Informing Evaluation Candidates for Endangered Species Act Listing: Bee Diversity and Abundance from USGS Pitfall Traps on the Seward Peninsula, Alaska

Final Report Prepared for: US Fish and Wildlife Service Agreement Number F20AS00071

September 20, 2023

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Acknowledgements

Thanks to Steve Matsuoka and other ecologists at USGS Alaska Science Center for conducting the original study that collected the invertebrates. Special thanks to Sabrina Kessakorn, ACCS Technician, for washing, processing, pinning, and databasing the bees. Dr. Derek Sikes and the University of Alaska Museum have curated the majority of specimens.

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Suggested Citation

Carlson, M.L. and J.R. Fulkerson. 2023. Informing Evaluation Candidates for Endangered Species Act Listing: Bee Diversity and Abundance from USGS Pitfall Traps on the Seward Peninsula, Alaska. Alaska Center for Conservation Science, University of Alaska Anchorage. 19 pp. plus appendix

Introduction

The Alaska Center for Conservation Science (ACCS), University of Alaska Anchorage, entered into a cooperative agreement (Agreement Number F20AC11537) with the US Fish and Wildlife Service (USFWS). The primary task of the agreement was to process previously collected insect specimens from northwestern Alaska to better inform our understanding of the distribution and abundance of bee species in the state. Specifically, the data associated with these collections are intended to inform conservation assessments of bumble bees and other bees in Alaska. Here, we report on our technical approach and summarize the results.

Species richness of wild bees and other pollinators has declined over the past 50 years, with some species undergoing major declines and some have gone extinct. Pollinators are at risk from various environmental threats such as habitat loss and alteration, invasive species, parasites and pathogens, pesticides, as well as climate change (Potts et al. 2010, Goulson et al. 2015). Dramatic declines have been well-documented in honey bees (Natural Research Council 2006), but have also been witnessed in bumble bees (Cameron et al. 2011), and solitary bees (Burkle et al. 2013).

As numerous species of bees have declined in abundance and range in recent decades, calls for effective conservation action have grown. The Western Bumble Bee, *Bombus occidentalis*, has dwindled in the southern portion of its range in the last few decades and is now considered imperiled enough to warrant IUCN Vulnerable Red List Category (Goulson et al. 2008, Hatfield et al. 2015, Sheffield et al. 2016) and was petitioned as a candidate for listing under the Endangered Species Act. Particular questions and information gaps in Alaska have been noted on the distribution and abundance of the *Bombus occidentalis* and its sister taxon that has been recently elevated from a subspecies of *B. occidentalis* to the species, *B. mckayi* (Sheffield et al. 2016, Graves et al. 2020, Williams et al. 2021). The distribution and abundance of both of these species in Alaska is not fully resolved (Graves et al. 2020) and efforts from previous studies in Alaska are suggestive that *B. mckayi* may not be declining in the north (Koch 2012, Pampell et al. 2015).

The diversity of bumble bees is reasonably known, yet a species that was new to science was recently described from Alaska and Yukon (Williams et al. 2016). In contrast, the diversity and distribution of non-social, or solitary bees, in Alaska is poorly known. For example, available data are concentrated around Fairbanks and the road system of interior Alaska, limiting our understanding of their presence and importance statewide (Figure 1). Wise management action is predicated on a foundation of relevant and high-quality information on the distribution, abundance, ecology, and potential threats. The Alaska Center for Conservation Science lists eleven bumble bee species as rare in the state and globally rare (Table 1), and it lists numerous species of solitary bees as rare in the state. In addition, there are many other species of bees in the state (currently estimated at 116 species, Carlson et al. unpublished data) that are known from collections from one or very few locations.

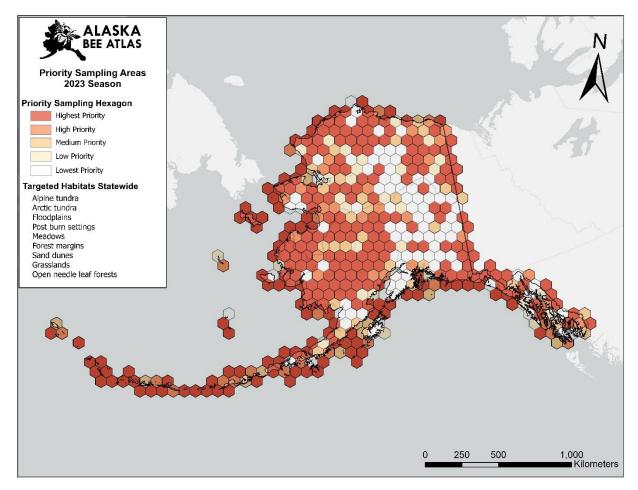


Figure 1. Bee sampling intensity in Alaska compiled by the Alaska Bee Atlas. Hexagons with no bee records are shown in red, intermediate number of records are shown in orange and yellow, and hexagons that are white are in the highest quartile of records (> 23).

