Ants and subterranean Sternorrhyncha in a native grassland in east-central Alberta, Canada

J.S. Newton, J. Glasier, H.E.L. Maw, H.C. Proctor, R.G. Foottit

Abstract—Little is known about the associations of ants (Hymenoptera: Formicidae) with subterranean aphids and mealybugs (Hemiptera: Sternorrhyncha: Aphididae and Pseudococcidae), particularly in Canadian grasslands. Knowledge of host plants for these sternorrhynchans is equally rare. We carried out a plant-based survey of ants and belowground aphids and mealybugs in a native fescue grassland in east-central Alberta, Canada. We found 23 species of ants, 12 of which (species of *Lasius* F., *Myrmica* Latreille, *Tapinoma* Förster, and *Temnothorax* Mayr) were in association with subterranean sternorrhynchans. Twelve species of aphids and mealybugs were collected; 3 are new records for Canada and 2 are possibly undescribed. Most ant species associated with sternorrhynchans were found with more than one species of sternorrhynchan, sometimes in the same nest. Almost all sternorrhynchans were found on graminoid hosts (Poaceae and Cyperaceae); there was little observed plant-specificity beyond this. There were no significant correlations between presence of subterranean sternorrhynchans and percent cover of different plant types, soil moisture content, slope, aspect, or visible entrances to ant nests.

Résumé—Les associations des fourmis (Hymenoptera : Formicidae) et des pucerons et cochenilles (Hemiptera : Sternorrhyncha : Aphididae et Pseudococcidae) hypogées sont mal connues, particulièrement dans les prairies canadiennes. Il y a de même peu de renseignements sur les plantes hôtes de ces sternorhynches. Nous avons mené un inventaire des fourmis et des pucerons et cochenilles hypogées en fonction des plantes dans une prairie indigène à fétuque dans le centre-est de l'Alberta, Canada. Nous avons trouvé 23 espèces de fourmis, dont 12 (des espèces de *Lasius* F., *Myrmica* Latreille, *Tapinoma* Förster et *Temnothorax* Mayr) sont associées aux sternorhynches hypogées. Nous avons récolté 12 espèces de pucerons et cochenilles; 3 représentent des nouvelles mentions pour le Canada et 2 sont potentiellement non décrites. La plupart des espèces de fourmis associées aux sternorhynches le sont avec plus d'une espèce de sternorhynches, quelquefois dans le même nid. Presque tous les sternorhynches se retrouvent sur des hôtes graminoïdes (Poaceae et Cyperaceae); il n'y a, par ailleurs, aucune spécificité additionnelle des plantes. Il n'existe aucune corrélation significative entre la présence de sternorhynches hypogées et le pourcentage de couverture des différents types de plantes, le contenu hydrique des sols, la pente, l'aspect général, ni la présence d'orifices visibles de nids de fourmis.

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Introduction

Ants (Hymenoptera: Formicidae) and various Sternorrhyncha species (Hemiptera) have a long history of close association (Johnson *et al.* 2001). Their relationship is mostly mutualistic whereby the ants benefit from "honeydew" (carbohydrate-rich secretions) produced by the sternorrhynchans and the sternorrhynchans are protected from

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predators. Four families of Sternorrhyncha commonly exhibit this "trophobiotic" form of mutualism with ants: Aphididae (Aphidoidea) and Coccidae, Pseudococcidae, and Stictococcidae (Coccoidea) (Delabie 2001). Most research has focused on this interaction above ground (reviewed by Way 1963; Buckley 1987; Stadler and Dixon 2005; Styrsky and Eubanks 2007) and has shown that the association of the tending ants and their symbionts ranges from facultative to obligate (Hölldobler and Wilson 1990; Lapolla *et al.* 2006). Interactions of tending ant species and root-feeding aphids and mealybugs are poorly understood, even though this phenomenon has long been recognized (*e.g.*, Lubbock 1882).

In a study conducted near Kinsella, Alberta, Canada, Coupe (2003) suggested that the ants *Tapinoma sessile* (Say), two species of *Lasius* F., and a species of *Myrmica* Latreille were possibly tending unidentified aphids. In this study we followed up on Coupe's observations to determine plant-insect associations, the specificity of interactions, and whether or not particular soil and landscape features provide clues to the presence of ants tending root-feeding sternorrhynchans.

