

STATUS UPDATE FOR BRISTLY CAVE CRAYFISH, *CAMBARUS SETOSUS*
(DECAPODA: CAMBARIDAE), AND RANGE EXTENSION INTO ARKANSAS

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ABSTRACT—This study updates the status, expands the range, and summarizes conservation activities to date for the bristly cave crayfish, *Cambarus setosus*. A new state record for this crayfish is reported after its discovery at 2 sites in 2 counties (Benton and Independence counties, Arkansas). This makes *C. setosus* the fifty-eighth crayfish species known from Arkansas. This is also the first record of *C. setosus* from the Salem Plateau of the Ozark Plateaus ecoregion; it was known previously only from the Springfield Plateau of Missouri. The range of *C. setosus* in Missouri includes the counties of Barry, Christian, Dade, Greene, Jasper, Lawrence, McDonald, Newton, and Stone; it is not presently known from Oklahoma, as previously reported. *Cambarus setosus* is probably a species complex with genetic variability at the subspecific level, especially in southwestern Missouri. *Cambarus setosus* is now known from 50 sites, but only 164 individuals (tallied from the most recent census at each site). This crayfish has been impacted by both habitat degradation and scientific study. Conservation organizations have begun preserving cave entrances, but conservation activity is lacking in subterranean stream recharge zones. Increased protection is recommended, including federal listing under the US Endangered Species Act.

RESUMEN—Este estudio actualiza el estado, expande la distribución, y resume acciones de conservación que hasta la fecha se han llevado a cabo a favor del cangrejo *Cambarus setosus*. Se reporta un nuevo registro estatal para este cangrejo luego de su descubrimiento en dos localidades en dos condados (los condados Benton e Independence, Arkansas). Esto hace a *C. setosus* la 58ava especie de cangrejo conocida de Arkansas. Es además el primer registro de *C. setosus* en el Salem Plateau en la ecoregión del Ozark Plateaus, conocido previamente sólo del Springfield Plateau de Missouri. La distribución de *C. setosus* en Missouri incluye los condados de Barry, Christian, Dade, Greene, Jasper, Lawrence, McDonald, Newton, y Stone; no se conoce en Oklahoma, como anteriormente fue reportado. *Cambarus setosus* es un complejo de especies con variabilidad genética a nivel subespecífica, especialmente en el suroeste de Missouri. *Cambarus setosus* es conocido en 50 localidades, pero solamente 164 individuos (contados en los censos más recientes en cada localidad). Este cangrejo ha sido impactado tanto por degradación del hábitat como por estudio científico. Organizaciones de conservación han comenzado la protección de entradas de cuevas, pero faltan estos esfuerzos en zonas de reabastecimiento de corrientes subterráneas. Se recomienda un incremento en esfuerzos de conservación, incluyendo su registro en el listado federal cubierto bajo el Acta de Especies en Peligro de Extinción de los Estados Unidos.

Crayfish biodiversity is in decline globally, and members of the genus *Cambarus* are particularly at risk, with 34 species on the Red List of Threatened Species (International Union for Conservation of Nature and Natural Resources, 2004), with 50 species ranked as critically imperiled, imperiled, or vulnerable by the National Heritage Network (NatureServe,

2005), and with 2 species on the United States Fish and Wildlife (USFWS) Threatened and Endangered Species List. The bristly cave crayfish, *Cambarus setosus* Faxon 1889 (Fig. 1) is 1 of at least 40 crayfishes restricted to, and physiologically adapted to, groundwater habitats (stygobites) in North America, and the first and most widespread of at least 7 stygobitic

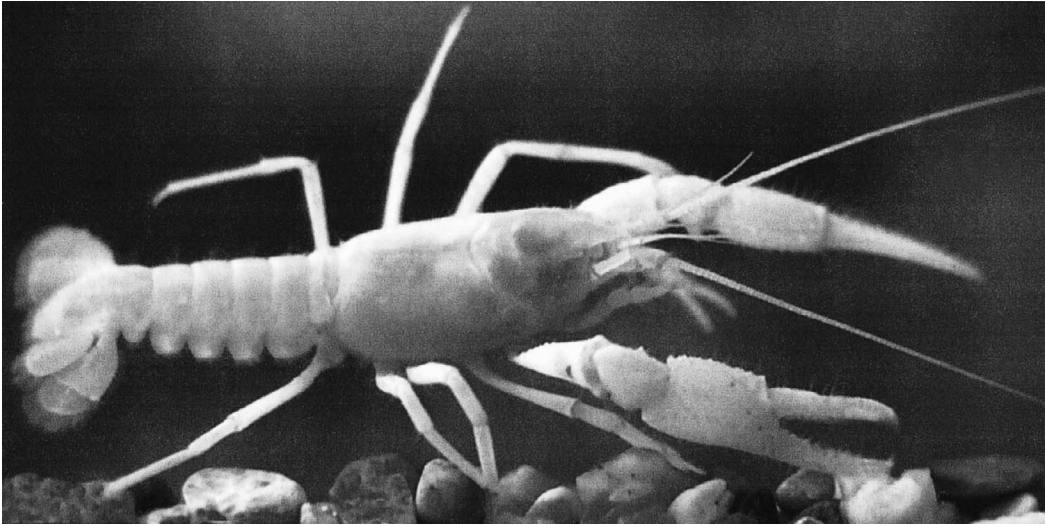


FIG. 1.—Photograph of the bristly cave crayfish (*Cambarus setosus*) from Tom Allen's Cave by J. Gunter (University of Arkansas).

species known from the Ozark Plateaus Ecoregion (Hobbs, 2001a). This study updates the status and expands the known range of this species, and summarizes conservation activities to date.

