

## Bee Inventory and Monitoring Plan for Alaska 2023



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## I. Introduction

## Importance of Pollinators

Pollinators play a key ecological role that directly and indirectly affects wildlife and their habitat. Many species of plants are completely or partially dependent on pollinators for fruit and seed production, including plants in northern latitudes (Fig.1). A wide range of mammals and birds are reliant on fruits that are the result of insect pollination in Alaska (e.g., cloudberries and blueberries). Pollinators indirectly affect wildlife diversity and populations by promoting seed production of keystone food resources. For example, many species of willows, which are the primary forage species for moose, are largely insect-pollinated and if early emerging bee and flower-visiting flies are not present, the willows will not be able to produce seeds and colonize new areas; thus limiting forage and habitat availability of moose.

Pollinating insects are wildlife of conservation concern in their own-right and the Bureau of Land Management (BLM) - Alaska identifies five species of bumble bees as <u>BLM Sensitive Species</u>, as well as five additional bumble bees and three butterflies as Watchlist Animals.

There have been significant declines in both honey bee and wild bee populations in Europe and North America for the past 50 years (Potts et al. 2010, Cameron et al. 2011, Koh et al. 2016). The scope of pollinator decline in North America is not entirely clear due to limited inventory and monitoring practices. Pollinator decline has been attributed to several factors that include, but are not limited to, habitat loss and fragmentation, loss of preferred floral resources, pesticide use, invasive plant species, and climate change (Potts et al. 2010, Scheper et al. 2014, Koh et al 2016).

*Bombus occidentalis* (Western bumble bee) has declined significantly in the southern portion of its range in the last few decades and is now listed under the International Union for Conservation of Nature and Natural Resources (IUCN) Vulnerable Red List Category (Goulson et al. 2008, Hatfield et al. 2015, Sheffield et al. 2016). Furthermore, the species has been petitioned as a candidate for listing under the Endangered Species Act. Previously, a subspecies (*B. occidentalis mckayi*) was considered to occur in Alaska and the Yukon but molecular evidence supports the bumble bee as a different species, *B. mckayi* (Williams et al. 2021). However, the northern distribution range and abundance of *B. occidentalis* is poorly known and appears to nearly reach the boundary of Southeast Alaska. Further sampling in Southeast Alaska is warranted.



Figure 1. Bombus flavifrons foraging on native lupine flowers.

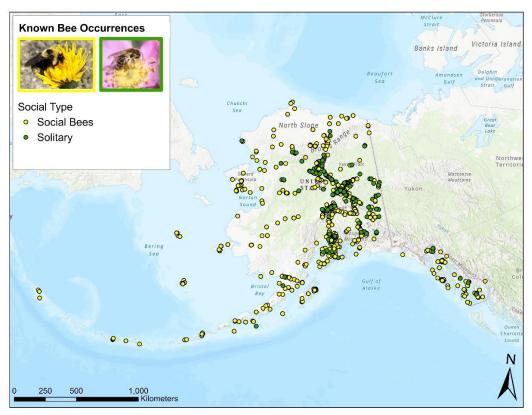


Figure 2. Distribution of bees collected across Alaska from 1877 to 2021. Data originate from museum collections and literature.

## Current State of Pollinator Knowledge in Alaska

There are 22 species of bumble bees (*Bombus* sp.) found in Alaska which have been well documented across Alaska in a multitude of habitats, though less in the western and arctic regions (Figure 2). Some of the earliest bumble bee collections for the region date back to the late 1800's. Even with a long history of collections, new species are still being discovered in Alaska and neighboring Yukon Territory (e.g., Williams et al. 2016, Sheffield et al. 2020).

While pollinators provide essential ecological and agricultural services to Alaska, only one monitoring program has been initiated in the state and it was solely focused on bumble bees in an agricultural setting. Bumble bee diversity and abundance varied significantly throughout and between the two collecting seasons (Pampell et al. 2015). The monitoring study found that *B*. *mckayi* had high rates of parasitic nematode infections.

An inventory of the National Park system documented pollinators in various habitats and elevations. The inventory program most frequently found *Bombus* and syrphid fly species in traps with solitary bees being less frequent (Rykken 2015, Rykken 2017). In Denali National Park, the alpine tundra habitats contained the highest diversity and abundance of pollinators compared to roadside and trails systems at lower elevations, though the alpine habitat had a higher intensity of sampling (Rykken 2015). Conversely, in Gates of the Arctic National Park, disturbed and riparian areas had the highest abundance compared to alpine tundra habitats (Rykken 2017).

While the diversity of bumble bees is reasonably known, the diversity and distribution of nonsocial, or solitary bees, in Alaska is very 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 (Fig. 2). As their name implies, solitary bees do not conform to a socialize colonies or hives and do not have a separate caste system of workers and queens (female solitary bees both provision nests and lay eggs throughout their short adult lives). There is an estimation of 4,000 species of bees in North America (Cane and Tepedino 2001), with solitary bees accounting 98% of the species (Asher and Pickering 2020). Previous research documented over 50 solitary bee species in Alaska. They are most commonly found in open and sunny habitats in lowland settings such as roadsides and floodplains but many are primarily associated with the Steppe Bluff ecosystem in eastern interior Alaska and the Copper River Basin (Armbruster and Guinn 1989, Bishop and Armbruster 1999). Solitary bees have been found in alpine and tundra systems in Alaska at a much lower frequency than bumble bees, but extensive sampling has yet to occur.

The Alaska Center for Conservation Science (ACCS) initiated a bee monitoring program in Interior Alaska from 2017–2019 (with support provided by our state partners at the Alaska Department of Fish and Game) focused on the Steppe Bluff habitats; an ecosystem suspected to harbor a large diversity and abundance of solitary bees (ACCS *unpublished data*). This inventory effort has provided substantial data on the temporal habitat use of this rare ecosystem by pollinators. ACCS collected at least ten new species records for Alaska, the first male *Osmia maritima* in North America, and have molecular support for potentially undescribed taxa. The current estimated number of bee species in the state, combining data from the University of Alaska Museum, National Park Service, and ACCS is approximately 111.

With new state records for solitary bees, syrphid flies, and new species to science being found in Alaska in recent years, the diversity and distribution of pollinators of the region are not yet fully known. Efforts to initiate a statewide biodiversity collection protocol are therefore warranted given the limitations of baseline knowledge, coupled with conservation concerns and ecological importance of the group.

