Peck's Cave Amphipod
(Stygobromus pecki)
5-Year Status Review:
Summary and Evaluation

U.S. Fish and Wildlife Service
Austin Ecological Services Field Office
Austin, Texas
March 28, 2024

5-YEAR REVIEW

Species reviewed: Peck's cave amphipod (*Stygobromus pecki*)

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1.0 GENERAL INFORMATION

1.1 Reviewers:

Lead Regional or Headquarters Office:
Vanessa Burge, Recovery Biologist, Southwest Regional Office, Albuquerque, New Mexico, vanessa_burge@fws.gov

Lead Field Office:
Amelia Hunter and Michael Warriner, Fish and Wildlife Biologist, Austin Ecological Services Field Office, Austin, Texas, amelia_hunter@fws.gov, michael_warriner@fws.gov

Cooperating Field Office(s):
Not Applicable

Cooperating Regional Office(s):
Not Applicable

1.2 Purpose of 5-Year Reviews:

The U.S. Fish and Wildlife Service (Service or USFWS) is required by section 4(c)(2) of the Endangered Species ESA (ESA) to conduct a status review of each listed species once every 5 years. The purpose of a 5-year review is to evaluate whether or not the species' status has changed since it was listed (or since the most recent 5-year review). Based on the 5-year review, we recommend whether the species should be removed from the list of endangered and threatened species, be changed in status from endangered to threatened, or be changed in status from threatened to endangered. Our original listing as endangered or threatened is based on the species' status considering the five threat factors described in section 4(a)(1) of the ESA. These same five factors are considered in any subsequent reclassification or delisting decisions. In the 5-year review, we consider the best available scientific and commercial data on the species and focus on new information available since the species was listed or last reviewed. If we recommend a change in listing status based on the results of the 5-year review, we must propose to do so through a separate rule-making process including public review and comment.

1.3 Methodology used to complete the review:

The Service conducts status reviews of species on the List of Endangered and Threatened Wildlife and Plants (50 CFR 17.12) as required by section 4(c)(2)(A) of the ESA (16 U.S.C. 1531 et seq.). The Service provides notice of status reviews via the *Federal Register* and requests new information on the status of the species (e.g., life history, habitat conditions, and threats). Data for this status review were solicited from interested parties through a *Federal Register* notice announcing this review on May 5, 2021 (86 FR 23976) with a subsequent correction for that notice published on February 26, 2024 (89 FR 12868). The Austin Ecological Services Field Office conducted this review and considered both new and previously existing information from federal and state agencies, municipal and county governments, non-governmental organizations, academia, and the public. The primary sources of information used in this analysis was the final rule listing the Peck's cave amphipod as endangered (62 FR 66295), revised critical habitat ruling for the Peck's cave amphipod (78 FR 63100), research published in scientific journals, and unpublished reports and data.

1.4 Background:

1.4.1 FR Notice citation announcing initiation of this review:

86 FR 23976 May 5, 2021

1.4.2 Listing history:

Original Listing
FR notice: 62 FR 66295
Date listed: December 18, 1997
Entity listed: Peck's cave amphipod (*Stygobromus pecki*)
Classification: Endangered

Revised Listing, if applicable
FR notice: Not applicable
Date listed: Not applicable
Entity listed: Not applicable
Classification: Not applicable

1.4.3 Associated Rulemakings:

In a petition dated September 9, 1974, the Conservation Committee of the National Speleological Society requested that the Service to list *Stygobromus* (=*Stygonectes*) *pecki*. The species was included in a notice of review published on April 28, 1975 (40 FR 18476). A "warranted but precluded" finding regarding several species in that petition was made on October 12, 1983, and published on January 20, 1984 (49 FR 2485). The same determination was repeated for Peck's cave amphipod in subsequent years.

The species was included as a category 2 candidate in comprehensive notices of review published on May 22, 1984 (49 FR 21664), January 6, 1989 (54 FR 554), and November 21, 1991 (56 FR 58804). Category 2 candidates were species for which data in the Service's possession indicated that listing was possibly appropriate, but substantial data on biological vulnerability and threats were not known or on file to support proposed rules. Peck's cave amphipod was elevated to category 1 status in the 1994 notice of review (59 FR 58982). Category 1 candidates were those species that the Service had on file substantial information on biological vulnerability and threats to support a proposal to list. As published in the *Federal Register* on February 28, 1996 (61 FR 7596), candidate category 2 status was discontinued, and only category 1 species are currently recognized as candidates for listing purposes.

Critical habitat for Peck's cave amphipod was revised on November 22, 2013, in areas of occupied, spring-related aquatic habitat with designations for surface and subsurface critical habitat (78 FR 63101). The original critical habitat designation encompassed only surface critical habitat and did not include any designation for subsurface critical habitat (72 FR 39248). Springs, associated streams, and underground spaces immediately inside of or adjacent to springs, seeps, and upwellings are the primary components of the physical or biological features essential to the conservation of this species (50 CFR 17.95).

1.4.4 Review History:

Not applicable

1.4.5 Species' Recovery Priority Number at start of 5-year review:

2C

1.4.6 Recovery Plan or Outline

Name of plan or outline: Not Applicable
Date issued: Not Applicable
Dates of previous plans/amendment or outline, if applicable: Not Applicable

2.0 REVIEW ANALYSIS

Section 4 of the ESA (16 U.S.C. 1533) and its implementing regulations (50 CFR part 424) set forth the procedures for determining whether a species meets the definition of "endangered species" or "threatened species." The ESA defines an "endangered species" as a species that is "in danger of extinction throughout all or a significant portion of its range," and a "threatened species" as a species that is "likely to become an endangered species within the foreseeable future throughout all or a significant portion of its range." The ESA requires that we determine whether a species meets the definition of "endangered species" or "threatened species" due to any of the five factors described below.

Section 4(a) of the Act describes five factors that may lead to endangered or threatened status for a species. These include: A) the present or threatened destruction, modification, or curtailment of its habitat or range; B) overutilization for commercial, recreational, scientific, or educational purposes; C) disease or predation; D) the inadequacy of existing regulatory mechanisms; or E) other natural or manmade factors affecting its continued existence.

The identification of any threat(s) does not necessarily mean that the species meets the statutory definition of an "endangered species" or a "threatened species." In assessing whether a species meets either definition, we must evaluate all identified threats by considering the expected response of the species, and the effects of the threats—in light of those actions and conditions that will ameliorate the threats—on an individual, population, and species level. We evaluate each threat and its expected effects on the species, then analyze the cumulative effect of all of the threats on the species as a whole. We also consider the cumulative effect of the threats in light of those actions and conditions that will have positive effects on the species—such as any existing regulatory mechanisms or conservation efforts. The Service recommends whether the species meets the definition of an “endangered species” or a “threatened species” only after conducting this cumulative analysis and describing the expected effect on the species now and in the foreseeable future.

2.1 Distinct Population Segment (DPS) policy (1996):

Not Applicable

2.2 Updated Information and Current Species Status

2.2.1 Biology and Habitat

2.2.1.1 New information on the species’ biology and life history:

Background

Peck’s cave amphipods are groundwater obligate crustaceans that inhabit subterranean habitats, have restricted ranges, and can potentially occupy deep groundwater niches (Holsinger 1967, p. 119; Arsuffi 1993, p. 14). The species was first collected in 1964 at Comal Springs in Comal County, Texas and later collected at Hueco Springs, 7 kilometers (4 miles) north of Comal Springs, in 2003 (Holsinger 1967, pp. 117, 119; Fries et al. 2004, p. 5; Gibson et al. 2008, pp.76-81). This is the first 5-Year Review for the amphipod since the species’ listing in 1997.

Biology

The Peck's cave amphipod, despite its shallow groundwater and spring-associated distribution, exhibits both surface and deeper groundwater characteristics (Holsinger 1967, p. 143; Fries et al. 2004, p. 7). It has no eyes and lacks mechanoreceptors for prey detection beyond direct interaction but can detect and avoid light (Nowlin et al. 2015, pp. 49-50; Nowlin et al. 2016, p. 30; Kosnicki and Julius 2019, p. 21). In both captivity and natural habitats, these amphipods typically inhabit the space beneath leaf substrate or interstitial spaces between rocks, displaying a preference for shelter rather than swimming freely or exposed at the surface (Arsuffi 1993, p. 14; Fries et al. 2004, p. 8).

The Peck’s cave amphipod, a top invertebrate predator in food webs at both spring run 3 and Spring Island locations within the Comal Springs ecosystem, consumes organic from both the surface (e.g., photosynthetic) and groundwater ecosystems (e.g., chemolithoautotrophic) (Hutchins et al. 2016, p. 1536; Kosnicki and Julius 2019, pp. 20-21; Nair et al. 2020, p. 10; Nair et al. 2021, p. 239, 242). These food sources vary based on local vegetation and environmental characteristics, impacting the amphipod's dietary options (Nair et al 2021, p. 242). Therefore, this species is considered a shallow phreatic zone specialist, adept at adapting its feeding strategy to different environments and available food resources and may be able to switch to alternative food sources when environmental conditions are reduced or altered (Nowlin and Worsham 2015, pp. 45, 49, 51). This may explain why *Stygobromus sp.* amphipods from the Comal Springs ecosystem have orange hues due to carotenoid-rich food resources compared to the opaque hues of individuals observed at Hueco Springs ecosystem (Fries et al. 2004, p. 5; Gibson et al. 2008, p. 77).

