U.S. Fish and Wildlife Service
U.S. Department of the Interior

Voluntary Survey and Monitoring Technical Assistance and Best Survey Practices for the Rusty Patched Bumble Bee (*Bombus affinis*)

Version 3

September 29, 2025

Photograph by Susan Day

Contents

Survey and Monitoring Technical Assistance Development

USFWS biologists developed survey protocols for *B. affinis* that are effective in meeting survey and monitoring objectives and unlikely to negatively impact the species. We obtained guidance from bumble bee experts including biologists from State and Federal agencies; scientific and academic institutions; and conservation organizations. In addition, several State, Federal, academic, and conservation organization biologists gathered for a May 2025 U.S. Geological Survey (USGS) long term monitoring workshop at the Powell Center in Fort Collins, Colorado, for the rusty patched bumble bee workshop, which informed much of this document. We considered the best available information for all aspects of the technical assistance and will continue to work with surveyors to collect additional data on the distribution and ecology of *Bombus affinis*. This technical assistance document is subject to continual improvement and modification.

Suggested Citation

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Introduction and Purpose

Once widespread and abundant, the rusty patched bumble bee (*Bombus affinis*) has undergone precipitous declines and was listed as an endangered species under the Endangered Species Act on January 11, 2017(FR50 CFR 17 38116 January 11, 2017; USFWS 2017). At the time of listing, we were aware of extant *B. affinis* records in isolated places within 13 states and 1 province since 2000 (USFWS 2017). Since the 2017 listing, we have become aware of 2 additional states and 1 additional province with historical records(USFWS 2022). Sightings of *B. affinis* were common prior to the early 1990s, and it was considered relatively abundant across 28 states, the District of Columbia and three Canadian provinces). Since then, it has experienced a swift and dramatic decline in abundance and distribution of approximately 90 percent or more. Similar declines have occurred in Canada (COSEWIC 2010) where it was listed as Endangered on Schedule 1 of the Species at Risk Act in 2012 (https://www.canada.ca/en/environment-climate-change/services/species-risk-act-accord-funding/listing-process/wildlife-species-amendment-list-schedule-1.html).

While we recognize the great importance of conducting surveys for bees (and other insects), we need to be particularly cautious in areas that may contain *B. affinis*. We must carefully consider the benefits of surveys versus their potential to negatively impact the rusty patched bumble bee. Therefore, we recommend non-lethal protocols in areas where we may encounter the species (*e.g.*,

areas within **High Potential** and Low **Potential Zones** refer to definitions below and online at https://www.fws.gov/species/rusty-patched-bumble-bee-bombus-affinis).

Survey effort throughout the historic range of *B. affinis* has not been systematic and the occupancy of some sampling units is uncertain. For these reasons the Service recommends non-lethal survey protocols in areas where there is a relatively high potential of encountering the species and in areas where there is a low, but still some, potential of encountering the species. We have developed a high potential zone model (the **High Potential Zone Model** is described online at https://www.fws.gov/media/high-potential-zone-model-rusty-patched-bumble-bee) that identifies geographic areas to focus survey effort and where to be cautious regarding potential take associated with surveys.

The objectives of the survey protocols are to: (1) find and document new *B. affinis* locations; (2) determine if *B. affinis* are still extant at new or previously documented locations where federal projects are proposed; (3) monitor bumble bee populations to determine long-term occupancy or abundance; and (4) provide protocol recommendations for areas locations we believe are unoccupied by *B. affinis*.

Because *B. affinis* surveys can result in take (by capture), such surveys should only be conducted by a qualified biologist[1]. Generally, a recovery permit for *B. affinis* authorizes the capture of bees for identification and handling of bees for photography. Following this guidance will meet standard USFWS requirements for conducting surveys and monitoring under a federal scientific recovery permit under section 10(a)(1)(A) of the ESA. However, surveyors also need to ensure they meet all applicable state permitting and reporting requirements. For further information about obtaining a federal permit and surveyor qualifications please refer to **Obtaining a permit under the Endangered Species Act and Frequently Asked Questions** available online at (https://www.fws.gov/media/recommendations-obtaining-rusty-patched-bumble-bee-recovery-permit).

This document provides the U.S. Fish and Wildlife Service's (USFWS) recommended survey methodology and outlines additional reporting requirements for federal scientific recovery permit holders. Future changes to this document may occur and will be posted on the USFWS rusty patched bumble bee website (https://www.fws.gov/media/survey-protocols-rusty-patched-bumble-bee) by March 15th of each year. Before conducting surveys, please check the website to ensure use of the most current version of this document. Contact the USFWS Field Office near you (https://www.fws.gov/locations) if you are interested in collecting data that is not discussed in this document.

[1] A qualified biologist is an individual who holds a USFWS Recovery Permit (Federal Fish and Wildlife 10(a)(1)(A) Permit) for *B. affinis* in the state/region in which they are surveying and/or has been authorized by the appropriate state agency to capture and handle *B. affinis*. See **Obtaining a Permit under the Endangered Species Act and FAQs** available online at https://www.fws.gov/media/recommendations-obtaining-rusty-patched-bumble-bee-recovery-permit.

Definition of Terms used in this Document

- **Sampling Unit.** For the purposes of this document, a **sampling unit** is defined as a record (observation point) and the surrounding area (collectively, a "patch"). There may be one or more sampling units within any of the zones described below. A standard sampling unit as a 3.14 ha patch (100m radius circle) around a point location, although your unit area may be smaller.
- **Extant Sampling Units** are defined as sampling units where *B. affinis* has been documented in 2007 or later, unless surveyed sufficiently to be considered unoccupied. Ideally, extant sampling units will have multiple years with records of the species. If no or little effort has been spent attempting to relocate the species at the sampling unit, then additional surveys are needed before considering the sampling unit to be unoccupied.
- **High Potential Zones** are based on the habitat connectivity model described in detail online (refer to **Habitat Connectivity Model** at https://www.fws.gov/media/high-potential-zone-model-rusty-patched-bumble-bee). These High Potential Zones contain **extant sampling units** (refer to definition below) and the surrounding area considered to have highest potential for the species to be present, as generated from the model. These zones are not of uniform size and have discrete boundaries that can be used to determine where non-lethal surveys or scientific recovery permits may be recommended (Fig. 1 and https://www.fws.gov/media/high-potential-zone-model-rusty-patched-bumble-bees). The zones range from about 1 to 2 miles (1.6 to 3.2 km) from extant sampling units. For recommendations regarding protocols and scientific recovery permits in High Potential Zones, refer to Fig. 2.
- **Low Potential Zones** include: **Uncertain Zones** (defined below and online at https://www.fws.gov/species/rusty-patched-bumble-bee-bombus-affinis) and **Primary Dispersal Zones** surrounding **High Potential Zones** (Fig.1). For recommendations regarding protocols and scientific recovery permits in Low Potential Zones, refer to Fig. 2.
- **Primary Dispersal Zones** are based on the habitat connectivity model described in detail online (refer to **Habitat Connectivity Model** online at https://www.fws.gov/species/rusty-patched-bumble-bee-bombus-affinis). Primary Dispersal Zones considers the maximum dispersal potential from known sampling units and across the range. These zones, although not of uniform size, have discrete boundaries that can be used to determine where non-lethal surveys are recommended and where a scientific recovery permit for surveys is recommended (refer to Figs. 1 and 2).
- **Uncertain Zones** have records of the *B. affinis* in 2000 to 2006 out to and including the surrounding dispersal area, but with fewer than 3 years of negative survey data with sufficient effort[2] since the last known record. If no or little effort has been spent attempting

[2] Sufficient effort would consist of five to six approximately equally spaced sampling periods during the peak flight season (mid-June to late-August); 30-minutes of search time per 100m radius sampling unit focusing on blooming foraging habitat (defined below) using non-lethal techniques. This document provides further details on methods, techniques, and best practices and is subject to continual improvement and modification.

to relocate the species at the sampling unit, then additional surveys are needed before considering the sampling unit to be unoccupied. Areas remain uncertain until they have either (1) a positive detection, when the sampling unit would be considered extant or (2) have at least 3 years of negative survey data with sufficient effort, when they would be considered unoccupied.

- **Unoccupied Zones** are areas where the likelihood of encountering *B. affinis* is so low that the area is considered unoccupied and includes areas outside of the High Potential and Primary Dispersal Zones; sampling units where last known *B. affinis* record is from before 2000 and unoccupied sampling units (defined below) with at least 3 years of negative surveys with sufficient effort since the last known record. For recommendations regarding protocols and scientific recovery permits in Unoccupied Zones, refer to Table 1.
- **Unoccupied Sampling units** are where the species has been previously documented since 2000, but where there have been at least 3 years of negative surveys since the last known record (*i.e.*, occupancy or abundance methods). Sampling units that only have records older than 2000 are also assumed to be unoccupied unless new information (*e.g.*, recent positive *B. affinis* detection) suggests otherwise.