Visual censuses of crayfish were performed using headlamps and diving lights. One to 3 surveyors moved slowly upstream, counted crayfish in all accessible aquatic habitats, and used snorkeling gear where necessary. Census and habitat data were obtained from this study and other cited studies, as well as unpublished data from the Missouri Natural Heritage database and Missouri Cave Life Database (Missouri Department of Conservation), the Oklahoma Natural Heritage Inventory database (Oklahoma Biological Survey, E. Bergey, pers. comm., 2002), the Missouri Caves and Karst Conservancy files (J. Beard, pers. comm., 2005); the Ozark Subterranean Biodiversity Database, and cave trip reports from S. Todd and L. Willis (University of Arkansas). Collection of voucher specimens was performed under the following permits: USFWS Recovery Permit TE834518-3, Arkansas Game and Fish Commission Scientific Collecting Permit 1476, and the Missouri Department of Conservation Wildlife Collector's Permit 10975.

NEW STATE RECORDS—*Cambarus setosus* Faxon (1889)

Arkansas: Benton County, Tom Allen's Cave; 1 male collected 5 April 2000. The population of stygobitic crayfish in Tom Allen's Cave (consisting of several conduits in a bluff) was discovered by S. Todd (Arkansas Game and Fish Commission) on 12 November 1990 during an Ozark cavefish census. The landowners (T. Allen and family) also observed the crayfish in the cave stream resurgence infrequently throughout the 1900s. These shallow cave streams are actively dissolving the bedding planes of Mississippian-aged cherty limestone of the Boone Formation in the Springfield Plateau within the Spavinaw Creek watershed (tributary to the Neosho River). One male (Form I) specimen was collected by GOG, MES, and AVB in Tom Allen's Cave #2 on 5 April 2000. Specific determination was made by GOG using type specimens from the United States National Museum (USNM) and later confirmed by HHH. The specimen is currently in the personal collection of HHH, and will be deposited in the USNM for permanent curation. Four other individuals of *C. setosus* were noted in this cave and 2 others in the adjacent habitat of Tom Allen's Cave #1 on 5 April 2000; the 2 habitats are probably hydrologically connected and, thus, these individuals are part of a single population.

Arkansas: Independence County, Blowing Cave (or "Dozen's Den"); 1 male collected 22

March 2001. For several decades, local residents have noticed stygobitic crayfish in cave streams along Poke Bayou, a tributary of the White River, north of Batesville, Arkansas. Numerous caves are developed in the Ordovician-aged dolostones of the Plattin Formation in this portion of the Salem Plateau, but specimen collection has been hindered by dangerous hydrological conditions, lack of available male specimens, and the fear of the local community of government land seizure. Credit for notification of the scientific community is given to members of the Dirty Dozen Caving Club (M. Broyles, B. Merrill, G. Molhke, G. De Loss-Molhke, D. Thompson, M. Wann, and H. Bryant) and to R. Redman (Arkansas Soil and Water Conservation Commission).

On 12 December 2000 and again on 22 March 2001, GOG, MES, and B. Wagner (Arkansas Game and Fish Commission) inventoried biota of the accessible (non-submerged) sections of Blowing Cave. Although the Dirty Dozen Caving Club reported dozens of crayfish on caving trips in the 1990s, a maximum of 2 individuals was seen on our visits. Crayfish burrows on streamside mud banks were observed by GOG and MES, and on earlier caving trips by H. Bryant. Although *C. setosus* was not actually seen in the burrows, no other crayfish species is known from this stream habitat. The genus *Cambarus* is known to burrow in the sediments of springs and cave streams (Hobbs and Barr, 1960), and burrowing crayfish and stygobitic crayfish are able to spend extended periods of time out of the water because of the humid environments afforded by saturated air in burrows and caves, respectively (Hobbs, 2001*b*).

One male (Form II) specimen was collected on 22 March 2001 in Blowing Cave. Because the specimen was not in breeding condition (Form I), which is necessary for taxonomic separation from related species, the specimen was held in an environmental chamber under similar conditions to its habitat: native water held at 14°C, complete darkness, and limestone-cobble substrate. After the specimen molted into Form I several months later, it was euthanized by lowering body temperature in a freezer to 3°C and subsequently preserved in 70% ethanol. The specific determination of *C. setosus* was made by HHH by comparison of type specimens of other stygobitic cambarids

from the USNM. The specimen is currently in the personal collection of HHH and will be deposited in the USNM for permanent curation. Directly across Poke Bayou from Blowing Cave is another wet cave called Poke Cave; H. Bryant and the Dirty Dozen Caving Club counted 24 stygobitic crayfish in Poke Cave on a caving trip in the 1990s; because of its proximity to Blowing Cave, we tentatively assign the specific name *C. setosus* to this population until non-lethal genetic analysis can be performed.

