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Characterization of Arthropod Assemblages Attracted to Canine Feces Using Pitfall Traps and Rearing Experiments

Jennifer Albrecht

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University of Nebraska – Lincoln

Abstract

The decomposition of fecal matter is critical for an ecosystem, turning matter into a form that is usable by other organisms; the removal of feces also eliminates breeding sites for pests and prevents waste from entering waterways. The aim of this study was to survey arthropod populations that are attracted to dog feces in suburban and residential settings. Due to their established role in nutrient recycling, an emphasis was placed on identifying dung beetle species present. Monitoring was conducted in Lancaster County, Nebraska from July to September 2017 using pitfall traps and rearing experiments. It was hypothesized that southeast Nebraska supports a low level of paracoprid dung beetle activity. Furthermore, it was predicted that arthropods commonly found in association with vertebrate feces (e.g. calliphorid and sarcophagid species) would be recovered during rearing experiments. Arthropods from four classes and ten orders were collected during this evaluation; a total of seven scarab beetle species were identified. Results from this study were used to estimate species richness and abundance at each of the three sites evaluated. This information was then used to identify future work required to assess whether native arthropod populations could be augmented to recycle canine waste in Lancaster County, Nebraska.

Characterization of Arthropod Assemblages Attracted to Canine Feces Using Pitfall Traps and Rearing Experiments

Organisms responsible for the decomposition of fecal matter provide a valuable ecological service by turning waste into a form that is usable by other organisms (Speight et al., 2008). A number of arthropods have evolved mechanisms to exploit dung for nutrition, development, or protection from predators and competitors. In particular, beetles in the subfamily Scarabaeinae are largely coprophagous detritivores; these species feed on the liquid fraction of feces and use the fibrous portion to form brood balls for raising larvae (Nichols et al., 2008). Three general nesting strategies have been observed among dung beetles. Rollers (telecoprids) produce balls of dung and move them away from the food source prior to burial. Tunnelers (paracoprids) dig burrows and push dung into nests that are formed directly underneath the fecal matter. Dwellers (endocoprids) live and develop within the original dung pad or at the soil-feces interface (Nervo et al., 2014).

Waste recyclers, such as dung beetles, act as ecosystem engineers, regulating the function and productivity of a habitat in several ways (Boze et al., 2012; Lumaret et al., 1992). Aboveground dung movement incorporates organic matter into the soil and distributes nutrients throughout the area, improving soil fertility (Barragán et al., 2011). The rolling process also initiates changes in the chemical and microorganismal composition of soil, improves forage palatability, and facilitates the dispersal of plant seeds (Boze et al., 2012). The tunneling and burial activities of dung beetles incorporate nitrogen into the soil, improve aeration, and enhance vegetation productivity; bioturbation also reduces erosion and surface water runoff (Brown et al., 2010).

The removal or incorporation of fecal matter into the soil disrupts breeding sites for parasites and pestiferous insects (Boze et al., 2012). The survival of dung-breeding flies, for example, is reduced through mechanical damage to eggs, habitat alteration, and greater intraspecific competition (Nichols et al., 2008). Similarly, cattle raised on lands without dung beetles are four times more likely to acquire endoparasites than those in areas with a natural dung beetle population (Nichols et al., 2008). Furthermore, it has been estimated that dung beetles decrease the persistence of feces in pastures by 19% (Losey et al., 2006). The removal of fecal matter from pastures avoids spoilage of foraging material and loss of rangeland due to fouling (Losey et al., 2006).

Several studies have examined dung beetle associations with a range of vertebrate hosts (Carpaneto et al., 2005; Dormont et al., 2010; Fincher et al., 1970; Gittings et al., 1998; Halffter et al., 1999; Martín-Piera et al., 1996; Wagner, 2016; Whipple, 2011). However, there is a paucity of data related to the utilization of canine feces, particularly in urban or residential settings. Carpaneto et al. identified dung beetle species that have shifted their host associations from sheep to dogs due to changes in land use, decreases in local biodiversity, and a lack of alternative food sources (2005). Similarly, Ramírez-Restrepo et al. reported a potential shift towards dog feces in an urban setting (2016). Neither study examined the attractiveness of canine feces relative to other hosts.

The rate of dog ownership has increased markedly among industrialized countries in recent years (Cinquepalmi et al., 2013). This trend naturally corresponds with an increase in the amount of pet waste generated in an urban environment (Nemiroff et al., 2007; Ramírez-Restrepo et al., 2016). It has been estimated that approximately 10 million tons of canine and feline fecal waste is produced annually in the United States alone (as cited in Nemiroff et al., 2007). Many municipalities have ordinances that require the prompt removal and disposal of pet waste; however, it is not uncommon to encounter unmanaged waste in public or residential settings.

Unmanaged animal waste can pose health risks for humans and animals alike. Feces can contain microorganisms that are pathogenic (e.g. *Campylobacter*, *E. coli* O157:H7, *Listeria*, *Leptospira*) or resistant to antibiotics. The genes responsible for conferring resistance are readily transferred between humans and animals, compounding the potential threat to public safety (Sobsey et al., 2006). Furthermore, animal waste may contain parasites, such as *Giardia* and *Cryptosporidia* (Cinquepalmi et al., 2013). The close physical proximity between pets and their owners increases the likelihood that pathogens and parasites will be transmitted to humans (Damborg et al., 2016).

Animal waste is readily transported to local waterways via runoff, posing a threat to water quality in urban and recreational settings. Contamination of urban water systems decreases overall quality, limits recreational value, and contributes to the formation of algal blooms capable of killing aquatic organisms (as cited in Whipple, 2011). One study found that 95% of coliform bacteria present in urban storm water was not of human origin; the majority of these organisms were determined to be of canine or livestock origin (as cited by the Center for Watershed Protection, 1999).

The present study was designed to characterize native arthropod populations that live in association with canine feces; due to the ecological services rendered by dung beetles, an emphasis was placed on the identification of species that are associated with waste recycling. It was hypothesized that Lancaster County, NE supports a low level of paracoprid dung beetles; due to the high desiccation rate of canine feces, this nesting strategy is thought to be favored over others (Carpaneto et al., 2004). Furthermore, it was predicted that arthropods commonly found in association with vertebrate feces (e.g. calliphorid and sarcophagid species) would be recovered during rearing experiments. Tourist species, such as ants, were expected be recovered in pitfall traps due to their widespread terrestrial activity. The results of this evaluation were then used to make recommendations for future studies, with the ultimate goal of reducing pet waste in urban, recreational, and residential settings through augmentation.