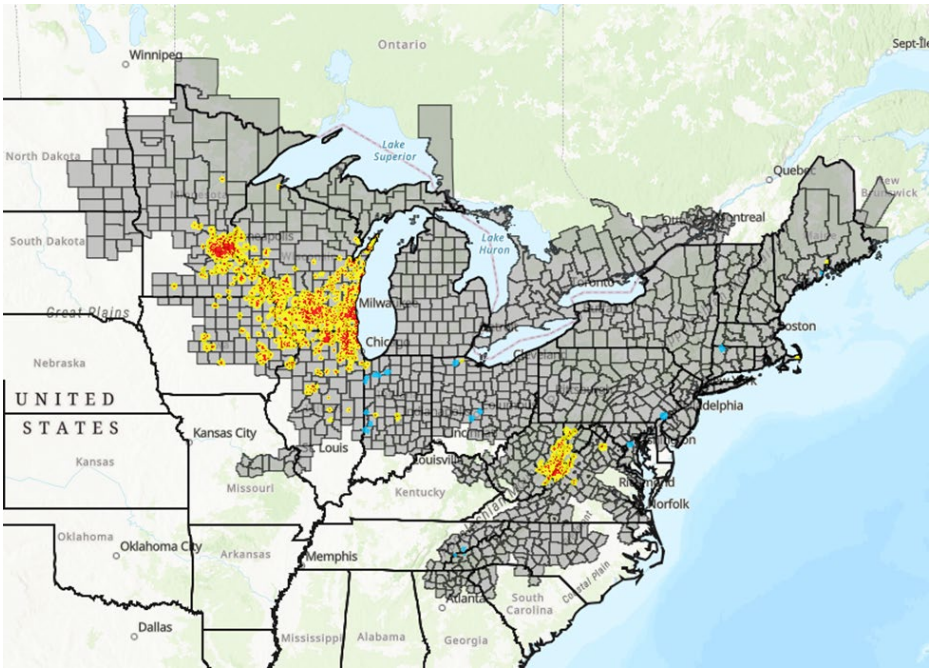

Figure 1. "**High Potential Zones**" (red dots), Uncertain Zones (blue), and Primary Dispersal Zones yellow) (together, the Uncertain and Primary Dispersal Zones comprise the "**Low Potential Zones**") for *B. affinis*, based on a habitat connectivity model and on species survey data compiled through the previous field season. The gray shaded area is the historical range of *B. affinis* and is called the "**Unoccupied Zone"**. This map is updated regularly, so we recommend that surveyors visit the FWS *B. affinis* website for the most current information. Description of the habitat model, an interactive map, and downloadable shapefiles are available on the *B. affinis* website (https://www.fws.gov/species/rusty-patched-bumble-bee-bombus-affinis, accessed September 3, 2025).

Endangered Species Act (ESA) scientific recovery permits within USFWS mapped zones

Below are recommendations for obtaining scientific 10(a)(1)(A) recovery permits under the Endangered Species Act (ESA) for intentional take for recovery and scientific purposes within the High Potential, Low Potential, and Unoccupied Zones (also, refer to Figs. 1 and 2).

High Potential Zone (HPZs)- With respect to typical foraging distances and potential dispersal movements of *B. affinis*, the High Potential Zones (HPZs, Fig. 1) provide a reasonable basis for describing where the species is likely to be present and as such, researchers planning to handle bumble bees in HPZs are advised to obtain a scientific recovery permit (refer to Fig. 2). Federal projects and projects with a federal nexus that intersect with HPZs consult with the USFWS to evaluate the potential effects of their actions.

Low Potential Zone (LPZs) – Low Potential Zones (LPZs, Fig. 1) buffer high potential zones and are much less likely to support existing populations. However, based on the last few years of *B. affinis* observations and the observed dispersal distances of a closely related species (*B. terrestris*, 1 to 10 km(0.6 to 6.2 mi) (Kraus et al. 2009, p. 249, Lepais et al. 2010, pp. 826-827) the species may be found in some of these areas. Therefore, since there is some chance that a surveyor looking for *B. affinis* may encounter the species, we recommend that surveyors in LPZs obtain a scientific recovery permit (Fig. 2).

Unoccupied Zones – We believe the likelihood of finding a *B. affini*s in unoccupied areas (refer to Figs. 1 and 2) and scientific recovery permits for rusty patched bumble bee are not required in these areas. However, if *B. affinis* is observed or collected in unoccupied zone, please stop lethal activities in that area and promptly notify the USFWS.

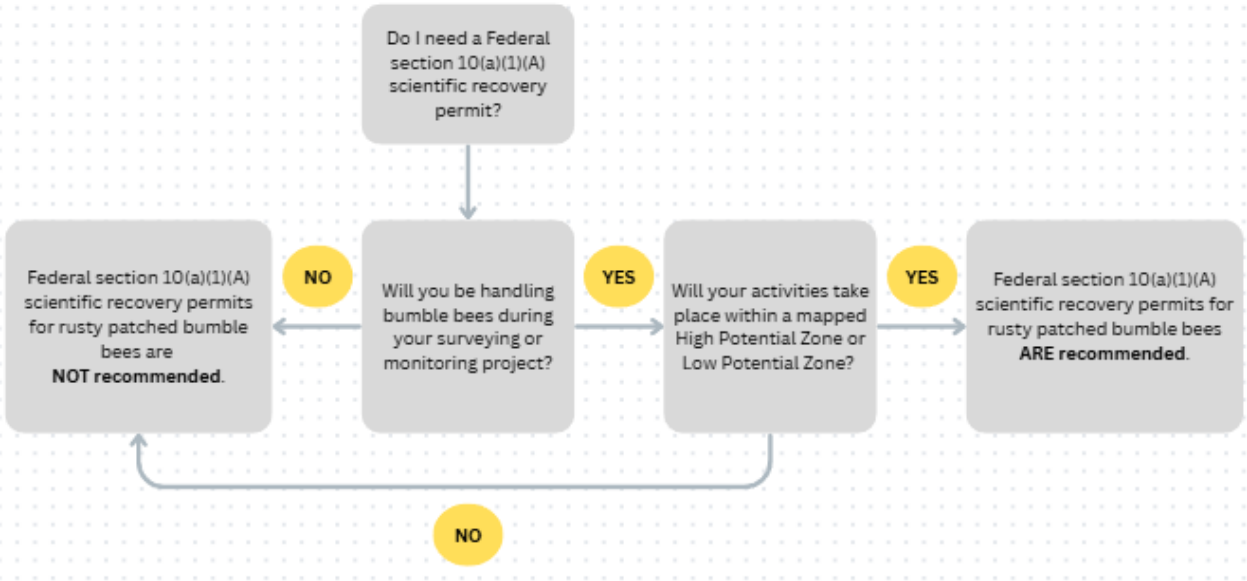

Figure 2. Flow chart to help determine whether a federal ESA section 10(a)(1)(A) scientific recovery permit is recommended.

Best Practices for Surveying and Monitoring

We recommend following best practices when surveying or monitoring for rusty patched bumble bees:

- ***Adhere to the Endangered Species Act and determine if you need a survey permit.*** Details on how to apply for a permit are provided online at -**Obtaining a Recovery Permit under the ESA and FAQs** (https://www.fws.gov/media/recommendations-obtaining-rusty-patched-bumble-bee-recovery-permit). USFWS only has regulatory authority over federally listed species. Keep permits with you while conducting surveys.
- ***Obtain land-owner permission*.** Written permission is recommended. Keep permission paperwork and landowner contact information with you while surveying.
- ***Conduct surveys during suitable survey conditions*** (below).
    - **Time of Year** - Surveys must be conducted between mid-June and late-August, for the highest detection probability (e.g., Otto et al. 2023) and to reduce potential impacts to *B. affinis* queens.
    - **Weather -** Surveys should take place when temperatures are above 60°F (15.5°C) and not during wet conditions (*e.g.*, foggy, raining, or drizzling). Wait at least 1 hour after rain subsides before conducting a survey. Sunny days with low wind speeds (less than 8 mph) are optimal. Partially cloudy days or overcast conditions are permissible if you can still see your shadow or see flying bumble bees.
    - **Time of Day -** Surveys should be conducted at least 2 hours after sunrise and 2 hours before sunset.

- ***Netting Technique.*** Collect bumble bees directly from flowers. Videos on netting techniques are available here: https://www.youtube.com/watch?v=SwYbv5bySPQ. If possible, capture one bee at a time or immediately separate bees into individual containers.
- ***Handling.*** Do not hold a bee in a container for longer than 15 minutes, unless you place it in a cooler with ice to later photograph the bee. Hold only one bee per container. Do not hold bees in a cooler with ice for more than 2 hours and do not place bees directly onto ice. If the air temperatures are above 90°F (32°C), do not hold for longer than 5 minutes. Bumble bees can easily overheat, so do not keep vials or bags in direct sunlight. Keep the bees in the shade, if possible.
- ***Photographs.***
    - Take several photographs of each *B. affinis* (and other species of interest in your area, *e.g., B. terricola* and *B. pensylvanicus*). Photographs are required to verify *B. affinis* observations.
    - Photograph the bee from the top (dorsal view) showing the entire bee, including the top of thorax and abdomen. Also photograph the face from the front and top, and side view of thorax and abdomen (**refer to Appendix A - Standardized Bee Photography**). Short videos of *B. affinis* are also recommended.
    - Photographic documentation requirements for non-target *Bombus* may vary by project objective(s).
    - In some cases, visual surveys (*e.g.*, using butterfly binoculars) to count or estimate species may be the preferred technique for individuals highly experienced in identifying bumble bees. Photographic documentation is a necessary secondary component to visual surveys (*e.g.*, if *B. affinis* is observed).
- ***Identify and Verify.*** Identify the bumble bees or submit photographs for verification.
    - Refer to **Appendix B – Bee Identification.**
    - Refer to **Appendix C - How to Verify Records.**
- ***Marking.*** Depending on the specifics of your survey, you may choose to use blue chalk dust, paint pens, or nail polish to temporarily mark bees, other than *B. affinis*, to avoid recapture of individual bees (*e.g.*, if you process bees right away). Do not mark bees on the wings. In some cases, marking *B. affinis* may be permitted for individuals highly experienced in marking bumble bees - contact your state's USFWS Ecological Services Field Office (https://www.fws.gov/locations) if this is something you are considering.
- ***Release.*** Release bees back on or near flowers on which they were found. If you held bees in a cooler with ice, first transfer bees to a warm container after being in cooler (as the original container will fog up when it hits the warm humid air). Place the open vials (or bags) in a shaded area to allow the bees time to warm up and fly away. Release *B. affinis* within 15 minutes of capture, if possible.
- ***Disinfect.*** Use a disinfecting or sterilizing agent (e.g., isopropyl, bleach solution) to clean all equipment used to handle bees between sampling units. If temporarily holding bees (e.g., in vials), use one per individual bee and clean between sampling units.
- ***Reporting.*** Report sightings of *B. affinis* as described below and as required on your permit(s), if applicable. Any collections or sightings of *B. ashtoni* should also be reported as

this is a nest parasite of rusty patched bumble bee (and *B. terricola*) and thus could potentially indicate the presence of *B. affinis* in the area. Although not required for several protocols discussed below, USFWS is also requesting that surveyors report observations of other species of conservation concern and the plant species the bee was using.

