

LARVAL DEVELOPMENT OF LABORATORY-REARED ROSYLIP SCULPIN, *ASCELICHTHYS RHODORUS* (COTTIDAE)

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ABSTRACT

Larvae which hatched from egg masses collected at southwest Vancouver Island, Canada, were identified as *Ascelichthys rhodorus* and were successfully reared through transformation. A developmental series from yolk-sac larvae through newly settled juveniles (5.9-17.6 mm SL) is described and illustrated. Larvae hatch at approximately 6.0 mm SL and the yolk is absorbed by 6.5 mm SL. Notochord flexion begins between approximately 8.8 and 9.0 mm SL and is usually completed by 11.0 mm SL. Transformation to the juvenile stage begins between 12.0 and 13.0 mm SL and is complete in most of our larger specimens (15.0-16.0 mm SL).

Ascelichthys rhodorus larvae possess the following distinguishing characters: 1) pigment patterns along the ventral body and gut, 2) a pointed snout and moderately slender body as compared to other cottids, and 3) four prominent preopercular spines. A series of larvae is examined for meristic structures, including fin ray, vertebral and caudal development, and sequence of bone ossification. All structures except the caudal complex are ossified in our largest specimen (17.6 mm SL). Head and preopercular spination is discussed.

Minimum egg incubation time was 24 days (10°C); the minimum spawning period was 25 days. Larvae were examined for swimming behavior; older larvae maintained a relatively high speed schooling behavior throughout the planktonic phase. Settlement of juveniles started at 55-60 days, with ambivalence over reentry to the plankton until about 90 days, when settlement became permanent.

The rosy lip sculpin, *Ascelichthys rhodorus*, is a small (11-15 cm) intertidal and subtidal cottid species distinguished by smooth skin, a low spinous dorsal fin, the absence of pelvic fins, and a single hooked preopercular spine (Hart 1973). Little is known of its biology and development or its relationships within the family Cottidae (Howe and Richardson 1978³). The geographic range of *A. rhodorus* extends from Moss Beach, Calif., northward to Sitka, Alaska (Miller and Lea 1972), and localized populations are commonly found throughout this range (Howe and Richardson footnote 3).

We provide here the first published description of the larvae of *A. rhodorus* with notes on the development and behavior of the species in the aquarium environment.

MATERIALS AND METHODS

Egg Collection and Laboratory Rearing

On 23 March 1979, nine unidentified egg masses were collected from under boulders on a cobble beach, at the 0.9 m tide level, at Jordan River (southwest Vancouver Island, Canada; lat. 48°25'20"N, long. 124°03'30"W). The egg masses were incubated in flowing seawater of about 10°C and 27‰ salinity at the Vancouver Public Aquarium. Only three tanks were available for rearing larvae, so some larvae that hatched on different dates were mixed. Rearing tanks were of 1,000 l volume, with inflow rates of over 1 tank volume/day. Newly hatched *Artemia salina* nauplii were fed in excess numbers to larvae once daily. Larvae were killed and preserved (3% Formalin⁴ with sodium borate buffered seawater of 15‰ salinity) at weekly intervals until settlement from the planktonic stage started at 55-60 d. All preserved specimens were from two rearing tanks, one with a single sibling group and the other with a mixture of larvae from two separate hatching dates (Table 1). Surviving

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³Howe, K., and S. L. Richardson. 1978. Taxonomic review and meristic variation in marine sculpins (Osteichthys: Cottidae) of the northeast Pacific Ocean. Final Rep., NOAA NMFS Contract No. 03-78-MO2-120, 1 January 1978 to 30 September 1978, Northwest and Alaska Fisheries Center, National Marine Fisheries Service, NOAA, 2725 Montlake Blvd. East, Seattle, WA 98112. Unpubl. rep.

⁴References to trade names do not imply endorsement by the National Marine Fisheries Service, NOAA.

TABLE 1.—Age and sibling relationships for *Ascelichthys rhodorus* larvae used for descriptions.