Monitoring was conducted using pitfall traps and rearing experiments. Pitfall traps are commonly used to estimate the diversity and abundance of ground-dwelling arthropods. Rearing experiments were performed to better characterize species that dwell within feces, as well as those that would otherwise not be captured by pitfall traps.

Materials and Methods

Study sites. Studies were conducted at three sites located in Lancaster County, Nebraska:

Location 1 is a residential property in Waverly, NE that was constructed in 1998; prior to this time, the area was zoned for agricultural use. The neighborhood has few trees and landscaping is minimal. The area immediately behind the property is currently undeveloped and undergoes minimal maintenance. The property owners and several residents in the surrounding neighborhood have dogs as pets.

Location 2 is a residential property in Waverly, NE that was constructed in 1977. The neighborhood has numerous trees that are well established, and the landscaping is extensive. The property is surrounded by other homes on all sides. Nearby residents have dogs and cats, although the property owners do not have pets.

Location 3 is a heavily wooded area located on the outskirts of Lincoln, NE that experiences little maintenance or human traffic throughout the year. A small creek runs nearby, making the area prone to flooding after heavy rains. Nearby property owners have dogs; the region also supports a variety of waterfowl, and intermittent coyote activity has been reported.

These sites were selected to provide information for a variety of habitats located within southeastern Nebraska.

Pitfall trap monitoring. A preliminary evaluation was conducted at Location 1 using two trap designs; see Appendix A for a detailed account of this work. While no dung beetle activity was observed during this evaluation period, the open trap design was selected for use in the official study due to its ease of use and the comparatively high number of organisms recovered.

Two pitfall trap designs were selected for use in the official study. For the first design, traps were constructed as described in Appendix A (Figure A3B). The second design was identical in construction, but omitted the bait pouch to evaluate whether it had a negative impact on the attractiveness of feces (Figure 1). Monitoring was conducted at the three study sites listed above. Traps were placed at intervals of at least 3 m; for each study site, two replicates were tested per trap design, and one non-baited trap was included to provide baseline data (i.e. total of five traps set per site).

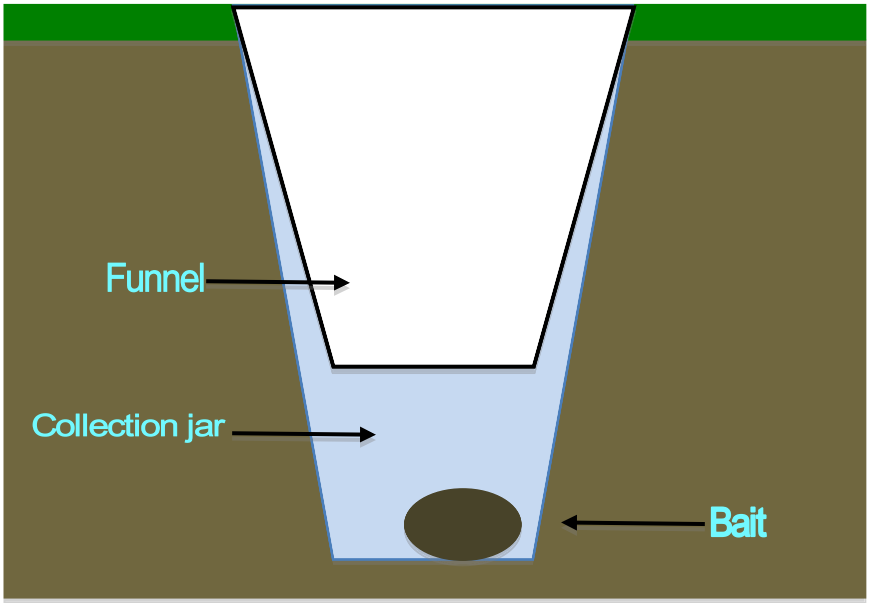

Figure 1. Pitfall trap diagram, omitting bait pouch.

Feces used to bait traps were collected within two hours of deposition; based on personal observations made during the preliminary evaluation period, this timing minimizes desiccation and correlates with higher arthropod activity. Throughout the study, unused feces were removed from the surrounding area on a daily basis to concentrate arthropod activity near the traps. Preservatives, such as ethylene glycol, were not used to ensure the safety of pets and other animals in the vicinity.

Monitoring was conducted during the months of July – September 2017. Traps were observed daily for arthropod activity; due to the high rate of desiccation observed, baits were collected and replaced daily. At each monitoring site, the placement of each test condition (i.e. baited/pouch, baited/no pouch, and non-baited control) was periodically rotated to account for local variation in arthropod activity and eliminate position bias. Following collection, baits were carefully examined for the presence of organisms dwelling on the surface of and within the feces; the number of organisms recovered per trap was tabulated. Where possible, scarab beetles were identified to the species level according to the identification guide published by Ratcliffe et al. (2008). All other insects were identified to the order or family level, and non-insect arthropods were identified to at least the class level according to the guide published by Borror et al. (2005).

Rearing experiments. Enclosures used for rearing studies were constructed using a modification of the method recommended by Hardy et al. (1982, pp. 1-3). Plastic containers (3.5" H x 4.5" D x 7" W; 1,890 mL capacity) were filled with approximately 2 cm of sand, followed by 3 cm of soil that was obtained from Location 1. The soil was packed and lightly moistened with water. Feces were collected 0, 1, 2, 4, 6, and 8 hours following deposition and were placed into separate enclosures. A control enclosure was prepared as described above, omitting the feces. Lids were placed over the opening of each container, and enclosures were stored outdoors at ambient temperature. Containers were observed periodically to assess the moisture level and evaluate arthropod activity. All rearing experiments were conducted during the months of August and September 2017.

Feces used for pitfall trap and rearing experiments were obtained at Location 1 from dogs receiving preventative treatment for internal parasites, fleas, and ticks. Each dog was administered 1000 mg fluralaner (Bravecto®; Merck), 23 mg milbemycin oxime, and 460 mg lufenuron (Sentinel® flavor tabs®; Virbac) according to label instructions. No other medications were administered throughout the study period. Animals are medium-sized dogs of mixed breed, and were fed a standard diet consisting of commercial dry dog food. Scarab and hister beetle activity was periodically observed on feces prior to collection for use. All beetles observed were removed before traps were baited to avoid bias.