- ***Salvage.*** *B. affinis* that are found dead or killed accidentally may be salvaged. Prior to collecting specimen(s), photograph the specimen to document the condition and circumstances. Notify the USFWS within 48 hours. Preserve the specimen(s) in a freezer including proper identification and data [include date, complete scientific and common names, and geographic location (latitude and longitude in decimal degrees), and place in a freezer until you receive further instructions. USFWS will provide further instructions for possible scientific uses of specimens (e.g., pathogen or genetic analyses). All specimens of *B. affinis* collected under your federal permit are the property of the United States Government and should clearly be identified as such. Dead specimens that are not being used for additional analyses should be sent to a public scientific or educational facility or museum in the state the individuals were collected along with a copy of the permit(s) under which they were collected. Specimens kept at scientific or educational facilities should be preserved using standard museum practices[3].

Equipment basics

We suggest bringing the following equipment:

- **Permits.** Individuals planning to handle bumble bees in High or Low Potential Zones are recommended to obtain a federal scientific recovery permit that covers *B. affinis* prior to conducting surveys (https://www.fws.gov/media/recommendations-obtaining-rusty-patched-bumble-bee-recovery-permit).
- **Nets.** Use cloth aerial hand nets. Do not use sweep, beater, or wire nets. Netting should be fairly transparent.
- **Bags and/or clear vials.** If using vials, make sure vials allow for clear photographing of specimens inside (test before you go into the field). If using plastic bags, bring extra bags in case moisture builds up or bags get wet. There will be sufficient air in any of these containers while taking photographs.
- **Binoculars**. Butterfly binoculars are recommended for visual surveys.
- **Camera or Phone with camera** (refer to **Appendix A - Standardized Bee Photography**).
- **Cooler** with ice, if applicable.
- **Disinfectant** (e.g., bleach solution, isopropyl) for equipment (e.g., vials).
- **GPS unit or phone with GPS capabilities.**
- **Field data sheets** (digital or paper, for both bee surveys and sample location assessments).
- **Pencils.**

[3] You may find tutorials and protocols online, for example, see methods on the University of Minnesota webapge http://www.extension.umn.edu/youth/mn4-H/projects/environment/entomology/collecting-and-preserving-insects/

- **Timer or stopwatch.**
- **Hand lens or loupe.**
- **Long measuring tape (100m) or rangefinder.**
- **Flags or stakes to mark sampling site boundaries**, if applicable.
- **Safety gear**, as appropriate (e.g., first aid kit, sunscreen, EpiPen).
- Water and snacks.
- **Regionally appropriate bumble bee** and **plant species identification guides.**
- All Federal, State, and Local **Permits**, if applicable.

Surveys and Monitoring within the High and Low Potential Zones

The following methods and techniques are common to all the recommended protocols in this document. Methods specific to each protocol are provided under each protocol description, below these initial steps.

Initial Steps

Before you begin, conduct the following initial steps:

1. **Determine your survey purpose or objectives**. Figure 3 is a useful tool for this exercise.
2. **Determine your area(s) of interest.** Area(s) of interest will depend on the objectives and purpose of your study. Examine recent aerial photography using Google Earth or other tools to identify potential habitat for bees (refer to **Appendix D – Suitable *B. affinis* foraging habitat**). Examine and consider vegetation surveys or floral lists, if available for the location.
3. Prior to survey date, (if time permits), field-truth the selected area. Check the area for floral resources.
4. **Select Method and Protocol** for your objective (refer to **Fig. 3**). You may add additional methods at your discretion, as appropriate given your objective(s).

Objectives

This document is set up to accommodate several objectives (refer to Fig. 3) including 1) finding **new *B. affinis* locations**, 2) **assessing proposed projects** for *B. affinis* presence, 3) assessing the ***Bombus* community**, 4) informing *B. affinis* **recovery plan** targets (USFWS 2021), 5) assessing *B. affinis* **habitat or population management**, or 6) **improving scientific understanding** of *B. affinis*. Our recommended methods and protocols provide for standardized data gathering that can be supplemented with other techniques and methods to meet your specific project objectives. For example, if you are interested in assessing *B. affinis* populations for a proposed habitat management project, we recommended using the occupancy protocol before and after the management. You may also choose to conduct an in-depth floral assessment to assess floral diversity or implement a

genetics study to assess the genetic health of bumble bees at your location, for example. The baseline occupancy data can contribute to larger landscape level assessments that also use the occupancy protocol.

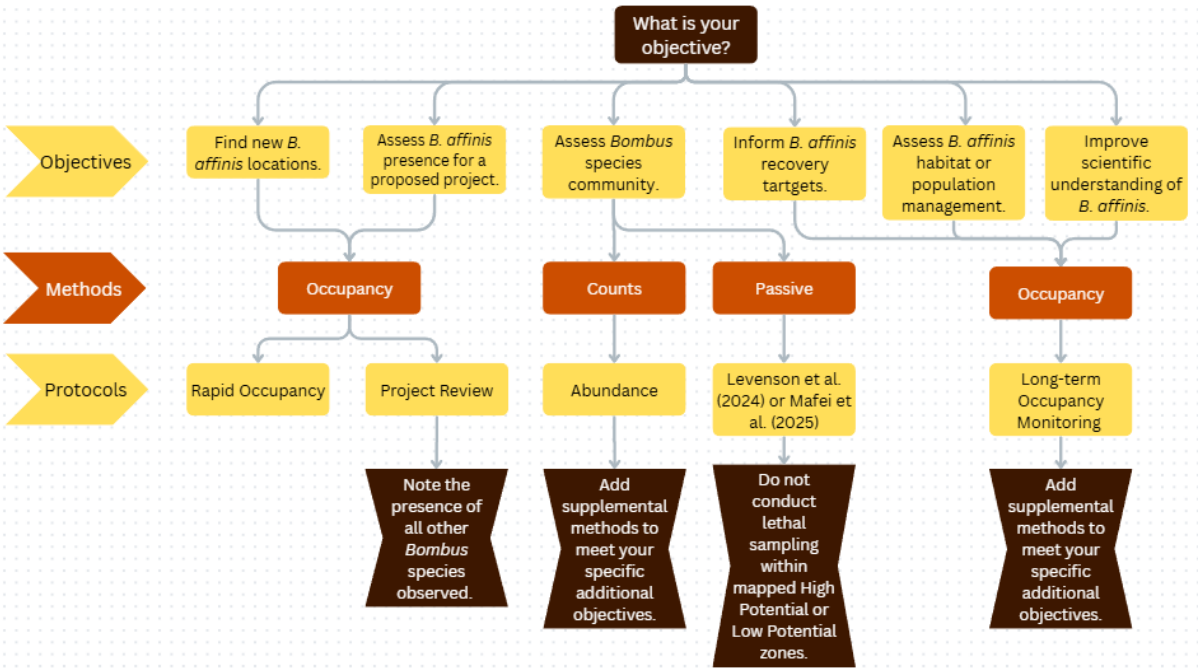

Figure 3. Flow chart to help determine the recommended method and protocols based on your fundamental objective(s).

Methods and Protocols

Use Figure 3 to determine which method and protocol are appropriate given your study objective(s). Conduct survey or monitoring accordingly. Below are brief descriptions of two methods that may be conducted within the mapped High Potential and Low Potential Zones. Specific protocols are provided after the brief method descriptions.

Occupancy Method

- Use non-lethal (e.g., photograph, netting, vial) techniques.
- Each observer should conduct a meandering survey walk (or similarly sized transect) for 30 minutes within a 100m radius (3.14 acre) sampling unit (or equivalent size).
- Ensure that each observer is independently conducting the survey (e.g., do not walk side by side, start at opposite ends of the sampling unit). Record the effort and results of each observer separately.
- **Record detection and non-detection of *B. affinis***. Photographic documentation of *B. affinis* is required to verify the record.
- **Record all castes *B. affinis* detected** and the plant species that the species was using (refer to the Sample Data Sheet in **Appendix E**).

- Note the time of the first *B. affinis* detection (e.g., *B. affinis* detected after 12 minutes), if applicable (refer to the Sample Data Sheet in **Appendix E**). Record the times of additional *B. affinis* detections if possible.
- Photographic documentation of *B. affinis* is required to verify the record.
- Collect core data fields (Table 2). These data match the information requested on the **Rusty patched bumble bee reporting form** (available online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form) to quantify survey conditions.

Count Method

- Use non-lethal (e.g., photograph, netting, vial) techniques.
- Each observer should conduct a meandering survey walk (or similarly sized transect) for 30 minutes within a 100m radius (3.14 acre) sampling unit (or equivalent size).
- Ensure that each observer is independently conducting the survey (e.g., do not walk side by side, start at opposite ends of the sampling unit). Record the effort and results of each observer separately.
- **Record detection and non-detection of *B. affinis***. Photographic documentation of *B. affinis* is required to verify the record.
- **Record the caste and number of any *B. affinis* detected** and the plant species that each caste was using (refer to the Sample Data Sheet in **Appendix E**).
- Note the time of each *B. affinis* detection (e.g., *B. affinis* detected after 12 minutes), if applicable (refer to the Sample Data Sheet in **Appendix E**).
- **Count all *Bombus* species observed** and note the species of plant(s) the other species are using (refer to the Sample Data Sheet in **Appendix E**).
- You may use visual surveys to count or estimate numbers of each species, however, this method is only advised for individuals highly experienced in identifying bumble bees in the field and is recommended only for commonly observed species. Indicate degree of confidence in identifications on the field sheets and data forms.
- The data sheet will provide space to tally the numbers of each species observed. Note how these were counted (i.e., actual counts or estimates) on the field sheets and data forms, to ensure that numbers are not misinterpreted.
- Collect core data fields (Table 2). These data match the information requested on the **Rusty patched bumble bee reporting form** (available online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form) to quantify survey conditions.

