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Conservation Status and Nesting Biology of the Endangered Duskytail Darter, *Etheostoma percnurum*, in the Big South Fork of the Cumberland River, Kentucky

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ABSTRACT

In September 1995 and May and June 1998 and 1999 we conducted an intensive survey of a middle reach of Big South Fork of the Cumberland River in Kentucky with the goal of finding the duskytail darter, *Etheostoma percnurum*, a federally endangered species. Seventy-one specimens were observed in a 19-stream km reach from the mouth of Station Camp Creek, Scott County, Tennessee, to the mouth of Bear Creek, McCreary County, Kentucky. Using underwater observation and a kick-seining technique around slabrocks, we concur with others that the primary habitat of *E. percnurum* includes clear, silt-free pools immediately above riffles where it seeks cover under cobbles and slabrocks. Most Kentucky specimens (31 of 35) and all nests were found in a 3-km reach from just upstream of the mouth of Troublesome Creek to the mouth of Oil Well Branch. On 26 May 1998 five nests were found at two sites. All nests were located immediately above riffles in silt-free glides with slabrock and cobble substrates and were guarded by males. Eggs, deposited in a monolayer on the underside of slabrocks, numbered from 79-103 eggs per nest. Slabrocks with eggs had mean dimensions of $24 \times 19 \times 4.1$ cm and were located in shallow water (51-70 cm) in areas of low flow (5-14 cm/s). Because of the rarity of this darter within its restricted range in Kentucky, we recommend that it be added to Kentucky's list of protected species as endangered. Morphological comparison of *E. percnurum* from across its range revealed that specimens from Big South Fork have more lateral-line scales, are larger, and are shaped differently than specimens from other populations. The morphological and biological comparisons, along with zoogeographic evidence, indicate that the Big South Fork population of *E. percnurum* is an independent evolutionary unit.

INTRODUCTION

Etheostoma percnurum (Perciformes: Percidae), the duskytail darter, is one of 18 species in the darter subgenus *Catonotus* (Braasch and Mayden 1985; Page et al. 1992), a group characterized by a derived spawning habit of clustering eggs in a monolayer on the underside of slabrocks (Braasch and Mayden

1985; Page 1985). It is a member of one of three recognized complexes in the subgenus, the *flabellare* complex, which includes the stripetail darter, *Etheostoma kennicotti*, and the fantail darter, *Etheostoma flabellare*, in addition to *E. percnurum*. The duskytail darter, long known only by its common name, was formally described and distinguished from its

closest relative, the wide-ranging *E. flabellare*, by R. E. Jenkins in 1994 (Jenkins and Burkhead 1994:877-881). At that time, *E. percnum* was known from six relict populations in drainages of the Cumberland and Tennessee rivers: one in Virginia, Copper Creek; and five in Tennessee, Citico Creek, Abrams Creek, Little River, South Fork Holston River, and Big South Fork of the Cumberland River. Populations in South Fork Holston River and Abrams Creek are believed extirpated (Etnier and Starnes 1993; Jenkins and Burkhead 1994). The only known site of occurrence in the Cumberland River drainage is Big South Fork. Because of this relict distribution, the presumed extirpation of two populations, and threats to water quality in streams it is known to inhabit, *E. percnum* is listed as Federally Endangered (Biggins 1993). Biggins and Shute (1994), Burkhead and Jenkins (1991), Etnier and Starnes (1993), Jenkins (in Jenkins and Burkhead 1994), Layman (1984, 1991), and Simon and Layman (1995) summarized aspects of life history, development, distribution, and abundance based largely on populations in either Copper Creek or Little River.

Despite numerous and intensive fish collections made over the past 40 years, *E. percnum* has been reported from only one locality on Big South Fork, the mouth of Station Camp Creek, Scott County, Tennessee. In 1995, we were contracted by the Kentucky Department of Fish and Wildlife Resources (KDFWR) to conduct a status survey of *E. percnum* in the Kentucky reach of Big South Fork. In this paper we document the status of this species in Kentucky by presenting distributional, abundance, reproduction, and recruitment data. We describe the habitat and nesting biology for the species in Kentucky. Finally, we identify unique morphological, behavioral, and ecological features that indicate the Big South Fork population may be an independent evolutionary unit.

