First Record and Ecological Observations on the Blister Beetle *Tricrania sanguinipennis* (Say, 1824) (Coleoptera: Meloidae) in Manitoba.

Deanna Dodgson[1] and Robert E. Wrigley[2]

[1]ddodgson@shaw.ca, [2]Corresponding author, robertwrigley@mts.net

INTRODUCTION

On March 19, 2021, Dodgson (DD) was searching for early-season arthropods in the Portage Sandhills Wildlife Management Area, 18 km south of Portage la Prairie (in south-central Manitoba), when she noticed several red and black meloid beetles crawling along three sandy trails in aspen-oak savanna. She was able to identify the species that evening as *Tricrania sanguinipennis*, which had never been reported previously in Manitoba (Figure 1). She contacted Wrigley (RW) who followed up by checking the sites the following day and found large numbers of live and dead beetles along the trails. In the subsequent days, both authors continued to make observations along these and other trails.

The discovery of a population of *T. sanguinipennis* in southern Manitoba came as a surprise, since the nearest records for this distinctive and flightless species are Minnetonka, Hennepin County, Minnesota (Bugguide.com) and Cedar Creek Ecosystem Science Reserve (https://www.inaturalist.org), both in the Minneapolis area, 650 km to the southeast.

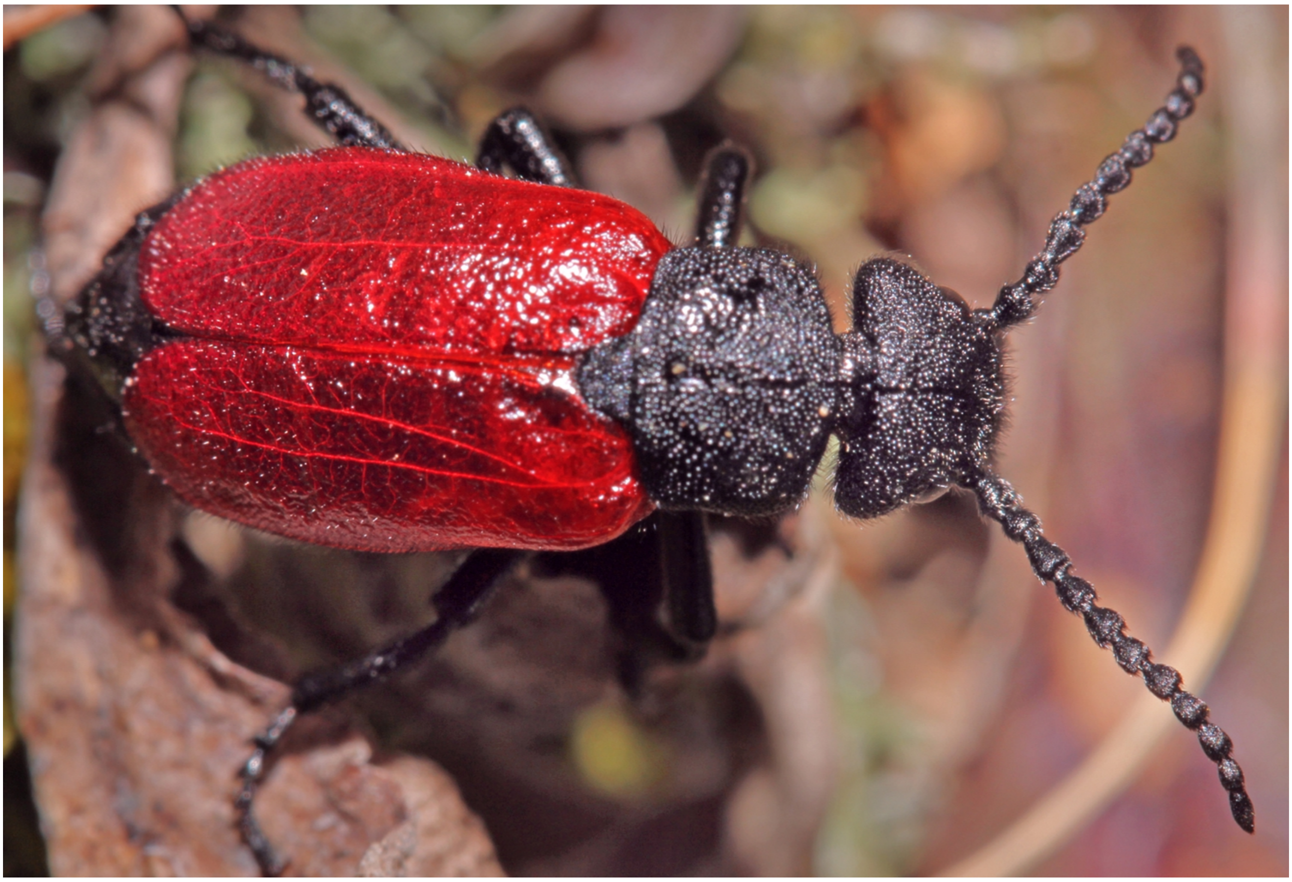

Figure 1a. Dorsal view of *Tricrania sanguinipennis* photographed at Rock Bridge Memorial State Park near Columbia, Boone Co., Missouri, on February 22, 2012. (Jon Rapp). The elytra of Manitoba specimens were typically reddish-brown.

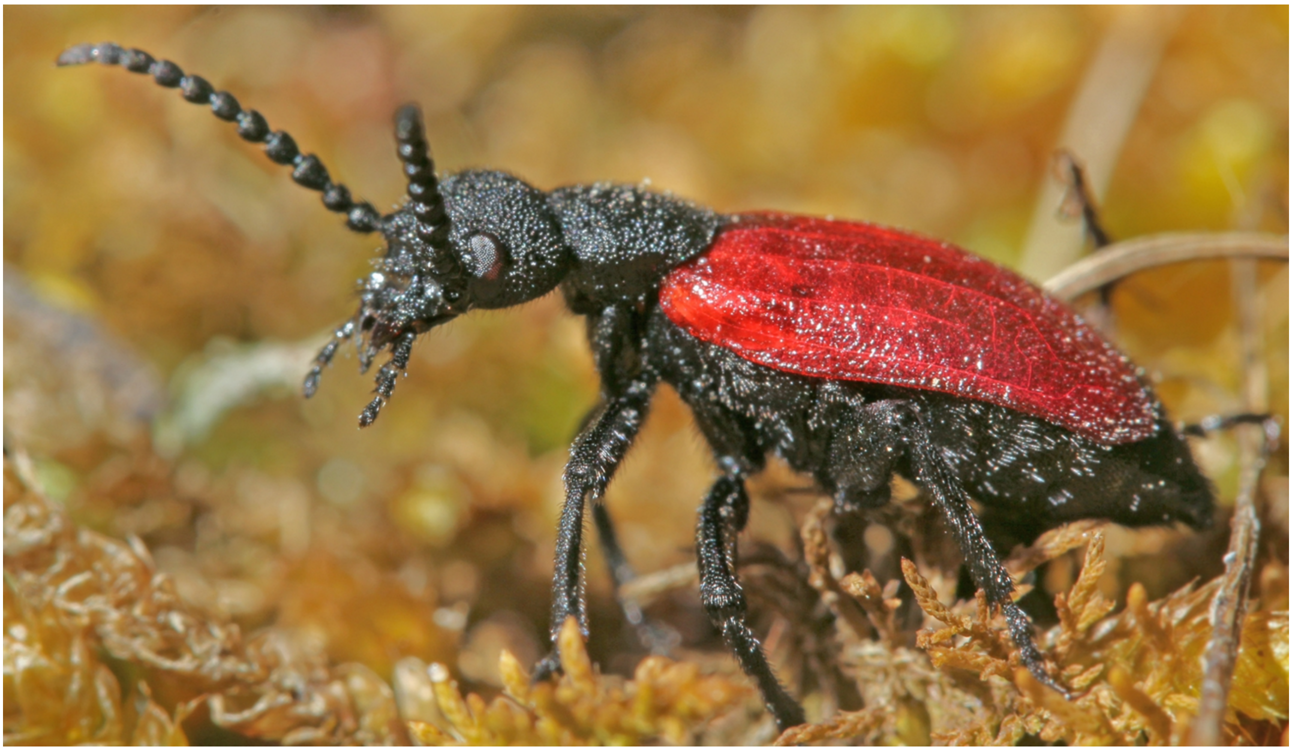

Figure 1b. Lateral view of *Tricrania sanguinipennis.* (Accreditation as per Fig. 1a)

Say described this species in 1834 (as *Horia sanguinipennis*); “body black, short, robust, elytra sanguineous.” He based it on a specimen from Pennsylvania, stating: “This species is an interesting addition to the catalogue of North American insects, as it is the only one of its very limited genus, yet found here. It must be rare, only a single specimen having yet occurred.”

T. sanguinipennis ranges in size from 7.5–15 mm (our measurements) and has a black body with red, rugose elytra, which fades to brownish red after death. The elytra cover all or almost all of the abdomen. The flight wings are entirely absent. The body is densely covered in short black, erect setae, becoming sparse on the elytra. The head and pronotum are heavily punctured. The antennae are short and submoniliform. The scientific name of this flightless blister beetle, *T. sanguinipennis,* well describes this species, in reference to its triangular head and flexible, blood-red elytra. No common name exists, so we refer to it as the Eastern Red-winged Blister Beetle, distinct from its flighted western counterpart, *Tricrania stansburyi* (Haldeman, 1852), the only two species in the genus. A third species, *Tricrania murrayi* Leconte 1860, was synonymized with *Tricrania stansburyi* (Cline and Huether 2011).

T. sanguinipennis has been recorded throughout eastern North America from southern Quebec and Ontario (Bousquet et al. 2013), and New Brunswick (Webster et al. 2020), south to Florida, and from Minnesota south to Kansas and Texas (Evans 2014, Bugguide.net). Both eastern and western species of *Tricrania* occur in Texas, but their ranges do not overlap there or elsewhere (Quinn 2015). Torchio and Bosch (1992) reported on the biology of the western *Tricrania stansburyi.*

T. sanguinipennis is an obligate brood parasitoid, having lost the ability to acquire provisions for its young, and therefore relies on an appropriate host to serve as a food provider (Litman 2019). Its triungulin larva attacks bees in the families Colletidae, Megachilidae and Apidae (Parker and Boving 1924, Cline and Huether 2011).

METHODS

Beetles and their habitats were photographed by both of us, and RW collected 325 specimens (live and dead) by hand while walking along sandy trails and blowouts. Ruts in the trail formed by motorcycles and ATVs acted as traps, since the beetles had difficulty climbing out. A number were also crushed by the wheels.

While an early spring and drought conditions existed in 2021, the record snowfall and rain events into late spring in 2022 resulted in delayed warm weather and serious flooding in southern Manitoba. Many roads in the study area became impassable. Consequently, we were unable to access some research sites as frequently as in the previous year, but were still able to determine early emergence of the population.

In determining sex ratios, males were identified by their longer and significantly stouter antennae compared with those of the females (Figure 2). Four specimens were collected alive by RW on March 29, 2021, and maintained in a terrarium for observation. Series of specimens are being donated by RW to the JB Wallis-RE Roughley Museum of Entomology, Manitoba Museum, Canadian National Collection of Insects, Arachnids and Nematodes, Canadian Museum of Nature, and the Royal Ontario Museum. Additional specimens are available to other museums upon request.