Range size, occupancy, abundance, and habitat condition are key pieces of information that inform conservation ranks (see ACCS & <u>NatureServe</u> conservation ranking system) – with limited geographic sampling species may appear rarer than they are. Targeting sampling in underrepresented locations that are likely to produce relatively large number of bee species results in conservation ranks that better reflect their true status. Alternatively, processing and reviewing collected material of previous unpublished studies from under-studied regions can also be significant. The USGS implemented a pitfall trap design to capture insects in an under-studied area in the Seward Peninsula to study potential biomass diets of the black-bellied and lesser golden plovers. The insect collection was summarized for their study, however the thousands of insects collected had notes of 'bumble bees' but not identified to species. For this agreement, we further investigate the insect collection to identify the bee fauna to enhance conservation rankings for this under-studied region.

Table 1. Global and state conservation status ranks of rare bees in Alaska. Conservation status ranks follow the NatureServe conservation ranking system.

Species Name	Global Rank	State Rank
Andrena algida	G5 – Secure	S1 – Critically Imperiled
Andrena barbilabris	G5 – Secure	S1 – Critically Imperiled
Andrena clarkella	GNR – Not Ranked	S2S3 – Imperiled to Vulnerable
Andrena costillensis	GNR – Not Ranked	S1 – Critically Imperiled
Andrena costillensis	GNR – Not Ranked	S1 – Critically Imperiled
Andrena frigida	GNR – Not Ranked	S3 – Vulnerable
Andrena thaspii	G5 – Secure	S3 – Vulnerable
Bombus bohemicus	G4 – Apparently Secure	S3 – Vulnerable
Bombus distinguendus	GNR – Not Ranked	S1 – Critically Imperiled
Bombus insularis	G3 – Vulnerable	S4 – Apparently Secure
Bombus johanseni	GU – Unrankable	SU – Unrankable
Bombus kluanensis	G3 – Vulnerable	S1 – Critically Imperiled
Bombus natvigi	G3 – Vulnerable	S4 – Apparently Secure
Bombus neoboreus	G3 – Vulnerable	S3S4 – Vulnerable to Apparently Secure
Bombus perplexus	G5 – Secure	S2 – Imperiled
Bombus rufocinctus	G4 – Apparently Secure	S2 – Imperiled
Bombus sitkensis	G4 – Apparently Secure	S3 – Vulnerable
Bombus vancouverensis	G5 – Secure	S3 – Vulnerable

Our current knowledge of bees in the Seward Peninsula region is limited to several collecting events at 13 total locations in 2015 and 2016 (Figure 2, Alaska Bee Atlas 2020). Less than 100 bumble bees have been collected from these events and include two vulnerable species: *Bombus bifarius* and *B. neoboreus*. These two taxa are ranked as 'Vulnerable-at moderate risk of extirpation in Alaska' by ACCS, however they are on the close to being ranked as 'Secure within Alaska'. Several more occurrence records can justify lowering the conservation ranking of the species. Such data are possibly within unsorted material, such as the USGS Seward Peninsula insect collection. Furthermore, a specimen identified as "*B. occidentalis*" has previously been found in Unalakleet, about 200 km from the USGS collections – this specimen is likely what would be called *B. mckayi* now. It is possible for *B. mckayi* to occur within the Seward Peninsula given



similar ecoregion as Unalakleet and the paucity of collecting events and would be useful in evaluation of the ESA petition for listing.

Figure 2. Previous bee collection data from the Seward Peninsula compiled by the Alaska Bee Atlas (2020).

Project Location:

The locality of the USGS insects samples originate from the Seward Peninsula in western Alaska (Figure 3). The tundra habitat ranges from near sea-level to approximately 300 m in elevation and include riparian habitats, low shrub-forb tundra and fell field tundra. The samples were processed at the University of Alaska Anchorage with representative samples permanently retained at the University of Alaska Museum of the North (UAM) in Fairbanks.

Project Objectives:

Our primary objective was to address geographic data gaps in bee biodiversity knowledge by processing, identifying, and databasing bees collected in western Alaska on the Seward Peninsula by the USGS. These specimens were collected for a broader ecological study and are uniquely preserved with associated habitat, location, and date information.