METHODS

Status Survey

From 7-9 Sep 1995 we conducted a nearly comprehensive survey of a middle reach of Big South Fork Cumberland River, Kentucky and Tennessee. We surveyed 14 sites judged to have suitable habitat for *E. percnum* in a

19-km reach from the mouth of Station Camp Creek, Scott County, Tennessee, to the mouth of Bear Creek, McCreary County, Kentucky. Four of these sites were resampled and two new sites in Big South Fork were surveyed in spring 1998 and 1999 during trips designed to gather information on the nesting biology of *E. percnum*. Near the Tennessee-Kentucky border, Big South Fork is a medium-sized river, 30-50 m wide, that flows through a deep (200-300 m) gorge of the Cumberland Plateau. Pools are long and deep, with house-sized boulders and bedrock substrates; riffles are fast, well defined, and flow over a substrate of cobbles, boulders, and some pea gravel and coarse sand. The mainstem has a completely forested riparian zone and is protected as a National River and Recreation Area under management of the National Park Service. Because of limited access in this area, nearly all sites required travelling via canoe.

Underwater visual sampling with snorkeling gear was used at all sites for locating individuals of *E. percnum*. As many as 10 people were involved in underwater sampling at one time, thus increasing the efficiency of the search. In suitable habitat, snorkelers fanned out and turned over slabrocks in pools above and below riffles, macrohabitats known to harbor the species (Jenkins and Burkhead 1994; Layman 1991). This method was supplemented at selected sites by kick-seining (Jenkins and Burkhead 1994) isolated rocks serving as potential cover for *E. percnum*. About 20-60 minutes of snorkel and/or kick-seining time were spent at each site. Standard physical habitat features (width, depth, area sampled) were recorded at each site. The size of large specimens observed was measured or judged to the nearest millimeter with a plastic ruler. Young-of-the-year (YOY) generally were noted but not measured. Initially, identifications of *E. percnum* observed by snorkeling were confirmed by capturing individuals with a dipnet. We quickly discovered that even YOY were easily identifiable while we were snorkeling because of the distinctive appearance of *E. percnum*, the only member of *Catnotus* present in Big South Fork mainstem.

Nesting Biology

Our initial status survey aided in the identification of potential nesting sites for *E. perc-*

nurum. Six sites were surveyed for nests in a 27-km reach between Station Camp Creek and Blue Heron, McCreary County, Kentucky, on 25–26 May 1998, 24 Jun 1998, and 18 Jun 1999. Underwater visual observation with snorkeling gear was used to locate nesting adults. Snorkelers concentrated on appropriate habitat above riffles, turning over rocks suitable for use as nesting substrate. About 60–230 minutes of snorkeling time were spent at each site, and up to six people were involved in underwater visual observation. All *E. percunurum* adults observed were captured with dipnets, measured, photographed, and released. The number of eggs in a nest was counted, and the diameter of eggs, nest rocks, and other physical parameters of nest sites were measured with a small plastic ruler or meter tape. Current velocity was measured over the nest with a Swoffer model 2100 flowmeter at 0.6 of the depth above a nest site.

Systematics

To better understand the evolutionary units under protection, we examined and compared specimens from the drainages of the Cumberland and Tennessee rivers. Seventeen meristic and 27 morphometric variables were taken from 65 and 39 specimens, respectively, of *E. percunurum*. Measurements and counts of meristic features follow the methods of Hubbs and Lagler (1974) except that scales above the lateral-line were counted diagonally from the origin of the second dorsal fin. Vertebrae were visualized by the aid of soft x-rays (3A, 30 mv, 15 seconds) and were counted using the methods of Jenkins and Lachner (1971). Cephalic lateral pore counts followed the methods of Page (1983).

Truss-geometric protocol (Humphries et al. 1981; Strauss and Bookstein 1982) was used in part to archive body form and included 17 measurements distributed among three sagittal truss cells with appended anterior and posterior triangles. Ten additional measurements were included in the morphometric analysis. Multivariate analysis of the morphometric data was accomplished using sheared principal component analysis (PCA) (Bookstein et al. 1985; Humphries et al. 1981) to eliminate overall size effects. Principal components were factored from a covariance matrix of log-transformed morphometric variables following the

recommendations of Bookstein et al. (1985). Multivariate analyses were conducted with programs available in SAS 6.01 (SAS Institute, Inc. 1985) and as modified by D. L. Swofford.