Peck's cave amphipods can adapt their feeding strategy to reduced or altered environmental conditions by switching to an alternative food source between the locations at Comal Springs spring run 3 (wood biofilm-based food chain) and Spring Island (periphyton-based food chain) (Nowlin and Worsham 2015, pp. 45, 49, 51). In laboratory studies, Peck's cave amphipod showed lower metabolic rates and better energy reserves when starved compared to surface amphipod species, *Sicifera* (*Synurella*) sp., indicating their metabolic strategies match that of deep phreatic organisms of low or infrequent food accessible systems, despite its association with shallower groundwater habitat (Nair et al. 2020, pp. 9-10). This suggests a possible evolutionary history of the Peck's cave amphipod occurred at deeper depths over an undetermined period (Nair et al., 2020 p. 11).

Life History

The mating behavior of this amphipod is unknown, but larger females have been known to cannibalize smaller males (Nowlin et al. 2016, p. 31). In captivity, wild adult females can produce approximately 10 eggs per female, with a hatching success rate of 24 percent and an average incubation time of 49.7 ± 12.4 days (Fries et al. 2004, p. 9; Kosnicki and Julius 2019, p. 11). Brooding females have been observed cannibalizing juveniles outside of the marsupium (i.e., a type of brooding pouch of a female crustacean) or when eggs drop with agitation (Nowlin et al. 2016, p. 31; Kosnicki and Julius 2019, p. 12; Service 2019, p. 57).

The eggs require more than 32 days to survive their first molting event to become neonates (i.e., newborns), and they go through multiple molts over an average period of 50 days to reach the final adult life stage under stressful captive conditions (Kosnicki and Julius 2019, pp. 11-12, 20). Juvenile Peck's cave amphipods reach sexual maturity between the sixth and eight instars, which likely depend on available food resources and temperatures (Kosnicki and Julius 2019, p. 19).

Subterranean amphipods in general have life cycles that vary from 4–10-year life spans (Wellborn et al. 2015, p. 788). Wild-caught Peck's cave amphipods have survived in captivity for three years and have successfully achieved F2 generations, which is an indication that habitat conditions in captivity are suitable and promising for future reintroduction efforts (BIO-WEST, Inc. 2007, p. 40).

2.2.1.2 Abundance, population trends (e.g. increasing, decreasing, stable), demographic features (e.g., age structure, sex ratio, birth rate, seed set, germination rate, age at mortality, mortality rate, etc.), or demographic trends:

Little is known about limiting factors that may impact the abundance and distribution of the Peck's cave amphipod because the subterranean habitats they inhabit are largely inaccessible to humans aside from these wells and springs. Current abundance estimates only include samples collected at the surface.

The species' reclusive nature and life history adds complexity to determine abundance, as individuals spend most of their lives underground. Thus, no population estimates are available for the Peck's cave amphipod. Mature and immature life stages have been collected only near spring outlets, from seeps along the spring runs, and from a shallow groundwater well in Panther Canyon, further complicating such estimation efforts (Gibson et al. 2008, p. 76).

A 1992 study indicated these cave amphipods were abundant in Comal Springs (spring runs 1, 2, 3, and 4) driftnet samples, with 271 individuals, and one specimen at a new locality, Hueco Springs, over 96 hours of combined drift time (Barr 1993, pp. 37, 56-57). The species was abundant at all spring runs but spring run 4 (spring run 1: 78; spring run 2: 62; spring run 3: 130; spring run 4: 1) (Barr 1993, p. 56).

Surveys in 2003 collected an average of 9.2/day were collected at Comal Springs (spring runs 1, 2, and 3) and an average of 1.2/day at Hueco Springs (Fries et al. 2004, pp. 6-7; Gibson et al. 2008, p. 79). Individual cave amphipods were more abundant and easily accessible via hand collection or driftnetting at Comal Springs compared to Hueco Springs, with other sampling evidence to suggest Peck's cave amphipods inhabit a deeper section at Hueco Springs compared to the Comal Springs sampling locations (Fries et al. 2004, p. 7).

Biomonitoring for all benthic macroinvertebrates in the Comal Springs system was established in 2000 and occurs every spring and fall using driftnets (BIO-WEST, Inc. 2003, pp. 37-41). The Peck's cave amphipod was discovered at the western shoreline and upwellings in Landa Lake (BIO-WEST, Inc. 2004, p. 37). Between 2017 and 2021 with over 29 sampling events, the long-term median number of Peck's cave amphipods collected per cubic meter (m^3) of water is $0.25/m^3$ (8.8 per cubic foot [ft^3]) (BIO-WEST, Inc. 2021, pp. 39-40). However, without access to their subterranean habitat, little can be evaluated in terms of their actual population sizes or abundances within their entire habitat at this time using current available methodology.

2.2.1.3 Genetics, genetic variation, or trends in genetic variation (e.g., loss of genetic variation, genetic drift, inbreeding, etc.):

Genetic analysis using mitochondrial DNA sequences indicated high levels of differentiation within and among Peck's cave amphipod localities, but they were found to contain sequences from two distinct haplotype groups with deep divergence (Ethridge et al. 2013, p. 233, 235). The two haplotypes were not geographically separated, and they co-occurred and often in similar proportions. This observation, in addition to the hydrogeology of the Comal ecosystem, suggests Peck's cave amphipod is composed of two sub-populations that at one time were separated and now converge between surface habitats at Comal Springs and migration present within the Comal ecosystem (Nice and Lucas 2015, pp. 18, 22; Lucas et al. 2016, pp. 8, 12).

Measurements of genetic diversity across populations of *Stygobromus* spp. show Peck's cave amphipod to be comparable to populations of congeneric species from central Texas. Future sampling would be beneficial for estimation of population size (Nice and Lucas 2015, pp. 42-44).

2.2.1.4 Taxonomic classification or changes in nomenclature:

The original description of the Peck's cave amphipod (*Stygobromus pecki*) placed this species in the genus *Stygonectes* (Holsinger 1967, entire), which was later synonymized by into the genus *Stygobromus* and placed into the *flagellatus* group (Holsinger 1967, entire). This species is also referred to in some references as the "Peck stygobromid" or "Peck's cave scud" (40 FR 18477; McLaughlin et al. 2005, p.145).

2.2.1.5 Spatial distribution, trends in spatial distribution (e.g. increasingly fragmented, increased numbers of corridors, pollinator availability, etc.), or historic range (e.g. corrections to the historical range, change in distribution of the species' within its historic range, etc.):

Various researchers have examined amphipod assemblages from springs, caves, and wells from neighboring counties, without finding the Peck's cave amphipod elsewhere, beyond the known occurrences at Comal and Hueco spring ecosystems (Holsinger 1967, entire; Holsinger and Longley, 1980 entire; Barr 1993, entire; Gibson et al. 2008, entire). This suggests that individuals of the species may be confined to small areas surrounding the spring openings and are not distributed throughout the aquifer.

The collection of Peck’s cave amphipods at Panther Canyon well lends support to early characterizations of the *flagellatus* group suggesting they inhabit deeper groundwater niches compared to other amphipod groups found above the water table and in hyporheic (i.e., saturated sediments near a streambed gravel or river) habitats (Holsinger 1967, pp. 143, 159).This distinction in partitioned niche habitat zones were also exhibited at Hueco Springs, with Peck’s cave amphipods found more prevalent at deeper sites than others in the genus, *Stygobromus russelli* (Gibson et al. 2008, p. 80).

To what extent the subterranean connections between Hueco and Comal Springs are inhabited by this amphipod are unknown (72 FR 39255). Presumably an interconnected area, the subterranean portion of this habitat provides for feeding, growth, survival, and reproduction of the Peck's cave amphipod. Both springs have local and regional groundwater contributions, with Comal Springs having a more phreatic, older origin than Hueco Springs (Ogden et al. 1986, pp. 80, 124; Rothermel and Ogden 1987, p. 76). These groundwater sources can intermix when aquifer levels are high and separate during severe droughts. This regional flowpath connection could explain the distribution of Peck’s cave amphipods over these two spring systems (Gibson et al. 2008, p. 75).

2.2.1.6 Habitat or ecosystem conditions (e.g., amount, distribution, and suitability of the habitat or ecosystem):

Peck’s cave amphipod inhabits the shallow, subterranean spaces associated with thermally stable spring orifices issuing from the Edwards (Balcones Fault Zone) Aquifer (herein referred to as the “Edwards Aquifer”) (Holsinger 1967, p. 119). It is unknown if this species can re-enter the subterranean aquifer once it has emerged or discharged through the springs (Barr 1993, p. 52). Specific springflow requirements and how much subterranean habitat this species uses is unknown; management relies on assuring historical conditions are maintained within the natural habitat for the species (LBG-Guyton and Associates et al. 2004, pp. C-4–C-5).

