# THERMAL BIOLOGY, OVERWINTERING, AND A DEGREE-DAY MODEL FOR THE ADULT EMERGENCE OF THE RED SUNFLOWER SEED WEEVIL (COLEOPTERA:

# CURCULIONIDAE)

A Thesis Submitted to the Graduate Faculty of the North Dakota State University of Agriculture and Applied Science

By

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In Partial Fulfillment of the Requirements for the Degree of MASTER OF SCIENCE

> Major Department: Entomology

> > April 2022

Fargo, North Dakota

# North Dakota State University Graduate School

# Title

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## ABSTRACT

Red sunflower seed weevil, *Smicronyx fulvus* LeConte, is a univoltine seed-feeding weevil that is a primary pest of cultivated sunflower, *Helianthus annuus*, in North America, especially within the upper Great Plains region. This pest spends its final larval stage overwintering in the soil before pupating in the summer and emerging as an adult to feed on sunflower pollen and lay eggs in developing achenes. Larval feeding can damage of up to 83% of seeds on a head. Growers typically manage *S. fulvus* by using insecticides; however, early planting dates have also been demonstrated to reduce weevil damage. It is important to understand how overwintering conditions impact larval development and develop a model to predict adult emergence. Larvae were found to overwinter in the soil at depths that are not subject to temperatures near their freezing point, and a degree-day model was created for emergence.

## ACKNOWLEDGMENTS

Thank you to my advisors Dr. Jarrad Prasifka and Dr. Deirdre Prischmann-Voldseth for their guidance throughout the duration of this project. A global pandemic is a memorable time to for everything, including graduate school, so thank you for taking on a student in this uncertain time. The assistance provided by my other committee members, Dr. Joe Rinehart and Dr. Mark Boetel, has been very beneficial and I am grateful for their insight. Thank you to Dr. Beth Ferguson, for your influence in the project and for the endless amounts of both work and life advice. I am thankful for the NDSU entomology department and faculty for continuing my education, whether that be virtually or in-person as well as the National Sunflower Association for funding this project. Finally, I would like to acknowledge all of the support I have received from friends and family, especially Dennis and Connie Olson and Kirsten Erickson. Thank you to all of the prior mentioned people for seeing this project through, every step of the way.

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# CHAPTER 1. OVERWINTERING BEHAVIOR AND THERMAL BIOLOGY OF THE RED SUNFLOWER SEED WEEVIL (COLEOPTERA: CURCULIONIDAE) 1.1. Introduction

### 1.1.1. Life History

The red sunflower seed weevil, *Smicronyx fulvus* LeConte (Coleoptera: Curculionidae), is a native seed-feeding pest of cultivated sunflower, *Helianthus annuus* L. (Asteraceae), and several wild *Helianthus* spp. Its range includes much of the central United States, from Texas to North Dakota and continuing into Canada (Anderson 1962, Oseto and Braness 1979, Charlet et al. 1992). A single generation occurs each year, with adult weevils typically emerging from soil and appearing in early summer on blooming wild or cultivated sunflowers (Charlet et al. 1997, Roseland et al. 1990). Female weevils require sunflower pollen for oogenesis (Korman and Oseto 1989) and lay eggs singly within achenes, ovipositing through the sunflower hull and onto developing seeds (Brewer 1991). After each larva ecloses, it feeds on a single seed for two to three weeks until its final (5<sup>th</sup>) instar (Oseto and Braness 1979). Larvae subsequently chew small exit holes in the hull, fall to the ground and burrow downward, into the soil to overwinter (Oseto and Charlet 1981).

Spending approximately 10 months in the soil presents a challenge for *S. fulvus* survival (Oseto and Charlet 1981). In North Dakota, life table analysis estimated that near 87% of total mortality occurred while *S. fulvus* were overwintering in the soil, and most of the mortality was not explained by predation, parasitism, or pathogens (Pinkham and Oseto 1988, Charlet and Seiler 1994). Because most cultivated sunflower production in the United States occurs in a region where soil freezes (USDA-NASS 2021), often for many months, this unexplained mortality may be related to cold stress. Mechanisms for *S. fulvus* to survive during winter may be

both behavioral and physiological. Since soil temperatures are more moderate with increasing depth (Márquez et al. 2016), moving deeper into the soil is a behavior that could limit acute or chronic effects of cold temperatures, thus increasing survival. Physiological mechanisms that limit insect mortality during the winter include tolerance and avoidance. Tolerance is an ability to survive the formation of ice crystals within body tissues, whereas avoidance is the accumulation of cryoprotectants to lower the freezing point of the organism's body, which is also known as supercooling (Block et al. 1990). For insects that survive freezing temperatures using cryoprotectants, the supercooling point (SCP) is the temperature in which ice formation can occur spontaneously in the body, ultimately causing mortality with continued cold temperatures (Rozsypal and Košťál 2018).

## 1.1.2. Overwintering Strategy of S. fulvus

Prior research on overwintering of *S. fulvus* includes both behavioral and physiological aspects. After dropping out of the achenes, the larvae exhibit little horizontal movement (Oseto and Charlet 1981). Gednalske and Walgenbach (1984) found that soil cores collected in spring had 93% of the larvae within 7.5 cm of the soil surface and no larvae below 15 cm. However, because those authors only collected samples once per year, it is unclear whether larvae move vertically between their initial entry into the soil in early fall and pupation in the following spring. Prior research on weevils collected in North Dakota suggested that *S. fulvus* is physiologically freeze-avoidant under field conditions, with a supercooling point near  $-24^{\circ}$ C (Rojas et al. 1991); however, when larvae were in contact with water, the supercooling capacity of *S. fulvus* was reduced by as much as 10°C (Rojas et al. 1992).

# **1.1.3.** Project Objectives

Though prior research gives key insights into *S. fulvus* ' overwintering, some results appear incomplete or in conflict. For example, overwintering mortality is very high (Pinkham and Oseto 1988), but the causes of this mortality are unclear; additionally, supercooling by larvae appears to protect against temperatures lower than those larvae are exposed to in the soil ( $\geq$  -17°C, NDAWN 2022). This implies that overwintering mortality is not due to freezing. A series of three experiments was designed to characterize overwintering behaviors and cold tolerance of *S. fulvus* larvae. The first objective was to re-evaluate its SCP under two different types of larval storage conditions (i.e., indoors with constant cold and outdoors with fluctuating conditions). The second objective was to assess vertical movement of *S. fulvus* larvae between the onset of overwintering in the fall through pupation in late spring. The third goal was to evaluate whether exposure to realistic soil temperatures (i.e., warmer than estimated SCP) could explain observed patterns of overwintering mortality.