Preliminary morphometric analysis revealed strong sexual dimorphism and seasonal variation associated with reproduction in the Copper Creek specimens (the only ones collected in the spring). To reduce confounding variation associated with reproduction, we removed the Copper Creek material from the analysis, and only compared material from collections made outside of the breeding season (August–February).

RESULTS

Status Survey

We observed 60 individuals of *E. percunurum* in September 1995 and 11 individuals in May 1998 in the 19 stream km reach of the Big South Fork between the mouth of Station Camp Creek, Scott County, Tennessee, and the mouth of Bear Creek, McCreary County, Kentucky (Figure 1). Environmental conditions were ideal because the river was at base flow, water clarity was excellent (at least 2 m), direct sunlight was present, and water temperature was warm, averaging 22.2°C in September 1995 and 22.5°C in May 1998. All sites sampled in Tennessee produced 1–10 *E. percunurum*, but only 6 of 10 sites sampled in Kentucky produced individuals, ranging from 1–11 including both adults and YOY (Table 1). Most (31 of 35) individuals observed in Kentucky were in a 3-km reach from just above the mouth of Troublesome Creek to the mouth of Oil Well Branch.

Etheostoma percunurum, not common at any site, was probably the least common darter species observed. Considering that we adequately sampled only 25–50% of the suitable habitat at any one site, the species is presumably more abundant than our results (Table 1) might otherwise suggest. Our professional judgment is that at least 5–10 times the numbers we observed almost certainly inhabit a given site. This conservative estimate would yield a total population of 300–600 individuals of *E. percunurum* in a 19-km stretch of Big South Fork.

Underwater observation proved to be a productive method of finding and observing *E.*

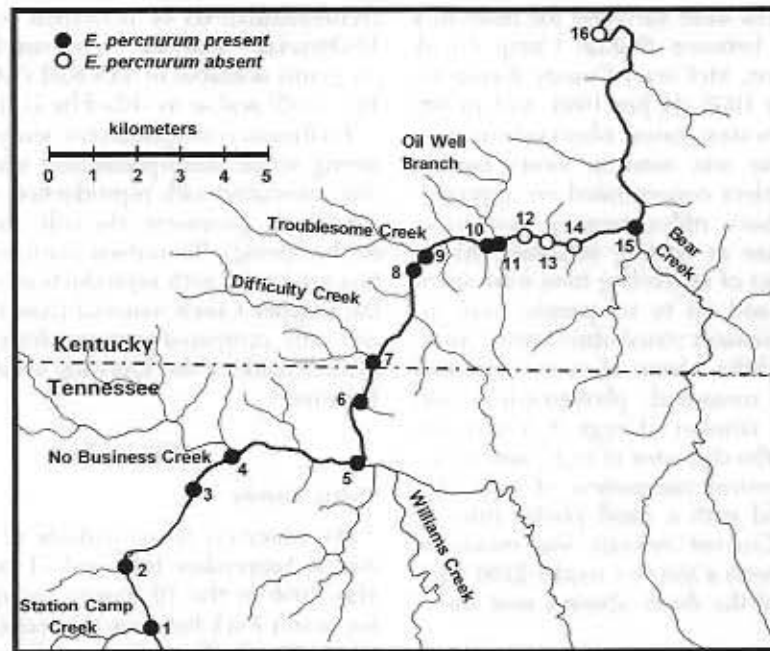


Figure 1. Sampling sites for *Etheostoma percnurum* along Big South Fork of the Cumberland River, Scott County, Tennessee, and McCreary County, Kentucky.

percnurum in most habitats. For comparison, we kick-seined isolated rocks judged to potentially harbor *E. percnurum* at three sites in Kentucky. Initially, our success rate was high, as 3 of 6 rocks sampled yielded *E. percnurum* (site 8). Subsequently, at sites 9 and 11 only 2 of 14 rocks and 1 of 14 rocks sampled, respectively, yielded *E. percnurum*. At these three sites our yield per unit effort for kick-seining (1 fish per 23.3 person-minutes) was considerably higher than for underwater observation (1 fish per 80.8 person-minutes). However, the stacked slabrocks and boulders of most areas precluded sampling by the kick-seining method.

