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Microctonus aethiopoides Loan (Hymenoptera: Braconidae), Fecundity and Longevity of an Endoparasitoid Attacking *Hypera postica* (Gyllenhal) (Coleoptera: Curculionidae)

دراسة خصوبة و إعاشة حشرة الطفيل الداخلي *Microctonus aethiopoides* Loan لحشرة سوسة البرسيم *Hypera postica* (Gyllenhal)

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Abstract: Average time required for *Microctonus aethiopoides* development from egg to adult was 24.5 ± 0.98 d (Mean ± SE) at a temperature regime that fluctuated between 15 and 23.3 °C. Average longevity of *M. aethiopoides* females was 15.4 ± 1.2 d (Mean ± SE). Following emergence, mated female *M. aethiopoides* were provided with 25 laboratory reared adult weevils per day until parasitoid death. Oviposition commenced the day of adult emergence, and mean fecundity was 97.0 ± 7.3 (Mean ± SE) eggs per female. Superparasitism was common but declined with parasitoid age. Life table statistics assuming 50% female progeny were calculated. Using total eggs laid $(m_x)_t$, life table statistics (Mean ± SE) were gross reproductive rate (GRR) = 51.4 ± 3.7, net reproductive rate (R_o) = 49.4 ± 3.8, intrinsic rate of increase (r_m) = 0.153 ± 0.003, and mean generation time (T) = 26.3 ± 0.2. Using only effective eggs laid $(m_x)_e$ (i.e. excluding superparasitism), life table statistics were GRR = 41.6 ± 3.5, R_o = 39.4 ± 3.3, *rm* = 0.142 ± 0.003, and T = 26.9 ± 0.3. The usefulness of these values as indices of relative effectiveness of *M. aethiopoides* against adult *H. postica* are discussed.

Keywords: *Microctonus*, solitary parasitoid, alfalfa weevil, proovigenic, superparasitism, life tables.

المستخلص: لقد خلصت الدراسة إلى أن الوقت اللازم لمراحل نمو الطفيل من البيضة الى الطور البالغ يستغرق في المتوسط 24.5 يوما و ذلك عند درجة حرار تراوحت ما بين 15°م و 23.3°م. كما بينت الدراسة ان متوسط حياة أنثى طفيل حشرة *M. aethiopoides* قد بلغ 15.4 يوما. دلت الدراسة انه بعد إكتمال التطور و إنبثاق الطور البالغ لأنثى الطفيل تبداء بوضع البيض مباشرة بعد تزاوجها. و عند تعريض أنثى الطفيل من اليوم الأول من أكتمال نموها لعدد 25 من الطور البالغ لسوسة البرسيم و جد انها تضع ما متوسطه 97 بيضة بصورة يومية فترة حياتها . لوحظ من الدراسة أيضا ان انثى الطفيل تقوم بالتطفل على العائل أكثر من مرة واحدة حيث تعرف هذه الظاهرة بالتطفل المتكرر (superparasitism). و على الرغم من أن ظاهرة التطفل المفرط كانت شائعة الحدوث في بداية حياة الطفيل إلا أنها تتناقصت مع التقدم في العمر للطفيل . تم خلال الدراسة حساب جدول الحياة لأنثى طفيل *M. aethiopoides* بناء على فرضية ان نسبة الأناث الى الذكور هي 50%. و باستخدام عدد البيض الكلي $(m_x)_t$ فإن إحصائيات جدول الحياة مثل التكاثر الكلي قد بلغ 51.4 بينما بلغ معدل التكاثر الصافي قد بلغ 49.4 ، و قد بلغ معدل الزيادة الآتية 0.153 r_m، و متوسط مدة الجيل 26.3 (T) يوما . و عند استخدام عدد البيض الفعلي الذي تم وضعه $(m_x)_e$ (باستثناء التطفل الفائق) فإن إحصائيات جول الحياة كان التكاثر الكلي قد بلغ 41.6، بينما بلغ معدل التكاثر الصافي 39.4، بلغ معدل الزيادة الآتية 0.142، و متوسط مدة الجيل 26.9. و يناقش البحث فائدة هذه القيم كمؤشرات لمدى فاعلية النسبية لحشرة طفيل *M. aethiopoides* في مكافحة حشرة سوسة البرسيم .

كلمات مدخلية: سوسة البرسيم، الطفيل الداخلي، التطفل المتكرر، *Microctonus aethiopoides*، جدول الحياة.

