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SYSTEMATICS, STATUS, AND LIFE HISTORY ASPECTS OF THE ASHY DARTER, *ETHEOSTOMA CINEREUM* (PISCES: PERCIDAE)

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Abstract.—Etheostoma cinereum, an endemic of the Cumberland and Tennessee River drainages, is redescribed and diagnosed. Although the species has historically been collected in 15 tributary systems, it has been taken from only 7 in the past 20 years. Sexual dimorphism exists in breeding coloration, size and shape of the genital papilla, tuberculation, and several body proportions. Geographic trends in meristic and morphometric characters indicate that somewhat distinct populations exist in the Cumberland, Duck, and upper Tennessee drainages; no subspecies are recognized. *Etheostoma cinereum is* a pool-inhabiting species often associated with cover in the form of boulders, snags, and stands of *Justicia.* The maximum life-span is 52 months, maximum size is 100 mm SL. A sex ratio of 1.5 females to 1 male was found. Spawning apparently occurs from late January to mid-April. Females at the peak of the spawning season contain an average of 53 mature ova per gram adjusted body weight. The diet consists primarily of chironomids, *Ephemera* larvae, and oligochaetes. Individuals are parasitized by fluke metacercariae, encysted nematodes, and acanthocephalan worms.

The ashy darter, *Etheostoma cinereum* Storer, is one of the largest, most spectacular, and poorly known members of the genus *Etheostoma.* Since its description in 1845 by D. H. Storer from specimens collected near Florence, Alabama, this uncommon fish has been taken in fewer than 80 collections for a total of less than 250 specimens. It is sporadically distributed in medium-size to large streams of the Cumberland and Tennessee River drainages in Alabama, Georgia, Kentucky, Tennessee, and Virginia. Historically, *E. cinereum* has been recorded from 15 tributary systems; recently, however, it has been taken with regularity in only four: Rockcastle River, Big South Fork of the Cumberland River, Buffalo River, and Little River.

Etheostoma cinereum is considered extirpated in Alabama (Smith-Vaniz 1968, Ramsey 1976), and was last collected in Georgia in 1955 (Dahlberg and Scott 1971) and in Virginia in 1964 (Jenkins and Musick 1980). It is on the endangered list in Kentucky (Branson *et al.* 1981) and of special concern in Tennessee (Starnes and Etnier 1980). Nationally, the Endangered Species Committee of the American Fisheries Society (Deacon *et al.* 1979, Williams 1981) recognized *E. cinereum* as a species of special concern because it is presently threatened by destruction, modification, or curtailment of its habitat and range.

Nearly 50 years after its original description, Kirsch (1893) rediscovered and described additional specimens of *E. cinereum* from the Obey River, Tennessee. Bailey, in Bailey and Gosline (1955), placed *E. cinereum* in the monotypic subgenus *Allohistium* based on several unusual morphological features and noted that the species appeared to be most closely related to the subgenera *Not honotus* and *Oligocephalus.* Collette (1965) did not find breeding tubercles on a male with well developed testes from the Cumberland River drainage. Page and Whitt (1973) discovered that *E. cinereum* was the only *Etheostoma* species with electrophoretic mobility of the LDH B_4 isozyme (apparently present in all *Percina).* Based on aspects of the lateralis system, Page (1977) considered *Allohistium* to be a primitive subgenus, as did Collette and Bănărescu (1977). Recently, Page (1981) amplified and augmented Bailey's original description of *Allohistium.* Other than these brief accounts and its inclusion in checklists and state fish-books, very little has been published on the systematics or life history of *E. cinereum.*

The paucity of information about all aspects of this unusual darter prompted this study. Its goals were to: 1) document the historical and present geographic range of the species based on museum specimens and additional collecting; 2) redescribe the species and discuss its geographic variation; and 3) provide information on the life history of the species.

Methods.—Counts and measurements followed the methods of Hubbs and Lagler (1964) except as follows. Squamation on the breast, nape, belly, cheek, and opercle were estimated to the nearest 10%. Postdorsal length was the distance from the posterior margin of the second dorsal fin base to the end of the hypural plate. Terminology and counting procedures of the cephalic lateralis system followed Hubbs and Cannon (1935) and Page (1977).

Methods of studying aspects of the life history were those employed by Page (1974) except as follows. Aging to month was done by using March, the month with the ripest females, as month zero; thus a specimen collected in October and having no annuli on its scales was considered to be 6 months of age. Eighty-three specimens were dissected to obtain information on diet, internal parasites, and reproduction.

In the statistical analysis of data, regression coefficients were calculated by the method of least squares using the General Linear Models procedure of SAS. F tests were used to test the significance of group differences in regression coefficients and in homogeneity of regression. Correlation coefficients were all Pearson product-moment correlations *(r).* Morphometric characters were tested for significant differences between populations at the 0.05 level by regression analysis of the characters on standard length (SL). The effect of sexual dimorphism on differences in regression between drainage populations was reduced by testing the semipartial contribution of drainage to the correlation after partialing out the effects of sex. Determinations as to which populations differed significantly from others were accomplished using a priori orthogonal comparisons. Sexual dimorphism in morphometric characters was analyzed by testing the semipartial contribution of sex to corrleations between the characters and SL. For tabular presentation, measurements were expressed as thousandths of SL for specimens greater than 40 mm SL.

Means of meristic characters were tested for significant differences between drainages at the 0.05 level using Duncan's multiple range test. Raw meristic and morphometric data were transformed to natural logarithms and subjected to SPSS discriminant analyses using the WILKS stepwise method (Nie *et al.* 1975). A priori groupings of specimens were by tributary system. Because small sample sizes can artificially inflate group separations due to random effects (Shaklee and

Tamaru 1981), only those tributary systems with more than 5 specimens were included in the analyses. Meristic and morphometric data were analyzed both together and separately.

For the analysis of variation in some morphological characters and life history aspects, tributary populations were combined into 3 or more inclusive drainage units: Cumberland, Duck, and upper Tennessee. Throughout this paper, "upper Tennessee drainage" refers to the Tennessee River drainage exclusive of the Duck River drainage.

Material Examined.-The following material was examined. The number of specimens in each series is in parentheses. Abbreviations for institutions are identified in Acknowledgments. Complete collection data are on deposit at the Department of Zoology, SIUC.

CUMBERLAND DRAINAGE. ROCKCASTLE RIVER. Kentucky, Rockcastle-Laurel Co.: INHS 87452 (1), 28 May 1981; SIUC 6612 (2), 23 Oct 1979; 2108 (2), 28 Oct 1980; 2131 (2), 27 Oct 1980; 3863 (1), 25 Sep 1981; 3923 (2), 25 Sep 1981; 3989 (4), 12 Apr 1981; 3993 (1), 12 Apr 1981; 4048 (5), 26 Sep 1981; 7456 (9), 5 Mar 1983. BIG SOUTH FORK RIVER. Kentucky, McCreary Co.: SIUC 6633 (1), 24 Oct 1979. Wayne Co.: EKU 1130 (1), 31 Oct 1980; REJ 555 (2), 18-19 May 1972; USNM 25106 (7), 7 Sep 1891; 46212 (1), 7 Sep 1891. Tennessee, Scott Co.: INHS 83895 (1), 30 Sep 1978; SIUC 4128 (1), 30 Sep 1978; UF 17509 (1), 16 Sep 1968; UT 91. 175 (2), 26 May 1968; 91.402 (1), 8 Sep 1969; 91.430 (5), 30 Aug 1968; 91.431 (5), 16 Sep 1968; 91.658 (1), May 1972; 91.1463 (5), 1 Nov 1977; 91.2174 (5), 22 Sep 1968; WCS 015-02 (1), 7 Aug 1969; 455-13 (1), 4 Oct 1975. BUCK CREEK. Kentucky, Pulaski Co.: UMMZ 171557 (5), 14 Sep 1955; 171590 (2), 14 Sep 1955. OBEY RIVER. Tennessee, Overton Co.: EKU 655 (1), 6 Jul 1970; INHS 75541 (1), 6 Jul 1970; USNM 70674 (9), 28 Aug 1891. Pickett Co.: USNM 70675 (6), 28 Aug 1891. ROARING RIVER. Tennessee, Jackson Co.: UT 91.266 (1), 30 Aug 1968. RED RIVER. Tennessee, Robertson Co.: UMMZ 175061 (3), 21 Jun 1957.