Results

Pitfall traps. A total of 78 scarab beetles from seven species were collected over the course of 26 trap-days (Table 1). *Copris fricator* (Fabricius) was the species most commonly recovered at Locations 1 and 2, while the primary species collected at Location 3 was *Pseudocanthon perplexus* (LeConte). At Locations 1 and 2, scarab beetles demonstrated a preference for traps baited with uncovered feces rather than a baited pouch. No clear preference was observed at Location 3.

The recovery of *C. fricator* at Location 1 peaked in mid-August (Figure 2). No other trends could be seen in the prevalence of individual species at any of the three sites evaluated. The overall abundance of scarab beetles also peaked in mid-August (Figure 3A). No correlation was observed between overall abundance and either temperature or precipitation during the study period (Figure 3A-C). Weather data obtained from the National Oceanic and Atmospheric Administration for the Lincoln, NE airport is provided in Appendix B. This airport is located 10-14 miles from each of the three study sites.

Aside from scarabaeids, arthropods from a total of four classes and ten different orders were collected during this study. The most common orders recovered from baited traps at each site were: Orthoptera and Carabidae (Location 1), Staphylinidae and Araneae (Location 2), and Carabidae and Araneae (Location 3). It is important to note that Araneae were collected in non-baited control traps at a frequency similar to or greater than that seen for baited traps at each site.

Table 1. Total Number of Arthropods Collected Per Location

Identification	**Location 1**			**Location 2**			**Location 3**		
	Control	**P**	**N**	**Control**	**P**	**N**	**Control**	**P**	**N**
Class Arachnida: Subclass Acari	1	1	0	0	2	2	0	0	0
Class Arachnida: Order Araneae	19	13	19	14	31	25	20	14	34
Class Diplopoda	0	0	0	0	1	0	0	0	0
Class Insecta									
Order Coleoptera: Carabidae	6	21	57	0	1	0	14	42	64
Order Coleoptera: Chrysomelidae	0	0	1	0	1	0	0	0	0
Order Coleoptera: Dermestidae	0	2	3	0	3	6	3	14	9
Order Coleoptera: Elateridae	0	0	3	0	0	0	1	0	0
Order Coleoptera: Histeridae	0	0	1	0	0	0	0	0	0
Order Coleoptera: Lampyridae (larva)	0	0	0	0	0	0	0	1	0
Order Coleoptera: Meloidae	0	0	0	0	0	1	0	0	0
Order Coleoptera: Nitidulidae	0	1	0	0	0	4	1	1	0
Order Coleoptera: Scarabaeidae									
Ataenius spretulus (Haldeman)	0	0	0	0	0	1	0	1	0
Copris fricator (Fabricius)	0	10	40	0	5	5	0	0	0
Onthophagus hecate (Panzer)	0	0	2	0	0	3	0	0	0
O. orpheus pseudorpheus (Howden & Cartwright)	0	1	2	0	0	0	0	0	0
Phyllophaga sp.	0	0	1	0	0	0	0	0	0
Popillia japonica (Newman)	0	0	0	0	1	1	0	0	0
Pseudocanthon perplexus (LeConte)	0	0	0	0	0	3	0	2	2
Order Coleoptera: Staphylinidae	0	4	8	0	20	41	2	4	8
Order Coleoptera: Trogidae	0	0	1	0	0	0	0	0	0
Order Diptera	3	0	2	1	0	10	1	1	3
Order Hemiptera: Cercopidae	0	0	4	0	0	0	0	0	0
Order Hemiptera: Lygaeidae	0	0	3	0	0	0	0	0	0
Order Hemiptera: Pentatomidae	0	0	0	0	0	2	0	0	0
Order Hemiptera: Tingidae	0	2	1	0	0	0	0	0	0
Order Hymenoptera	2	26	25	2	27	18	0	0	0
Order Lepidoptera	0	1	1	0	1	4	1	1	0
Order Orthoptera	7	31	69	1	10	12	5	6	8
Class Malacostraca: Order Isopoda	5	8	6	0	22	25	0	0	1
Total No. Organisms Collected per Test Condition	**43**	**121**	**249**	**18**	**125**	**163**	**48**	**87**	**129**

Control: Non-baited Trap (One Replicate) **P**: Baited Trap/Pouch (Two Replicates) **N**: Baited Trap/No Pouch (Two Replicates)