This cave amphipod is likely an omnivore and upon reaching the surface, consumes terrestrial-derived organic matter from riparian vegetation sources (78 FR 63100; Nair et al. 2021, p. 3). In the Comal ecosystem, this cave amphipod occupies a higher trophic level as a predator consuming other surface aquatic crustaceans (Nowlin et al. 2017, pp. 15-16). Therefore, riparian areas adjacent to the spring ecosystem provide a necessary role in the nutrient cycle for the food web of this invertebrate and influence its habitat distribution.

The principal habitat at Comal Springs (spring runs and Landa Lake) maintains a fairly stable water temperature at both locations (69.3 and 75 °F [20.7 and 23.9 °C]) and conductivity (579-587 micro-Siemens/centimeter), low levels of dissolved oxygen (5.06-5.23 milligrams/liter [mg/L]), with few detections of contaminants, such as personal care products and pharmaceuticals (BIO-WEST, Inc. 2021, p. 18; EAA 2013, p. 62; EAA 2018, p. 5; EARIP HCP 2021, pp. 27-36, 45-47). Nevertheless, other anthropogenic contaminants have been identified as concerning and may be relevant to the species. For example, nitrate runoff from surface water recharge results in elevated nitrate concentrations within the aquifer. Nitrate levels exceeding 1 mg/L suggest the presence of anthropogenic inputs and urbanization in the recharge zone, a trend documented historically at Comal Springs. Over the past 70 years, these concentrations have doubled (median concentration 2 mg/L), posing a concerning threat to the ecological health of Comal Springs and highlighting the detrimental impact of human activities on the local aquifer system (Dubrovsky et al. 2010, p. 79; Musgrove et al. 2016, pp. 462, 465, 467; Castaño-Sánchez et al. 2020, p. 6).

Information for habitat conditions at Hueco Springs are incomplete due to lack of access. The best available information indicates that the shallow spring waters at Hueco Springs are relatively constant with near neutral (pH 6.8 to 7.0), range in temperature between 69.3-70.7 °F (20.7-21.5 °C), supersaturated with oxygen (5.0-6.8 mg/L; over 100 percent saturation), and few detections contaminants, such as of personal care products and pharmaceuticals (Fries et al. 2004, pp. 4, 13; EAA 2015, pp. 56-58; EAA 2018, p. 5).

2.2.1.7 Other:

Biological Constraints and Needs

Peck's cave amphipod occurs in a limited range at a small number of localities with little or no ability to disperse between or beyond these localities. These characteristics make them susceptible to local extirpation and extinction (McKinney 1997, p. 499; O'Grady et al. 2004, p. 514). A severe drought or water contamination event could eliminate many or all the existing sub-populations. Having a high number of individuals at a site provides no protection against extinction due to stochastic events. Dispersal beyond their extant range is unlikely, given the isolated nature of the spring headwater system dynamics and aquifer hydraulic connectivity that limit movement of individuals.

The areas inhabited by individuals of the species can be protected through localized conservation measures (e.g., intact riparian zones, springflow protection measures); however, the groundwater that provides water quality and quantity for the species can originate a significant distance from these habitats, and efforts that protect or conserve groundwater may be variable in their success and implementation. Although some of the threats can be adequately addressed, the inherent problems associated with narrow endemics in isolated habitats will always be present. Even with the most effective management and recovery plans in place, the species remains vulnerable to devastating stochastic events such as floods or droughts that could eliminate the species.

Hueco Springs Private Ownership

The primary spring within the Hueco Springs ecosystem is on undeveloped land, but other satellite springs are located within a privately owned campground (78 FR 63109). Of the two major spring orifices, the large spring on the west side stops flowing during severe drought events, and the spring on the east side of River Road typically stops flowing during the driest months each year (Puente 1976, pp. 25-27; Guyton and Associates 1979, p. 46; Ogden et al. 1986, p. 122; Barr 1993, p. 36). These springs are located on private property, and researchers are rarely granted access to this site. Thus, evaluation of habitat conditions, current sub-populations or demographic data, documented changes in land-use activities, or ability to conduct future recovery actions and activities are not achievable at this time.

2.2.1.8 Conservation Measures:

Groundwater Quantity

The Edwards Aquifer Authority (EAA) is charged with protecting terrestrial and aquatic life, domestic and municipal water supplies, the operation of existing industries, and the economic development of the entire Edwards Aquifer (Chapter 626, Laws of the 73rd Texas Legislature, 1993). Aquifer management since these rules were implemented has been successful at controlling groundwater withdrawals to maintain springflows. By EAA estimates, Comal Springs would have likely ceased flowing during the 2014 drought period without current regulations (EAA 2015, p. 62). Currently, these regulations have been effective in managing the Edwards Aquifer and reducing the risk of substantial declines in spring flows at Comal Springs.

Another important conservation measure is implementation of the City of San Antonio’s Edwards Aquifer Protection Program (Stone and Schindel 2002, pp. 38-39; City of San Antonio 2023, pp. 3, 6). In 2000, the voters of San Antonio passed Proposition 3, a $65 million sales tax initiative, to fund the acquisition (i.e., fee-simple and conservation easements) of open space to protect the contributing and recharge zones of the aquifer in Bexar County (Romero 2018, p. 2). Protection of open space has the potential to reduce the impacts of development (e.g., run-off form impervious cover, fertilizer applications, and wastewater) on maintain aquifer recharge (Reilly and Carter 2018, pp. 3-2, 3-6; Romero 2018, pp. 5-6). That program was re-approved in 2005, 2010, and 2015 with additional funds to acquire open space (Reilly and Carter 2018, pp. 1-3–1-5). The effort was later expanded to acquire lands in Medina and Uvalde counties that contain larger portions of the contributing and recharge zones (Romero 2018, pp. 5-6, 8). The dedicated sales tax expired in 2021 with 97,124 hectares (240,000 acres) acquired under the Edwards Aquifer Protection Program (Siglo Group 2022, pp. 51-52). The City of San Antonio recently approved an alternative funding stream to support land acquisitions through the commitment of $100 million over ten years (City of San Antonio 2023, pp. 3, 6).

Groundwater Quality

There are several laws and regulations to protect water quality that apply to the Edwards Aquifer. The Federal Safe Drinking Water Act of 1974, as amended, regulates pollution and sedimentation of public drinking water sources, including the Edwards Aquifer. This legislation mandates enforcement of drinking water standards established by the Environmental Protection Agency. The Texas Commission on Environmental Quality (TCEQ) is responsible for enforcement of these standards in Texas. Under the authority of the Texas Administrative Code (30 TAC § 213), the TCEQ regulates activities having the potential for polluting the Edwards Aquifer and hydrologically connected surface streams through the Edwards Aquifer Protection Program or "Edwards Rules." The Edwards Rules require a number of water-quality protection measures for new development occurring in the recharge zone and portions of the contributing zone of the Edwards Aquifer. The TCEQ also prohibits facilities such as municipal solid waste landfills and waste disposal wells from being built in the recharge or transition zones.

Discharge from non-point residential or agricultural sources is one of the primary sources of pollution in the Edwards Aquifer. Texas has an extensive program for the management and protection of water that operates under State statutes and the Federal Clean Water Act. The Program includes regulatory programs such as the following: Texas Pollutant Discharge Elimination System, Texas Surface Water Quality Standards, and Total Maximum Daily Load Program (under Section 303(d) of the Clean Water Act).

The TCEQ's Texas Pollutant Discharge Elimination System program regulates discharges of pollutants to Texas surface water. Through the Pollutant Discharge Elimination System program, the TCEQ authorizes the discharge of stormwater and non-stormwater to surface waters in Texas associated with storm sewer systems and construction sites, which must meet the requirements of the Edwards Rules.

A watershed protection plan was accepted in 2018 by TCEQ for the Dry Comal Creek and Comal River Watershed by the City of New Braunfels. Dry Comal Creek has not met state water quality standard for bacteria, and the watershed protection plan is intended to address and reduce the elevated bacteria levels through management (TCEQ 2020, p.1).

The EAA has additional regulations (EAA rule 713) that apply to the recharge zone and five miles upgradient of the recharge zone. Much of the contributing zone occurs outside of the EAA's jurisdiction (Edwards Aquifer Habitat Conservation Plan 2020, pp. 1-4, 1-5) and is not subject to these regulations.

New development in the Edwards Aquifer recharge, transition, or contributing zones is reviewed by the TCEQ Edwards Aquifer Protection Program (30 TAC § 213.1). For the contributing zone, the rule covers activities that disturb more than two hectares (five acres) in Medina, Bexar, Comal, Kinney, Uvalde, Hays, Travis, and Williamson counties (30 TAC § 213.20). The contributing zone in Bandera, Kerr, and Kendall counties does not have additional protections under either program.

Several other entities also have measures to protect groundwater from contamination including the EAA's Aboveground Storage Tank Program, Agricultural Secondary Containment Assistance Program, and Abandoned Well Program among others (EAA 2022, entire). The San Antonio Water System implements several water quality protection measures including development regulations (i.e., Aquifer Quality Protection Ordinance No. 81491) for properties over the contributing and recharge zones, review of building permits and master development plans, regulation of underground storage tanks, commercial/industrial compliance, and an abandoned well program (San Antonio Water System 2022, unpaginated).