Habitat

All *E. percnurum* were observed in silt-free pools or raceways with low, but evident flow, immediately above riffles where cobbles, boulders, and slabrocks were available. These pools averaged about 25 × 40 m in area and about 54 cm deep, although specimens were observed as deep as 1.5 m. All individuals were under cover of cobbles, boulders, or slabrocks. Cover rocks ranged from fist-sized

cobbles to 76 × 76 cm slabrocks and boulders, with an average thickness of 5 cm. Eleven other darter species were found in association with *E. percnurum*, including *Etheostoma baileyi*, emerald darter; *Etheostoma blennioides*, greenside darter; *Etheostoma camurum*, bluebreast darter; *Etheostoma caeruleum*, rainbow darter; *Etheostoma cinereum*, ash darter; *Etheostoma sanguifluum*, bloodfin darter; *Etheostoma stigmaeum*, speckled darter; *Etheostoma tippecanoe*, tippecanoe darter; *Etheostoma zonale*, banded darter; *Percina caprodes*, logperch; and *Percina copelandi*, channel darter.

Nesting Biology

Five nests were located in 44.7 person-hours of snorkeling, for a rate of one nest per 8.9 hours of snorkeling. Three nests were found at site 8, just above the mouth of Troublesome Creek, and two nests were found at site 10, mouth of Annie Branch, all on 26 May 1998 (Table 2). All nests were in pools and raceways, 5–50 m above riffles. Nests were in water 51–70 cm (mean = 62 cm) deep with current velocity at 5–14 cm/s (mean = 10 cm/

Table 1. Geographic location, date, number of individuals observed while snorkeling, unit of effort (snorkelers \times minutes), and approximate size of specimens of *Etheostoma percnurum*, Big South Fork Cumberland River, Kentucky and Tennessee. Geographic location numbers correspond to those in Figure 1. Sexable adults are divided into males (M) and females (F).

Geographic location	Date	Number of individuals (sex)	Unit of effort	Size of specimens (mm, TL)
1. Mouth of Station Camp Creek, Scott Co., TN	7 Sep 1995 25 May 1998	6 2F	9 \times 60 8 \times 30	35-50 + YOY 50
2. Mouth of Parched Corn Creek, Scott Co., TN	7 Sep 1995	6	9 \times 60	40-50
3. Halfway between Cold Spring and Big Branch, Scott Co., TN	7 Sep 1995	10	9 \times 60	30-50 + YOY
4. Big Island, Scott Co., TN	7 Sep 1995	10	9 \times 90	35-50 + YOY
5. Just above mouth of Williams Creek, Scott Co., TN	8 Sep 1995	1	9 \times 60	30
6. Near mouth of Hurricane Creek, Scott Co., TN	8 Sep 1995	1	9 \times 60	35
7. Mouth of Difficulty Creek, McCreary Co., KY	8 Sep 1995 25 May 1998	1 2	9 \times 60 6 \times 60	40 35
8. 1 km above mouth of Troublesome Creek, McCreary Co., KY	8 Sep 1995 25-26 May 1998	11 3M, 1F	9 \times 110 6 \times 180	40-60 YOY 50-65
9. Mouth of Troublesome Creek, McCreary Co., KY	9 Sep 1995	7	10 \times 75	30-55 + YOY
10. Mouth of Amie Branch, McCreary Co., KY	26 May 1998	2M, 1F	6 \times 35	54-67
11. Mouth of Oil Well Branch, McCreary Co., KY	9 Sep 1995	6	10 \times 20	40-60 + YOY
12. Huling Ford, McCreary Co., KY	9 Sep 1995	0	10 \times 20	—
13. Mouth of second unnamed tributary below Huling Ford, McCreary Co., KY	9 Sep 1995	0	10 \times 30	—
14. Mouth of tributary near Slaven's Branch Trail, McCreary Co., KY	9 Sep 1995 26 May 1998	0 0	10 \times 30 2 \times 20	— —
15. Mouth of Bear Creek, McCreary Co., KY	9 Sep 1995	1	10 \times 120	50 mm
16. Blue Heron, McCreary Co., KY	24 Jun 1998 18 Jun 1999	0 0	2 \times 90 2 \times 150	— —

Table 2. Summaries of physical habitat features and nest characteristics of five nests of *Etheostoma percnurum* at sites 8 and 10 (see Figure 1 and Table 1) in Big South Fork Cumberland River, Kentucky, 26 May 1998.