OTHER STYGOBITIC CRAYFISH SIGHTINGS—Missouri: Christian County, Avin Sink Cave, Garrison Cave #2, and Sonrise Cave. Avin Sink Cave is postulated to be a former upstream input for the Breakdown Cave karst system (until its input stream was pirated to a lower level that now takes flood waters to Blue Spring). This 64-m cave ends in a perennially wet sump; one crayfish was observed on 23 September 1994 by J. Beard in this sump. We tentatively assign the specific name *C. setosus* to this population because this habitat is contiguous with known *C. setosus* habitat. Garrison Cave #2 contains extensive subterranean stream passage (ca. 7 km) and has a substantial population of stygobitic crayfish: in 1989 D. Figg counted 4 and J. Beard counted 40 in 2000 and 38 in 2005. Sonrise Cave is on the same property as Smallin Cave, but they are several hundred meters apart. Sonrise Cave apparently was connected to Smallin Cave before ceiling collapse. On 7 February 1982, J. Beard reported one stygobitic crayfish in the subterranean stream and, on 4 June 2005, saw 4 individuals. We tentatively assign the specific name *C. setosus* to this population because this habitat is contiguous with known *C. setosus* habitat.

Missouri: Dade County, Pump Cave. J. Beard observed 2 stygobitic crayfish on 18 March 1995 and again on 22 July 1995 approximately 92 m deep into the cave in a perennially wet pool. The entrance sinkholes to Pump Cave and Carrico Cave are only several hundred meters apart, and both systems have been dye-traced to Bishop Springs. Because Pump Cave habitat is hydrologically connected to a known *C. setosus* site, we tentatively assign this same specific name to this new population.

Missouri: Greene County, Sam's Cave. J. Beard observed 8 stygobitic crayfish on 29 June 1996. This population is suspected to be *C. se-*

tosus because of its proximity to Fantastic Caverns.

Missouri: Lawrence County, Wilson (Stinson) Cave. J. Beard observed 2 stygobitic crayfish in 2001 in the subterranean stream.

Missouri: Newton County, Capp's Creek Well. MES, WRE, and S. Samoray observed 2 stygobitic crayfish on 19 November 2004.

Missouri: Polk County, Leith Cave. J. Beard observed one stygobitic crayfish on 17 January 1981 in the subterranean stream; this is the first stygobitic crayfish reported from this county.

DISCUSSION—*Cambarus setosus* is the fifty-eighth crayfish species known from Arkansas (Bouchard and Robison, 1982; H. Robison, pers. comm., 2002). While it is predicted that *C. setosus* occurs in Oklahoma because of proximal locations in Missouri (Creaser and Ortenberger, 1933; Hobbs and Barr, 1960; Reimer, 1969), it is currently incorrect to include *C. setosus* in the fauna of Oklahoma, and checklists such as those by Pflieger (1996) and Taylor et al. (2004) should be revised. Stygobitic crayfish populations discovered in the 1900s in both Arkansas and Oklahoma were tentatively assigned to *C. setosus* (Hobbs and Barr, 1960; Hobbs et al., 1977; Mehlhop-Cifelli 1990), but later were described as new species (Hobbs and Cooper, 1972; Hobbs and Brown, 1987; Hobbs, 1993), or they remain undetermined. Hobbs et al. (1977) assigned *C. setosus* to Twin Cave (Delaware County, Oklahoma), but Hobbs (1993) later described it as a new species, *Cambarus subterraneus*, and extended its range to 2 other caves in the same county: Star Cave and Jail Cave. *Cambarus tartarus* was described from January-Stansberry Cave, Delaware County (Hobbs and Cooper, 1972) and is now known from 2 other caves in the same county: Long's Cave and McGee Cave (Graening et al., 2006a). Specific determination is still needed for the elusive, stygobitic decapods in at least 9 caves in the Oklahoma counties of Adair, Delaware, Mayes, and Ottawa (Hobbs and Barr, 1960; Black, 1971; Tafanelli and Russell, 1972; Willis, 1984; Brown, 1987; USFWS, 1989; Mehlhop-Cifelli, 1990).

Cambarus setosus was known previously only from Mississippian-period carbonate bedrocks of the Springfield Plateau and its unconfined aquifer. The discovery of the population in In-

dependence County, Arkansas, extends its range into the Salem Plateau (Ordovician-period dolomitic bedrock) and its unconfined Ozark aquifer. The 2 discoveries also extend its range by watershed, adding the middle White River and Spavinaw Creek (tributary to the lower Neosho), whereas they were known previously only from the James River and the Bull Shoals Reservoir area (tributary to the upper White River), the Sac River (tributary to the Osage River), and the Elk and Spring rivers (tributaries to the upper Neosho River).

Genetic analyses of *C. setosus* demes by Koppelman and Figg (1995) revealed sufficient polymorphism to propose subspecific differentiation of southwestern Missouri demes (particularly those in McDonald County) from other demes. The new populations in Arkansas have not yet been analyzed, and non-lethal tissue sampling is recommended for these populations and the new populations found by J. Beard and colleagues. The genetic and meristic similarity of the stygobitic members of the subgenus *Jugicambarus* indicates a common ancestry (Hobbs and Brown, 1987; Koppelman and Figg, 1995). The current hypothesis posits that during the Pleistocene epoch, ancestral cambarids dispersed from the Gulf of Mexico into the Ozark Plateaus via the White River, colonizing subterranean channels en route and speciating to various degrees (Hobbs and Barr, 1960; Hobbs and Brown, 1987). This hypothesis explains the apparently disjunct distribution of *C. setosus*, as well as the overlapping range of this species with other stygobitic crayfish, such as *C. aculabrum*.

