

June 1998

Development, Survival and Phenology of the Sweetclover Weevil Parasitoid, *Pygostolus Falcatus* (Hymenoptera: Braconidae)

Lindsey R. Milbrath
North Dakota State University

Michael J. Weiss
University of Idaho

Follow this and additional works at: <https://scholar.valpo.edu/tgle>

 Part of the [Entomology Commons](#)

Recommended Citation

Milbrath, Lindsey R. and Weiss, Michael J. (1998) "Development, Survival and Phenology of the Sweetclover Weevil Parasitoid, *Pygostolus Falcatus* (Hymenoptera: Braconidae)," *The Great Lakes Entomologist*: Vol. 31 : No. 2 , Article 6.
Available at: <https://scholar.valpo.edu/tgle/vol31/iss2/6>

This Peer-Review Article is brought to you for free and open access by the Department of Biology at ValpoScholar. It has been accepted for inclusion in The Great Lakes Entomologist by an authorized administrator of ValpoScholar. For more information, please contact a ValpoScholar staff member at scholar@valpo.edu.

DEVELOPMENT, SURVIVAL AND PHENOLOGY OF THE SWEETCLOVER
WEEVIL PARASITOID, *PYGOSTOLUS FALCATUS*
(HYMENOPTERA: BRACONIDAE)

Lindsey R. Milbrath^{1,2} and Michael J. Weiss^{1,3}

ABSTRACT

Biennial sweetclovers were widely used for soil improvement and as forages in the first half of the 1900s. The introduction of the sweetclover weevil, *Sitona cylindricollis*, caused a drastic decline in sweetclover acreage. In North Dakota, yellow sweetclover, *Melilotus officinalis*, is still the legume of choice on organic farms. In an effort to control the weevil, the thelytokous parasitoid *Pygostolus falcatus* was imported. Parasitoids were studied for temperature-dependent development, and adult longevity as influenced by temperature and availability of provisions. Development from egg to adult at 15, 20, 25 and 30°C was 58, 28, 22 and 21 d, respectively. No parasitoids were reared out at 10°C, although diapausing first instars were present. Longevity of adult parasitoids provided honey, water, sweetclover and sweetclover weevils at 15, 20, 25 and 30°C was 29, 22, 12 and 6 d, respectively. Adults provided the following combinations of provisions at 25°C survived for: nothing—2 d; water—2 d; honey—4 d; honey and water—6 d; honey, water and sweetclover—11 d; honey, water, sweetclover and hosts—12 d. Field cage releases and a degree-day model developed for the parasitoid demonstrated that poor synchrony between *P. falcatus* and the sweetclover weevil hinders its usefulness as a biological control agent.

Biennial sweetclovers, *Melilotus* spp. Miller, were introduced into North America from Eurasia in the early 1700s, but not until the end of the 1800s did sweetclover gain value in agriculture for soil improvement and, later, as a forage (Smith and Gorz 1965). The sweetclover weevil, *Sitona cylindricollis* Fåhraeus, was first recorded in 1924 in Quebec and soon spread across Canada and the northern states (Brown 1940, Bird 1947, Munro et al. 1949). Subsequently, sweetclover acreage significantly declined in many areas, e.g. in some parts of North Dakota acreage was reduced up to 75% after the weevil's arrival in the state (Munro et al. 1949). Damage to the plant is caused by the adult weevil feeding on the foliage, with seedling stands being at greatest risk to destruction during the spring (Bird 1947, Munro et al. 1949).

¹Department of Entomology, North Dakota State University, Fargo, North Dakota 58105.

²Current address: Department of Entomology, Kansas State University, Manhattan, KS 66506.

³Current address: Department of Plant, Soil and Entomological Sciences, University of Idaho, Moscow, ID 83844.

Heavy feeding in the late summer of the establishment year can also significantly reduce forage yields the following season (Craig 1978). Larvae feed on the root system of second-year sweetclover but are not considered an economic problem (Bird 1947). Few farmers still use sweetclover due, in part, to the sweetclover weevil and the advent of synthetic fertilizers (Smith and Gorz 1965). However, yellow sweetclover, *Melilotus officinalis* Lam., is still the legume of choice by organic farmers in North Dakota for use in their rotations.