## National and International Coordination

This Alaska Bee Atlas is designed to be compatible with the forthcoming U.S. National Program for Monitoring Native Bees (Woodard et al. 2020). In 2016, the Governmental Accountability Office issued a Report of Bee Health (GAO 2016) (https://www.gao.gov/products/GAO-16-220), which recommended that the USDA coordinate with other agencies to develop a federal pollinator monitoring plan. The plan includes action areas on gathering and cataloging data, identifying optimal survey methods, and identifying priority areas for monitoring. While this Alaska plan will be completed before the National Plan is published and implemented, this plan has been designed to fit within the general action areas of the National Plan.

Additionally, the Arctic Council-Conservation of Arctic Flora and Fauna (CAFF) State of the Arctic Terrestrial Biodiversity Monitoring Report (CAFF 2021) has identified pollinators as a focal ecosystem component to prioritize for monitoring across all Arctic Nations. Since many bee

species are shared throughout the boreal forests and arctic tundra, the Alaska Bee Atlas can contribute to arctic-wide knowledge on bees.

## Who Benefits and How Will the Data be Used?

ACCS has produced rarity assessments of all 23 known bumble bee species, four mining bee species, and all 80 known butterfly species in the state. Having basic distribution, area of occupancy, and habitat associations for these species is necessary data for assessing state conservation status ranks, which facilitate and inform federal land managers of sensitive species or those of greatest conservation concern. Additionally, fundamental baseline data would provide scientific support for those taxa under review for listing under the Endangered Species Act. ACCS disseminates these data online as part of public education/outreach and have presented results to state and federal land managers and at scientific meetings. These data can be viewed online at (https://accs.uaa.alaska.edu/wildlife/pollinator-diversity/).

## Sampling Plan

In Alaska, bee sampling is heavily skewed toward the major cities of Anchorage and Fairbanks, as well as the highway system of interior Alaska (Figure 2). The intention of the Alaska Bee Atlas is to coordinate a baseline collection program to determine biodiversity of bees in Alaska before implementing a region wide monitoring program that has yearly sampling events. This will allow us to identify regions, habitats, and species that are of conservation concern and more effectively target specific monitoring actions. The same methods outlined here can be adapted to monitoring plans to detect local population declines.

A Priority Sampling Grid was created by synthesizing known bee occurrences in 5,000 km<sup>2</sup> grids across the state. Hexagons with more than 22 data points were categorized in the Lowest Priority Grids compared to hexagons with 0 data points (Highest Priority Grids) or 1 to 2 data points (High Priority Grids) (Figure *3*). The sampling grid map is updated each spring. For the 2022 season, twelve grids changed in priority levels, indicating successful collections away from the road system. For this reason, sampling under this plan needs to be coordinated through ACCS to ensure efficient sampling and available funding for specimen identification. Participants will need to sign up in the spring of each year at the Alaska Bee Atlas website (www.alaskabeeatlas.org). Participants staying within the road system are still encouraged to participate but are recommended to sample the targeted habitats summarized in Table *1*.

Please note: ACCS will only accept sampling efforts from participants who are in coordination with the appropriate land management agency for the proposed sampling area.

Table 1. Alaska habitats with data gaps.

Target Habitats in Lowest Priority Grids						
Alpine and arctic tundra	Post burn settings	Forest margins	Grasslands			
Floodplains	Meadows	Sand dunes	Open needle leaf forests			

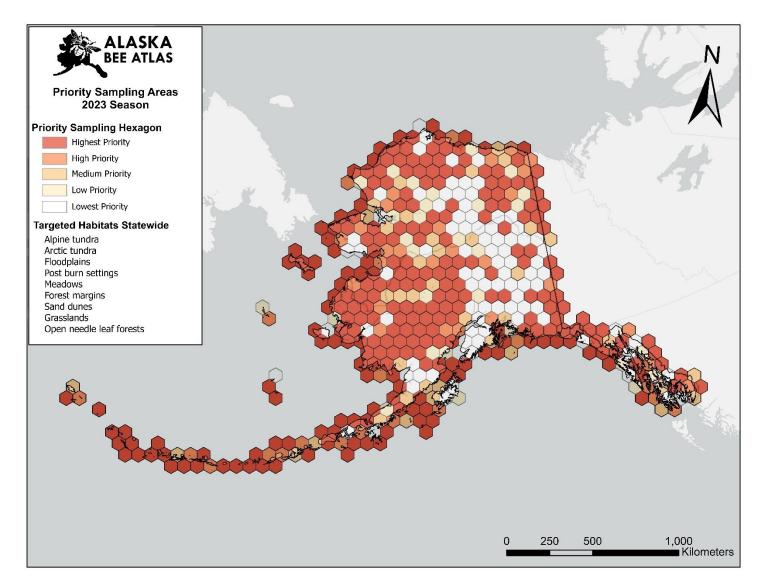


Figure 3. Sampling areas of priority in Alaska based on the number of bees collected in the past. Areas marked 'Lowest Priority' can still be sampled in high priority habitats or if a yearly monitoring station is established. An interactive map is available on the AK Bee Atlas website.

## II. Sampling Protocol

We describe three survey methods that participants can choose based on their time commitment, skills, and abilities. The methods can be done indecently or together. We strongly encourage the use of all three methods at a sample location because this has been shown to thoroughly document the bee diversity of a location, however it requires more effort. Details on each method are in the following sections and printable instructions are in the Standard Operating Procedures at the end of this document.

- **Bee Bowls** Bee bowl traps are placed in a habitat for 24 to 72 hours. The site must be revisited to collect the traps.
- **Blue Vane Traps** A blue vane trap (BVT) is placed at a site location for 5 to 7 days. The site must be revisited to collect the trap.
- **Net Capture** This will be an active 20-minute sampling effort of netting bees found on flowers. If a site will not be revisited later, we suggest this method.

## Materials and Supplies

Below is a list of supplies and materials needed to perform the bee survey. ACCS can provide many of the materials upon request. Photos of the various containers/jars included in Appendix A.