DUCK DRAINAGE. UPPER DUCK RIVER. Tennessee, Bedford Co.: AU 4375 (1), 7 Oct 1971; 4408 (4), 7 Oct 1971; UT 91.229 (1), 20 Oct 1968; 91.736 (1), 4 Oct 1972. BUFFALO RIVER. Tennessee, Lewis Co.: INHS 79389 (2), 14 Apr 1978; NLU 29692 (5), 10 Apr 1974; 39945 (6), 8-9 Apr 1978; SIUC 4127 (1), 22 Mar 1978.

UPPER TENNESSEE DRAINAGE. CLINCH RIVER. Virginia, Russell or Scott Co.: VPI 2153 (1), 1964. EMORY RIVER. Tennessee, Morgan Co.: UT 91.224 (1), 5 Aug 1969; 91.690 (1), 1 Jul 1972. LITTLE RIVER. Tennessee, Blount Co.: CU 65136 (1), 31 Jul 1980; EKU 717 (2), 18 Oct 1975; NLU 47492 (6), 8 Apr 1981; OAM 6279 (1), summer 1966; SIUC 3517 (1), 10 Aug 1979; 4141 (5), 13-14 Mar 1982; 7466 (1), 5 Mar 1983; UAIC 4148.15 (2), 5 Jun 1975; 5742.01 (1), 1 Nov 1977; 5743.01 (1), 23 May 1979; 5818.16 (7), 13 Oct 1979; 5953.08 (1), 13 Mar 1980; 6334.16 (4), 25 Jan 1981; UF 19172, 17 Aug 1972; 30676 (1), 10 Apr 1981; UT 91.273 (1), 26 Oct 1968; 91.376 (1), 23 Oct 1969; 91.418 (2), 28 Dec 1969; 91.445 (2), 15 May 1970; 91.499 (4), 21 Jul 1970; 91.512 (1), 20 Oct 1970; 91.593 (10), 5-7 Oct 1971; 91.732 (1), 26 Oct 1972; 91.1205 (4), 17 Apr 1976; 91.1206 (1), 6 Nov 1975; WCS 572-02 (1), 18 Apr 1976; 1071-01 (4), 16 Jun 1979; 1132-02 (4), 25 Aug 1979; 1422-01 (1), 1 Aug 1979. ELK RIVER. Tennessee, Lincoln Co.: TVA uncat. (1), 28 Apr 1981.

Systematics

Etheostoma cinereum Storer, 1845
Fig. 1

Types.—The type(s) of *Etheostoma cinereum* were "caught in deep still water" (Storer 1845) by Charles A. Hentz from Florence, Alabama, some time prior to 1845. It is unclear from the wording just prior (p. 48) to the original description (p. 49) whether Storer based the description on actual specimens, on drawings and a description sent to him by Hentz, or both. Careful reading of the minutes of the Boston Society of Natural History meeting where Storer presented the original description, and a personal letter from Hentz to Storer accompanying some drawings, indicates that no specimens were sent to Storer and none were saved by Hentz. Hentz's original fish drawings are not at the Boston Museum of Science (K. E. Hartel, pers. comm.). This may explain why Collette and Knapp (1966) were unable to find any types of *Etheostoma cinerea,* or of *E. tessellata* described from the same set of drawings and descriptions sent to Storer by Hentz.

The following counts for the missing type(s) or original drawings are from the original description: 11 dorsal spines, 13 dorsal rays, 15 pectoral rays, 6 pelvic rays, 10 anal-fin elements, 17 caudal rays.

Diagnosis.- A species of *Etheostoma* and only member of subgenus *Allohistium* as diagnosed by Page (1981). A large darter reaching a maximum SL of 100 mm characterized by thick, papillose lips; snout long in relation to head length ($\bar{x}$ = 32% of head length); greatly reduced gill rakers on arches 1-3, occurring in 7-9 discrete clusters per arch; breast and nape unscaled (or few scales posteriorly on nape); absence of palatine teeth; usually 6 branchiostegal rays; separate or narrowly joined branchiostegal membranes across isthmus; mid-lateral row of 10-13 small black rectangles expanding into grayish-brown diagonal bands, darkest just below lateral line and forming interrupted lateral stripe extending through eye to tip of snout; usually 4 irregular rows of orange dots forming thin horizontal stripes on upper sides.

The most distinctive features of *Etheostoma cinereum* are those characteristic of mature males including a greatly elongate and distally rounded second dorsal fin reaching nearly to caudal base when depressed (mean second dorsal fin length 32% of SL); rust or blood red on lips (in Cumberland River populations), margin of first dorsal fin, and interradial membranes of both dorsal fins; brilliant, iridescent blue on rays of anal and pelvic fins, that is less intense on interradial membranes of anal and pelvic fins, chin, branchiostegals, lower opercle, breast, belly, and lower caudal peduncle and as a median band in first dorsal fin.

Description.-Certain counts and measurements are presented in Table 1. General body shape is illustrated in Fig. 1.

Dorsal spines 10(6 specimens), 11(78), 12(83), 13(15), 14(1); dorsal rays 11(5), 12(89), 13(78), 14(11); total dorsal fin elements 22(7), 23(59), 24(44), 25(54), 26(16), 27(3); anal spines 1(1), 2(182); anal rays 7(6), 8(142), 9(34); left pectoral rays 14(35), 15(135), 16(12). Pelvic fins with 1 spine and 5(183) rays. Principal caudal rays 15(3), 16(163), 17(2). Lateral line on body complete, rarely a few pores lacking posteriorly. Lateral-line scales 50(1), 51(5), 52(3), 53(14), 54(21), 55(27), 56(25), 57(24), 58(27), 59(14), 60(14), 61(7), 63(1); scale rows above lateral line 6(14), 7(109), 8(50), 9(5), 10(1); scale rows below lateral line 9(7), 10(37),