Table 1. Effort and area for each protocol.

Protocol	Method	Sample unit area (ac)	Effort per sampling event (minutes per independent observer)	Minimum number of sampling events per season	Minimum number of years
Rapid Occupancy	Occupancy	3.14	30	1	1
Project Review	Occupancy	3.14	30	5-6	1
Abundance	Counts	3.14	30	5-6	1
Long-term Occupancy	Occupancy	3.14	30	5-6	3
Levenson et al. (2024) or Mafei et al. (2025)	Passive	NA	NA	refer to protocol	refer to protocol

Protocols

Four protocols are recommended for rusty patched bumble bees, which should be chosen based on your study objectives (refer to Fig. 2).

All Protocols

The following steps are common to all Protocols on the day(s) of the sampling event. Specific protocol descriptions follow this section.

1. **Select and define the sample unit**
    a. Determine the area (**sampling unit**) that you would like to survey. The size of the sampling unit is at your discretion, but sufficient effort should be applied to the amount of area surveyed. We define a standard sampling unit as a 3.14 ha patch (100m radius circle) around a point location. If used, transects should contain a similar total area.
    b. If you have discretion on where you will conduct surveys, we recommend prioritizing your survey locations in areas with high quality habitat in your geographic area of interest in the **Unoccupied Zones**. Contact the USFWS Field Office (https://www.fws.gov/locations) to help find priority areas within in your area of interest or use our guidance to help identify **priority survey areas** (online at https://www.arcgis.com/home/webmap/viewer.html?webmap=feab9e66608840818795c2dfef93993b&extent=-95.4873,36.0689,-69.5815,46.6361).
    c. **Distance between Sampling Units** – If you would like to survey multiple sampling units, we suggest that surveyed sampling units be spaced at a minimum of 1 km (0.62 mi) apart to ensure independence of surveys.
    d. **Core.** Record the name and size of the surveyed area (radius or ac) and sketch the sampling unit on your field data form (e.g., field sheet, digital survey 1-2-3 form). Delineate the sampling unit (e.g. use ArcGIS to draw polygons around the sample unit).
    e. **Core.** Use a portable GPS device to record the coordinates (decimal degrees is preferred) at the approximate **center of the sampling unit**.
        i. For **long-term monitoring, the start point should remain constant**, though different areas may be searched within the sampling unit as floral resources change throughout the sampling seasons.
2. **Core.** Record the date of the sampling event, the field observer name, and permit number if applicable.
3. **Record environmental, habitat, and floral resource data**
    a. **Core.** Record basic information about the dominant habitat type, management, and major stressors at the sampling unit.
    b. **Core.** Collect the percentage of your sampling unit that is covered in flowering resources in bloom (refer to **Appendix D**).
    c. **Recommended.** On the day of the sampling event(s), take representative photographs, showing areas of suitable (high quality) and unsuitable (poor) habitat.

d. **Optional.** If not guided otherwise by a particular protocol, you may conduct more detailed habitat assessments at your discretion, for example, use Robinson's (2024) "project-specific bumble bee habitat assessment " or the Xerces Rusty Patched Bumble Bee Habitat Assessment Form and Guide (Xerces Society for Invertebrate Conservation 2017) depending on your project objectives. For example, the Xerces assessment helps conservation planners and landowners prioritize conservation actions and quantify habitat or land management improvements for *B. affinis*. This assessment also helps identify specific actions for habitat improvement and management practices to help protect *B. affinis* from potential threats.

1. Rapid Occupancy Protocol

Surveys to find new populations should place emphasis on maximizing the possibility of finding new *B. affinis* locations through surveying many areas, rather than repeated sampling in one area (refer to Fig. 2). Rapid surveys may be used for quick searches for *B. affinis*.

a. **Methods.** Use **occupancy methods** as described above.
b. **Effort.** Each observer should conduct a meandering survey walk for **30 minutes** within the 100m radius (3.14 ac) sampling unit. Record the effort of each observer separately. Only **one visit** during the height of the flight season (ideally July or August) is needed for a rapid survey, however repeated sampling is encouraged and necessary if there is a desire to have confidence in non-detection data.
c. **Conduct** the sampling event.
    1. **Start** your timer when you begin the sampling event.
        - **Core.** Record the start time.
    2. Search for bumble bees on flowers within the **highest quality foraging habitat** in your survey area. Spend minimal time in lesser quality habitat. Scan the areas for bumble bee activity and move towards those areas. Walk from flower patch to flower patch looking for active bumble bees and record detection and non-detection of *B. affinis*.
    3. **Core. Record the data fields as noted in Table 2 for the Rapid Protocol.**
    4. **Catch or photograph bumble bees**, preferably one at a time. Move bees from aerial nets to clear vials or plastic bags for easier identification.
    5. Pause your timer while you are not actively searching for bees (*e.g.*, while you are putting bees in your cooler, walking between habitat patches, or taking photographs).
    6. Representative photographs of each bumble bee species in each sampling location may be preferred depending on your specific project objectives but is not required for rapid occupancy surveys solely intended to find new *B. affinis* locations.
    7. **Core. Record the data fields as noted in Table 2 for the Rapid Occupancy Protocol.**
        - **Core.** Record the **plant species that *B. affinis* was using,** if applicable. If you are unsure of the plant species or need help with plant identification, photograph the flower and diagnostic parts (*e.g.*, stem and leaves) to aid with later identification.
    8. **Stop** your timer when you have finished the sampling event.

- **Core.** Record the stop time and the survey duration.

e. **Contact the FWS Ecological Services Field Office** near you (https://www.fws.gov/locations) within 48 hours if you find and verify a *B. affinis* observation if it is observed outside of a High Potential Zone. Report all other observations, including non-detection data, via the required reporting form for permittees. If you do not hold a federal permit, by submit photographs to a science app such as www.iNaturalist.org or www.BumbleBeeWatch.org. Do not submit to multiple sites to avoid duplication of records.

f. Release

- Process and release *B. affinis* within 15 minutes of capture, if possible.
- Release all *Bombus* at site of capture, on or near flowers (refer to *Release* in **General Guidelines and Best Practices for Surveys**, above).

g. **Report** data on all surveys (positive detection of *B. affinis* and sampling events where the species was not detected) as required on state and federal permits.

- Federal *B. affinis* recovery permits require annual reports via submitting the **Rusty patched bumble bee reporting form** (available online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form) for annual permit reports.

2. Project Review Surveys

Surveys to confirm extant or uncertain sampling unit locations should have a standard level of effort at any given visit, plus repeated sampling during at least one year of the active flight season (refer to Figs. 1 and 2). Because the purpose of these surveys is to detect *B. affinis*, sampling may be biased towards finding that species, however a full *Bombus* species list is also requested. Surveys should be conducted within a year of project initiation, however locations can be surveyed for multiple years (e.g., if you want to be reasonably certain that the species no longer exists in the area, refer to definitions of an unoccupied zones and unoccupied sampling units, above). More information is given in **Appendix G –About Surveys for Proposed or Ongoing Actions that May Impact *B. affinis*.**

a. **Methods.** Use **occupancy** or **count methods** as described above.

b. **Effort.** Each observer should conduct a meandering survey walk for **30 minutes** within the 100m radius (3.14 ac) sampling unit. Record the effort of each observer separately.

- Conduct surveys during five-six evenly (approximately) spaced sampling periods during the sampling season (as defined in the **General Guidelines and Best Practices** section, above). For each sampling event, survey each suitable habitat patch for **30 minutes in the highest quality habitat** in your survey[4].
- Six, 30-minute surveys in July is sufficient for achieving a 0.95 cumulative detection probability of *B. affinis* at 3-ha (100m radius) sampling units in the Midwest (Otto et al. 2023). Five surveys are sufficient for achieving a 0.90 cumulative detection

[4] Refer to Appendix D.

probability of *B. affinis* at 3-ha (100m radius) sampling units in the Midwest (Otto et al. 2023). Therefore, we recommend five to six surveys to have demonstrated sufficient evidence that *B. affinis* is not present if not observed during those surveys.
    - Recognizing that five to six surveys during July may not be feasible and annual weather variation may affect detectability, it is reasonable that five to six 30-minute between mid-June and late August will provide high levels of confidence (i.e., 90-95% certain that the species is not there if not detected).
    - Surveys should be conducted within a year before the project initiation for negative survey results to remain valid for the duration of the project unless new information (e.g., new positive surveys) suggests that the species is likely to be present in the action area.
    - Although not required for project review, repeat sampling events in the same areas (if still suitable) for at least 3 consecutive years are advisable if you are trying to be reasonably certain that the species no longer occupies the area (refer to **Unoccupied** sampling unit in **Definitions** section, above).