Introduction

Only a few parasitoid species use the adult stage of Coleoptera (particularly weevils) as a host resource (Clausen, 1940; Tobias, 1965). One of these parasitoids is *Microctonus aethiopoides* Loan, a species of European origin, which attacks the adult stage of the alfalfa weevil, *Hypera postica* (Gyllenhal). *Microctonus aethiopoides* has an ovipositional behavior designed to overcome a mobile and sclerotized adult weevil, both of which provide a general means to overcome antiparasitism mechanisms as observed in other host-parasitoid relationships (Tobias, 1965; Gross, 1993; Mohamed, 2003). Although *M. aethiopoides* is characterized by ovipositing into moving adult weevils, not all weevils are equally susceptible to parasitism. In addition to heavy sclerotization, and movement that acts as a deterrent to parasitism, other defense mechanisms such as tightness of the abdominal sclerites, kicking the attacking wasp, squatting behavior, remaining motionless, act as detriment to parasitoid attack even when they are encountered and accepted by female parasitoid (Mohamed, 2003). Superparasitism is another factor that represents a loss of parasitoid fitness. Such a phenomenon in a solitary endoparasitoid such as *M. aethiopoides* will always be considered a 'waste' because ovipositing in previously parasitized hosts represents a loss in reproductive potential compared with ovipositing in unparasitized hosts (van Alphen and Visser, 1990, Godfray, 1994). Altogether or independently these factors can affect the outcome of the parasitoid-host interactions.

A number of studies have been carried out on *M. aethiopoides* establishment and biology in the United States of America (Loan and Holdaway, 1961; Drea, 1968; Fusco and Hower, 1973, 1974; Sunjaya, 1975; van Driesche and Gyrisco,1979; Morales and Hower, 1981; Bryan, *et al.* 1993; Kingsley, et al. 1993). However, while there is a good deal of information on the general biology of *M. aethiopoides* as it relates to *H. postica*, there is little information regarding the fecundity and longevity. To complement the earlier work (e.g. Neal, 1970; Fusco and Hower 1973, 1974), this study determined longevity and fecundity for developing a life table for *M. aethiopoides* and then applied the data to estimate the parameters for population increase.

Materials and Methods

- Host and parasitoid culture

Field-collected alfalfa weevil larvae were reared in the laboratory to provide a supply of unparasitized adult weevils. Alfalfa weevil larvae were placed on field collected alfalfa, secured in a paper bag and kept on a bench under a room temperature of 21 °C. Fresh alfalfa was added every three or four days as needed. After 15 to 20 days, about 5000 newly emerged adult weevils were divided into groups; each consisted of 25 adults placed into vented 1.0 litre ice cream containers and fed alfalfa until they were needed for exposure to a parasitoid (see below).

M. aethiopoides were reared from 150 field collected, overwintered alfalfa weevil adults. Weevils were divided into six groups, each consisting of 25 adults held in vented 1 litre ice-cream containers with cheese cloth at the bottom to facilitate parasitoid pupation. The weevils were fed and maintained in an environmental chamber at a temperature regime that fluctuated between 15-23.3 °C and 16: 8 light dark regimen To prevent disruption to parasitoid pupation once parasitoid larvae began to emerge from *H. postica* adults, weevils were transferred to new containers in accordance with parasitoid emergence. The pupae were held in the environmental chamber until adult parasitoids eclosed.

From the eclosed parasitoid adults, 14 were dissected to determine the number of ovarioles and eggs counted under a light binocularscope at x12 magnification.

- Longevity and fecundity of *M. aethiopoides*

Following emergence, female *M. aethiopoides* were mated and caged immediately with alfalfa. Cages used for oviposition were made from round transparent plastic containers, 18.7 cm x 8.0 cm deep with a mesh lid, similar to the design of Loan and Holdaway (1961). Each cage was also provided with two cotton dental wicks about five cm long soaked in 10% honey solution, with one placed on the top of the cage and the other in a Petri dish at the bottom inside of the cage to act as a feeding source for the parasitoid. Ten *M. aethiopoides* females were used, each considered a replicate. Each parasitoid was confined with 25 laboratory reared adult weevils, which were replaced daily until the parasitoid died. After 24 h each group of exposed weevils was moved to another cage, fed and kept alive for a week to allow for parasitoid egg hatch. Weevils were then placed in a Petri dish for dissection under a binocular microscope. Each weevil's sex, presence of parasitoid larva, and parasitoid remains (i.e., head capsules and dead bodies of supernumeraries resulting from super-parasitism) were determined (Fig. 1). The total