Nest parameter	Mean	Range
Length of nest rock	186 mm	150-300 mm
Width of nest rock	240 mm	150-400 mm
Thickness of nest rock	41 mm	37-50 mm
Depth of nest	62 mm	51-70 mm
Height of nest rock cavity	21 mm	15-25 mm
Size of guardian male	57 mm	55-58 mm
Water temperature	22.5 C	22.5 C
Length and width of egg mass	40 \times 50 mm	30 \times 40 mm-50 \times 75 mm
Diameter of eggs	2-3 mm	2-3 mm
Number of eggs in nest	101	79-132
Current velocity	9.6 cm/s	5.0-14 cm/s

Table 3. Frequency distribution of lateral-line scales in four populations of *Etheostoma percnurum*, Virginia and Tennessee.

Population	39	40	41	42	43	44	45	46	47	48	49	50	n	Mean	SD
Copper Creek, VA	2	1	5	3	5	3	1						20	42.05	1.67
Big South Fork, TN						2	1	1	2	1	—	1	8	46.38	2.07
Little Rock, TN		3	1	5	6	8	4	2	—	1	1		31	43.55	2.08
Citico Creek, TN			1	1	—	1	1	2					6	44.00	2.10

s). All nest rocks were slab-sided and ranged from 15 × 18 cm to 30 × 40 cm and averaged 4.1 cm in thickness. A cavity 15–25 mm deep was between the substrate (sand, coal, and detritus) and the bottom of the nest rock. Nests were located in a relatively small area at each site; nests ranged from 2.4–7.0 m apart. Water temperature was 22.5°C at both sites.

Eggs were adhesive, round, 2–3 mm in diameter, and amber. They were deposited in a monolayer on the underside of the nest rocks except one nest had one egg laid on top of another egg. Four of the five clutches contained "eyed" embryos rapidly moving inside their chorions. The number of eggs in the nest clutches (or the complement of eggs) ranged from 79–132 (mean = 101). Clutches were oblong to round, ranged in size from 30 × 40 mm to 55 × 75 mm, and generally placed near the center of the underside of the nest rock.

Each of the five nests was guarded by a single male with nuptial colors and morphology similar to other members of *Catonotus*. The knobs on the tips of the first dorsal fin were bright gold to amber, and the edges of the pectoral, soft dorsal, and caudal fins were distinctly peppered with black margins. All males had strong vertical bar development on their sides and blackened heads. The bases of their caudal and pectoral fins were light amber to salmon; pelvic fins were an iridescent white. Standard length (SL) for the five guarding males ranged from 55–58 mm (64–67 mm total length). The four adult females (50–54 mm SL) were found under rocks well away from nests. None of the females were swollen with mature ova and apparently already had spawned.

Parasites

In May 1998 we observed black-spot disease in five of six specimens examined closely. The number of black spots ranged from 1–4, except that one female had about 25 spots.

This female was covered in fungus and appeared to be near death. Black-spot disease was present in only three of the eight specimens vouchered in September 1995; the number of black spots ranged from 1–5.

Systematics

The Big South Fork population had higher mean lateral-line scales than other populations (Table 3). Specimens from Citico Creek were distinctive in having fewer principal caudal rays, scales above the lateral line, scales below the lateral line, scales around the caudal peduncle, and lateral-line scales and more pored lateral-line scales (Table 4). Other meristic characters examined showed little intraspecific variation. Principal component analysis of the meristic variables was not informative.

Sheared PCA of the morphometric variables separated individuals from the Cumberland and Tennessee drainages into non-overlapping clusters, with most separation occurring along the sheared PC 2 axis (Figure 2). Examination of loadings indicates Big South Fork specimens have shorter soft dorsal and anal fins, a shorter anal fin base, a more posteriorly placed anal fin, and a more robust body (Table 5). In addition, a larger maximum size was attained by males from Big South Fork (58 mm SL) and Little River (56 mm SL; Etnier and Starnes 1993) than males from Copper Creek (48 mm SL; Jenkins and Burkhead 1994). Females attained a larger maximum size in Big South Fork (maximum 54 mm SL) than Copper Creek (45 mm SL, Jenkins and Burkhead 1994) and Little River (47 mm SL; Layman 1991).