<u>Items needed for all methods</u>	<u>Additional for BVT</u>			
Sealable plastic bags	Blue vane trap*			
Specimen cups*	Zip ties*			
Permanent marker	Stake or pole at least 1 meter height			
Pencil	Hammer			
Scissors	Fumigant strip*			
Flagging tape	Insect net			
Camera	Killing jar			
GPS unit	Killing agent			
Data sheet (end of this manual)				
Preservative*: ethanol, propylene glycol, or denatured alcohol	Additional for Net Capture			
	Insect net			
Additional for Bee Bowls	Killing jar			
15 Bee bowls*	Killing agent			
1 Liter of water	Timer/stopwatch			
Plain, unscented dish soap*	Optional Non-kill methods for Net Capture			
Small strainer*	Photo Chamber*			
Plastic spoon*	OR			
	Ice & ice chest or insulated box			
	Vials*			

\*Materials that can be provided by ACCS.

## Seasonality Sampling

Collecting pollinators can occur anytime between late April and mid-August for most of Alaska. Even with most of the early spring vegetation looking brown, willows are often the first to flower and provide important food resources for bees. Pollinator activity diminishes in windy, rainy, foggy, or cold weather. Plan the bee collecting activity around favorable weather when bees are more active. Specifically, sample on sunny to partly sunny days when the temperature is between 50 and 90 °F (10 - 32 °C) and no rain is expected. Wind should be no greater than a gentle breeze (<12 mph). Visual cues of wind at this speed are where leaves and twigs are in constant motion, fallen leaves blow around, or flags extend out (NWS 2020).

The early spring marks the time for bumble bee queens to emerge and search for a new nesting location and floral resources. Lethal sampling efforts at this time frame may have negative consequences since killing a queen in this early stage will disrupt colony development. The timing of this early sampling period varies across the state and special coordination with ACCS should occur. We recommend the Net Capture with non-kill methods during this time frame in habitats that are particularly good for bumble bees (alpine and arctic tundra, boreal and temperate rain forests, and forest margins). Bee bowls do not catch a large number of bumble bees, however BVTs can catch large numbers of bumble bees (Sikes and Rykken pers. comm.).

## Bee Bowl Traps

Bee bowl traps, also known as pan traps, are a standardized method for bee monitoring in the U.S. and are a favored method due to their low cost and high efficiency (Westphal et al. 2008, Droege 2015). A standardized bee monitoring program for the United States is in the early stages but we follow the standardized guideline for bee monitoring as described by Droege (2015). The primary author has successfully used this method to monitor bees in southwest Idaho (Fulkerson and Kinter 2013) and adapted the methods for a pollinator study of the Steppe Bluff ecosystem of Alaska (Fulkerson et al. *in prep.*).

Bee bowl traps are 4-ounce translucent plastic cups painted white, fluorescent yellow, or fluorescent blue that reflect UV light (Figure 4). All three colors of bowls are used within the study because not all bee species are equally attracted to the same colors. We also use an asterisk design with black marker drawn on the base to mimic UV floral guides to enhance attraction (Wilson et al. 2016).

The bee bowls are filled <sup>1</sup>/<sub>4</sub> full with a soapy water solution. Soap is used to break the water tension, allowing bees to be trapped in the bowl. The soapy water solution does not need a consistent amount of soap; a simple unmeasured squirt is sufficient. Any brand will suffice, but **please use a non-scented dish soap.** Scented soaps can artificially attract unwanted insects and mammals.



Figure 4. Blue bee bowl with collected bees. Permanent marker included for size reference.

## **Trap Duration Time**

We highly encourage bee bowls be left within the study area for 20 to 24 hours, though shorter time frames of 8 to 12 hours are acceptable if the length of time is recorded. Lengths greater than 72 hours are not recommended as captured insects will degrade sitting in the trap, making identifications more difficult. The bowls can be reused many times and therefore several sampling events can occur if participants are in a given area for a long period of time.

We have observed bumble bees foraging in the late evening during the Alaska midnight summer sun, though at a lower frequency. Evening and night foraging behavior of bees has not been studied in Alaska, but we expect overnight bee bowls to enhance collection due to the longer daylight hours.

## Sample Size

A sample size of 30 bee bowls has been recommended for estimating species richness and diversity of pollinators, however the sample size is subjective to regional habitat and fewer bowls have been found to be effective in some areas (Shapiro et al. 2014). In our previous work, we limited our sample size to 15 bowls per habitat due to the size of the microhabitat. There is insufficient data to recommend a specific sample size for habitats in Alaska; however, we suggest participants use 15 bowls for a sampling event.

There is no evidence in literature that the use of traps negatively impacts pollinator populations or non-target arthropods, even when monitoring methods are used bi-weekly for long term monitoring (Lebuhn et al. 2015, Gezon et al. 2015). However, we still suggest a well-planned collection effort that implements responsible collecting.



Figure 5. Example of a bee bowl transect. Note that the distant bowls are not clearly visible from the camera view.

## **Bowl Placement**

Bee bowls are placed on the ground in a transect line in alternating colors (Figure 5). The transect line location is up to the discretion of the collector with consistency in mind. The transect should be placed within a homogenous area or habitat. For example, a transect should occur entirely within a forest system, not crossing over into other habitat types such as tree line into shrubland or alpine tundra. Another example, if sampling a river floodplain, keep the entire transect within the river scrub floodplain, not partially in both the floodplain and forest setting.

Avoid sampling in dense tall vegetation unless extra effort is used to increase visibility of the bee bowls by trimming vegetation or attaching the bowls to a stake. Generally, *if you can see the bee bowl, so can the bees.* Vegetation should be trimmed or pushed aside to be able to see the bowls from overhead from about 0.5 meters away. Alternatively, the bowls can be secured to a stake in a cup holder made from PVC pipe to be elevated off the ground. Avoid mountain ridges or other areas exposed to consistent windy weather, as this is typically less favorable for pollinator flight. Warm, sheltered microsites typically harbor more bees. In Alaska, forested habitats have yielded fewer numbers of bees compared to open meadows but are still acceptable sampling habitats.

We have experienced the occasional loss of a bee bowl due to either wind or animals. Please record the number of bowls placed and retrieved on the data form.

## Blue Vane Traps

Blue vane traps (BVT) consist of a plastic jar with and a blue funnel screw cap with interconnecting vanes (Figure 6). The BVT has been a successful passive collecting method for bees in lower latitudes but recently has been replaced by for cheaper and quicker traps such as the bee bowls. The use of BVT has had mixed results in Alaska from successfully collecting many bees to collecting no bees (Pampell 2010, Rykken 2015). The BVT does appear to be a favorable trap method for bumble bees.

