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A COMPARISON OF PAN TRAP AND BLUE VANE SAMPLING METHODS FOR DETERMINING BEE DIVERSITY

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Abstract

Bees (family: Apidae) are important ecologically and economically as primary pollinators of many natural and agricultural systems. Given current concerns about declining numbers of bees, many survey-oriented studies are being conducted to understand factors that contribute to the loss of bees and to direct conservation efforts. For these studies to be informative, researchers must be aware of biases associated with different sampling methods. Given their effectiveness at capturing a large number of insect individuals, blue vane traps are becoming a widely used method for sampling bees.

We investigated sampling biases associated with blue vane traps versus the more commonly used pan traps at two different high elevation sites in the Front Range of Northern Colorado. At each site, (1) sampling efficiency (number of species and abundance of bees associated with each method) were compared and (2) the overlap in species sampled by each method determined. We also examined the degree to which potential biases in the bees sampled by each method was associated with life history characteristics of the bees (level of sociality, floral specialization, nesting habits, and body size). At both surveyed sites, blue vane traps captured roughly 4 times more individuals and 1.5 times more species than paired sets of pan traps. Individual-based rarefaction curves, a calculation of species richness per individual sampled, however, showed that for an equal number of individuals sampled by both methods, pan traps would actually sample more species. To survey an equal number of individuals between trap types, it is recommended to employ 4 times as many sets of pan traps than blue vane traps. The degree of similarity between species associated trap types was

approximately 20%. Differences in species associated with each trap type were not caused by bee sociality, floral specialization, nesting habits, or body size. Given the differences in sampling efficiency and the low overlap of species captured by the two sampling methods, caution is suggested when comparing studies that employed only one of these methods. Rather, the use of both methods is suggested to better represent the species present within a given area.

Introduction

Insects, particularly bees, are important ecologically and economically due to their role as primary pollinators of most crops and plants. Over 35% of crops used in global food production require the services of pollinators like bees (Klein et al 2007), the value of which is estimated to be billions of dollars annually (NRC 2007). Alarm over evidence of global pollinator decline has prompted a myriad of studies assessing the degree to which factors such as habitat loss (Brown and Paxton 2009; Winfree et al. 2008), climate change (Williams et al. 2007), pesticide use (Holzschuh et al. 2007; Brittain et al. 2009), and parasites (Sammataro et al. 2000) might explain this loss. Because of their importance, bees have also become the focus of many studies to inform conservation efforts (Brown and Paxton 2009). However, these studies rarely take into account or acknowledge the sampling biases of their own studies or of those to which they compare their results. This is problematic because biases in the types of bees collected could lead to erroneous conclusions about the presence or absence of bees that may be independent of any potential factor being studied. Thus, it is important to develop a better understanding of bee sampling methods in an effort to improve the accuracy of study results.

Bee communities can be sampled using active (based on seeking out and catching bees) and passive (trapping) methods. Commonly used active sampling methods include a skilled person sweeping, hand netting, and vacuuming (Cane et al 2000; Roulston and Smith 2007), while passive sampling methods rely on bees being collected by the use of malaise, pan, and vane traps (Stephen and Rao 2005; Campbell and Hanula 2007; Kimoto et al. 2012). Although attempts have been made to

standardize sampling protocols among studies (LeBuhn et al 2003), no official and widespread standards exist. Rather, studies tend to vary in the combination of methods employed, sizes and characteristics of traps used, the number of replicates employed, and the frequency with which a site is surveyed (Matteson et al. 2008; Hopwood 2008), thus making results difficult to compare and interpret.

It is essential to acknowledge the documented sampling biases of different survey methods to understand the types of bees that may be over or underrepresented and the degree to which results of studies using different methods can be compared (Aguar and Sharkov 1997; Bartholomew and Prowell 2005; Kimoto et al. 2012). For example, it has been shown that oligolectic bees (that collect pollen from single plant genus) prefer certain pan trap colors (Waser and Price 1981; Haslett 1989) and that pan traps under sample the presence of bumblebees, honeybees (Roulston and Smith 2007) and cavity nesters (Westphal et al. 2008). Also, active sampling methods may be more strongly associated with observer bias, an unconscious influence by an investigator (Rosenthal and Fode 2007). Findings such as these play an essential role in improving studies and a better understanding their implications.