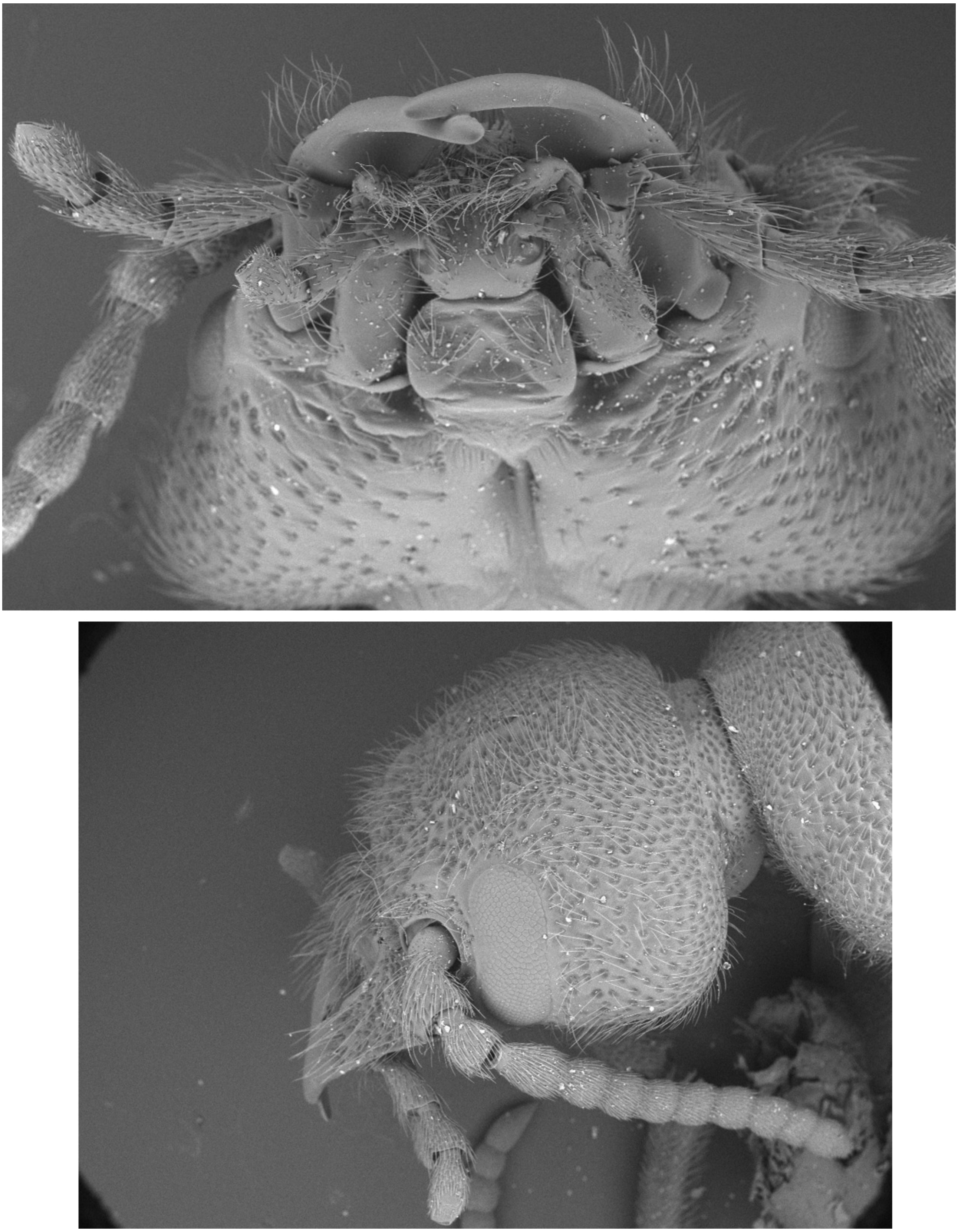

Figure 2. Scanning Electron Microscope images of a male (top) and female (bottom) *Tricrania sanguinipennis*. (Erwin Huebner)

FIELD SITES
The Portage Sandhills Wildlife Management Area lies on the eastern edge of a massive delta of sand that formed where the ancient Assiniboine River flowed into Glacial Lake Agassiz. This 1600-ha, hilly area is crisscrossed by countless kilometres of motorized-vehicle trails (Figure 3) and blowouts (Figure 4), within a larger sandy region of mixed-grass prairie, aspen-oak savanna, and agricultural lands. Motorized vehicles are banned in this wildlife management area from March 1 to November 30 (mainly the snow-free period); however off-road vehicles were present on many of the observation days. The frequent passage of these vehicles over time has kept prairie vegetation from encroaching onto the sandy trails. The trail-side slope below the drop-off of the prairie sod also offered ideal, well-drained habitat for the burrows of *Colletes inaequalis* Say bees, the local hosts of *T. sanguinipennis*.

Dominant plants in Manitoba sandhills include *Bouteloua gracilis*, *B. curtipendula*, *Prunus virginiana*, *Opuntia fragilis*, *Euphorbia virgata*, *Juniperus horizontalis*, *Rosa acicularis*, *Arctostaphylos uva-ursi*, *Quercus macrocarpa*, *Populus tremuloides*, *P. balsamifera* and *Picea glauca*.

We accessed the main trails and blowouts on Road 54N at the following sites: 49.79849/-98.2169
49.798476/-98.236328
49.798284/-98.239994, and
49.798241/-98.254973.
DD visited these sites in April of the two previous years, and from May 17 to 24 over three years, without observing this blister beetle. Other nearby locations in south-central Manitoba where we found beetles in 2021 were:
49.79387/-98.24202 Road 54N
49.88681/-98.26582 Road 60N
49.88744/-98.34106 Road 60N
49.88784/-98.29788 Road 60N
49.82870/-98.27392 Road 36W
49.69381/-98.97643, 10.2 km N, 6.6 km W Holland. This sandy area is 55 km west of the Portage Sandhills, and is the eastern edge of the Carberry Sandhills.

Additional distributional records found in 2022:
49.79792/-98.60742W; 3.2 km S, 0.8 km E Rossendale, isolated sandy hill on Road 54N (north side of the Assiniboine River)

49.739494/-99.416616, 18 km from the CFB Shilo Garrison, 14.5 km SW Carberry (provided by Sherry Punak-Murphy, and currently the northwestern peripheral record for the species).

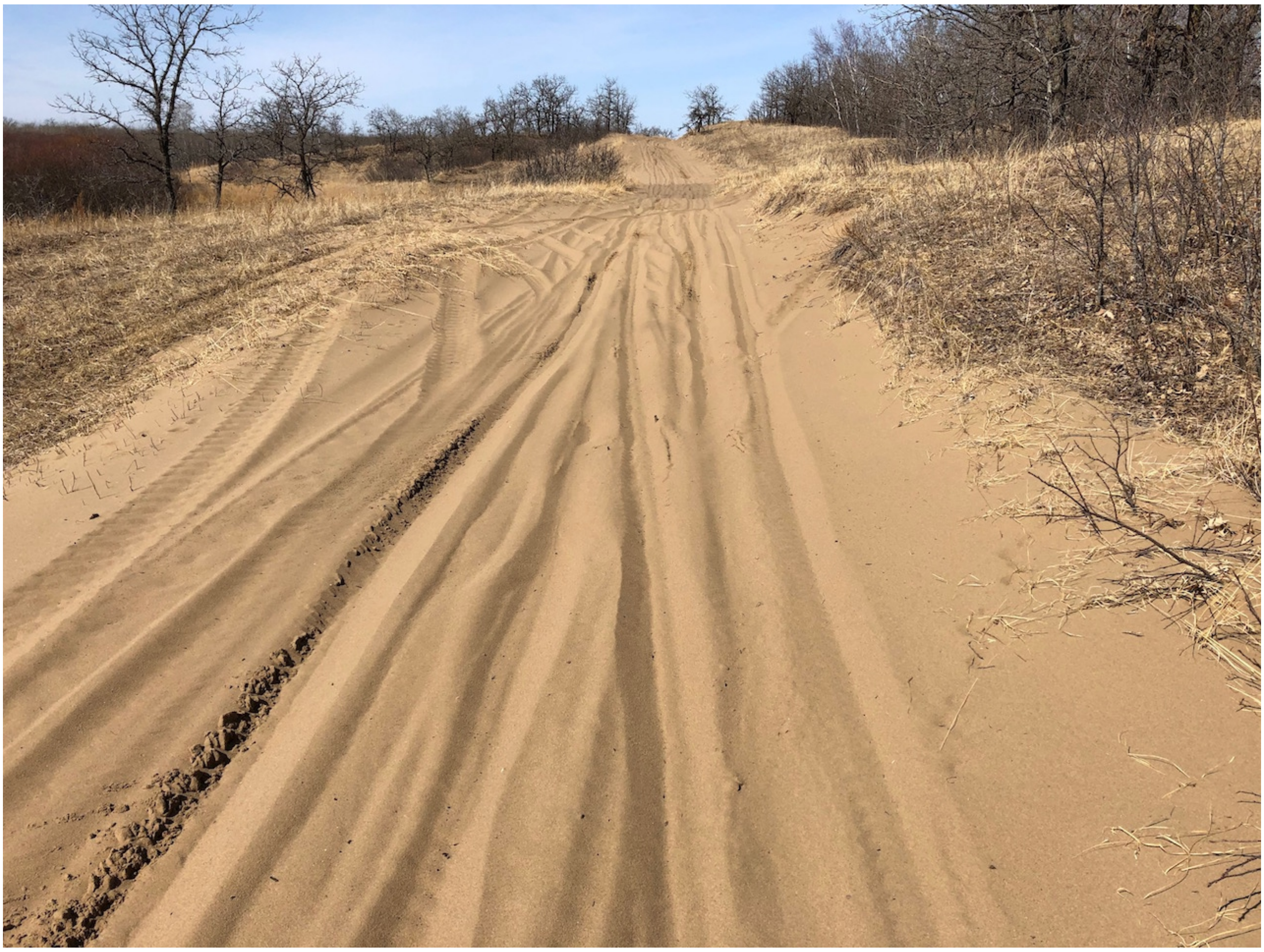

Figure 3. The trail in the Portage Sandhills Wildlife Management Area where most of the *Tricrania sanguinipennis* specimens were collected. (Robert Wrigley)