In addition to these state and federal regulations, a significant number of local regulations to protect water quality were implemented as part of the Edwards Aquifer Recovery Implementation Program Habitat Conservation Plan (EARIP HCP; see sub-section below). Additionally, Texas Water Code (Chapter 36) allows groundwater districts, but not cities, to regulate groundwater, including groundwater quality. However, cities can regulate pollution at the surface that ultimately impacts groundwater quality.

Habitat Conservation Plan

The EARIP HCP was finalized in 2013, amended in 2020, and covers incidental take of these species for groundwater withdrawal, recreation, and other activities through 2028 (EARIP HCP 2020, entire). Permittees to the plan include the EAA, City of San Antonio acting through the San Antonio Water System, City of New Braunfels, City of San Marcos, and Texas State University (National Research Council 2015, pp. 25–26). The EARIP HCP includes activities to minimize and mitigate impacts and contribute to the recovery of the eleven Covered Species and addresses a variety of aquifer management issues, including ensuring springflow during a repeat of the drought of record (Payne et al. 2019, p. 200; EARIP HCP 2020, pp. 4-57–4-59, 4-62–4-66). Long-term commitments to protect listed species in the Edwards Aquifer beyond the HCP and the term of its associated section 10(a)(1)(b) permit are not currently in place. However, a new habitat conservation plan is expected in 2028.

The current EARIP HCP biological goal centers on water quality for the Peck's cave amphipod is: "Not exceed a 10 percent deviation (daily average) from historically recorded water quality conditions (long-term average) within the

Edwards Aquifer as measured issuing from the spring openings at Comal Springs"; there are no habitat biological goals or biological objectives specific to this species.

A captive refugia (operation and maintenance) and associated research is funded by the EARIP HCP through a contract (Contract # 16-822-HCP) with the Service at facilities in San Marcos and Uvalde, Texas (EARIP HCP 2020, p. 5-3). The contract was established to protect species left vulnerable to extirpation throughout a significant portion of their range due to a limited geographic distribution of the population and will preserve the capacity for these species to be re-established in the event of the loss of a sub-population due to a catastrophic event, such as the unexpected loss of springflow or a chemical spill. Research activities expand knowledge on habitat requirements, biology, life histories, and effective reintroduction techniques for the species.

2.2.2 Five-Factor Analysis (threats, conservation measures, and regulatory mechanisms):

2.2.2.1 Present or threatened destruction, modification or curtailment of its habitat or range:

Water Quantity

A primary threat to the habitat of the Peck's cave amphipod is the potential loss of springflows and reduced water quantity underground brought on by groundwater withdrawals from the southern segment of the Edwards Aquifer and other activities. Springflows at Comal and Hueco springs are tied inseparably to water usage for the southern segment of the Edwards Aquifer. Groundwater pumping to meet municipal, industrial, and irrigation uses is a widely recognized threat to the persistence of subsurface and surface groundwater-dependent ecosystems (Danielopol et al. 2003, pp. 109-112; Eamus et al. 2016, pp. 317, 333-335; Mammola et al. 2019, pp. 645-646). Removal of groundwater from an aquifer leads to water level decline, especially if discharge of groundwater significantly exceeds recharge (Theis 1940, pp. 278-280; Alley et al. 2002, pp. 1,986; Foster and Chilton, 2003 pp. 1961-1962). Declining aquifer levels can result in springflow decline or failure, loss of stream and creek base-flow, and/or drying of water-filled caverns (Springer and Stevens 2009, pp. 9-10; Eamus et al. 2016, pp. 316-318, 333-335).

If not replenished through recharge, groundwater discharged through wells and springs is removed from aquifer storage (i.e., total amount of water in aquifer), and with absent or much reduced recharge, persistent groundwater removal would initially lead to decline and/or cessation in springflows (Lindgren et al. 2004, p. 41). Like other karst aquifers, water levels of the Edwards Aquifer fluctuate with recharge (i.e., distribution, amount, and intensity of rainfall) and discharge (i.e., wells or springs) (Petitt and George 1956, p. 49; Buszka 1987, pp. 24-27; Maclay 1995, pp. 48, 52; Worthington et al. 2003, p. 4; Lindgren et al. 2004, pp. 40-41, 45). Prolonged dry periods result in declines in aquifer water levels but rebound rapidly with return of precipitation (Petitt and George 1956, p. 49). Groundwater pumping has exceeded recharge multiple times with water levels rebounding with increased rainfall (Petitt and George 1956, p. 49). The longest period was the drought of record (a three-year period when aquifer recharge was at its lowest recorded level) during the mid-1950s (Arnow 1959, pp. 27-29). At one point, Comal Springs stopped flowing from June 13 through November 3, 1956, during the drought of record (Puente 1976, p. 22).

In the early 1990s, federal litigation (i.e., *Sierra Club v. Secretary of the Interior* [No. MO-91-CA-069] United States District Court for the Western District of Texas) resulted in the creation of the EAA in 1993 by the State of Texas to manage groundwater withdrawals (i.e., by nonexempt wells) from the southern segment and limit Edwards Aquifer pumping authorized through permits (National Research Council 2015, pp. 24-26; Hardberger 2019, pp. 193-194; Payne et al. 2019 p. 199). During the 2007 legislative session, the Texas Legislature increased the annual maximum amount of pumping that could be authorized by permits to 705,551 megaliters (572,000 acre-feet) and directed the EAA to adopt and enforce a "Critical Period Management" plan establishing targeted withdrawal reductions during times of drought to achieve the water, species, and species habitat conservation goals established in the agency's enabling legislation (80th Texas Legislature, 2007, Senate Bill 3). Aquifer management since these rules were implemented have been successful at reducing groundwater withdrawals, but currently do not account for future droughts that may be worse than the drought of record. The Stage V Critical Period Management that currently exists is also tied to the Edwards Aquifer Habitat Conservation Plan (EARIP HCP) but could be subject to change after species recovery.

Springflows have been protected at Comal Springs during recent droughts in the 2000s and 2010s because of groundwater pumping restrictions from the EAA during periods of drought. During the 2008-2009 drought, springflows remained at sufficient levels to maintain resiliency for species (above 80 cubic feet per second [cfs] (2.3 cubic meters per second (m^3/s)]) (USGS station 08169000). By EAA estimates, Comal Springs likely would have gone dry during the 2014 drought without the enforcement of Critical Period Management (EAA 2015, pp. 1, 62).

However, regardless of pumping, Hueco Springs may receive water from the Trinity Aquifer (Otero 2007, pp. 18, 21). Of the two major spring orifices, the large spring on the west side stops flowing during severe drought events and the spring on the east side of River Road typically stops flowing during the driest months each year (Puente 1976, pp. 25-27; Guyton and Associates 1979, p. 46; Ogden et al. 1986, p. 122; Barr 1993, p. 36).

Groundwater will continue to be a source of water in the future as city populations increase. For the four counties within the San Antonio pool (i.e., Hays, Comal, Bexar, Medina), predicted water demands increase 48 percent in the year 2070, insufficient to fulfill using existing supplies (Texas Water Development Board 2021, p. A-2–A-3). The State of Texas and Groundwater Conservation Districts for these counties have identified surface and groundwater management supply strategies that could supplement the forecasted needs of each county but are contingent on funding and infrastructure availability (Texas Water Development Board 2021, entire).

While a repeat drought of record has not occurred, modeling indicates that the Critical Period Management plan during Phase II (current phase) of the EARIP HCP will maintain springflows above 30 cfs (0.85 m^3/s) at Comal Springs and above 45 cfs (1.3 m^3/s). However, the Critical Period Management plan is currently unable to return springflows at either spring system to 80 cfs (2.3 m^3/s) within six months (EARIP HCP 2020, pp. 4-58, 4-66), which is necessary to reduce threats of prolonged lowered springflows on population viability. Future droughts may also be more severe than the drought of record and current aquifer management does not account for this.

Springflows needed to sustain resilient populations is species-specific, and contingent on habitat use and requirements. The biological opinion (Service 2013, p. 129) associated with the EARIP HCP concluded that the issuance of the Incidental Take Permit for the EARIP HCP is not likely to jeopardize the continued existence of the Peck’s cave amphipod or destroy or adversely modify their designated critical habitat. Modeled springflows for conditions during Phase II projected Comal Spring flows to remain at approximately 50 cfs (1.4 m^3/s) during a repeat drought of record (Service 2013, pp. 32, 91, 100). The 27 cfs (0.8 m^3/s) at Comal Springs is greater than the springflows during the drought of record, when springflows ceased for four months in 1956.

Springflows for the Peck’s cave amphipod were not included in the 1995 recovery plan or quantitative delisting criteria. The springflows that affect the Peck’s cave amphipod and its habitat may differ from other surface species. For example, at 30 cfs (0.9 m^3/s) at Comal Springs, runs 2 and 3 do not provide surface habitat for invertebrates (EARIP HCP 2020, pp. 4-97–4-98). The Service determined that 30 cfs (0.9 m^3/s) during a repeat drought of record is not likely to jeopardize the Peck’s cave amphipod (Service 2013 p. 129). Water from Panther Canyon well, seeps along the western shoreline of Landa Lake, and within upwellings near Spring Island are expected to continue to provide habitat during low flow conditions. The Peck’s cave amphipod may be able to use subterranean habitat, but it is possible genetic diversity at some sub-populations may be lost (Service 2013, pp. 100, 104, 110; Lucas et al. 2016, pp. 6, 12).