Introduced in 2005, blue vane traps are a relatively new passive sampling method that has grown in popularity due their ease of use and effectiveness at capturing large numbers of individuals (Stephen and Rao 2005, 2007; Kimoto et al 2012). Despite the increased popularity of blue vanes in current research, little is known about their sampling biases relative to other techniques. This is important because some surveys use only blue vanes (Stephen et al. 2009, Kimoto et al. 2012), thus remaining unaware of potential biases involved.

In the present study, we compare potential sampling biases associated with blue vane traps relative to pan traps; the most commonly used passive sampling method. To approach this question, we (1) compare sampling efficiency (number of species and abundance of bees associated with each method) and (2) determine the overlap in species sampled. We also examined the degree to which these two sampling methods caught bees with different life history characteristics, such as sociality, floral specialization, nesting habits, and body size. We sampled at two different sites to determine whether any patterns found were consistent. The results of this study will be useful in distinguishing advantages and/or biases of pan traps versus blue vane traps, which will inform researchers as to which method most accurately assesses abundance and diversity of bee populations.

Materials and Methods

Study Area

Differences in sampling efficiency and biases of pan traps and blue vane traps were assessed by sampling two montane bee communities located in the Front Range of northern Colorado, USA. These two sites are known as A1 (2195 m; 40°15'–105°376') and B1 (2591 m; 40°0219'–105°453') and are associated with the open meadow areas within the lower and upper montane life zones, respectively. The average yearly temperatures at A1 are higher than at B1 (7.93 vs. 5.98C at A1 and B1, respectively) and the season length is longer at A1 than B1 (164.5 vs 148 days at A1 and B1,

respectively) (McGuire et al. 2012). By personal observation, it appears that the plant communities differ between sites.

Bee Sampling

Bees were sampled once per week in the summer of 2014 (from May 6 to September 25), totaling 20 sampling rounds at each site. Three sets of traps were used during each survey with each set consisting of a blue vane trap and three plastic pan traps (one of each of the following colors: blue, yellow, and white) placed roughly 0.5 m apart. Each set of traps was placed in an open grassy area approximately 30 m apart to maximize their exposure to pollinators. Blue vane traps were attached to a wooden stake using a screw/band hose clamp with the base of the traps being roughly 35.5 cm off the ground. Blue vane traps consist of a 15 cm diameter x 15 cm height plastic collecting jar with a polypropylene funnel attached to two polypropylene cross vanes on top (SpringStar™ brand). An insect landing on a blue vane trap slides down the funnel and into the attached collecting jar where it is often unable to escape. Relative to a single pan trap, the collecting jar on a blue vane trap is extremely large. No pheromones or attractants were used.

The pan traps used in this study were plastic, 12 oz. Solo brand colored bowls (blue, yellow, and white) filled with approximately 140 mL of dish soap, water, and salt solution. Trap color preferences have been observed in bees (Leong and Thorp 1999), and to account for this variability, white, blue, and yellow bowls were used to reflect the most commonly chosen colors in studies that use pan traps. These colors are selected

to obtain a greater diversity of bees, as each color attracts different types. No additional paint or UV coating was used.

All traps were placed in the field during the early mornings (roughly 09:00 or 10:00h) and collected 48 hours later. This sampling strategy was employed because preliminary samples at these sites had suggested that the number of bees collected over a 24-hour period would be low. Once retrieved, all specimens were frozen until they could be sorted, pinned, labeled, and identified. All processed specimens are currently housed within the Entomology Section of the University of Colorado's Museum of Natural History.

Life history characteristics

For all species identified, information on sociality, floral specialization, and nesting habits were determined using the literature (Scott et al 2011). These functional groups are standard when studying bee life history characteristics (Williams et al. 2010, Rodriguez-Girones and Bosch 2012). For sociality, species were categorized as either eusocial (with cooperative brood care, overlapping generations, and division of labor), parasocial (species that share a single nest), solitary (do not live in colonies), or parasitic. For floral specialization, species were categorized as either polylectic (collect pollen from many unrelated plants), oligolectic (collects pollen from a single plant genus), monolectic (collects pollen from a single plant species), or other. For nesting habits, species were categorized as ground nesting, cavity nesting, or other. Bees were placed in an "other" category for both floral specialization and nesting habit if their

precise characteristic was unknown, if their identification was not specific enough to determine a more precise characteristic, or if their characteristic encompassed multiple or neither of the available options.