The BVT is attached to a stake or post one meter from the ground and sets on its side, parallel to the ground (Figure 6). This reduces rain and water accumulation in the trap. The BVT can be either on its own or used in conjunction with the bee bowls by placing it at the end of the bee bowl transect. Fumigant is placed inside the white bowl to kill insects that are trapped inside. The fumigant is a small (2cm x 1cm) cut strip of HotShot brand pesticide strip. When the participant collects the BVT at the end of the sampling time frame, a net might be needed to transfer any insects still alive to a killing jar.

We have observed poor capture rates with BVT that are set for less than 3 days. We only suggest BVT use if they can be left out at a sample location for 5 to 7 days, but no longer than 10 days. Traps have differing success based on bee species preferences and having multiple trap type enhances the biodiversity of the collecting event.



Figure 6. A properly secured blue vane trap attached to a post horizontally with plastic zip ties and string.

## Net Capture

Not all bees are attracted to passive traps or may not see a particular trap, therefore they are not caught. Active netting increases the sampling ability to capture the most reflective diversity of bees in the sampling site. It also provides an alternative to collect data without killing bumble bees.

An aerial insect net is required for this activity. If participants are within the Anchorage area, they may be able to borrow a net from ACCS for their sampling plan if available. Alternatively, insect nets can be purchased from various online sources. Insect nets come in a different sizes and features, but we suggest purchasing one with at least a 12" diameter net, 3-foot handle, and a standard aerial bag (not a sweep or muslin bag). For those with limited space, collapsible nets are an ideal effective tool.

From Forestry Suppliers, we suggest the following nets: #53725 Economy Series Insect Net #53746 Professional Insect Net

Participants will spend 20 active minutes walking in a defined habitat searching for bees and flies on flowers. The survey area should be no larger than 25 meter  $\times$  25 meter square. If the netting activity is in addition to the passive traps, the meander should occur within 50 meters of the transect, within the same sampling ecosystem. A stopwatch is needed to time the active search. The stopwatch should be paused during the transfer of bees to a killing jar and data recording. The stopwatch will resume until another successful capture or the 20-minute time is completed. Participants should be actively walking within the sampling site and not a 'wait and see' approach with an individual flower.

The netting activity can be performed solo or with multiple people. The 20 minutes should be divided equally among the participants who follow the netting survey instructions independently. For example, if two participants are netting, then each will have 10 minutes to perform the netting activities. If three participants, then 6:40 for each participant.

There are many techniques to successfully capture pollinators and we highlight two here. With both techniques, the participant will first capture the pollinator, then transfer it to a killing jar or perform a non-kill method.

- 1. A common netting technique uses quick swings to snatch pollinators from flowers, but this damages the plant and takes some practice. After a forceful swing, immediately rotate the net handle to close off the net opening and then inspect the net to see if a successful capture occurred.
- 2. An alternative approach is to hold the net handle in one hand and the tip of the net outstretched and taught with the other hand. Slowly place the taught net over the flowers with the pollinator. Bees are typically focused on pollen and nectar collection and often do not notice the net. However, flies are quick to escape with this method. Insects will naturally fly or crawl upwards to the net tip, at which point the middle of the net is grabbed and closed off with your hand, holding the pollinator in the end of the net.

Next, the netted insects are transferred to a killing jar or a photo chamber or vial if non-lethal methods are used (see below). At this point the stopwatch should pause so the participant can perform the data collection. Netting requires some patience, finesse, and sometimes luck. Try several methods to find what best suits your skills and abilities.

The Netting Procedure is in <u>Standard Operating Procedure C. Net Capture for Bees</u> with some video references at the end of this document.

## Preserving the Samples

## Killing Jars and Killing Agent

A killing jar is needed for collecting pollinators in the Net Capture approach. An optional non-kill method is described in the next section but should only be applied to bumble bees. The killing jar is glass jar with screw lid and often contains plaster at the bottom to absorb and dissipate a killing agent. If such a kill jar is not available, a simple 1-quart canning jar with a layer of loose tissue paper and cotton balls on the bottom is sufficient. DO NOT USE PLASTIC CONTAINERS, unless specifically designed for this purpose. Plastic will melt when in contact with most killing agents.

The most common killing agent is ethyl acetate, which can be obtained as nail polish remover. Alternatively, the killing jar can be placed in the freezer for an hour. To create a killing jar:

- 1. Charge the jar: add about 1 teaspoon of ethyl acetate to the kill jar and keep the lid closed. The jar will remain charged for several days.
- 2. If the jar is not labeled with a warning, do so with prominent lettering such as:

## **Poison- ethyl acetate**

3. Add an additional crumpled paper towel or face tissue to absorb excessive moisture and to provide some separation of the insects to reduce damage.

*If the participant is flying to field locations:* Ethyl acetate is a hazardous material but allowed in checked or carry-on luggage in small quantities. Please refer to TSA guidelines for updated information.

## Preservative

The preferred preservative for insects collected in the field is ethanol. <u>Ethanol</u> maintains the integrity of the specimen, is easy to wash and remove, and minimizes DNA degradation. Pure grain alcohol, such as Everclear, can be used when diluted to 70% with water. Alternatively, <u>propylene glycol</u> (PG) can be used to preserve collected insects and minimizes DNA degradation but requires more lab work for processing the insects. PG is a non-toxic sugar alcohol that is used in non-toxic anti-freeze, food additives, and e-cigarette fluids. Lastly, <u>denatured alcohol</u> can be used, however insect samples experience DNA degradation resulting in slightly lower performance for DNA barcoding and should only be used if ethanol or PG can't be found. Klean Strip brand of clean burning fuel is available at local big box stores and should be diluted with water to create a 70% mixture. **Note that rubbing alcohol is not the same and should not be used**. Please adhere to the brand names mentioned for consistency and safety.

Typically, 30 mL (1 ounce) of preservative is needed for each sampling event. Pour just enough preservative to cover or saturate the collected insects in the specimen cup.

