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Introduction

This document outlines the Service's survey requirements under a section 10(a)(1)(A) permit for conducting presence/absence surveys for federally listed endangered, terrestrial karst invertebrate species (herein referred to as "karst invertebrates") in Travis, Williamson, and Bexar Counties, Texas (Table 1). Section 10(a)(1)(A) permits, also referred to as recovery, enhancement of survival, or scientific permits, allow for "take" of listed species that may or will occur while conducting activities to further the recovery of a listed species (see *When a Section* 10(a)(1)(A) *Scientific Permit is Needed*) below. In addition to describing survey requirements, this document includes:

- information to be included in final reports, which includes positive and negative karst invertebrate survey results;
- information to be included in recovery project proposals (such as, monitoring plans for locations known to be occupied by these species), which must be approved in writing by the Austin Ecological Services Office; and
- minimum qualifications for personnel conducting presence/absence surveys for endangered karst invertebrates under a section 10(a)(1)(A) permit.

This document supersedes any previous guidance from the Austin Ecological Services Office on conducting presence/absence surveys for these species. Information that relates to the effectiveness of these survey guidelines in conserving endangered karst invertebrates is welcome. As additional information becomes available regarding survey techniques and effectiveness, these guidelines may be updated.

A karst invertebrate survey, when conducted according to these requirements is an attempt to determine presence or probable absence of these species in locations where the species have not been previously located. It does not provide sufficient data to determine population size or structure nor to determine absence from locations where endangered species have been previously found. Following these requirements will standardize survey procedures. It will help maximize the potential for detection of karst invertebrates at previously undocumented sites at a minimum acceptable level of certainty. Although the detection of karst invertebrates confirms their presence, failure to detect them does not absolutely confirm their absence. For example, during in-cave monitoring from 2002 to 2013 at one karst invertebrate preserve, *Rhadine exilis* was observed in 2007 only (SWCA 2014). Also, in-cave monitoring at another preserve from 2002 to 2013 found that *Rhadine infernalis* was observed from 2002 through 2005 and then not again until 2012 (SWCA 2014). This exemplifies how even multiple years of sampling without observing a species does not mean the species is extirpated.

When a Section 10(a)(1)(A) Scientific Permit is Needed

Activities that are being conducted to benefit endangered species but would also result in "take" are prohibited under section 9 of the Endangered Species Act of 1973, as amended, unless the "take" is covered under a section 10(a)(1)(A) scientific permit. "Take" is defined as "to harass, harm, pursue, hunt, shoot, wound, kill, trap, capture, or collect, or to attempt to engage in any such conduct." Activities that could result in take and that could occur while conducting karst invertebrate surveys include:

- collecting individuals;
- crushing individuals inadvertently;
- compacting habitat and oviposition sites;
- destroying webs;
- disturbing cover objects;
- excavating caves, which could result in inadvertent removal of individuals or removal or alteration of habitat;
- harming or harassing individuals by introducing noise, light, chemicals, or biological substances (such as microbes) that are normally found on the surface or in other caves; and
- possibly other actions that would cause individuals to flee, seek shelter, or alter or ceasenormal feeding, breeding, and sheltering behavior.

Since one of the first steps in determining presence/absence of endangered karst invertebrates is to survey for karst features that may have suitable habitat, this document also outlines the Service's recommendations for conducting surveys for these features. Since no "take" of endangered species is anticipated while conducting initial surface walking karst feature surveys, this activity does not necessitate a section 10(a)(1)(A) permit. However, the potential for "take" exists with entry into a void or cave where endangered karst invertebrates may occur. Therefore, the Service recommends that all activity that is being conducted to benefit the species and that involves excavating, entering, or collecting in a void or cave that may contain suitable habitat for these species be covered under a valid 10(a)(1)(A) permit for the endangered karst invertebrates in the county being surveyed. If personnel are conducting excavation only, then they can receive coverage by being supervised by someone with a 10(a)(1)(A) permit, and the permittee should also be onsite to collect specimens discovered during excavation.

For information on how to apply for a 10(a)(1)(A) permit contact permitsR2ES@fws.gov or go to our permits webpage at: https://fwsepermits.servicenowservices.com/fws.

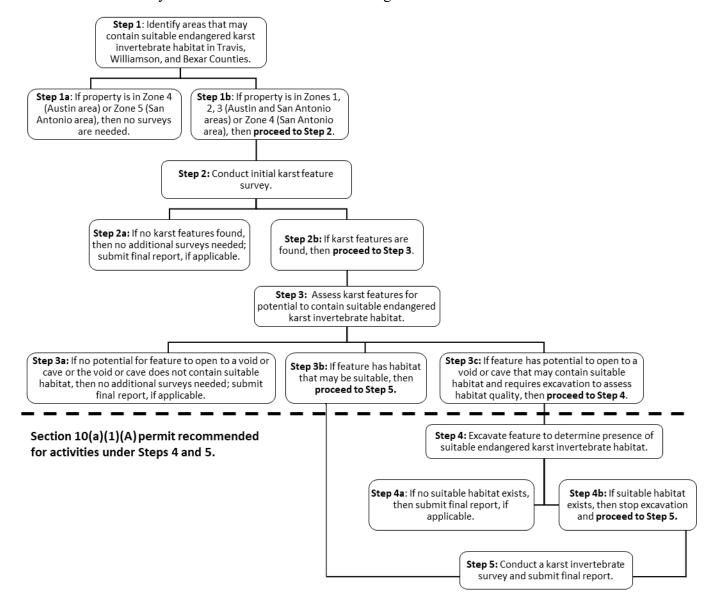
Table 1. Federally endangered terrestrial karst invertebrates from central Texas (Final Rule for Travis and Williamson Counties - 53 FR 36029-36033; Bexar County - 65 FR 81419-81433; technical corrections - 58 FR 43818-43820).