To estimate body size of each species, a single representative female bee specimen was selected and measured from each species caught. We chose to measure females, as they are typically involved in pollen collection and are more commonly represented than males. If a female of a given species or genus was not present, a specimen from the University of Colorado Entomology Collection was measured in its place. Of all body size measurements, 5.2% were estimated from using specimens from the Entomology Section of the University of Colorado. Body size of each species was measured with calipers and was defined as the intertegular span, the distance between the plates covering the base of the wings, an acceptable proxy for body size (Cane 1987). When male specimens could not be matched to a given species in the collection, the body size metric was excluded from the analysis. This occurred for approximately 16.6% of specimens.

Data analyses

To determine whether the efficiency of the two traps differed, both with respect to total number of individuals and specimens collected by each trap type, we used chi-square analyses with the expectation that the distribution of individuals and species would be 50% of the total for each trap type. To control for the number of individuals caught in each trap, individual-based rarefaction curves were generated. To determine

whether the likelihood of being caught by a given trap type was independent of selected life history characteristics, contingency table analyses were conducted in which all species captured by each trapping method were classified according to their levels of sociality (eusocial, parasocial, solitary or parasitic), levels of specialization on floral types (polylectic, oligolectic, monolectic, or other), or nesting habits (ground nesting, cavity nesting, or other). These analyses were conducted independently for each site to determine whether results were consistent between sites. To determine whether the size of bee species differed between trap types across both sites, we conducted a two way ANOVA with body size being explained by both trap type (pan or blue vane trap) and site (A1 or B1). Finally, we used the Jaccard's index to determine the similarity of bee species captured in each trap type at each site.

Results

A total of 361 bee specimens were collected which reflected a total of 57 potential species (33 taxonomic units were identified to the species levels and 24 additional ones were only identified to the genus level (Appendix 1). Of the specimens collected, 83.4% were identified to species and 16.6% to genus levels. Of the specimens collected, 1.3% were too damaged to identify and were excluded from analysis.

Trap efficiency

At both sites, blue vane traps captured roughly 4 times more individuals (A1, $\chi^2=51.496$, $P < 0.001$; B1, $\chi^2=15.074$, $P < 0.00001$) and 1.5 times more species (A1, $\chi^2=0.56$, $P = 0.45$; B1, $\chi^2=0.9$, $P=0.34$) than pan traps (Table 1). Given the greater sampling efficiency of blue vane traps relative to pan traps, more species were detected using blue vane traps. However, when controlling for the actual number of specimens collected in both the two trap types, pan traps were associated with more species, suggesting that while fewer individuals may be collected by pan traps, pan traps may actually be associated with a greater number of species caught (Gotelli and Colwell 2011) (Figure 1).

Sampling biases and life history characteristics

At both A1 and B1, more bee species were collected in blue vane traps than in pan traps. A comparison of the species present in these different traps showed that there was little overlap in the species present in blue vane traps versus pan traps at both A1 ($J=0.25$) and B1 ($J =0.15$) (Table 2). Since blue vane and pan traps exhibited little overlap in the number of species caught, are significantly dissimilar in terms of the number of species caught.

The dissimilarity of species present within each of the trap types could not be attributed to the specific life history characteristics of sociality, floral specialization, and nesting habits (Table 3). There was also no difference between mean body sizes of

species collected in blue vane traps versus pan traps ($F_{1,50} = 2.37$, $p = 0.13$) or between bees collected at the two sites ($F_{1,50} = 3.07$, $p = 0.09$) (Figure 2). There was, however, a weak trend towards bees caught at A1 being slightly larger than those caught at B1.

Discussion

In the present study, we compared the sampling efficiency and biases associated with the use of pan and blue vane traps. Blue vane traps were shown to catch more individuals and more species than pan traps. However, based on individual-based rarefaction curves, it was found that pan traps catch more species for a given number of individuals caught. Approximately 4 times as many pan traps need to be used to match the individual counts of blue vane traps. It can thus be concluded that pan traps catch a larger variety of bee species, but in smaller numbers. Consequently, pan traps sample bee diversity better than blue vane traps, but must be set out in greater numbers to compensate for low individual counts. Our data are consistent with the finding that pan traps have a low capture rate (Calabuig 2001).