Ethanol is classified as a hazardous material. If the participant is flying to field locations, there are two options:

- 1. Store collected insects in a cold cooler for up to 5 days or freeze the collection. Immediately preserve the insects with preservative when returning to the office.
- 2. Use PG: either food grade or non-toxic anti-freeze (PEAK Sierra Brand <u>only</u>). PG is not hazmat and documentation is not needed. Either the full strength or a 50/50 blend is suitable.

## Non-kill Methods

Non-kill methods are not recommended for solitary bees since these species are identifiable only by microscopy or DNA barcoding. Non-kill methods can be used for bumble bees since identification can be achieved with photographs for most bumble bee species but error rates are higher than having physical specimens. A monitoring plan for bumble bees is possible with this method.

We adopt the approach of the Pacific Northwest Bumble Bee Atlas for the non-kill methods and summarized here as two options, 1. the ice method and 2. photo chamber method. The chilled method requires access to ice or very cold streams. Transporting ice to remote locations in Alaska is not particularly feasible and best reserved for near populated areas or the road system.

- 1. **Ice Method:** After netting a bumble bee, the bumble bee is transferred to a vial and placed in a cooler with ice (crushed is best) for 10-12 minutes before photographing the bumble bee. If ice is not available, the vials can be placed in remnant snowbanks, or in very cold running streams for 10-15 minutes or until the bees are cold and not moving fast.
- 2. **Photo Chamber Method:** After netting a bumble bee, the bumble bee is transferred to a photo chamber vial for photography.

Clear macro photographs **are essential** for this method to be successful (Figure 7). Please review the detailed methods in Standard Operating Procedure C. Net Capture for Bees and photography tips from the Pacific Northwest Bumble Bee Atlas (https://www.pnwbumblebeeatlas.org/photo\_tips.html).



Figure 7. A useful photo of a bumble bee using the Ice Method (left). A non-useful photo of a bumble bee in a vial, an identification cannot be determined with high confidence (right).

## **Delivering Data and Samples**

Deliver all samples, data sheets, and photos to ACCS for processing. Generally, the samples should have as little fluid in the cup as possible. The preservative liquid should be poured off and placed in a double secure resealable plastic bag with some paper towels to absorb accidental residual moisture. A Standard Operating Procedure is at the end of this document.

## Contact:

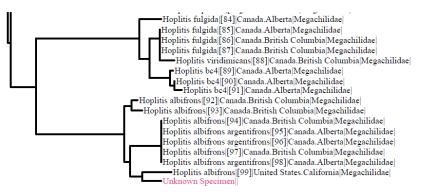
Justin Fulkerson UAA/ACCS BMH 3211 Providence Drive Anchorage, AK 99508 907-786-6387 | jrfulkerson (at) alaska.edu https://www.alaskabeeatlas.org

## III. Identification and Data Processing

After ACCS receives samples, they are processed with a washing and blow dry method then pinned to standard museum curation practices. Specimen data will be recorded and housed with databases at ACCS and displayed online. Final pinned specimens will either be retained at the UAA Entomology Collection or sent to the University of Alaska Museum of the North Entomology collection for preservation.

Identification of bees is the most difficult and time intensive aspect of the Alaska Bee Atlas. There are relatively few taxonomic experts in North America available for bee identification. Ecologists at ACCS have been identifying bees from previous studies and sending material to experts for identification. This has allowed ACCS to develop a reference collection of Alaska bees for identification purposes.

DNA barcoding is an additional approach for bee identification that is gaining in utility. ACCS has implemented this method with positive results. Small amounts of tissue from the bee, such as an antenna, are fixed in ethanol and sent to a lab for DNA sequencing. An online database processes the sequences and make comparisons to other taxa in online databases. For those specimens that might be difficult to identify or are of poor quality, the result is an accurate identification to the species level (Figure 8).





## IV. Products

The initial effort will be focused on developing a network of participants, refining protocols, and ensuring the workflow is operating efficiently. Second, the project aims to summarize the biodiversity and biogeographic data to the public in an online portal. A significant goal is to document all bee species known to the region. ACCS and the Alaska Museum of the North Entomology Collection have been compiling a comprehensive list of bee species, but recent work has shown new species and data gaps. The list of Alaska bumble bees and some solitary bees with their biogeographic data can be found at: <a href="https://accs.uaa.alaska.edu/wildlife/pollinator-diversity/">https://accs.uaa.alaska.edu/wildlife/pollinator-diversity/</a>. Third these data will be used to update statewide conservation ranks that inform sensitive species lists with federal agencies.

After bee samples are curated by ACCS, bees are deposited in the UAA Entomology Collection and the Alaska Museum of the North Entomology Collection. Museum data will be displayed in the online ARCTOS database: <u>https://arctosdb.org/.</u> DNA barcoding data are publicly available to scientists at online accessible databases.

## V. Management Implications

Management of native bee species and their habitat in Alaska has been hindered due to a lack of knowledge of species distribution, status, trend, habitat associations, and floral resource preferences. Proactive management will be facilitated with additional information on native bees. While initially designed with BLM land management in mind, there will be immediate and long-term benefits to other managers in Alaska with this monitoring plan and additional data collection. Increasing coordinated monitoring will benefit all land and species managers by better informing statewide trends in the conservation status of pollinators and their habitats. A better understanding of native bee species will allow for more effective habitat management and could help avoid the need to list species through the Endangered Species Act.

Suitable data that are useful to inform management decisions is costly, and the patchwork of land managers make it difficult for any single agency to collect all the data necessary. By working together with accepted sampling protocols to efficiently collect and analyze bee data, land and species managers can more effectively collaborate toward the common goals of proactively managing these species and minimizing negative effects of other authorized land uses.

## VI. Outreach and Recruiting

We plan to continue and expand the bee monitoring effort and collaborate with agency partners and community (citizen) scientists in the following years. This will enhance data collection from a larger region to generate more accurate conservation status ranks. Community scientists trained by experts has been a successful model in developing a long-term bumble bee and butterfly monitoring project in Norway (Åström et al. 2019). Specifically, these data have detected changes in communities and abundance across time and space in a statistically defensible framework. See the Alaska Bee Atlas Community Science Plan (2021) developed by ACCS, BLM, FWS, and ADFG for more information.

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## Standard Operating Procedure A. Bee Bowl Traps

Bee bowls traps are established for a single or multiple sampling events.

Trap time length: 24 to 72 hours suggested. No more than 72 hours.