# 1.2. Materials and Methods

# **1.2.1. Weevil Collection**

Larvae used in laboratory and field experiments were reared from field-collected adult *S*. *fulvus* in order to obtain parasitoid-free larvae, and to standardize larval size and health. Adult *S*. *fulvus* were collected from wild sunflower, *H. annuus*, growing in roadside ditches in agricultural areas near Strasburg, North Dakota in July and August of 2019–2021 by visually inspecting wild sunflower heads for beetles with red-brown elytra. Sunflower heads with adult *S. fulvus* were cut directly under the head using pruning shears and placed into  $28 \times 28$  cm cotton cloth bags, which were placed in a handheld cooler and transported to a laboratory in Fargo, ND where they were removed from bags and placed in handmade plywood cages (0.036 m<sup>3</sup>) with polyethylene mesh

for ventilation. Cut *H. annuus* heads were placed into cages and held at room temperature for one to four days until they could be processed. Processing involved gently shaking each sunflower head to encourage adult *S. fulvus* to drop out, then collecting fallen weevils using an aspirator, after which 30 or 50 adults were transferred to 50–mL plastic centrifuge tubes along with one to three sunflower ray petals. Within 24 h of larvae being collected, tubes were sealed with ventilated caps and transported in coolers to a sunflower field near Casselton, ND with self-pollinating sunflower lines (i.e., HA 89, HA 434, HA 456; Jan 2006; Miller et al. 2004, 2006). Adults were shaken out of the tubes onto the disc florets of blooming sunflower heads at growth stage 5.3–5.6 (Schneiter and Miller 1981), with one tube of weevils added per head. Sunflower heads were covered with a woven polyethylene bag (43 × 52 cm, Delnet<sup>TM</sup>, Schweitzer-Mauduit International, Alpharetta, GA) to restrict weevil movement, but allow light penetration and air flow. Bags were secured at the base of the head using plastic zip ties.

When sunflower heads reached physiological maturity (i.e., growth stage R9; Schneiter and Miller 1981), stalks were cut just below the sunflower head using pruning shears, and heads were subsequently transported to the laboratory. The polyethylene bags were removed and a thin plastic stake was pushed through the back of each sunflower head, making sure to not to disturb the seeds and larvae. Heads were then suspended on racks so that each hung approximately 3 cm above the bottom of a plastic tub ( $46 \times 33 \times 13$  cm). Heads were left undisturbed at room temperature ( $22 \pm 2^{\circ}$ C) and standard indoor humidity ( $55 \pm 5\%$ ) for five to seven days to collect mature larvae exiting the achenes. Tubs were checked daily with contents emptied into a sieve (size 14, 1.40 mm opening, Thermo Fisher Scientific, Hampton, NH). The sieve helped exclude dried plant material that had fallen into the tub, but the small openings also required larvae to actively crawl downward in order to be collected for later use. Larvae were further cleaned by

transferring them onto damp paper towels and rolling larvae back and forth to remove plant fine debris, after which they were counted and weighed using an analytical balance (Secura125-1S, Sartorius, Göttingen, Germany). Depending on the experiment, cleaned larvae were either placed outdoors the day they were collected or placed into plastic bags partially filled with moistened sand (17.7 cm × 18.8 cm, Ziploc, S. C. Johnson, Racine, WI). These self-sealing plastic bags contained a mixture of autoclaved sand and distilled water as described in Prasifka et al. (2015; 9:1 sand to water ratio), with 300 larvae ( $\approx$  1300 mg) added per bag. Bags containing larvae were stored indoors in the laboratory at room temperature for five to seven days until larval collection was complete.

# **1.2.2. Supercooling Point**

To evaluate the ability of overwintering *S. fulvus* to tolerate freezing temperatures, the larval supercooling points were measured over the winters of 2019–2020 and 2020–2021, with each year using a different larval cold storage method. In 2019–2020, larvae were collected into bags with moistened sand as previously described and held in cold storage in a refrigerator at  $\approx$  4°C until needed. For 2020–2021, larvae were placed into small cylindrical plastic containers (r = 5 cm, h = 13 cm) with brass cloth (0.305 mm wire diameter, 0.541 mm opening) on the bottom for drainage that had been filled with field soil from their overwintering location at nearby agricultural research plots (North Dakota State University's Main Station) in Fargo, ND. In both winters, containers with larvae were removed from storage for supercooling point measurement in November, January, April and June, with containers holding the larvae sorted through using a sieve (size 14, 1.40 mm opening, Thermo Fisher Scientific, Hampton, NH) and larvae selected at random.

After removal from overwintering media (sand or soil), each larva was placed inside a 1.5–mL microcentrifuge tube. A thermal wire was attached to the outside of each tube on the bottom using laboratory tape. The thermal wires were connected to a multi-thermocouple reader and logger that used a microcontroller circuit board (Arduino.cc) with a thermocouple breakout, a type K thermocouple, and an attached display to observe real-time temperature readings. To gradually cool *S. fulvus* larvae, weevil-filled microcentrifuge tubes were placed into Corning® CoolCell LX<sup>TM</sup> freezing containers (432138, Corning, Corning, NY), and then transferred to a –80°C freezer (Stirling Ultracold, Athens, OH). The freezing containers limited the rate at which the temperature dropped to 2.5±0.5°C per minute. Larvae were cooled until the temperature was below –30°C, which was past the expected freezing point (Rojas et al. 1991). This process was repeated with groups of four tubes at a time until data were collected for at least 20 individuals per testing period.

Time-series temperature data were read (40 data points per minute) from the memory card attached to the circuit board and graphed to identify a rapid increase in temperature that interrupted the gradual cooling. The sharp increase is from heat released (i.e., heat of fusion) as larvae freeze (Block et al. 1990). The supercooling point for each larva was noted as the temperature immediately before the increase (Figure 1).



**Figure 1**: Freezing of an individual *S. fulvus* larva cooled by  $2.5\pm0.5$ °C per minute, with the supercooling point spike around -21°C. SCP is noted with an arrow.

# **1.2.3.** Overwintering Depth

The depths at which *S. fulvus* larvae are found determines the range of temperatures weevils are exposed to while overwintering. 'Depth tubes' were constructed to determine how far below the soil surface *S. fulvus* larvae overwinter. Tubes (n = 16) were made by using white polyvinyl chloride (PVC) pipe (outside diameter 8.9 cm, inside diameter 7.5 cm) cut into 40–cm long sections. Holes were drilled into standard PVC pipe covers and mesh fabric placed on the inside to allow for drainage of excess moisture. These modified caps were then pushed onto the bottom of each tube to prevent the soil from shifting and weevils being lost from the depth tubes during collection.