It was also found that blue vane versus pan traps caught significantly dissimilar species. That is, a large difference exists between the kinds of species being caught in each trap. The dissimilarity was not explained by level of bee sociality, floral specialization, nesting habits, or body size. Given that these life history characteristics did not explain the dissimilarity between trap types, it is recommended that future work focus on other possible factors that might account for this difference.

Among the variety of species found, one noteworthy specimen—an individual identified as *Bombus occidentalis*—was caught in a blue vane at A1. *Bombus occidentalis* has experienced rapid decline since the 1990s (Cameron et al 2010) and is therefore of great interest. This finding

Though it was found that groups of bees caught in blue vane traps are significantly dissimilar from those in pan traps, it is still unclear what accounts for this difference. Possible areas of consideration for further research might include (1) the use of killing agents in blue vanes, (2) trap placement, and (3) UV paints. Firstly, this study did not use any killing agents. Since blue vane traps do not kill trapped specimens right away, it is possible that some species are able to escape and go unnoticed by researchers. Some potential factors that could cause this are body hair, bee flexibility, or shape. Secondly, in this present study all traps were placed on the ground. Results may differ if traps are elevated or arranged differently, as some studies do. Thirdly, the present study used standard Solo bowls as pan traps. It is possible that UV-fluorescent painted bowls, as are used in some studies (Saunders and Luck 2013), lead to different results. Considering these factors, (1) the use of killing agents in blue vanes, (2) trap placement, and (3) UV paints provide opportunities to better understand biases associated with blue vane traps relative to pan traps and potentially explain the biases between them. For example, killing agents in blue vanes may prevent some species from escaping, thus increasing the number of species. Traps elevated to different heights may capture bees with different foraging habits, as was shown in one study (Tuell and Issacs 2008). Lastly, sensory perception of different bee species may be sensitive to UV paints.

It is also important to acknowledge the potential for error in specimen identification in this study. Because sexually dimorphic bee species (with radical differences in the physical features of males and females) can easily be misidentified as two different species, this introduces some error into statistical analyses. Thus, efforts to more accurately identifying specimens might be worthwhile, although this can prove to be quite complicated in some species.

Conclusion

Though many studies exist with the intention of improving bee conservation efforts, they are subject to error if no consideration is given to the limitations of their sampling methods. In order to improve the accuracy of studies that survey bees, it is important to examine the effectiveness of commonly-used sampling tools. By gaining a better understanding of the biases inherent to different sampling methods, scientists can design better studies and obtain more reliable results. In summary, we found that blue vane traps catch more individuals than pan traps, that pan traps catch more species for a given number of individuals, and that, although traps are dissimilar in species caught, these differences are not due to bee sociality, floral specialization, nesting habits, or body size. Based on this information, we recommend that pan traps and blue vane traps be used together to best sample an area, and represent species that prefer a given type of trap. Pan traps should also be used in greater numbers to compensate for low individual counts—about 4 sets of pan traps for every blue vane trap. The present study, as well as additional studies of this kind, will improve sampling efforts of bee

communities and assist in assessing efficacy and biases associated with blue vane traps.

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Tables

Table 1. The number of species and individuals associated with blue vane trap and pan traps at A1 (1a) and B1 (1b).

1a

A1

	Blue Vane Trap	Pan Trap
# Species	26	19
# Individuals	160	29

1b

B1

	Blue Vane Trap	Pan Trap
# Species	22	14
# Individuals	81	26

Table 2. Numbers of unique and shared species present in blue vane traps and pan traps at A1 and B1.