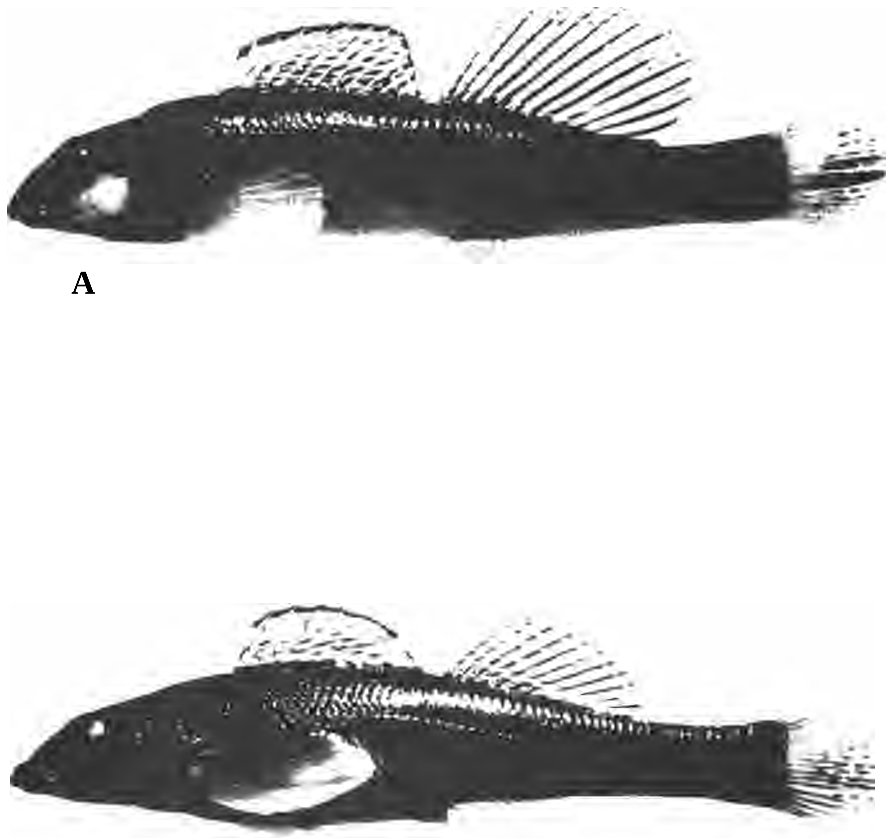

Fig. 1. *Etheostoma cinereum:* A, Male, 77 mm SL, Rockcastle R., Rockcastle-Laurel Co. line; KY, 28 May 1981 (INHS 87452); B, Female, 66 mm SL, as above, 5 Mar 1983 (SIUC 7456). Photos by L. M. Page and B. M. Burr.

11(80), 12(38), 13(14), 14(3); transverse scale rows to first dorsal fin 18(7), 19(27), 20(42), 21(40), 22(34), 23(18), 24(14), 25(1); caudal-peduncle scale rows 20(1), 21(1), 22(12), 23(59), 24(72), 25(32), 26(5). Cheek scales small, slightly ctenoid, moderately to deeply imbedded; cheek, belly, and opercle always fully scaled; nape usually naked, rarely as much as 20% scaled; breast and prepectoral area naked. Modified midventral scales absent. Branchiostegal rays 5:6(1), 6:6(177), 7:6(2), 6:7(1), 6:5(4). Gill rakers greatly reduced and arranged in 7-9 discrete clusters, those on first arch more reduced than on subsequent arches. Vertebrae 39(1), 40(2), 41(48), 42(75), 43(40), 44(1). Prevomerine teeth present; palatine teeth absent. Canals of cephalic lateralis system complete. Infraorbital pores 7(2), 8(174), 9(3); preoperculomandibular pores 9(1), 10(175), 11(3); supratemporal pores 3; supraorbital pores 4; lateral pores 5.

Body elongate, moderately deep (mean body depth 17% of SL), and somewhat compressed (mean body width 93% of body depth), and deepest at nape. Caudal peduncle shallow (mean caudal peduncle depth 10% of SL). Snout long and pointed

Table 1.-Comparisons of means and ranges of selected meristic and morphometric (in thousandths of SL) characters among populations of *Etheostoma cinereum* from three drainages. M is the modal count for each meristic character.

Character	Cumberland drainage				Duck drainage				Upper Tennessee drainage			
	M	$\bar{x}$	Range	n	M		Range	n	M	$\bar{x}$	Range	n
Dorsal spines	11	11.16	10-12	87	12	11.86	11-12	21	12	12.04	10-14	75
Dorsal rays	12	12.15	11-13	87	13	12.57	12-13	21	13	12.93	11-14	75
Dorsal spines and rays	23	23.31	22-25	79	25	24.43	23-25	21	25	24.97	23-27	75
Anal spines	2	2.30	2-3	87	2	2.00	2	21	2	1.99	1-2	75
Anal rays	8	8.14	7-9	87	9	8.57	8-9	21	8	8.08	7-10	75
Lateral-line scales	57	57.18	51-63	87	58	58.33	55-61	21	55	54.97	51-60	75
Scales above lateral line	7	7.44	6-10	85	7	7.33	6-8	21	7	7.07	6-8	73
Scales below lateral line	11	11.21	9-14	85	12	11.86	10-14	21	11	10.84	9-13	73
Transverse scale rows	20	21.13	18-25	87	22	22.33	20-24	21	21	20.47	18-24	75
Caudal peduncle scales	23	23.52	20-26	87	24	24.15	23-26	21	24	23.88	22-26	75
Vertebrae	41	41.29	39-42	75	43	43.00	42-44	21	42	42.27	41-43	71
Body depth		164	90-184	76		172	159-184	21		182	161-201	75
Body width		114	94-138	66		123	113-134	21		125	86-204	67
Predorsal length		330	293-361	76		331	307-398	21		331	200-360	67
Postdorsal length		223	188-278	66		214	188-237	21		206	171-311	67
Caudal peduncle depth		96	79-112	76		99	93-108	21		101	78-114	67
Caudal peduncle length		270	199-299	66		266	244-292	21		262	189-324	67
Head depth		155	135-168	66		161	151-175	21		175	122-195	67
Head width		137	102-191	76		137	123-159	21		152	109-183	67
Snout length		97	79-117	76		99	88-111	21		95	81-112	67
Eye length		65	56-79	66		68	61-76	21		66	57-77	67
D_1 base length		212	163-261	76		226	158-268	21		233	184-270	67
D_1 base length		199	163-239	76		206	178-254	21		214	159-252	67
Longest dorsal spine length		101	71-179	66		90	75-107	21		103	64-136	67
Longest anal spine length		51	34-70	66		49	30-60	21		57	36-81	67
Pectoral fin length		219	149-267	75		209	170-239	21		235	187-283	67
Upper jaw length		84	76-95	66		86	77-95	21		87	72-100	67
Gape width		67	37-89	76		70	54-86	21		77	59-90	67
Cheek length		115	100-154	66		110	96-120	21		110	88-136	67
Suborbital width		26	16-34	66		29	19-36	21		29	16-43	67
Interorbital width		33	21-46	76		40	29-44	21		39	26-53	67

(mean snout length 32% of head length). Lips thick and fleshy with flattened papillae on inner surfaces. Premaxillary connected to snout by broad frenum. Lower jaw included in upper jaw. Gill membranes separate to narrowly connected. Pectoral fins short (mean pectoral fin length 22% of SL). Posterior margin of caudal fin straight-edged or slightly emarginate.

Coloration.—The description is based on fresh and preserved material including specimens in breeding condition collected in Little River, Tennessee, 13-14 March 1982, in Rockcastle River, Kentucky, 5 March 1983, and on color notes provided by Noel M. Burkhead on specimens collected 11 February 1978 in Little River. A color photograph of a breeding male from Little River is shown in Deacon *et al.* (1979).

Ground color of nonbreeding specimens straw-yellow dorsally, white or cream ventrally. Midlateral row of 10-13 small black rectangles, darkest for about 4 scale rows below lateral line, expanding into fainter grayish-brown diagonal bands forming an interrupted lateral stripe which adjoins a dark stripe on head extending through eye to margins of frenum. Usually 4 irregular rows of orange dots on upper sides of body forming thin horizontal stripes. Dorsum crossed by 7 or 8 faint blotches extending ventrally only as far as uppermost horizontal stripe. Subocular bar absent. First dorsal fin with rust-orange blotches on interradial membranes, and blood-red marginal band. Second dorsal fin with several orange blotches on interradial membranes proximally, melding into blood red vertical bars distally. Caudal fin with brown pigment over rays in blotches, sometimes forming 2 or 3 distinct bands, more prominent proximally; blood red blotches present on interradial membranes distally. Pectoral, pelvic, and anal fins usually clear.