Depth tubes were moved to research plots (North Dakota State University's Main Station) in Fargo, ND during late summer. Each tube was partially buried with the capped end 30 cm below ground level, 60 cm apart from neighboring tubes, and then filled with the soil removed when each tube was buried. Water was poured inside and around the tubes and left to dry, and then soil was added if needed until soil within the tubes was level and soil moisture appeared equal to the surrounding area. On September 14, 2020, the soil surface inside each tube was lightly scored with a dinner fork and 300 *S. fulvus* larvae were poured onto soil surface in each tube. To prevent loss of larvae by birds or other predators, tubes were then covered with a woven polyethylene bag (43 × 52 cm, Delnet<sup>TM</sup>, Schweitzer-Mauduit International, Alpharetta, GA), which was secured to the tube with rubber bands. Larvae typically burrowed into the soil within a few hours after being placed on soil surface, allowing bags to be removed the following day.

Depth tubes were destructively sampled four times during the 2020–2021 larval overwintering period (November 18, January 15, April 20, and June 21) to investigate whether *S. fulvus* overwintering depth changed throughout the year. Four randomly selected tubes were destructively sampled on each date by breaking up the ground around the tube with a spade and pulling the entire tube out of the ground. Once tubes were removed from the soil, each tube was cut lengthwise using a table saw. The cut section of PVC was then removed to access the soil within. Sections of soil were measured out and separated every 2 cm starting from the top of the soil column at the soil surface. Each section was washed and sifted through a sieve (size 14, 1.40 mm opening, Thermo Fisher Scientific, Hampton, NH) to separate *S. fulvus* larvae from soil. Numbers of live larvae in each section were counted and recorded on the same day the tubes were pulled from the ground. Any dead, hardened larvae were discarded. After counting live

larvae in each section on November 18, subsequent counts combined all soil below 8 cm due to the low numbers of larvae at increasing depth (0–2 cm, 2–4, 4–6, 6–8, and 8–30 (i.e., the bottom of the tube). Overwintering depth evaluation was repeated during the 2021–2022 larval overwintering period, but with fewer (n = 8) depth tubes and just two sample times (January 24 and April 4) due to the interest in mortality between those dates in the previous year. To aid in interpretation of results, soil temperature data at 5 cm below the soil surface were downloaded for the nearest weather station (Fargo, ND; NDAWN 2022).

#### **1.2.4. Cold Bath Exposure**

An additional study in 2021–2022 was carried out to test whether week-long exposure to realistic soil temperatures (i.e., warmer than estimated SCP) could explain observed patterns of *S. fulvus* overwintering mortality. Bags (n = 12) containing 300 larvae each in moistened sand as previously described were stored at 4°C in September 2021. Excluding the control (constant 4°C) treatment, groups of three bags were later placed into a cold bath of 40% ethylene glycol (AP07R-40-A11B, PolyScience, Niles, IL) for one week. Exposure of larvae in bags to temperatures of  $-4^{\circ}$ C (November 1–8),  $-8^{\circ}$ C (November 8–15), and  $-12^{\circ}$ C (November 15–22) was followed by a return to storage at 4°C. This storage temperature was selected because *S. fulvus* larvae can be stored at 4°C for extended periods of time without negative impacts on survival (Prasifka et al. 2015). Two months after each group of *S. fulvus* larvae was returned to 4°C, larvae were sifted through sieves and the number of survivors counted for each bag. Surviving larvae from the 4°C controls were counted on the same day as those subjected to the  $-8^{\circ}$ C treatment.

#### **1.2.5. Statistical Analysis**

All statistical analyses were conducted using SAS (SAS Institute Inc., 2013). A two-way analysis of variance (ANOVA; PROC GLM) tested whether *S. fulvus* supercooling points were affected by differing storage conditions in 2019 and 2020 (year), time within year (month), and the year  $\times$  month interaction. Tests for the effect of month were made separately for each year because the initial analysis generated a significant year  $\times$  month effect.

Overwintering depth data for *S. fulvus* larvae were collected as counts within ranges (e.g., 56 larvae from 4–6 cm), requiring a non-parametric analysis. Counts were summarized in five depth categories (i.e., 0–2, 2–4, 4–6, 6–8, and >8 cm), and data from all replicate columns were pooled. Resulting depth data for *S. fulvus* removed from tubes in 2020–2021 were analyzed using a Chi-squared goodness-of-fit test (PROC FREQ) with the depths as columns and dates (November, January, April, June) as rows. Data for 2021–2022 were analyzed in the same way, except there were only two sample dates (January and April).

Data on *S. fulvus* survival following week-long exposure to realistic soil temperatures in the cold bath were analyzed by a one-way ANOVA (PROC GLM). Post-ANOVA comparisons were made only between the control (i.e., constant  $4^{\circ}$ C) and the other three treatments (-4, -8, and -12°C) using Dunnett's test.

# 1.3. Results

## **1.3.1. Supercooling Point**

Supercooling points for individual *S. fulvus* larvae tested over two years were typically between -20 and  $-23^{\circ}$ C (67 % of n = 197 larvae), though values as low as  $-27.0^{\circ}$ C and as high as  $-17.2^{\circ}$ C appeared to be valid (i.e., void of severe outliers or errors). The two-way ANOVA showed significant effects of year (*F* = 9.52; df = 1, 189; *P* = 0.002) and a year × month interaction (F = 5.73; df = 3, 189; P < 0.001) on the temperature at which larvae froze. Across all months, mean SCP for *S. fulvus* larvae stored at a constant 4°C indoors during 2019–2020 (-21.09°C) were about 1°C warmer than larvae overwintering outdoors under natural conditions in 2020–2021 (-22.09°C). When a potential effect of month was examined separately in each year, a significant effect was found for 2020–2021 when *S. fulvus* were overwintered outdoors (F = 5.41; df = 3, 189; P = 0.001); this month effect (and the year × month interaction) reflects a loss of cold tolerance between the first three months of testing ( $\approx -22.6$ °C in November, January, and April), and the final sample date (-20.9°C in June) (Table 1).

**Table 1**: Supercooling points (mean  $\pm$  SE) for *S. fulvus* larvae overwintered indoors at 4°C (2019–2020) or outdoors (2020–2021) with larvae removed for testing four times each year (month).