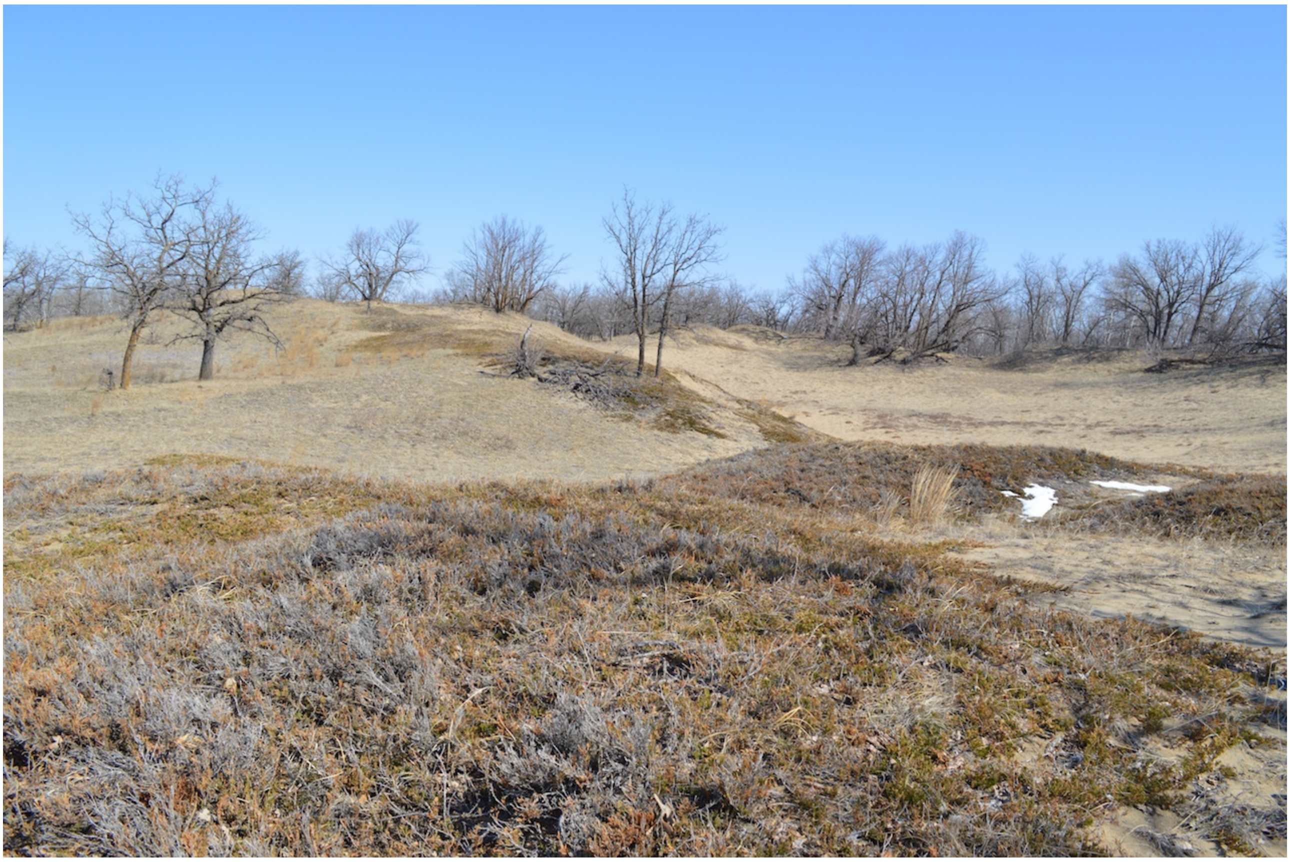

Figure 4. Sandy blowout in the Portage Sandhills Wildlife Management Area in which *Tricrania sanguinipennis* was common. (Robert Wrigley)

RESULTS

The synchrony of emergence and of the brief period of activity of both the beetle and its bee host is a critical factor in the beetle's successful reproduction and continued survival of the population. Consequently, the following log provides details of the daily climatic conditions and natural-history observations of the beetles and host bees as the spring seasons progressed in 2021 and 2022. Due to unusually early spring in 2021 (ground thaw and beetle emergence in late March) and unusually late spring in 2022 (snow cover delayed emergence until late April), there was a significant difference (about five weeks) in the timing of emergence of both the beetle and its host bee.

March 19, 2021. Sunny, 9° to 15°C during the mid-day hours of observations. Road 54N. DD observed 31 live specimens, including three mating pairs and several dead individuals. A few specimens were in the sparse short grass by the trailside. No bees were yet active. Snow was present in a few drifts in low-lying areas (sand trails were clear and thawed), and no new plant growth was noted. The first beetles must have emerged from hibernation several days earlier.

March 20. Sunny, 6°C to 17°C. Road 54N. RW and his son Mark collected 129 specimens, mostly alive, but many dead. Numerous beetles were on their back, struggling to regain their feet, and were trapped in ruts in the sand. When touched, the beetles always feigned death for about five seconds, and then resumed crawling slowly. No individuals were seen in the mat of grass on either side of the trails, but limited time was spent searching there. By far the majority of beetles (groupings of three to eight) were found on the upper-hill, west-facing side of the trail, as if the beetles had been moving downhill and then tumbled over the break in the prairie sod to the sand below. No bees were active. The sex ratio was 70% male to 30% female (N=129).

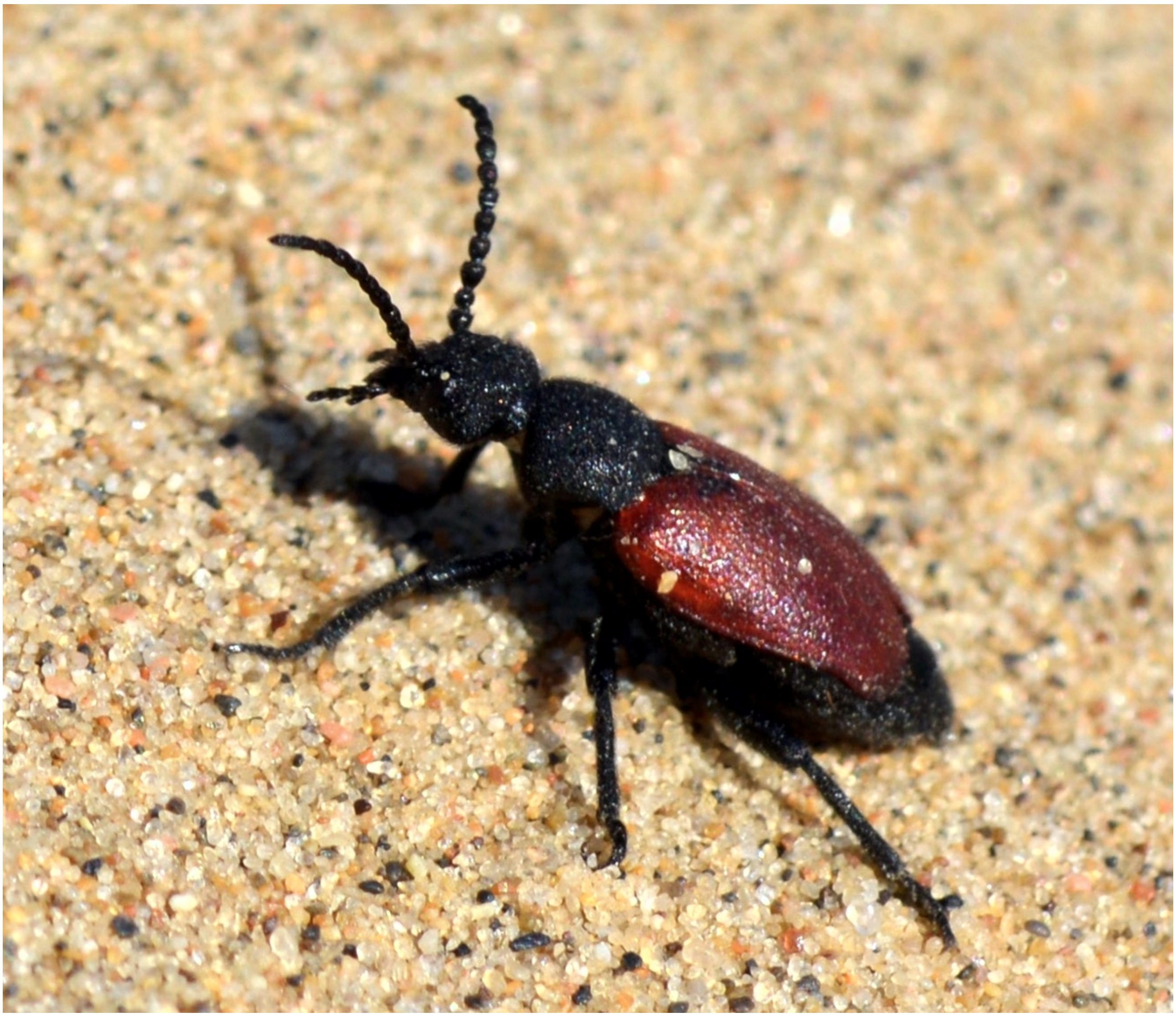

Figure 5. *Tricrania sanguinipennis* attempting to climb up a slope. Individuals often fell over backwards and spent minutes before righting themselves. (Deanna Dodgson)

March 22. Cloudy, 9°C, Road 54N. DD observed 35 live specimens and many others dead.

The next 10 days experienced extreme weather fluctuations, from sunny and 20°C one afternoon to snow and -10°C that same night. Two days later a second low front brought another light snowfall.

March 25. Sunny, 11°C. DD searched a sand pit and surrounding areas in Birds Hill Provincial Park (50.01229, -96.92921) and barren sand dunes in Mars Hill Wildlife Management Area (50.27816, -96.56581), both northeast of Winnipeg, but found no blister beetles.

March 29. Heavy cloud and windy, 6°C at 10:00 am. Road 54N. A few beetles were active but moving very slowly. RW collected 120 beetles by 2:00 pm at which time the temperature had reached 15°C. Dozens of individuals were on their backs, struggling to right themselves, and several were seen being tumbled over the sand surface by wind gusts. Over 50% of the beetles were dead, mostly small individuals (8 mm). The sex ratio was 67% male, 33% female. RW checked another sandhill area west of St. Claude (49.653714, -98.460918) on Provincial Highway 2 at 4:00 pm (mostly cloudy and 20°C), but no beetles were observed. That night, the temperature dropped to -10°C, with rain turning to snow, and -15°C the following night. Many beetles likely froze on the sand surface when it became too cold to continue searching for a burrow.

Among four live specimens collected for observation by RW, two individuals were observed mating the following day. Slices of grape were offered to provide moisture and nourishment, but the beetles showed no interest. They were often on the move, and two had expired by day four, one made it to day five, all likely having exhausted their energy reserves from frequent bouts of activity. This species is not known to eat, and therefore lives for only a week or two (Parker and Boving 1924).

April 1. 10°C with a partly cloudy sky. DD searched sandhills for four hours at a site 10.2 km N, 6.6 km W Holland, and managed to photograph three *T. sanguinipennis* crawling through sparse vegetation and clear patches of sand. This site is 55 km west of the Portage Sandhills. Still no bees or plant growth.