Tank	Date killed (1979)	Age (days)	Relationship ¹
1	26 Mar	3	1
1	9 Apr	14/17	2
1	13 Apr	18/21	2
Not reared	17 Apr	<1	—
2	17 Apr	8	1
1	17 Apr	22/25	2
1	26 Apr	31/34	2
2	1 May	22	1
1	1 May	36/39	2
1	1 May	36/39	2
1	9 May	44/47	2
2	23 May	44	1
1	23 May	58/61	2

¹1 = siblings, all one age, same source.

2 = 2 sibling groups, mixed age, different source.

juveniles from both tanks were reared to maturity at the Vancouver Public Aquarium.

Taxonomic Specimens

Measurements

The following measurements were made on 64 unstained larvae of *A. rhodorus* (5.9-15.8 mm SL) using an ocular micrometer in a stereomicroscope:

Standard length (SL)—Snout tip to notochord tip prior to development of caudal fin, then to posterior margin of hypural bones.

Head length (HL)—Snout tip to posterior margin of opercle.

Snout to anus length—Distance along body midline from snout tip to a vertical line through center of anal opening.

Body depth at pectoral—Vertical distance from dorsal to ventral body margin at pectoral fin base.

Meristic Structures

A total of 49 larvae was cleared and stained for observation of various meristic structures and sequence of bone ossification. The following size ranges inadvertently were not preserved and are not represented in our discussion: 9.5-10.0 mm SL and 11.5-12.5 mm SL. Bone terminology follows Richardson and Washington (1980).

Specimens were stained using alizarin red and alcian blue (Dingerkus and Uhler 1977). Structures were considered ossified even if only slightly stained with alizarin red. Counts on stained larvae were made of dorsal fin spines and rays, anal fin rays, left pectoral fin rays, caudal

fin rays, branchiostegal rays, and abdominal and caudal vertebrae (including the terminal ural centrum). Counts of caudal fin rays in juvenile and adult *A. rhodorus* were made from radiographs of six specimens (46-101 mm SL) from the collections in the College of Fisheries, University of Washington, Seattle. Twenty adult specimens (57-99 mm SL) were also cleared and stained for examination of the caudal fin.

The problems and inconsistencies of head spination terminology in cottid larvae have been discussed by Richardson and Washington (1980). We follow their terminology by using names proposed for *Sebastes* spp. (Richardson and Laroche 1979). Head spines for *A. rhodorus* larvae were examined on cleared and stained specimens in order to determine the origin of the spines.

Illustrations of larvae were made with the aid of a camera lucida.

IDENTIFICATION OF *ASCELICHTHYS RHODORUS*

The eggs of *A. rhodorus* range from 1.7 to 2.0 mm in diameter. Larvae hatch at approximately 6.0 mm SL and the yolk is absorbed by 6.5 mm SL. Notochord flexion begins between approximately 8.8 and 9.0 mm SL and is usually complete by 11.0 mm SL. Transforming larvae (about 12.0-13.0 mm SL) were distinguished by a combination of characters including changes in pigmentation and ossification of fin rays. Our largest specimens (15.0-18.0 mm SL) were newly settled and exhibited increased juvenile pigmentation.

The work of Richardson (1981) attempts to organize the cottid genera from the northeast Pacific that have been divided into phenetic groupings based on larval characters. In the northeast Pacific, larvae of 25 of 40 genera are described and most of the genera can be placed in 6 groups. Several genera are ungrouped (e.g., *Enophrys*, *Gymnocanthus*, and *Myoxocephalus*).

The present study indicates that *Ascelichthys* is most similar to the genera of Richardson's Group 2 (*Paricelinus*, *Triglops*, *Icelus*, *Chitonotus*, and *Icelinus*) which all possess the following characters: 1) moderately slender body form; 2) pointed snout; and 3) four prominent preopercular spines. Most members of this group also have postanal ventral midline melanophores sometimes extending along the caudal fin base. Although Richardson considers Group 2 coher-

ent, some differences are found among the genera in degree of gut pigmentation, head spination, number and position of postanal ventral melanophores, and myomere counts.