The Peck's cave amphipod survived the drought of record during the mid-1950s, which resulted in cessation of flow at Comal Springs from June 13 through November 3, 1956, and were not extirpated (Arnow 1959, pp. 27-29; Barr 1993, p. 61). However, given that they are fully aquatic and that no water was present in the springs for a period of several months, they were probably negatively impacted by the unregulated aquifer pumping during this record drought in the 1950s. Hueco Springs is documented to have gone dry in the past and dries yearly in the summer, but due to lack of access we cannot determine the health and numbers of the sub-population within this spring ecosystem (Barr 1993, p. 36; U.S. Geological Survey 2023a, unpaginated).

This cave amphipod is not likely adapted to surviving long periods of drying or stagnation (depending on the duration and severity), especially if the current water management plan for the Edwards Aquifer that accommodates the needs of these invertebrates were to cease.

Water Quality

Water quality at Comal and Hueco springs are influenced by groundwater and surface water. Both systems depend on groundwater flow from the southern segment of the Edwards Aquifer. This segment of the aquifer is fed by many stream systems that enter the aquifer through recharge features.

The Edwards Aquifer is vulnerable to contamination because the limestone and carbonate rocks are highly permeable and exposed at the surface in the recharge zone (Clark 2000, pp. 1-2, 8-9; Burri et al. 2019, p. 150). Contaminants, commonly linked to urban and suburban activities such as residential and commercial development, industrial operations, transportation infrastructure, and waste disposal, tend to accumulate in higher concentrations within the shallow areas of recharge zones, especially in regions characterized by urban land uses (Wilson 2011, pp. 1-2; Lin and Gong 2016, pp. 384-385; Opsahl et al. 2018, p. 58).

There are currently no established groundwater quality standards for subterranean ecosystems, and the concentrations of pollutants that could harm subterranean species remain unclear (Hinsby et al. 2008, p. 10; Manenti et al. 2021, p. 2). However, subterranean fauna are likely to exhibit greater vulnerability to contaminants and a longer recovery period from stochastic events compared to surface fauna because of their inherent limitations, including a lack of adaptations to pollutants, isolation within their habitat, and restricted dispersal abilities, all of which render them sensitive to environmental disturbances (Hose 2005, p. 961; Di Lorenzo et al. 2019, pp. 293–294, 300; Hose et al. 2022, p. 2206).

Although water quality in the Edwards Aquifer is generally good, several studies have detected contaminants in groundwater from the southern segment including nitrates, herbicides, pesticides, and polycyclic aromatic hydrocarbons, among many others (Fahlquist and Ardis 2004, pp. 7-8, 10; Johnson et al. 2009, pp. 10-13, 23-26, 31-35; Musgrove et al. 2014, pp. 67, 69-71; Opsahl et al. 2018, p. 58; Opsahl et al. 2020, pp. 17-30). For example, contaminants have exceeded public drinking water standards in springwater and surface water recharging the aquifer, including antimony, arsenic, lead, lithium, and tetrachloroethene (Johnson et al. 2009, p. 45). However, groundwater contamination has not been shown to be widespread or with large numbers of substances present in concentrations that exceed drinking water standards (Bush et al. 2000, pp. 1-2, 14-21; Fahlquist and Ardis 2004, pp. 7-8, 10; Johnson et al. 2009, 44, 47; Opsahl et al. 2018, p. 58; Opsahl et al. 2020, pp. 17-30; EARIP HCP 2020, pp. 3-40-3-42).

Some of the sources of water quality degradation include impervious cover and stormwater runoff, construction activities, recharge from irrigation return flow (i.e., water that is not lost from evapotranspiration on laws or to stream runoff), wastewater discharge, transportation infrastructure, and hazardous materials spills resulting from development within the watersheds that contribute flows to subterranean habitats (Passarello et al. 2012, pp. 29–34; Lapworth et al. 2012, entire). Hueco Springs is situated adjacent to River Road, a popular route for recreational activities along the Guadalupe River. Due to its high recreational traffic, there is a potential vulnerability to road runoff and spills associated with the frequent passage of vehicles (62 FR 66295).

Forested land with limited human disturbances contributes to high-quality recharge (Dudley and Stolten, 2003, pp. 11, 58; Shah et al. 2022, p. 120396), while rural and exurban land uses contribute to groundwater contamination from leaking sewage, refuse dumping, and dead livestock (Sui et al. 2015, p. 21; Katz 2019, p. 565; EARIP HCP 2020, pp. 5-43). Septic systems are a likely source of nutrients (EARIP HCP 2020, p. 5-43; Sui et al. 2015, p. 21). Once a source of pollution enters groundwater, it can be difficult if not impossible to track, intercept, and remediate because of karst conduit complexity (Humphreys 2011, p. 297). Since water quality in the Edwards Aquifer is generally good, this indicates that local sources of water pollution can disproportionately affect water quality in portions of the aquifer.

Oil and gas transmission pipelines are another potential source of hazardous material spills on the contributing and recharge zones of the aquifer. The "development and production of oil, gas, or a geothermal resource within the jurisdiction of the Texas Railroad Commission" are not considered regulated activities "having the potential for polluting the Edwards Aquifer and hydrologically connected surface water in order to protect existing and potential uses of groundwater and maintain Texas Surface Water Quality Standards" (Texas Natural Resource Conservation Commission 1996 p. 1). Consequently, the construction and maintenance of these pipelines are not subject to guidance mitigating impacts to karst features such as voids, and development of these pipelines are not subject to the Edwards Aquifer rules (Texas Natural Resource Conservation Commission 1996, entire).

Abandoned groundwater wells are a source of potential contamination from shallow groundwater into subsurface habitat. Shallower wells (< 300 m [< 984 ft]) are less likely than deeper wells to intercept older groundwater that received cumulative, diluted inputs of pollutants across the aquifer and therefore are more likely to intercept anthropogenic contaminants coming directly from the surface than deeper wells (Musgrove et al. 2014, pp. 69, 73). The EAA funds a needs-based abandoned well closure assistance program to assist well owners with proper well plugging in cooperation with San Antonio Water System to locate and plug abandoned wells (EAA 2021, pp. 50-53). Likewise, former oil wells require maintenance decades after plugging (cement plugs in a steel pipe) and can blowout underground and break free under artesian pressure if not properly maintained (Gold 2022, entire).

Nitrogen is highly soluble and a threat to groundwater quality and a stressor to subterranean taxa (Castaño-Sánchez et al. 2020, pp. 6, 11; Banerjee et al. 2023, pp. 3–6). Panther Canyon well (State well number 6823302), recorded nitrate (2 mg/L) present in 2003 (Texas Water Development Board 2023, unpaginated). Nitrates and orthophosphate consistently emerge from spring run 1 at Comal Springs, and they are typically present at low concentrations (2 mg/L) (U.S. Geological Survey 2023b, unpaginated). The current drought has significantly decreased flow and thus dilution of contaminants are slowed at Comal Springs and recent data resulted in 3 mg/L of nitrate measured at spring run 2 at Comal Springs (West 2023, unpaginated). While safe for humans, it is unknown what effect these elevated nutrients will have over time within the aquifer food web and if conditions would become more favorable for surface species to colonize further underground (Notenboom et al. 1994, pp. 482–484, 490; Opsahl et al. 2018, p. 3). The cave amphipod’s environmental tolerances are unknown, hindering quantitative assessments of this stressors' impact on its sub-populations. Additionally, there are no established groundwater quality standards for subterranean ecosystems, making pollutant concentrations' harm unclear.

Volatile organic compounds have been detected at these spring ecosystems but are rare (Johnson and Schindel 2014, p. 21). There is one documented diesel spill (i.e., naphthalene) that occurred in 2000 at spring run 7 at Comal Springs and emerged at Hueco Springs, further validating their groundwater connection (Ogden et al. 1986, p. 126; Gibson et al. 2008, p. 75). It is unknown what effect this had on the subterranean community.

Urban and agricultural land uses dominate the artesian zone in the southern segment. Low- to high-density urban development occurs across much of the former, while agriculture dominates the latter county. Land use across the southern segment of the Edwards Aquifer plays a major role in groundwater and surface water quality. The presence of agriculture, residential and commercial developments, industrial facilities, military installations, and transportation infrastructure are correlated with increased presence of many contaminants (Bush et al. 2000, pp. 6-9; Fahlquist and Ardis 2004, p. 7; Johnson et al. 2009, p. 46; Wilson 2011, pp. 1-2; Musgrove et al. 2014, pp. 69-71; Opsahl et al. 2018, p. 58; Opsahl et al. 2020, pp. 17-30).