Common Name	Species	County of Occurrence
Madla Cave meshweaver	Cicurina madla	Bexar
Robber Baron Cave meshweaver	Cicurina baronia	Bexar
Braken Bat Cave meshweaver	Cicurina venii	Bexar
Government Canyon Bat Cave meshweaver	Cicurina vespera	Bexar
Government Canyon Bat Cave spider	Tayshaneta microps	Bexar
Cokendolpher cave harvestmen	Texella cokendolpheri	Bexar
Ground Beetle (no common name)	Rhadine exilis	Bexar
Ground Beetle (no common name)	Rhadine infernalis	Bexar
Helotes mold beetle	Batrisodes venyivi	Bexar
Bee Creek Cave harvestmen	Texella reddelli	Travis
Kretschmarr Cave mold beetle	Texamaurops reddelli	Travis
Tooth Cave pseudoscorpion	Tartarocreagris texana	Travis
Tooth Cave spider	Tayshaneta myopica	Travis and Williamson
Tooth Cave ground beetle	Rhadine persephone	Travis and Williamson
Bone Cave harvestmen	Texella reyesi	Travis and Williamson
Coffin Cave mold beetle	Batrisodes texanus	Williamson

How to Determine if Karst Invertebrates May be Present

Figure 1 outlines a five-step approach for determining presence/absence of endangered karst invertebrates and karst features that may contain suitable habitat for endangered karst invertebrates in central Texas. This should be used in conjunction with the text following this figure for a more complete description of each step.

Figure 1. Five-step approach for determining presence/absence of endangered karst invertebrates and karst features that may contain suitable habitat for endangered karst invertebrates in central Texas.



Step 1. Identify areas that may contain suitable habitat for endangered karst invertebrates in Travis, Williamson, and Bexar Counties

Four karst zones have been delineated in the Austin area (Travis and Williamson Counties) (Veni 1992) and five karst zones have been delineated in the San Antonio area (Bexar County) in Texas (Veni 1994). The karst zones in the San Antonio area were updated and revised in Veni (2002). These karst zones are a useful first step in determining if karst features containing endangered invertebrates are likely to occur on a property. The karst zone maps are available online at https://www.fws.gov/office/austinecological-services or upon request from the Austin Ecological Service Field Office.

Table 2. Definitions of Karst Zones (modified from Veni 1992; 1994; 2002)

Zone	Austin Area	San Antonio Area
1	Areas known to contain endangered karst invertebrate species	Areas known to contain endangered karst invertebrate species
2	Areas having a high probability of containing suitable habitat for endangered karst invertebrate species	Areas having a high probability of containing suitable habitat for endangered karst invertebrate species
3	Areas that probably do not contain endangered karst invertebrate species	Areas that probably do not contain endangered karst invertebrate species
4	Areas, both cavernous and non- cavernous, that do not contain endangered karst invertebrate species	Areas that require further research but are generally equivalent to Zone 3, although they may include sections that could be classified as Zone 2 or Zone 5 as more information becomes available
5	Not applicable	Areas, both cavernous and non-cavernous, that do not contain endangered karst invertebrate species

Step 1a. If the subject property is in Zone 4 (Austin area) or Zone 5 (San Antonio area), then it lies within an area, either cavernous or non-cavernous, that does not contain the endangered karst invertebrates, and no surveys are needed. Note, that the karst zones could be revised in the future based on survey data and site-specific mapping.

Step 1b. When conducting a karst invertebrate survey under a section 10(a)(1)(A) permit, the surface survey for karst features is not expected to result in "take." However, while not required under a 10(a)(1)(A) permit, certain procedures for surface surveys are recommended as part of the scientifically sound process for assessing the presence/absence of karst invertebrates. In karst zones 1,2, 3 (Austin

and San Antonio areas), and 4 (San Antonio area), we recommend an initial karst feature survey be conducted on the entire property within these zones; **proceed to Step 2** for more on conducting these surveys.

Step 2. Conduct an initial karst feature survey

If you are in zones 1, 2, or 3 in Austin or San Antonio or zone 4 in the San Antonio area (where sections of Zone 2 may occur), we recommend that a survey be conducted by a qualified karst geologist or karst biologist with demonstrated experience identifying karst features.

Techniques to assess the presence of karst features and potential endangered species habitat: To conduct karst feature surveys, follow methods outlined in section II-A of *Procedure for Conducting a Geologic Assessment* in TCEQ's *Instructions to Geologists for Geologic Assessments (GA)*, as revised October 1, 2004. Applicable portions of those procedures are included here in Appendix I. We intend for you to use the GA to locate features only and not to assess whether a feature has the potential to contain karst invertebrate habitat. Guidance on assessing a feature's potential to contain suitable karst invertebrate habitat is discussed in Step 3 below. If you have questions regarding the GA you may contact the TCEQ Austin Regional Office (512-339-2929), theSan Antonio Regional Office (210-490-3096), or via the internet at *https://www.tceq.texas.gov/*.

If a GA has previously been conducted on the subject site following TCEQ's October 1, 2004, guidelines, then it may serve as an initial karst feature survey. If a GA is not required on the subjectsite by TCEQ (for example, the site is not located on the Edwards Aquifer recharge or transition zones) then we recommend that the initial karst feature survey be conducted following the methods outlined in those portions of section II-A of *Procedure for Conducting a Geologic Assessment* in TCEQ's GA (October 1, 2004) that are contained in Appendix I herein. All surveys should beconducted such that the likelihood of overlooking any karst feature is very low.

In addition to the methods in the TCEQ GA, remote sensing techniques, such as Lidar, video cameras, or geophysical techniques such as, electrical resistivity, microgravity, ground penetrating radar, or natural potential, may be helpful in assessing the presence of a void or the extent of a known feature that may contain suitable habitat. However, the use of such techniques cannot determine the absence of endangered invertebrates. If using these techniques detects inaccessible voids that have potential tolead to a cave, boreholes should be drilled in and near the voids to allow for baiting (see Baiting underStep 5 below). Note: drilling boreholes has the potential to result in take, and therefore, we recommend this activity be covered by a 10(a)(1)(A) permit if it is being implemented as part of these protocols.

Step 2a. If no karst features are found during the initial karst feature survey, no permit report is required. Because surface surveys for karst features are not expected to cause take and can be conducted by someone without a 10(a)(1)(A) permit, this work is not required in a 10(a)(1)(A) annualreport. However, we would appreciate receiving karst feature reports because this information will increase our understanding of these species and will assist in making decisions on management and conservation and in evaluating and refining scientific survey procedures for determining presence/absence. Also, the results may be needed or useful to document that environmental protection compliance requirements have been met (for example, documenting that development would not result in incidental take of endangered species.