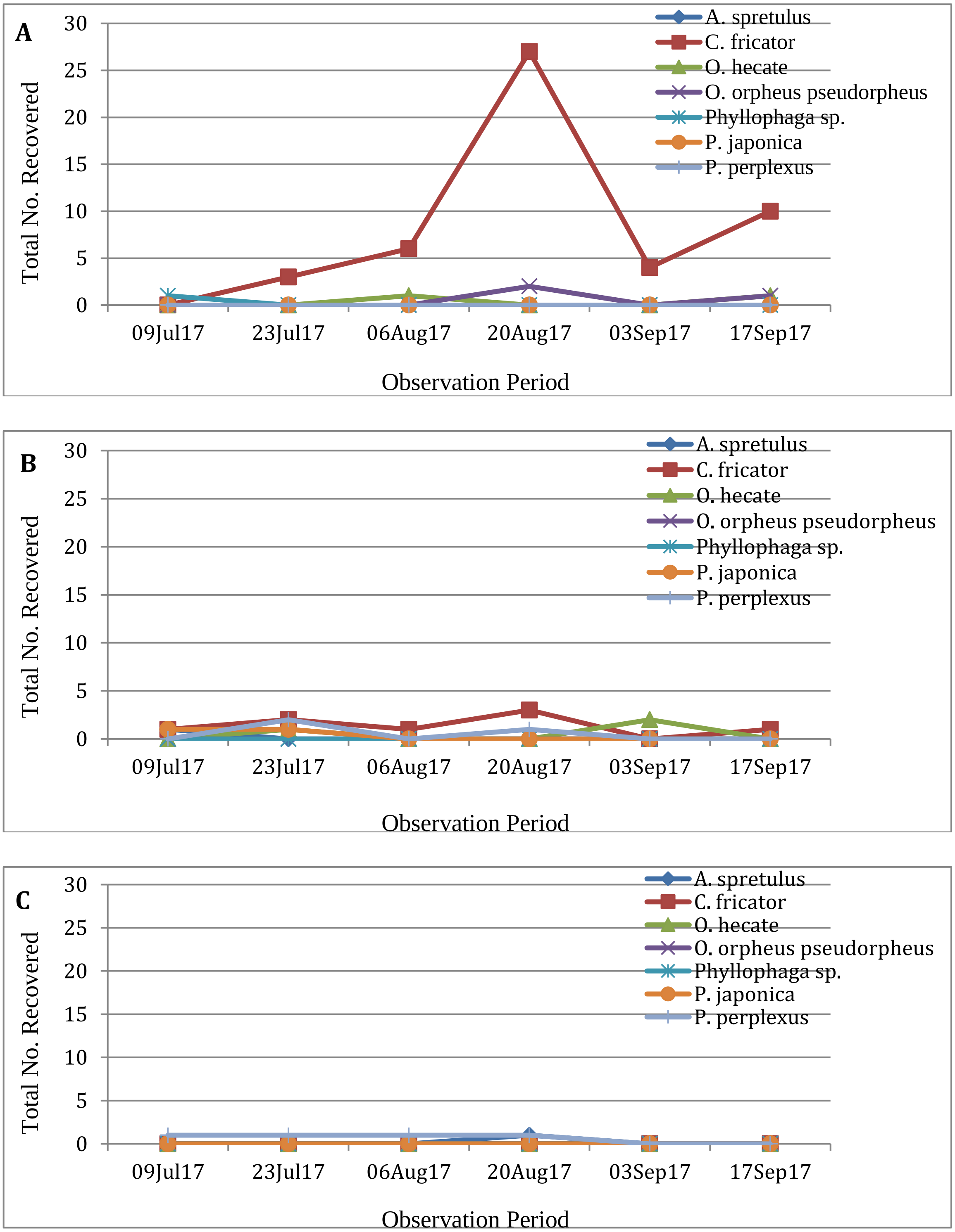

Figure 2. Total number of each scarab beetle species collected at A) Location 1, B) Location 2, and C) Location 3.

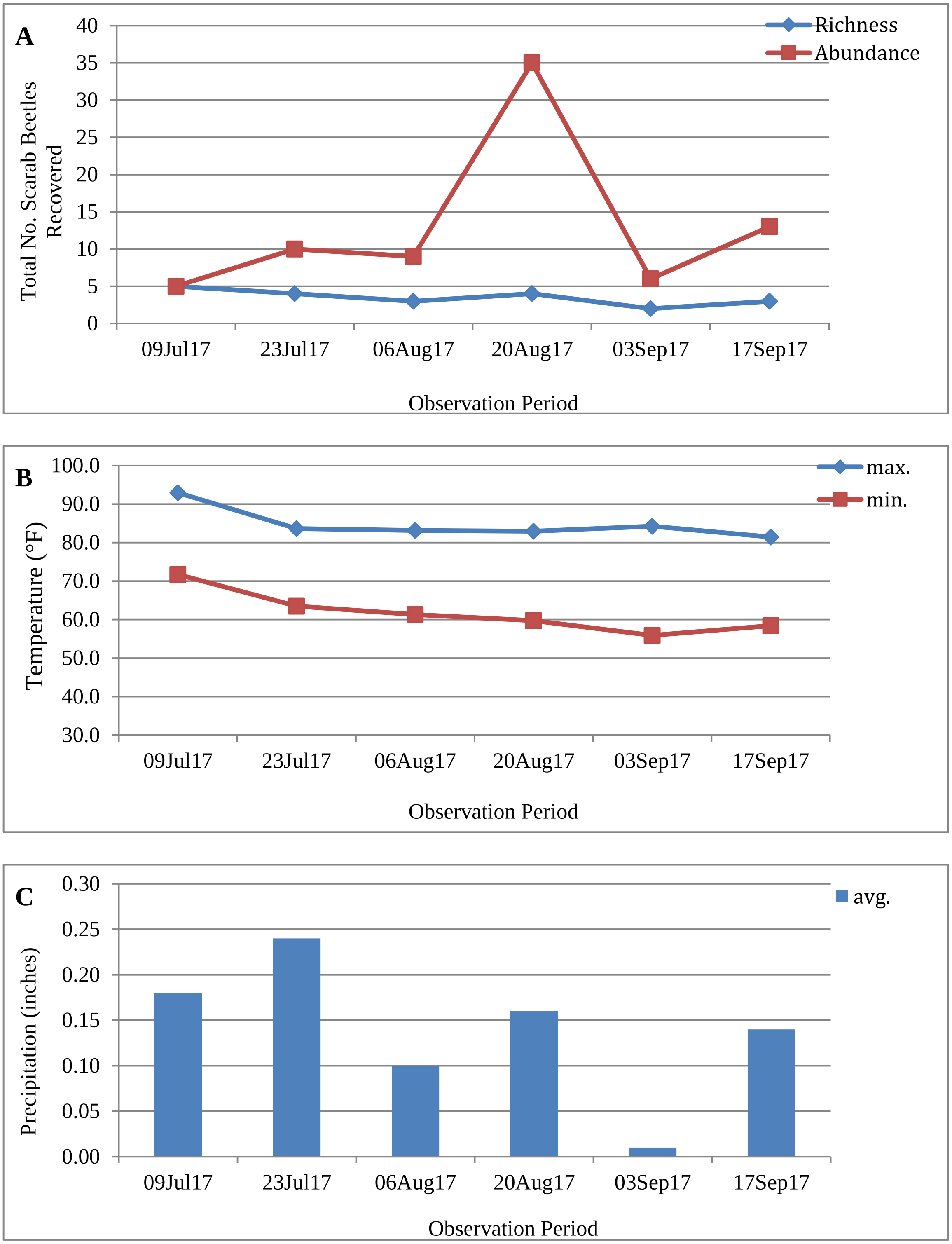

Figure 3. Scarab beetle and abiotic dynamics at each study location. A) Overall species richness and abundance; B) Minimum and maximum temperatures, and C) Average precipitation for July – September 2017.

A strong correlation was observed between scarab beetle recovery and study site, with more than 70% of the specimens collected at Location 1. A total of four species were collected at Location 1, five species at Location 2, and two species at Location 3 (Table 2).