c. **Conduct** the sampling event.
    1. **Start** your timer when you begin the sampling event.
        - **Core.** Record the start time.
    2. Search for bumble bees on flowers within the **highest quality foraging habitat** in your survey area. Spend minimal time in lesser quality habitat. Scan the areas for bumble bee activity and move towards those areas. Walk from flower patch to flower patch looking for active bumble bees and record detection and non-detection of *B. affinis*.
    3. **Core. Record the data fields as noted in Table 2 for the Project Review Protocol.**
    4. **Catch or photograph bumble bees**, preferably one at a time. Move bees from aerial nets to clear vials or plastic bags for easier identification.
        - **Core.** Photograph or capture (if handling, refer to Fig. 2) bees that match descriptions of *B. affinis.*
        - **Core.** Note **each caste** observed. Take representative photographs of each *B. affinis*. caste observed.
        - **Core. Note each species of *Bombus*** that is detected**.** Take representative photographs of each *Bombus* species detected.
        - Pause your timer while you are not actively searching for bees (*e.g.*, while you are putting bees in your cooler, walking between habitat patches, or taking photographs).
        - **Core.** Record the **plant species that *B. affinis* was using,** if applicable. If you are unsure of the plant species or need help with plant identification, photograph the flower and diagnostic parts (*e.g*., stem and leaves) to aid with later identification.
    5. **Stop** your timer when you have finished the sampling event.
        - **Core.** Record the stop time and the survey duration.

d. **Contact the FWS Ecological Services Field Office** near you (https://www.fws.gov/locations) within 48 hours if you find and verify a *B. affinis* observation if it is observed outside of a High Potential Zone. Report all other observations, including non-detection data, via the required reporting form for permittees. If you do not hold a federal permit, by submit

photographs to a science app such as www.iNaturalist.org or www.BumbleBeeWatch.org. Do not submit to multiple sites to avoid duplication of records.

e. Release
- Process and release *B. affinis* within 15 minutes of capture, if possible.
- Release all *Bombus* at site of capture, on or near flowers (refer to *Release* in **General Guidelines and Best Practices for Surveys**, above).

f. **Report** data on all surveys (positive detection of *B. affinis* and sampling events where the species was not detected) as required on state and federal permits.
- Federal *B. affinis* recovery permits require annual reports via submitting the **Rusty patched bumble bee reporting form** (available online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form) for annual permit reports.

3. Abundance

If you are interested in assessing the entire *Bombus* community at a location, you may choose the non-lethal **abundance** protocol (refer to Fig. 2).

a. **Methods.** Use **count methods** (see description above), which requires tallies of individuals of each species detected**.**

b. **Effort.** Each observer should conduct a meandering survey walk for **30 minutes** within the 100m radius (3.14 ac) sampling unit. Record the effort of each observer separately
- Conduct surveys during five-six evenly (approximately) spaced sampling periods during the sampling season (as defined in the **General Guidelines and Best Practices** section, above). For each sampling event, survey each suitable habitat patch for **30 minutes in the highest quality habitat** in your survey[5].
- Six, 30-minute surveys in July is sufficient for achieving a 0.95 cumulative detection probability of *B. affinis* at 3-ha (100m radius) sampling units in the Midwest (Otto et al. 2023). Five surveys are sufficient for achieving a 0.90 cumulative detection probability of *B. affinis* at 3-ha (100m radius) sampling units in the Midwest (Otto et al. 2023). Therefore, we recommend five to six surveys to have demonstrated sufficient evidence that *B. affinis* is not present if not observed during those surveys.
- Recognizing that five to six surveys during July may not be feasible and annual weather variation may affect detectability, it is reasonable that five to six 30-minute between mid-June and late August will provide high levels of confidence (i.e., 90-95% certain that the species is not there if not detected).

c. **Conduct** the sampling event.
1. **Start** your timer when you begin the sampling event.
   - **Core.** Record the start time.
2. Search for bumble bees on flowers within the **highest quality foraging habitat** in your survey area. Spend minimal time in lesser quality habitat. Scan the areas for bumble bee

[5] Refer to Appendix D.

activity and move towards those areas. Walk from flower patch to flower patch looking for active bumble bees and record detection and non-detection of *B. affinis*.

3. **Core. Record the data fields as noted in Table 2 for the Abundance Protocol.**
4. **Catch or photograph bumble bees**, preferably one at a time. Move bees from aerial nets to clear vials or plastic bags for easier identification.
    - **Core.** Photograph or capture (if handling, refer to Fig. 2) bees that match descriptions of *B. affinis.*
    - **Core.** Note **each caste** observed. Take representative photographs of each *B. affinis*. caste observed.
    - **Core. Count individuals of each species of *Bombus*** that is detected**.** Take representative photographs of each *Bombus* species detected.
    - Pause your timer while you are not actively searching for bees (*e.g.*, while you are putting bees in your cooler, walking between habitat patches, or taking photographs).
    - **Core.** Record the **plant species that *B. affinis* was using,** if applicable. If you are unsure of the plant species or need help with plant identification, photograph the flower and diagnostic parts (*e.g*., stem and leaves) to aid with later identification.
    - **Core.** Record the plant species that each species was using. Refer to the example data sheet in **Appendix E.**
5. **Stop** your timer when you have finished the sampling event.
    - **Core.** Record the stop time and the survey duration.

d. **Contact the FWS Ecological Services Field Office** near you (https://www.fws.gov/locations) within 48 hours if you find and verify a *B. affinis* observation if it is observed outside of a High Potential Zone. Report all other observations, including non-detection data, via the required reporting form for permittees. If you do not hold a federal permit, by submit photographs to a science app such as www.iNaturalist.org or www.BumbleBeeWatch.org. Do not submit to multiple sites to avoid duplication of records.

e. Release

- Process and release *B. affinis* within 15 minutes of capture, if possible.
- Release all *Bombus* at site of capture, on or near flowers (refer to *Release* in **General Guidelines and Best Practices for Surveys**, above).

f. **Report** data on all surveys (positive detection of *B. affinis* and sampling events where the species was not detected) as required on state and federal permits.

- Federal *B. affinis* recovery permits require annual reports via submitting the **Rusty patched bumble bee reporting form** (available online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form) for annual permit reports.

4. Long-term Monitoring

Long-term monitoring should be conducted in extant sampling units or any sampling unit where you are interested in 1) informing the *B. affinis* Recovery Plan targets (USFWS 2021) such as evaluating occupancy and trends, identifying healthy populations, and identifying population clusters; 2) informing population or habitat management or 3) improving scientific understanding, such as understanding the effects of drought on the species)(refer to Fig. 2). We recommend a standard level of effort which can be applied to any of the objectives above. Surveys should be conducted within a fixed sampling unit, multiple times per year, ideally for multiple years.

Background – long term recovery monitoring can be conducted to detect trends in known populations at **extant sampling units or at other sampling units at your discretion**. The quantity of bumble bees changes throughout the warm months as worker populations increase or decrease. For example, bees are subject to many environmental and health factors that impact the number of workers produced by each bumble bee colony. Additionally, over time, bumble bee habitat suitability changes as floral landscapes change composition. Because suitable habitat may change locations from one year to the next and bumble bee numbers fluctuate throughout the season, quantifying populations can be difficult, and those variabilities should be accounted for in your monitoring effort.

a. **Methods.** Use **occupancy** or **count methods** as described above.
b. **Effort.** Each observer should conduct a meandering survey walk for **30 minutes** within the 100m radius (3.14 ac) sampling unit. Record the effort of each observer separately.
    - Conduct surveys during five-six evenly (approximately) spaced sampling periods during the sampling season (as defined in the **General Guidelines and Best Practices** section, above). For each sampling event, survey each suitable habitat patch for **30 minutes in the highest quality habitat** in your survey[6].
    - Six, 30-minute surveys in July is sufficient for achieving a 0.95 cumulative detection probability of *B. affinis* at 3-ha (100m radius) sampling units in the Midwest (Otto et al. 2023). Five surveys are sufficient for achieving a 0.90 cumulative detection probability of *B. affinis* at 3-ha (100m radius) sampling units in the Midwest (Otto et al. 2023). Therefore, we recommend five to six surveys to have demonstrated sufficient evidence that *B. affinis* is not present if not observed during those surveys.
    - Recognizing that five to six surveys during July may not be feasible and annual weather variation may affect detectability, it is reasonable that five to six 30-minute between mid-June and late August will provide high levels of confidence (i.e., 90-95% certain that the species is not there if not detected).
    - **Repeat sampling events in the same area** (i.e. using the same center point and moving to the highest-quality foraging area within the fixed area) for a minimum of 3 consecutive years, ideally much longer (i.e., decades).
c. **Conduct** the sampling event.

[6] Refer to Appendix D.

- **Start** your timer when you begin the sampling event.
    - **Core.** Record the start time.

1. Search for bumble bees on flowers within the **highest quality foraging habitat** in your survey area. Spend minimal time in lesser quality habitat. Scan the areas for bumble bee activity and move towards those areas. Walk from flower patch to flower patch looking for active bumble bees and record detection and non-detection of *B. affinis*.
2. **Core. Record the data fields as noted in Table 2 for the Long-Term Monitoring Protocol.**
3. **Catch or photograph bumble bees**, preferably one at a time. Move bees from aerial nets to clear vials or plastic bags for easier identification.
    - **Core.** Photograph or capture (if handling, refer to Fig. 2) bees that match descriptions of *B. affinis.*
    - **Core.** Note **each caste** observed. Take representative photographs of each *B. affinis*. caste observed.
    - **Optional** for long-term monitoring**. Count individuals of each species of *Bombus*** that is detected**.** Take representative photographs of each *Bombus* species detected.
    - Pause your timer while you are not actively searching for bees (*e.g.*, while you are putting bees in your cooler, walking between habitat patches, or taking photographs).
    - **Core.** Record the **plant species that *B. affinis* was using,** if applicable. If you are unsure of the plant species or need help with plant identification, photograph the flower and diagnostic parts (*e.g*., stem and leaves) to aid with later identification.
    - **Optional.** Record the plant species that each species was using. Refer to the example data sheet in **Appendix E.**
4. **Stop** your timer when you have finished the sampling event.
    - **Core.** Record the stop time and the survey duration.

d. **Contact the FWS Ecological Services Field Office** near you (https://www.fws.gov/locations) within 48 hours if you find and verify a *B. affinis* observation if it is observed outside of a High Potential Zone. Report all other observations, including non-detection data, via the required reporting form for permittees. If you do not hold a federal permit, by submit photographs to a science app such as www.iNaturalist.org or www.BumbleBeeWatch.org. Do not submit to multiple sites to avoid duplication of records.

e. Release

- Process and release *B. affinis* within 15 minutes of capture, if possible.
- Release all *Bombus* at site of capture, on or near flowers (refer to *Release* in **General Guidelines and Best Practices for Surveys**, above).

f. **Report** data on all surveys (positive detection of *B. affinis* and sampling events where the species was not detected) as required on state and federal permits.