Step 2b. If karst features are found during the initial survey, **proceed to Step 3**.

Step 3. Assess karst features for suitable endangered karst invertebrate habitat

The potential for each identified karst feature to contain suitable habitat for endangered karst invertebrates should be assessed by a qualified biologist or a geologist with demonstrated experience in karst geology and the ability to identify cave-adapted invertebrate species.

- If a site seems likely to contain culturally or paleontologically significant materials, action related to the feature should first be coordinated with the Texas Historical Commission (THC), before determining if any excavation is appropriate (http://www.thc.state.tx.us).
- If a feature is filled, or partly filled, by leaf litter, loose soils, and a few rocks, it should be subjected to a reconnaissance excavation, which involves hand removal of loose soil, rocks, and leaf litter not exceeding 30 centimeters (cm) (1 foot (ft)) in depth and 30 cm (1 ft) in diameter prior to its evaluation for potential to lead to a cave. A rod at least 30 cm (1 ft) long should be used to probe into the soils of a feature in search of shallow voids to further estimate the feature's origin and permeability. If reconnaissance excavations are conducted as described above they are not expected to result in "take."
- If a feature exhibits airflow, channelized recharge of water, development by soil or bedrock collapse, loose soil or rock fill to a depth of at least 30 cm (1 ft), or clean-washed rocks at its base, then it may lead to a void.
- The presence of cave crickets (*Ceuthophilus cunicularis* or *C. secretus*) or *Cicurina varians* spiders found during the initial examination or reconnaissance excavation also indicates the presence of a void.

If none of the above factors are present, then any combination of at least two of the following factors should be considered justification for further excavation (Step 4) or examination of a feature (Veni and Reddell 2002):

- The feature is developed along or near a fracture.
- The feature extends at least 1 meter (m) (3.3 feet) beyond the original surface expression.
- The feature morphology is similar to the pre-excavation appearance of a nearby known cave in the same geologic setting.
- The feature contains a visible humanly or potentially humanly enterable void.

Other factors that should be considered justification for further excavation (Step 4) or examination of a feature (Veni and Reddell 2002):

- The feature is close to caves known to contain endangered species.
- The feature is in karst zone 1 or 2.
- The feature has physical characteristics (for example, size, appearance, catchment basin, conduits, air flow, and mammal etchings) that suggest the presence of a cave.
- The feature contains fill that does not match surrounding area, which may indicate the presence of artificial material in the feature.
- The feature contains roots and/or black soil.
- The feature is located near structural features that may promote cave and karst features to

- develop, such as a fault, photolineament (possible bedrock feature indicated by aerial photographs), or an area of relatively high fracture density.
- The vegetation near the feature includes trees (for example, cedar elm trees) that may grow in cave entrances and other karst features.

Characteristics of karst features not likely to contain suitable habitat for endangered karst invertebrates: The following is based on Veni and Reddell (2002) and is provided as guidance for determining when karst features are not likely to contain habitat for endangered karst invertebrates. Each factor below indicates conditions unfavorable to karst invertebrates, but individually, none of these factors rule out their occurrence. A conclusion that no surveys are needed requires that all of these factors occur together, making habitat for the endangered karst invertebrates unlikely.

- Features that have all, or nearly all, floors, walls, and ceilings covered with calcite speleothems (secondary geologic deposit in caves) are unlikely to provide habitat for karst invertebrates. The calcite speleothems may block the species and nutrients for the species from entering the feature.
- The absence of non-listed troglobites or troglophiles suggests conditions are unsuitable for the endangered troglobites. Troglobites are a species of animal that is restricted to the subterranean environment that typically exhibits morphological adaptations to that environment, such as elongated appendages and loss or reduction of eyes and pigment (Veni 2002). Troglophiles are a species of animal that may complete its life cycle in the subterranean environment but may also be found on the surface (Veni 2002). To determine if this characteristic is met, the evaluation must be conducted or directly supervised by someone with experience recognizing these species.
- The feature lacks evidence of water-formed features that may indicate episodic moisture, such as recently formed scallops and pitting of sediments and bedrock, sediment depositional patterns exhibiting flow or ponding, or recent speleothem growth.
- There is an absence of discernible airflow, which suggests the feature may not connect to a cave or or or or or or or or of airflow usually indicates the existence of such voids, its absence does not indicate the opposite. Several factors may prevent airflow when voids are present.
- The feature is not collapse-formed or related to a collapse. If a feature is part of a collapsed area of bedrock, it is part of a deeper, more extensive cave or series of voids that produced the collapse and are more likely to contain suitable habitat for the endangered karst invertebrates.

Based on the assessment of the karst feature to provide suitable habitat, follow the appropriate step below (either 3a, 3b, or 3c):

Step 3a. If, after a thorough assessment, you determine there is no potential for the feature to open to a void or cave, then no additional surveys are needed.

Step 3b. If the karst feature is a cave or has habitat that may be suitable for endangered karst invertebrates, then **proceed to Step 5**.

Step 3c. If the karst feature has potential to open to a void or cave that may contain suitable habitat for endangered karst invertebrates and requires excavation to assess habitat quality, then proceed to Step 4.

Step 4. Excavating features

Considering that excavation of features could result in "take," we recommend that surveyors conducting excavations beyond the scope of a reconnaissance excavation hold a valid 10(a)(1)(A) permit. If personnel are conducting excavation only, then they can receive coverage by being supervised by someone with a 10(a)(1)(A) permit, and the permittee should also be onsite to collect specimens discovered during excavation. The permittee should determine if the feature leads to a cave or void and will require removal of fine sediments, collapsed rocks, calcite deposits, and/or bedrock. Excavation with hand tools should be used whenever possible to minimize disturbance of a feature's environment. Explosives may be needed to excavate collapsed rocks, calcite deposits, and/or bedrock but should be used strategically under the supervision of experienced personnel and in small amounts to selectively remove obstructions. Backhoes or related heavy machinery may be needed where large rocks or volumes of sediments are impractical and/or unsafe for removal by hand. The size of excavations should be kept as small as possible while allowing space for efficient excavation efforts and creating an area safe for entry. Multiple entrances dry out caves; therefore, only one entrance should be excavated. Additional entrances should be sealed with natural fill equivalent in permeability to what was excavated. (Also see *Restoring Excavated Features* below). To minimize promotion of red-imported fire ant (Solenopsis invicta) activity and siltation of streams, excavated material from all features should be evenly distributed downslope of and at least 5 m (16 feet) from the features. Sediments should be distributed in thicknesses of no more than 1-2 cm (0.39-0.78 inches) to allow rapid integration into the existing soils and stabilization by vegetation.