Table 2. Percent Abundance of Scarab Beetle Species

	% Total		
Species	Location 1	Location 2	Location 3
A. spretulus (Haldeman)	0.00	5.88	20.00
C. fricator (Fabricius)	89.29	47.06	0.00
O. hecate (Panzer)	3.57	17.65	0.00
O. orpheus pseudorpheus (Howden & Cartwright)	5.36	0.00	0.00
Phyllophaga sp.	1.79	0.00	0.00
P. japonica (Newman)	0.00	11.76	0.00
P. perplexus (LeConte)	0.00	17.65	80.00
Sum	100	100	100
Total Number of Scarab Beetles Collected	56	17	5
Total Number of Species Collected	4	5	2

Voucher specimens of the scarab beetles collected were deposited with the Entomology Department at the University of Nebraska in Lincoln, NE. Due to an oversight, the *Phyllophaga* specimen was not retained; for this reason, it could only be identified to the genus level. Photographs of representative specimens are provided in Appendix C.

Rearing experiments. After 51 days of incubation, no arthropod activity was observed in the control enclosure (i.e. no feces). For all remaining conditions tested, the fecal matter developed mycelial growth within 2-3 days. Two attempts were made to perform these experiments as described above, with each attempt resulting in an overgrowth of fungus. During the exposure period, dipterans in the families Calliphoridae, Sarcophagidae, Scatopsidae, and Scathophagidae were routinely observed. No other data could be recovered from this effort.

Discussion

This study confirmed that several scarab beetle species native to southeastern Nebraska are attracted to canine feces. *Copris* and *Onthophagus* species are tunnelers (Carpaneto et al., 2005), while *P. perplexus* is a telecoprid (Reyes et al., 2007). As explained by Carpaneto et al., canine feces are prone to desiccation and undergo rapid microclimatic alterations; these qualities favor organisms that remove and immediately bury dung over those that dwell within the feces (2005). The results obtained from this study appear to support this observation. The remaining scarab beetles collected (*A. spretulus*, *Phyllophaga*, and *P. japonica*) are phytophagous.

Other specimens captured by pitfall traps, such as trogid, hister, and dermestid beetles, are known to feed on decaying organic matter (Borror et al., 2005), indicating that these species play minor roles in the recycling of canine waste. However, many organisms recovered during this study are predaceous (e.g. carabids, arachnids) or phytophagous (e.g. pentatomids, tingids, cercopids) (Borror et al., 2005). It is likely that these specimens have an indirect association with canine feces (e.g. as a predator of waste recyclers) or are tourists that were inadvertently collected. No conclusions about the diversity of arthropods dwelling within canine feces could be drawn from the rearing efforts described above; however, dipteran species commonly associated with vertebrate dung were observed during these activities.

Limitations of These Studies

Limitations can result from sampling constraints, study design flaws, host attributes, and information gaps. Each constraint will be examined in greater detail below.

Sampling limitations. This study likely underestimated the number and diversity of organisms attracted to canine feces. The capture efficiency of pitfall traps is affected by factors such as trap size and shape, materials of construction, environmental factors, activation time, and the number of traps used; additionally, pitfall sampling is more reliable when trap designs are optimized for a particular habitat and application (Cheli et al., 2010). Furthermore, sampling was only conducted over a three-month period at three different sites; in order to obtain more robust data, sampling must be conducted over the span of multiple years and at numerous sites to account for seasonal and regional variation.

Pitfall traps are traditionally used to sample ground-dwelling organisms; however, they do not provide reliable data about other arthropod populations. Based on personal observations, highly mobile insects, such as dipterans and cercopids, were able to freely move in and out of the traps. Additionally, soil-dwelling organisms (e.g. Collembola) were difficult to enumerate due to their ability to disperse through drainage holes; such specimens were excluded from the study for practicality. Alternative methods may enhance the sampling accuracy and more fully characterize organisms that are attracted to canine dung.

In the present study, scarab beetle recovery was highest for pitfall traps that were constructed as depicted in Figure 1 (i.e. no bait pouch). Dr. Brett Ratcliffe (personal communication) indicated that the pore size of towels used to construct these pouches might have been too small. Dung beetle orientation is largely accomplished through the olfaction of volatile compounds released by feces (Dormont et al., 2010). As such, the fabric may have interfered with their ability to detect host odors, negatively impacting the number of beetles recovered.

Study design limitations. All attempts to rear organisms dwelling within feces were unsuccessful. Dr. Ratcliffe (personal communication) suggested that these results were not surprising: numerous efforts to rear dung beetles have failed due to the difficulty of mimicking natural conditions. It is important to note that feces were left exposed during the daytime prior to collection. If rearing efforts had been successful, results would only have reflected diurnal species attracted to canine feces.

Host limitations. Dogs associated with this study were administered preventative medications that are known to be toxic to insects. Commercial veterinary drugs often contain active ingredients that have a negative impact on arthropod feeding or survival. Ivermectin, for example, causes sensory and locomotive problems in dung beetles at low concentrations, impairing olfaction, foraging behavior, and intraspecific communication (Verdu et al., 2015); this compound has also been shown to reduce survival and fecundity rates in some species (Cruz Rosales et al., 2012). Ivermectin has a slow decomposition rate, with 62-98% of the active ingredient excreted without degradation (as cited in Verdu et al., 2015). Due to its environmental persistence, ivermectin may exert effects on non-target organisms attracted to feces over an extended period of time (Madsen et al., 1990). The two active ingredients in Sentinel®, milbemycin oxime and lufenuron, are a GABA agonist and inhibitor of chitin synthesis, respectively (Yu, 2014). As a GABA agonist, milbemycin oxime is expected to exert effects similar to those described above for ivermectin. In contrast, Bravecto® contains fluralaner, which acts as a GABA antagonist (Yu, 2014). Differences in the chemical and nutritional composition of a canine's diet may also influence dung beetle preference (Scholtz et al., 2009). The impact of canine medications or diet on the results obtained cannot be properly evaluated without further studies.