DISCUSSION

Conservation Status

Etheostoma percnurum occupies a greater range in the Big South Fork than previously known. We have established the existence of the species over a 19-stream km reach at six

Table 4. Meristic counts displaying little intraspecific variation of 65 *Etheostoma percnurum* from four populations in the drainages of the Tennessee and Cumberland rivers, Virginia and Tennessee. Means are followed by ranges in parentheses.

Meristic	Copper Creek, VA n = 20	Big South Fork, TN n = 8	Little Rock, TN n = 31	Citico Creek, TN n = 6
Dorsal spines	6.9 (6-8)	7	6.61 (6-7)	6.50 (6-7)
Dorsal rays	11.50 (11-13)	11.75 (11-12)	11.62 (11-13)	11.67 (11-12)
Pectoral rays	12.75 (12-13)	13	12.52 (12-13)	12.67 (12-14)
Pelvic rays	6	6	6	6
Anal spines	1.95 (1-2)	2	2	2
Anal rays	7.25 (6-8)	7.25 (7-8)	7.19 (7-8)	7.50 (7-8)
Principal caudal rays	17.85 (17-18)	17.25 (16-18)	17.87 (17-18)	16.33 (16-17)
Pored lateral-line scales	24.20 (17-28)	27.63 (25-30)	25.94 (23-30)	31.00 (29-34)
Scales above lateral line	6.80 (6-8)	6.63 (6-7)	6.48 (6-7)	6.00 (5-7)
Scales below lateral line	8.95 (7-10)	8.25 (7-9)	8.71 (8-10)	8.00 (7-9)
Scales around caudal peduncle	24.05 (22-27)	23.13 (22-25)	23.52 (21-27)	21.83 (20-25)
Interorbital pores	6.05 (6-7)	6	6	6
Preoperculo-mandibular pores	10.00 (9-11)	10.13 (10-11)	10.03 (9-12)	9.50 (9-10)
Total vertebrae	33.55 (33-35; n = 11)	33.63 (33-35)	—	—
Precaudal vertebrae	13.82 (13-15; n = 11)	14.13 (13-15)	—	—
Caudal vertebrae	19.73 (19-20; n = 11)	19.50 (18-20)	—	—

sites in the Tennessee reach of Big South Fork (five of these not previously reported) and report it here for the first time from six sites in the Kentucky reach of Big South Fork. A recent survey by Shute et al. (unpublished data) located *E. percnurum* as far upstream as the mouth of Blevins Branch, Tennessee, expanding the known range in the Big South Fork to a 22-stream km reach. However, most Kentucky specimens are known only from a 3-km reach, and additional populations in Kentucky are unlikely to be present. The Big South Fork harbors the only known population of *E. percnurum* in the Cumberland River drainage.

The few other streams in this drainage that might harbor a relict population of this species have been well sampled (Burr and Warren 1986; Etnier and Starnes 1993). Our population estimate (300-600) over a 19-km stretch, though conservative, indicates considerably lower density in Big South Fork than Little River, Tennessee, where Layman (1991) estimated a population of 1023 *E. percnurum* in a 200-m reach in 1983. The highly restricted and localized distribution (mostly in about 3 stream km) of *E. percnurum* in Kentucky as well as its general rarity argue strongly for its immediate inclusion on the Kentucky state endangered/threatened species list as an endangered species.

The small distribution and population size of *E. percnurum* in Kentucky appears to be due to limited suitable habitat in Kentucky. Extensive alluvial streamside deposits (Pomeroy 1964) are present from the mouth of Oil Well Branch to about 0.5 km above the mouth of Troublesome Creek, the reach with the largest Kentucky populations of *E. percnurum*. Similar alluvial deposits are almost entirely absent along the remainder of unimpounded portions of Big South Fork in Kentucky. Below the mouth of Bear Creek, suitable habitat continues to decline. Big South Fork narrows and becomes a series of long rapids strewn with large boulders, essentially lacking cobble and small boulder shoals. At Blue Heron, the