Earlier attempts at biological control of the sweetclover weevil involved three braconids and one tachinid. All were oligophagous, solitary, adult endoparasitoids of *Sitona* spp. None of the parasitoids became established on the sweetclover weevil. Explanations offered included climatic unsuitability and poor synchrony (Munro et al. 1949, 1950; Loan 1961, Loan and Holdaway 1961a, 1961b; Loan 1965). Our current biological control project involved the importation of the parasitoid *Pygostolus falcatus* (Nees). This species was one of the four parasitoids that had been previously studied; however, certain aspects of its biology were incomplete. Therefore, we conducted additional laboratory studies on temperature-dependent development and adult wasp longevity as influenced by temperature and availability of provisions. A field cage study was also conducted to determine host-parasitoid phenology under field conditions in North Dakota, and therefore, to what extent lack of synchrony between *P. falcatus* and the sweetclover weevil is a problem.

MATERIALS AND METHODS

In cooperation with Dave Hogg, University of Wisconsin, Madison, Wisconsin; Walter Goldstein, Michael Fields Agricultural Institute, East Troy, Wisconsin; and Anton Poiras, Institute of Zoology, Chisinau, Moldova Republic, the thelytokous braconid *P. falcatus* was collected from Barnaul, Siberia, to ensure a good climatic match to North Dakota. Offspring of the original wasps collected were used to establish a research colony at North Dakota State University. Wasps were reared at 20–25°C using sweetclover weevils as hosts. Adult sweetclover weevils for both the laboratory and field studies were field-collected and stored at 10°C and 16:8 L:D with bouquets of sweetclover until used. Rearings and/or dissections of the weevils did not reveal any natural parasitism. Specimens are deposited in the North Dakota State Insect Reference Collection, Lot #4790-1544.

Temperature-dependent Development. The exposure/holding cages for the temperature-dependent development study were half-pint paper cages with a nylon organdy top. Honey was streaked inside the cage to provide food for the parasitoids and a bouquet of yellow sweetclover cv. Norgold was provided in a water-filled vial inserted through the side of the cage to provide food for the hosts. Ten sweetclover weevils and 1–2 *P. falcatus* females were held for either:

1) 24 h at 10, 15, 20, 25 or 30±1°C and 16:8 L:D. The wasps were then removed from the cages and the weevils remained at the initial temperature.

2) 6 h at 25°C. The weevils were then transferred to holding cages at 10, 15, 20, 25 or 30±1°C and 16:8 L:D. This second method was used because we felt that little to no parasitism would take place after 24 h at 10 or 15°C, as both the wasps and weevils were lethargic at these temperatures.

Following exposure, cages of weevils were checked daily for host death and parasitoid cocoons. Cocoons were removed, placed in shell vials plugged with cotton, and checked daily for adult eclosion. Dead hosts and unemerged

cocoons were dissected. Remaining live hosts were dissected 21–82 d after exposure to the parasitoids, depending on the rearing temperature.

Adult Longevity and Temperature. To determine adult longevity at different temperatures, adult *P. falcatus*, < 24 h old, were set up individually in exposure cages as previously described. Hosts were periodically replaced with fresh weevils. The wasps were held at the same temperature as during their larval/pupal development, i.e. 10, 15, 20, 25 or 30°C, and checked daily for survival. As zero to few adult wasps were reared out at 10, 15 and 30°C, additional wasps for these treatments were obtained from the research colony.

Adult Longevity and Availability of Provisions. To determine adult longevity when provided different provisions, cocoons (within a day of eclosion) or adult wasps (< 24 h old) of *P. falcatus* were individually placed in half-pint paper containers at 25°C and 16:8 L:D. Adults were held under six conditions: 1) empty cage, 2) water only, 3) honey only, 4) honey and water, 5) honey, water and a plant, and 6) honey, water, plant and hosts. Water was provided in a 15 ml shell vial, plugged with cotton. Honey was streaked inside the cage and replaced as needed. Plants were single, non-flowering stems of yellow sweetclover cv. Norgold, inserted into a water vial. Hosts were 10 sweetclover weevil adults, which were periodically replaced. Adult wasps were examined daily for survival.

Statistics. Data for the number of days for development and longevity were analyzed with analysis of variance. Means were separated using Tukey's HSD test (SAS Institute 1990).

Using the developmental data, a developmental equation was constructed from the linear regression of percent development per day on temperature (SAS Institute 1990). The equation was of the form $y = a + bx$, where y = percent development per day and x = temperature. The lower developmental threshold is the x -intercept of the line. When the equation is solved for 'y' at any given temperature (x), the expected number of days for development at that temperature is equal to $100*y^{-1}$.