In degree of gut pigmentation, *A. rhodorus* larvae have a moderate intensity of melanophores; the gut is not as dark as *Paricelinus* but is darker than *Chitonotus*. *Ascelichthys rhodorus* do not have as many head spines as some members of Group 2 (e.g., *Triglops* and *Paricelinus*), possessing only parietal and nuchal spines and lacking spines in regions of the postocular, post-temporal-supracleithrum, opercle, and cleithrum. There is much variation among Group 2 genera in the number of ventral melanophores ranging from none in some species of *Triglops* to over 40 in *Chitonotus* (Richardson and Washington 1980). Larvae of *A. rhodorus* are most similar to larvae of *Paricelinus* in ventral pigmentation by having approximately 20-30 melanophores in preflexion larvae and approximately 15-20 melanophores in postflexion larvae. Myomere counts may also be useful in distinguishing *A. rhodorus* larvae. Myomere counts for *A. rhodorus* are most similar to those reported for *Chitonotus* and *Icelinus* (<40). *Triglops*, *Icelus*, and *Paricelinus* have >40 myomeres (Howe and Richardson footnote 3).

The absence of pelvic fins in *A. rhodorus* does not distinguish the early larvae since in most cottids the pelvic fins are the last fins to develop. However, in larger postflexion specimens the lack of pelvic fins does help to distinguish the species.

DEVELOPMENT OF *ASCELICHTHYS RHODORUS*

Pigment Patterns

A total of 35 larvae was examined for changes in larval pigmentation (Fig. 1). The following discussion describes general trends in melanophore distribution.

In the head region, pigment on early preflexion larvae is usually scattered dorsally over the head and nape; posterior to the eye, heavy internal pigment occurs at the base of the brain (Fig. 1A). With development, pigment increases in the area of the head, snout, mouth, operculum, and internally around the brain (Fig. 1B-E). A distinct patch of melanophores occurs at the jaw angle, first appearing between 6.0 and 8.0 mm SL and then becoming less prominent as larvae

begin to transform (>12.0 mm SL). After 6.0 mm SL, pigment appears on the underside of the mouth along the median cartilage between the dentaries and urohyal (Fig. 1C). In the abdominal region, early larvae have a distinctly pigmented gut with large, stellate melanophores covering most of the abdominal cavity (Fig. 1A-C). Melanophores are also present on the isthmus and pectoral fin base of early larvae (Fig. 1B, C). With development, the external pigment covering the gut becomes more internal than external with only a few melanophores visible on the overlying skin (Fig. 1D).

An average of about 15 melanophores ($N=12$, range 11-22) line the ventral body midline in 0-8 d (6.1-7.9 mm SL) *A. rhodorus* larvae, beginning well posterior to the anus at about myomeres 11-15 (Fig. 1A-C). These ventral melanophores show much variation in size and spacing among individual specimens. In general, the spacing between melanophores decreases from anterior to posterior with the last few spots appearing close together. The size of melanophores does not follow any pattern although usually the first 2 or 3 anterior spots are larger than the posterior ones. In preflexion larvae between 22 and 36 d (8.8-9.5 mm SL), the ventral melanophores extend further forward beginning at about the fifth myomere posterior to the anus and increase in number to over 20 ($N=11$, range 23-28). Melanophores in the anterior half of the ventral midline pigment (about the first 12 spots) are more widely spaced and occur in the area where the anal fin is forming. In larger postflexion larvae at 44 d (10.2 mm SL), 15-20 ($N=25$, range 9-22) ventral midline melanophores are present with the pigment beginning just posterior to the anus (Fig. 1D, E). The anterior melanophores along the developing anal fin are larger and are becoming more diffuse as they extend into the fin. Transforming specimens have fewer ventral spots, usually about 10 ($N=23$, range 8-13), with most of them more internal than external (Fig. 1F). In these specimens, melanophores posterior to the completely developed anal fin appear more or less as a single row whereas those along the anal fin are aligned in a double row. In the tail region posterior to the ventral midline row of melanophores a group of caudal melanophores occurs near the tail tip on the early larvae (Fig. 1A). As the caudal fin develops, these melanophores begin to align in the area where the hypural bones are forming and in some specimens may extend onto the caudal fin (Fig. 1B, D).