Our primary objective was to sort, clean, identify using classic methods coupled with DNAbarcoding, pin/curate, and database these bees. This objective includes the following activities:

- 1) Sorting all invertebrate specimens and isolation of Hymenoptera
- 2) Sorting Hymenoptera into bees relative to wasps and ants
- 3) Washing the bees that were preserved in ethylene glycol
- 4) Blow-drying the bees and pinning for long-term curation
- 5) Generating specimen labels from data provided by USGS
- 6) Identifying all bee specimens by morphology
- 7) Sending specimens that were questionable to other expertsa. Or send tissue for DNA barcoding
- 8) Databasing all bee specimens and send to UAM for curation
- 9) Sharing data and short written summary of findings with USFWS and USGS

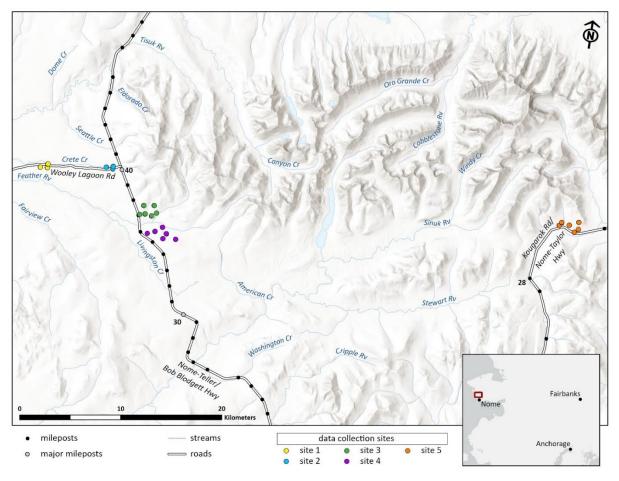


Figure 3. Sampling locations by the USGS on the Seward Peninsula, Alaska.

Methods

Data associated with the sampling sites, times, and personnel involved was supplied by the USGS. Arthropods were collected from pitfall traps from four sites on the Seward Peninsula in 2012. Pitfall traps were deployed in five-day periods from early June to mid-August. Arthropods were presorted by order and site.

Insect samples were preserved in vials of ethylene glycol. ACCS sorted the vials and extracted all bees. ACCS staff used a standard processing and curation technique of washing and drying bees

for museum quality curation (see Fulkerson et al. 2023). ACCS staff provided provisional specieslevel identifications based on morphology. In some cases, specimens were damaged or matted and unidentifiable and some species groups are difficult to identify morphologically (e.g., *Bombus lapponicus sylvicola* and *B. johanseni*; *B. polaris* and *B. kirbellus* and *B. neoboreus*). All of the questionable specimens had an antenna removed for DNA bar coding analysis. One or more representatives of all other species and specimens that are readily identifiable morphologically were also included for DNA bar coding analysis at the Canadian Centre for DNA Barcoding.

Data were summarized as count and abundance. A Sorensen two-way cluster analysis using PC-ORD was used to determine if there was similarity between the sites based on bees collected (McCune and Mefford 2011).

Results and Discussion

A total of 640 bee specimens were identified from the Seward Peninsula pitfall traps, encompassing 11 species from three subgenera (Figure 4, Table 2) and no solitary bee species were found. The most numerous species was *Bombus lapponicus sylvicola* (306 specimens), followed by *B. polaris* (109 specimens), *B. natvigi* 75 specimens, and *B. johanseni* (53 specimens). The least abundant species were *B. mixtus* (1 specimen), *B. cryptarum* (2 specimens), *B. melanopygus* (7 specimens), and *B. neoboreus* (9 specimens). Approximately ¹/₄ of the specimens were representatives of the tundra-specialist subgenus *Alpinobombus* that nests above ground and the remainder were subgenus *Pyrobombus*. A single representatives of subgenus *Bombus* (*B. cryptarum* of two specimens) was detected at two sites, Wooley Lagoon and Blume Creek. No *B. mckayi* nor *B. occidentalis* were identified in the samples.

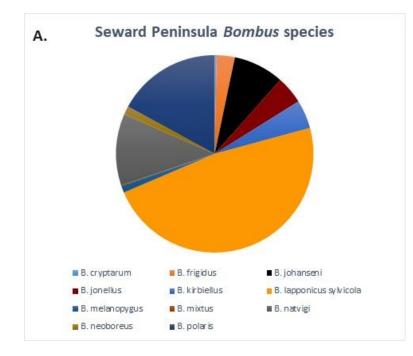
Bombus johanseni was rediscovered in Alaska recently (Sheffield et al. 2020) and was previously known from just two specimens in the state on the Arctic Coastal Plain. Collections from this study increase the number of known specimens to 55 and extends the known range size in Alaska substantially.