Excavation should cease upon encountering: (1) a cave (caves may require further excavation during biological surveys, see Step 5), (2) solid bedrock with no conduits, (3) packed clay with no airflow present (the passage should be checked several times under different surface temperature conditions (for example, cool mornings, warm evenings) before determining there is no airflow), (4) potential archaeological or paleontological materials, or (5) where continued excavation would be dangerous (for example, due to a large, overhanging rock or high levels of CO₂). If the CO₂ level is high, consider excavating (or surveying) during more favorable weather conditions (such as during the winter months, especially after strong cold fronts, which push O₂ deeper into the cave displacing CO₂).

If a void or cave that may contain suitable habitat for endangered karst invertebrates is encountered during excavation, excavation should stop and a qualified individual (see Appendix II) holding a valid section 10(a)(1)(A) scientific permit should survey for endangered karst invertebrates and, if needed, conduct or supervise further excavations within the cave. However, we recommend immediate collection of any karst invertebrates observed within the entrance area during the initial excavation by an individual holding a valid section 10(a)(1)(A) permit (see Appendix II). Also, while active excavation is ongoing, features and boreholes should be immediately covered with a tarp to prevent the feature from drying, which may cause species to retreat into mesocavernous voids. The tarp should be secured as tight as possible to prevent air from entering or exiting the feature. Therefore, the tarp should be held down by using a combination of large stones, sandbags, light-colored blankets (which are less likely to

¹ **NOTE:** Excavation or any other activity that alters or disturbs the topographic, geologic, or existing recharge characteristics of a site, is regulated under the TCEQ's Edwards Aquifer Program and may require a Water Pollution

Abatement Plan (WPAP). TCEQ's regional office should be consulted prior to either blasting or using a backhoe to excavate any feature occurring in the Edwards Aquifer recharge and transition zones. For more information, contact TCEQ at 512-239-1000 or access the Internet at https://www.tceq.texas.gov/.

absorb sunlight and heat), or other means that would not impact the habitat. Non-toxic materials should be used to accomplish this task, and metal plates should not be usedbecause they dry out the habitat by conducting heat into the feature.

<u>Restoring excavated features</u>: After all necessary biological surveys have been conducted, features, caves, or boreholes should not necessarily be refilled but should remain in, or be returned to, a state most beneficial for the karst ecosystem, which may include but is not limited to: (1) returning the entrance to its pre-excavated condition (to reduce air flow if the original entrance was small) or (2) installing a cave gate to prevent trespassing or large mammal access (for example, feral hogs).

Features that are excavated into caves should usually be left open enough that human access for biological surveys is possible. However, openings larger than 1 m (3.3 feet) to relatively small caves may be detrimental to the karst ecosystem by increasing drying and temperature fluctuation. Therefore, we recommend that those features have smaller openings and be covered with a boulder orother natural materials equivalent in permeability to what was excavated.

After excavating to determine if suitable habitat exists, follow the appropriate step below (either 4a or 4b):

Step 4a. If no suitable habitat for endangered karst invertebrates exists, then no further excavation is necessary. A final karst feature survey report should be provided to the Service's Austin Ecological Services Field Office if excavation is conducted under a 10(a)(1)(A) permit (see Appendix III for reporting requirements).

Step 4b. If suitable habitat for endangered karst invertebrates exists, then stop excavation and **proceed** to **Step 5.**

Step 5. Conduct a Karst Invertebrate Survey

Since collection of endangered species constitutes "take" and is a violation of section 9 of the Act without a permit, species surveys should be conducted by persons holding a valid 10(a)(1)(A) permit. The following section outlines the required survey methodology for conducting presence/absence surveys for endangered karst invertebrates in central Texas under a section 10(a)(1)(A) permit. Once the survey(s) are complete, a comprehensive report must be submitted whether endangered karst invertebrates were encountered or not (See Appendix III for reporting requirements).

NOTE: Any work in a cave is potentially dangerous. The presence of pits and ledges; large, unstable, overhanging rocks; and high levels of CO₂ present danger to researchers. Surveyors should use their best judgment to determine when conditions are safe to proceed with surveys. If invertebrate surveys are limited or discontinued due to safety concerns, this should be made clear in the report. Baiting (see Baiting below) may be recommended as an alternative under these conditions if it can be done safely by the permittee.

<u>Number and timing of surveys</u>: To assess the presence/absence of endangered karst invertebrates with a high level of confidence, survey all caves and significant features at least 14 times. The number of surveys to conduct is based on detection of the endangered *Rhadine exilis* (Krejca and Weckerly 2007). For a more thorough explanation for why we are requiring 14 surveys see Appendix V. All 14 surveys

should be conducted even if a listed karst invertebrate is found before the 14th survey because you may detect a different species of a listed karst invertebrate.

Each survey should occur at least 48 hours apart during suitable surveying conditions (see Suitable surveying conditions below). We encourage permittees to bait every survey unless red-imported fire ants are being observed. However, if no endangered species are detected by the eleventh survey, then baits **must** be used in addition to surveying (see Baiting below). Also, data loggers must be installed in caves and features (as far back into the cave or feature as possible) prior to the beginning of surveysand remain in place until all surveys are completed. We encourage that at least three surveys be conducted at night and at least three during daylight hours to account for possible daily variation in behavior.

Special instructions for features hit during construction: If the feature is humanly enterable, then conduct presence/absence surveys. If you are surveying an inaccessible feature encountered during construction, baiting must be conducted for a minimum of 14 days beginning when the baits are set and ending when the baits are removed. Baits must be checked at least six times during this period with no more than three days between visits (see Baiting below for methods). Data loggers must be installed during the entire two-week baiting period.