- Federal *B. affinis* recovery permits require annual reports via submitting the **Rusty patched bumble bee reporting form** (available online at

https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form) for annual permit reports.

Table 2. Data fields for the *B. affinis* surveys and monitoring protocols. Each of the core or project-specific data fields should be collected and recorded during each 30-min survey. Adapted from Otto et al. (2024) and (2024*b*).

Data Field	Rapid	Project Review	Abundance	Long-Term Occupancy
Sampling unit location (Lat/Long)	Core	Core	Core	Core
Sampling unit name	Core	Core	Core	Core
Survey date	Core	Core	Core	Core
Detection/Non-Detection of *B. affinis*	Core	Core	Core	Core
Field observer name	Core	Core	Core	Core
Permit number (if applicable)	Core	Core	Core	Core
Start time	Core	Core	Core	Core
End time	Core	Core	Core	Core
Survey duration	Core	Core	Core	Core
Area of sampling unit	Core	Core	Core	Core
B. affinis caste (worker, male, queen, unknown)	Core	Core	Core	Core
B. affinis floral host plant(s)	Core	Core	Core	Core
Time of *B. affinis* detection(s)	Core	Core	Core	Core
Percent of sample unit with flowering resources in bloom[7]	Core	Core	Core	Core
Dominant habitat type in sample unit (*e.g.*, prairie, developed, forest)	Core	Core	Core	Core
Sample unit stressors (*e.g.*, pesticide treatments, honeybee hives present)	Core	Core	Core	Core
Sample unit management (*e.g.*, restored prairie)	Core	Core	Core	Core
Other *Bombus* species detections	Recommended	Core	Recommended	Recommended
Other *Bombus* species floral host(s)	Recommended	Recommended	Recommended	Recommended
Survey-specific weather (wind, temperature, humidity, cloud cover, air quality index)	Recommended	Recommended	Recommended	Recommended

[7] Refer to Appendix D for more details.

Protocol Suggestions for Unoccupied Zones

*Here, we recommend established protocols for bumble bee surveys and inventories in areas within the **Unoccupied Zones**, within the historical county range, or beyond the boundaries where the species was thought to occur. Choose the methods and protocols based on the objectives of the study (note that the protocols you use are not necessarily limited to those listed here). The protocols discussed above may also be conducted outside of the Unoccupied Zones, if you desire.*

Passive Methods and Protocols

- Passive lethal sampling is not authorized within High Potential Zones or Low Potential Zones.
- For community-level bee data in unoccupied zones, consider using the standardized protocol by Levenson et al (2024*a*). Refer to the USFWS National Protocol Framework for the Inventory and Monitoring of Bees (Maffei et al. 2025) for more details if surveying on USFWS National Wildlife Refuges.
- Passive methods to sample the bee community are described in Levenson et al. (2024*a*).
- Refer to the USFWS National Protocol Framework for the Inventory and Monitoring of Bees (Maffei et al. 2025) for more details if surveying on USFWS National Wildlife Refuges.

Sampling unit Selection – If handling bumble bees, sampling units should be within the **Unoccupied Zones** (refer to High Potential Zone map, https://www.fws.gov/species/rusty-patched-bumble-bee-bombus-affinis), updated annually).

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Appendix A – Standardized Bee Photography

- Type of camera - Digital cameras, DLSR and phones with cameras (>= 10megapixels) will be the best options for use after aerial netting and moving the bee into a clear vial or clear baggie (zip net) to take photos.
- To properly identify a bumble bee, it is best to take photos that clearly show the entire top side of the abdomen, the side of the thorax/abdomen and the face/head. Take several photos of each specimen to show these various characteristics.
- Hair color patterns vary with lighting. Examine photos to ensure that coloration is clear and that shadows and the underlying integument are not creating deceptive color patterns.
- If you are not handling bees, photograph them as they forage, taking a series of photos of each individual to document the bee's characteristics clearly, as described above. Take a "spacer" photo between the series, so you can distinguish between individual bees when you archive photos and report findings.
- If you are handling bees, vials with a flat side may provide higher quality photos. Placing the bee in a cooler will slow them down making them easier to photograph. You need a scientific recovery permit to handle *B. affinis* within High and Low Potential zones (refer to Fig. 1 and Fig. 2).
- Do not hold a bee in a container for longer than a few minutes, unless you place it in a cooler with ice. Bumble bees can easily overheat, so do not keep vials/bags in direct sunlight. Do not keep bees in a cooler longer than two hours, and not directly on the ice.
- If using a cooler with ice, bees may take a few minutes to warm up before flying away. Place vials or bags in a shaded area to give the bees time to fly away.
- Archive photographs for long-term storage.
- Link each photograph to the corresponding specimen. Use standardized naming conventions that provide the following information:
    - Permit number
    - Unique sampling unit location name
    - Specimen number
    - Photograph number
    - Date
    - For example, label PERMITNUMBER_SAMPLE UNIT_SPECIMEN#_PHOTO#_DATE (*e.g.*, TE5555-7_STPAUL28_Specimen12_photo2_12July2025)

Appendix B – Bumble Bee Identification

This section lists basic key diagnostics for field identification. These should be documented clearly when using photography survey protocols. For detailed and technical descriptions, refer to **Bumble Bees of North America: An Identification Guide** (Williams et al. 2014); and Discover Life (https://wwww.discoverlife.org/mp/20q). Other helpful resources include, but are not limited to: (https://www.xerces.org/bumble-bees/identification), (https://ncipmhort.cfans.umn.edu/sites/ncipmhort.cfans.umn.edu/files/2020-05/guide%20to%20bumble%20bees%20lab.pdf), and (https://www.flickr.com/photos/usgsbiml/albums/72157664851159091/).

Bombus affinis gynes and queens are entirely yellow on the first two abdominal segments; the rest of the abdominal segments are black (Fig. B.1). In workers and males, the first abdominal segment is yellow, and the second has a medial patch of rusty hairs on the anterior portion of the segment, with yellow hairs on the posterior portion (Fig. B.1). *B. affinis* can vary from pale tan to a bright rusty brown. The other abdominal segments are black. Occasionally, abdominal segments 3-6 may have reddish hairs or a reddish patch.

B. affinis has a mostly yellow upper thorax with a black spot or band that may extend toward the posterior in a v-shape. The bottom of the thorax is black. Queens and workers have black hairs on the head and at the back of the head. Males have black hairs on the head, with some yellow hairs intermixed at the back of the head. Overall, hairs are moderately long and even. *B. affinis* is a short-tongued species and sometimes is observed nectar-robbing on tubular flowers. It has a short face, with cheek (oculo-malar area) slightly shorter than broad. *B. affinis* can be confused with *B. citrinus, B. griseocollis* (Fig. B.3), *B. perplexus,* and *B. vagans*.

B. affinis phenology in each region will determine when queens (or gynes), workers, or males will be observed in flight. For example, in southern Wisconsin, *B. affinis* gynes and queens, distinguished by larger size and other characteristics described above, are in flight in spring (roughly mid March - May) and then again in late summer and fall. Workers can be seen in the field several weeks after nest establishment, throughout the summer, into early fall (late June-September). Males are typically in flight during late summer and fall (August-September).

More difficult to observe, nesting occurs mid - March, April or May through September, and gynes overwinter from August until mid- March, April or May. If you happen to observe activity of *B. affinis* gynes and queens, take notes regarding behavior, habitat use, etc., as nesting and overwintering are little known. Notify USFWS as soon as reasonably possible.

Figure B.1. Illustrations of a *B. affinis* queen (left), worker (center), and male (right) by Elaine Evans, University of Minnesota.

Figure B.2. Photographs of *B. affinis* (a) top view, (b) side view and (c) side/top view. Photographs were taken by Dan Mullen (Creative Commons) (a,b) and USGS Bee Monitoring Laboratory (c).

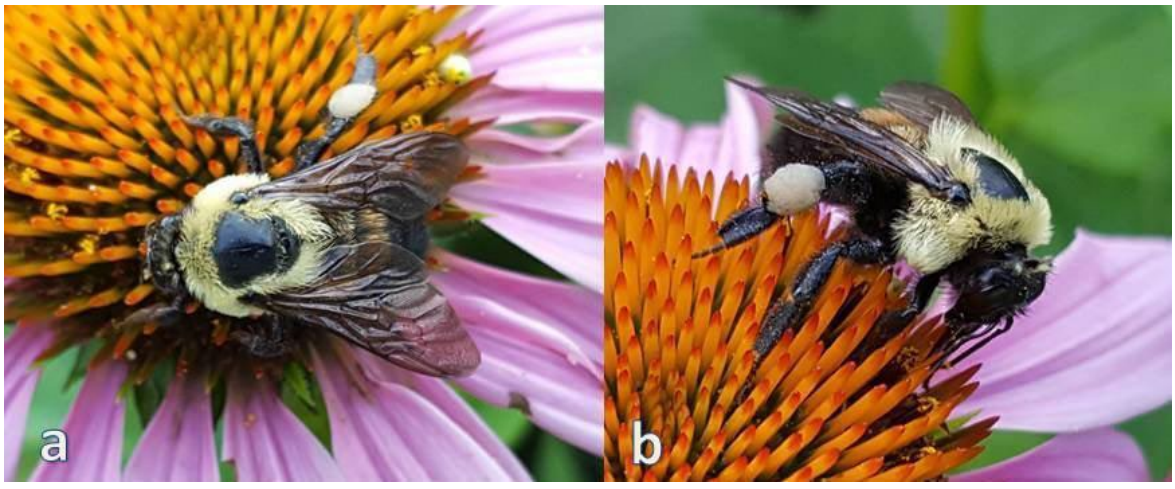


Figure B.3. Photographs of *B. griseocollis* (a) top view and (b) side view, which can be confused with *B. affinis*. Photographs were taken by Tamara Smith, USFWS.