Breeding males with intense iridescent blue over rays of pelvic and anal fins, interradial membranes with less intense blue or blackish pigment. Chin, lower opercle, breast, belly, branchiostegals, and lower caudal peduncle, iridescent blue. First dorsal fin with a distal blood red band and a median iridescent blue band, most intense over first 2 or 3 interradial membranes, becoming fainter posteriorly. Second dorsal fin with faint blue-black band over basal one-third of first 7-9 interradial membranes; distal two-thirds of fin with blood red on rays and red spots on interradial membranes. Pectoral fins darkly pigmented on rays, less so on interradial membranes. Caudal fin with red spots on interradial membranes and a dark distal edge. Males more darkly pigmented than females and nonbreeding males, taking on a bronze iridescent appearance. Anus and genital papilla white, contrasting sharply with darkly pigmented venter. Intense blood red on lips in Cumberland River populations only. Breeding females do not develop intense blue body and fin colors, but have red on margin of first dorsal fin and on interradial membranes of both dorsal fins and caudal fin and some blue on ventral edge of caudal fin base. Females and immature males from Cumberland drainage also develop red on lips, but not as intensely as breeding males.

Tuberculation.—Collette (1965) did not find tubercles on specimens of *Etheostoma cinereum* he examined. Since then, a 77 mm SL nuptial male (SIUC 7456) with tubercles on the pelvic and anal fins was collected on 5 March 1983 from Rockcastle River. On the anal fin the tubercles are small, white, and rounded and most abundant on the distal portions of rays 3-8, sparsely developed on rays 1-2. The tubercles on the pelvic fins are similar to those on the anal fin and are

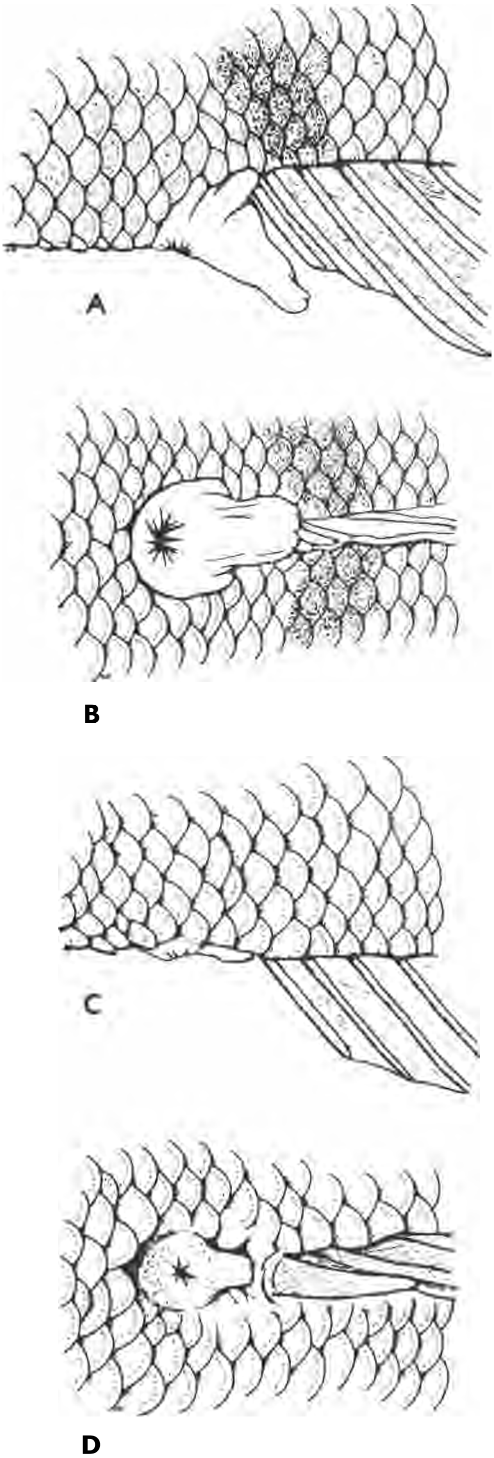

Fig. 2. Genital papillae of *Etheostoma cinereum.* **A, Lateral view and B, ventral view of papilla of gravid female collected 13 Mar 1982 (SIUC 4141); C, Lateral view and D, ventral view of papilla of nuptial male collected 10 Apr 1974 (NLU 29692).**

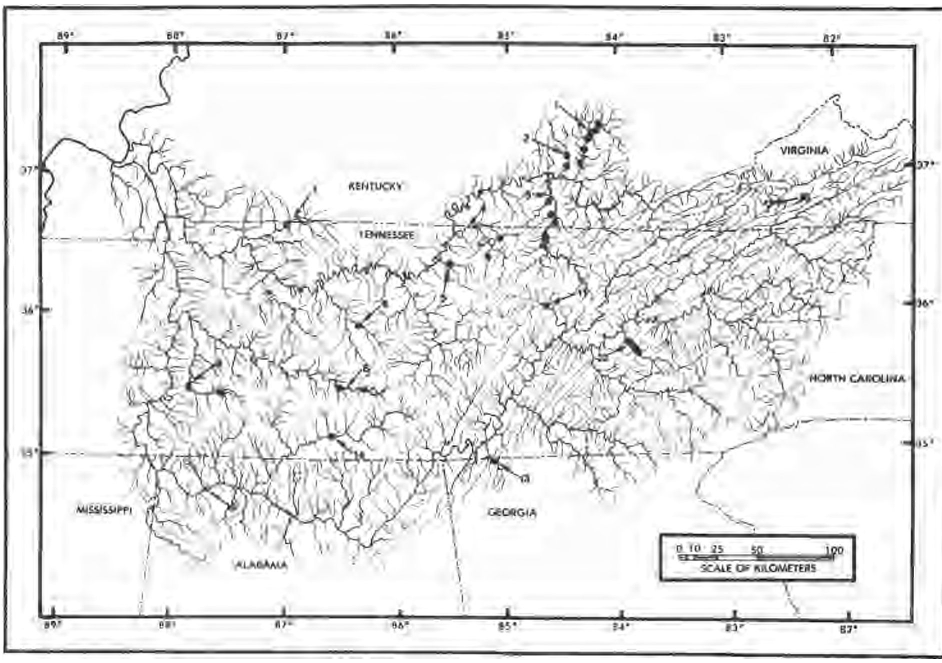

Fig. 3. Distribution of localities where *Etheostoma cinereum* has been collected. Open symbols represent localities presently impounded; the star represents the approximate type locality; the question mark represents an uncertain locality in the Clinch River. 1. Rockcastle R. 2. Buck Cr. 3. Big South Fork of the Cumberland R. 4. Obey R. system. 5. Roaring R. 6. Stones R. 7. Red R. 8. upper Duck R. 9. Buffalo R. 10. Clinch R. 11. Emory R. 12. Little R. 13. Chickamauga Cr. 14. Elk R. 15. Approximate type locality near Florence, Alabama.

located on the ventral sides of rays 2-3. Tubercles were not found on any other specimen examined.

Sexual dimorphism.—Sexual dimorphism in breeding coloration, tuberculation, and growth rate are discussed elsewhere in this paper. No sexual dimorphism in meristic characters was found. Of 30 morphometric characters tested (F-test) for sexual dimorphism, 3 displayed highly significant differences ($P < 0.005$), and 5 displayed less significant differences ($0.005 < P < 0.05$). The most significant characters were longer first dorsal, second dorsal, and anal fin lengths in males. Greater dorsal and/or anal fin lengths in males is a common sexually dimorphic trait in darters, e.g., *Etheostoma variatum, E. blennioides,* and *E. zonale* (Lachner *et al.* 1950); *E. moorei* (Raney and Suttkus 1964); *E. kennicotti* (Page 1975); *E. smithi* (Page and Burr 1976); *E. fonticola, E. microperca,* and *E. proeliare* (Burr 1978); and *E. blennius* (Burr 1979). The longer pectoral, pelvic, and caudal fins reported for males of several darter species were not found in *E. cinereum.* Measurements significantly greater in males at the $0.005 < P < 0.05$ level were caudal peduncle length, snout length, second dorsal fin base length, cheek height, and gape width.