Year	Month	Supercooling Point (Mean ± SE)	Sample size
2019–2020	November	$-21.13 \pm 0.23$	33
	January	$-21.20 \pm 0.19$	28
	April	$-21.59 \pm 0.18$	32
	June	$-22.50 \pm 0.31$	20
2020-2021	November	$-22.60 \pm 0.48$	20
	January	$-22.60 \pm 0.40$	21
	April	$-22.68 \pm 0.37$	23
	June	$-20.93 \pm 0.48$	20

# **1.3.2.** Overwintering Depth

In 2020–2021, most larvae (91.5%) removed from depth tubes were found within 6 cm of the soil surface. However, the distributions of larvae by depth were not equal across sample dates ( $\chi^2 = 904.70$ ; df = 12; P < 0.001), with larvae appearing to move 1–2 cm deeper between each of the first three sampling dates (November to January, January to April; Table 2). When tubes were removed during June, *S. fulvus* recovered were a mix of larvae (72%) and pupae (28%), indicating the depths at which larvae were found were representative of where pupation

occurred. Survival of larvae added to depth tubes in September (n = 300 per tube) also declined sharply between January (73.83%) and April (34.50%) 2021. Soil temperature data (5 cm depth) from the nearest weather station show February 10–18 included the lowest soil temperatures during the 2020–2021 overwintering period (daily minima between –8 and –12°C); minimum soil temperatures during the six-year period (winters from 2016–2021) were consistently lowest in January and February, but minimum daily temperatures varied across years from as high as  $-1^{\circ}$ C to as low as  $-17^{\circ}$ C (Figure 2). Average temperatures also differed over the same six years (Figure 3). In the winter of 2021–22, tubes were brought up and checked in January and April. As in the previous year, the distribution of larvae differed between sampling dates ( $\chi^2 = 102.10$ ; df = 4; *P* < 0.001), with apparent downward movement between January and April. However, no significant mortality was apparent between January and April 2022 (Table 3).

**Table 2**: Number of larvae recovered from each depth interval during each month. Counts of live *S. fulvus* recovered from soil-filled tubes at depth intervals during 2020–2021. Four tubes were removed on each date. Each tube initially contained 300 larvae added in September 2020 (=1,200 larvae).

		S. fulvus	Count by	Depth			
Month	0–2 cm	2–4 cm	4–6 cm	6–8 cm	> 8 cm	Total	% of Original Total
				CIII			Iotal
Nov 20	354	505	56	19	5	939	78.25%
Jan 21	59	409	297	106	15	886	73.83%
Apr 21	0	107	258	44	5	414	34.50%
Jun 21	9	82	138	15	1	245*	27.22%

\* June 2021 sample included only three tubes due to an accidental loss, and a 28% of *S. fulvus* recovered had begun pupation.

**Table 3**: Number of larvae recovered from each depth interval during each month. Counts of live *S. fulvus* recovered from soil-filled tubes at depth intervals during 2021–2022. On each date four tubes were removed. Each tube initially contained 300 larvae added in September 2020 (=1,200 larvae).

S. fulvus Count by Depth							
Month	0–2 cm	2–4 cm	4–6 cm	6–8 cm	> 8 cm	Total	% of Original Total
Jan 22	24	132	295	73	42	566	47.17%
Apr 22	6	82	370	214	118	790	65.83%



**Figure 2**: Minimum soil temperatures by month at 5 cm depth in Fargo, North Dakota from 2017 to 2021 (NDAWN 2022).



**Figure 3**: Average soil temperatures by month at 5 cm depth in Fargo, North Dakota from 2017 to 2021 (NDAWN 2022).

# **1.3.3. Cold Bath Exposure**

An ANOVA showed significant differences (F = 34.33; df = 3; P < 0.0001) in survival of overwintering *S. fulvus* larvae among treatments including a control (constant 4°C) and 4°C interrupted with week-long exposure to one of three realistic soil temperatures (-4, -8, and -12°C) in a cold bath. Post-ANOVA comparisons (Dunnett's test) showed the -8°C and -12°C treatments produced significantly greater mortality than the 4°C control (Table 4).

Treatment	% Survival Mean ± SE
Control	$92.58\pm6.17$
-4°C	$96.42 \pm 1.00$
-8°C	$71.75\pm7.09$
-12°C	$67.67 \pm 2.92$

Table 4: Survival of S. fulvus larvae after week-long exposure to colder temperatures.

### 1.4. Discussion

Previous research with *S. fulvus* provides information that naturally lead to additional questions. For example, life-table analysis conducted in North Dakota (Pinkham and Oseto 1988) suggested very high (87%) larval mortality in *S. fulvus* while overwintering in the soil, and total annual mortality so high (95%) that population growth may be zero or negative; the cause of overwintering mortality is mostly unknown, and the total mortality conflicts with *S. fulvus*' role as the most damaging seed-feeding insect in cultivated sunflowers. Freezing soil temperatures seem a likely explanation for overwintering mortality, but supercooling point tests suggested that *S. fulvus* larvae appear capable of tolerating temperatures 5–10°C lower (–24°C; Rojas et al. 1991) than they are likely to experience in the soil in the coldest of winters in the northern Great Plains region. Additional experiments in 2019–2021 provided some clarification on freeze-avoidance (supercooling) by *S. fulvus*, their overwintering depths (including movement and survival), and their ability to survive prolonged exposure to realistic soil temperatures in the northern Great Plains.

Estimates of *S. fulvus* supercooling points used larval storage under controlled (4°C in 2019–2020) and natural (outdoors in 2020–2021) conditions, with SCP means of -21°C and -22°C, respectively. Though the two years differed statistically from each other and appeared to differ from the -24°C estimate of Rojas et al. (1991), the 1-3°C range of means may not be

biologically meaningful because all SCP estimates are several degrees colder than temperatures *S. fulvus* larvae experience in soil under natural conditions. However, additional work by Rojas et al. (1992) noted freeze-avoidance by *S. fulvus* larvae can change by as much as 10°C because of contact with external moisture. The mechanism that explains the reduced supercooling ability observed by Rojas et al. (1992) is inoculative freezing, when ice formed on the outside of the insects' body may enter through any of several small openings in contact with external moisture (Rozsypal and Košťál 2018). Due to the potential for inoculative freezing, Rojas et al. (1992) recommended that estimates of survival be made with and without external moisture for insects that overwinter in moist habitats. However, it is unclear if inoculative freezing alone explains observed overwintering mortality for *S. fulvus*.

Placing *S. fulvus* larvae into tubes of field-collected soil and allowing them to move naturally within the soil provided two general insights on overwintering depth. First, most larvae were found near (2–6 cm depth) the soil surface during winter. Second, larvae appeared to move  $\approx 1-2$  cm deeper between November and January, and again between January and April. These observations agree with (but are more detailed than) data from Gednalske and Walgenbach (1984), which included soil collected in 7.5-cm increments and showed that, during spring, most (> 90%) or larvae were found at 0–7.5 cm depth. Though the tubes used to assess overwintering depth in 2020–2021 were not meant to study mortality, a decline in the numbers of live weevils recovered between January and April 2021 was notable. Examination of soil temperatures showed a distinctly colder week-long period in February with daily minima at 5 cm depth of  $-8^{\circ}$ C or colder, suggesting a duration and range of temperatures that cause mortality for *S. fulvus*. Given the observed mortality between January and April 2021, it is also possible that the apparent larval movement between those samples was actually mortality of larvae nearer the soil

surface, where temperatures would be even colder than at 5 cm depth. But data from 2021-2022 suggest apparent downward movement was real, as there was no evidence of significant weevil mortality between January and April 2022 (when soil temperatures never were lower than  $-7^{\circ}$ C).