Trap number: 15 bowls

**Trap placement:** on the ground in a transect line in alternating colors within a homogenous habitat. For example, entirely within a forest setting, not crossing in other habitats such as alpine tundra or into floodplains.

**Supplies needed:** Sealable plastic bags, Specimen cups, Permanent marker, Pencil, Scissors, Flagging tape, Camera, GPS unit, Data sheet, Preservative, 15 Bee bowls, 1 Liter of water, unscented dish soap, Small strainer, Plastic spoon

## Setting out bee bowls:

Preparation

- 1. Pick a warm sunny day if possible. Rainy and windy days can limit pollinator activity.
- 2. Prepare a stack of bowls, consistently alternating between the colors (e.g. Yellow, White, Blue, Yellow, White, Blue, etc.). The order of color does not matter. Have a few extra bowls of each color with you just in case some in your stack are broken.
- 3. Fill a bottle almost full with water and add a large squirt of dish soap. Shake until mixed and frothy. Top off with water.

#### Set up

- 4. At transect start, mark where the locality of the first bowl with flagging tape to help with relocation. Create a GPS point of the transect start.
- 5. Place the bee bowls on the ground in a straight line transect approximately three to five meters apart (~5 big steps apart is a good rule of thumb). As you set each bowl down, fill it <sup>1</sup>/<sub>4</sub> full with the soapy water (do not overfill). Ensure the bowl is set secure on the ground so that it doesn't tip over or blow away. If the site is sloped or uneven, use a trowel, knife, or spoon, to dig out the soil or vegetation for secure placement of the bowl.
- 6. Photograph the site and fill out the AK Bee Atlas Survey Form as completely as possible.
- 7. Leave the bowls out for the designated amount of time.

## Collecting samples from the bee bowls:

Preparation

- 1. Create a label: cut out a small rectangle of paper for each transect. A template is on the Data Sheet.
- Label it with the site code, date of collection (DD-Month-YYYY), BOWL, and collector name in <u>pencil.</u> Place the <u>label inside</u> the specimen cup. NOTE: The insects from the net, bee bowls, and BVT will have their own respective specimen cup if multiple methods are being used.
- 3. With a permanent marker, write the same information on the <u>outside</u> of the specimen cup.
- 4. All of the bowl contents from one transect will be lumped together into one specimen cup.
- 5. Record in hours on the Data Sheet, the length of time the bowls were deployed collecting insects.

## Collect the bee bowls

- 6. Pick up a bowl and dump the water and insects (all of them) into the strainer.
  - Place the empty bowl in a collecting bag.
  - Repeat with the remaining bowls.
- 7. Once all bowls are emptied, gently scrape the insects from the strainer into the specimen cup with the plastic spoon. Insects are quite sturdy and gentle pressure will not harm the integrity of the insect, but be careful to not smear them against the strainer.
- 8. Fill the specimen cup with *just enough* preservative to cover the insects, do not overfill. Tighten the specimen cup and place in a plastic resealable bag or the provided WhirlPak. Secure the bag so no leakage occurs.
  - Alternatively, the insects can be stored in a cooler for up to 5 days before being frozen or adding preservative.



In this photo, the sample cups have enough liquid to cover the insects, but are nor overly filled with liquid.

## Delivering data and samples:

• Review Standard Operating Procedures D. Deliver the samples, data sheets, and photos to ACCS for processing.

Contact: Justin Fulkerson UAA/ACCS BMH 3211 Providence Drive Anchorage, AK 99508 907-786-6387 | jrfulkerson (at) alaska.edu https://accs.uaa.alaska.edu/wildlife/pollinator-diversity/

## Standard Operating Procedure B. Blue Vane Trap

A blue vane trap (BVT) will be established with bee bowls or alone to collect pollinators

**Trap time length**: 5 days to 7 days. No more than 10 days.

**Trap number**: a single BVT at a sample site.

**Trap placement:** if using alone, the BVT can be placed anywhere. If using with bee bowls, follow the bee bowl operating procedure and place the BVT at one end of the transect line.

**Supplies needed:** Sealable plastic bags, Specimen cups, Permanent marker, Pencil, Scissors, Flagging tape, Camera, GPS unit, Data sheet, Preservative, Blue vane trap, Zip ties, Stake or pole (1 meter height), Hammer, Fumigant strip, Insect net, Killing jar, and Killing agent.

## Setting out the BVT

Preparation

- 1. Pick a warm sunny day if possible. Rainy and windy days can limit pollinator activity.
- 2. If using a BVT in conjunction with the bee bowls, please follow the Standard Operating Procedure A. Bee Bowls., Setting out bee bowls to step #6.
- 3. Assemble the two vanes and push the vane unit into the blue funnel so it clicks into place.
- 4. Screw the blue vane and funnel onto the white bowl.
- 5. Add fumigant strip to the white bowl.



Figure 9. A blue vane trap being properly secured.

## Set up the BVT

- 6. Set the BVT in the desired sample area alone or at the end of the bee bowl transect.
  - The BVT can be placed at the start or end of the bee bowl transect.
  - Set the BVT within 5 meters of the bee bowls.
- 7. Pound a stake or pole <u>securely</u> into the ground but leave at least 1 meter (approximately) above ground.
- 8. Two zip ties will be secured around the white base of the BVT. Secure a wide zip tie (white in Figure 9) around the white bowl if one is not already secured. Secure a smaller zip tie (black in Figure 9) under the wide zip tie but leave a large loop.
- 9. Slip the smaller zip tie around the fixed pole snug but not tight as in Figure 9.

- 10. Thread string into a hole at the end of the vane and secure to the post as shown in Figure 9. Tightly secure the small zip tie in place.
- 11. Photograph the site and fill out the AK Bee Atlas Survey Form as completely as possible.
- 12. Leave the traps out for the designated amount of time.

## Collecting samples from the BVT

#### Preparation

- 1) Create a label: cut out a small rectangle of paper. A template is on the Data Sheet. **NOTE:** *The insects from the bee bowls and BVT will have their own respective cup if both traps are being used.*
- Label them with the site name, date of collection (DD-Month-YYYY), 'BVT', and collector name in <u>pencil.</u> Place the <u>label inside</u> the specimen cup. NOTE: The insects from the net, bee bowls, and BVT will have their own respective specimen cup if multiple methods are being used.
- 3) With a permanent marker, write the same information on the <u>outside</u> of the specimen cup.
- 4) Prepare a killing jar: place cotton balls or several crushed paper towels in a glass jar with 1 tsp. of ethyl acetate (nail polish remover). One killing jar per collection method only.
- 5) Record in hours on the Data Sheet, the length of time the bowls were deployed collecting insects.