<u>Suitable surveying conditions</u>: The entire cave should be searched when conditions in the cave are appropriate for finding the endangered karst invertebrates, generally avoiding temperature extremes and low humidity.

Surveys may be conducted any time of year as long as the weather conditions below are met (see suitable weather conditions). Ideally at least one survey should be conducted in the fall and one in the spring to observe species that may be more active or visible in one season or the other. For example, *Rhadine* beetles appear to be more abundant in the spring, indicating that fall surveys may not be as useful for these species (James Reddell, Texas Memorial Museum, pers. comm. 2002). Also, a study by Weckerly (2010) assessed surveys conducted by the Balcones Canyonlands Preserve staff and found that *Texella* species were not detected in the fall and winter (however, there was not a statistically significant trend in seasonality of detection).

Suitable weather conditions include:

- Average weather (temperature and rainfall) for time of year (found at www.noaa.gov).
- Absence of recent, extensive, local flooding.

<u>Surveying diligence and thoroughness</u>: Since karst invertebrates are small, have low population sizes, and may have behaviors that make them difficult to find, such as retreating under rocks or into mesocavernous passages too small for humans, it is necessary to ensure that sufficient time and effort have been spent surveying. Before any endangered species are considered absent the following should be done:

- Search the void/cave thoroughly.
- Search times should be proportional to the size of the void/cave.
- Use a system of transects to ensure the entire cave is thoroughly searched.

- Search under all loose and easily moveable rocks. Rocks should be moved with care to ensure species are not injured and should be returned to their original position immediately after examination.
- Search under clumps of dried, cracked sediment, which should also be moved with care and returned to their original position after examination.
- Search in crevices, on ceilings, and walls.
- Hand-sift samples of loose sediment and look on, and in, scat and dead animals.
- Search all habitat types, not only those that are believed to be the preferred habitat of the endangered karst invertebrates. Since habitat profiles are incomplete, this will provide information on habitat selection by these species.
- Record species abundance for listed and non-listed species and the microhabitat that they were found on/in. See Appendix III for information to collect during each survey that should be included in annual reports.

<u>Specimen collection and preservation</u>: Since the endangered karst invertebrates may not be possible to distinguish in the field from closely related species, specimens should be collected for identification by a qualified taxonomist.

- Over the duration of the 14 presence/absence surveys, three specimens of each genus of an endangered species may be collected per cave. All collected specimens must be sent to a taxonomist within one month of collection for identification. They should be curated no longer than two months after collection. **NOTE**: Entry and collection in caves known to contain endangered karst invertebrates is not authorized, even under a section 10(a)(1)(A) permit, unless a monitoring or research plan (see Appendix IV for more information on this) has been submitted by a permittee and approved by Austin Ecological Services Field Office.
- These collections should be identified as specifically as possible and sent to the Texas Memorial Museum, in Austin, Texas or other appropriate museum or university listed in our Karst Invertebrate Taxonomy document found at: https://www.fws.gov/library/collections/terrestrial-karst-invertebrates for taxonomic determination and curation.
- All specimens should be preserved in 100 percent non-denatured ethanol because they may be used in future genetic studies.
- Specimens collected should be immediately placed in a cooler and kept there until transferred to a freezer. Before transfer to a freezer, the preservative should be discarded and replaced with new ethanol. All preserved specimens should be stored in a freezer at (-20°C (-4°F)) (Vink et al. 2005).
- All specimens should be stored in separate vials to prevent misidentification in the event that appendages become separated from the body.
- Immature specimens collected alive with the intent of rearing them to adulthood for positive identification (for example, blind *Cicurina* and *Texella* species) should be sent to a taxonomist immediately. To promote specimen viability, surveyors should coordinate shipments with taxonomists so they will know when to expect them and can prepare accordingly.

<u>Baiting</u>: Baits may attract red-imported fire ants into the cave; therefore, they should be used with caution as an invertebrate survey technique. Baits should be used in leads that are inaccessible for visual examination. When baiting is used, follow these conditions:

- Baits should consist of both sticky traps and bottle traps. The reason we are recommending two types of traps is because specimens can be damaged in sticky traps. However, capture probability may be higher in them than in bottle traps. This requirement may be revised if future data indicate that one trap is more desirable than the other.
- Bottle traps should be constructed by cutting the top off a plastic bottle, then inverting and inserting the lid back into the bottle. Prior to reinserting the lid, place cheese or another pungent bait inside the bottle and a funnel inside the lid. The funnel should not touch the bottom of the trap and should allow easy entry and difficult exit. You should also cut small holes in the bottle to allow for air flow; these holes should be as small as possible to prevent individuals from exiting the trap.
- Baits should be placed as far back in a void/cave as possible to reduce the chance of attracting surface species, but the baits must be retrievable.
- Baits can be deployed further into inaccessible features by tying a **strong** string to the bait and feeding the string through a 0.5-inch polyvinyl chloride (pvc) pipe. The string should be long enough to extend a few feet outside of the pipe pushing the bait into the feature with a pole. While deploying the trap, hold the end of the string tightly, once the bait is in place, let go of the string and pull the pipe out of the feature leaving the string for future retrieval. Strings should be inspected for deterioration during each visit to ensure that the trap will be retrievable and not lost in an inaccessible area and thus serving as a constant source of mortality after baiting is completed.
- Baits must be checked every other day; the surface area above the baits must also be checked to ensure that red-imported fire ant mounds are not present. Any red-imported fire ant mounds found prior to, or during, baiting should be immediately treated with boiling water.
- Data loggers must be installed in features prior to the beginning of baiting and remain in place until all baiting is completed. (See above for *Special instructions for features hit during construction*.)

Please note that some karst invertebrate species, such as *Texella* species, are less likely to be captured by baiting (George Veni, George Veni & Associates, in litt. 2003). However, we still recommend this technique, in addition to presence/absence surveys. Also, finding only non-endangered invertebrates in bait traps does not necessarily imply there are no endangered species present.