Cambarus setosus is currently known from 48 sites in Missouri and 2 in Arkansas with a total of 164 individuals (tallied from the most recent census of each site) (Table 1, Fig. 2). At least 9 other populations of stygobitic crayfish are hypothesized to be *C. setosus*. Currently, the largest population known is Smallin Cave (Christian County, Missouri) with at least 47 individuals (and historically, up to 48 individuals). Although statistical analyses cannot be performed upon these census data because of the lack of consistent census technique and effort, one trend is suggestive—the majority of sites have had higher counts in the past. Hobbs and Barr (1960) reported that *C. setosus* was more abundant in the first half of the 1900s. Habitat degradation remains the primary

TABLE 1—Summary of all known censuses of *Cambarus setosus* throughout its range, organized alphabetically by state, then county, with data columns as follows: date(s) censused to the nearest known day or year; count is number visually censused, number casually observed, or in a few instances, number collected; presence-absence of gate or fence, or unknown; property owner class—private but not commercial, commercial (“show cave”; Comm), government agency (Gov), non-governmental agency (NGO), or unknown; and data source citation.

Site	Date	Count	Gated	Owner	Citation
ARKANSAS					
Benton County					
Tom Allen’s Cave (# 1 and 2)	12 Nov 1990	1	No	Private	A
	5 Apr 2000	7			B
Independence County					
Blowing Cave	25 Aug 1993	3	No	Private	C
	1997	*1			D
	12 Dec 2000	2			B
	22 Mar 2001	1			B
MISSOURI					
Barry County					
Moore Cave	7 Apr 1992	1	—	—	E
Christian County					
Breakdown/Fitzpatrick Cave	8 Jan 1985	*1	Yes	Private	P
	1987	6			E
	1 Nov 1989	0			E
	10 Jan 1993	*1			P
	10 Apr 05	5			P
Cave Hollow Cave	1974	5	No	Private	E
	31 Aug 1989	1			E
Green Valley Creek Spring	16 Feb 1995	1	—	—	E
H’Doubler Cave	1979	*1	—	—	I
Hedgpeth Well	25 May 1995	5	No	Private	E
Minnick Spring Cave	13 Aug 1981	1	—	Private	J
Saunders Valley Cave	1986	1	No	Gov	E
	Nov 1989	0			E
	1933	*1	Yes	NGO	F
	1948	33			G
	1951	48			H
	1979	12			I
	1981	29			J
	12 Apr 1982	12			P
	24 Mar 1983	12			P
	6 Jul 1985	4			P
	1988	3			E
1 Oct 1989	4			E	
6 May 1992	2			P	
29 Oct 1997	2			P	
1 Feb 2004	2			P	
8 Jun 2005	47			P	
South Cave	10 Mar 2002	2	—	Gov	E
Wood (Woody) Cave	1977	*1	No	Private	K
	1979	0			E
	8 Nov 1989	2			E
	22 Sep 1999	1			E
	7 Jul 2000	2			E
Dade County					
Carrico Cave	1 Feb 1964	*1	No	Private	K
	1979	5			I
	14 Nov 1989	6			L
	18 Feb 1995	1			P

TABLE 1—Continued.

Site	Date	Count	Gated	Owner	Citation
Greene County					
Crystal Cave	1979	7	Yes	Comm	I
	8 Nov 1989	0			E
Doling City Park Cave	11 Aug 1987	1	Yes	Gov	P
	1993	2			E
	1994	2			E
	15 Aug 1996	1			E
	17 Feb 1997	1			P
	2 Mar 2004	4			P
	27 Jan 2005	2			P
Fantastic Caverns	1979	1	Yes	Comm	I
	20 Feb 1982	12			J
	23 Feb 1987	7			M
	15 Aug 1989	12			P
	1994	2			E
Fauna Cave	14 Mar 1996	8			E
	1979	7	Yes	Private	I
Fulbright Spring Cave	1989	1			L
	1979	5	Yes	Gov	I
Jackson Cave	8 Nov 1989	1			L
	1989	5	—	—	E
Jones Spring & Cave	1991	2			E
	31 Oct 1992	11			E
	1993	5			E
	1994	9			E
	17 Jul 1996	1			E
	7 Jul 2000	7			E
	12 Jun 1951	*1	No	Private	K
	1977	2			E
McCowan Cave	1979	0			I
	1989	1	Yes	—	E
Moore's Spring & Cave	1950	1			S
	1977	*1	Sealed	—	K
	1979	1			I
Pfaff (Brewer) Cave	12 Aug 1971	1	—	Private	I
	1980	0			E
	4 Aug 1981	5			J
	1985	2			O
	1989	0			E
	1 Nov 1991	1			E
Sequiota (Fishers) Cave & Spring	1977	*1	No	Gov	K
	19 Oct 1989	3			L
	19 Aug 1990	4			P
	21 Apr 2002	1			P
	18 Apr 2004	1			P
Steury Cave	1968	*1	Sealed	Gov	I
Taylor's Spring Cave	1984	2	—	Private	J
	1987	2			E
	8 Nov 1989	0			E
Jasper County					
Bristly Cave	9 Apr 1992	8	—	—	E
Cave Spring Cave	25 Jan 1957	1	—	—	I
Coolbrook Spring Cave	1977	*1	—	—	K
	1979	1			I
	1989	4			L

TABLE 1—Continued.