Cold bath exposure treatments were informed both by observed mortality in the overwintering depth experiment and soil temperature trends over the period that most *S. fulvus* mortality occurred (January–April 2021). Although data on supercooling points and soil temperatures suggest that some larvae may die from freezing, the cold bath confirmed that temperatures as high as –8°C can produce *S. fulvus* mortality if the period of cold extends over several days. Difference in soil temperatures at 5 cm below the surface between 2021 and 2022 suggests that in field conditions –8°C may also be causing some mortality.

Results from supercooling point, overwintering depth, and cold bath studies together suggest that mortality of overwintering *S. fulvus* is likely caused by continuous exposure to low temperatures that may not be cold enough to freeze larvae. A similar conclusion was reached for overwintering adults of the cabbage seedpod weevil, *Ceutorhynchus obstrictus* (Marsham) (Coleoptera: Curculionidae), in Canada; though its estimated SCP is -7°C, increasing cumulative exposure to  $-5^{\circ}$ C (in relatively dry [14% moisture] soil that may prevent inoculative freezing) decreased weevil survival in the laboratory, and cumulative time < 0°C in the field also was closely related to survival in field microcosms (Cárcamo et al. 2009). Similar predictions of *S. fulvus* overwinter survival may not be possible without additional research because of complicating effects of ground cover (including snow), soil moisture, and other factors.

One result that relates to overall management of sunflowers is the shallow overwintering by *S. fulvus*, which suggests farming operations like tillage (Vencill and Banks 1994) oreven herbicide incorporation (Harris et al. 1969) may have an added benefit to suppressing weevil

populations. Gednalske and Walgenbach (1984) also observed some forms of tillage may help limit survival of overwintering *S. fulvus*, but the effects of tillage for other crop-insect combinations appear inconsistent (Stinner and House 1990), and in recent years more sunflowers are grown in areas where no-till farming is common. As a result, a combination of integrated pest management strategies (e.g., cultural practices, monitoring, economic thresholds, insecticides) are likely needed to limit crop losses to *S. fulvus*.

# CHAPTER 2. DEVELOPMENT OF A DEGREE-DAY MODEL FOR ADULT EMERGENCE OF THE RED SUNFLOWER SEED WEEVIL, (COLEOPTERA: CURCULIONIDAE)

#### **2.1. Introduction**

#### 2.1.1. Pest Status and Crop Damage

Among many insects that feed on cultivated sunflower, *Helianthus annuus* L. (Asteraceae), in North America (Rogers 1992), recent crop survey data show the red sunflower seed weevil, *Smicronyx fulvus* LeConte (Coleoptera: Curculionidae), to be the most destructive seed-feeding pest (Buetow et al. 2020). This univoltine weevil can be found on *Helianthus* spp. across much of North America east of the Pacific mountain system (Anderson 1962), but infestations of >100 *S. fulvus* adults per head are sometimes observed in North Dakota and South Dakota, which lead the United States in sunflower production (USDA-NASS 2021). After larvae complete overwintering in the soil, adult weevils emerge in early summer, and females feed, mate, and oviposit through sunflower hulls onto the developing sunflower seeds (Korman and Oseto 1989, Brewer 1991).

An infested sunflower achene (= the pericarp or 'hull' + the seed contained inside) typically contains only a single larva (Oseto and Braness 1979), which has been estimated to consume 31% of the seed mass and reduce oil content by 25% (Oseto and Braness 1980). Peng and Brewer (1995) gave similar estimates of crop damage by each larva (i.e., 26% of seed mass and 30% seed oil lost in an infested seed), but also provided additional information that allowed for calculation of economic injury levels. In oilseed sunflowers, recent (2021) recommendations call for insecticides to be applied if 3–5 weevils are present per head. Confection sunflowers, which are sold as a snack food or for other premium use, effectively do not have an economic

threshold (ET) for *S. fulvus*; typically, >1% insect-damaged seed is unacceptable and confection sunflowers are grown under contracts that require the use of insecticides during sunflower bloom (John Sandbakken, pers. comm.).

### 2.1.2. Insecticide-based Management Options

Management practices for S. fulvus are intended to control adult weevils and prevent oviposition. Insecticide evaluations in small plots were described by Satterthwait (1945), and later by Oseto and Braness (1980) and Gednalske and Walgenbach (1984b). In field-scale plantings (often 16-64 hectares or more), insecticides are usually applied by plane. Aerial insecticide applications are effective, but have recently relied on products incorporating chlorpyrifos, an organophosphate insecticide that was revoked for food crop use by the Environmental Protection Agency in 2021 (US-EPA 2022); the elimination of chlorpyrifos means that insecticide options are largely limited to several pyrethroids (Knodel et al. 2022). Insecticidal seed treatments do not appear to provide any value for S. fulvus management (Bredeson and Lundgren 2015). Several field trials have suggested host plant resistance to S. *fulvus* is possible (Brewer and Charlet 1995, Gao and Brewer 1998, Charlet et al. 2010). Subsequently, DeGreef et al. (2020) reported on release of an inbred line that reduced weevil feeding damage by  $\approx$  70%. Shallow overwintering by *S. fulvus* larvae means that tillage can reduce survival and adult emergence (Gednalske and Walgenbach 1984a), though large areas of sunflower production involve no-till or limited tillage to conserve soil moisture (NRCS 2019). Planting early-maturing sunflowers on the perimeter of fields and applying insecticides to the field edges suggests this type of trap cropping can be effective (Brewer and Schmidt 1995). Trials in South Dakota (Gednalske and Walgenbach 1983) and North Dakota (Oseto et al. 1987)

indicate that a combination of early planting and early-maturing hybrids can limit the overlap of sunflower bloom and *S. fulvus* populations.

Though many complementary tools and tactics are available for integrated management of *S. fulvus*, discussions with sunflower growers and stakeholders suggest economic thresholds and insecticide applications are the only tools routinely used. The reasons for limited use of other viable strategies are not clear, but may be related to perceived difficulty in use. For example, early planting as a strategy relies on the facts that weevils are not attracted to sunflower once pollen is no longer available (i.e., when bloom is complete; Oseto and Braness 1979) and that weevil emergence in an area may extend over several weeks (Gednalske 1983). However, unlike some other sunflower insect pests (Charlet 1987, Merrill et al. 2010), no model for adult emergence of *S. fulvus* is available. A degree-day model for *S. fulvus* emergence and locationspecific maturity data for current sunflower hybrids in North Dakota (https://www.ag.ndsu.edu/ varietytrials/sunflower) or South Dakota (https://extension.sdstate.edu/south-dakota-sunflowerhybrid-performance-trials) could be used to find combinations that minimize overlap between adult weevil populations and sunflower bloom.