Reporting: Reports documenting activities under a section 10(a)(1)(A) scientific permit are to be provided to the Service annually. Reporting requirements are outlined in Appendix III.

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Appendix I: Geologic Assessments

Section II-A of the TCEQ Procedure for Conducting a Geologic Assessment TNRCC-0585-Instructions (Rev. 10-1-04) to Geologists for Geologic Assessments on the Edwards Aquifer Recharge/Transition Zones

[Note: We have appended applicable portions of Section II-A that we recommend using to locate karst features. In some places, the text here may be modified slightly from that in the TCEC document. For TCEQ purposes, please see their original and most current document.]

A. Procedure for Conducting a Geologic Assessment

The general procedure for conducting a geologic assessment is to: research information, perform a field survey, evaluate data, return to the site, if necessary, make conclusions, and make a report with your feature assessments and recommendations. A geologic map, notes, photographs and/or sketches should be made while in the field. These data may be used and included in your final report.

Research information

Published reports and maps of area geology should be studied prior to performing the field survey. A literature or database search should be conducted for the presence of documented caves or other karst features on the property or in proximity to the property boundary. Information may be found about known caves, such as mapped extent, depth or elevation or orientation, on the subject property or on adjacent tracts. Some commonly used data sources for geologic maps and cave location and interpretation are included in the "Citations for Sources of Further information" in these Instructions [See TCEQ, GA for these citations].

Evaluate former land use practices and modifications. Interview persons knowledgeable about historical activities such as well drilling, irrigation or water control ditches or trenches, pit or structure construction, episodes of brush clearing and tree pulling, and cave filling or excavation. In ranches that have been occupied for a long time, manmade features can be degraded and overgrown and be confused with natural features. Human activities also may obscure indicators of natural processes that otherwise could be used to determine the sensitivity of a feature. Aerial photos may be examined for the presence of structural features that should be field checked and plotted on the map.

Perform a field survey

The entire subject site must be walked to survey the ground surface for the presence of geologic and manmade features. It is recommended that the site be walked systematically in spaced transects 15.2 meters (50 feet) apart or smaller, paying close attention to streambeds and structural features observed on aerial photographs. The transect pattern should be adapted to ensure that the geologist is able to see features and will vary with topography and vegetation on the site. Streambeds, including dry drainages, are significant because runoff is focused to them. Not only are features in streambeds likely to receive large volumes of recharge, but they are likely to be part of hydrologically integrated flowpaths because past flow has preferentially enlarged and maintained conduits. Features in streambeds are likely to be obscured by transported soil or gravel (swallets or swallow hole). Structural features such as faults and fracture zones have influenced karst processes in the Edwards recharge zone, and awareness of these

structures may be helpful in completing a high-quality assessment. The assessment must include the path of any proposed sewer line that extends outside ofthe water pollution abatement plan (WPAP) assessed area plus 15.2 meters (50 feet) on either side. Any features identified should be marked where possible with flagging or stakes, accurately located, preferably using a GPS, and assigned a unique number with the location accurately plotted on the geologic map.

[Note: After all karst features are located and mapped, please return to Step 3 of the "United States Fish and Wildlife Service, Section 10(a)(1)(A) Scientific Permit Requirements for Conducting Presence/Absence Surveys for Endangered Karst Invertebrates in Central Texas" to determine if potential karst invertebrate habitat may be present.]

Appendix II: Surveyor Qualifications

United States Fish and Wildlife Service, Section 10(a)(1)(A) Endangered Karst Invertebrate Surveys: Surveyor Qualifications

The following levels of expertise are required for issuance of a section 10(a)(1)(A) scientific permit to conduct presence/absence surveys for endangered karst invertebrates in central Texas. The Service will consider on a case-by-case basis granting a section 10(a)(1)(A) scientific permit to individuals who do not meet these qualifications but who have demonstrated adequate/appropriate experience to conduct this work.

To be considered qualified by the Service to conduct unsupervised presence/absence surveys for listed karst invertebrates and to supervise others in the field, the conditions below should be met:

- The person has extensive experience identifying both listed and non-listed karst invertebrates in Texas with at least one year of experience collecting (under the on-site supervision of a biologist permitted to survey for karst invertebrates) and accurately identifying, at least to genus, the listed karst invertebrates in the county being surveyed.
- Also, the person can provide letters of recommendation from two individuals that hold a valid 10(a)(1)(A) permit to independently survey for karst invertebrates or a taxonomist that has received their collections. Individuals that write these letters should attest to the applicant's ability to properly collect and identify karst invertebrates to the genus level. Equivalent experience in caves outside of Texas may be acceptable. The Service will review these on a case-by-case basis.

NOTE: Other individuals may be permitted to accompany permittees into caves to gain experience or for the reasons of caving safety but must remain under the direct on-site supervision of the Permittee at all times. Also, a section 10(a)(1)(A) permit may be issued to a qualified geologist with demonstrated experience in karst geology covering "take" of endangered karst invertebrates that may occur during a habitat assessment and/or excavation and for the collection of endangered karst invertebrates encountered while conducting these activities. Note, these individuals may not conduct presence/absence surveys unless they are permitted for that activity.

Appendix III: Reporting Requirements

United States Fish and Wildlife Service, Section 10(a)(1)(A) Karst Feature and EndangeredKarst Invertebrate Surveys: Report Requirements

An annual permit report is required for 10(a)(1)(A) permit holders. For information that should be included in these reports see the Karst Invertebrate Survey Report section below.

A section 10(a)(1)(A) scientific permit is not required to conduct surface walking surveys to determine the presence/absence of karst features, as no "take" of listed species is likely to occur. However, we would appreciate receiving karst feature reports. These data are important, even if findings are negative, for the conservation and recovery of the species. See the Karst Feature Survey Report section below for information that we would find helpful in these reports.

KARST INVERTEBRATE SURVEY REPORT: This report is **required** by 10(a)(1)(A) permittees and should include, but is not limited to, the information below. This information will benefit the conservation of these species by furthering our knowledge of the biology and ecology.

Personnel

- List the names of all persons involved in the surveys (including baiting) and their duties.
- List each person's section 10(a)(1)(A) scientific permit number, if applicable.
- Provide a brief summary of experience, education, and certification for each person **NOT** holding a valid section 10(a)(1)(A) scientific permit.
- Identify the person(s) directly responsible for writing the report.