The development of the second dorsal fin of mature males of *E. cinereum* is the greatest of any darter. The second dorsal fins of both sexes develop at the same rate until about 50 mm SL. After this size the male's fin begins to lengthen

Table 2.—Status of selected populations of *Etheostoma cinereum* with a list of habitat modifications that may have adversely affected them. Numbers refer to localities as identified in Fig. 3.

Locality, date of last collection and no. of specimens taken on that date	Habitat modifications that may have adversely affected population	Present status and source
Cumberland R. drainage		
2. Buck Cr., Pulaski Co., KY, 1955:7	Acid wastes; pesticide and chemical pollutants from farming and coal mining	Extirpated; pers. observ., R. R. Cicerello, pers. comm.
4. Obey R. system, Overton Co., TN, 1973:3	Inundation of some localities by Dale Hollow Lake; coal mining pollution	Unknown, additional collecting needed; Carrithers (1971)
5. Roaring R., Jackson Co., TN, 1968:1	Unknown	Unknown, additional collecting needed; this study
6. Stones R., Rutherford Co., TN, 1961:1	Inundation of potential habitat downstream by Percy Priest Lake	Extirpated; this study
7. East Fork Red R., Robertson Co., TN, 1957:3	Heavy siltation; pesticide pollution	Extirpated; pers. observ., Warren and Cicerello (1984)
Tennessee R. drainage		
8. Upper Duck R., Bedford Co., TN, 1972:1	Inundation of some localities by Normandy and Columbia lakes	Unknown, additional collecting needed; Starnes and Etnier (1980)
10. Clinch R., Scott or Russell Co., VA, 1964:1	Alkaline spill in 1967; acid spill in 1970; inundation of potential habitat downstream by Norris Lake	Extirpated; Jenkins and Musick (1980)
11. Emory R., Morgan Co., TN, 1972:6	Coal mining pollution; acidic water; inundation of potential habitat downstream by Watts Bar Lake	Unknown, additional collecting needed; Tackett (1963)
13. Chickamauga Cr., Catoosa Co., GA, 1955:1	Siltation; domestic and industrial pollution	Extirpated; Etnier et al. (1981)
15. Florence, AL (type locality), 1845:?	Siltation; insolation; inundation by Pickwick, Wilson and Wheeler lakes	Extirpated; Ramsey (1976)

at a rate of about 1.5 times that of the female's, which grows at the same rate throughout life. The relationship between second dorsal fin length (Y) and SL (X) for males was $Y = -10.81 + 0.50X$, with $r = 0.94$, and for females was $Y = -3.55 + 0.34X$, with r 0.94.

There is a marked difference between the genital papilla of the female and that of the male. In nonbreeding females the genital papilla is a short thickened tube. During the breeding season, it becomes enlarged to form a conical tube, with a crenate flap of flesh on the distal anterior margin which overhangs the gonopore (Fig. 2). Papilla length (measured from anal orifice to tip of papilla) ranged from 3.7 to 4.7 mm, $\bar{x}$ 4.2 mm, in 6 ripe females 62 to 88 mm SL ($\bar{x}$ = 73 mm).

The genital papilla of both nonbreeding and breeding males is a short flattened tube (Fig. 2) which does not noticeably enlarge during the breeding season.

Distribution and Status

Etheostoma cinereum has been collected from 15 tributary systems in the Cumberland and Tennessee River drainages (Fig. 3). It has not been collected in seven of these since 1969, and in the past five years has been collected only from Big South Fork of the Cumberland, Rockcastle, Buffalo, Little, and Elk River systems. The fact that this fish has not been recently collected from a given tributary system is not necessarily an indication of its absence there. Even at localities where reproducing populations are known to exist, *E. cinereum* is often not taken unless special effort is made thoroughly to collect its habitat. The apparent elimination of *E. cinereum* from some tributary systems is perhaps an artifact of inadequate sampling in recent years.

Some populations of *E. cinereum* have apparently been extirpated and the species has not been collected from several localities in the last 11 years (Table 2). The status of *E. cinereum* in four localities is unknown (Table 2). The factors most seriously affecting the species are impoundments, siltation, coal mining, and domestic and industrial pollution (Table 2). Because *E. cinereum* is primarily a pool inhabitant, siltation may have a greater detrimental effect on its habitat than on that of riffle-dwelling darters where the current helps keep the riffles swept clean of silt deposits.

Reproducing populations showing some recruitment include Rockcastle River, Kentucky, Big South Fork Cumberland system, Kentucky and Tennessee, Buffalo River, Tennessee, and Little River, Tennessee. In the Cumberland drainage, *E. cinereum* has been collected from seven localities in Rockcastle River, Rockcastle-Laurel counties, in the past 10 years, and more than 50 specimens have been collected there in the past five years. The Rockcastle River watershed is partially protected in the Daniel Boone National Forest and water quality and species diversity remain high there. However, Branson (1977) has cited strip mining as a potential threat to fishes in this river. Comiskey and Etnier (1972) reported substantial numbers of *E. cinereum* in the Big South Fork of the Cumberland River and its larger tributaries. More recent collections reveal that the population is still substantial. The lower portion of the system in Kentucky is protected in the Daniel Boone National Forest, but there is considerable coal mining pollution in the upper watershed and the silt load is heavy there at times. Two streams, New River and lower Rock Creek, are badly polluted by strip mining (Comiskey and Etnier 1972).

In the Tennessee drainage, *E. cinereum* has been regularly collected in the Buffalo River in recent years, but only at the mouth of Grinders Creek, Lewis County, Tennessee. Specimens were also taken once in 1971 in Perry County at the Highway 13 bridge. The Buffalo River, one of the most unspoiled southeastern rivers (Anonymous 1973) contains apparently suitable habitat for *E. cinereum* throughout much of its length. It is thus expected that the species is more widespread in the Buffalo River. More specimens (>100) have been collected from Little River, Blount County, Tennessee, than any other system, with the largest single series (11) collected there in 1971. The headwaters of Little River are

Table 3.-Counts of total dorsal fin elements in *Etheostoma cinereum.*

Population	22	23	24	25	26	27	n	$\bar{x}$	SD	CV
Rockcastle R.	2	14	3	1			20	23.15	0.67	2.90
Buck Cr.	2	3	2				7	23.00	0.82	3.55
Big South Fork	1	29	7	2			39	23.26	0.59	2.56
Obey R.	1	6	6	4			17	23.76	0.90	3.80
Roaring R.				1			1	25.00		...
Red R.	1	2					3	22.67	0.58	2.55
Upper Duck R.			4	3			7	24.43	0.53	2.19
Buffalo R.		1	6	7			14	24.43	0.65	2.65
Clinch R.				1			1	25.00	...	...
Emory R.			1	1			2	24.50	0.71	2.87
Little R.		3	15	34	16	3	71	25.01	0.89	3.57
Elk R.		1					1	23.00		...

protected in the Great Smoky Mountains National Park, and *E. cinereum* faces no immediate threat from habitat modification in that area, although a substantial reach of lower Little River has been subject to recent development.

The only record of the species in the Elk River, Lincoln County, Tennessee, is a specimen collected by C. F. Saylor of the TVA near Fayetteville in 1981. The presence of this species in the Elk is surprising in light of water levels that vary more than 1 m daily due to power generation at Tims Ford Dam. Because this dam had been in operation for more than five years when the specimen was collected, there must have been recruitment in the population.

Geographic Variation

While considerable overlap among major drainage populations existed in the ranges of nearly all characters examined, univariate analyses revealed some significant differences and distinctive geographic trends. The limited range of *E. cinereum* and similarity of the streams it inhabits, particularly in regard to temperature regimes, suggest that genetic rather than environmental factors are responsible for most geographic variation found. Results of discriminant analyses

Table 4.-Counts of total vertebrae in *Etheostoma cinereum.* Counts of two specimens from the Obey River arc taken from Bailey and Gosline (1955).