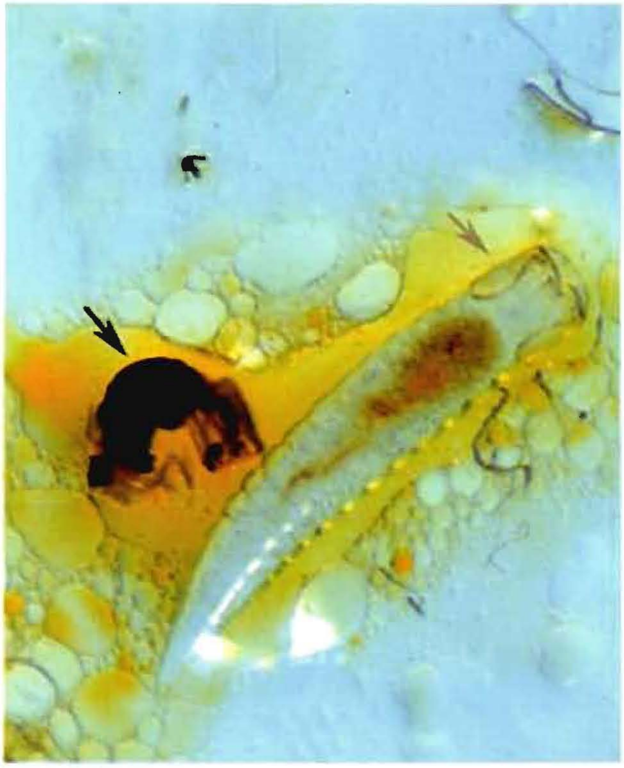

Fig. **1.** Two *M. aethiopoides* first stadium larvae dissected from superparasitized adult alfalfa weevil; dead melanized supernumerary (dark arrow), surviving larva (light arrow).

numbers of weevils exposed, numbers dissected, and numbers parasitized and superparasitized were recorded.

- *M. aethiopoides* life table

The longevity and fecundity of *M. aethiopoides* was used to calculate female parasitoid daily survivorship (l_x) and fecundity (m_x). This in turn was used to determine life table statistics for *M. aethiopoides,* as described by Messenger (1964). Adjustment of m_x was made to generate life tables with and without corrections to gross parasitoid reproduction, and assuming either 100% (i.e., hypothetical ratio) or 50% (based on a 3 yr field average; Mohamed, unpublished) female progeny. Gross parasitoid reproduction used the total eggs laid regardless of superparasitism and was referred to as $(m_x)_t$; whereas corrected reproduction considered only effective eggs laid, excluding all supernumerary eggs resulting from superparasitism, and was referred to as $(m_x)_e$ (Messenger, 1964). From these data, the life table statistics, most importantly net reproductive rate (R_o), the intrinsic rate of increase (r_m), and mean generation time (T) were computed. Standard errors of r_m were estimated using a jackknife statistical procedure (Meyer, *et al.* 1986).

Table 1. Total number of female and male adult *H. postica* exposed, parasitised and superparasitised by *M. aethiopoides.*

Total	
Total adult female recovered	1890
Total adult males recovered	1707
Total *H. postica* parasitized	**752**
No. of females parasitized	402
No. of males parasitized	350
Total *H. postica* superparasitized	**194**
No. of females superparasitized	112
No. of males superparasitized	82
Total eggs laid by *M. aethiopoides*	**970**
Total *M. aethiopoides* larvae alive	744
Total *M. aethiopoides* larvae dead	226

Results and Discussion

- Exposure Results

A total of 3850 weevils were exposed to parasitoids of which 1890 and 1707 female and male weevils were subsequently dissected to determine parasitoid preference for either weevil gender, superparasitism and parasitoid mortality (Table 1). No statistical differences were found in preference of *M. aethiopoides* parasitizing (x^2 = 0.23, df = 1, $P < 0.5$) or superparasitizing (x^2 = 1.6, df = 1, $P < 0.25$) either weevil sex. A total of 970 *M. aethiopoides* eggs were recovered from the dissected weevils of which 744 (76.7%) were viable, the remaining supernumerary eggs being atrophied. As a solitary endoparasitoid this result is not surprising and observed by Loan and Holdaway (1961), Fusco (1971), and van Driesche and Gyrisco (1979).