Location

- Provide the location of caves and features surveyed and the property boundaries on a USGS topographic map (7.5 minute or larger scale) or, if possible, in a GIS (Geographic Information System) layer with georeferenced location data (using global positioning system (GPS)), including references such as roads and political boundaries.
- Provide GPS coordinates (if GPS used) for each cave or feature surveyed. Also, report the GPS unit model, its accuracy, and if any real time correction or post processing was done.
- Provide georeferenced data (if collected) in lat-long (decimal degrees). North American Horizontal Datum 1983 (NAD 83) is preferred. If collected in an alternate coordinate system, please report the coordinate system and datum the information was collected in.

Methods

- Describe survey methodology using standards consistent with a scientific, peer-reviewed publication.
- Report whether the entire cave was surveyed or if surveys were conducted along transects or following another statistical sampling method and describe that methodology.
- Report the use of baiting. Include a description of the methodology used including the type(s) of bait used, the type and location of traps, and the amount of time traps were left out. Also, report the capture results by trap and date checked.
- Report total time spent searching (in person-hours) specifically for karst invertebrates.
- Report date and time of day each survey was conducted.

- Report temperature and humidity on the surface and at locations inside the feature as indicated below in the section titled "Caves and Karst Features." Indicate the brand and model of the equipment used and the equipment's accuracy (degree of accuracy).
- Report weather conditions on the survey day and previous week.
- Report temperature and humidity results of data loggers as well as the type of data logger and the location in the cave or feature where the date logger was installed.

Caves and Karst Features

- Describe each cave or feature surveyed and include a detailed, scaled cave map with plan and profile views.
 - Description or map should include:
 - The approximate passable length of the cave or feature.
 - Possible leads or breakdown areas that could contain mesocavernous invertebrate habitat that is not humanly passable.
 - The approximate heights and widths of passages.
 - Locations of any standing or flowing water.
- Describe the interior of each cave or feature surveyed including:
 - Principal formations and whether they are active.
 - Make-up of the cave floor in each section (for example, mud, breakdown with approximatesizes, powder).
 - Approximate area and depth for standing water and approximate width, length, depth, and flow rate.
 - Temperature (to the nearest 0.1°F) and relative humidity (to the nearest 1 percent). Indicate the brand and model of the equipment used and the equipment's degree of accuracy. Temperature and relative humidity should be taken at a minimum just inside the entrance and at the farthest humanly accessible part of the cave or feature. Several locations are preferred, particularly for large caves or those with multiple rooms, and should be referenced to labeled locations on the cave map.
 - Report any indications of "bad air," (for example, high CO₂ levels or any noxious gas) and reference to labeled locations on the cave map.
 - Report the result of any excavation, including reasons for discontinuing excavation.
 - Describe the methodology used for restoring excavated features, if applicable.

Species and Biotic Karst Community

Report the presence of all species, listed and non-listed, observed or collected during surveys or any other activity such as during the initial karst feature survey following the TCEQ GA, including:

- Identify species (vertebrate and invertebrate) as specifically as possible, preferably to species level,including:
 - Troglobites a species of animal that is restricted to the subterranean environment and typically exhibits morphological adaptations to that environment, such as elongated appendages and loss or reduction of eyes and pigment (Veni 2002).
 - Troglophiles a species of animal that may complete its life cycle in the subterranean environment but may also be found on the surface (Veni 2002).

- Trogloxenes a species of animal that inhabits caves but must return to the surface for food and other necessities (Veni 2002).
- Accidentals species that may wander into caves but cannot survive there.
- Report listed species behavior when observed (for example, feeding, sedentary, moving, etc.).
- Report the presence of dead specimens (vertebrate and invertebrate) and identify them to the lowest taxonomic level possible.
- Report numbers of each species (listed and non-listed) encountered on each survey date. For highly abundant species, approximations are acceptable.
- Describe the microhabitat where species (listed and non-listed) were found, including:
 - Type of substrate the specimen was found on (for example, large breakdown; dry, fine silt; under a fist-sized rock; on the ceiling).
 - Type of rock/soil the specimen was found on.
 - An approximation of how much organic material was found in the cave (for example, scat, bat or cricket guano, dead animals, plant material, fungus) with a reference on the cave map to where the organic material was found.
 - Proximity to water.
 - For listed species, indicate location(s) found on the cave map.
- Report any previous collections in the cave, regardless of the listing status of those species.
- Provide a description and sketch of the area immediately around the cave entrance (approximately 10 meters (32.8 feet)), including approximate percent cover by bedrock versus soil, approximate percent cover by trees or shrubs versus herbaceous plants, and approximate percent cover by deciduous versus coniferous trees.
- Also, report locations where caves/features were searched but no listed species were found and any additional information above that is available.

Species Identification

If specimens are tentatively identified as listed species in the field, they should still be sent to a taxonomist for verification. A list of taxonomists is in our Karst Invertebrate Taxonomy document at: https://www.fws.gov/library/collections/terrestrial-karst-invertebrates. The final report should include the results of the taxonomist's identification including accession numbers for each specimen. If taxonomic results are not back at the time your report is due, identify where the specimens were sent, the date they were sent, and how many specimens were included. The report should include a list of species collected (listed and non-listed species to the genus level) and/or encountered during collections, name of collector(s), date of collection, and method of preservation/storage. Once the taxonomist's results are received, they should be reported to the Service within two weeks.

Specimen Deposition

All specimens should be deposited with an appropriate curated museum collection as approved in writing by the Austin Ecological Services Office if different from our list in the Karst Invertebrate Taxonomy document referenced above.

KARST FEATURE SURVEY REPORT: While a report on the surface survey for features is not required, we would appreciate if you prepared and submitted a comprehensive written report following the completion of karst feature surveys. This information will increase our understanding of these species and will assist in making decisions on management and conservation and in evaluating and refining scientific survey procedures for determining presence/absence. In addition to the information required by the TCEQ's GA, the following information would be helpful to include:

Personnel

- Names of all persons involved in the surveys and their duties in the karst feature survey report.
- Each person's section 10(a)(1)(A) scientific permit number, if applicable.
- Person(s) directly responsible for writing the report.