Site	Date	Count	Gated	Owner	Citation	
Kellhauser's Cave	1957	*1	—	Private	E	
	9 Jul 1981	2			N	
	1984	3			J	
	14 Nov 1989	20			L	
	12 Jul 1994	2			E	
Lewis Well Sarcoxie Cave	29 Mar 1996	6	—	—	E	
	1977	*1	Yes	NGO	K	
	9 Jul 1981	5			J	
	4 Feb 1984	2			N	
	1985	3			O	
	21 Nov 1996	5			E	
	6 Apr 1997	4			P	
	31 Jan 2000	1			P	
	10 Feb 2001	1			P	
	8 Feb 2002	3			B	
	12 Apr 2003	1			P	
	18 Nov 2004	1			E	
	Wilson's (Whisner) Cave	1889	12	—	—	Q
		1977	*1			K
1979		10			I	
16 Jan 1982		3			J	
Apr 1984		7			E	
1985		5			O	
1987		5			E	
1989		*1			L	
1992		1			E	
Sep 1993		0			E	
May 1994		1			E	
1996		2			E	
Lawrence County Turnback Cave		1979	4	Yes	Gov	I
	24 Aug 1981	20			R	
	23 Feb 1987	24			N	
	1988	2			E	
	1989	0			E	
	1992	1			E	
	1995	7			E	
	28 Feb 1996	4			E	
	8 Nov 1997	3			P	
	7 Nov 1998	2			P	
	6 Nov 1999	*1			P	
	4 Nov 2000	4			P	
	3 Nov 2001	5			P	
	2 Nov 2002	5			P	
	1 Nov 2003	2			P	
19 Nov 2004	7			E		
McDonald County Bluff Dweller's Cave	1927	21	Yes	Comm	E	
	1979	2			I	
	1989	0			E	
Polar Bear Cave	3 Jun 1991	2	—	—	E	

TABLE 1—Continued.

Site	Date	Count	Gated	Owner	Citation		
Newton County							
Ben Lassiter Cave	16 Mar 1983	4	—	Private	N		
	5 Feb 1986	4			N		
	11 Jun 1986	2			N		
	26 Jun 1987	14			N		
	1989	3			E		
	1992	1			E		
	1993	7			E		
	1994	1			E		
	30 Mar 1995	4			E		
	Boulder Spring Cave	17 Apr 1997	1		—	—	E
	Elm Spring	1987	11		—	—	M
		1989	1				E
		Jun 1992	29				E
Jun 1996		5			E		
Hearrell Spring	Aug 2000	3			E		
	1989	*1	—	—	L		
	1992	1			E		
Hilldale Cave	1991	1	—	—	E		
	16 Mar 1995	0			E		
Neosho Big Spring Park	5 Feb 1986	2	Yes	Gov	N		
	26 Jun 1987	14			N		
Severn Well	11 Aug 1992	6	—	—	E		
Slaughters Well	March 1995	10	—	—	E		
Stroop Cave	1979	*1	—	—	I		
Stone County							
Culvert in Cave Spring	12 Dec 1989	2	—	—	S		
	23 May 1995	1			E		
Exsurgence on Schulyer Creek	23 May 05	1	No	—	E		
Elm Springs	17 Apr 1982	15	—	—	N		
	1984	5			J		
	1989	1			L		
Hayes Spring Cave	1979	4	—	—	I		
	1989	5			L		
	21 Jun 1994	2			S		
	14 Aug 2003	4			E		
	10 Aug 2004	2			E		

Notes: Asterisk indicates *C. setosus* was observed, but count not recorded. Where multiple censuses were performed in any one year, only the highest count is listed. Citation legend is as follows: A = S. Todd, pers. comm., 2002; B = Arkansas Subterranean Biodiversity Database; C = H. Bryant, pers. comm., 2002; D = R. Redman, pers. comm., 2002; E = Missouri Biospeleological Database; F = Creaser and Ortenburger, 1933; G = Burbank et al., 1948; H = Wells, 1959; I = Marquart, 1979; J = Willis, 1984; K = Hobbs et al., 1977; L = Koppelman, 1990; M = Brown, 1987; N = A. Brown, unpublished data; O = Willis and Brown, 1985; P = J. Beard, pers. comm., 2005; Q = Garman, 1889; R = Gardner, 1986; and S = Pflieger, 1996.

threat to this species. The population inhabiting Sequiota Cave (Greene County, Missouri), for example, was thought to be extirpated by sewage pollution from the city of Springfield (Gardner, 1986), but specimens were again sighted decades later (J. Beard, pers. comm., 2001). The population at Tom Allen's Cave

also might be at risk, because this subterranean habitat is located in the Spavinaw Creek watershed, which contains numerous confined animal feeding operations and several municipal sewage treatment outfalls, resulting in its designation as an impaired waterbody by the state of Oklahoma under Section 303(d) of the fed-

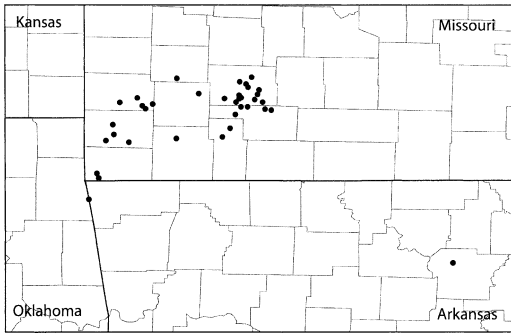


FIG. 2—Current distribution of the bristly cave crayfish (*Cambarus setosus*): black dots indicate confirmed habitat locations, black lines show state boundaries in central USA, and gray lines show county boundaries.

eral Clean Water Act (40 CFR 130.7) due to excessive nutrient loading.