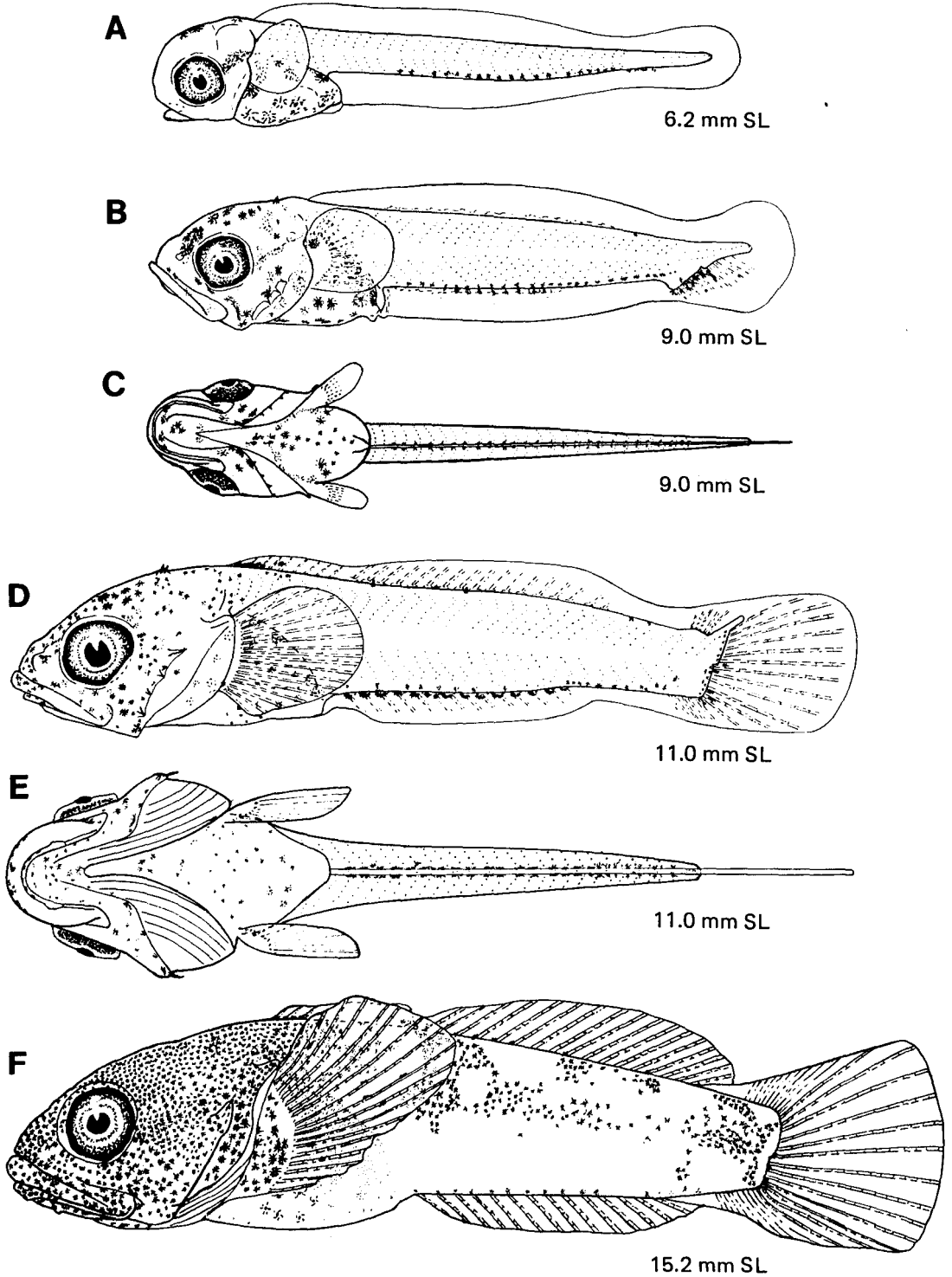


FIGURE 1.—Larval stages of *Ascelichthys rhodorus* showing changes in pigmentation: A. 6.2 mm SL; B. 9.0 mm SL; C. 9.0 mm SL (ventral view); D. 11.0 mm SL; E. 11.0 mm SL (ventral view); F. 15.2 mm SL.