Population	39	40	41	42	43	44	n	$\bar{x}$	SD	CV
Rockcastle R.			4	16			20	41.80	0.41	0.98
Big South Fork	1	2	32	3			38	40.97	0.49	1.20
Obey R.			9	9			18	41.50	0.51	1.24
Roaring R.			1				1	41.00		...
Upper Duck R.					6	1	7	43.14	0.38	0.88
Buffalo R.				1	13		14	42.93	0.27	0.62
Clinch R.					1		1	43.00		...
Emory R.					1	1	2	42.50	0.71	1.66
Little R.			2	47	18		67	42.24	0.50	1.18
Elk R.					1		1	43.00		

Table 5.-Counts of anal rays in *Etheostoma cinereum.*

Population	7	8	9	n	$\bar{x}$	SD	CV
Rockcastle R.	1	16	3	20	8.10	0.45	5.52
Buck Cr.		5	2	7	8.29	0.49	5.89
Big South Fork		31	8	39	8.21	0.41	4.99
Obey R.		16	1	17	8.06	0.24	3.01
Roaring R.		1		1	8.00	...	...
Red R.	1	2		3	7.67	0.58	7.53
Upper Duck R.		6	1	7	8.14	0.38	4.64
Buffalo R.		3	11	14	8.79	0.43	4.85
Clinch R.		1		1	8.00	...	...
Emory R.		2		2	8.00	...	...
Little R.	4	59	8	71	8.10	0.47	5.86
Elk R.		1		1	8.00		

provided further evidence of isolation and divergence of some drainage populations, and gave an indication of multivariate similarity among them.

Counts of dorsal spines, dorsal rays, total dorsal fin elements, and vertebrae were all correlated and displayed similar geographic trends (Tables 1, 3, and 4). Counts of vertebrae and dorsal spines were also highly correlated in a population of *Etheostoma exile* (Gosline 1947) and in *E. zonale* (Tsai and Raney 1974), suggesting that these characters may be linked in development, possibly involving the differentiation of somites (Gosline 1947). Specimens from the Cumberland drainage, with the exception of Rockcastle River, typically had 41 vertebrae, 11 dorsal spines, and 12 dorsal rays. Most specimens from the Rockcastle River had 42 vertebrae, but displayed the same modal dorsal spine and ray counts as other Cumberland drainage populations. Duck and upper Tennessee drainage specimens typically had 42 or 43 vertebrae, 12 dorsal spines, and 12 or 13 dorsal rays. Only 27% of specimens from the upper Tennessee drainage had 43 or more vertebrae, compared to 95% in the Duck drainage. The Buffalo River population was unique in having a modal anal ray count of 9 compared to 8 in all others (Table 5). Scale counts were generally correlated with each other, and to a lesser degree, with

Table 6.-Counts of lateral-line scales in *Etheostoma cinereum.*

Population	51	52	53	54	55	56	57	58	59	60	61	62	63	n	$\bar{x}$	SD	CV
Rockcastle R.					1	2	4	2	5	3	2	—	1	20	58.55	1.99	3.39
Buck Cr.					1	1	3	2						7	56.86	1.07	1.88
Big South Fork	2		3	5	7	9	5	3	2	2	1			39	55.85	2.27	4.06
Obey R.					1	1	2	5	3	3	2			17	58.47	1.66	2.84
Roaring R.										1				1	60.00	...	...
Red R.							1	1	1					3	58.00	1.00	1.72
Upper Duck R.						1	1	1	1	2	1			7	58.71	1.80	3.07
Buffalo R.					1	1	2	6	1	1	2			14	58.14	1.70	2.93
Clinch R.										1				1	60.00	...	...
Emory R.							1	1						2	57.50	0.17	1.23
Little R.	3	2	12	16	16	10	6	5	—	1				71	54.73	1.78	3.26
Elk R.									1					1	59.00	...	

counts of dorsal fin elements. Tsai and Raney (1974) found similar results for *E. zonale.* Little River specimens had significantly fewer lateral-line scales (modally 54, 55) than other tributary systems (Table 6). Most specimens from the Cumberland drainage, as well as the other Tennessee drainage tributaries, had 56 or more, while those from the Duck River drainage usually had 58 to 60. Other scale counts displayed the same trends (Table 1). No other significant variation in meristic characters was found.

As judged by mean values of body depth and body width (Table 1), specimens from the Cumberland drainage were typically less robust than those from the Duck and upper Tennessee drainages. These differences may reflect the condition of individual specimens at the time of capture more than genetically determined factors. Many Cumberland drainage specimens (particularly those collected in fall and winter) often appeared slightly emaciated. As noted in the section on diet, *Ephemera* larvae, a major food item for Cumberland drainage specimens only, were eaten only from late spring until early fall, possibly contributing to the emaciated appearance. Cumberland drainage specimens also differed from others in having significantly longer cheeks, caudal peduncles, and postdorsal lengths and in having shorter first and second dorsal fin bases, upper jaw lengths, suborbital widths, and interorbital widths (Table 1). Specimens from the upper Tennessee drainage differed from others in having significantly greater head depths, head widths, gape widths, pectoral fin lengths, and longest dorsal spine lengths (Table 1). Duck drainage specimens had no unique morphometric features and were more similar to specimens from the upper Tennessee drainage than to those from the Cumberland drainage in most body proportions.

A trenchant color difference may exist between breeding males in the Cumberland and upper Tennessee drainages. Based on examination of a small number of fresh specimens, color transparencies, and Noel M. Burkhead's color notes on a breeding male from Little River, mature males from the Cumberland drainage have blood red lips at least from early fall until late spring (possibly year-round). Males from Little River apparently develop no red on the lips even in peak spawning condition. Examination of more fresh material is needed to determine the distribution and consistency of this character from throughout the range of the species.

In the discriminant analysis with both morphometric and meristic characters included, five significant ($P < 0.001$) functions were derived, with the first two accounting for 70% of the variance between groups. While the variables with the highest value on the WILKS selection criterion were meristic, all of the discriminant functions were most heavily loaded for morphometric characters, e.g., body depth, postdorsal length, and predorsal length. The analysis correctly classified 97.2% of specimens, with all misclassified specimens being assigned to another tributary system in the same major drainage unit. The first two functions were sufficient to classify 97.9% of specimens into their correct major drainage, but only 78.2% into their proper tributary system. In a plot of individual scores on the first two functions (Fig. 4), the clusters formed by specimens from the three Cumberland drainage tributary systems widely overlap each other, as do the two Duck drainage clusters, while the Little River specimens formed a discrete cluster. The only overlap between major drainage units was between the Rockcastle River cluster and those formed by individuals from the two Duck drainage tributaries.