The four sites were similar in community composition, particularly the three western sites and with only minor differences between the adjacent Blume Creek and Feather Creek sites (Figure 5) The Kougarok site had a very high abundance of *B. lapponicus sylvicola* and *B. frigidus* and low abundance of *B. johanseni*. The Wooley Lagoon sites had some of the least common species, members of *Bombus* and *Pyrobombus* that typically have a more southerly and boreal distribution. Most of the sites at this location were at a low elevation floodplain near the coast and perhaps not surprising that this location would harbor these species. The higher elevation, fellfield tundra locations (Feather River and Blume River) had a larger proportion of subgenus *Alpinobombus*. The Blume and Feather sites also had a large contingent of *Bombus natvigi* that is a social parasite of *B. polaris* and probably other members of *Alpinobombus* (Williams et al. 2016).

Table 2. Bee species and number collected from USGS pitfall traps on the Seward Peninsula June – August 2012.

Species Name	Subgenus	Number of Specimens	Conservation Rank
Bombus cryptarum	Bombus	2	G5 S4
Bombus frigidus	Pyrobombus	19	G5 S5

Species Name	Subgenus	Number of Specimens	Conservation Rank
Bombus johanseni	Pyrobombus	53	G2 S2
Bombus jonellus	Pyrobombus	29	G4 S4
Bombus lapponicus sylvicola	Pyrobombus	306	G5 S5
Bombus melanopygus	Pyrobombus	7	G5 S5
Bombus mixtus	Pyrobombus	1	G5 S5
Bombus kirbellius	Alpinobombus	30	G4 S4
Bombus natvigi	Alpinobombus	75	G3 S4
Bombus neoboreus	Alpinobombus	9	G3 S3S4
Bombus polaris	Alpinobombus	109	G5 S4



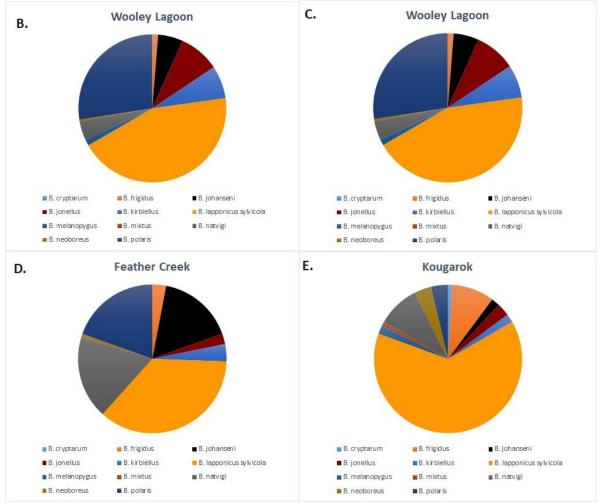


Figure 4. Relative abundance of bumble bee species for all locations collectively and for the four separate locations.

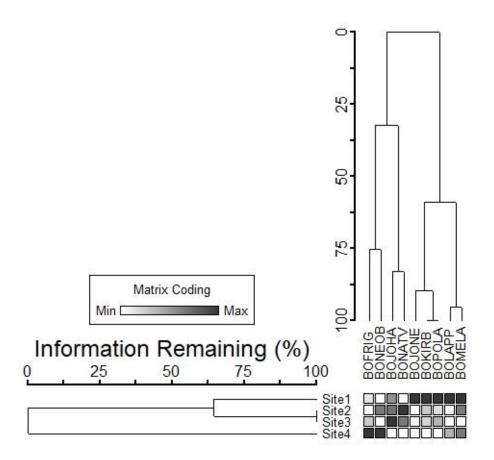


Figure 5. Two-way cluster analysis of the four sites and species of bumble bees, based on Relative Sørensen's Distance measure and Group Average Linkage method. Site 1 = Wooley Lagoon, Site 2 = Blume Creek, Site 3 = Feather Creek, and Site 4 = Kougarok Road. BOFRIG = B. frigidus, BONEOB = B. neoboreus, BOJOHA = B. johanseni, BOJONE = B. jonellus, BOKIRB = B. kirbellius, BOPOLA = B. polaris, BOLAPP = B. lapponicus sylvicola, BOMELA = B. melanopygus. Bombus cryptarum (2 specimens) and B. mixtus (1 specimen) were removed from the analysis.