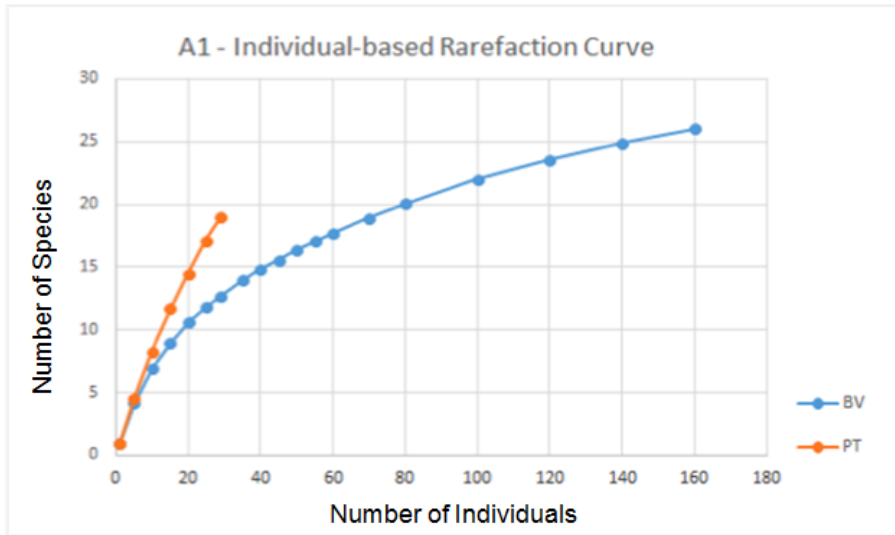
	A1		B1	
Total Species	36		30	
	Blue Vane	Pan Trap	Blue Vane	Pan Trap
Unique Species	17	10	16	8

Table 3. Chi-squared and p-values representing differences between the means of species numbers with regard to sociality (eusocial, parasocial, solitary, parasitic), floral specialization (polylectic, oligolectic, other), and nesting habits (ground, cavity).

Life History Characteristic	A1		B1	
	χ^2	p-value	χ^2	p-value
Sociality	5.063	0.17	2.262	0.52
Floral Specialization	0.47	0.79	1.421	0.49
Nesting Habit	0.86	0.65	1.106	0.57

Figures

1a



1b

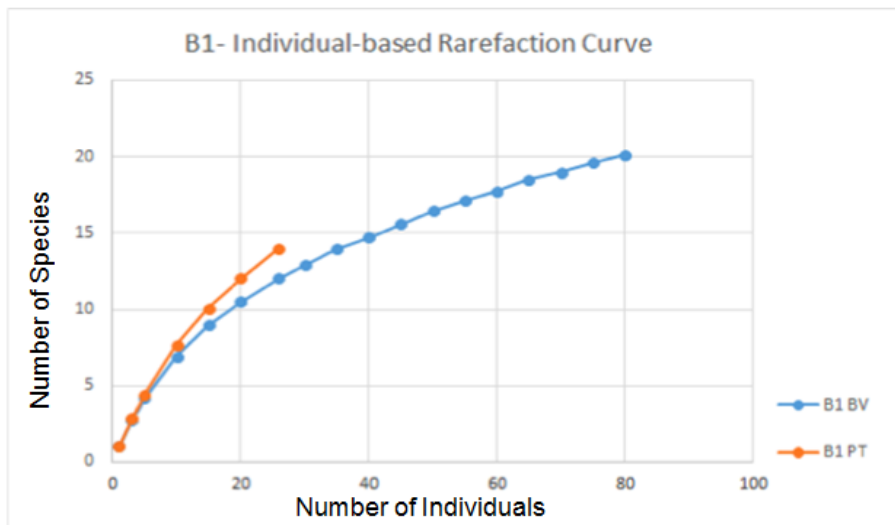


Figure 1. Individual-based rarefaction curves for sites A1 (1a) and B1 (1b).

