

April 1993

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Recommended Citation

Hoffman, George D. and Raffa, Kenneth F. 1993. "*Microctonus Pachylobii* (Hymenoptera: Braconidae): New Host Record From *Hylobius Radicis* (Coleoptera: Curculionidae), and Additional Notes on Its Biology," *The Great Lakes Entomologist*, vol 26 (1)
Available at: <https://scholar.valpo.edu/tgle/vol26/iss1/6>

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MICROCTONUS PACHYLOBII (HYMENOPTERA: BRACONIDAE):
NEW HOST RECORD FROM HYLOBIUS RADICIS
(COLEOPTERA: CURCULIONIDAE),
AND ADDITIONAL NOTES ON ITS BIOLOGY

George D. Hoffman¹ and Kenneth F. Raffa¹

ABSTRACT

The endoparasite *Microctonus pachylobii* was discovered parasitizing a new weevil host, *Hylobius radialis*. Thirteen of the 154 *H. radialis* adults collected were parasitized (8.5%). The median numbers of parasites per weevil were 26 (\bar{x} = 22.5) during the period April through June, and 4 (\bar{x} = 9.4) during August and September. The median male:female sex ratio was 0.91 (\bar{x} = 0.65). Males emerged approximately 1 day earlier than females. Median parasite mortality while in the cocoon was 10.2% per parasitized weevil (\bar{x} = 11.8%). *Microctonus pachylobii* was not found parasitizing two previously recorded weevil hosts from field samples, *Hylobius rhizophagus* and *H. pales*, and a laboratory study suggests that the parasite may have difficulty parasitizing the latter species.

Microctonus pachylobii Muesebeck was originally described as a parasite of the pitch-eating weevil, *Pachylobius picivorus* (Germar) (Muesebeck 1961). Its reported host range and geographical distribution were long confined to *P. picivorus* collected from southeastern United States (Krombein et al. 1979). Recently *M. pachylobii* was found parasitizing the pine root tip weevil, *Hylobius rhizophagus* Millers, Benjamin and Warner, and the pales weevil, *Hylobius pales* (Herbst), collected from central and northwest Wisconsin (Rieske et al. 1989). These two weevil species, plus *P. picivorus* and the root collar weevil, *Hylobius radialis* Buchanan, are pests in Christmas tree plantations in the Great Lakes region. The discovery of *M. pachylobii* in Wisconsin has led to questions regarding its potential as a biological control agent against these weevil pests. There is currently little information in the literature on the biology of this parasite.

We recently discovered *M. pachylobii* parasitizing a new weevil host, *H. radialis*, during the course of a study at the site in central Wisconsin where Rieske et al. (1989) had previously found *M. pachylobii*. Data were collected on the incidence of parasitism of the three *Hylobius* species, the number of parasite progeny per parasitized weevil, differences between female and male parasite pupal development time, parasite mortality during the pupal period, and the parasitization rates of the three *Hylobius* species under laboratory conditions.

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MATERIALS & METHODS

Endoparasitic larvae were initially recovered from *H. radicans* during routine dissections of adults of the three *Hylobius* species. The weevils were collected in 1991 as part of a study on weevil reproductive biology. The study site was a red pine, *Pinus resinosa*, and jack pine, *P. banksiana*, plantation 10 miles south of Milston, Jackson County, Wisconsin. Living weevils were collected from either pitfall traps baited with turpentine and ethanol, or screen traps wrapped around tree trunks. After the initial discovery of the parasite larvae a portion of the *Hylobius* weevils were held alive for parasite emergence. These individuals were not a random sample of all the collected weevils, but were from weekly collections in which parasitized weevils were found during dissections. The adult weevils were placed in individual diet cups containing a section of fresh red pine twig (weevil food) and peat moss (pupation medium for emerging parasite larvae). They were held at 22°C and 16:8 L:D photoperiod. The diet cups were checked for the presence of parasite cocoons every three to four days for two months. When cocoons were found they were separated from the peat moss, placed individually in gelatin capsules, and held at the same temperature and photoperiod. The peat moss was placed in a petri dish and observed for emergence of parasites from any undiscovered cocoons. The adult weevil, usually dead, was dissected and any unemerged parasite larvae recorded. Cocoons were checked daily for adult parasite eclosion. Cocoons from which parasites did not emerge were opened to determine the developmental stage at which the parasite died.