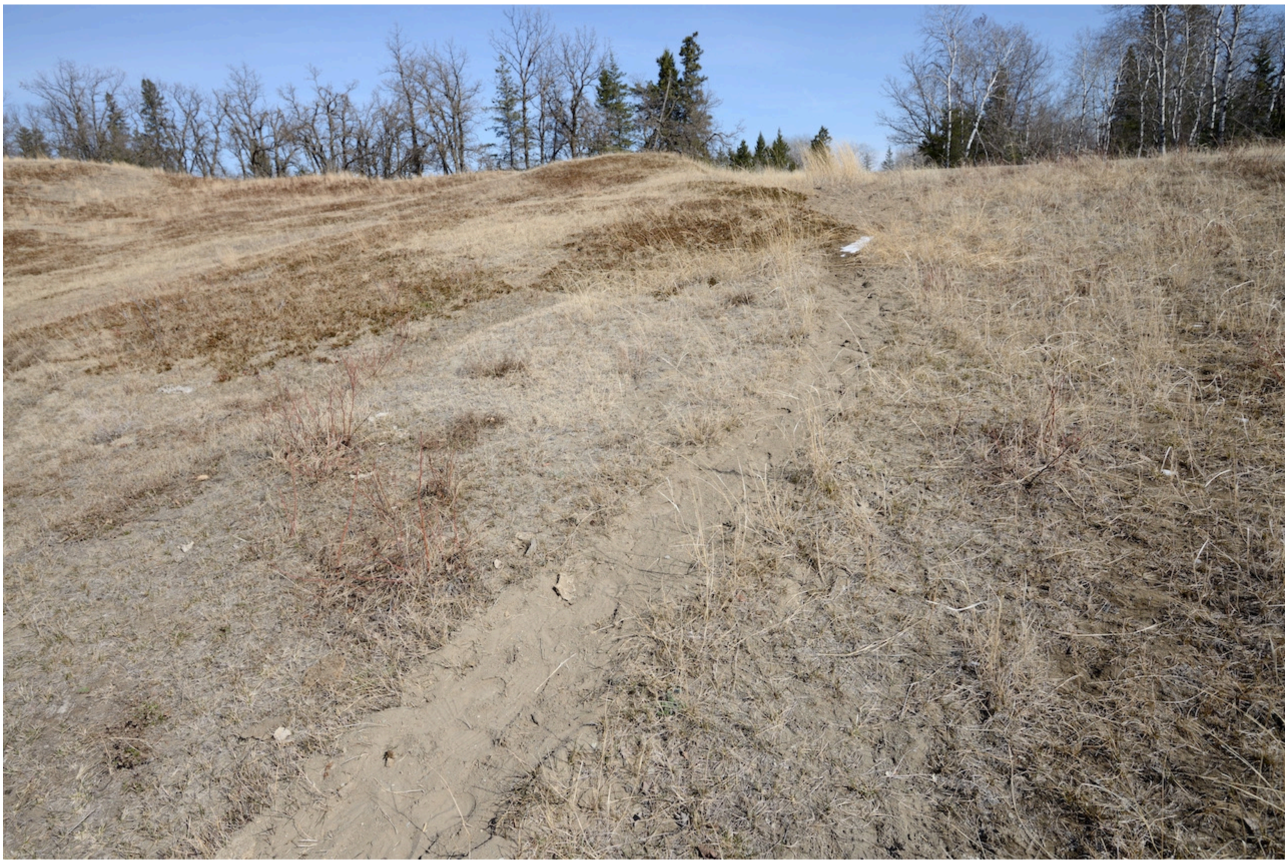

Figure 6. Sandhill habitat at the Holland site. (Garry Budyk)

April 4. 15°C. RW collected at four sites along two roads 54N and 60N, and one site in Rock Road ATV Park on road 36N. A total of 22 specimens were collected, with a sex ratio of 73% males, 27% females. The first observed *C. inaequalis* bee was collected at the latter site.

April 7. 12 to 16°C, DD checked three sites on Road 54N and two sites on road 60N. She observed 9 live and 26 dead beetles, and 10 *C. inaequalis* bees. *Cicindela tranquebarica* and *C. scutellaris* (potential predators of the blister beetles) were observed for the first time this spring.

April 11 to 13. A third storm dropped 12 to 20 cm of snow in the region, freezing every night and with a low of -9°C. The snow cover remained on the ground for over a week.

April 17. Snowfall of 2 cm, temperature rising to 2°C

April 20. -6°C, snowfall of 1 cm; the fifth snow event of the study period.

April 22. 21°C, sunny; low that night of -3°C. RW visited two sites on Road 54N and collected 20 *T. sanguinipennis* (sex ratio 85% males, 15% females), two-thirds of which were dead. *C. inaequalis* bees were abundant at several nesting colonies (2 to 28 burrows at each site) on the upper, west-facing bank of the trail. One mating swarm of bees was observed at a 'rendezvous site' in the sand, about 2 metres away on the other side of the trail from the bee burrows. With bees arriving and departing, it appeared there were about 12 individuals hovering and landing on the spot at a time. None was aggressive to RW.

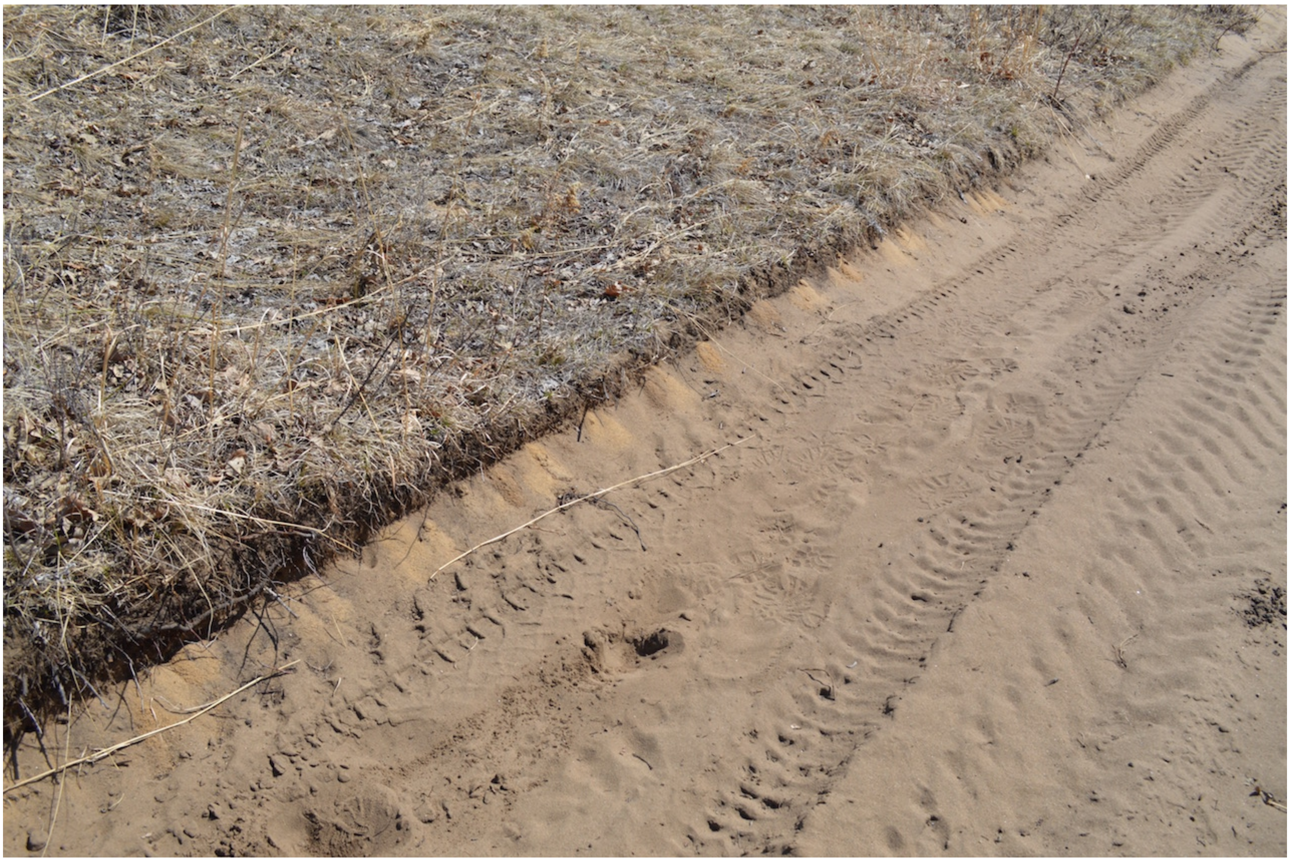

Figure 7. Over two-dozen active burrows (marked by plumes of light-coloured sand) of just one colony of *C. inaequalis* along a trail in the Portage Sandhills. Female bees were observed continually entering and leaving most burrows. (Robert Wrigley)

No *T. sanguinipennis* triungulins were present on the bees of either sex at this early date. Two live adult *T. sanguinipennis* were captured within a few metres of a bee colony. Two *Cicindela tranquebarica* and two large spiders were also collected, and several others of each were seen. The shape and colour of the beetles so closely resembled the fallen reddish-brown winter bud scales of poplars, and the dried fruits of wild rose (both of which littered the ground) that RW had to repeatedly pick up these items to determine if they were beetles. No new plant growth was evident yet. Most of the dead specimens were

again of small size; perhaps they had depleted their energy reserves sooner than did the larger specimens.
April 27. Sunny, 12-18°C, DD revisited the Holland site and observed no *Tricrania* or *Colletes.*

May 12. Sunny, 23°C. RW collected 14 *C. inaequalis* bees, two females of which had a single black triungulin attached, one on the yellow pollen basket, the other on the dorsum. An *Andrena wellesleyana* bee also had a triungulin attached to the dorsal setae between the thorax and abdomen. No *T. sanguinipennis* were seen on the trail, dead or alive, so the life span of adults on the surface appeared to be over. A half hour was devoted to searching for beetle eggs and triungulins, but the sheer abundance of debris on the sand soon proved this exercise fruitless.

June 12. Sunny, 25°C, windy. RW checked the trail and no *T. sanguinipennis* beetles or *Colletes* bees were observed; even the bees' burrow entrances and excavated sand piles were no longer discernible.

2022, April 28, 4°C, DD found 5 *T. sanguinipennis* (no dead ones) at the previous year's two main sandy trails on Road 54N in the Portage Sandhills. This appears to be the start of the beetle's emergence. Some snow in the ditches, but the trails are thawed.

May 4, 2022. DD, RW, Garry Budyk, and Tim Arendse searched for beetles on the same two trails, and RW collected 34 live specimens and observed another 20. Many of the females have swollen abdomens, presumably with eggs. One *Cicindela scutellaris* and two *C. tranquebarica* were collected.

May 5, 2022. DD found 3 specimens at an isolated sandhill on Road 54N, 3.2 km S, 0.8 km E Rossendale, on the north side of the Assiniboine River. Two were mating, and the other was mangled with elytra missing; possibly predated. At this site, about 50 *C. inaequalis* were active.

May 24, 2022. One dead specimen was discovered by biologist Sherry Punak-Murphy on the Canadian Forces Base Shilo. The specimen was situated under a plywood board placed in this sandy mixed-grass prairie for the purpose of studying the Northern Prairie Skink (*Plestiodon septentrionalis*) and the Western Hognose Snake (*Heterodon nasicus*). No skinks or snakes were present under the board on the day she found the beetle (Figure 8).