Degree-day requirements for development were calculated using the equation $K = D(T - t)$, where $D = 100*y^{-1}$ at temperature T , and t is the lower developmental threshold.

Field cage study. The study site was located on the Dave Podoll farm near Fullerton, North Dakota at approximately 46°16' N latitude. Field-collected sweetclover weevils were exposed to parasitoids in the laboratory. Both parasitized and unparasitized weevils were released into two Saran® screen field cages (Synthetic Industries, Gainesville, GA 30503), 1.8 m on a side, on 25 September 1996 and left to overwinter.

Jars of sugar-water with wicks were hung in the cages in spring 1997 as a supplement to any flowering plants. Periodic visits were made during the 1997 field season to see if and when adult *P. falcatus* were present in the cages and to collect caged sweetclover weevils for rearing and dissections. Sweetclover weevils were also collected outside the cages and around Fargo, North Dakota to track their phenology.

The presence of parasitoid cocoons and occurrence of wasp flights in the field was estimated using the degree-day model:

Daily degree-day accumulation = $(\text{Max. temp } [\leq t^U] + \text{Min. temp}) / 2 - t^L$, where t^U = upper developmental threshold and t^L = lower developmental threshold. Weather data was obtained from an automated weather station located 26 km SE of the study site.

RESULTS AND DISCUSSION

Temperature-dependent Development. No parasitoids emerged from hosts at 10° C; however, based on dissections, first instar larvae were present in the hemocoel and were assumed to be in diapause for two reasons. First, diapausing larvae have little to no gut content and exhibit no enlargement of the ventral lobe of the head as compared to developing first instars (Loan and Holdaway 1961b). Second, calculating from the developmental equation (described below), it would take *P. falcatus* approximately 175 d to complete its development at 10°C. As hosts at this temperature were not dissected until at least 82 d later, the parasitoid would have completed 47% of its development. Based on data from Loan and Holdaway (1961b), one would expect to find third or fourth instars instead of first instars.

Developmental rate decreased significantly from 15 to 25°C. However, development was not significantly different between 25 and 30°C (Table 1). Therefore, data for 30°C were not used in determining the developmental threshold. Wasps' wings at 15 and 30°C were sometimes crumpled, and prepupal/pupal survival was low (38%) at 30°C (Table 1). The optimal rearing temperature for the Siberian strain of *P. falcatus* appears to be 20–25°C.

Two developmental equations were calculated for *P. falcatus*: from egg deposition to spinning of the cocoon ($y = -3.067 + 0.388x$, $R^2 = 0.80$) and from egg deposition to adult eclosion ($y = -2.201 + 0.277x$, $R^2 = 0.89$). For both equations the lower developmental threshold was determined to be 7.9°C (Figure 1). This compares favorably with our experimental results in that some development occurred at 10° C before diapause intervened. The developmental rate plateaued at about 25°C (Figure 1); therefore, we made this value the upper developmental threshold. This is important to take into account when accumulating degree-days, as daytime summer temperatures regularly exceed 25°C. Development from egg to cocoon and from egg to adult for non-diapausing *P. falcatus* was calculated to take 258 and 363 degree-days, respectively. This compares favorably to data taken from Loan and Holdaway (1961b), whereby a Swedish strain of the parasitoid took an estimated 370 DD to develop from egg to adult.

Because *P. falcatus* overwinters as a diapausing first instar inside the host, the above degree-day values cannot be used to predict the first field appearance of the wasp. However, based on laboratory rearings, cocoons and adults were obtained 154–250 and 259–355 degree-days, respectively, after the removal of parasitized hosts from cold storage at 5°C.

Table 1. Development of *Pygostolus falcatus* at Different Temperatures with the Sweetclover Weevil, *Sitona cylindricollis*, as Host

Temperature (°C)	Developmental Time (Days, Mean±SD)					
	Egg to Cocoon (N)		Adult Emergence from Cocoon (N)		Egg to Adult (N)	
10	Only diapausing first instars recovered					
15	40.7 ± 3.5 (7)	a	16.8 ± 0.4 (5)	a	57.6 ± 4.0 (5)	a
20	19.8 ± 2.0 (10)	b	8.1 ± 0.6 (9)	b	28.1 ± 1.8 (9)	b
25	15.5 ± 2.3 (24)	c	6.4 ± 0.7 (21)	c	21.6 ± 1.9 (21)	c
30	15.2 ± 2.2 (13)	c	6.2 ± 0.8 (5)	c	21.0 ± 1.0 (5)	c

Means within columns followed by the same letter do not differ significantly ($P < 0.05$, Tukey's HSD test).