#### 2.1.3. Objectives

Development of a degree-day model for adult emergence of *S. fulvus* would encourage use of early planting or early-maturing hybrids as tools to complement the use of economic thresholds and insecticides. Progress towards a useful model was made in three steps. First, new and existing (Prasifka et al. 2015) data on *S. fulvus* emergence under laboratory conditions were used to estimate the lower threshold for post-diapause development and cumulative degree-days to 50% weevil emergence. Second, types of available temperature data from a state-wide weather network (North Dakota Agricultural Weather Network or 'NDAWN') were evaluated as

resources to describe thermal conditions experienced by *S. fulvus* larvae and pupae prior to adult emergence. Third, observations of *S. fulvus* emergence in field plots were compared to the laboratory-based model of adult emergence.

### 2.2. Methods

## 2.2.1. Smicronyx fulvus Collection and Rearing

To obtain *S. fulvus* for laboratory and field emergence tests, larvae were reared from field-collected adults. Adult weevils were collected from infested heads of wild sunflower, *H. annuus*, near Strasburg, North Dakota during late July and early August of 2019–2021. Weevils from the wild sunflowers were counted and transported to research plots in Casselton, ND, at which point groups of 30 or 50 weevils were confined onto heads of cultivated sunflower using woven polyethylene bags and plastic zip ties. After 30–40 d when all larvae had reached maturity; (Oseto and Braness 1979), heads were returned to the laboratory and suspended over plastic tubs. Larvae exiting sunflower achenes were collected daily and placed into small bags of moistened, autoclaved sand. Larvae from these bags, which varied in quantities of sand and *S. fulvus* larvae as needed, were used for laboratory or field tests of weevil emergence. Additional details on the collection and handling of *S. fulvus* adults and larvae were as described in Chapter 1.

#### 2.2.2. Developmental Threshold and Degree-day Model

To estimate a minimum temperature for post-diapause development (i.e., a lower threshold) and the cumulative number of degree-days required for 50% of *S. fulvus* adults to emerge, larvae were overwintered outdoors and moved indoors in early spring to emerge under a range of constant temperatures. On September 14, 2020, 12 self-sealing plastic bags containing 500 *S. fulvus* larvae in 750 g of moistened sand were moved to a research plot at North Dakota

State University's Main Station in Fargo, ND (46.897301, -96.814568). The contents of bags were emptied into cylindrical plastic containers (r = 5 cm, h = 13 cm) containing soil from the plot and buried at ground level, with 2 m between neighboring containers. Drainage of excess soil water was permitted by brass cloth (0.305 mm wire diameter, 0.541 mm opening) fused to the bottom of each container.

Containers were taken out of the field April 6, 2021. After being covered with lids ventilated with brass cloth (to allow air exchange but prevent adults from leaving a container) and randomly placed into incubators (MyTemp<sup>TM</sup> Mini, Model H2200-HC, Benchmark Scientific, Sayreville, NJ) set at 10, 14, 18, or 22°C (n = 3 containers per temperature). Set temperatures were also verified using a thermocouple on the inside of each incubator. Containers were checked twice weekly until the first adult *S. fulvus* had emerged, after which time containers in each incubator were checked daily. Distilled water was added as needed to each container to prevent soil from drying out. Emerged adults were counted and collected using an aspirator. After *S. fulvus* emergence decreased to <1 adult per day, emergence at 25°C (Prasifka et al. 2015; following 365 d overwintering at 6°C) were also used to supplement the observations from 2021.

A lower threshold for post-diapause development of *S. fulvus* larvae was estimated using a least variability method (Arnold 1959). Specifically, a range of potential developmental thresholds (2°C to 12°C) were used to calculate total degree-days to first emergence and 50% emergence under the different temperature treatments. Because temperatures were constant, each day added a fixed number of degree-days to a cumulative total. For example, at 22°C, a developmental threshold of 2°C would add 20 degree-days for each day leading up to (first or

50%) adult emergence. The correct lower threshold was considered to be the temperature for which accumulated degree-days under different temperature treatments had the lowest % coefficient of variation ( $100 \times [$ standard deviation / mean]).

# 2.2.3. Soil Temperature Data

Optimal use of a degree-day model for adult emergence of *S. fulvus* depends on qualitycontrolled soil temperature data that covers the primary sunflower production areas of North Dakota and South Dakota. The public weather networks of NDAWN (North Dakota's Agricultural Weather Network) and South Dakota's Mesonet provide similar sources of soil data that can be used to calculate degree-days for predicting *S. fulvus* emergence. Research on overwintering depths of *S. fulvus* larvae (Gednalske and Walgenbach 1984b and Chapter 1) show most larvae within  $\approx$  6 cm of the soil surface. Soil temperature data from NDAWN that are likely similar to what *S. fulvus* larvae experience are available from (a) a thermocouple 5 cm below soil with unmanaged vegetation (hereafter '5 cm turf'), (b) a thermocouple 10 cm below soil with unmanaged vegetation ('10 cm turf'), and (c) a thermistor placed 10 cm beneath soil kept free of plant material ('10 cm bare'). Note that NDAWN refers to (a) and (b) as "deep soil temperatures" and (c) is listed as "bare soil temp" online.

To assess the quality and consistency of NDAWN soil temperature data for predicting *S*. *fulvus* emergence, hourly data (5 cm turf and 10 cm turf) or daily means (10 cm bare) were downloaded for 2017–2021 for the 'Fargo NW' site (located 120 m east of plots used to overwinter *S*. *fulvus* larvae). The single-sine method was used to estimate degree-days (Roltsch et al. 1999). Based on the lower threshold indicated by emergence under controlled conditions, degree-days (°C) for each day were calculated as ((daily maximum – daily minimum)/2) – 5 (5 cm turf and 10 cm turf) or daily mean – 5 (10 cm bare), with any value < 0 recorded as zero

degree-days. Because *S. fulvus* adults are often first noted in the second half of July in North Dakota, cumulative degree-days from the start of the year, January 1, to July 15 were calculated for each year (2017–2021) and sensor (5 cm turf, 10 cm turf, 10 cm bare) combination. An analysis of variance (SAS Institute Inc., 2013; PROC GLM) was used to test for effects of year and sensor type on cumulative degree-days from January 1 to July 15.