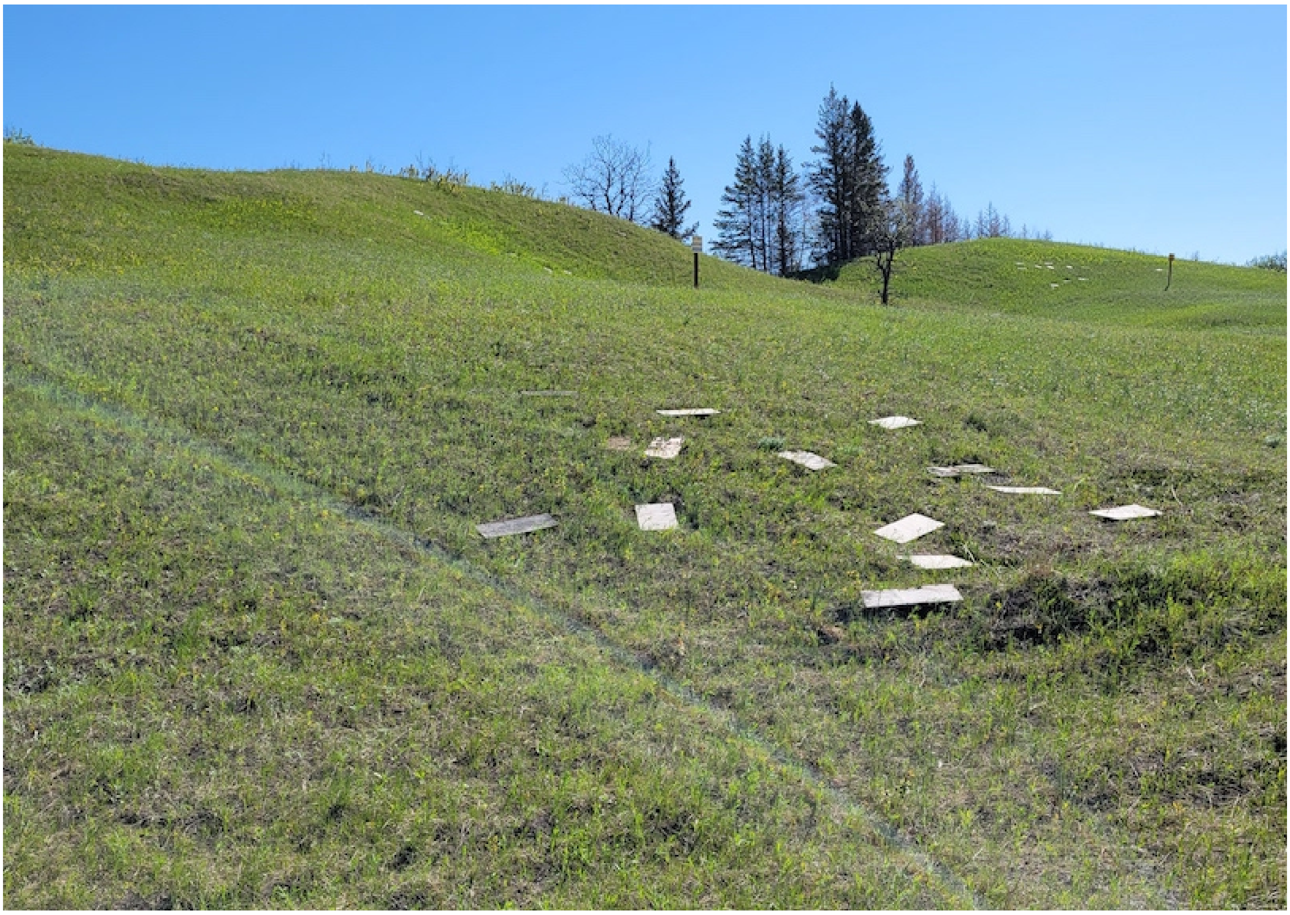

Figure 8. Habitat of the CFB Shilo specimen, the northwest peripheral record for the species. (Sherry Punak-Murphy)

In summary, approximately 350 live and dead beetles were observed over 34 days from March 20 to April 22, 2021, and 58 from April 8 to May 24, 2022. The sex ratio for 291 specimens was 74% males to 26% females. Assuming an equal sex ratio of larvae, perhaps adult females were less active than males while on the surface, with the latter expending more time and energy searching for females to mate. Other researchers (e.g., Frost 1912, Cline and Huether 2011) also noted that males were far more abundant than females, and found large numbers of dead specimens (50%), as we report in our study.

Since many beetles were observed in our study on March 20, 2021, the earliest emergence of some individuals would have begun days earlier. Consequently, the period of beetle activity on the surface that year would have extended for about five weeks (estimated from March 15 to April 22), commencing soon after the snow had melted from the trails, leaving the frozen sand exposed to the warming rays of the sun.

LIFE HISTORY OF *TRICRANIA SANGUINIPENNIS*

This beetle is a parasitoid of several species of solitary, ground-nesting bees including the genus *Colletes,* such as *C. inaequalis* (Figure 7)*, C. validus,* and *C. thoracicus* (Batra 1980). Ghoneim (2013) presented a worldwide review of blister beetle behaviour, focusing on reproduction, but *T. sanguinipennis* was not specifically mentioned. Turco et al. (2003) described the characteristics of sexual behaviour in the meloid subfamily Nemognathinae (of which *Tricrania* is a member) as courtship dorsal without display, and copulation dorsal and brief. Cantharidin produced by the male is transferred to the female in the male's sperm packet.

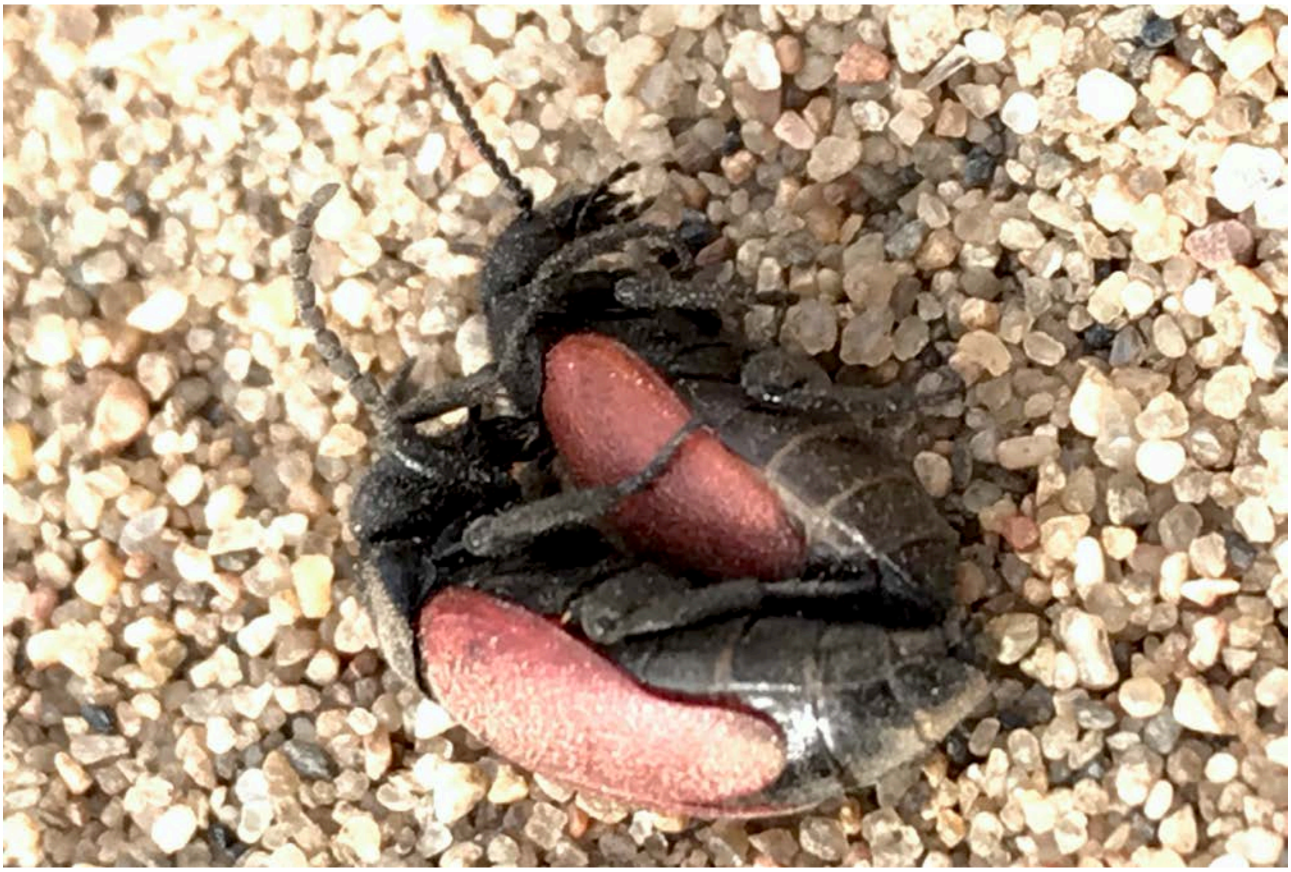

Figure 9. One of several mating pairs of *Tricrania sanguinipennis* observed on March 20*.* Note copulation in the dorsal position. (Mark Wrigley)

Figure 10. Mating in a ventral position. (Deanna Dodgson)

Parker and Boving (1924) found that the peak of the adult beetle's emergence in spring in the District of Columbia occurred in only one or two days, and was dependent on temperature. Mating and oviposition followed immediately, with females of the population laying daily for two to three weeks. The exhausted females died within two to four days, since little or no food was ingested. This, plus freezing overnight temperatures, likely explain why we found so many dead beetles on the trails. In most other meloid genera (e.g., *Epicauta, Lytta*), adults emerge later in the spring, feed actively on plants, and live for three months or more. Females typically mate and oviposit periodically throughout their adult lives (Selander and Fasulo 2020).

Cline and Huether (2011) stated that mating is rarely observed, and they did not find a single mating pair in spite of observing and collecting over 350 individuals over two seasons. Frost (1912) reported observing and collecting 32 males and 13 females (71% males, 29% females) in a few minutes on March 30 in a sandy pasture in Sherborn, Massachusetts, but no sign of mating. We noted over a dozen mating pairs, and when individuals were placed in close proximity, they immediately began copulating.

In Wisconsin, Marschalek (2013) observed specimens from March 10 to April 21, with occasional pitfall records from May 28 to July 28. Our records in southern Manitoba extend from March 19 to May 5 (over the two springs), but some beetles were no doubt active several days before and after our visits to the sites.