- Potential fecundity

The reproductive system of female *M. aethiopoides* observed in this study was similar to that described by Loan and Holdaway (1961) and Barratt *et al.* (1999). Dissections of recently emerged *M. aethiopoides* revealed the presence of fully matured oöcytes in their ovarioles, thus characterizing the species as proovigenic (sensu Flanders, 1950). The parasitoids had 90.2 ± 3.25 (Mean ± SE, range 74-110) mature eggs in their ovaries, apparently ready to be laid (see below). Most of the parasitoids had paired ovaries each with four ovarioles. However, of the 14 females

Table 2. Longevity and ovipositional characteristics of female *M. aethiopoides* (n = 10) at 15-23.3°C and 16: 8 light dark regimen.

Characteristics	Mean ± SE
Time to adult (days)	24.5 ± 1.00
Adult longevity	15.4 ± 1.20
Total eggs/female	96.0 ± 7.08
Eggs/female/day	6.9 ± 0.90
Eggs/parasitized adult alfalfa weevil	1.1 ± 0.03

dissected one was found to have a total of 12 ovarioles, and two were found to have 9 ovarioles. This number of eggs is similar to that reported by Drea (1968) who found approximately 100 fully matured oöcytes in newly emerged female parasitoids. However, depending on the parasitoid biotype, the number of ovarioles and the eggs can vary considerably ranging from 6 to 12 ovairoles, and 18 to 90 eggs, respectively (Loan and Holdaway, 1961; Barratt, *et al.* 1999).

● Longevity and fecundity

The longevity and oviposition characteristics of *M. aethiopoides* are presented in Table 2, and age-specific survivorship (l_x) and natality (m_x) curves are shown in Fig. 2. The first adult death was recorded on day ten, and the average longevity of 15.4 d for *M. aethiopoides* females was slightly higher than the estimate of 12.1 d under a constant 23.0 °C reported by Loan and Holdaway (1961) and Fusco and Hower (1974). The longevity of adult female *M. aethiopoides* appears to be consistent with other *Microctonus* species, with longevity ranging from 6 to 21 d (Smith, 1956; Drea, *et al.* 1972; Luff, 1976; Goldson, *et al.* 1995).

In *M. aethiopoides*, oviposition commenced on the day of adult parasitoid emergence. Most parasitoid females laid ≈ 32% of the eggs within the first two days after eclosion and declined gradually thereafter. This is in accordance with other laboratory studies of *M. aethiopoides* (Fusco and Hower, 1974; van Driesche, 1975). On average, the total number of eggs laid by a female during its lifetime was 96.0 (Table 2), which was similar to the average potential fecundity of 90.2 eggs found from dissection of newly emerged parasitoids. From these observations, it can be concluded that *M. aethiopoides* is a proovigenic species.

No relationship was detected between the fecundity of a *M. aethiopoides* and the age at death (Fig. 3). The fecundity of *M. aethiopoides* found in