#### Collect the BVT

- 6) If bees are still alive and actively flying in the BVT, a net and killing jar will be needed. *If not proceed to Step #7*.
  - a) Place the BVT under the net.
  - b) Unscrew the blue funnel/vane while under the net and allow the active bees to be captured in the net.
  - c) Transfer the bees to a prepared killing jar.
- 7) Collect all insects in the BVT by unscrewing the funnel and transferring the bees to the sampling cup.
- 8) Fill the specimen cup with *just enough* preservative to cover the insects, do not overfill. Tighten the specimen cup and place in a plastic resealable bag. Secure the bag so no leakage occurs.
  - a) Alternatively, the insects can be stored in a cooler for up to 5 days before being frozen or adding preservative.
- 9) Disassemble the BVT.

## Delivering data and samples

• Review Standard Operating Procedures D. Deliver the samples, data sheets, and photos to ACCS for processing.

## Contact:

Justin Fulkerson, UAA/ACCS BMH 3211 Providence Drive Anchorage, AK 99508 907-786-6387 | jrfulkerson (at) alaska.edu | https://www.alaskabeeatlas.org

## Standard Operating Procedure C. Net Capture for Bees

Participants will net bees within the survey area and use a killing jar or non-kill method to sample the bees. Please note that the non-kill method is only useful for bumble bees. We adopt the approach of the Pacific Northwest Bumble Bee Atlas (<u>www.pnwbumblebeeatlas.org</u>) for the non-kill methods.

**Time length**: 20 active minutes. If done in conjunction with bee bowls or BVT, perform this after the traps are set.

**Supplies needed:** Sealable plastic bags, Specimen cups, Permanent marker, Pencil, Scissors, Camera, GPS unit, Data sheet, Preservative, Insect net, Killing jar, Killing agent, Timer/stopwatch. *If using non-kill methods*: a photo chamber OR vials and cooler of ice

## Collecting samples

Preparation- choose one of the following

A. Killing methods: Prepare one or more killing jars.

- a. Place several cotton balls and/or crumpled paper towel in a killing jar.
- b. Add about 1 tsp of killing agent (ethyl acetate/nail polish remover) to the paper material.
- B. *Non-killing methods:* Prepare ice chest or insulated lunch box with ice and vials **OR** use the photo chamber which is ready to use.

Sampling with the net

Time the netting survey for 20 active minutes. Multiple people can help with the netting survey but the time should be adjusted for each person to total 20 person minutes. For example, if four people are netting, then each person will independently walk and net for 5 minutes.

- 1. Start a stopwatch
  - a. Walk slowly throughout the survey area, approximately a  $50m \times 50m$  square, OR survey up to 50 meters away from the middle of the bee bowl transect/BVT.
  - b. Maintain the walk within the same habitat as the bee bowl transect.
- 2. Find a bee or fly on a flower? Capture it!
- 3. Gently place the net over the flower, while holding the tip of the net up. This is often effective–insects typically will fly or crawl upwards when disturbed into the net. Using strong sweeping motions can damage plants and often results in missed captures.
- 4. After the insect is captured, close the net to prevent escape. Either turn the net over or close the opening or use your hand. *Please view this video tutorial from a colleague of the PNW Bee Atlas*: <u>https://youtu.be/mVOi-jRpw2E</u>
- 5. **Stop the stopwatch** and proceed with one of the following methods
  - a. <u>Kill method</u>
    - i. Transfer the pollinator to the kill jar while it is inside the net. You will need to slip the cover over on the jar before removing it from the net. Additional pollinators will be added to the jar.

- b. <u>Non-killing method ICE</u>
  - i. Transfer the bumble bee to a vial.
  - ii. Cover the vial with ice in the cooler for at least 10 minutes to slow down the bee for photographing. For a video demonstration please view: <u>https://youtu.be/TjaOpZWNF4M</u>
  - iii. Photograph the bee following the notes below.
- c. Non-Killing method PHOTO CHAMBER
  - i. Transfer the bumble bee to the photo chamber
  - ii. Push the rod or foam block to the clear plastic to lightly squash the bee for photographing. See photo tips below. For a video demonstration please view: <u>https://youtu.be/Wva3qKUwIsE</u>
  - iii. Photograph the bee following the notes below.

## 6. **Restart the stopwatch**

- 7. Resume the meander and repeat with new pollinators until the allotted time is complete. All pollinators collected from this 20 min. survey will go into the same killing jar or separate ice vials.
- 8. Photograph the site and fill out the AK Bee Atlas Survey Form as completely as possible.

Timing notes:

- > Watching and waiting for an *active* pollinator to land on a flower is acceptable.
- > Chasing after a pollinator is acceptable.
- Sitting and watching one flower for 20 minutes is <u>not</u> acceptable.

## Preservation from killing jars

- 1. At the end of the active time, transfer all collected pollinators to a specimen cup if they are dead. It's best to not keep insects in the kill jar for longer than 24 hours. If the insects are not dead, place the jar in the freezer for 1 hour, then transfer to the specimen cup.
- Create labels: cut out a small rectangle of paper and label it with the transect name, collection date (DD-Month-YYYY), 'NET', and collector name in <u>pencil.</u> Place the label <u>inside</u> the specimen cup. NOTE: The insects from the net, bee bowls, and BVT will have their own respective specimen cup if multiple methods are being used.
- 3. Fill the specimen cup with *just enough* preservative to cover the insects, do not overfill. Label the outside of the cup with the same information, tighten, and place in a plastic resealable bag. Secure the bag so no leakage occurs.

## Photographing for non-kill methods

Comprehensive photography tips and examples are at the PNW Bee Atlas project (<u>https://www.pnwbumblebeeatlas.org/photo\_tips.html</u>). Some general requirements for good photographs are:

- 1. Place the bee on a solid background such as the AK Bee Atlas Data Sheet. Write a number next to the bee so it can be identified as the same bee if your photo series.
- 2. Use an 8 Megapixel camera. Cell phone cameras are generally acceptable and handy for review of photos.