Little pigment is added until the onset of transformation, except on the head, nape, and in the dorsal, anal, and caudal finfolds. Pigmentation changes occurring at the beginning of transformation are visible as early as 44 d after hatching (10.0-11.0 mm SL) but are not consistently visible until the 47th day (13.0 mm SL). During transformation *A. rhodorus* larvae show a rapid increase in pigmentation of all areas of the head and nape, and on the anterior dorsal body surface over the gut. A few melanophores appear in the dorsal portion of the postanal body becoming patches of pigment in the upper region dorsally and laterally (Fig. 1F). Melanophores also appear in the posterior caudal peduncle area. Early juveniles of the transformed, newly settled *A. rhodorus* have small, densely concentrated melanophores on the entire head, and several spots on the overlying skin over the gut cavity in addition to internal melanophores (Fig. 1F). The juveniles also have internal pigment along the notochord, and several distinctive groups of melanophore patches in the postanal body region along the upper body and in the caudal peduncle area. Dorsal body pigment on the largest specimens (17.0 mm SL) occurs in about five patches located under the posterior portion of the first dorsal fin, at the anterior, posterior, and center of the second dorsal fin, and in the posterior caudal area.

Morphology (Tables 2, 3)

Head length of *A. rhodorus* as a proportion of standard length increases with development and becomes almost one-third the standard length in early juveniles. Head length increases from 21.3% SL in preflexion larvae to 25.4% SL in larvae undergoing flexion. Values for head length continue to increase to 29.5% SL in postflexion larvae and 31.1% SL in transforming specimens. The head length of adult rosy lip sculpin is slightly larger than our postflexion and transforming larvae; adult head lengths are generally about 37% SL (Hart 1973).

Eye diameter as a proportion of head length decreases with development. Preflexion larvae have diameters over half the size of the head (50.8% HL), decreasing to 36.6% HL in transforming larvae. Eye diameter continues to decrease in adult rosy lip sculpin, usually measuring about 25% HL (Hart 1973).

Snout to anus length as a proportion of standard length increases with development. Snout

TABLE 2.—Morphometrics (in millimeters) of larvae and juveniles of *Ascelichthys rhodorus*. Approximate interval of notochord flexion is between dashed lines and interval of transformation is between solid lines.