Because *M. pachylobii* was not found parasitizing field collected *H. pales* and *H. rhizophagus* during this study, we investigated the ability of the parasite to successfully parasitize the three *Hylobius* species under laboratory conditions. Parasites that emerged in the laboratory were mated when two days old as adults. Four females were placed with four males in a diet cup containing a honey and water source. Two days later, individual females were placed with individual *H. radicans* (n = 8), *H. rhizophagus* (n = 4), and *H. pales* (n = 10) under the above conditions. The former two weevil species had been collected in the field and held in the laboratory for 3 months to assure they were not already parasitized. The *H. pales* adults were from a laboratory colony. Twenty four hours later the parasites were removed and the weevils held for 3 months while observed twice weekly for parasite emergence. Weevils that died during this period were dissected and checked for parasite larvae. Weevils still alive after three months time were dissected and checked for the presence of diapausing first instar parasite larvae. *Microctonus* species diapause as first instar larvae within the host (Loan 1967).

RESULTS

Microctonus pachylobii larvae were found in 7 of the 118 dissected *H. radicans*, and larvae emerged from 6 of the 36 isolated weevils. The total parasitization rate was 8.5%. No parasites were found in the 52 dissected and 16 isolated *H. rhizophagus*, or the 77 dissected and 10 isolated *H. pales* weevils.

The data on the number of parasites per parasitized *H. radicans* are separated into two groups based on the date the weevils were collected. We report results using the median and range for data from small sample sizes that appear to be non-normally distributed. The conventional mean and its standard error are also reported. The first group is comprised of weevils collected during the early peak of weevil activity (April through June). The second group consists of root collar weevils collected during the second peak of activity (August and September). Weevil activity is very low from mid-July to early

August (Raffa and Hall 1988). The median number of parasites (larvae or cocoons plus unemerged larvae) in the April-June group (6 female and 2 male weevils) was 26, with a range of 9 to 32 ($\bar{x} = 22.5 \pm 2.9$). The median number in the August-September group (2 female and 3 male weevils) was 4, with a range of 1 to 22 ($\bar{x} = 9.4 \pm 4.2$).

A few parasite larvae did not successfully emerged from their hosts. Two of the isolated weevils, from which 23 and 14 larvae emerged and pupated, contained 2 and 1 unemerged parasite larvae, respectfully.

The median male:female sex ratio of the brood irrespective of weevil collection date was 0.91, with a range of 100% female to 1.14 (n = 6) ($\bar{x} = 0.65 \pm 0.18$). The median parasite mortality while in the cocoons was 10.2% per weevil (n = 6), with a range of 4.3% to 23.1% ($\bar{x} = 11.8\% \pm 2.5$). Five of the 14 dead parasites died as larvae or shortly after spinning the cocoon, three died during the pupal stage, and six of the dead were completely formed adults. Some of these adults appeared to have been unable to completely free themselves from the pupal skin.

Because isolated weevils were not checked every day for parasite emergence, the exact length of the pupal period is unknown for most of the parasites. Our data suggests that on a per weevil basis (n = 5), male parasites (n = 43) eclose 0.82 days earlier (median) than females (n = 55). Parasite larvae were seen emerging from a laboratory parasitized *H. rhizophagus*. In this incident the average length of the pupal period was $11.6 \pm .12$ days for the all male brood (n = 17).

Four of the 8 *H. radialis* weevils confined with female *M. pachylobii* were parasitized. One of the parasitized weevils died, possibly due to its food drying out, and the parasite larvae were discovered upon dissection of the cadaver. All male parasite progeny emerged from the other three weevils. Three of the four *H. rhizophagus* weevils were parasitized. Parasite larvae were found in one moribund weevil, and the other two weevils gave rise to all male parasites. None of the *H. pales* weevils were parasitized (n = 10). Dissections of the nonparasitized weevils of all three species revealed no diapausing first instar parasites. In this laboratory parasitism test the mean duration of the egg plus larval stage of *M. pachylobii* in *H. radialis* and *H. rhizophagus* was 41.0 ± 1.14 days (n = 5).