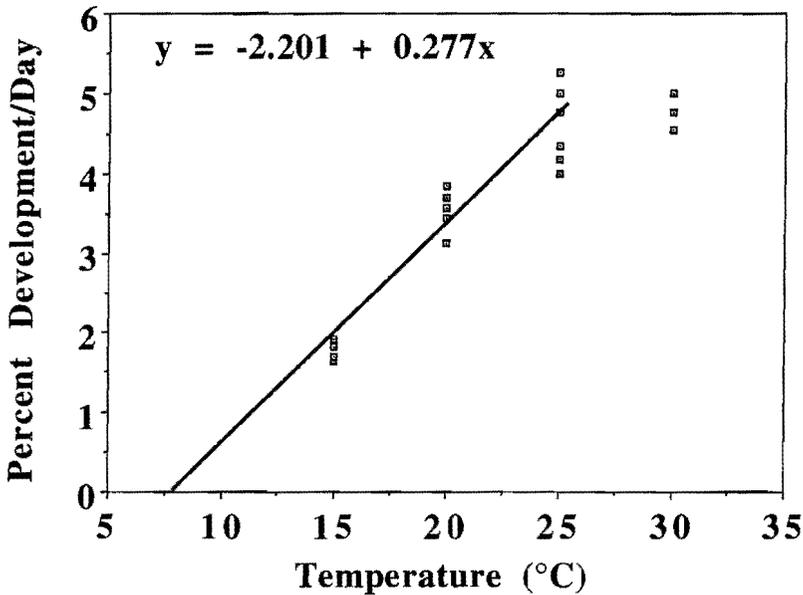


Figure 1. Developmental rate (egg to adult) for *Pygostolus falcatus* reared on sweetclover weevil hosts at various temperatures. Linear equation based on data from 15–25°C.

Adult Longevity and Temperature. At temperatures $\leq 15^\circ\text{C}$ when provided honey, water, sweetclover and hosts, adult *P. falcatus* survived a maximum of 39 d (Table 2). Longevity decreased significantly as temperatures increased to 25°C. Although not significantly different from the 25°C data, parasitoids were relatively shorter lived at a constant 30°C, regardless of the conditions of their larval development (Table 2).

Table 2. Longevity of Adult *Pygostolus falcatus* at Different Temperatures when Provided Honey, Water, Sweetclover and Sweetclover Weevil Hosts.

Temperature (°C)	Longevity (Days)		
	Mean \pm SD ^a (N)		Range
10 ^b	32.6 \pm 7.6 (5)	a	20–39
15 ^b	32.4 \pm 7.7 (5)	a	19–38
15	29.3 \pm 6.8 (3)	ab	24–37
20	22.1 \pm 7.1 (10)	b	8–30
25	12.3 \pm 2.0 (16)	c	9–16
30	6.0 \pm 2.0 (3)	c	4–8
30 ^b	8.3 \pm 0.5 (4)	c	8–9

^aMeans followed by the same letter do not differ significantly ($P < 0.05$, Tukey's HSD test).

^b Adult parasitoids obtained from a laboratory colony, 20°C. All other adults held at the same temperature as during their larval/pupal development.

Table 3. Longevity of Adult *Pygostolus falcatus* when Given Different Provisions, 25°C

Treatment	Longevity (Days)		
	Mean±SD ^a (N)		Range
Empty cage (no provisions)	2.1 ± 0.6 (10)	a	1-3
Water	2.4 ± 0.8 (10)	a	1-3
Honey	4.3 ± 1.5 (10)	ab	2-7
Honey & water	6.2 ± 1.3 (10)	b	4-8
Honey, water & plant ^b	11.0 ± 3.2 (9)	c	6-17
Honey, water, plant & hosts ^b	12.3 ± 2.0 (16)	c	9-16

^aMeans followed by the same letter do not differ significantly ($P < 0.05$, Tukey's HSD test).

^bPlant was a non-flowering stem of yellow sweetclover. Hosts were sweetclover weevils.