Scientific study, beginning with Parker (1890), has also negatively impacted this species. R. Hoppin, credited with the discovery of *C. setosus*, intensively collected the species in its type locality (Wilson's Cave, Jasper County, Missouri) stating, "The first things that caught the eye were a lot of white crayfish, a dozen in all, like those I took from the wells. It seemed as if I might take every one of them" (Garman, 1889:227). At least 31 voucher specimens have been collected from the type locality by various scientists (Hobbs et al., 1977), yet the most recent (1992) population count was only 2 individuals. In Civil War Cave, 33 *C. setosus* individuals were asphyxiated by Burbank et al. (1948: 366) in a study proving the greater toleration of lowered oxygen tension of cave crayfish compared to their epigeic counterparts, and then warned that such lethal experiments "must not be carried out on such a large scale as to deplete to extinction or near-extinction the population of the cave being studied." However, an additional 48 individuals were taken (and their eyestalks amputated) from this same habitat by Wells (1959) for physiological studies, and another dozen sacrificed for similar research by Fingerman et al. (1964) and Larimer (1966), yet subsequent population censuses in the 1970s (Marquart, 1979) and 1980s (USFWS, 1989) censused only 12 and 29 total individuals, respectively.

Several other mortality factors exist for this species. The caves along Poke Bayou receive

heavy recreational use, and crayfish at Blowing Cave and Poke Cave could be inadvertently trampled by pedestrian traffic, as documented in other cambarid habitats (Graening et al., 2006b). Expulsion by flood waters from the subterranean environment might be a common, natural mortality factor (Graening et al., 2006b); R. Hoppin (Garman, 1889) reported that the landowners of Wilson's Cave often found *C. setosus* flushed out of the cave after flooding. Predation and competition pressure by other species is probable; Brown et al. (1994) reported predation of *C. aculabrum* by banded sculpin (*Cottus carolinae*).

Conservation measures implemented by organizations, such as the Missouri Department of Conservation, the Arkansas Game and Fish Commission, and The Nature Conservancy, have focused primarily upon land acquisition and restriction of traffic through the caves by gating or fencing of entrances; yet only 10 of the 50 sites are currently owned by conservation organizations and only 14 are known to have restricted access (Table 1). Because of this lack of habitat protection, continuing habitat degradation, and the small number of total known individuals, *C. setosus* is still at risk of extinction. We recommend further implementation of conservation measures and further protective status, especially designation as a threatened species under the U.S. Endangered Species Act of 1973, as amended.

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RANGE EXTENSION AND STATUS UPDATE OF THE ENDANGERED HELL CREEK CAVE CRAYFISH, *CAMBARUS ZOPHONASTES* (DECAPODA: CAMBARIDAE)

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ABSTRACT—The range of the endangered Hell Creek Cave crayfish (*Cambarus zophonastes*) is expanded to include a second population, determined by genetic analyses. This crayfish is still restricted to Stone County, Arkansas, and is known from only 14 individuals at Hell Creek Cave and 9 individuals at Nesbitt Spring Cave. Despite this range extension, *Cambarus zophonastes* remains vulnerable to extinction. Water quality sampling of Hell Creek Cave did not detect any major contamination, although numerous potential point and nonpoint source pollutants exist in the watershed. Habitat degradation remains an important threat to this species. Recovery plan implementation status and conservation activities were reviewed and recommendations made.

RESUMEN—La distribución geográfica del cangrejo en peligro de extinción *Cambarus zophonastes*, se incrementa al incluir una segunda población, determinada por análisis genético. Este cangrejo se limita a Stone County, Arkansas, y se conoce solamente de 14 individuos en Hell Creek Cave y 9 individuos en Nesbitt Spring Cave. A pesar de esta extensión geográfica, *Cambarus zophonastes* continúa vulnerable a la extinción. Muestras de la calidad del agua de Hell Creek Cave no detectaron contaminantes trascendentales, aunque numerosas fuentes potenciales localizadas y difusas de contaminación existen en la cuenca. La degradación del hábitat continúa siendo una amenaza importante a esta especie. El estatus de un plan de recuperación y actividades de conservación fueron revisadas y se hicieron recomendaciones.



FIG. 1.—First published photograph of the Hell Creek Cave crayfish (*Cambarus zophonastes*), Nesbitt Spring Cave, Stone County, Arkansas, by D. Fenolio.

As part of the multi-agency initiative to document the subterranean biodiversity of the Ozark Plateaus (the Ozark Subterranean Biodiversity Project), the status and distribution of the groundwater-adapted (stygobitic) crayfishes of this ecoregion were updated. *Cambarus zophonastes* is one of the rarest of all crayfishes, known only from its type locality (Fig. 1) and a maximum of 15 individuals. Because of its rarity and susceptibility to disturbance, it was designated endangered by the U.S. Fish and Wildlife Service (USFWS) (Stewart, 1987), critically endangered by the International Union for the Conservation of Nature and Natural Resources (2003), and critically imperiled by the Natural Heritage Network (NatureServe, 2004). The *Cambarus zophonastes* Recovery Plan cited limiting factors as destruction of habitat, scientific collection, disturbance by amateur cavers, and lack of reproduction (USFWS, 1988). The recovery plan also detailed specific objectives needed for the recovery and delisting of the species, and this report summarizes the status of these objectives to date. The following recovery plan recovery objectives were

implemented in this study: surveys for additional populations of Hell Creek Cave crayfish (objective 2); regular censuses of each population (objective 3); and analyses of habitat quality (objective 4).