- 3. Photograph the bee next to the written number. Specific photos of the bees are needed. Move the bee around with a pencil to orientate the bee.
  - a) FACE at a slight side angle that shows the check length & colors
  - b) SIDE & ABDOMEN (tail/butt) at an angle getting the segment colors. The last few segments are crucial. It is helpful to place the bee on its back. Extra photos of this section are encouraged.
  - c) THORAX color patterns, easiest to take photo at a top view.
- 4. <u>Ice method:</u> after at least 10 minutes, the bumble bee should be cold and inactive. Uncap the vial and gently place the bee on a solid background. We have found Alaska bumble bees are cold tolerant and need more time to cool down. Add 3 min. until the bee is not actively moving or flying in the vial. Depending on the outdoor temperature, it may take up to 10 min. to warm up and fly away. There is plenty of time to take excellent photographs. Direct sunlight will warm the bumble bee up sufficiently, however in cloudy or shady conditions if you feel the bumble bee is taking too long to become active, cup your <u>warm</u> hands over the bee without touching it.
- 5. <u>Photo chamber method:</u> you will not be able to manipulate the bee to get the full range of photos, but please do your best. Pulling the foam back from the viewing window will allow the bee to move around slightly giving you another chance to reposition the bee for additional/better photo angles. Avoid glare from the acrylic window.



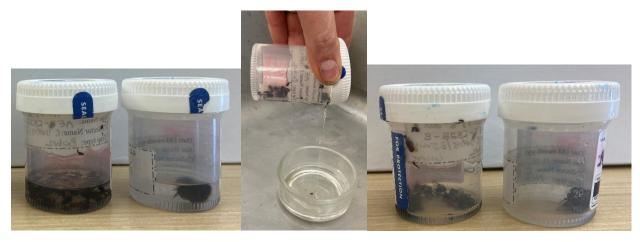
*Top photos*: These are very good photos of the abdomen and side view. *Bottom Left*: Excellent close-up view of the abdomen. *Bottom Right*: A poor photo quality, there is low light and far away.

# Standard Operating Procedure D: Shipping Bees and Data Delivery

After returning from the field, the data sheets can be scanned and emailed to ACCS or included with the bees during mailing. Please email photos of the site location and action photos of hard-working people.

## Prepare the sample cups for shipping

- 1. Unscrew the cap.
- 2. Using the cap as a catchment sieve, pour out as much of the preservative liquid as possible. Residual liquid will be enough to keep the insects moist. Ethanol and propylene glycol are non-hazardous and nearly food grade, therefore they can be poured down the drain or outside.
- 3. Retighten the cap as securely as possible.
- 4. Place up to two sampling cups in the provided WhirlPak with at least one crumpled paper towel and secure tightly. Place inside another resealable bag. If the WhirlPak is not available, use a resealable bag.
- 5. Ensure the sample cups are double bagged and secure.
- 6. Place the bag and data sheets in a padded envelope or cardboard box with padding. Mail the package to the address below immediately. First class postage, flat rate box, or UPS Ground are acceptable.



Left: sample cups with preservative. Middle: preservative being poured out. Right: sample cups with very little preservative remaining.

*Mail to:* Justin Fulkerson UAA/ACCS BMH 3211 Providence Drive Anchorage, AK 99508

## **Appendix A. Supply Photos**





First row left: Specimen cup
First row middle: Preservative bottles
First row right: Dish soap bottle
Second row left: Killing jar
Second row middle: Non-kill vial (left) photo chamber (right)
Second row right: collapsible net pieces
Third row left: assembled collapsible net



Collector Name:

Trap type:

Contact: J. Fulkerson, jrfulkerson@alaska.edu www.alaskabeeatlas.org



Collector Name:

Trap type:

## AK Bee Atlas Survey Form 2023 Survey Type: Bowls / BVT / Net Capture

Survey Date (day-month-yy):

## Site Information the 'site' is composed of the traps and a 100m buffer radius in the same habitat

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Cloud cover %:	Cloudy	Partly Sun	wind.	light breeze (4-7 mph)		gentle breeze (8-12 mpl		
Habitat Informa	tion				-	· • •	-	
Please take a photo	o of the ha	bitat with th	e traps or i	the netting a	rea.			
General Habitat T	ype:							
Agriculture	Alpine		Grassland	d	Dunes		Coastal beach	
Developed/Roadsid			Meadow		Marsh/Bo	g	Floodplai	
City/Village	Forest m	0	Shrub/Sc	rub	Lake		Post-Burn	1
Garden/Park	Woodlar	nd/Forest	Bluff	ff River/Stream		eam		
other description:								
The vegetation of t	he samplin	g area is pr	rimarily con	mposed of (p	ick ONE):			
Conifer tree	Tall shru	ıb (>1.5 m)	Low shru	ıb (0.2-1.5 m	l)	Grass/Sed	lge	Forbs
Broadleaf tree Barren ground Dwa						Moss/Lich	oss/Lichen	
Vegetation Classifi	ication (op	tional) Vier	eck or NVC	CS:				
Floral Resource	es							
How many differen	nt flower sp	oecies do yo	u see in the	e survey area				
1 2	3	4	5	6	7 to 9	10 to 12	13 to 15	>15
Flowers observed			-		•		-	
abundant plants in	•	-	•		•			
photo numbers, the							-	
observerved, recor	d the type	0 0		ly, bumble be	ee, bettle, or	0.00	-	
Common Name	or	Scientific	Name			Photo #	Floral Vi	sitor
Cut Out Labels				es: BOWL, E				
NOTE: If multiple s			-					cup
Cut out this label a	<u>^</u>	inside the			s template fo	or additional		
Date (dd-month-yy	y):			-month-yy):				month-yy):
Site Name:			Site Nam	ne:			Site Nam	e:

Collector Name:

Trap type:



Contact: J. Fulkerson, jrfulkerson@alaska.edu www.alaskabeeatlas.org



## AK Bee Atlas Survey Form

Flowers observed within the site continued from page 1. Use an additional sheet if needed.

Common Name	Scientific Name	<b>Floral Visitor</b>

**Notes or Site Sketch:** (optional) sketch the site with transect line of traps and other helpful features to help retrieve bowls and display the site.