Adult Longevity and Availability of Provisions. Adult *P. falcatus* given no provisions or water only lived an average of 2 d (Table 3). Surprisingly, wasps provided honey or honey and water lived an additional 2-4 d only. Wasps provided honey, water and a plant (with or without hosts) lived significantly longer, 11-12 d on average, than adults from all other treatments (Table 3).

A moist carbohydrate source is necessary to prolong the parasitoid's life. The shortened life span of adult *P. falcatus* when provided only honey or honey and water was most likely due to the honey drying out in the cage. The addition of a plant apparently increased the humidity within the cage, as the honey did not dry out as fast. Also, the wasps were often seen resting under a leaf, possibly enjoying a cooler microclimate.

Field cage study. The life cycles of the sweetclover weevil and *P. falcatus* were determined by Bird (1947) and Loan and Holdaway (1961b), respectively. The sweetclover weevil is univoltine in North America. The adult weevil overwinters in sweetclover fields, and mating and oviposition occur from spring through mid-summer. By August almost all these overwintered weevils have died (Bird 1947). The larvae feed on the roots and root nodules, and the summer generation of adult weevils emerges beginning in late-July and peaking in mid-August. These weevils feed until cold weather begins (Bird 1947).

P. falcatus is bivoltine. The parasitoid overwinters as a first instar inside the adult host. Larval development is completed in the spring and the mature larva exits the host to spin a cocoon, killing the host (Loan and Holdaway 1961b). The first flight of the adult wasps occurs in late spring. Reproduction is parthenogenic and there is no pre-oviposition period. The female wasps parasitize other overwintered adult weevils, and a second flight of wasps takes place in mid-summer (Loan and Holdaway 1961b). Ideally, this second generation of *P. falcatus* parasitizes weevils of the summer generation and both host and parasitoid overwinter. It is at this point that synchrony between host and parasitoid is in question (Loan 1963b).

The Siberian strain of *P. falcatus* successfully overwintered in North Dakota. Based on field cage observations, host rearings and dissections, 1997 weather data and extrapolations of our degree-day model, a phenology for *P. falcatus* in southern North Dakota was constructed. The first appearance of cocoons and the first flight of wasps was estimated to begin May 31 and June 10, respectively. This is based on both rearings of caged hosts and laboratory rearings of parasitoids that had been artificially overwintered. The emer-

gence of adult wasps coincided with the first flowering of yellow sweetclover. If the wasps do indeed forage on sweetclover, synchrony with this nectar source is fairly good. Various crucifers are usually flowering in the field at this time, too. However, if no plant is flowering in the field, then these first generation wasps will need to forage elsewhere as they will not survive more than a few days without food (Dyer and Landis 1996). With adequate food resources, the adult wasps may survive through the latter part of June, as the average daily temperature is 20°C (see Table 2).

The second appearance of cocoons and adult wasps was estimated to begin 2 and 12 July, respectively. This was verified by the presence of wasps in the cages on 16 July. A few summer generation sweetclover weevils were present at this time in Fargo, but they did not become abundant until the end of July. At the field cage site, new weevil adults, collected outside the cages, did not appear until early August. The Fargo sites generally had thinner stands of sweetclover and better soil drainage. The field cage site, located near a slough, supported very dense stands of sweetclover and the soil remained moist throughout the summer. This combination of heavily shaded ground and moist conditions most likely resulted in cooler soil temperatures and therefore delayed larval development of the weevils as compared with the Fargo population. Cage collections made on 23 July yielded no wasps and only old, overwintered weevils, some of which contained diapausing first instar parasitoids. This incidence of diapause in overwintered hosts had been reported previously by Loan and Holdaway (1961b). They found 66–83% of the larvae entering diapause in July, with the result that a substantial proportion of larvae were eliminated by natural mortality of the overwintered hosts. Furthermore, adult *P. falcatus* will live no more than three weeks at this time of the year, as the average daily temperature is 22°C (see Table 2). Most wasps will therefore die prior to the main emergence of summer weevils in August; only a minor percentage of *P. falcatus* will oviposit in such hosts. Most *P. falcatus* will instead encounter overwintered weevils and waste their eggs on hosts that will die instead of overwintering, if the parasitoid larva enters diapause, or hosts that will die before the parasitoid larva completes its development.