Informational limitations. Two Japanese beetles (*Popillia japonica*) were recovered at Location 2 near grapevines; several adults were also observed in the surrounding area on multiple sampling dates. For these reasons, the property owner administered Sevin® (Bayer) on 07Aug17 to susceptible host plants according to label instructions. This insecticide, a member of the carbamate family, is a neurotoxin that affects movement and orientation in sensitive species (Haynes, 1988; Yu, 2014). Sevin® demonstrates high environmental persistence and is resistant to translocation via rainfall. Due to the wide spectrum toxicity of carbamates for a variety of arthropods (Hulbert et al., 2011), the impact of this treatment on subsequent monitoring conducted at Location 2 is unknown. No pesticides or herbicides were applied at Location 1 during the course of this study. No information is available for Location 3, or for areas adjacent to any of the three study sites.

Conclusions and Future Study

Unmanaged animal waste poses a threat to public health, and is a potential source of contamination for water systems. As such, the prompt removal of pet waste from urban and residential settings is critical. Dung beetles play an important role in the elimination of fecal matter from the environment; their feeding activity reduces the persistence of waste, suppresses pest and parasite populations, and diminishes the amount of feces available for transport to local waterways (Boze et al., 2012). Dung beetles also act as ecosystem engineers, making resources available to other organisms through changes in the biotic or abiotic environment (Boze et al., 2012). Preservation of these services is vital for the healthy functioning of an ecosystem.

The ultimate goal of this project is to identify future studies needed to assess whether native dung beetle populations can be augmented to facilitate the removal of feces. First and foremost, this strategy requires a robust method for mass rearing of dung beetles. In the present study, all efforts to rear arthropods from canine feces were unsuccessful. Artificial enclosures can produce microclimates that are unrepresentative, or they may alter an organism's behavior (Speight et al., 2008). As such, future efforts should be adjusted to more closely mimic natural conditions.

Due to the limited scope of this evaluation, additional sampling should be performed at more sites over a longer period of time. Also, alternative trap designs should be considered. In this study, the bait pouch itself appeared to have a negative impact on scarab beetle recovery at two of the study sites; design modifications may further improve recovery and provide a more thorough depiction of coprophagous arthropods. If results are found to be reproducible, efforts should be made to identify the environmental attribute(s) responsible for increased scarab beetle recovery at Location 1.

Similarly, the accuracy of these results would improve with repetition over multiple years to account for climatic variation. Temperature and moisture content dictate the number and types of insects that are able to exploit this resource, with microbial activity altering fecal quality over time (Hanski, 1987). As explained by Hanski (1987), "physiochemical and biotic conditions in these microhabitats change rapidly and select for fast exploitation" (p. 838). Weather conditions, such as rain, snow, frost, sun, and wind, ultimately determine how long the resource will remain attractive (Lumaret et al., 1992). Likewise, dung beetles often demonstrate a preference for the chemical composition, size, and desiccation rate of fecal matter (Lumaret et al., 1992; Nichols et al., 2008). As such, results may vary if traps are baited with feces from other dog breeds, or animals receiving different medications or diet.

Augmentation efforts may be confounded by several factors. Due to the spatial and temporal patchiness of fecal matter, long-term persistence of released organisms will likely require metapopulation-level dynamics (Roslin et al., 2001). Although clear preferences have been observed in some species, most coprophagous dung beetles are thought to be opportunistic feeders (Dormont et al., 2010; Halffter et al., 1999; Nervo et al., 2014). If food is patchy in distribution within a given habitat, it is likely that dung beetles will seek alternative sources that are more reliable.

Likewise, human activities directly or indirectly impact species richness and abundance within an ecosystem (Barragán et al., 2011; Ramírez-Restrepo et al., 2016). The widespread use of pesticides and herbicides has a deleterious effect on an ecosystem and can lead to the development of resistant populations (Speight et al., 2008). Similarly, changes in land use (e.g. agricultural intensification, urbanization, globalization) contribute to habitat degradation and instability, increasing the likelihood of local extinction events through fragmentation (Roslin et al., 2001). Scarabs are sensitive to environmental disturbances, and habitat preferences have been documented for several species (Roslin et al., 2001). Climate change is expected to cause shifts in insect distribution, alter the balance between competing species, and affect relationships on multiple trophic levels (Speight et al., 2008). Numerous authors have hypothesized that such factors may be responsible for past or future shifts in dung beetle-host associations (Barragán et al., 2011; Carpaneto et al., 2005; Radtke et al., 2008; Ramírez-Restrepo et al., 2016). Finally, anthropogenic drivers of change can act together in a synergistic manner to exacerbate the threat to biodiversity (Speight et al., 2008).

Augmentation, by definition, is intended to be a short-lived alteration to the composition of a given habitat (Hajek, 2004). However, any form of ecological manipulation has the potential to cause unintended consequences. Such introductions can alter inter- and intraspecific dynamics, potentially changing the community structure or impacting the overall functioning within an environment (Hajek, 2004). For these reasons, the intentional release of an organism should only be considered once a thorough ecological risk assessment has been completed. Augmentation efforts must also be performed in conjunction with public education to maximize the likelihood of success. It is hoped that this study can serve as a starting point for future evaluations, and that a feasible strategy for employing dung beetles for the removal of pet waste can be developed.

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Appendix A

Preliminary Evaluation of Pitfall Trap Designs

Pitfall traps were constructed from 16 oz. plastic drinking cups (Solo Cup Company Catalog No. P16RLR; internal diameter of 3.5 inches at the opening). Small holes (~2 mm diameter) were punched in the bottom of each cup to facilitate drainage. The bottom of a 7 oz. plastic cup (Solo Cup Company Catalog No. 806A) was cut off to produce a funnel. Bait containers were constructed by cutting cotton flour sacks into strips; the sides were sewn together to produce an open-ended pouch with approximate dimensions of 3.5” x 5” (Figure A1).

Figure A1. Photograph of representative bait pouch

Dog feces were collected within six hours of deposition and were loaded into bait pouches. Funnels were inserted into 16 oz. drinking cups such that the upper edge of a bait pouch was trapped between the funnel and cup, suspending the pouch near the opening (Figure A2A). Holes were dug into the ground so that the top of each trap was roughly flush with the soil surface. Plastic garden fencing (Quest Brands Inc. SKU 1721200; 1 ¼” nominal mesh size) was cut into ~5”x5” squares. These squares were secured over the opening of each trap using garden staples (Yardworks Catalog No. 268-9979) to occlude larger organisms (Figure A2B).