The type locality (Hell Creek Cave, Stone County, Arkansas) is a phreatic conduit formed in limestone of the Plattin Formation, Ordovician Period. This karst system pirates surface water from the Hell Creek watershed, collects it in a dendritic conduit system, and discharges the groundwater to springs downstream, where Hell Creek resurfaces and eventually joins the White River. At least 3,150 m of non-submerged passage have been mapped (D. Taylor, Association for Arkansas Cave Studies, Incorporated [AACS], pers. comm., 2002). A second entrance, created in the early 1900s by boring for mineral exploration, acts as a funnel, trapping and dropping a diverse array of epigean fauna into the base of the shaft near the terminal sump. Beyond this sump, only 300 m of passage have been mapped and censused, and extensive habitat is predicted to occur upstream, but will require more technical diving

and logistical support (J. Disler, pers. comm., 2000).

The first published records of *C. zophonastes* in Hell Creek Cave were the collection of 5 specimens in 1961 by Hobbs and Bedinger (1964), and 2 more by Hobbs and colleagues in 1972 (Smith, 1984, unpublished report to USFWS). On 23 August 1983, USFWS sponsored the first census, and Arkansas Natural Heritage Commission (ANHC) employees counted 15 crayfish with the help of cave diver T. Ernst, who used self-contained underwater breathing apparatus (SCUBA) in the submerged passages of Hell Creek Cave (Smith, 1984, unpublished report to USFWS). Another census occurred on 5 October 1990, when S. Snyder counted 13 crayfish (ANHC Natural Heritage Database, C. Osborne, pers. comm., 2001). The latest census of *C. zophonastes* (objective 3.1) was performed using headlamps and dive lights underwater; 1 to 3 surveyors moved slowly upstream, counting crayfish in all accessible aquatic habitats. For submerged passages, certified cave divers were employed after receiving biological training. This census of *C. zophonastes* was performed during 2 site visits because of the difficulty involved in transporting SCUBA throughout the subterranean habitat. On 16 September 2000, we assisted J. Disler (Mid-Ozark Sump Team) in transporting SCUBA into the first cave pool ("Dipping Vat"), where Disler sighted 2 crayfish. We then proceeded upstream to the first sump (the terminus for non-divers) and counted 4 more in the non-submerged passages. On 8 April 2001, members of AACS assisted the Mid-Ozark Sump Team in transporting SCUBA to the first sump to continue the census. Disler swam upstream of the first sump approximately 300 m into unmapped passage and counted 8 additional crayfish, completing the census with a tally of 14 individuals. Another partial census on 20 February 2003 by MES, BKW, D. Fletcher, and D. Kampwerth detected 6 individuals.

Recovery objective 2 identifies the need to survey additional caves for the possible presence of the Hell Creek Cave crayfish. We surveyed an additional 30 cave streams in Stone County (Graening et al., 2001, unpublished report to Arkansas Game and Fish Commission and ANHC; Graening et al., 2004). We found stygobitic crayfish, determined from morphological characters to be members of the genus

Cambarus, in only 2 locations: Cave River Cave and Nesbitt Spring Cave. Because of their proximity to Hell Creek Cave, we suspected that these crayfish might be additional populations of the *C. zophonastes*. Cave River Cave was censused on 24 November 2002; however, the landowner denied permission to use SCUBA or to collect any crayfish. We sighted a single crayfish in Cave River Cave and 5 others in the upstream karst window named Flitterin' Pit. Future, non-lethal genetic sampling of this population will be attempted for specific determination.

In 1992, during the first SCUBA exploration of Nesbitt Spring Cave, "dozens" of stygobitic crayfish were reported (J. Disler and J. Fant, pers. comm., 2001). On 30 March 2002, we censused the non-submerged passages of Nesbitt Spring Cave, saw 2 crayfish, and collected one of them (male Form I) for specific determination. On 5 March 2005, we censused the entire known habitat with the assistance of the Ozark Cave Diving Alliance. Cave diver S. Wallace sighted 9 stygobitic crayfish between the second and third sumps, and carefully captured 6 live crayfish using a suction device, transported the specimens back to D. Kampwerth (USFWS), who then removed a pereopod from each individual. Five of the crayfish were released back into the stream and one was preserved as a voucher specimen. The tissue samples were stored in a preservative of 75% ethanol and 10% 0.5M EDTA. An approximately 502-base-pair (bp) region of the 16S ribosomal DNA (rDNA) from the mitochondrial genome was amplified and sequenced (ABI 3700 sequencer with M13 primers). Pairwise percent sequence differences between specimens from the type locality and Nesbitt Spring Cave ranged from 0 to 1.83% for the same 492-bp fragment. The greatest differences (9 bp) were seen between the Hell Creek Cave specimen and 2 of the Nesbitt Spring Cave samples; however, within Nesbitt Spring Cave, base differences ranged from 0 to 7, so there was a continuum of variation among all the samples. Therefore, the Nesbitt Spring Cave site is considered to represent an additional site for *C. zophonastes*. Voucher specimens will be deposited in the National Museum of Natural History, Smithsonian Institution.