Methods

The study site (centered around 53°01.1′N, 111°32.1′W) is located at the University of Alberta Research Ranch in Kinsella. The ranch is situated in native fescue grassland within aspen parkland, geographically positioned between grasslands to the southeast and boreal forest to the northwest. The landscape consists of knob and kettle topography with short slopes interspersed with intermittent wetlands. Soils are characterized as orthic black and dark brown chernozems (Howitt 1988). The vegetation is dominated by graminoids (*Festuca hallii* (Vasey) Piper and species of *Bromus* L., *Elymus* L., *Hesperostipa* (Elias) Barkworth, and *Poa* L. (Poaceae) and species of *Carex* L. (Cyperaceae)), low-growing forbs and shrubs (*Amelanchier alnifolia* (Nutt.) Nutt. *ex* M. Roem. and *Rosa arkansana* Porter (Rosaceae), *Artemisia frigida* Willd. (Asteraceae), *Elaeagnus commutata* Bernh. *ex* Rydb. (Elaeagnaceae)), and patches of trembling aspen (*Populus tremuloides* Michx. (Salicaceae)) (S.R. White, University of Alberta, personal communication).

In mid-June and late July 2009, 230 soil cores were taken from a variety of locations at the ranch over an area of approximately 50 ha. Nineteen transects of 50 or 100 m length were selected within this area to maximize plant diversity and variation in aspect and slope, but avoiding aspen stands. A 1 m^2 quadrat was sampled every 10 m along each transect, resulting in a total of 115 quadrats. To avoid visual bias, each quadrat was blindly placed approximately 1–3 m from the transect, alternating between left and right.

Percent cover of "grasses", "forbs", "shrubs" V, and "bare ground" in each quadrat was determined by rough visual estimation. Other measurements taken were soil moisture content (Theta probe type ML2x, Delta-T Devices, Cambridge, England), slope category (none, shallow, steep), aspect, and number of visible entrances to ant nests. Two soil cores approximately 15 cm × 20 cm wide and 15 cm deep were collected per quadrat using a spade and a hand shovel, placed in plastic bags, and returned to the laboratory for examination. We attempted to maximize the number of plant species included within each coring site.

Each core was manually broken apart in a white tray and carefully inspected for ants, aphids, and mealybugs. Cores that had gone through ant nests were recognized by the presence of chambers, ant eggs and pupae, and a relatively high abundance of ants. Plants with sternorrhynchans on their roots or root crowns were identified to the lowest level possible with the aid of local botanists. Ants were identified using Wheeler and Wheeler (1963), Bolton (1995), and unpublished keys to Albertan species created by J.G. Sternorrhynchans were identified by H.E.L.M. using reference collections at the Canadian National Collection of Insects, Arachnids and Nematodes, Agriculture and Agri-Food Canada, Ottawa. Ant-sternorrhychan associations were categorized as "direct" (sternorrhynchans found in an ant nest or observed being carried by ants) or "indirect" (sternorrhynchans and ants simply found in the same quadrat or soil

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core). We looked for evidence of associations among insects, plants, and environmental variables using bivariate correlation tests in SPSS (SPSS Inc. 2008).

Results

A total of 23 of the 87 species of ants reported for Alberta (Glasier 2011) were collected. Eleven of these were not observed in association with sternorrhynchans: *Camponotus modoc* Wheeler, *Formica hewitti* Wheeler, *F. lasioides* Emery, *F. limata* Wheeler, *F. neogagates* Viereck, *F. neorufibarbis* Emery, *F. obscuriventris* Mayr, *F. oreas* Wheeler, *F. podzolica* Francoeur, *Leptothorax muscorum* (Nylander), and *Myrmica brevispinosa* Wheeler. The remaining 12 species (Table 1) were considered to be directly or indirectly associated with subterranean Sternorrhyncha. Most of the ants listed in Table 1 were found in association with more than one root-feeding sternorrhynchan species, sometimes in the same ant nest.