To examine projected land-use changes in the urban centers intersecting Edwards Aquifer groundwater, we used the EPA's (2019, unpaginated) Integrated Climate and Land-Use Scenarios. These outputs produce spatially explicit projections of population and land-use that are based on the Intergovernmental Panel on Climate Change's Special Report on Emissions Scenarios. The combination of SSP5-RCP8.5 illustrates a higher population growth and higher emissions, and a faster rate of human population growth consistent with the Texas Demographic Center population projections for Bexar County and the San Antonio-New Braunfels Metropolitan Area (Environmental Protection Agency 2017, pp. 34-35, 46; Texas Demographic Center 2022, unpaginated). Within the Edwards Aquifer artesian, recharge, and contributing zones (543,498 ha^2 [134,3014 ac^2]), developed land-use classes are projected to grow from 21 percent in 2020 to 27 percent developed by 2050. When examining delineated areas at a finer scale around Comal Springs using the Integrated Climate and Land-Use Scenarios, population is also projected to increase in development from 66-82 percent developed. These areas may be important to assess more immediate impacts from groundwater contamination. Alternatively, the area around Hueco Springs is not projected to have noticeable changes in development use classes.

Based on the Integrated Climate and Land-Use Scenario results, projections of developed land-uses and population growth will continue to expand outward outside of the major metropolitan areas, San Antonio and Austin, Texas. Over time, these alterations have the potential to affect recharge rates, leading to deteriorating groundwater quality as a result of heightened runoff from impervious surfaces in suburban and urban areas or septic systems that are poorly managed and prone to leakage in exurban areas (Berube et al. 2006, pp. 10, 38; Barkfield 2022, p. 2).

The U.S. Census Bureau (2020, unpaginated) ranked several of the counties in the recharge and contributing zones of Comal Springs among the fastest growing in the United States from April 2010 to July 2019: Hays County was the second fasting growing county with a 46.5 percent population increase, Comal County the fourth fastest growing county with a 43.9 percent population increase, and Kendall County the fifth fastest growing county with a 42.1 percent population increase. Since 2000, these three counties have doubled in population and have seen substantial associated development. Projections indicate that the human population of Bexar, Comal, Hays, and Kendall counties will continue to increase substantially over the next three decades.

Conversion of natural habitat to urban, suburban, and exurban development is likely to accompany this population growth. Under a high human population growth scenario, land use projections suggest that large areas west and north of Bexar County will be converted to increasingly more urbanized land-use classes by 2100 (EPA 2019, unpaginated). Much of the exurban and suburban development is postulated to occur outside of municipal boundaries in unincorporated areas of counties where land use regulations (e.g., restrictions on impervious cover) are non-existent (Siglo Group 2022, pp. 13-14). Run-off from existing and expanded impervious cover in sensitive areas of the aquifer could affect groundwater quality over time. New contaminant sources are expected to be added to the region as increased human populations and expanded development continues; many existing contaminant sources will persist.

Land-use changes, particularly increases in impervious cover, are known stressors to aquatic systems and are difficult to predict, model, and remediate (Sharp 2010, p. 3; Coles et al. 2012, p. 65). Future development in the recharge and contributing zones are likely to decrease water quality because of the increased risk of contamination entering the aquifer. Additionally, nitrate runoff from surface water recharge leads to increased nitrate concentrations in the aquifer, and concentrations over 1 milligram per liter (mg/L) are indicative of anthropogenic inputs, which have been recorded historically at Comal Springs and have doubled over the last 70 years (median concentration 2 mg/L) (Dubrovsky et al. 2010, p. 79; Musgrove et al. 2016, pp. 462, 465, 467; Castaño-Sánchez et al. 2020, p. 6). These changes in water quality in streams and groundwater correspond with increases in impervious cover over a watershed (Kaushal et al. 2005, p. 13518; Baker et al. 2019, pp. 6494–6495; Castaño-Sánchez et al. 2020, p, 6).). These water quality parameter changes may be a long-term indication of urbanization that has already occurred across the recharge zone.

A review of research studies found that impacts to aquatic species are seen with impervious cover of 10 percent or more (Center for Watershed Protection 2003, p. 97). Although the studies were focused on stream systems, we assume that shallow groundwater habitats would have similar impacts because shallow groundwater ultimately flows into streams through discharge features. While physical parameters may be different (e.g., higher oxygen, lower temperatures, higher conductivity) in the shallow groundwater, pollutants entering both systems would be the same.

The EAA does not have explicit impervious cover limits in the recharge zone, with the intent that structural best management practices will protect water quality (Greater Edwards Aquifer Alliance 2010, p. 3). The TCEQ shares responsibility in protecting the Edwards Aquifer through impervious cover limits through a construction permit review process for development proposals of more than 20 percent impervious cover that includes structural best management practices (30 TAC § 213). Additionally, Comal County has goals to minimize impervious cover within the city of New Braunfels to limits of 26 percent per parcel (Design Workshop, Inc. 2012, pp. 4–5).

These percentages are all higher than 10 percent, and each project approval does not account for the cumulative impact of combined impervious cover amounts within each county. Likewise, most lands over the contributing zone are not managed with land use regulations (e.g., impervious cover restrictions) (Siglo Group 2022, pp. 13–14).

Habitat Disturbance- Flooding

Surface habitat modification can occur as the result of flooding. Flash flooding is common throughout the Edwards Plateau (Woodruff and Wilding 2008, pp. 614-616). However, channel modification and the elimination of riparian zones can increase the severity of flooding (Schoof 1980, p. 697). Depending on the severity of floods, they can either deposit or increase suspended sediment loads over species habitat or scour substrate and vegetation from species habitat under high velocities (Griffin 2006, pp. 57-58, 61, 64; BIO-WEST, Inc. 2016, p. 26; BIO-WEST, Inc. 2019a, pp. 14, 17; Schwartz et al. 2020, pp. 12). During wet periods, flows at Hueco Springs are highly responsive to storm events. These events increase flows and dilutes higher quality springflows with greater proportions of local recharge, which may include increased loads of contaminants (Ogden et al. 1986, pp. 118, 125, 127; Musgrove and Crow 2012, pp. 53, 56-57). It is possible that individuals of species may also be washed away in floods, though this has not been studied for the Peck's cave amphipod. Floods have deposited finer sediments (e.g., silt) over invertebrate surface habitat within the Spring Island area within Comal Springs, reducing springflow and quality of habitat (BIO-WEST, Inc. 2002, p. 11; Gibson 2022, pers. comm.).

2.2.2.2 Overutilization for commercial, recreational, scientific, or educational purposes:

Peck's cave amphipod specimens are collected for scientific study and two refugia populations. Such collections which have not been documented to negatively impact total wild population numbers. At present, this species is not recognized for their commercial worth, and there is no evidence of overexploitation, making overutilization insignificant as a threat.

2.2.2.3 Disease or predation:

Disease and parasitism is rarely observed for the Peck's cave amphipod. A nematode (*Amphibiocapillaria texensis*) and an acanthocephalan (*Dendronucleata americana*) parasite have been observed in Texas blind and San Marcos salamanders (*Eurycea rathbuni*; *E. nana*) and *Hyalella* amphipod species (likely as an intermediate host), which other *Stygobromus* taxa may serve as a possible intermediate host within the parasites' life cycle (Moravec and Huffman 2000, entire; Worsham and Gibson 2022, pers. comm.).

A facultative ectoparasite (e.g., rotifers, Phylum Rotifera) can be found on the gills of other amphipod taxa of this aquifer ecosystem but has not been observed in Peck's cave amphipods and needs further investigation (Worsham and Gibson 2022, pers. comm.).

Seen in other members of the subphylum Crustacea (e.g., prawn, crab, and lobster juveniles and adults), a rickettsia-like bacterium causes milky haemolymph disease and can be treated (Nunan et al. 2010, p. 105, 111). This syndrome has been identified in other *Stygobromus* sp., softening exoskeletons and killing the individual (Worsham and Gibson 2022, pers. comm.). It is unknown if the Peck's cave amphipod is affected by this disease and what extent contact with other infected freshwater crustaceans at the surface have on this species.

The amount of predation that occurs in the wild has not been examined for this species. Blind, fragile subterranean species such as the Peck's cave amphipod may be more susceptible to predation once they enter surface waters (Barr 1993, pp. 63-64). Fishes compete for prey expelled from the aquifer at discharge features (e.g., spring openings). Researchers have seen Mexican tetras (*Astyanax mexicanus*), sunfish (*Lepomis* sp.), and mosquitofish (*Gambusia* sp.) congregating at spring openings waiting for the driftnet to be removed and consume the bycatch, including subterranean invertebrates (BIO-WEST, Inc. 2003, p. 42). Macroinvertebrates are a part of the food chain, and it is assumed any number of individuals removed from the Peck's cave amphipod sub-populations through typical levels of predation are negligible.

