	1.9	6	6 (1)	6.3
Strath Taieri						
Hartfield Rd ^{1,2}	99	201	1.5	5	12 (1)	5.3
Island ¹	97	32	3	3	2	3.9
Middlemarch A ^{1,2}	75	9	3.5	7	2	10.7
Middlemarch B ^{2,3}	99	51	1.2	3	4 (1)	4.0
Sutton ³	99	143	1.4	2	2 (1)	1.2
Sutton Stream ³	98	0	-	10	0	10.2
Waitaki						
Dog Kennel Rd ¹	49	6	2	1	0	1.8
Ben Ledi Homestead ¹	0	2	♂ only	0	0	0.0
Mt. Stalker ⁴	0	14	1.8	0	0	0.0
Lindis Pass ²	50	7	0.4	0	0	0.0
Wedderburn ¹	0	2	1	0	0	0.0
TOTAL	977	640	1.5	63	45 (6)	6.3

¹Pasture. ²Roadside exotics. ³Lucerne. ⁴Shrubland.

weevil activity levels is suggested as a possible explanation for the result obtained, perhaps because male *R. conicus* have to seek females for mating.

Most parasitoids examined during weevil dissections showed no sign of a host immune response and were developing in the presence of teratocytes. The absence of teratocytes from the few parasitised females that contained mature eggs suggests that in these individuals, the parasitoid was unable to successfully partition the host's resources. These

FIGURE 1: Size distribution of parasitised male and female *Rhinocyllus conicus*.

parasitoids, although apparently healthy, may have failed to develop successfully in the absence of teratocytes.

The implications of *R. conicus* parasitism by *M. aethiopoides* remain undetermined. *Rhinocyllus conicus* is widely established and can significantly reduce nodding thistle seed production (Jesep 1975) but evidence that this translates into reduction of nodding thistle populations is lacking. It is therefore debatable whether the parasitoid reduces the weevil population sufficiently to compromise nodding thistle control, especially as *R. conicus* is only one of three biocontrol agents introduced to control the weed.

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