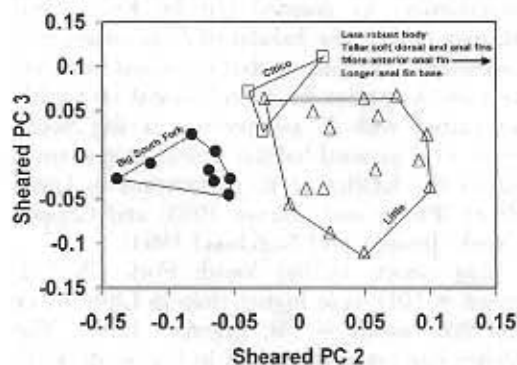


Figure 2. Morphometric scores on sheared PC axes 2 and 3 for 27 *Etheostoma percnurum* from Big South Fork of the Cumberland River, Little River, and Citico Creek, Tennessee.

Table 5. Sheared principal component loadings for 27 morphometric variables for 27 *Etheostoma percnurum* from Big South Fork Cumberland River, Little River, and Cúico Creek, Kentucky and Tennessee.

Measurement	Sheared PC 2	Sheared PC 3
Standard length	-0.005	-0.023
Head length	-0.115	0.067
Gape width	0.190	0.303
Pectoral fin length	0.103	0.222
Pelvic fin length	0.144	0.179
First dorsal fin height	-0.162	0.251
Second dorsal fin height	0.392	0.337
Anal fin height at third ray	0.225	0.367
Interorbital width	0.089	-0.069
Snout to occiput	-0.220	0.066
Snout to origin of pelvic fin	-0.044	0.010
First dorsal fin base length	0.143	0.034
Second dorsal fin base length	0.042	-0.161
Pelvic fin origin to anal fin origin	-0.344	0.068
Anal fin base length	0.334	-0.258
Second dorsal fin insertion to hypural	-0.037	0.118
Anal fin insertion to hypural	0.055	-0.290
First dorsal fin origin to anal fin origin	-0.133	-0.012
Pelvic fin origin to second dorsal fin origin	-0.054	0.097
Second dorsal fin origin to anal fin insertion	-0.175	-0.161
Anal fin origin to second dorsal fin insertion	0.296	-0.197
Occiput to pelvic fin origin	-0.025	-0.126
First dorsal fin origin to pelvic fin origin	-0.138	-0.045
Second dorsal fin origin to anal fin origin	-0.007	-0.108
Second dorsal fin insertion to anal fin insertion	0.110	-0.411
Head width	-0.271	0.022
Body width under second dorsal fin origin	-0.352	0.103

river widens, and some suitable habitat is present, although *E. percnurum* was not observed from this area. Below Blue Heron are the impounded waters of Lake Cumberland, certainly unsuitable for *E. percnurum*.

Although the mainstem of Big South Fork is protected from disturbances by the National Park Service, several tributaries (e.g., Bear Creek) are discharging low-quality because of mining in their watersheds. On 26 May 1998, following a rain the previous night, we observed extremely turbid water discharging from Bear Creek into the otherwise clear Big South Fork. Improvement in these impacted streams will help maintain the high-quality habitats in Big South Fork that are required by *E. percnurum* and other species (e.g., *Notropis* sp. "sawfin shiner" and *E. cinereum*) with restricted distributions in Kentucky.

We suggest periodic monitoring of the distribution and abundance of Kentucky *E. percnurum*. We consider both underwater observation and kick-seining around potential rock cover, our primary means of sampling in Big South Fork, as effective and non-lethal. We

recommend that electrofishing not be used to sample this rare species because of the potential harm it can do to fishes (Snyder 1995).

Natural History

Medium to large streams with silt-free rocky pools in the vicinity of riffles seem to be requirements for viable populations of *E. percnurum*. As pointed out by Etnier and Starnes (1993), the habitat of *E. percnurum* is essentially the same as that occupied by *E. cinereum*, a species we found almost invariably associated with *E. percnurum* in Big South Fork. Our general habitat description is similar to the habitat of *E. percnurum* in Little River (Etnier and Starnes 1993) and Copper Creek (Jenkins and Burkhead 1994).