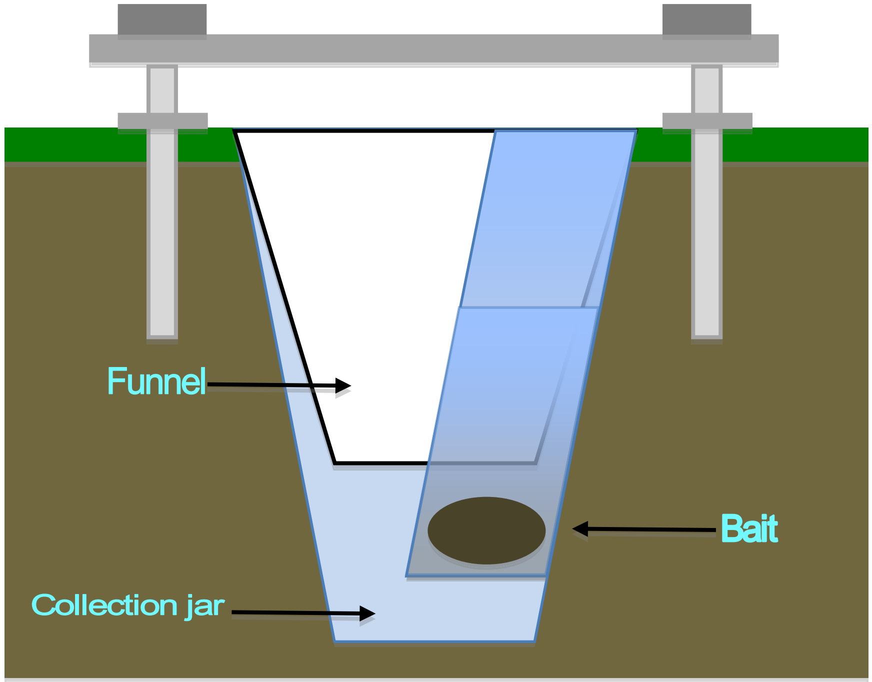
A  **B** 

Figure A2. Photographs of a constructed pitfall trap. A) Following insertion of bait pouch; B) Following installation into the ground.

Two trap designs were evaluated. For the first design, 6”x6” cover plates constructed from aluminum sheeting were secured over the mouth of a trap (Figure A3A). The second design omitted the metal roof (Figure A3B). No preservatives were used with either trap design.

Monitoring was conducted at Location 1 over a three-week period in June 2017. Traps were monitored daily for arthropod activity, and baits were replaced as deemed necessary. Throughout the evaluation period, unused feces were removed from the surrounding area on a daily basis to concentrate arthropod activity near the traps. Following collection, baits were carefully examined for the presence of organisms dwelling on the surface of and within the feces; the number of organisms recovered per trap was tabulated. Specimens were identified to the order or family level according to the identification guide published by Borror et al. (2005).

A



B

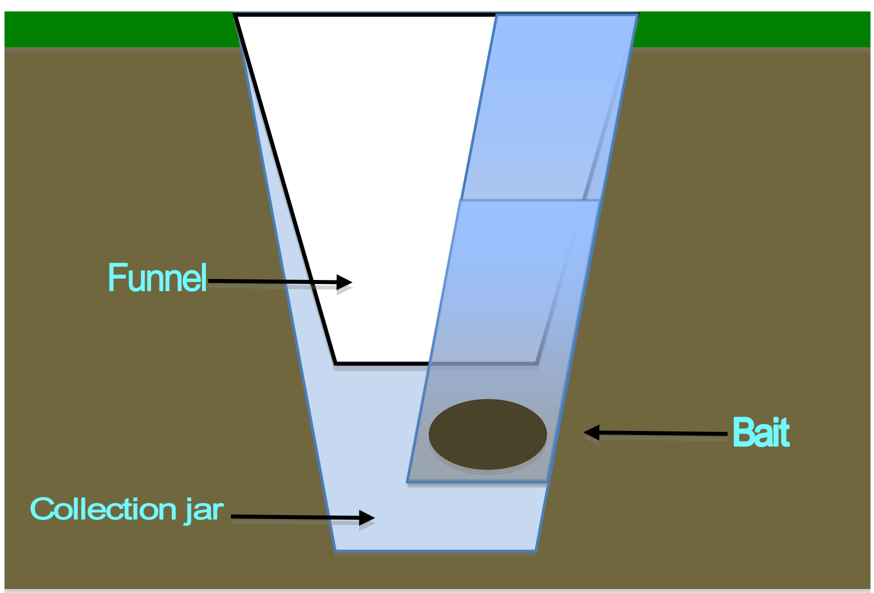
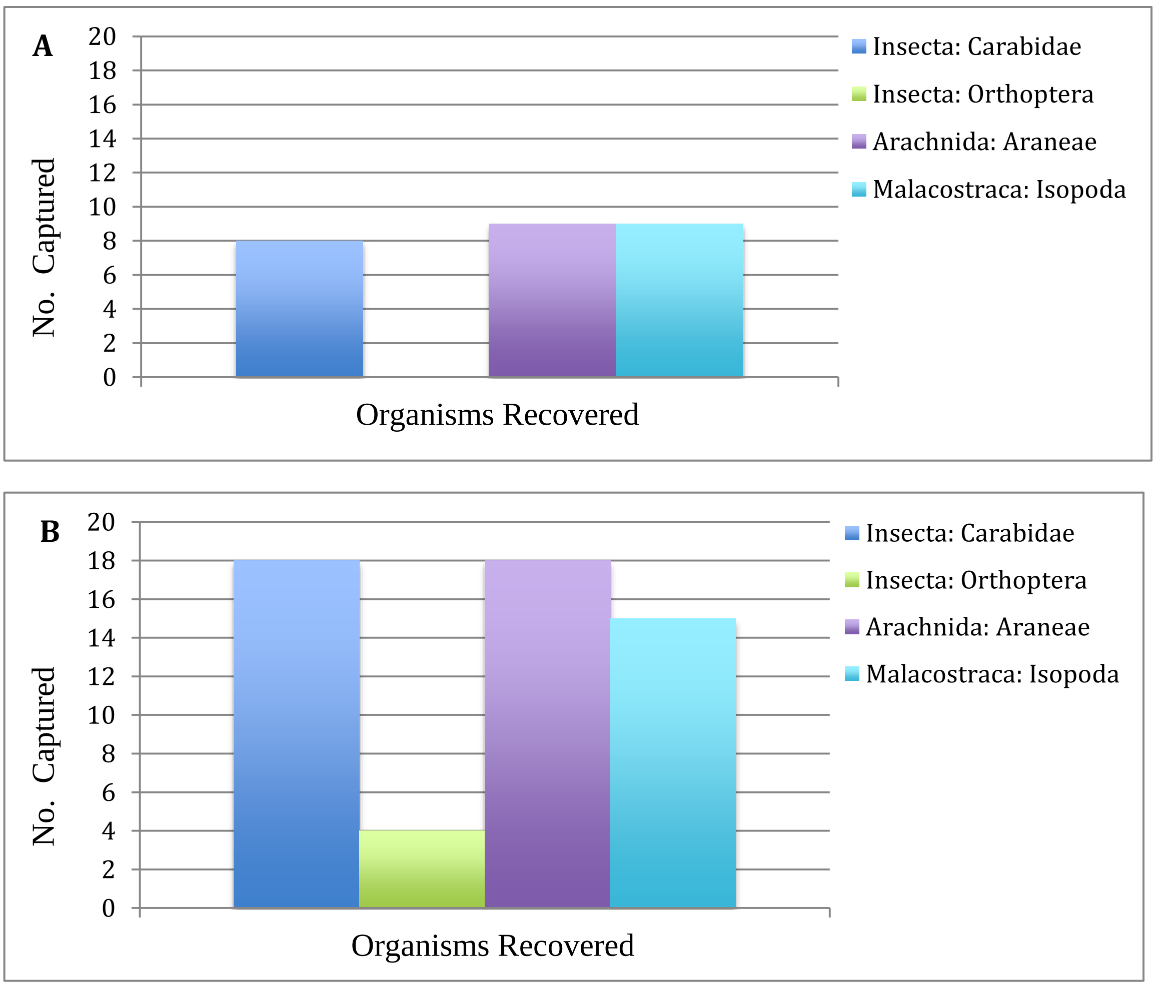