Feature Survey Methodology

Describe survey methodology using standards consistent with a scientific, peer-reviewed publication. Please include in the report:

- Total time spent searching for karst features and spacing and direction of all transects.
- A map of the survey location with transects and features identified.
- Results of reconnaissance excavations and methodology used for restoring excavated features, if applicable. (Note: for excavations that go below 30 cm (1 ft) deep, we recommend the surveyor have a 10(a)(1)(A) permit because "take" is more likely to occur below this depth.)

Supporting information

- Citations for all references used or consulted in the final report.
- Definitions of any terminology that would not be common knowledge to persons with general scientific, non-geology specific backgrounds including terminology specifically used by or for agencies other than the Service, for example, the TCEQ.
- Results of any additional studies related to the karst investigations, for example, biological observations, remote sensing for subsurface voids, hydrological studies, etc.

Appendix IV: Recovery Project Proposals

United States Fish and Wildlife Service, Section 10(a)(1)(A) Endangered Karst Invertebrate Recovery Project Proposal Requirements

Permittees are prohibited from entering or collecting from locations known to contain endangered karst invertebrates until they submit a detailed recovery project proposal (previously referred to as monitoring plans) and receive written approval of that proposal from the Austin Ecological Services Office (Recovery and Candidate Conservation Branch). We approve recovery project proposals only if the proposed activities and any associated "take" will provide a conservation benefit to the species. Recovery project proposals should be submitted with the following sections and information below. These projects may include research, monitoring, or other conservation-related activities. Recovery project proposals that do not include this information will be returned to the permittees for revision.

Purpose

Thoroughly explain the purpose or hypothesis of the proposed project and how it will benefit the conservation of the endangered karst invertebrates. Include any pertinent background information that supports the need for the proposed project. Explain feasibility of the research/project.

Location

- Location of caves and features to be entered and the property boundaries on either a USGS topographic map (7.5 minute or larger scale) or, if possible, in a GIS (Geographic Information System) layer with georeferenced location data (using global positioning system (GPS)), including references such as roads and political boundaries.
- Include GPS location information for each cave or feature to be entered. Also, report the GPS unit model and its accuracy, and if any real time correction or post processing will be done.
- Georeferenced data should be collected in lat-long (decimal degrees). North American Horizontal Datum 1983 (NAD 83) is preferred.

Personnel

- Names of all persons that will be involved in the recovery project and their precise duties.
- Each person's section 10(a)(1)(A) scientific permit number, if applicable.
- A brief summary of experience, education, and certification for each person **NOT** holding a section 10(a)(1)(A) scientific permit who will be involved in the research.

Methods

- Describe any proposed research methodology using standards consistent with a scientific, peerreviewed publication.
- Describe the statistical analysis that will be used to assess results such as species abundance, diversity, or other parameters.
- Describe if the entire cave will be involved in the recovery project or if transects or another sampling method will be used and how many times the cave will be entered.
- Discuss if bait traps will be used and why they are necessary for the recovery project. Include a description of the proposed baiting methodology including the type(s) of bait used, bait trap locations, the amount of time baits will be left out, and intervals when they will be checked.

Final Recovery Project Reports

Final projects should include all information from Appendix III and thorough results and discussion sections consistent with a peer-reviewed scientific publication. These reports must be included in each permittee's annual report; however, if the report is available sooner than the due date for annual reports, we request that permittees submit them sooner. If the recovery project is amulti-year project, then a progress report should be included in each permittee's annual report.

Appendix V: Protocol Revisions

United States Fish and Wildlife Service, Section 10(a)(1)(A) Endangered Karst Invertebrate Survey Protocol Revisions

As part of our May 8, 2014, revision, we changed the number of surveys required to determine presence/absence of endangered karst invertebrates in central Texas from 3 to 14. We based the revised number of required surveys on a study by Krejca and Weckerly (2007). This study used the program PRESENCE (Mackenzie et al. 2006) to assess detection probabilities of three troglobitic species including *Chinquipellobunus madlae*, *Batrisodes unicornis*, and the endangered *Rhadine exilis*. They concluded that 10 to 12, 22, and 14 surveys, respectively, are needed to have a 95 percent confidence interval that those species were detected if present at a given site.

Prior to our survey protocol revision, we conducted an independent peer review of Krejca and Weckerly (2007). Peer review comments support that more than three surveys (and possibly more than 14) are needed to detect the sixteen endangered karst invertebrates in central Texas; however, they indicated that the number of surveys needed to detect each species may vary. Species that are relatively more abundant and conspicuous may be easier to detect and require fewer surveys than species that are relatively more cryptic, which may require many more surveys.

Although peer reviewers identified some issues with the Krejca and Weckerly (2007) report, they all indicated that more than three surveys are needed to conclude presence/absence with a high degree of confidence. Therefore, absent additional information and analysis, we are requiring 14 surveys to conclude presence/absence of the endangered karst invertebrates. In addition, we are seeking advice from biostatisticians on how to analyze survey data in the future to further evaluate and potentially revise this requirement. We plan to adapt the number of required surveys, as needed, based on future analyses.

One exception to the requirement to conduct 14 surveys was included in the May 8, 2014, revisions. This exception allowed presence/absence surveys being conducted for individuals who planned to participate in the Williamson County Regional Habitat Conservation Plan (RHCP) to end when either *Texella reyesi* or *Batrisodes texanus* were detected in the North Williamson County or Georgetown Karst Fauna Regions. This was because the amount of mitigation when participating in the RHCP is the same if one or both species are found and because no other listed karst invertebrates are known from these two regions. In our April 27, 2022, revision, we removed this exception as we believe genera such as *Batrisodes* are being underrepresented in surveys because they are more difficult to detect. Although the mitigation amount for applicants is the same regardless of species, the purpose of section 10(a)(1)(A) permits, also referred to as recovery, enhancement of survival, or scientific permits is to produce information upon which to base decisions and actions for the conservation of the listed species. The knowledge gained during these surveys is important to help us better understand the range of these species and their habitat requirements as well as to facilitate recovery.