The oviposition behavior of *M. pachylobii* during the first five to 10 minutes after contact with *H. radialis* and *H. rhizophagus* did not seem conducive to successful parasitization. The parasites were highly attracted to feeding weevils, and less so to feeding scars. Many of the parasites would repeatedly thrust their ovipositor at the mouthparts of feeding weevils. Weevils which were not moving were less attractive to parasites than feeding scars. Parasites would routinely walk over stationary weevils without making attempts to oviposit. We saw a few parasites chase after walking weevils, but no complete ovipositional thrusts were made, even after the weevil had stopped walking. Two parasites were seen to hang from the lower abdomen of a weevil and thrust their ovipositor forward. The ovipositors did not appear to penetrate the intersegmental membrane, but we had a poor view of the parasites. Weevil behavior did not appear to be influenced by the presence of the parasite. We did not observe the behavior of parasites placed with *H. pales*.

DISCUSSION

Finnegan (1962) and Shenefelt & Millers (1960) each reported the rare occurrence of a larval parasite of *H. radialis*. This study is the first to identify a Hymenopteran parasite of adult *H. radialis*. The expansion of the host range of

M. pachylobii to include *H. radialis* is not surprising given that the parasite's range includes two weevil genera, and the fact that *H. radialis* and *H. rhizophagus* are sibling species. What is surprising is that we did not find parasitized *H. pales* and *H. rhizophagus* in our field collections. Four years earlier, Rieske et al. (1989) found parasitized *H. pales* and *H. rhizophagus* at this site, but not parasitized *H. radialis*. The laboratory parasitism test showed that *M. pachylobii* can parasitize *H. rhizophagus* as readily as *H. radialis*, but may have difficulty parasitizing *H. pales*.

The 8.5% parasitism rate of *H. radialis* is a pooled value combining all collections over the summer. Depending upon the life history of the host, parasitism rates expressed in this manner can misrepresent the total loss to parasitism for a life stage over a generation (Van Driesche et al. 1991). The long adult life and seasonal history of *H. radialis* (Wilson and Millers 1983) will reduce the magnitude of this error.

The time at which *H. radialis* weevils were collected from the field appears to affect the number of larvae found in both female and male weevils. The April-June peak of weevils was primarily composed of reproductive individuals. Female weevils were filled with eggs in May and June, and ovipositing parasites may be able to detect a host with substantial nutritional reserves. Females collected in August-September were almost all postreproductive or newly emerged adults. Conversely, males collected during both peaks were mostly sexually mature with fully enlarged gonads (G.D. Hoffman unpublished data). This suggests that differences in nutritional reserves between weevils collected during the April-June and August-September peaks may not explain the difference in the number of parasites per weevil during these two periods. It is not known how many generations *M. pachylobii* has each year, but the long juvenile development period, and length of time over which parasitized weevils were found, suggests there are two generations a year.

The all male *M. pachylobii* progeny that resulted from the laboratory parasitism test suggests that female parasites were not fertilized during the two days they were confined with males. We observed male and female parasites coupling soon after the two sexes were placed in the same vial. We did not investigate the duration of copulation or note behaviors of the parasites which could have disrupted fertilization. Alternative explanations are that the parasites can control the sex ratio of their progeny, or that the male parasites were infertile.

The life histories of *H. rhizophagus* (Kearby and Benjamin 1969) and *H. pales* (Finnegan 1959, Bliss and Kearby 1970) are similar to that of *H. radialis* (Wilson and Millers 1983); and same is true for the annual pattern of ovary and teste development (G.D. Hoffman unpublished data). This suggests that constraints imposed on the parasite's biology by the life history and physiology of the three weevil species should be similar.

The data from this and other studies suggest that *M. pachylobii* will not be an effective biological control agent of *Hylobius* weevils in the Great Lakes region. We recorded low parasitism rates of *H. radialis*, and found that the parasite does not consistently parasitize a given weevil species at a single location. Of particular interest is that during the six-year period in which our laboratory has studied these four weevil pests, *M. pachylobii* was never found parasitizing *P. piceivorus*, despite high local densities of this weevil. Nor has *M. pachylobii* been found to parasitize *H. pales* in the southern United States (Lynch 1984).

ACKNOWLEDGMENTS

We thank Steven Krauth, University of Wisconsin, and Paul Marsh, Systematic Entomology Laboratory, USDA, for the identification of *M. pachylobii*. Richard Hofstetter and Todd Patton assisted with the weevil collections. We thank Dan Mahr and Michael Strand for reviewing the manuscript. This study was supported by The University of Wisconsin-Madison Graduate School, the University of Wisconsin-Madison College of Agricultural and Life Sciences Center for Integrated Agricultural Systems, The Wisconsin Christmas Tree Producers Association, and USDA Competitive Grants 86-CRCR-1-2077, and 86-FSTY-90210.

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