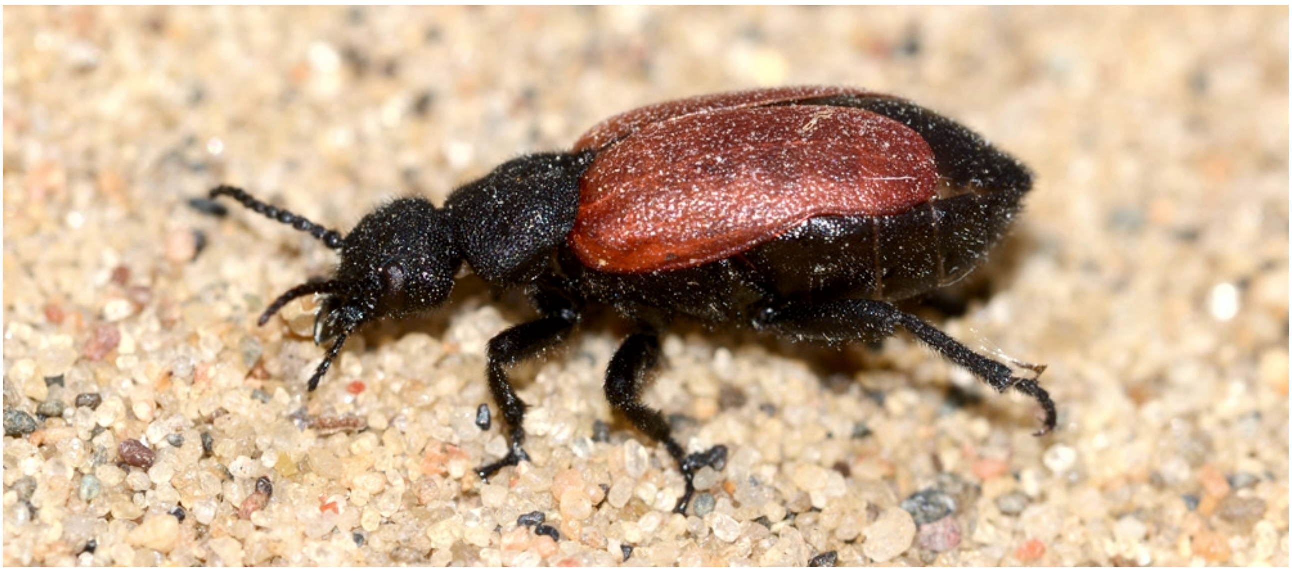

Figure 11. A female with abdomen presumably swollen with eggs. (Deanna Dodgson)

Parker and Boving (1924) noted that the female lays its eggs under debris, such as stones and cow dung, each egg measuring 0.8 by 0.3 mm. Four females mated in the lab produced from 933 to1295 eggs over a period of 13-16 days (from April 28-May 9) and then died 2-4 days later. The large number of eggs is related to the hazards faced by the triungulins, and the low probability of attaching to hosts. Interestingly, the triungulins were found not to enter bee burrows directly, but only via host bees. The authors pointed out that fluctuating temperature, violent rain, or drying winds under natural conditions would affect egg production. With the great amount of debris scattered over the sand at our sites, we spent limited time searching for eggs and triungulins, and found none.

Figure 12. *Colletes inaequalis.* (Reid Miller)

The beetle larva ecloses from the egg in from 12 to 21 days after oviposition (Parker and Boving 1924, Cline and Huether 2011) and is found in the underground cell of cellophane bees *C. inaequalis* Say, 1837, and *C. thoracicus* Smith, 1853 (was *C. rufithorax* Swenk, 1906). Triungulins have also been noted on *Megachile* sp and *Andrena* sp (Cline and Huether 2011), but they are unlikely hosts because bee egg location and nest construction prevented beetle triungulin survival (Parker and Boving 1924).

In our study, we found only two female *C. inaequalis* (Figure 9) and one *Andrena wellesleyana* (Figure 10) with single triungulins attached. On May 29, 1920, Parker and Boving (1924) found that 60% of both male and female *Colletes thoracicus* were infested with 1 to 4 triungulins. In males, most larvae were attached to setae on the posterior underside of the head, while in females, most were attached to setae on the posterior dorsal part of the head, suggesting that the larvae transferred from the male to the female during dorsal copulation. They concluded that highly active male bees, searching for females on the ground, were therefore an active agent enabling the parasitoid to reach and attach itself to the female.

Figure 13. A female *Colletes inaequalis* with a black triungulin attached to a pollen-covered leg. (Reid Miller)

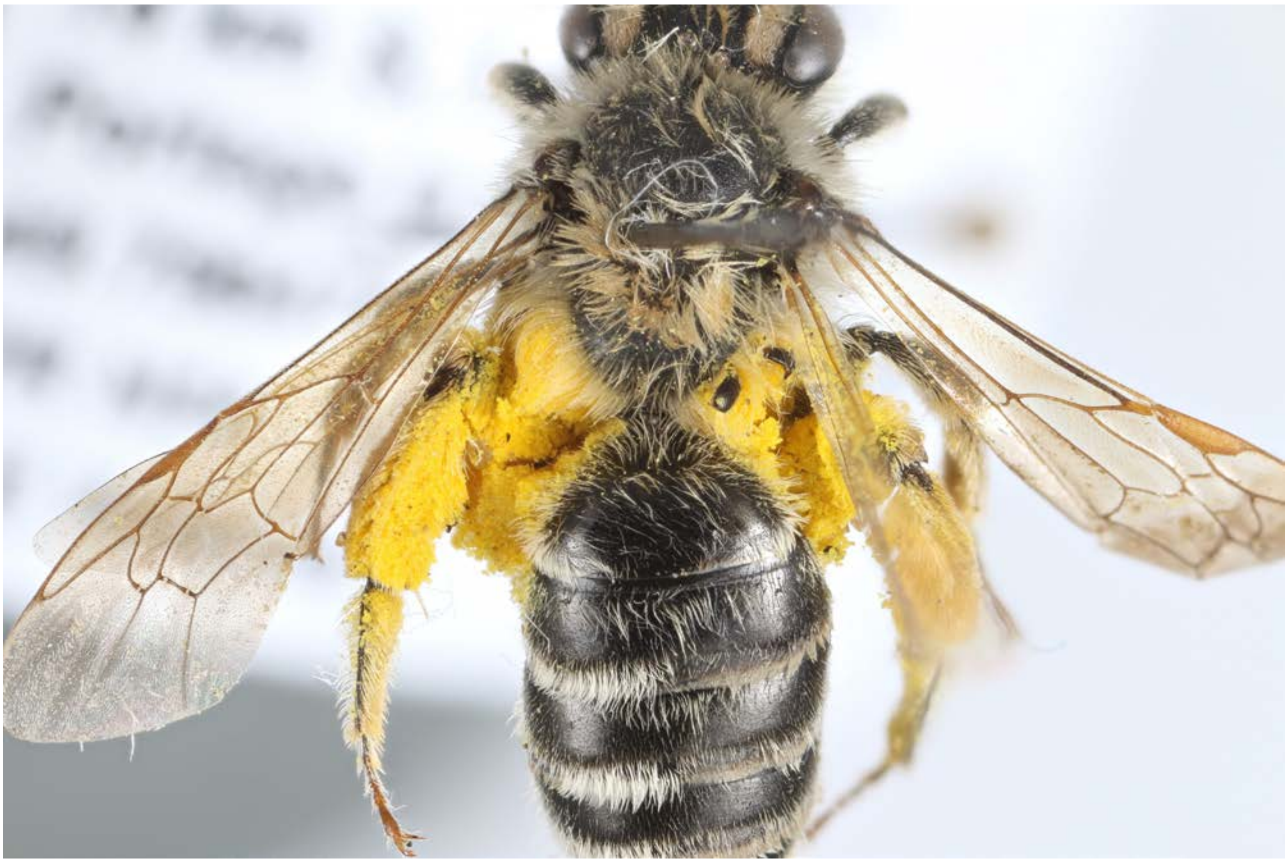

Figure 14. A female *Andrena wellesleyana* with a black triungulin (right side) attached to a pollen-covered leg. (Reid Miller)

The first instar larva of *T. sanguinipennis* is a highly mobile, sclerotized, hypermetamorphic triungulin, followed by four weakly sclerotized grub-like instars, referred to as feeding grubs. A triungulin is depicted in Cline and Huether (2011). Instars six and seven are followed by the pupal stage, lasting 24 days (Arnett et al. 2002). C.V. Riley (in Snodgrass 1930) described the activity of *Epicauta vittata* triungulin larvae as follows; “At night, or during cold or wet weather, all those of a hatch huddle together with little motion, but when warmed by the sun they become very active, running with their long legs over the ground, and prying with their large heads and strong jaws into every crease and crevice in the soil, into which, in due time, they burrow and hide. As becomes a carnivorous creature whose prey [acridid grasshopper eggs] must be industriously sought, they display great powers of endurance, and will survive for a fortnight without food in a moderate temperature.”

The survival period of *T. sanguinipennis* triungulins in the field is unknown, however Parker and Boving (1924) found them active at 11 days in the lab. Neither is it known whether the triungulin produces a chemical cue to attract a male bee, which mimics the sex pheromone of a female bee, as reported for the meloid *Meloe franciscanus* (Saul-Gershenz and Millar 2006). The adult of *T. sanguinipennis* overwinters in the brood cell of the host (Parker and Boving 1924, Batra 1980). Parker and Boving (1924) reported that the maximum emergence of adult beetles occurred on April 12 at Brookland, Washington D.C.

LIFE HISTORY OF *COLLETES INAEQUALIS* AS IT RELATES TO *TRICRANIA SANGUINIPENNIS*

T. sanguinipennis provides a fascinating example of phoresy leading to parasitoidism with cellophane bees, *Colletes* in particular*. Colletes inaequalis* is a ground-nesting species active from March to May (while there is often still some snow remaining on the ground), which occurs in aggregations in southern Manitoba. It uses primarily Red Maple *Acer rubrum* pollen in Maryland (Batra 1980; https://val.vtecostudies.org/projects/vtbees/colletes/). At our study sites in southern Manitoba, pollen of willow (*Salix*) and chokecherry (*Prunus*) are usually available by mid-to-late March. Batra (1980) observed the first bee burrows from March 19 to 21, mating was seen on March 22, eggs were laid from April 22 to May 2, after which males died off, and the females by May 24. Triungulin larvae of *T. sanguinipennis* were found on the medial tuft of setae on the first abdominal sterna of female bees on May 18 and 25.

Soil texture and slope are critical factors in determining where ground-nesting bees choose to nest, and many species, including those in the genus *Colletes*, prefer sand and sandy loam on warm southwest-facing, and well-draining slopes (Antoine and Forrest 2021). Smith (1901) found that *C. inaequalis* began nesting in sandy soil in New Jersey pine barrens by March 12, as soon as the ground thawed. Mating occurred by March 22, and provisioned brood cells by April 1. The membranous cells were generally found at the end of radiating burrows from 45 to 60 cm deep, but may extend down to 86 cm. Batra (1980) noted that eggs were laid from April 22 to May 2 in Maryland, first larvae were discovered on April 23, and adult activity ceased by May 13. Mature larvae developed by July 1. Triungulins were common from May 18 and 25 on the medial tuft of the first abdominal sterna of female bees. While triungulin larvae actively roam on the sand in search of a bee to gain entrance into a burrow, Christine Young video-taped adult beetles mating and; “...wandering in and out of bee holes in the dirt.” (Bugguide.net April 24, 2022, Woodbury, Connecticut)

The Dufour's gland of a cellophane bee secretes a polyester-like substance, which is spread over the soil in the brood cells via the bee’s tongue. This lining helps protect the larval provisions from bacteria and fungi, and because the provisions are watery, it helps keep the provisions from leaking out of the brood cell. While many other bee species lay the egg directly on top of the provisions, a cellophane bee lays the egg suspended above the provisions in the brood cell, which helps prevent the beetle triungulin from becoming trapped in the sticky food (Batra 1980, Antoine and Forrest 2021). Cline and Huether (2011) reported a dramatic drop (40%) in the number of *C. inaequalis* in 2007 following a year of "extreme pressure put upon them by the immense *Tricrania* boom."

In our study, *C. inaequalis* began activity in the first week of April in 2021, about two weeks after beetle emergence. There were still a few snow drifts remaining in ditches, but the sand on the trails and blowouts had thawed. There was no new growth of ground plants, however aspen trees had catkins with pollen by mid-April. The first *C. inaequalis* was collected on April 4, and by April 22, numerous bees were observed entering and investigating burrows in the bank. One mating swarm was observed at a 'rendezvous site' close to one series of burrows. Batra (1980) believed that a mandibular gland secretion of the males attracts females to a site for mating. Paxton (2005) described mating behaviour of *Colletes cunicularius* at rendezvous sites located in the vicinity of female nest entrances. High competition of patrolling males provoked 'scramble' behaviour during attempts to pounce on newly emerging virgin females. Adult bees were not observed on a June 12, 2021 visit to the main site, presumably their life cycle was completed. In 2022, with much later spring temperatures, the first bees (50 active) were observed on May 5.