Recovery objective 4.1 is the establishment

of baseline data on water quality. Grab samples were collected at Hell Creek Cave at the Dipping Vat on 20 February 2000 and 7 April 2001 at base-flow conditions. Sampling techniques and analytical procedures followed U.S. Environmental Protection Agency standard methods; appropriate quality assurance and quality control measures were taken. Metal analyses were performed by the University of Arkansas at Fayetteville Central Analytical Laboratory and all others by the University of Arkansas at Fayetteville Arkansas Water Resources Center Water Quality Laboratory. The subterranean stream had the following mean physical and chemical characteristics: pH of 6.5 ± 0.5 units; temperature of $14.5 \pm 0.5^\circ\text{C}$; turbidity of 1 ± 0.5 NTU; conductivity of 265 ± 1 $\mu\text{S}/\text{cm}$; water hardness of 128 mg/L, with a mean concentration (mg/L) of ions of 49.3 for calcium, 1.2 for magnesium, 4.2 for chloride, 0.01 for fluoride, 3.0 for sodium, 0.5 for potassium, 0.1 for fluoride, and 3.2 for sulfate. The following nutrient parameters were sampled and are reported as the mean value: total organic carbon 0.5 mg/L; total Kjeldahl nitrogen < 0.05 mg/L; nitrate-nitrogen 0.7 mg/L, ammonia-nitrogen < 0.01 mg/L; total phosphorous 0.08 mg/L; ortho-phosphate 0.023 mg/L; total coliform density of 1,000 colony forming units/100 mL; and *Escherichia coli* density of 50 colony forming units/100 mL. Metals and other dissolved elements were sampled, and the following were detected, expressed as a mean concentration ($\mu\text{g}/\text{L}$): barium 15.1, boron 2.0, copper 1.3, iron 7.0, and zinc 7.5. Aluminum, antimony, arsenic, beryllium, cadmium, cobalt, chromium, lead, manganese, molybdenum, nickel, selenium, and vanadium were not detected at a detection limit of 1.0 $\mu\text{g}/\text{L}$. These water quality parameters did not exceed Arkansas State Water Quality Standards (Arkansas Pollution Control and Ecology Commission, 1998) and were comparable to regional levels reported by National Water Quality Assessment Program (Petersen et al., 1998). Future, periodic environmental quality sampling of both habitats (objective 4.2) will be coordinated by ANHC and USFWS.

Protection of Hell Creek Cave and its recharge zone (objective 1) is being implemented. The Hell Creek Cave groundwater recharge area was defined (objective 1.2) by Aley and Aley (1985, unpublished report to ANHC

and The Nature Conservancy) as a 906-ha zone. The groundwater recharge zone of Nesbitt Spring Cave should be delineated as soon as possible. Lands adjacent to Hell Creek Cave are protected within the 65-ha Hell Creek Natural Area, and ANHC is attempting to expand their natural area by acquisition of additional acreage in the recharge zone (recovery objectives 1.1 and 1.3). A site conservation and management plan has been prepared jointly by The Nature Conservancy and ANHC. Installation of cave gates, fencing, and monitoring has secured the entrances to Hell Creek Cave, although trespass and vandalism continue to occur. No conservation activities have occurred at Nesbitt Spring Cave or within the Rocky Bayou watershed, but the Nesbitt Family remains dedicated to the protection of the groundwater resource. Recovery objective 1.5 specifies the implementation of an outreach and education program, which should begin immediately. The most efficient approach would be to coordinate with the cave education program of the U.S. Forest Service at nearby Blanchard Springs Caverns. Outreach efforts also should focus on watershed awareness.

Although its range is now extended, the Hell Creek Cave crayfish remains vulnerable to extinction. The recovery plan cited lack of reproduction as a threat; however, no data on population dynamics exist to evaluate this threat. Another threat listed in the recovery plan is over-collection. While this was a significant mortality factor at the time (7 sacrificed out of the observed maximum of 15 individuals), scientific collection has ceased and no amateur collection is known. The 2 other threats cited in the recovery plan—recreational caving impacts and habitat destruction—continue to be potential factors threatening the viability of this species. Inadvertent trampling of stygobitic crayfish by cavers has been documented in another Ozark cave (Graening et al., 2006), so pedestrian traffic in the streambed of both caves should be minimized regardless of reason for visitation. Aley and Aley (1985, unpublished report to ANHC and The Nature Conservancy) conducted a threat analysis of the Hell Creek Cave recharge zone and documented several potential pollution sources, including illegal refuse dumping areas, salvage yards, and malfunctioning septic systems. Other potential habitat stressors include the dis-

charging of urban storm-water or treated municipal sewage from the town of Mountain View, or hazardous material releases from major transportation corridors in the vicinity (Arkansas State Highways 9 and 14). Even though water quality sampling during this study did not detect excessive nutrient, bacteriological, or metal contaminants, source-water protection programs should be implemented for both the Hell Creek watershed and the Rocky Bayou watershed. Additional legal protection for these habitats could be provided by designating these waterbodies as "Extraordinary Resource Water Body" by the Arkansas Department of Environmental Quality and by designating both recharge zones as critical habitat by the USFWS.

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