2a

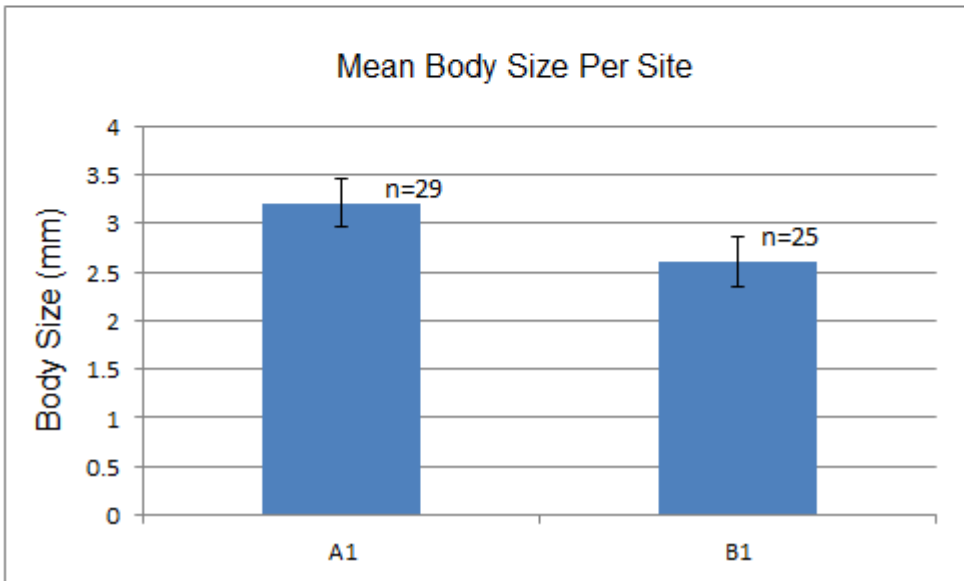
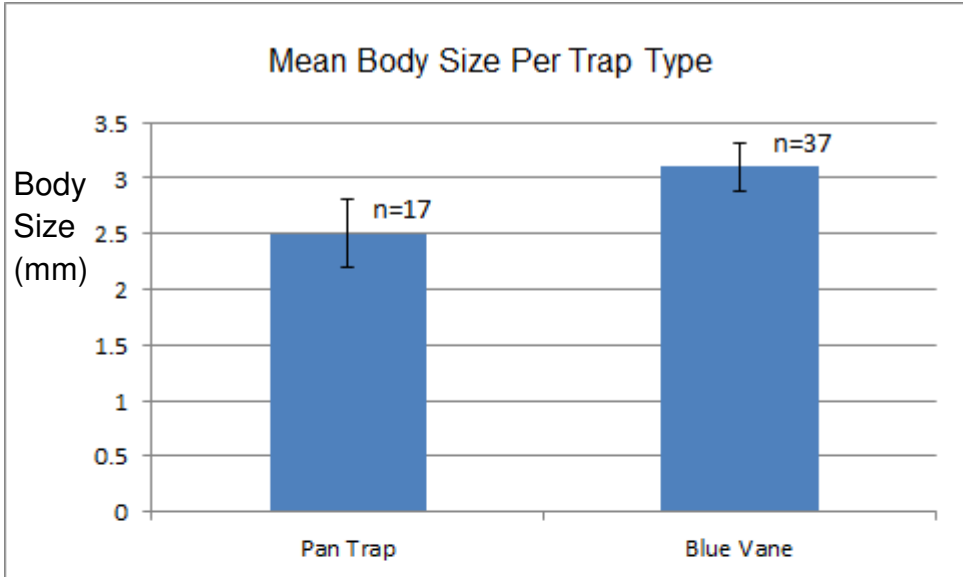


Figure 2. Mean body size comparisons between traps (2a) and between sites (2b).

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Appendix

Appendix 1. Species collection and life history information.

Species	BV A1	PT A1	BV B1	PT B1	Sociality	Nesting Habits	Floral Specialization	Female Size
<i>Bombus bifarius</i>	17	1	12	4	Eusocial	Cavity	Polylectic	3.57
<i>Osmia bucephala</i>	1	1	1	-	Solitary	Cavity	Oligolectic	4.1
<i>Melissodes #1</i>	1	-	-	-	Solitary	Cavity	Oligolectic	-
<i>Melissodes #2</i>	-	1	-	-	Solitary	Cavity	Oligolectic	-
<i>Melissodes #3</i>	2	-	-	-	Solitary	Cavity	Oligolectic	-
<i>Melissodes #4</i>	-	-	6	-	Solitary	Cavity	Oligolectic	-
<i>Melissodes #5</i>	2	-	3	1	Solitary	Cavity	Oligolectic	-
<i>Bombus insularis</i>	2	-	-	-	Parasitic	Cavity	Other	5.49
<i>Coelioxys</i>	-	-	-	1	Parasitic	Other	Other	-
<i>Triepeolus</i>	-	-	1	-	Parasitic	Other	Other	-
<i>Nomada sp.</i>	1	-	-	-	Parasitic	Other	Other	1.2
<i>Xeromalecta sp.</i>	1	-	-	-	Parasitic	Other	Other	-
<i>Stelis #1</i>	-	-	-	1	Parasitic	Other	Other	1.1
<i>Stelis #2</i>	-	1	-	2	Parasitic	Other	Other	1.8
<i>Stelis #3</i>	-	-	1	-	Parasitic	Other	Other	-
<i>Osmia montana</i>	-	-	-	1	Solitary	Cavity	Oligolectic	3
<i>Svastra obliqua</i>	2	-	-	-	Solitary	Ground	Oligolectic	3.58
<i>Melissodes agilis</i>	15	2	4	-	Solitary	Cavity	Oligolectic	2.2
<i>Dufourea maura</i>	5	2	1	-	Solitary	Ground	Oligolectic	2
<i>Lithurgopsis apicalis</i>	5	-	-	-	Solitary	Other	Oligolectic	3.8
<i>Hylaeus basalis</i>	-	-	-	1	Parasocial	Cavity	Polylectic	1.19
<i>Hylaeus #1</i>	-	-	-	2	Solitary	Cavity	Polylectic	0.84
<i>Andrena pronum</i>	-	1	-	-	Parasocial	Ground	Other	-
<i>Andrena #1</i>	-	-	-	1	Parasocial	Ground	Other	-
<i>Andrena #2</i>	-	2	-	-	Parasocial	Ground	Other	-
<i>Andrena #4</i>	-	-	-	4	Parasocial	Ground	Other	-
<i>Andrena #5</i>	-	1	-	-	Parasocial	Ground	Other	-
<i>Andrena #7</i>	-	1	-	-	Parasocial	Ground	Other	-
<i>Andrena #8</i>	-	1	-	-	Parasocial	Ground	Other	-