Seasonal Abundance

All species had a similar pattern of abundance across the summer (Figure 6). Abundance was intermediate at the start of the sampling period in mid-June and declined to mid-July, when it increased dramatically. It then declined strongly and began rebounding in mid-August. This pattern is consistent with mated queens foraging and nest hunting at the beginning of the period, followed by somewhat lower activity as the queens have provisioned nests and are involved in thermoregulation of the nests (see Heinrich 1979). After approximately three to four weeks the worker generation matures and begins foraging in mid-July. This generation then provides the floral resources for the production of queens and drones at the end of the summer in mid-August. A pulse of males also appears in mid-July and may be the result of switching to producing reproductive offspring early in the summer, workers succeeding at reproduction and laying eggs, or overwintered queens that were unsuccessful in mating laying haploid eggs (L. Richardson 2023 pers. comm.).

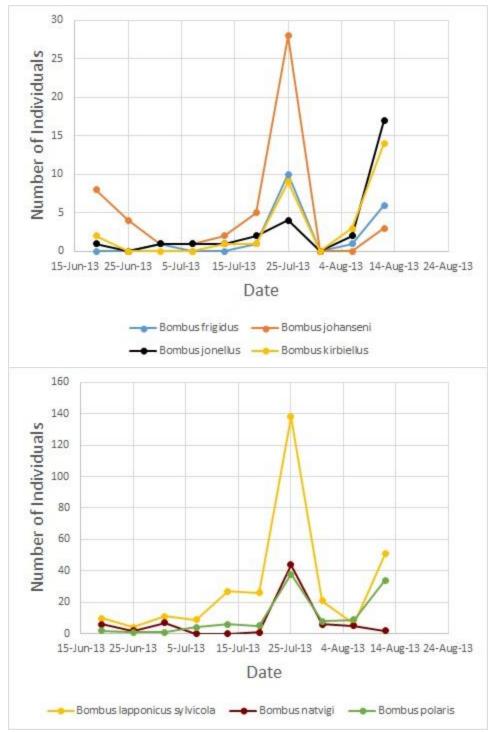


Figure 6. Seasonal abundance of bumble bee species on the Seward Peninsula, Alaska; top figure shows four species of intermediate abundance (>10 and < 75 records); bottom figure shows three species of high abundance (> 75 records).

Bombus johanseni distribution

There was one occurrence of *Bombus johanensi*, the type locality, from the Toolik Field Station at the base of the Brooks Range at the start of this project (Figure 7). The distribution of the species was unknown but suspected to be more widespread across the North Slope Alaska. The collection from the USGS specimens significantly enhanced the knowledge of the distribution, occurrence, and timing of *B. johanseni*. Due to the paucity of data, *B. johanseni* was unrankable and therefore had a conservation rank of GUSU. This additional dataset contributed to the updated conservation rank of G2S2. Since this project started, there was one additional record within the Arctic National Wildlife Refuge that was collected for the Alaska Bee Atlas. The two North Slope records would have justified a conservation rank of G1S1. With the disjunction of the North Slope records and Seward Peninsula records, we suspect *B. johanseni* to be found in northwest Alaska and connect to the other North Slope populations.



Figure 7. Distribution of Bombus johanseni in Alaska.

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Appendix. A. Two-way Cluster Analysis Raw Results

USGS – SewardPenBees_Analysis_2023_02

```
PC-ORD, 6.08
usgs bee2023_09
Linkage method: GROUP AVERAGE
Distance measure: Relative Sorensen
Total sum of squares:
                     5174.250
Cluster cycle
  1 Combined group
                     3 into group
                                  2 at level 1.0324E-02
  2 Combined group
                     2 into group
                                  1 at level 5.2499E-02
                     4 into group
  3 Combined group
                                  1 at level 1.2756E-01
Relativizing matrix by column maximum.
Distance measure: Relative Sorensen
Total sum of squares:
                     4.789223
Cluster cycle
  1 Combined group
                     10 into group
                                   5 at level 5.1541E-03
  2 Combined group
                     7 into group
                                  6 at level 2.0957E-02
  3 Combined group
                     5 into group
                                  4 at level 3.9853E-02
  4 Combined group
                     8 into group
                                  3 at level 6.1287E-02
  5 Combined group
                     9 into group
                                  1 at level 8.5978E-02
  6 Combined group
                     2 into group
                                  1 at level 1.3794E-01
  7 Combined group
                     6 into group
                                  4 at level 1.9177E-01
  8 Combined group
                                  3 at level 2.8917E-01
                     4 into group
  9 Combined group
                     3 into group
                                  1 at level 4.3446E-01
Percent chaining = 21.25
```