Eight species of Aphididae were collected, the most common being *Forda marginata* Koch and a member of the *Geoica utricularia* (Passerini) species complex. Specimens of *Anoecia* Koch collected from *Elymus* roots differ from all described species in this genus but match specimens previously known only from roots of *Hordeum jubatum* L. (Poaceae) in Winnipeg (see Blackman and Eastop 2006). However, intraspecific variation in many species of *Anoecia* is poorly documented, and we are unsure that this is indeed an undescribed species. Also, a single specimen of *Geoica* Hart found on *Hesperostipa curtiseta* (Hitchc.) Barkworth fell outside the documented variation within species of *Geoica* known to occur in North America. Finally, we collected *Pleotrichophorus pseudoglandulosus* (Palmer) and *Pseudoepameibaphis tridentatae* (Wilson). Both are known as aboveground feeders on sage (*Artemisia* L. (Asteraceae)) (Foottit and Richards 1993), but we collected them from unidentified graminoid roots and root crowns, respectively.

Four species of Pseudococcidae were collected. One specimen of *Heliococcus osborni* (Sanders), found on the soil surface and not associated with ants, represents a considerable northern extension of the distribution of this species and the first record for Canada (Kosztarab 1996). The other three species were ant-associated. *Chnaurococcus trifolii* (Forbes) is widespread in North America and has been previously reported from Alberta (Ben-Dov 2010*a*, citing Ben-Dov 1994 (but no such record exists in Ben-Dov 1994)). *Tridiscus sporoboli* (Cockerell) was previously known only from New Mexico and Nebraska (Ben-Dov 2010*b*). *Phenacoccus solenopsis* Tinsley is a widespread pest of a variety of economically important plants (CAB International 2011); our collection is the first record of a population in Canada under field conditions (as opposed to greenhouse collections; H.E.L.M., personal observation). *Phenacoccus solenopsis* was the only sternorrhynchan in this study found on non-graminoids (*Artemisia frigida* (Asteraceae) and *Rosa arkansana* (Rosaceae)). Graminoid hosts plants for the various pseudococcids were species of *Elymus*, *Festuca*, *Hesperostipa*, *Poa*, and *Carex*. There was no evidence that graminoid-feeding sternorrhynchan species were restricted to one host species.

None of the environmental data collected were significantly correlated with ants or subterranean sternorrhynchans ($P > 0.10$). Although we frequently found ants without sternorrhynchans (Fig. 1), only four samples had root-feeding aphids (*Geoica utricularia* and *Anoecia* sp.) without tending ants, and all subterranean pseudococcids (Table 1) were found with ants.

Discussion

Although sampling intensity and study-area size were limited, a large number of target species were collected, including new records for Alberta or Canada. This demonstrates the current gap in knowledge of the diversity of subterranean arthropods in Canada. This may be due in part to a lack of taxonomists specializing in these taxa, but the inherent difficulty of locating, sampling, and observing the behaviour of cryptic soil fauna also plays a

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Table 1. Associations of ant species (Formicidae) and Sternorrhyncha.

Aphididae	*Lasius coloradensis* Wheeler	*Lasius crypticus* Wilson	*Lasius fallax* Wilson	*Lasius flavus* (F.)	*Lasius neoniger* Emery	*Lasius niger* (L.)	*Lasius pallitarsis* (Provancher)	*Myrmica fracticornis* Forel	*Myrmica latifrons* Starcke	*Tapinoma sessile* (Say)	*Temnothorax ambiguus* (Emery)	*Temnothorax rugatulus* (Emery)
Forda marginata Koch	j	—	—	**Q**, **Z**, **FF**	B	—	**N**, **S**, **U**, **X**, cc, **HH**	d, **J**	gg	—	q	cc
Anoecia sp.	—	—	—	—	—	—	**V**, cc	—	—	—	—	cc
Aphis middletonii Thomas	—	—	—	—	—	—	X	—	—	—	—	—
Geoica utricularia (Passerini) complex	j	M	—	**E**, **K**, **Z**, **AA**, dd, **EE**	—	—	**L**, o, **R**, **Y**, cc, dd, **HH**, **II**	d, **J**	r	—	t	cc
Geoica sp.	—	—	—	E	—	—	—	—	—	—	—	—
Pleotrichophorus pseudoglandulosus (Palmer)	bb	—	—	—	—	—	—	—	—	bb	—	—
Pseudoepameibaphis tridentatae (Wilson)	—	—	—	—	—	—	L	—	—	—	—	—
Tetraneura sp.	—	—	—	—	—	—	—	W	—	—	—	—
Pseudococcidae												
Tridiscus sporoboli (Cockerell)	—	—	—	—	—	—	—	—	—	A	a	—
Chnaurococcus trifolii (Forbes)	j		—	k, q, **FF**	C	—	**L**, **R**	j	f, r		q	—
Phenacoccus solenopsis Tinsley	—	—	h	e, p	—	N	—	h	g	h, i	—	—