2.2.2.4 Inadequacy of existing regulatory mechanisms:

Under this factor, we examine the stressors identified within the other factors as ameliorated or exacerbated by any existing regulatory mechanisms or conservation efforts. Section 4(b)(1)(A) of the ESA requires that the USFWS consider "those efforts, if any, being made by any State or foreign nation, or any political subdivision of a State or foreign nation, to protect such species…". In relation to Factor D under the ESA, we interpret this language to require the USFWS to consider relevant Federal, State, and Tribal laws, regulations, and other such binding legal mechanisms that may ameliorate or exacerbate any of the threats we describe in threat analyses under the other four factors or otherwise enhance the species' conservation. Our consideration of these mechanisms is described in detail within each of the threats or stressors to the species (see discussion under the other Factors). Much of the information under Section 2.2.2.1 should also be considered as relevant here because it is often the inadequacy of existing regulations that contributes to habitat loss and degradation for these species.

The recharge and contributing zones to the Edwards Aquifer continue to experience rapid human population growth and conversion of natural habitat to development, which continues to threaten water quality. Much of the contributing zone is not under the same regulations as the recharge zone to protect water quality, even though much of the water that recharges the aquifer originates in the contributing zone. Regulatory mechanisms that protect water in the Edwards Aquifer are crucial to the future survival of the Peck's cave amphipod. Federal, State, and local laws and regulations have improved water quality and quantity protection but could be insufficient to prevent ongoing impacts to the species and their habitats from water quality degradation, reduction in water quantity, and surface disturbance of spring sites, and are unlikely to prevent further impacts to the species in the future. Knowledge of the source, accumulation, and transport of these compounds in the aquifer are lacking and investigations into their effects on the habitat quality are necessary for the recovery of the Peck's cave amphipod and for sustainable use of the aquifer (Danielopol et al. 2004, pp. 187-188; Opsahl et al. 2018, p. 2).

Under Texas Parks and Wildlife Code (Chapter 68) and TAC (31 TAC § 65.171-65.176), the Texas Parks and Wildlife Department is authorized to add species to the agency's List of State Threatened and Endangered Nongame Species and List of Endangered, Threatened, and Protected Native Plants. The seven species in this plan are also state listed. The Texas Parks and Wildlife Department prohibits the taking, possession, transportation, or sale of any animal species that are state listed as threatened or endangered. State law prohibit commerce in threatened and endangered plants, and also prohibits collection of listed plant species from public land without a permit. However, prosecutions for these prohibited actions are rare and the burden of proof to prosecute is high, which can result in unauthorized take of state listed species. In addition, it is likely that at the time of recovery they would no longer be state listed. Because the Peck's cave amphipod is conservation reliant, it would be expected that delisting would increase threats identified in the listing determination, unless there are other mechanisms to continue conservation efforts.

While the EAA was granted regulatory authority by the Texas Legislature, there have been several legal challenges to the EAA permitting program. For example, in court cases *Edwards Aquifer Authority v. Day* (2012, Supreme Court of Texas No. 08-0964) and *Edwards Aquifer Authority v. Bragg* (2013, Court of Appeals of Texas No. 04-11-00018-CV), courts awarded landowners compensation for groundwater permits that were denied by the EAA due to lack of historical usage. The ruling for *Edwards Aquifer Authority v. Day* by the Texas Supreme Court argued that there was no reason to treat groundwater differently than oil and gas and recognized groundwater as real property. In both cases, landowners owned the land prior to enactment of new groundwater pumping regulations. There remains a lack of clarity with Texas groundwater law that results in ongoing legal challenges regarding groundwater regulation, and these could impact the EAA’s ability to regulate the aquifer in the future.

The EAA manages and issues permits for groundwater withdrawals within the Edwards Aquifer through conservation and drought management. The EAA’s jurisdiction is limited to the Edwards Aquifer in Uvalde, Medina, Bexar, and portions of Comal, Guadalupe, Hays, and Caldwell counties. The contributing zone in Bandera, Kerr, and Kendall counties do not have additional protections under either program. Thus, the EAA’s its water quality regulations do not protect most of the contributing zone, which may ultimately reduce the water quality of the Edwards Aquifer.

As described above, TCEQ regulates activities that have the potential to pollute the Edwards Aquifer and hydrologically connected surface streams under the same Edwards Aquifer Protection Program or “Edwards Rules” and for the same counties. This means areas of the contributing zone do not have additional protections that could affect the amount and quality of recharge that enters the Edwards Aquifer, resulting in lower water quality protection for the aquifer and the Comal and Hueco ecosystems.

Likewise, this agency does not address development or other land use, impervious cover limitations, some nonpoint source pollution, or application of fertilizers and pesticides over the recharge zone (30 TAC § 213.31). Changes to how surface water and the Trinity Aquifer are managed are likely to change the amount that can be sustainably pumped from the Edwards Aquifer during drought conditions. For example, the Hays-Trinity Groundwater Conservation District also manages groundwater that influences the water at San Marcos Springs.

2.2.2.5 Other natural or manmade factors affecting its continued existence:

Global climate change is already affecting many regions’ biodiversity, with stressors driven by increasing temperatures and extreme climatic events and will continue to in the near-term (Intergovernmental Panel on Climate Change 2023, pp. 5, 15). Over the last 115 years, the global averaged surface air temperature has increased by 1.0°C (1.8°F) with recent decades being the warmest in 1,500 years (Vose et al. 2017, pp. 186, 188). With the highly karstic permeability of the Edwards Aquifer, climate change and variability strongly influence this vulnerable aquifer that relies heavily on rainfall for recharge (Mace and Wade 200,8 p. 659; Taylor et al. 2013, p. 312; Ding and McCarl 2019, p. 11; Nielsen-Gammon et al. 2020, p. 9). The Fourth U.S. National Climate Assessment (U.S. Global Change Research Program 2018, pp. 1,002-1,003) presents the Edwards Aquifer as a case study in vulnerability to climate change, citing the shallow karst aquifer as especially sensitive to climate change, and the regional population growth and development as exacerbating the effects of decreased water supply during droughts. While average rainfall is not projected to change significantly in central Texas, the distribution of precipitation is anticipated to change with more extreme droughts and extreme rain events (Geos Institute 2016, pp. 14-15).

Increasing temperatures will also create drier conditions due to increased evapotranspiration (Loáiciga and Schofield 2019, p. 224). Extreme droughts in Texas are more likely than they were 40-50 years ago (Rupp et al. 2012, p. 1,054; Nielsen-Gammon et al. 2020, entire). A recent study predicts megadroughts in Texas, more severe than have been seen for the past thousand years, that will occur before 2100 (Nielsen-Gammon et al. 2020, entire). Droughts worse than the drought of record occurred since the 1600s and are not uncommon in the region (Mauldin 2003, entire; Cleaveland et al. 2011, entire). It is not possible to ensure that there will be adequate flow to these springs without planning for more extreme droughts than the drought of record (Loáiciga and Schofield 2019, p. 236; Mace 2019, p. 212). The sustainable water yield for the Edwards Aquifer will decrease in a dry climate (EARIP HCP 2020, pp. 3-12, 3-31, 3-43; Loáiciga and Schofield 2019, pp. 223, 235-236) while human demand for groundwater will increase (EARIP HCP 2020, pp. 3-10-3-11), making it more challenging to balance groundwater use for human needs and ecosystem function. In 2010, Texas set a record for lowest rainfall (March–May; June–August) and with similar conditions persisting until 2013 (Nielsen-Gammon 2012, p. 59; National Research Council 2015, p. 168). Heavy rainfall leading to floods may also become more common from extreme precipitation events and may result in increased habitat disturbance due to movement of materials and scouring.

Air temperature in Texas has risen 1°C (2°F) since the early 1900s (Geos Institute 2016, p. 4). Future air temperature changes will depend on the amount of future greenhouse gas emissions (U.S. Global Change Research Program 2018, p. 995). Based on current projections of greenhouse gas emissions, air temperature is projected to increase 2.0-2.8°C (3.6-5.1°F) by 2050, and 2.4–4.7°C (4.4–8.4°F) by 2100 for the southern Great Plains (U.S. Global Change Research Program 2018, p. 995). Projections expect a greater rise in air temperature by 2100, 2.7–5.6°C (5–10°F) (Sharif 2018, p. 4). Studies have not explicitly addressed groundwater temperature increases for the Edwards Aquifer. Based on other research into changes in groundwater temperature, it is reasonable to expect that groundwater temperature will increase as air temperature increases, with a possible lag in groundwater temperature increase (Mahler and Bourgeais 2013, p. 295). Groundwater temperature also increases with urbanization and vegetation removal (Benz et al. 2017, entire). This could further increase groundwater temperatures as more development occurs. Groundwater temperature typically increases with depth due to geothermal heat flow, although this also varies locally with other variables such as vertical groundwater flow (Bense and Kurylyk 2017, pp. 1, 8). This suggests that deeper water would not provide a long-term buffer to increasing temperatures.

Some subterranean-adapted species would likely be incapable of adapting to modified temperatures in the medium to long-term and less capable, due to restricted dispersal capabilities, to flee rising temperature conditions than surface-adapted species (Culver and Pipan 2009, pp. 207–208; Taylor et al. 2013, pp. 324–325; Mammola et al. 2019, p. 646). Subterranean-adaptations in ectothermic animals allow for small fluctuations in temperature, but increased temperatures due to climate change can affect subterranean diversity by altering mobilization of contaminants (i.e., change in recharge rates through the unsaturated zone) and disruption to biogeochemical processes (e.g., carbon and nitrogen cycle) (Kløve et al. 2014, p. 263; Castaño-Sánchez et al. 2020, p. 7). Water quality at the subsurface and surface is also likely to decrease with increased water temperature. For example, as dissolved oxygen decreases and microbial activity increases (Bates et al. 2008, p. 43). Therefore, the adaptive capacity ectothermic animals have to environmental changes is presumed to be low.