Based on our developmental model and cage observations, it appears that the host-parasitoid synchrony in North Dakota is not good. Poor synchrony has been reported for various braconids and their adult hosts: *P. falcatus* and *Sitona* spp. in Sweden (Loan and Holdaway 1961b), *Centistes excrucians* Haliday and *S. scissifrons* Say (Loan 1963a) and *Microctonus sitonae* Mas. and *S. scissifrons* (Loan 1963b). Our model predicts a yearly crash in *P. falcatus*'s population in late summer, with the consequence that parasitism levels will start out every spring at low levels. Because much feeding damage and oviposition by the sweetclover weevil occurs in the spring, it is questionable whether a late season build-up in parasitism will prevent economic loss. Mass rearing the parasitoids for augmentative releases is not feasible at present due to difficulties rearing the host. Although techniques have been presented for culturing hosts (e.g. Hans 1961, Byers 1995), our success with artificial rearing and breaking diapause in artificially overwintered sweetclover weevils has been poor.

ACKNOWLEDGMENTS

We thank Anton Poiras, Institute of Zoology, Moldova Republic, for collection of the parasitoids; Dave Podoll for his cooperation and support of this project; and Ken Swan, USDA-ARS-BIRL, for his assistance in quarantine. This study was supported by the Northern Plains Sustainable Agriculture

Society, SARE/ACE project LNC93-62/ANC93-19 (in cooperation with D. Hogg, Univ. of Wisconsin) and North Dakota Agricultural Experiment Station Project No. 1544.

LITERATURE CITED

- Bird, R. D. 1947. The sweetclover weevil, *Sitona cylindricollis* Fahr. Can. Entomol. 79: 5-11.
- Brown, W. J. 1940. Notes on the American distribution of some species of Coleoptera common to the European and North American continent. Can. Entomol. 72: 65-78.
- Byers, R. A. 1995. Factors affecting rearing of clover root curculio (Coleoptera: Curculionidae) in cone containers. J. Econ. Entomol. 88: 407-414.
- Craig, C. H. 1978. Damage potential of the sweetclover weevil, *Sitona cylindricollis* (Coleoptera: Curculionidae), in the Canadian prairies. Can. Entomol. 110: 883-889.
- Dyer, L. E. and D. A. Landis. 1996. Effects of habitat, temperature, and sugar availability on longevity of *Eriborus terebrans* (Hymenoptera: Ichneumonidae). Environ. Entomol. 25: 1192-1201.
- Hans, H. 1961. Termination of diapause and continuous laboratory rearing of the sweet clover weevil, *Sitona cylindricollis* Fahr. Entomol. Exp. Appl. 4: 41-46.
- Loan, C. 1961. Introduction of European parasites of *Sitona* spp. for control of the sweetclover weevil, *Sitona cylindricollis*, in Canada. J. Econ. Entomol. 54: 1026-1031.
- Loan, C. C. 1963a. Observations on the biology of *Centistes excrucians* Haliday (Hymenoptera: Braconidae). Proc. Entomol. Soc. Ont. 94: 56-61.
- Loan, C. C. 1963b. The bionomics of *Sitona scissifrons* (Coleoptera: Curculionidae) and its parasite *Microctonus sitonae* (Hymenoptera: Braconidae). Ann. Entomol. Soc. Am. 56: 600-612.
- Loan, C. C. 1965. Status of *Pygostolus falcatus* as a parasite of *Sitona* spp. following releases in Manitoba and Ontario. J. Econ. Entomol. 58: 798-799.
- Loan, C. and F. G. Holdaway. 1961a. *Microctonus aethiops* (Nees) auctt. and *Perilitus rutilus* (Nees) (Hymenoptera: Braconidae), European parasites of *Sitona* weevils (Coleoptera: Curculionidae). Can. Entomol. 93: 1057-1079.
- Loan, C. and F. G. Holdaway. 1961b. *Pygostolus falcatus* (Nees) (Hymenoptera, Braconidae), a parasite of *Sitona* species (Coleoptera: Curculionidae). Bull. Entomol. Res. 52: 473-488.
- Munro, J. A., M. A. Leraas and W. D. Nostdahl. 1949. Biology and control of the sweet clover weevil. J. Econ. Entomol. 42: 318-321.
- Munro, J. A., R. L. Post and G. Badger. 1950. New facts about sweet clover weevil control. Annu. Rep. ND Agric. Exp. Stn., Bull. 356: 66.
- SAS Institute. 1990. "SAS/STAT User's Guide," Version 6, 4th ed. SAS Institute, Inc., Cary, NC.
- Smith, W. K. and H. J. Gorz. 1965. Sweetclover improvement. Adv. Agron. 17: 163-231.