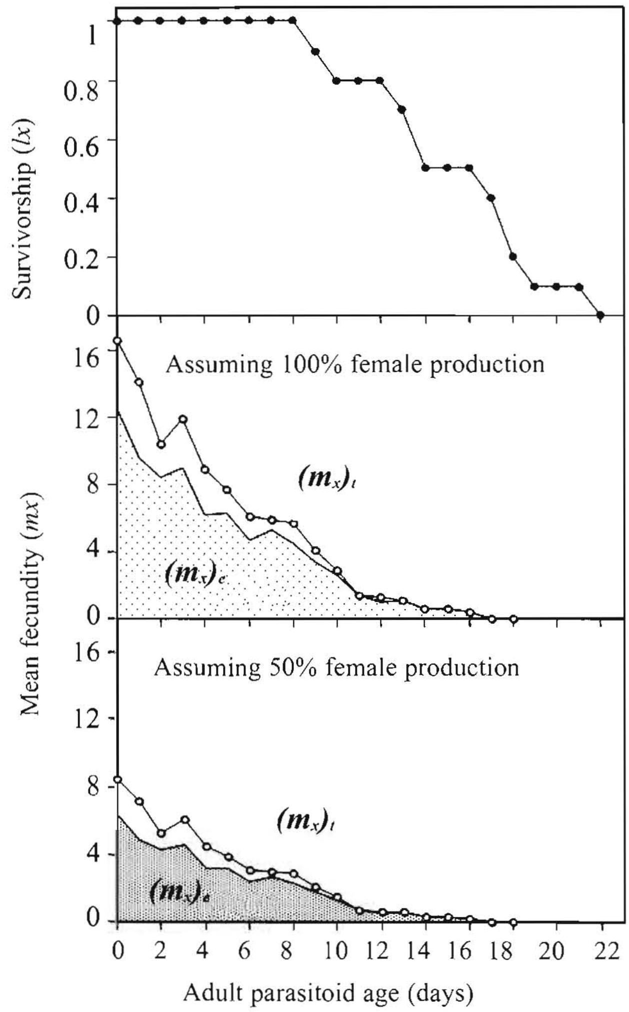

Fig. 2. Age-specific survivorship (l_x) and fecundity (m_x) showing the total eggs laid per day $(m_x)_t$, and effective eggs laid per day $(m_x)_e$ when 100% and 50% of progeny females were considered.

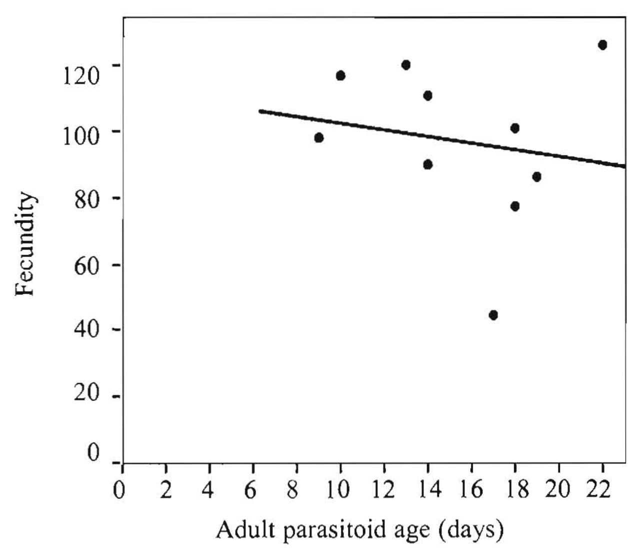

Fig. 3. Relationship between total eggs laid and survivorship for each female *M. aethiopoides* (r^2 = 0.037; F = 0.31; P = 0.59; y = 114.04 - 1.15x).

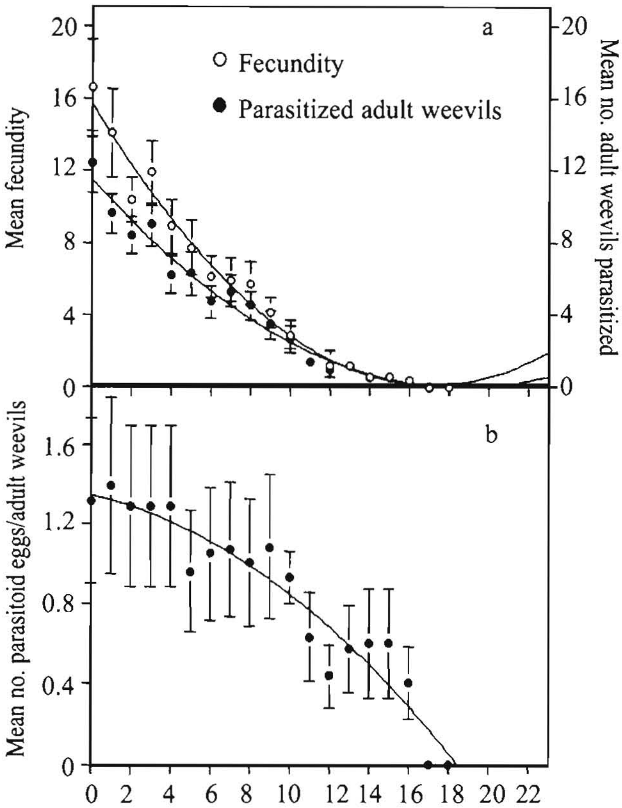

Fig. 4. a) Daily total fecundity (Mean ± SE) with adult weevil parasitized. b) Number of egg(s) (Mean ± SE) per parasitized weevil.

this study was greater than the average fecundity of 59.3 eggs reported by Fusco and Hower (1974) under similar experimental conditions. Reported fecundities of other *Microctonus* species include: *M. hyperodae*, 42.7 eggs (Goldson, *et al.* 1996), *M. vittatae*, 132 eggs (Smith, 1952), and *M. caudatus*, 152 eggs (Luff, 1976).