<i>Perdita #1</i>	-	-	1	-	Parasocial	Ground	Other	1.1
<i>Perdita #2</i>	-	-	1	-	Parasocial	Ground	Other	1
<i>Colletes #1</i>	1	-	-	2	Solitary	Ground	Other	2.81
<i>Colletes brevicornis</i>	1	-	-	-	Solitary	Ground	Other	2.71
<i>Colletes nigrifrons</i>	-	-	2	-	Solitary	Ground	Other	1.71
<i>Colletes #2</i>	1	-	-	-	Solitary	Ground	Other	
<i>Bombus nevadensis</i>	23	-	1	-	Eusocial	Cavity	Polylectic	5.4
<i>Bombus appositus</i>	31	1	9	-	Eusocial	Cavity	Polylectic	4.69
<i>Bombus centralis</i>	13	-	12	-	Eusocial	Cavity	Polylectic	3.55
<i>Bombus flavifrons</i>	1	-	-	-	Eusocial	Cavity	Polylectic	3.36
<i>Bombus californicus</i>	3	-	4	1	Eusocial	Cavity	Polylectic	3.5
<i>Bombus fervidus</i>	2	-	-	-	Eusocial	Cavity	Polylectic	4.33
<i>Bombus huntii</i>	-	2	-	-	Eusocial	Cavity	Polylectic	3.36*
<i>Bombus occidentalis</i>	1	-	-	-	Eusocial	Cavity	Polylectic	4.91
<i>Bombus rufocinctus</i>	12	6	7	2	Eusocial	Cavity	Polylectic	2.88*
<i>Hoplitis albifrons</i>	2	1	1	3	Solitary	Cavity	Polylectic	2.3
<i>Hoplitis fulgida</i>	1	2	-	3	Solitary	Cavity	Polylectic	2.1
<i>Ashmeadiella cactorum</i>	-	-	-	1	Solitary	Cavity	Polylectic	1.1
<i>Halictus ligatus</i>	-	-	1	-	Eusocial	Ground	Polylectic	1.3
<i>Ceratina neomexicana</i>	3	-	6	1	Parasocial	Cavity	Polylectic	1.2
<i>Ceratina nanula</i>	-	1	-	-	Parasocial	Cavity	Polylectic	0.9*
<i>Anthophora bomboides</i>	3	-	1	-	Solitary	Ground	Polylectic	3.1
<i>Anthophora terminalis</i>	-	-	1	-	Solitary	Ground	Polylectic	2.4
<i>Agapostemon sp #1</i>	1	-	-	-	Parasocial	Ground	Polylectic	2.1
<i>Agapostemon viresceus</i>	-	1	-	-	Parasocial	Ground	Polylectic	2.9
<i>Anthophora walshii</i>	2	-	2	-	Solitary	Ground	Polylectic	3.2
<i>Anthophora montana</i>	3	2	-	1	Parasocial	Ground	Polylectic	4.21