Appendix C – Observation Verification

- **Appendix B – Bee Identification** (above) provides useful information to help with identification. Photographic documentation of *B. affinis* is required to verify the record.
- Non-permitted individuals to submit their photographs to a science app such as www.iNaturalist.org or www.BumbleBeeWatch.org, where independent taxonomists may assist with species identifications.
- Verification requests may be submitted to the USFWS Ecological Services Field Office (FO) in your state. An online directory of USFWS FO(s) is available online at https://www.fws.gov/locations. Documentation should include geographic coordinates (decimal degrees). Photographs should meet the standards in **Appendix A - Standardized Bee Photography**.
- If assistance with identifications are requested of the USFWS, the surveyor acknowledges that data may be shared with third parties in order to accomplish this. The USFWS may submit by email the images and county level locations to a qualified expert for identification assistance, or if project information or locations are non-sensitive, the USFWS may submit the necessary information (including photos and locations) to other qualified experts (*e.g.*, taxonomic experts from www.BumbleBeeWatch.org). Once species identification has been made, the surveyor will be informed of the results by USFWS. Adjustments to this approach may be subject to change if, for example, these requests require a significant amount of time and resources.

Appendix D – *Bombus affinis* Foraging Habitat

Bombus affinis has been observed in a variety of habitats, including prairies, oak savanna, forests, prairies, parks, and residential areas (Colla and Packer 2008, p. 2008, Colla and Dumesh 2010, p. 46) *B. affinis* needs areas that contain sufficient food (nectar and pollen from diverse and abundant flowers), nesting sites that are predominantly free from ground-disturbing activities and near floral resources, and overwintering sites for hibernating queens (Potts et al. 2010, p. 349, Goulson et al. 2015, p. 2). It is a generalist forager for pollen and nectar like other bumble bees (Jepsen et al. 2013, pp. 27-28), but relies on diverse and abundant flowering plant species (Potts et al. 2010, p. 349, Goulson et al. 2015, p. 2). Due to the early emergence of *B. affinis* (roughly, mid-March through April), forested areas that support early blooming spring ephemerals are likely important habitats (Colla and Dumesh 2010, pp. 45-46), especially when they are near open areas that are also used for summer foraging. *B. affinis* nests are typically in abandoned rodent nests or other similar cavities, one to four feet below ground (Plath 1922, pp. 190-191, Macfarlane et al. 1994, p. 4). *B. affinis* nests have also been occasionally observed above ground (Plath 1922, p. 190). Little is known about the overwintering habitats of *B. affinis* foundress queens (refer to Herrick and Carpenter 2025), but other species of *Bombus* typically form a chamber in soft soil, a few centimeters deep (often in forests or in forest edges) and sometimes use compost or mole hills to overwinter (Goulson 2010, p. 11). Overwintering (roughly, mid-October through mid-March) sites may typically be in loose, uncompacted and often sandy, moss-covered soils on northwest exposures (E. Evans, University of Minnesota, pers. comm. 2017).

While *B. affinis* forage on early spring floral blooms within forests and forest edge, also use open natural areas (e.g., prairies, oak savannah) with flowering resources during their active flight season. Consequently, potentially suitable foraging habitat can be defined as any open, vegetated habitat within your survey region, such as prairies, roadsides, and meadows (Figs. D.1, D. 3 and D.4). Open habitats with high floral diversity and abundance should be considered "high quality" habitat (Figs. D.1, D. 3 and D.4). Areas dominated by grasses or sedges with few flowering plants or are considered low quality.

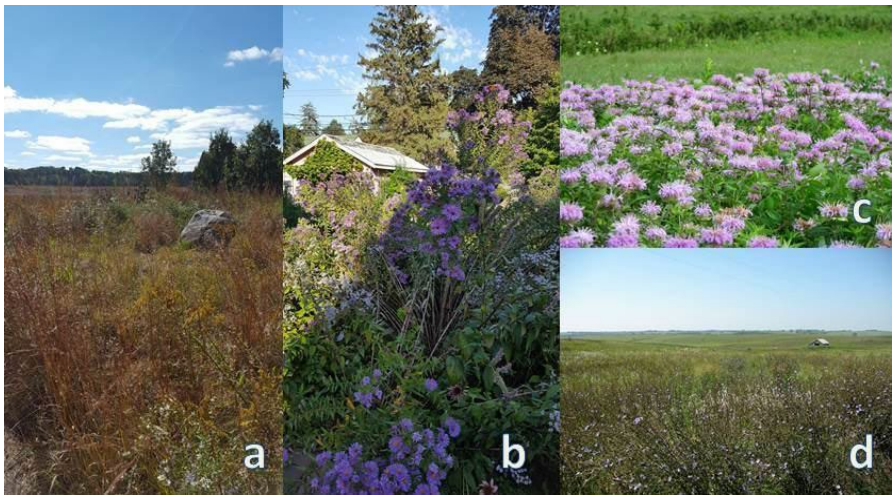
Habitat can be characterized in part by percent of vegetative cover that is forbs, flowering shrubs, or pollinator-friendly trees; especially areas that bloom all season long. Estimates of the number of species of forbs, flowering shrubs, or pollinator-friendly trees in bloom during each survey can help define "high quality" habitat and can help surveyors focus their effort (refer to **Rusty patched bumble bee reporting forms** available online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form).

Surveyors should spend less time in poor and/or questionable habitat. Questionable habitat includes areas that are not clearly poor habitat and quickly checked for bumble bee activity (Figs. D2.and D2.b) before moving on to better habitat or deciding not to survey an area. Poor foraging habitat can be defined as areas without an abundance of floral resources, areas with compacted

soils, sod-forming grasses, or large monoculture agricultural fields (although some flowering agricultural crops, like alfalfa, may provide temporary foraging habitat). Some examples of poor foraging habitat include open water, regularly maintained turfgrass monocultures, and pavement.

Special note on urban areas: Some of the last refuges for *B. affinis* appear to be in large urban areas, such as Minneapolis/St. Paul, Madison, Milwaukee and Chicago. From a landscape perspective, these cities have a network of natural areas that include parks, greenways, public gardens and other public or undeveloped lands. Interspersed among these natural areas are residential areas - - yards, gardens and boulevards that provide additional sources of flowering plants (Fig. Dd2.b). Areas considered high quality habitat in urban areas have the same characteristics as high-quality habitat outside of urban areas. They are generally open areas with an abundance and diversity of plants that flower from mid-March through mid-October; that have undisturbed areas without landscaping mulch or landscape fabric; and that are managed with minimal use of pesticides; particularly insecticides and fungicides.

In the urban landscape, high quality foraging habitat is most likely in or near natural areas that support open, or mostly open, habitats such as prairie, savannas, grasslands, or grassland/shrub mix (Fig. D1.a). Forested lands can also provide foraging habitat and may provide important early spring habitat if they support spring ephemerals or early spring blooming trees and shrubs. Natural areas within urban areas may be in blocks (small or large) or may be linear. The value of any of these tracts is higher if surrounding areas also provide suitable habitat for nesting and/or overwintering, and flowering native plants.



Figures D1 a - d. Examples of high-quality foraging habitat for *B. affinis*; (a) native prairie habitat within an urban landscape, (b) large patch of diverse floral resources within an urban landscape, (c) large patch of wild bergamot, and (d) open meadow with an abundance of floral resources. Photographs were taken by T. Smith, USFWS (a, b) and Rob Jean, ESI (c, d).

Figures D2 a - b. Examples of low-quality foraging habitat for *B. affinis*; (a) area that appears dominated by sod grass (b) predominantly grassy area with little or no floral resources. These areas should be checked briefly for bumble bee activity during the flight season. If no activity is observed, we would not recommend conducting surveys in these areas. Photographs were taken by Rob Jean, ESI.

Appendix E – Sample Field Sheets

Data forms for permittees can be found online at https://www.fws.gov/media/rusty-patched-bumble-bee-reporting-form. These example field sheets were developed by USGS Fort Collins Science Center.