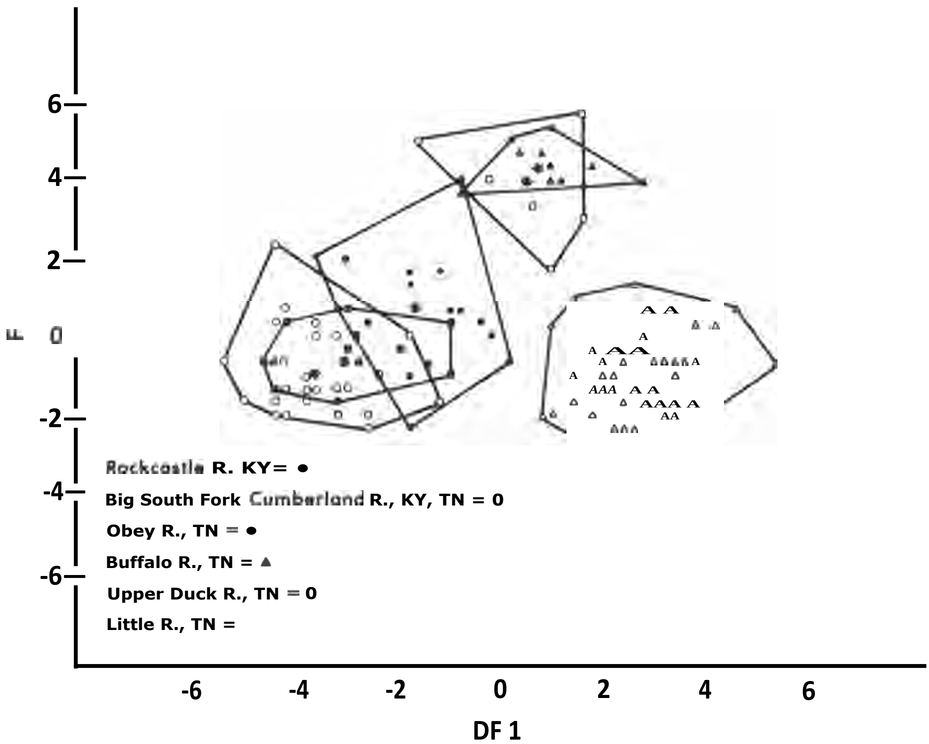

Fig. 4. Projection of 141 specimens of *Etheostoma cinereum* from 6 tributary systems on the first 2 discriminant functions in an analysis using 8 meristic and 26 morphometric characters. Asterisks represent group centroids. Solid lines connect outermost specimens of each tributary system cluster. Where 2 specimens overlap, only 1 is shown.

A discriminant analysis using only meristic characters classified 85.6% of specimens correctly, with 94% being placed in their proper major drainage. All five functions were significant ($P < 0.001$), with the first two accounting for 82% of the separation between groups. The first function was most heavily loaded for vertebrae, dorsal rays, and lateral-line scales. Those characters along with dorsal spines and transverse scale rows were also heavily loaded on the second function.

The analysis using only morphometric characters correctly classified 87.9% of specimens, assigning 93% to their correct major drainage. Of five significant ($P < 0.001$) functions derived, the first two accounted for 74% of the variance between groups. The first two functions were most heavily loaded for postdorsal length, pectoral fin length, caudal peduncle length, body depth, and head width.

Based on the results of the univariate and discriminant analyses, *E. cinereum can* most equitably be divided into three somewhat distinct populations: Cumberland drainage, Duck drainage, and upper Tennessee drainage. In most meristic and morphometric characters, the Duck drainage population is more similar to the upper Tennessee population than the Cumberland population, suggesting that it has experienced either more gene exchange with the former or that the populations in the two drainages have experienced similar selection pressures (Ehrlich and Raven 1969). Although no population is distinct enough in any character to warrant separate taxonomic status, examination of more fresh breeding material may eventually reveal color differences which might justify the naming of subspecies.

Aspects of Life History

Habitat.—*Etheostoma cinereum* has been most consistently collected from clear pools or eddies with sluggish current in water 0.50-1.75 m deep over silt-free sand or gravel substrates in medium-size to large upland streams. The species is often taken close to shore and is usually associated with cover such as boulders, snags, and stands of water willow *(Justicia)* above and below riffles. The close association with cover makes conventional seining techniques inefficient, and may partially explain why *E. cinereum* has usually been collected in low numbers ($\bar{x}$ number specimens per collection = 2.6). On several occasions more than one individual has been collected from the same restricted area of habitat; in one instance (SIUC 4048, 26 Sep 1981) four individuals were collected from the same pool area of approximately 10 m^2, in another, (SIUC 7456, 5 Mar 1983) nine individuals were collected from a pool about 30 m in length in one hour. This suggests that the species may not be as uncommon and solitary in its principal habitat as the low collection numbers indicate. However, the areas meeting the habitat requirements of *E. cinereum* in the streams it inhabits are often rare and localized, contributing to the low population numbers of this fish.

Age and growth.—The oldest specimen examined (EKU 655) was a 100 mm SL, 52-month-old male collected in the Obey River. The oldest female (NLU 29692) was 49 months old, measured 89 mm SL, and was collected in the Buffalo River. Most males (68.2%) and females (79.7%) were less than two years old, with only 20.5% of males and 18.8% of females being in their third year of life (Table 7). Of six specimens more than three years old, five were males. Males also dominated older year classes in other large darters including *Percina sciera* (Page and Smith 1970), *Etheostoma squamiceps* (Page 1974), and *E. kennicotti* (Page 1975). The few species of darters in which females have been found to be more long-lived than males are small species in which very few individuals of either sex live more than 18-20 months, e.g., *E. proeliare* (Burr and Page 1978) and *E. microperca* (Burr and Page 1979).

Within the size range examined (29-89 mm), *E. cinereum* grows at a decreasing rate throughout life. The relationship between SL (Y) and age in months (X) was Y = 1.52 + 20.18 Log_e X, with $r = 0.88$. One-half of the first year's length (26 mm) was reached in approximately 13 weeks. There may be an indirect relationship between growth rate and maximum adult size among darters. The smallest species, e.g., *E. microperca*, and *E. proeliare*, reach one-half of the first year's mean SL in eight weeks or less (Burr and Page 1978, 1979). Larger species, e.g., *E. squamiceps* (Page 1974) and *E. kennicotti* (Page 1975), typically reach one-half of the first year's mean growth in 12 weeks or more.

Significant differences were found between growth rates of specimens from the Duck, upper Tennessee, and Cumberland River drainages. Those from the Duck drainage grew fastest (Y = —3.68 + 23.08 Log_e X, $r = 0.83$), followed by those from the upper Tennessee drainage (Y = 1.33 + 20.45 Log_e X, $r = 0.82$), and Cumberland drainage (Y = 7.87 + 19.05 Log_e X, $r = 0.73$). These differences in

Table 7.- Distribution of sexes among year classes in *Etheostoma cinereum,* with corresponding ranges and means of standard length. Only 108 individuals were sexed.

Year class	Sex	n	Percent	Range SL	Mean SL	SD
0	M	4	9.1	39.0-49.5	43.7	4.92
		5	7.8	32.6-47.2	38.6	5.79
1	M	26	59.1	39.2-66.6	57.6	6.03
		46	71.9	39.7-66.2	56.8	6.47
2	M	9	20.5	62.4-79.1	73.9	5.76
	F	12	18.8	60.5-82.1	70.6	5.17
3	M	4	9.1	79.9-88.6	84.8	3.62
	F				...	
4	M	1	2.2	100.0	100.0	
	F	1	1.6	89.4	89.4	

growth rate are reflected in the mean and range values of SL of each year class for the major drainages compared (Table 8). Differences in the diets of the Cumberland and Tennessee drainage populations, which may be partially responsible for the differences in growth, are described elsewhere in this paper. Other factors known to effect the growth rates of fish and which may play some role in effecting the growth of *E. cinereum* include temperature, competition, and genetically determined growth potential (Weatherley 1976).

Although males of each year class measured larger than females (Table 7), regression analysis of 104 specimens from several localities failed to reveal significant differences between the growth rates of males and females. Since specimens used in the regression analysis were lumped from widely separated localities, variation between localities may have obscured differences between the sexes.

Reproductive biology, males.-No male less than 50 mm SL had well developed testes or enlarged second dorsal and anal fins associated with maturity. All males greater than 50 mm SL and collected during the breeding season had white spongy testes and secondary sexual characteristics suggesting maturity. All 2-year-old males, but only 88% of 1-year-old males, were mature. In most small species of darters (e.g., *E. microperca,* Burr and Page 1979; *E. striatulum,* Page 1980; *E. simoterum,* Page and Mayden 1981) all males, regardless of size, are capable of spawning at one year of age. Among several larger, more long-lived species of darters only males above some minimum size are capable of spawning, including: *E. olmstedi* and *E. longimanum* (Raney and Lachner 1943); *E. variatum, E. zonale,* and *E. blennioides* (Lachner *et al.* 1950); *E. squamiceps* (Page 1974); and *E. olivaceum* (Page 1980).