Note: Letter codes (a–z, aa–ii) represent individual quadrats. Ant-sternorrhynchan pairs with the same letter code came from the same quadrat; boldfaced capital letters denote an observed direct association (either sternorrhynchans found in an ant nest, or ants observed transporting sternorrhynchans); lower-case letters denote co-occurrence in the same quadrat, but no observed direct association, *e.g.*, *Anoecia* sp. found in direct association with *L. pallitarsus* (**V**), but also in indirect association with *L. pallitarsus* and *T. rugalutus* in one quadrat (cc), along with *F. marginata* and *G. utricularia* (cc).

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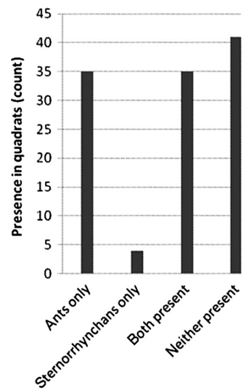
role. For example, not all ant species collected with sternorrhynchans (notably *Myrmica latifrons* Starcke, *Tapinoma sessile*, both species of *Temnothorax* Mayr, and more surprisingly *Lasius coloradensis* Wheeler and *L. fallax* Wilson) were directly observed interacting with their associated sternorrhynchans (Table 1). To our knowledge, no published record exists of a species of *Temnothorax* tending sternorrhynchans above or below ground, but species of *Lasius* are generally known to tend sternorrhynchans (Fisher and Cover 2007). *Lasius coloradenis* is particularly noted as being entirely subterranean and dependent on symbiotic relationships with root-feeding sternorrhynchans (Hölldobler and Wilson 1990; Fisher and Cover 2007).

The lack of observed direct interaction may be a function of sample size or miscategorization of some of the indirect associations. Breaking up of soil cores occasionally made it difficult to identify ant tunnels or nest chambers, and the disturbance caused by sorting may have caused ants to abandon their sternorrhynchan associates. Almost all subterranean sternorrhynchans were found in the presence of ants and, for those species that were occasionally found alone, there were other samples in which they were tended by ants. Perhaps the pattern shown in Figure 1 is caused by a high density of ants in the area, resulting in almost all Sternorrhyncha co-occurring with ants. However, this seems unlikely, as only 60% of all sampled quadrats contained ants above or below ground. We suggest that the very small number of Sternorrhyncha found alone, limited to *G. utricularia* and *Anoecia* sp. in 3.4% of all quadrats, were either temporarily not tended by ants or may be the only two species in our study area to not always have an association with ants. This implies that although the association may be facultative for ants, it might be obligatory for the other belowground sternorrhynchans.

Although we had hoped to find environmental cues that would increase the likelihood of our locating ants together with sternorrhynchans, there were no significant correlations with plant cover, soil moisture content, slope, aspect, or presence of visible entrances to ant nests. This implies that either there are none, we

Fig. 1. Frequencies of occurrence and co-occurrence of ants and subterranean sternorrhynchans in 115 quadrats at Kinsella Ranch, Alberta, Canada.



did not measure the relevant environmental factors, or our sample size was not adequate. Lastly, as previously noted for some taxa by Vogel and Kindler (1980), none of the sternorrhynchans in our study were found to be specific to a host-plant species.

Biological surveys of belowground ant and sternorrhynchan symbioses are rare and behavioural studies often limited to a few taxa (*e.g.*, Lapolla *et al.* 2002; Stuart and Polavarapu 2002; Kishimoto-Yamada *et al.* 2005), suggesting that such associations may be uncommon. However, our data suggest that they may be more widespread than was previously thought. We feel that this is an overlooked relationship that merits more attention from taxonomists and ecologists.

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