For instance, in periods of low rainfall, two main spring outlets at Hueco Springs cease their flow, particularly during droughts and the driest months annually (Puente 1976, pp. 25-27; Guyton and Associates 1979, p. 46; Ogden et al. 1986, p. 122; Barr 1993, p. 36; Otero 2007, pp. 18, 21). However, little is known how this affects the sub-population of Peck's cave amphipods because this site is located on private property, and researchers are rarely granted access.

Surface water temperature will also increase during warm months. Data from the EAA indicates greater temperature fluctuations downstream from the springs due to increased exposure time to ambient temperatures and runoff from rain events (BIO-WEST, Inc. 2019b, p. 20). Low spring discharge is also a mechanism that increases the water's exposure time to ambient temperature. Thus, both future droughts and increased ambient temperature are likely to increase the surface water temperature. Continuous temperature data for the springs began in 2000, and groundwater temperature at Comal Springs is relatively constant (BIO-WEST, Inc. 2019b, p. 20). Continuous water temperature monitoring in the Comal River should indicate whether water temperatures rise in the future.

There is currently no information on whether increased temperatures can affect different life stages or reproduction of the Peck's cave amphipod, or how quickly water temperature will change in their habitat into the future. For ectothermic animals (e.g., macroinvertebrates), overall vulnerability to climate change will depend on thermal sensitivity and how quickly their buffered environment changes (Pallarés et al. 2021, p. 487; Delić et al. 2022, p. 2). Species with similar tolerances and adaptive traits have no opportunity to migrate and are unlikely to successfully relocated due to its specific habitat requirements (Kløve et al. 2014, p. 263; Castaño-Sánchez et al. 2020, p. 7; Simčič and Sket 2021, entire; Becher et al. 2022, pp. 4–5). We are uncertain if this species could flee from undesirable conditions caused by catastrophic drought in their habitat. There could be voids that become de-watered, and we assume the species make attempts to follow the water down into the aquifer as drying occurs.

An assessment by U.S. Geological Survey evaluated the projected future vulnerability through 2050 of the Peck's cave amphipod and rated it as moderately vulnerable to climate change (Stamm et al. 2015, pp. 1, 40, 42, 47). Moderately vulnerable is defined as "abundance and/or range extent within geographical area assessed likely to decrease by 2050". There is currently no information indicating whether increased temperatures would affect different life stages or reproduction of the Peck's cave amphipod, or how quickly groundwater temperature will change in the Edwards Aquifer in response to climate change at the surface. Without more information, it is unknown to what extent these temporally delayed changes to the aquifer would have on this cave amphipod and if they would have sufficient time and have appropriate traits to adapt. These are important factors that require more research globally to fully understand vulnerability of these aquifer ecosystems and their subterranean communities (Mammola et al. 2019, pp. 646–647; Hose et al. 2022, entire).

2.3 Synthesis

There are currently two sub-populations of the Peck's cave amphipod in Texas. There is no recovery plan or species status assessment to fully evaluate species viability published at this time. However, available demographic data, captive refugia research, and the five-factor threats analysis (Section 2.2.2) are collectively not indicative for a change in listing status recommendation for the Peck's cave amphipod. Peck's cave amphipod sub-populations rely on continuous management and protective measures to preserve habitat, prevent silt accumulation and manage groundwater pumping for optimal springflow, supply terrestrial organic matter for the food web, and maintain sufficient water availability and quality for overall ecosystem health. In conclusion, it is our recommendation that a change in classification is not warranted at this time.

3.0 RESULTS

3.1 Recommended Classification:

No change is needed

3.2 New Recovery Priority Number (indicate if no change; see 48 FR 43098):

No Change Recommended; see 48 FR 43098, September 21, 1983 & 48 FR 51985, November 15, 1983 - Correction)

Brief Rationale:

The primary stressors are the loss of spring flows and decreases in subsurface habitat due to drawdown of the Edwards Aquifer and reductions in water quality from development and land-use changes. Research suggests that contamination of groundwater has not been historically widespread, is at relatively low concentrations currently, and the subterranean ecosystems do not exhibit significant signs of degradation (Hutchins 2018, pp. 481–482). Current conservation, flow protection, and water quantity optimization measures in place have been effective in meeting biological objectives for the Covered Species, including the Peck's cave amphipod, under which the EARIP HCP and regulations are reducing groundwater withdrawal pressure (National Research Council 2018, p. 109). Given the projected increases in development and climate change-induced droughts in South Central Texas, the impact on groundwater habitat quality and aquifer recharge into the future remains uncertain (Loáiciga and Schofield 2019, p. 224; National Oceanic and Atmospheric Administration 2022, unpaginated). The sustainable water output for the Edwards Aquifer could decrease in a dry climate while human demand for groundwater would increase, making it more challenging to balance groundwater use for human needs and ecosystem function, and thus, the Peck's cave amphipod's viability (Loáiciga and Schofield 2019, pp. 223, 235–236; EARIP HCP 2020, pp. 3-10–3-11, 3-12, 3-31, 3-43; Nielsen-Gammon et al. 2020, pp. 9–10).

In terms of viability, the Peck's cave amphipod occupies a restricted range of two sub-populations as a narrow endemic species (redundancy) only occurring in the Edwards Aquifer and associated spring ecosystems (representation) and are highly susceptible to extinction from perturbations that would affect water quantity and quality in the Edwards Aquifer and ongoing management is needed to maintain resiliency. Further, the absence of data to inform how these threats directly impact Peck's cave amphipod sub-populations precludes a more detailed assessment of these impacts. Thus, our analysis does not warrant a change in recommended classification or recovery priority number.

Therefore, we recommend the Peck's cave amphipod retain its classification as endangered due to its conservation-reliant status.

3.3 Listing and Reclassification Priority Number, if reclassification is recommended (see 48 FR 43098):

Reclassification (from Threatened to Endangered) Priority Number:
Reclassification (from Endangered to Threatened) Priority Number:
Delisting (Removal from list regardless of current classification) Priority Number:

Brief Rationale:

Not Applicable

4.0 RECOMMENDATIONS FOR FUTURE ACTIONS

- If possible given workload and priorities, a Species Status Assessment could be conducted to guide the development of a revised recovery plan.
- Continue to plan and implement regular surveys that monitor Peck's cave amphipod occurrence, habitat condition, groundwater and surface water quality, as well as any

potential threat to the Peck's cave amphipod from disease and parasitism (Section 2.2.2.3).

- Status survey at the Hueco Springs ecosystem in Comal County to assess species persistence, abundance, and habitat health of this sub-population, in addition to improving habitat conditions and landowner cooperation. Currently, the status of this sub-population is unknown.
- Continue to investigate the extent of groundwater watersheds between the Comal and Hueco ecosystems, including eDNA or physical sampling in between the sites when available, in order to get a more accurate representation of drainage areas and habitat connectivity and gene flow.
- Incorporate habitat-centered biological goals and objectives during EARIP HCP renewal process to promote protection of suitable habitat quality and quantity and species resiliency.
- Establish conservation easements or fund land purchases within the contributing and recharge zones of the Edwards Aquifer for the benefit of the Peck's cave amphipod and to ensure adequate springflow is sustained through droughts. Additionally, a site-prioritization tool could be developed to support decision making about strategic land acquisitions.
- Research to reduce sources of nitrate into the Comal ecosystem through coordination with agencies, public education, and other non-governmental organizations.
- To the extent possible, prevent or reduce increases in impervious surfaces or clearing of forest within the recharge areas supporting the species.
- Continuation of the captive propagation research:
    - Conduct ongoing research to enhance captive propagation techniques.
    - Develop the capacity to produce offspring on-demand, anticipating standard operating procedures to inform action for potential catastrophic events or extirpation in the wild.
    - Formulate a comprehensive reintroduction plan based on research findings, ensuring the ability to replenish populations as needed.
- Continue water quantity and quality monitoring at accessible spring and well sites within and areas that recharge the Comal ecosystem.
- Continue to measure genetic variability among sub-populations of the Peck's cave amphipod in order to evaluate gene flow, population structure, and estimate population sizes. These data can inform captive husbandry practices to preserve genetic diversity in the refugia population and future recovery plan implementation.

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U.S. FISH AND WILDLIFE SERVICE

5-YEAR REVIEW of the Peck's cave amphipod

Current Classification: Endangered

Recommendation resulting from the 5-Year Review:

No change needed

Appropriate Listing/Reclassification Priority Number, if applicable: Not applicable

FIELD OFFICE APPROVAL:

Lead Field Supervisor, Fish and Wildlife Service, [Austin Ecological Services Field Office]

Approve KAREN MYERS Digitally signed by KAREN MYERS Date: 2024.03.29 12:06:18 -05'00'