Superparasitism by *M. aethiopoides* declined with parasitoid age (Fig. 4a,b). Arguably, the occurrence of superparasitism in this study was influenced by the size of the arena, plus we observed that parasitized weevils tended to be more vulnerable to subsequent attack than were unparasitized weevils that displayed behavioral and/or morphological defences against parasitism (Mohamed, 2003). However, host discrimination by *M. aethiopoides* was not considered in the current study.

- *M. aethiopoides* life tables

Variation in the values of life table statistics can be attributed mainly to the different progeny sex ratios and effect of superparasitism (Table 3). Both sex ratio and superparasitism had a large influence on gross reproductive rate (GRR) and net reproductive rate (R_o) but no measurable effect on mean generation time (T). In the case of intrinsic rate of increase (r_m), progeny sex ratio seemed to have a larger effect than superparasitism; however, values represent instantaneous rates which are more difficult to evaluate than the other life table statistics. Although the growth rate of *M. aethiopoides* appears rather low, published life table information for other Euphorinae species is unavailable for comparison.

The utility of life tables statistics, most prominently the intrinsic rate of increase (r_m), provides an index of a parasitoid's potential as a biological control agent (Messenger, 1964). However, this is probably not true in the case of *M. aethiopoides*. First, *M. aethiopoides* is bivoltine with discrete generations, hence statistics such as that apply to populations with overlapping generations and a stable age distribution have limited predictive value. Second, the outcome of complex behavioral interactions between *M. aethiopoides* and its host *H. postica* (Mohamed, 2003) may overshadow information in the life table. Finally, several attributes of *M. aethiopoides* make it a particularly effective biological control agent for *H. postica*: the parasitoid completes two generations during the single *H. postica* generation per year, and parasitism by *M. aethiopoides* causes sterilization as well as ultimately death of its weevil host (van Driesche, 1975; Sunjaya, 1975; Mohamed, (unpublished).

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References

van Alphen, J.J.M. and Visser, M.E. (1996) Superparasitism as an adaptive strategy for insect parasitoid. Ann. Rev. Entomol. **35**: 59-79

Barratt, B.I., Evans, A.A., Stoltz, D.B., Vinson, S.B. and Easingwood, R. (1999) Virus-like particles in the ovaries of *Microctonus aethiopoides* Loan (Hymenoptera: Braconidae), a parasitoid of adult weevils (Coleoptera: Curculionidae). J. Inver. Path. **73:** 182-188

Bryan, M.D., Dysart, R.J., and Burger, T.L. (1993) Releases of introduced parasites of the alfalfa weevil in the United States, 1957-1988. Misc. Publ. 1504. USDA, Washington DC.

Table 3. Life table statistics (Mean ± SE) for *M. aethiopoides* (n = 10) parasitizing *H. postica* analyzed using the jackknife statistical procedure.

	When sex ratio 100% females		When sex ratio 50% females	
	Total eggs laid	**Effective eggs laid**	**Total eggs laid**	**Effective eggs laid**
GRR	96.0 ± 7.08	73.6 ± 5.40	51.4 ± 3.73	41.6 ± 3.45
R_0	94.2 ± 7.24	72.5 ± 5.41	49.4 ± 3.75	39.4 ± 3.34
r_m	0.179 ± 0.003	0.168 ± 0.003	0.153 ± 0.003	0.142 ± 0.003
T	26.1 ± 0.23	26.2 ± 0.22	26.3 ± 0.23	26.9 ± 0.28

GRR: gross reproductive rate; R_0: net reproductive rate; r_m: intrinsic rate of increase; **T:** mean generation time.

Clausen, C.P. (1940) Entomophagous insects. McGraw-Hill, New York.

Drea, J.J. (1968) Castration of the male alfalfa weevil by *Microctonus* spp. J. Econ. Entomol. **61:** 1291-1295.

Drea, J.J., Dysart, R.J, Coles, L.W., and Loan, C.C. (1972) *Microctonus stelleri* (Hymenoptera: Braconidae, Euphrinae), a new parasite of the alfalfa weevil introduced into the United States. Can. Entomol. **104:** 1445-1456.

van Driesche, R.G. (1975) An evaluation of *Microctonus aethiops* (Nees) (Hymenoptera: Braconidae) as a parasitoid of the adult alfalfa weevil, *Hypera postica* (Gyll.) (Coleoptera: Curculionidae) in Central New York. Ph.D. Thesis, Cornell Univ. (unpublished).

van Driesche, R.G. and Gyrisco, G.G. (1979) Field studies of *Microctonus aethiopoides*, a parasite of the adult alfalfa weevil, *Hypera postica*, in New York. Environ. Entomol. **8:** 238-244.