Figure 15. View of active burrows of *Colletes inaequalis.* Females were observed continuously departing and returning to their own entrance.(Robert Wrigley)

DISCUSSION

The population of *Tricrania sanguinipennis* in south-central Manitoba is located 650 km northwest of the nearest records in the Minneapolis area (Minnetonka, Hennepin County, Minnesota and Cedar Creek Ecosystem Science Reserve. Cline and Huether (2011) discussed two questionable records – in South Dakota (state name only) and an Omstead County, Minnesota record in an unpublished report but no voucher specimen. Interesting questions arise about how and when this flightless species arrived in the sandhills of southern Manitoba, with no records in between. The legs of *T. sanguinipennis* are relatively short compared with those of species in other meloid subfamilies, and consequently its locomotor and dispersal abilities are feeble. The beetle crawls slowly and often falls over on its back when encountering a modest incline, then righting itself with difficulty. It is also active as an adult for only a short period of a week or two in early spring, offering but a brief opportunity for adult dispersal.

Manitoba and the adjacent states of Minnesota and North Dakota were covered by the Laurentide Ice Sheet during the Wisconsinan Glacial Maximum (21,400 years ago). Following the retreat of the ice sheet and subsequent draining of Glacial Lake Agassiz, which persisted from 12,000 to 8000 years ago, the southern part of the province was successively invaded by taiga, spruce forest, and grassland. During the mid-Holocene, roughly 9500 to 5000 years ago, this region was significantly warmer and dryer than at present, enabling a mixed-grass prairie/aspen-oak savanna biota to expand hundreds of kilometres farther north than its current distribution (Nielsen et al. 1996, NOAA).

We surmise that during this period, *T. sanguinipennis* was part of the grassland community that spread into southern Manitoba. As environmental conditions returned to a cooler and wetter phase, this prairie/savanna biota retreated southward, leaving behind relict populations of the beetle in certain sandhill deposits. Other similar isolated, relict populations of species in these sandhills include the Northern Prairie Skink, Western Hognose Snake, and Ghost Tiger Beetle (*Ellipsoptera lepida*). An additional possible mode of range expansion of the beetle over many generations may have been via triungulin travel while attached to the setae of dispersing host bees, which are capable of flying long distances (Selander and Bouseman 1960). Of interest is our discovery of the beetle in an isolated sandhill north of the Assiniboine River, which would present a complete barrier to a weak-crawling, non-flighted beetle with a two-week adult stage.

We found this blister beetle at a number of sites in several distinct sandhill areas in south-central Manitoba, and it will likely be found in other nearby sandhills. It was not observed in a southern outlier of the Portage Sandhills west of St. Claude (largely covered by forest), and large glacial-moraine sand deposits at Birds Hill Park and Mars Hill Wildlife Management Area (the latter two northeast of Winnipeg). In occasional surveys in recent years from late April to July, for photography and arthropod collection by the authors, along trails in the Portage and Carberry sandhills, no *T. sanguinipennis* had been seen, perhaps because the adults' presence on the surface is limited to such a brief period, and therefore easily missed. We may also have been fortunate to find the species in the two years with unusually high populations. We plan to check these and other sandhill sites for the beetle in successive springs. An interesting distributional note is that the prominent entomologist, Norman Criddle, who collected many common and rare insects for decades in the late 1800s and early 1900s, while operating his federal Entomology Lab at his Aweme home (along the western edge of the Carberry Sandhills), did not report the presence of this species. It is possible that the species did not occur in the area during this period?

Another question is what evolutionary selective force(s) could have led to the loss of flight wings in the eastern North American *T. sanguinipennis*, while its identical-looking, western, sister species, *T. stansburyi,* maintained its ancestral ability to fly. Flight wings in *T. sanguinipennis* are not just reduced, but are absent entirely. In his review of flightlessness in insects, Roff (1990) identified a number of factors that might lead to this adaptation, such as the species inhabiting specific and isolated habitats like sand dunes, and permitting greater allocation of the female's resources to egg production. "In some species alternate modes of migration, viz. phoresy and ballooning, have evolved. Phoretic transport (i.e., via other animal species) may be important in those species in which it occurs, but it appears to have evolved relatively rarely."

Bologna and Pinto (2001) stated that; "Too little is known of bionomics in Meloinae to identify correlations between phoresy and other life history traits. There is certainly no obvious relationship to winglessness." However, Saul-Gershenz and Millar (2006) documented in flightless *Meloe franciscanus*; "... a suite of complementary semiochemical and behavioral characters in response to the challenge of locating their hosts' nests in changeable environment in which host bees, bee nests, and bee floral sources are both patchy and ephemeral. The sand dune habitat represents a formidable barrier to dispersal and host location by the small (2 mm in length) and flightless triungulins, and, to circumvent this barrier, the triungulins have commandeered their hosts' sexual communication system."

Dickinson and Dudley (2003) noted that; "One common feature of the otherwise diverse manifestations of flightlessness is a reduced need for locomotor mobility. Selection for maintaining flight may be weak if this capability is not required for dispersal, reproductive behaviour, or predator avoidance." These factors appear to be applicable to flightlessness and limited mobility in *T. sanguinipennis.* Most of the live beetles we observed were on the move, presumably searching for the scent trails of other individuals with which to mate.

Parker and Boving (1924) reported that the maximum emergence of adult beetles occurred on April 12 at Brookland, Washington D.C.. Our data demonstrated that the maximum occurred much earlier in 2021, around March 20, and later, around May 1 in 2022. Southern Manitoba's weather is much colder and with snow cover well into spring. Most species of meloids (e.g., in the genera *Lytta* and *Epicauta*), whose adults feed on leaves, emerge and reproduce mainly in June and July, with some extending from May to August in the northern states and southern Canada (Blodgett 2010, Wrigley and Reimer 2013). The two *Tricrania* species, whose adults do not eat, and therefore live only one to two weeks (Torchio and Bosch (1992), and whose larvae are parasitoids of bees, emerge earlier, mainly in March and April, timed so that their triungulin larvae have the best opportunities to attach to early-breeding ground-nesting bees. Consequently, the beetles are only found in the neighbourhood of their bee hosts.

In the past, RW had his fingers blistered badly from the caustic agent cantharidin (released by reflex bleeding when the beetle is disturbed), when he picked up large numbers of the Striped Blister Beetle *Epicauta vittata*. This species is noted for its higher concentration (about 5 mg) than most other species of meloids (about 0.5 mg) (Townsend 2011). No such burns were experienced in this study when picking up the several hundred specimens of *T. sanguinipennis,* likely due to the relatively small size and possibly low concentration of the toxin in this species. Aoun et al (2018) stated that cantharidin concentration differs significantly in meloids according to species, sex (only males produce it, before transferring it to females during mating), and age.

The black and red colouration of *T. sanguinipennis* is likely an aposematic adaptation, warning predators of its noxious taste and toxicity (Eisner 2003). The chemical deterrent cantharidin precludes the need to fly or crawl rapidly when attempting to escape predation by small mammals, birds and insects (e.g., robber flies). Abundant tracks in the sand of the Deer Mouse (*Peromyscus maniculatus*) and Wild Turkey (*Meleagris gallopavo*) revealed the presence of both of these prime insectivores, which would have presumably impacted the beetle population without the latter's chemical defense. We also observed several species of tiger beetles (*Cicindela formosa*, *C. scutellaris*, *C. tranquebarica*) and large spiders (*Geolycosa, Phidippus* and *Habronattus*), all potential predators of the blister beetles. Cline and Huether (2011) reported a number of dead *T. sanguinipennis* beetles wrapped with spider silk.

Figure 16. Individuals of *Tricrania sanguinipennis* were often challenging to spot among the numerous and similarly coloured poplar winter bud scales and rose hips that littered the sand. Note the poplar bud beside the lower pair of breeding individuals. (Robert Wrigley)

The beetle's small size and bicoloured pattern, resembling the winter bud scales of aspen and balsam poplar (shed in great numbers simultaneously with beetle emergence), provide some degree of camouflage protection. Were it not for the beetle's habit of nearly incessant crawling, we would not have noticed many specimens among these bud scales, rose hips, and other small pieces of debris littering the sand. Interestingly, this species' aposematic colour pattern (black body and legs, and red elytra) displays a strikingly close resemblance to numerous other insects (e.g., mutillid wasps) and beetles (some armed with toxins, others not) in meloid and many other families. As examples, *Lytta fulvipennis* and *Palaestra rufipennis* (Meloidae), *Zenodosus sanguineus* (Cleridae), *Brachyleptura champlaini* and *Anastrangalia rubida* (Cerambycidae), *Anthocomus equestris* (Melyridae), *Platycis minutus* (Lycidae), *Ampedus sanguineus* (Elateridae), *Bostrychus capucinus* (Bostrychidae), *Melasoma populi* (Chrysomelidae), *Aphodius fimetarius* (Scarabaeidae), and *Omophlus lepturoides* (Tenebrionidae)

In this study, 325 specimens of *T. sanguinipennis* were collected (and dozens of others seen, totaling about 400 individuals) along several trails and in blowouts, with over 200 found on just 0.8 km of one trail. The Portage and Carberry sandhills are crisscrossed with hundreds of trails and abundant blowouts, providing apparently suitable habitat for populations of the beetles and host bees. However, a search along many sandy trails and blowouts did not reveal the presence of the beetle or bees. The beetles and bee colonies were concentrated on certain elevated, well-drained trails and blowouts at specific sites consisting of pure sand in prairie and savanna. As soon as a trail dipped to a slightly lower elevation, and shrubs and poplar-oak forest commenced (marked by dark humous mixed in the sand), no beetles were observed. This is likely related to their host bee colonies which are restricted to sites of sunny, well-drained, open ground. The large emergence of *T. sanguinipennis* observed in the present study is typical of a number of meloid species in the genera *Epicauta* and *Lytta* (Wrigley and Reimer 2013), often related to outbreaks of prey species such as grasshoppers (Bilbertson and Horsfall 1940).

We found that this beetle comes out of hibernation at a remarkably early date compared to other coleoptera, their emergence timed so that mating and oviposition have taken place, and hatched larvae are active when host bees emerge, mate and lay their own eggs. Snow storms and temperatures dropping well below freezing on most nights in March, April and May must have presented repeated challenges to the survival of adults and triungulin larvae. In March, 2021, we recorded a remarkable drop in temperature of 30°C (+20°C in the afternoon, then plummeting to -10°Ç with snow overnight). The high proportion of dead adults in the latter half of March (over 35%) is indicative that this Manitoba population is highly susceptible to extreme weather events, and may become evidence of a dramatic negative effect of climate change (i.e., more unpredictable and extreme weather events) on beetle survival. The bee hosts have access to their burrows to avoid freezing temperatures at night, as well as snow, but the limited locomotory and burrowing abilities of the beetles would leave many adults exposed to freezing temperatures on the surface. We could find no information on freeze-tolerant or cold-hardening adaptations of this species.