## **2.2.4. Emergence and Predicted Emergence**

To assess emergence of *S. fulvus* adults under natural conditions and compare to the degree-day model developed under controlled conditions, larvae were released to overwinter in the soil of a research plot on North Dakota State University's Main Station in Fargo, ND. For 2019–2020, a total of six sites in the plot were spaced 2 m apart into an area between widely-spaced plantings of *Silphium integrifolium* Michx., but with soil at and around each site maintained free of volunteer *S. integrifolium* or other weeds. At each site, the top half of a plastic bucket (d = 18 cm, h = 17.5 cm) was pushed downward 4–5 cm into the soil and marked with stakes on the north and south edges. On September 24, 2019, a plastic bag containing 500 *S. fulvus* larvae in 750 g of moistened sand was emptied into each bucket, and covered with mesh fabric to prevent predation by birds or other animals. One week after larvae were introduced, buckets were removed and larvae were left to overwinter.

On June 15, 2020, an emergence trap made from the top half of a larger (28 cm diameter) plastic bucket was centered over the stakes placed at each site in the fall and pushed downward 4–5 cm into the soil. A circular opening (18 cm diameter) was cut into the plastic lid of each trap to allow air, light, and precipitation to pass through, but confine any emerging weevils using a section of mesh fabric (70-mesh, 0.21 mm openings) held in place where the bucket and lid snap together (Figure 4). Each trap was checked twice weekly to monitor for the first emerged adult.

Once the first adult *S. fulvus* was found, adults were collected daily from each trap by using an aspirator. Trap monitoring was continued for two weeks after the last emerged adult was observed.



**Figure 4**: Emergence trap for the outdoor emergence of *S. fulvus* from the side (A), the underside of the trap (B), and the top of the trap (C). Mesh fabric (70-mesh, 0.21 mm openings) was secured by the lid of the bucket.

The release of weevil larvae and monitoring of adults was repeated during the 2020–2021 overwintering and growing season, but with two significant modifications. First, the number of *S. fulvus* released at each site on September 16, 2020, was increased from 500 to 800 larvae. Second, the number of sites was increased to nine, with four of the sites maintained free of vegetation as in 2019–2020, and spring wheat (ND VitPro, 15 cm rows, 3.7 million seeds / ha) planted in and around five of the sites on April 28, 2021.

Weevil emergence data were pooled across similar traps, creating three cumulative weevil emergence curves (2020 [bare soil], 2021 [bare soil], 2021 [spring wheat]). To test whether observed *S. fulvus* emergence differed from that predicted by the degree-day model, Kolmogorov-Smirnov tests (SAS Institute Inc., 2013; PROC NPAR1WAY) were used to compare emergence under controlled conditions (18°C) to accumulated degree-days from the 5cm turf and 10-cm bare sensors located 120 m east of the plots.

### 2.3. Results

## 2.3.1. Developmental Threshold and Degree-day Model

Average soil temperatures (bare soil, 10 cm depth) for the week preceding April 6, 2021 was 3°C, when tubs containing overwintering *S. fulvus* larvae in soil were removed from field plots and transferred to environmental chambers in the laboratory. Larvae held in a chamber set to 22°C emerged between 57 and 100 days later, while those at 18°C emerged between days 76 and 106. No emergence of larvae at 14°C or 10°C occurred during 116 days of observation. When emergence data from 22°C and 18°C treatments were combined with similar data (larvae overwintered for 365 d at 6°C and moved into 25°C) obtained from Prasifka et al. (2015), a lower developmental threshold of 5°C minimized variation (lowered CV) in accumulated degreedays among treatments (Arnold 1959). No adults emerged in the 10°C and 14°C incubators. The same lower threshold appeared optimal whether degrees-days were calculated to first emergence or 50% emergence (Figure 5). Across the three temperatures using the 5°C lower threshold, first emergence of *S. fulvus* adults occurred after 969–980 degree-days (starting January 1) and 50% emergence from 1156–1190 degree-days.



**Figure 5**: Coefficient of variation at lower developmental threshold temperatures. The lower developmental threshold with the lowest coefficient of variation for both first and 50% emergence of adult *S. fulvus* is at 5°C.

### **2.3.2. Soil Temperature Data**

Using the 5°C lower threshold, accumulated degree-days from January 1 to July 15 in each of five years (2017–2021) were significantly affected by year (F = 17.75, df = 4, P < 0.001) and the type of soil temperature data (F = 15.18, df = 2, P = 0.002). A thermocouple 10 cm below soil with unmanaged vegetation (10 cm turf) accumulated fewer degree-days than a similar sensor at 5 cm (5 cm turf) or a thermistor placed 10 cm beneath soil kept free of plant material (10 cm bare; Table 5). Average soil temperatures during July (5 cm turf or 10 cm bare) of about 23°C, suggest the data from the 10 cm turf thermistor would predict *S. fulvus* emergence about six days later than either of the other measures. Though mean accumulated degree-days from January 1 to July 15 were statistically similar for 5 cm turf and 10 cm bare sensors over 2017–2021, degree-day totals for each year could produce estimates for emergence that differed by as much as four days (i.e., 75 degree-days) depending on whether 5 cm turf and 10 cm bare soil data were used.

**Table 5**: Accumulated degree-days to July 15 from 2017 to 2021 for *S. fulvus* using a lower threshold of 5°C in three different soil temperature data sets; 5 cm below turf, 10 cm below bare soil, and 10 cm below turf.

Year	5 cm turf	10 cm bare soil	10 cm turf
2017	990	936	866
2018	1103	1155	972
2019	872	948	767
2020	1064	1025	995
2021	1079	1120	981
Average	1022	1037	916

### **2.3.3. Emergence and Predicted Emergence**

In 2020, adult *S. fulvus* emerged into traps in field plots between 6–28 July, reaching 50% emergence on 13 July. Adults recovered in traps (n = 357) represented 12% of the weevil larvae placed into the plots in fall 2019. When cumulative weevil emergence in the field during 2020 was plotted against accumulated degree-days and compared to emergence in the laboratory (18°C data), the distribution of weevil emergence in the field differed from the degree-day model, with actual emergence several days in advance of prediction; this result was true whether field degree-days used 5 cm turf (Kolmorgorov-Smirnov D = 0.51, P < 0.001) or 10 cm bare soil (D = 0.64, P < 0.001) data (Figure 6).



**Figure 6**: Cumulative emergence in 2020, comparing degree-days for the 2020 outdoor emergence of adult *S. fulvus* between two different outdoor soil temperature data sets, 5 cm below turf and 10 cm below bare soil, and the indoor laboratory model. Each data point represents a 24-hour period.