Flanders, S.E. (1950) Regulation of ovulation and egg disposal in the parasitic Hymenoptera. Can. Entomol. **82:** 134-140.

Fusco, R.A. (1971) Productivity of the alfalfa weevil parasite *Microctonus aethiopes* (Nees) in relation to various environmental, host, and parasite factors. Ph.D. thesis, Penn. State Univ.

Fusco, R.A. and Hower, A.A. (1973) Host influence on the laboratory production of the parasitoid, *Microctonus aethiops* (Nees). *Environ. Entomol.* **2:** 971-975.

Fusco, R.A. and Hower, A.A. (1974) Influence of parasitoid-host density and host availability on the laboratory propagation of *Microctonus aethiops* (Hym.: Braconidae) parasitoid of *Hypera postica* (Coleoptera: Curculionidae). *Entomophaga.* **19:** 75-83.

Goldson, S.L., McNeill, M.R., Proffitt, J.R. and Hower, A.A. (1995) An investigation into the reproductive characteristics of *Microctonus hyperodae* (Hym.: Braconidae), a parasite of *Listronotus bonariensis* (Col.: Curculionidae). Entomophaga. **40:** 413-426.

Gross, P. (1993) Insect behavioral and morphological defences against parasitoids. Ann. Rev. Entomol. **38:** 251-273.

Guald, I.D. (1988) Evolutionary patterns of host utilization by ichnuemonoid parasitoids (Hymenoptera: Ichneumonoidae and Braconidae). Biol. J. Linn. Soc. **35:** 351-377.

Kingsley, P.C., Bryan, M.D., Day, W.H., Burger, T.L., Dysart, R.J. and Schwalbe, C.P. (1993) Alfalfa weevil (Coleoptera: Curculionidae) biological control: spreading the benefits. Environ. Entomol. **22:** 1234-1250.

Loan, C. and Holdaway, F.G. (1961) *Microctonus aethiops* (Nees) acutt. and *Perilitus rutilus* (Nees) (Hymenoptera: Braconidae), a European parasites of *Sitona* Weevils (Coleoptera: Curculionidae). Can Entomol. **93:** 1057-1079.

Luff, M.L. (1976) The biology of *Microctonus caudatus* (Thomson), a braconid parasite of the ground beetle *Harpalus rufipes* (Degeer) Ecol. Entomol. **1:** 111-116.

Messenger, P.S. (1964) Use of life table in a bioclimatic study of an experimental aphid-braconid wasp host-parasite system. Ecol. **45:** 119-131.

Meyer, J.S., Ingersoll, C.G., McDonald, L.L. and Boyce, M.S. (1986) Estimating uncertainty in population growth rates: Jackknife vs. Bootstrap techniques. Ecol. **67:** 1156-1166.

Mohamed, A.M.A. (2003) Studies of parasites of adult alfalfa weevil, *Hypera postica* (Coleoptera: Curculionidae) Ph.D. thesis, Univ. of Wisconsin-Madison (unpublished).

Morales, J. and Hower, A.A. (1981) Thermal requirements for development of the parasite *Microctonus aethipoides* on the alfalfa weevil, *Hypera postica* integrated control. Environ. Entomol. **10:** 279-284.

Neal, J.W. (1970) A study of *Microtonus aethiops* (Nees), a braconid parasite of the alfalfa weevil, *Hypera postica* (Gyllenhal). Ph.D. thesis. University of Maryland (unpublished).

Smith, O.J. (1952) Biology and behavior of *Microctonus vittatae* Muesebeck (Braconidae). University of California Publication. Entom. **9:** 315-344.

Sunjaya, P.I. (1975) *Microctonus aethiopes* (Nees), a hymenopterous parasitoid of the adult alfalfa weevil. Ph.D. thesis, Univ. Wisconsin-Madison (unpublished).

Tobias, V.I. (1965) Generic groupings and evolution of parasitic Hymenoptera of the subfamily Euphorinae (Hymenoptera: Braconidae). Part 1. Entomol. Rev. **44:** 494-507.

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