Males collected in February, March, and April had the most darkly pigmented fins and bodies, and the proportionally largest testes. Males collected from May until October had small, thin, clear testes, and nearly clear anal and pelvic fins. Large males collected in late October had slightly enlarged whitish testes and had developed some dark pigmentation on the fins.

Reproductive biology, females.-No female less than 55 mm SL had ovaries containing mature ova. All 2-year or older individuals and 65% of 1-year-olds

Table 8.-Distribution of year classes in *Etheostoma cinereum* from three drainages, with corresponding ranges and means of standard length.

Year class	n	Percent of individuals	Range SL	Mean SL	SD
		Cumberland Drainage			
0	15	19.5	29.6-47.3	39.8	5.00
1	36	46.8	39.2-67.1	56.1	7.15
2	25	32.5	58.5-78.9	67.7	5.50
3					
4	1	1.3	100.0	100.0	
		Duck Drainage			
1	12	57.1	49.8-64.2	57.1	4.00
2	6	28.6	67.8-82.1	72.1	5.45
3	2	9.5	79.9-88.5	84.2	6.12
4	1	4.8	89.4	89.4	
		Upper Tennessee Drainage			
0	23	30.7	32.6-49.5	42.7	4.56
1	42	56.0	42.9-66.2	56.6	6.59
2	8	10.7	60.5-79.1	70.6	6.16
3	2	2.7	85.1-85.9	85.5	0.57

collected during the breeding season had ovaries containing mature ova. The highest gonadosomatic indices were found in two females collected in March ($\bar{x}$ GSI = 225). Mature ova in these specimens were orange or translucent, averaged 2.0 mm in diameter (range = 1.7-2.4 mm, N = 20) and lacked the indentation on one side described for *E. fonticola*, *E. microperca*, and *E. proeliare* (Burr and Page 1978, 1979), and *E. olivaceum* and *E. striatulum* (Page 1980). The two ripest females contained 150 and 249 mature ova (Table 9), with an average of 53 mature ova per gram adjusted body weight (ABW). Small, short-lived species of darters, which spawn only once, typically have higher numbers of ova per gram ABW, e.g., 209 ova per gram ABW calculated for *E. proeliare* from data in Burr and Page (1978), and 114 eggs per gram ABW calculated for *E. simoterum* from data in Page and Mayden (1981). Larger species which spawn in more than one breeding season have values closer to that calculated for *E. cinereum*, e.g., 66 eggs per gram ABW calculated for both *E. squamiceps* from data in Page (1974) and *E. kennicotti* from data in Page (1975).

Females collected in May had the lowest gonadosomatic indices ($\bar{x}$ GSI for May = 2.4). Ovaries grew at an increasing rate from May until the peak breeding season in March and April. The relationship between GSI (Y) and month (X) with May taken as month 1, for females greater than 55 mm SL, was $\text{Log}_e\ Y = 0.30 + 0.15X$, with $r = 0.93$.

Sex ratio.-Of 104 individuals sexed, 62 were females and 42 were males, giving an overall ratio of 1.5 females to 1 male ($X^2 = 3.85$, $P < 0.10$). This skewed sex ratio, however, may be a manifestation of small sample size and/or sampling bias. Females outnumbered males in collections from the upper Tennessee drainage (34 to 15) and Duck drainage (13 to 2). However, more males than females (25 to 15) were collected in the Cumberland drainage. Most species of darters do not deviate significantly from a 1:1 sex ratio, except for the territorial and po-

Table 9.—Relationship between size, age, and ovary weight of *Etheostoma cinereum* females and the number of mature ova. An age of 1 year = 11-13 months, 2 years = 23-25 months.

Standard length (mm)	Adjusted body weight (g)[a]	Month of collection	Age in years	Weight of ovaries (g)	Number of mature ova	GSP
58	2.15	January	2	0.16	149	74
63	2.94	March	1	0.35	150	119
70	4.38	March	2	0.98	249	224
64	2.83	March	2	0.64	106	225
58	1.47	April	1	0.08	43 (partly spent)	54
60	1.62	April	1	0.08	51 (partly spent)	49

[a] Adjusted weight is the specimen's weight after removal of the ovaries, stomach, intestine, and liver.
[b] Equals weight of ovaries x 1000/adjusted body weight.

lygamous species of the subgenus *Catonotus,* in which females outnumber males (Lake 1936; Page 1974, 1975; Page and Burr 1976; Page 1980).

Spawning.—As judged by size, weight, and color of gonads, and period of breeding coloration, the spawning season of *E. cinereum* probably extends from late January to mid-April, with peak spawning probably in mid-March. Spawning habitat and spawning behavior are unknown, but the conical-shaped genital papilla of ripe females (Fig. 2) indicates that the species may be an egg burier or egg attacher (following the classification of Page, 1983).

Diet.—Of the 83 stomachs examined, only 72% contained at least 1 food item. The most common food items were chironomid larvae, larvae of the burrowing mayfly *Ephemera,* and oligochaetes. The abundance of *Ephemera* larvae and oligochaetes in the diet correlates the observation that the principal habitat of *E. cinereum* is sandy-bottomed pools where these organisms most frequently occur (Needham *et al.* 1935; Pennak 1978). Although the feeding behavior of *E. cinereum* was not observed, the elongate snout and papillose lips may be specializations for feeding on burrowing organisms. Less common items in the diet were, for the most part, either terrestrial organisms which had fallen into the water (e.g., formicids and cercopids), or aquatic organisms which are uncommon in the principal habitat of *E. cinereum* (e.g., *Ferrisia* and *Stenonema).*

Substantial differences were found among food items of specimens from the Cumberland (N = 39) and upper Tennessee (N = 35) drainages. Chironomids accounted for a larger proportion of the diet of upper Tennessee drainage specimens (84%) than of Cumberland drainage specimens (52%). *Ephemera* accounted for 25% of the diet and occurred in 46% of the stomachs examined in Cumberland drainage specimens but were only 3.3% of the diet of upper Tennessee drainage specimens. Oligochaetes also comprised a larger proportion of the diet of Cumberland drainage specimens (15.3%) than of upper Tennessee drainage specimens (2.2%). Too few specimens (2) from the Duck drainage were examined for stomach contents to make comparisons with other populations.

Food habits varied among size classes. Specimens less than 40 mm SL fed almost exclusively on chironomid larvae. Chironomids were less important in the diets of 41 to 60 mm SL specimens, with larger food items such as ephemeropterans and oligochaetes accounting for 30% of the diet. Only 50% of stomachs of specimens larger than 60 mm SL contained chironomids, with 40% of the diet being ephemeropterans, plecopterans, trichopterans, and other large food items. Microcrustaceans, important in the diets of smaller size classes of many species of darters, were not found in the stomachs of any specimens of *E. cinereum,* possibly because few specimens smaller than 40 mm SL were examined.

Seasonal variation in the importance of several food items in the diet was apparent. While chironomid larvae were eaten every season, *Ephemera* larvae were a major food item only from late spring through early fall, and plecopteran larvae were present in the diet only from April until June.

Parasites.—The only external parasites found on *E. cinereum* were fluke metacercariae; 76% of individuals examined had at least 1 metacercarial cyst, and 27% had 10 or more. The most heavily infested individual had 75 cysts. Cysts were present on individuals from all months in which the species has been collected.

Of the 83 specimens dissected, encysted nematodes were found on the livers and intestines of nine, with the most heavily infested individual containing five cysts. The intestines of three specimens each contained one acanthocephalan worm.

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