Occupancy and Abudance Survey Datasheet (v4)						
Date (mm/dd/yyyy)	Site Name	Latitude (6-digit, decimal degree)		Longitude (6-digit, decimal		
Survey Type (occupancy, abundance, partial)			Survey Duration (min)			
Observer Name		Start Time	End Time	Survey Distance (m)		
% Flowering of Site						
Rusty Patched Bumble Bee info						
		Count	Voucher IDs	GPS	Notes	
B. affinis	W					
	M					
	Q					
	U					
All other Bumble Bee info						
BB species	Count	Voucher ID	BB species	Count	Voucher ID	
B. impatiens			B. vagans			
B. griseocollis			B. sandersoni			
B. bimaculatus			B. citrinus			
B. auricomus			B. insularis			
B. pensylvanicus			B. perplexus			
B. fervidus			B. terricola			
B. rufocinctus			Other:_____			
Evidence of Disturbance (describe)						

Floral Resources (Species: Abundance Category [single, sparse, occasional, frequent, very common])			
Plant Species	Abund. Cat.	BB presence	RPBB presence
Survey Notes:			

Site Description Datasheet (v1)			
Date Assessed (mm/dd/yyyy)	Site Name	Latitude (6-digit, decimal degree)	Longitude (6-digit, decimal degree)
Does site meet minimum requirements? (Circle One) Yes / No / Unsure			
Site Permissions (Describe)			
Access Issues (Describe)			
Habitat Type (circle any that apply)			
meadow, wetland, urban, lawn, forest, forest clearing, savannah, roadside, orchard, cropland, other_____			
Site Management (if known or suspected - circle any that apply)			
grazing, mowing, burning, prairie restoration, construction of roads or buildings, timber harvest or clearing, other _____			
Site History (describe if known)			
Site Map (Draw or specify image)			
Other Notes			

Appendix F – Core: Percentage of Foraging Habitat

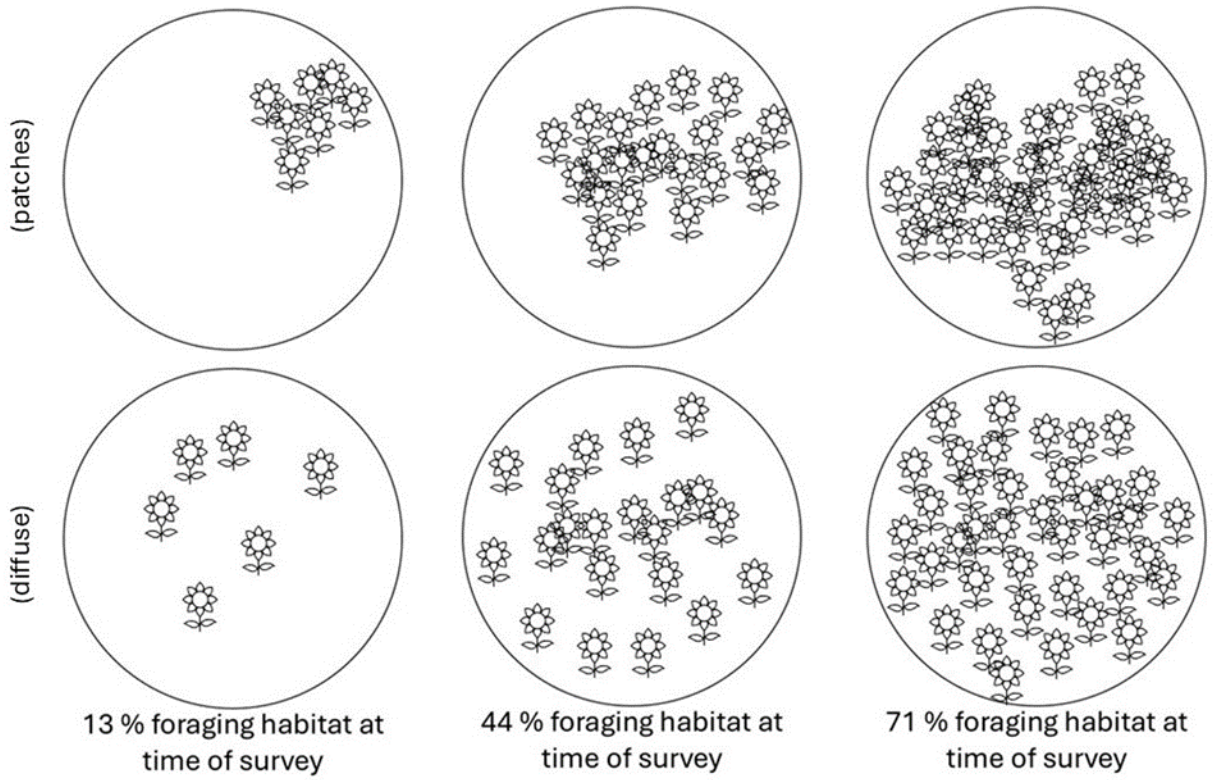
Developed by the *B. affinis* Long Term Monitoring Habitat Working Group

Objective of the core module habitat question: Understand how RPBB occupancy dynamics and detection probability are related to changes in floral habitat and use of a sampling unit (i.e., 3.14ha circle) within a 10 x 10 km grid cell.

Question for field form: Of the area you surveyed, what percent represents bumble bee foraging habitat? *Both native and non-native flowering resources actively blooming at time of survey within the boundary of the sampling unit should be considered in your assessment.*

Data field type: numeric, value between 0-100

Guidance to include in survey protocol for this question: Give your best estimate of the % of flowering resources both native and introduced species that are actively blooming at the time of your survey within the boundary of your sampling unit. Typically, this will be a 100-m radius circle around your center point (i.e. 3.14 ha sampling unit). A helpful way to visualize this is to think if you had a bird's eye view of your 100m radius circle, what % of that area is covered with actively flowering plants?



Note that you can select any integer value between 0 and 100 for your percentage estimate. You do not need to bin estimates as 25%, 50%, or 75%, nor should you spend a lot of time determining incrementally small differences such as what constitutes 13% vs 14% foraging habitat – it is a coarse estimate. Some flowering species may show patched distribution, occurring at high density within a patch, while other species may be spread more diffusely throughout the site, and most sites will have a combination – just remember to try to aim for estimating the overall footprint of actively flowering plants.

Important – do not increase the percentage of foraging habitat just because you detected a rusty-patched bumble bee during the survey or observed a lot of bumble bee activity. Weather and other factors can affect bumble bee detection, independent of habitat quality. Our habitat quality metric is meant to provide a rapid and coarse estimate of floral resource availability during each survey. This information will be useful for understanding how changes in floral resources affect RPBB detection and occupancy.

Do not overthink this – give your best estimate and try to remain internally consistent in how you estimate this across different sampling units and surveys – do not worry if another monitor has a different estimate, and do not worry about matching your estimate to that of other monitors – observer variation is normal and factored into modelling of survey data

Why is the habitat measure so simple? Does it collect any useful data?

It does - it can capture the effects of phenological shifts in flora and management actions on floral availability. Changes in the percentage of floral resources estimated by the observer is correlated with bumble bee foraging activity and the likelihood that bumble bees are detected during surveys. It is also allows more sampling units to be monitored – some monitors or organizations may have limited time, limited plant identification skills, or already have in-house plant protocols that they do not want to change, so this level of simplicity ensures that sampling units that might not otherwise be monitored are included in regional-scale survey data tracking RPBB vital rates such as persistence, colonization, and extirpation, *allowing for greater precision in measuring how those vital rates are affected by habitat changes through time.*

Organizations that have the interest and capacity to include additional recommended habitat modules to include more detailed data are very welcome to, but we ask all monitors to record the habitat percentage defined here as the baseline measurement of habitat.

Appendix G - About Surveys for Proposed or Ongoing Actions that May Impact *B. affinis*

As a reminder, for those interested in seeking incidental take coverage through Section 7 or Section 10(a)(1)(B) of the Endangered Species Act, it is important to first review the Section 7 technical assistance document (https://www.fws.gov/media/esa-section-7a2-voluntary-implementation-technical-assistance-rusty-patched-bumble-bee). The Service considers the rusty patched bumble bee likely to be present only in "**high potential" zones**" as described in the Section 7 (https://www.fws.gov/media/esa-section-7a2-voluntary-implementation-technical-assistance-rusty-patched-bumble-bee).

To determine if *B. affinis* may be in an action area, action agencies or project proponents may screen projects online via the USFWS's Information for Planning and Conservation website (IPaC, https://ipac.ecosphere.fws.gov/user/login), review the *B. affinis* map (available online https://www.arcgis.com/home/webmap/viewer.html?webmap=2716d871f88042a2a56b8001a1f1acae&extent=-100.6667%2c29.7389%2c-48.8551%2c50.9676)), or contact the USFWS Ecological Services Field Office for their area. If this screening indicates that a project is within an area where the rusty patched bumble bee is likely present (*i.e.*, the High Potential Zone), action agencies and others should coordinate with their state's USFWS Ecological Services Field Office (FO). An online directory of USFWS FO(s) is available online at https://www.fws.gov/locations.

If a project action area is within an area where *B. affinis* is likely present ("High Potential Zones" or HPZs), refer to HPZ map online at https://www.fws.gov/species/rusty-patched-bumble-bee-bombus-affinis), an action agency or project proponent may choose to conduct a survey to verify presence (note that negative surveys cannot verify absence with 100% confidence, but can at best predict likelihood of occupancy). The results of a survey, if they are negative and are carried out in accordance with FWS-recommended survey protocol (*i.e.*, Project Review Protocol), would indicate that the species would not be exposed to stressors associated with the action area. Project Review Protocol recommends one sampling season of surveys with sufficient effort[8] to support a determination that the species is not likely present in the portion of the action area surveyed. Although not required for project review, repeat sampling events in the same areas (if still suitable) for at least 3 consecutive years are advisable if you are trying to be reasonably certain that the species no longer occupies the area.

[8] Sufficient effort (to get a 95% probably that the species is not there) would consist of six approximately equally spaced sampling periods during the sampling season (mid-June to late-August); one-person hour of search time per three acres of suitable high-quality habitat (defined below) using non-lethal netting techniques. This document provides further details on methods, techniques, and best practices and is subject to continual improvement and modification.

Note that surveys should be conducted within a year before the project is initiated for negative survey results to remain valid throughout the duration of the project unless new information (*e.g.*, new positive surveys) suggests that the species is likely to be present in the action area. In that case, action agencies and the FWS field office (https://www.fws.gov/locations) should work together to ensure that the best available information is considered.

If an action agency or project proponent chooses to conduct a survey to verify presence or likely absence, we recommend that they and their surveyors develop a proposed survey strategy in coordination with the USFWS FO(s) so that all parties fully understand which methods will be deployed, what assumptions will be made, and what the various outcomes would be based on the survey results. If not already required by federal permit, we recommend that survey results are submitted (negative or positive) to the USFWS FO(s) in the state(s) where the surveys took place. We strongly encourage this coordination as it improves the USFWS' understanding of (1) the level of survey effort underway and (2) the distribution of the species.