During 2021, for traps placed over bare soil (as in 2020), weevils emerged from July 4 to July 26, reaching 50% emergence on July 9. Traps placed in locations where spring wheat was planted (April 28) showed slightly delayed emergence, with adults collected from July 7 to July 27, and reaching 50% emergence on July 16. Recovery of adults from weevil larvae placed in the plots in fall 2020 was substantially lower for traps over bare soil (n = 62,  $\approx 2\%$ ) and spring wheat (n = 19, <1%) than in the previous year. Cumulative weevil emergence into traps over bare soil (D = 0.58, P < 0.001) temperature data. Observed cumulative emergence of weevils where spring

wheat was planted differed from the degree-day model using temperatures from 5 cm turf (D = 0.39, P = 0.010), but was similar to predicted emergence using 10 cm bare soil (D = 0.26, P = 0.192) temperature data (Figure 7).



**Figure 7**: Cumulative emergence in 2021, comparing degree-days using a lower threshold of 5°C for outdoor emergence of adult *S. fulvus* in spring wheat between two soil temperature data sets, 5 cm below turf and 10 cm below bare soil, and the indoor laboratory model. Each data point represents a 24-hour period.

# 2.4. Discussion

Emergence data for *S. fulvus* under controlled conditions (18, 22, and 25°C) produced a degree-day model with a lower developmental threshold of 5°C and estimated  $\approx$  970 degree-days

to first emergence and  $\approx$  1160 degree-days to 50% emergence. A comparison of available soil

temperature data showed the accumulated degree-days at 5 cm below turf and 10 cm beneath

bare soil were similar over five years (2017–2021), but greater than the cumulative degree-days at 10 cm below turf. Observed emergence of weevils in field plots occurred several days earlier than predicted (using either 5 cm turf or 10 cm bare soil temperature data) when soil was kept free of vegetation, but was similar to predicted emergence when plots were shaded by spring wheat.

Degree-day models for other insects often incorporate more extensive datasets (year  $\times$ location combinations) than that used to predict S. fulvus emergence, but have similar challenges in both model development and use. For example, a model for emergence of the soiloverwintering rice water weevil (Lissorhoptrus oryzophilus Kuschel) was developed using 14 yr of light trap captures; validation of predicted emergence sometimes showed very little (1 or 2 d) error, but one year emergence occurred 7 d before the model prediction (Zou et al. 2004). Merrill et al. (2010) monitored emergence of adult sunflower stem weevils, Cylindrocopturus adspersus (LeConte), which overwinter as larvae within sunflower stalks, across 17 location-years. Due to differences in how emergence was estimated at different locations, they developed two separate emergence models that were later combined to create a 'consensus' model. Skinner et al. (2004) developed degree-day models for emergence of two chrysomelid flea beetles (Aphthona spp.) introduced as weed biological control agents. They estimated lower developmental thresholds and emergence dates by measuring emergence under controlled conditions (six constant temperatures). Validation of model predictions generally showed a lack of bias, but predicted and actual dates of first emergence could differ by  $\approx 20$  d, with error attributed to a wide range of climatic and environmental variation among sites in Montana, Minnesota and North Dakota (Skinner et al. 2004). A degree-day model for S. fulvus is intended to be used as a tool to predict adult emergence in areas where S. fulvus is a significant pest. The value of the model is

influenced by whether available soil temperature data (e.g., NDAWN [North Dakota's Agricultural Weather Network], South Dakota's Mesonet) reflect the conditions experienced by *S. fulvus* in the soil in spring and early summer. For weevils emerging from vegetation-free plots in 2020 and 2021, degree-day accumulations using soil temperature data from NDAWN predicted emergence several days later than what was observed. This bias likely occurred because ground cover (for 5 cm turf) and increasing soil depth (10 cm bare soil) delay warming of soil in spring (Pierson and Wight 1991). However, predicted and observed emergence were similar when spring wheat was planted into the plots in 2021. Examination of crop data layers (https://nassgeodata.gmu.edu/CropScape/) shows that in central South Dakota (where *S. fulvus* pest problems are currently most challenging), sunflowers are most likely to be followed with spring wheat or another spring-planted crop, a pattern that suggests 5 cm turf or 10 cm bare soil temperature data may be adequate for predicting *S. fulvus* emergence.

Survival of overwintering *S. fulvus* varied considerably between the two years of field observations. Much lower survival in 2021 (< 2%) compared to 2020 ( $\approx$  12%) could be considered a concern for modeling cumulative weevil emergence. However, the contrast in weevil survival might more appropriately be assessed as a confirmation of prior research. For example, at a site  $\approx$  40 km from the location used in 2020–2021, Pinkham and Oseto (1988) observed 87% mortality of larvae and pupae in the soil, which is virtually identical to overwintering survival in this study in 2020. It is also clear from Chapter 1 that lower survival in 2021 may be attributable to a period of very low temperatures in February 2021; though observed soil temperatures (between -8 and -12°C) were not cold enough to freeze overwintering *S. fulvus*, prolonged exposure to those temperatures causes significant mortality. For plots into which spring wheat was planted in 2021, adult emergence was further reduced, but shallow

overwintering by *S. fulvus* and narrow rows (15 cm spacing) of spring wheat seemed to produce mortality similar to that observed under conventional tillage by Gednalske and Walgenbach (1984). In combination, substantially lower winter temperatures and more frequent use of conventional tillage may help explain why recent populations of *S. fulvus* adults in North Dakota are more than ten-fold lower than some areas in South Dakota (NDSU 2021). Though prolonged low soil temperatures and disturbance in the spring seem to explain overwintering survival for *S. fulvus*, other factors including diet (Berthiaume et al. 2020), high temperatures prior to overwintering (Han and Bauce 1998), temperature fluctuations (Rozsypal and Košťál 2018), soil moisture (Zheng et al. 2013), or natural enemies such as predators, parasitoids, or pathogens (Pinkham and Oseto 1987) can cause variable overwintering survival. Differences in survival of overwintering larvae to adult emergence between this study and Prasifka et al. (2015), may be related to mortality from natural enemies, which were certainly killed when Prasifka et al. (2015) autoclaved sand used for overwintering larvae.

Several improvements to the degree-day model seem possible. Incorporation of additional locations and years may provide the most important potential enhancement. Although collection of new data on *S. fulvus* emergence in South Dakota could be justified, detailed observations on emergence by Gednalske (1983) could also be used to add more years and locations. However, soil temperature data for South Dakota's Mesonet are limited to 2015 and later years. If ambient temperature data could be related to accumulated degree-days in the soil, data from Gednalske (1983) could be added, which would allow prediction of emergence in any areas where local soil temperature data are not available. Also, if the degree-day model is being used to avoid weevil damage (versus strictly predicting *S. fulvus* emergence), data on reproductive development in female weevils suggest an additional 5 d after emergence should be

added to allow for oocyte development (Korman and Oseto 1989) when assessing the risk to a developing sunflower crop. Lastly, a web-based tool that estimates overlap of weevil populations and sunflower bloom (e.g., a web-based tool that allows user inputs), would support grower use of early planting and early-maturing sunflowers, which have received renewed interest as chemical options for insect management become more limited.

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