Figure A3. Pitfall trap diagrams. A) With metal roof; B) Without metal roof.

Results

No scarab beetles were recovered using either trap design. The overall species richness and abundance was higher for the trap that omitted the metal roof (Graph A1). Based on personal observations, more organisms were recovered with feces collected within two hours of deposition and when baits were replaced daily.



Graph A1. Preliminary evaluation of trap design. A) Metal roof; B) No metal roof

Appendix B

Weather Data Obtained from the National Oceanic and Atmospheric Administration

Date	Maximum Temperature (°F)	Minimum Temperature (°F)	Precipitation (inches)
09Jul17	95	68	0.00
10Jul17	93	77	0.00
11Jul17	97	73	0.00
12Jul17	89	70	1.32
13Jul17	84	69	0.87
14Jul17	88	65	0.00
15Jul17	91	70	0.00
16Jul17	95	69	0.00
17Jul17	93	71	0.00
18Jul17	91	72	0.15
19Jul17	96	75	0.00
20Jul17	96	74	0.00
21Jul17	100	79	0.00
22Jul17	93	72	Trace Amount
23Jul17	91	65	0.00
24Jul17	89	62	0.00
25Jul17	97	71	0.00
26Jul17	83	72	2.83
27Jul17	84	66	0.00
28Jul17	84	63	0.00
29Jul17	83	64	0.00
30Jul17	81	68	0.00
31Jul17	82	63	0.00
01Aug17	87	65	0.00
02Aug17	88	64	0.00
03Aug17	77	56	Trace Amount
04Aug17	79	50	0.00
05Aug17	69	60	0.26
06Aug17	75	62	0.00
07Aug17	81	59	0.00
08Aug17	83	56	0.00
09Aug17	77	61	Trace Amount
10Aug17	82	59	0.00
11Aug17	81	57	0.01
12Aug17	83	55	0.00
13Aug17	83	65	0.07
14Aug17	87	66	0.60
15Aug17	89	68	0.01
16Aug17	83	66	0.41
17Aug17	85	62	0.00
18Aug17	87	64	Trace Amount
19Aug17	88	58	0.11
20Aug17	87	65	1.82

Appendix B

Weather Data Obtained from the National Oceanic and Atmospheric Administration, *Continued*

Date	Maximum Temperature (°F)	Minimum Temperature (°F)	Precipitation (inches)
21Aug17	85	67	Trace Amount
22Aug17	78	56	0.00
23Aug17	80	53	0.00
24Aug17	82	58	0.00
25Aug17	84	65	0.01
26Aug17	88	66	Trace Amount
27Aug17	84	63	0.08
28Aug17	79	56	0.00
29Aug17	80	54	0.00
30Aug17	83	52	0.00
31Aug17	85	57	0.00
01Sep17	80	61	0.01
02Sep17	86	63	0.00
03Sep17	91	61	0.00
04Sep17	79	62	0.00
05Sep17	72	48	0.00
06Sep17	72	43	0.00
07Sep17	83	45	0.00
08Sep17	89	51	0.00
09Sep17	84	59	0.00
10Sep17	85	65	Trace Amount
11Sep17	86	59	0.00
12Sep17	86	55	0.00
13Sep17	90	55	0.00
14Sep17	95	64	0.00
15Sep17	92	67	0.01
16Sep17	75	58	0.17
17Sep17	73	46	Trace Amount
18Sep17	76	63	0.27
19Sep17	92	66	0.00
20Sep17	82	60	0.00
21Sep17	95	59	0.00
22Sep17	97	75	0.00
23Sep17	93	70	0.00
24Sep17	88	63	0.59
25Sep17	64	57	0.79
26Sep17	66	52	Trace Amount
27Sep17	74	46	0.00
28Sep17	80	48	0.00
29Sep17	81	51	0.00
30Sep17	79	62	0.01

Appendix C

Photos of Representative Specimens Collected

Guide to photo list:

1. *Ataenius spretulus*
2. *Copris fricator*
3. *Onthophagus hecate* (♂)
4. *Onthophagus hecate* (♀)
5. *Onthophagus orpheus pseudorpheus*
6. *Popillia japonica*
7. *Pseudocanthon perplexus*

1.

2.

3.

4.

5. 

6. 

7.