Another potential factor limiting survival is a mismatch in seasonal timing of spring emergence of the beetle and its bee host due to a potential shift in phenology, although the same above-zero degrees ground temperature would appear to be the joint stimulus controlling emergence of both insects. “Where an organism’s survival is closely tied to the phenology of another species, there should be strong selection for the two to use the same cues, or at least cues that have historically been strongly correlated (Forrest and Thomson 2011).

Interestingly, Bale and Hayward (2010) stated that; “Rapid cold hardening was initially regarded as physiologically interesting but ecologically irrelevant because in natural environments insects do not experience the sudden changes in temperature that cause ‘cold shock’ mortality, e.g. a rapid transfer from 20°C to -8°C...that is commonly used in laboratory experiments to induce the RCH response...However, rapid cold hardening is now recognized as a common occurrence, manifest across a range of responses, and of undoubted ecological importance.” In our study, the wildly fluctuating environmental conditions in the springs of 2021 and 2022 appear to have resulted in high mortality in a species whose adults emerge from diapause remarkably early, apparently to coincide their reproduction and dispersal with their early-emerging bee hosts. Some of these adults (as non-feeders) may also have died due to their short life span of around two weeks while on the surface.

The chance discovery of several widespread populations of *T. sanguinipennis* in south-central Manitoba (with native prairie greatly impacted by agriculture and development), after over a century of occasional surveys in the region by numerous collectors, is indicative of the research that needs to continue to gain a more-complete knowledge of the provincial fauna.

ACKNOWLEDGEMENTS
We thank Garry Budyk, Mark Wrigley, Tim Arendse, and Sherry Punak-Murphy for helping locate specimens in the field. Dr. Jason Gibbs (University of Manitoba, JB Wallis/RE Roughley Museum of Entomology) identified bee samples and provided suggestions to improve the manuscript. We greatly appreciate the editorial comments provided by Drs. Cedric Gillott and Donna Giberson. Jon Rapp provided his exceptional photos of *Tricrania sanguinipennis*, and Reid Miller photographed the triungulin larvae on the bees. Dr. Erwin Huebner kindly provided the SEM images of the blister beetle. Patrice Bouchard (Canadian National Collection of Insects, Arachnids and Nematodes), David Holder (University of Manitoba), and Dr. Randy Mooi (Manitoba Museum) confirmed that no Manitoba specimens of *T. sanguinipennis* were present in their respective collections. We thank Doug Collicutt for hosting this article on NatureNorth.com.

REFERENCES

Antoine, C.M. and J.R.K Forrest. 2021. Nesting habitat of ground-nesting bees: a review. Ecological Entomology 46:143-159.

Aoun, O., Francois, M., Demoncheaux, J-P., and Rapp, C. 2018. Morning blisters: cantharidin-related meloid burns. Journal of Travel Medicine, 25(1): tay045. https://doi.org/10.1093/jtm/tay045 (accessed 3 April 2021)

Arnett, R.H. Jr., Thomas, M.C., Skelley, P.E., and Frank, J.H. 2002. American Beetles. Polyphaga: Scarabaeoidea through Curculionoidea. CRC Press, New York. Volume 2: 861 pp.

Bale, J.S., and Hayward, S.A.L. 2010. Insect overwintering in a changing climate. Journal of Experimental Biology, 213(6): 980-994. https://doi.org/10.1242/jeb.037911 (accessed 6 May 2021)

Batra, S.W.T. 1980. Ecology, behavior, pheromones and management of the sympatric vernal bees *Colletes inaequalis*, *C. thoracicus* and *C. validus*. Journal of the Kansas Entomological Society, 53: 509-538.

Bilbertson, G.I., and Horsfall, W.R. 1940. Blister beetles and their control. Entomology Department, Agricultural Experiment Station, South Dakota State College, Brookings. Bulletin 340: 1-23.
https://openprairie.sdstate.edu/cgi/viewcontent.cgi?article=1339&context=agexperimentsta_bulletins (Accessed 25 April 2021)

Blodgett, S., Denke, P.M., and Knerr, V. 2010. Blister beetles of Montana. Montana State University, Agriculture and Natural Resources (Pest Management). http://animalrange.montana.edu/documents/extension/MT200209AG.pdf (accessed 4 April 2021).

Bologna, M.A., and Pinto, J.D. 2001. Phylogenetic studies of the Meloidae (Coleoptera), with emphasis on the evolution of phoresy. Systematic Entomology, 26: 33-72.

Bousquet, Y., Bouchard, P., Davies, A.E., and Sikes, D. 2013. Checklist of Beetles of Canada and Alaska. Pensoft Series Faunistica No. 109, Sofia-Moscow. 402 pp. https://doi.org/10.3897/zookeys.946.52489 (accessed 24 May 2021)

Bugguide.net. https://bugguide.net/node/view/93509 (accessed 21 May 2021).

Cline, A.R., and Huether, J.P. 2011. Revision of the nearctic blister beetle genus *Tricrania* LeConte, 1860 (Coleoptera: Meloidae: Nemognathinae). Zootaxa, 2832: 1-43. https://www.gbif.org/species/119398031 (accessed 24-March 2021).

Dickinson, M., and Dudley, R. 2003. Flight. *In*; Encyclopedia of Insects. Editors Resh, V.H., and Carde, R.T. Academic Press, New York. pp 416-426.

Eisner, T. 2003. For Love of Insects. The Belknap Press of Harvard University Press, Cambridge, Massachusetts. 448 pp.

Evans, A. 2014. Beetles of Eastern North America. Princeton University Press, Princeton. 560 pp.

Forrest, J.R.K., and Thomson, J.D. 2011. An examination of synchrony between insect emergence and flowering in Rocky Mountain meadows. Ecological Monographs 81(3): 469-491. https://esajournals.onlinelibrary.wiley.com/doi/pdfdirect/10.1890/10-1885.1 (accessed 5 May 2021)

Frost, C.A. 1912. Note on *Tricrania sanguinipennis* Say (Coleoptera)Psyche: A Journal of Entomology, 19: 208. https://doi.org/10.1155/1912/36486 (accessed 30 March 2021)

Gerber, G.H., and Church, N.S. 1976. The reproductive cycles of male and female *Lytta nuttalli* (Coleoptera: Meloidae). The Canadian Entomologist, 108(10): 1125-1136. DOI: https://doi.org/10.4039/Ent1081125-10 (accessed 21 April 2021)

Ghoneim, K.S. 2013. Behavioural characterization of blister beetles (Coleoptera: Meloidae) in the world: A bibliographic review. http://www.academeresearchjournals.org/journal/ijsbs (accessed 21 April 2021)

Litman, J.R. 2019. Under the radar: Detection avoidance in parasitic bees. Philosophical Transactions of the Royal Society of London, Series B, Biological Sciences, 374(1769):20180196. doi: 10.1098/rstb.2018.0196 (accessed 21 April 2021)

Marschalek, D.A. 2013. Blister beetles (Coleoptera: Meloidae) of Wisconsin: Distribution and ecology. https://depot.library.wisc.edu/repository/fedora/1711.dl:CG32FRISRZ5KL87/datastreams/REF/content. (accessed 29 March 2021)

Nielsen, E., McLeod, K.D., Pip, E., and Doering, J.C. 1996. Late Holocene environmental changes in southern Manitoba – Field Trip Guidebook A2, Geological Association of Canada/Mineralogical Association of Canada Annual Meeting, Winnipeg, Manitoba, May 27-29. https://www.manitoba.ca/iem/info/libmin/gacmac/guidebook_a2.pdf (accessed 30 March 2021)

NOAA National Centres for Environmental Information. https://www.ncdc.noaa.gov/global-warming/mid-holocene-warm-period (accessed 30 March 2021)

Parker, J.B., and Boving, A.G. 1924. The blister beetle *Tricrania sanguinipennis* -- biology, descriptions of different stages, and systematic relationship. Proceedings of the United States National Museum, 64: 1-40.

Paxton, R.J. 2005. Male mating behaviour and mating systems of bees: An overview. Apidologie, 36: 145-157. https://www.apidologie.org/articles/apido/pdf/2005/02/M4066.pdf (accessed 12 May 2021)

Quinn, M. 2015. http://texasento.net/TXMeloidae.html (accessed 2 April 2021).

Roff, D.A. (1990). The evolution of flightlessness in insects. https://doi.org/10.2307/1943013 (accessed 23 May 2021)

Saul-Gershen, L.S., and Millar, J.G. 2006. Phoretic nest parasites use sexual deception to obtain transport to their host's nest. https://www.pnas.org/content/103/38/14039 (accessed 20 April 2021)

Say, T. 1891. The complete writings of Thomas Say on the entomology of North America. Edited by J.L. Le Conte. Volume II, p166. A.E. Foote, Philadelphia. https://www.biodiversitylibrary.org/item/79303#page/27/mode/1up (accessed 3 April 2021)

Selander, R.B., and Bouseman, J.K. 1960. Meloid beetles (Coleoptera) of the West Indies. Proceedings of the United States National Museum, 111(3428): 197-226.

Selander, R.B., and Fasulo, T.R. 2020. Featured creatures; Blister beetles. University of Florida, Entomology and Nematology. http://entnemdept.ufl.edu/creatures/urban/medical/blister_beetles.htm (accessed 21 April 2021)

Snodgrass, R.E. 1930. Insects, Their Ways and Means of Living. Smithsonian Scientific Series, 5: 362 pp. SSS_05_Snodgrass_1930.pdf (accessed 20 April 2021)

Torchio, P.F., and Bosch, J. 1992. Biology of *Tricrania stansburyi*, a meloid beetle kleptoparasite of the bee *Osmia lignaria propinqua* (Hymenoptera: Megachilidae. Annals of the Entomological Society of America, 85(6): 713-721. *https://doi.org/10.1093/aesa/85.6.713* (accessed 29 March 2021)

Townsend, L.H. 2011. Blister beetles. Entomology at the University of Kentucky. College of Agriculture, Food and Environment. Entfact-102. https://entomology.ca.uky.edu/ef102 (accessed 25 April 2021)

Turco, F., Di Giulio, A., and Bologna, M.A. 2003. Sexual and cleaning behavior and related morphology in the genus *Cerocoma* (Coleoptera: Meloidae). Journal of Insect Behavior, 16(2): 257-278.

Webster, R.P., Tonnancour, P de., Sweeney, J.D., Webster, V.L., Kostanowicz, C.A., Hughes, C., Anderson, R.S., Klymko, J., Chantal, C., and Vigneault, R. 2020. New coleoptera records from eastern Canada, with additions to the fauna of Manitoba, British Columbia, and the Yukon Territory. ZooKeys, 946:53-112. https://zookeys.pensoft.net/article/52489/ (accessed 24 May 2021).

Wrigley, R.E., and Reimer, J. 2013. Striking oil -- beetles, that is. The Entomological Society of Manitoba Newsletter, 40(1): 7-9.

AUTHORS' BIOS

Deanna Dodgson is an avid amateur entomologist and nature photographer living in Winnipeg. She has contributed many of her photos and species records to on-line nature websites.

Dr. Robert E. Wrigley is an ecologist, amateur entomologist, and former Curator and Museum Director at the Manitoba Museum, founding Director of the Oak Hammock Marsh Interpretive Centre, and former Curator of the Assiniboine Park Zoo.