Egg counts in Big South Fork (79-132; mean = 101) were higher than in Little River (23-200; mean = 79; Layman 1991). The higher egg counts per nest in Big South Fork may be attributed to the larger body size of those females. Using the equation of Layman (1991) $\log C = -1.154 + 1.686 \log SL$, females observed in this study would have 51-

61 mature ova, as compared to the 19–44 ova for the smaller females of Little River. Also, nest rock size typically was larger in Big South Fork (mean = 24 × 19 cm) than in Little River (mean = 16 × 12 cm; Layman 1991). Larger nest rock size in Big South Fork may simply reflect an abundance of larger nest substrata available. Alternately, this may be a behavioral adaptation in choosing more stable nest rocks in an area with high flows and prone to flash flooding. Additional studies are needed to explore these possibilities. Other nesting biology observations are generally consistent with those of Layman (1991) from Little River, Tennessee and Jenkins and Burkhead (1994) from Copper Creek, Virginia, except Jenkins and Burkhead reported nuptial males from moderate to swift runs.

The heavy infestations and high rate of infection of the black-spot disease observed in this study indicate that it may be an important source of mortality for *E. percnum*. Although not previously documented for *E. percnum*, the disease has been reported from many species of North American freshwater fishes. We observed belted kingfishers (*Megaceryle alcyon*) and snails, required intermediate hosts for the strigeid flukes that cause black-spot disease (Berra and Au 1978), to be common in Big South Fork. Heavy infestations have been reported to cause mortalities, particularly during the winter months, in *Esox lucius*, northern pike (Harrison and Hadley 1982), *Lepomis macrochirus*, bluegill (Lemly and Esch 1984), and *Campostoma anomalum*, central stoneroller (Ferrara and Cook 1998). Monitoring of the Big South Fork population of *E. percnum* should include assessment of the extent of black-spot disease.

Systematics

The differences in squamation, body shape, maximum size, and nesting biology reported here indicate that Big South Fork populations certainly represent an independent evolutionary unit. Considering the relict distribution of this species it seems unlikely that any gene flow has occurred between Cumberland and Tennessee forms in thousands of generations. The morphological variation uncovered in this analysis supports the presence of deep phylogenetic partitions in *E. percnum*. This pattern suggests that most of the overall diversity

is located among the populations rather than within populations (Meffe and Vrijenhoek 1988). Because the loss of even one of the remaining four populations of *E. percnum* would cause a substantial loss in diversity of the species, conservation efforts should be directed to preserving as many populations as possible. Protection of the Big South Fork population seems particularly important in maintenance of diversity of *E. percnum* because of the population's unique morphology and ecology.

ACKNOWLEDGMENTS

Success in finding *E. percnum* would not have been possible without the assistance of many people and agencies. We are especially appreciative of the underwater expertise of J. R. Shute, P. W. Shute, P. L. Rakes, and K. Harpster (Conservation Fisheries, Inc.). Steve Bakaletz and R. Emmott (National Park Service) provided logistical support, canoes, and river guidance. We thank T. Slone and D. Stephens (Kentucky Department of Fish and Wildlife Resources) and K. M. Cook, D. B. Henry, J. B. Ladonski, K. R. Piller, and J. G. Stewart (Southern Illinois University at Carbondale), K. A. McCafferty, (Morehead State University), and L. V. Eisenhour for assistance in field work. Permits to study *E. percnum* were provided by Kentucky Department of Fish and Wildlife Resources. R. E. Jenkins, via D. W. Nelson and D. A. Etnier, loaned specimens of *E. percnum* for comparative purposes.

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APPENDIX

Specimens of *E. percnurum* used in morphological comparisons. Museum abbreviations follow Poss and Collette (1995). Parenthetical numbers after catalog numbers refer to the number of specimens used in the meristic and morphometric analyses, respectively.

Big South Fork of the Cumberland River. McCreary County, Kentucky: SIUC 24744 (1,1), SIUC 24761 (1,1), SIUC 24773 (5,5). Scott County, Tennessee: SIUC 24739 (1,1). Copper Creek. Scott County, Virginia: UMMZ 22038 (20,12). Little River. Blount County, Tennessee: UT 91.2719 (7,2), UT 91.2720 (9,4), UT 91.2721 (15,10). Citico Creek. Monroe County, Tennessee: UT 91.2558 (3,3), UT 91.4573 (3,0).