Age ¹ (days)	Standard length	Head length	Eye diameter	Snout to anus length	Body depth at pectoral
0	5.9	1.2	0.7	2.0	1.2
0	6.2	1.2	0.7	2.0	1.2
0	6.2	1.3	0.7	2.0	1.3
0	6.3	1.3	0.7	2.0	1.2
0	6.3	1.3	0.7	2.0	1.2
0	6.3	1.3	0.7	2.0	1.2
0	6.3	1.3	0.7	2.0	1.2
0	6.3	1.3	0.7	2.0	1.3
0	6.4	1.3	0.7	2.2	1.2
8	5.4	1.0	0.6	2.0	1.2
8	5.8	1.2	0.7	1.8	1.1
8	6.1	1.4	0.6	2.2	1.3
8	6.8	1.5	0.7	2.4	1.1
8	6.9	1.6	0.7	2.4	1.1
8	7.1	1.5	0.7	2.2	1.2
8	7.1	1.6	0.7	2.6	1.2
8	7.2	1.5	0.7	2.3	1.2
8	7.2	1.7	0.7	2.4	1.1
8	7.6	1.8	0.7	2.7	1.3
8	7.9	1.8	—	2.5	1.3
22	8.5	2.2	0.9	3.3	1.9
22	8.5	2.1	0.9	3.1	1.9
22	8.5	2.1	0.9	3.3	1.8
22	8.8	2.5	0.9	3.6	2.0
22	8.9	2.1	0.9	3.4	1.9
22	8.9	2.1	0.9	3.6	2.0
22	9.0	2.5	0.9	3.4	2.0
22	9.0	2.3	0.9	3.4	2.1
22	10.5	3.0	1.0	4.6	2.5
36/39	8.5	2.1	0.9	3.0	1.8
36/39	8.5	2.0	0.9	3.3	1.7
36/39	8.6	2.2	0.9	3.3	1.9
36/39	8.6	2.2	0.9	3.5	1.8
36/39	8.8	2.2	0.9	3.3	1.9
36/39	8.8	2.1	0.9	3.5	2.0
36/39	8.8	2.1	0.9	3.5	1.9
36/39	8.9	2.2	0.9	3.5	2.0
36/39	9.5	2.5	—	3.9	2.1
36/39	9.5	2.4	0.9	3.6	2.0
44	10.1	2.8	1.0	4.5	2.6
44	10.1	2.9	1.1	4.5	2.2
44	10.1	3.0	1.1	4.5	2.4
44	10.2	3.0	1.1	4.6	2.4
44	10.3	3.0	1.1	4.5	2.6
44	10.5	3.0	1.1	4.7	2.4
44	10.5	3.3	1.2	4.9	2.6
44	10.9	3.4	1.2	5.3	2.6
44	11.0	3.4	1.2	5.2	2.5
44	11.0	3.1	1.2	4.9	2.6
44/47	12.8	3.8	1.5	6.1	3.3
44/47	13.0	3.9	1.5	6.5	3.6
44/47	13.3	3.7	1.5	6.5	3.7
44/47	13.3	4.0	1.5	6.4	3.6
58/61	13.3	3.9	1.6	6.5	3.8
58/61	13.8	4.9	1.7	7.0	3.4
58/61	16.8	4.8	1.7	8.5	3.9
58/61	16.0	5.2	1.7	8.2	4.2
58/61	17.6	5.1	1.8	9.1	4.8
58/61	13.0	4.0	1.5	6.3	2.9
58/61	13.1	4.0	1.6	6.3	2.9
58/61	14.0	4.9	1.6	7.0	3.2
58/61	15.0	5.0	1.7	7.7	3.4
58/61	15.8	5.2	1.7	7.8	3.9

¹Two ages separated by a slash indicates a mixed age group, different sibling groups (see Methods).

length to anus length increases from one-third the standard length (33.2% SL) in preflexion larvae to 39.0% SL in larvae undergoing flexion.

TABLE 3.—Body proportions of larvae and juveniles of *Ascelichthys rhodorus*. Values given for each body proportion are expressed as percent of standard length (SL) or head length (HL): mean, standard deviation, and range.

Body proportion	Preflexion	Flexion	Postflexion	Transforming
Sample size	21	19	10	14
Standard length (mm)	6.5±0.6 (5-8)	8.9±0.5 (8-11)	10.5±0.4 (10-11)	14.3±1.6 (13-18)
Head length/SL	21.3±1.4 (18-24)	25.4±1.6 (24-29)	29.5±1.3 (28-31)	31.1±2.4 (28-36)
Eye length/HL	50.8±6.4 (38-60) ¹	40.5±3.6 (30-45) ²	36.6±1.1 (35-39)	36.6±3.0 (33-41)
Snout to anus/SL	33.2±2.0 (31-37)	39.0±2.0 (35-44)	45.4±1.6 (44-49)	49.6±1.3 (48-52)
Body depth at pectoral fin base/SL	18.5±1.9 (15-22)	22.0±0.9 (20-24)	23.8±1.2 (22-26)	25.2±2.3 (22-29)

¹Sample size = 20.

²Sample size = 17.

Values for snout length to anus length continue to increase in postflexion larvae to 45.4% SL and to almost half the body length (49.6% SL) in transforming larvae.

Body depth at the pectoral fin base increases only slightly with development. Preflexion larvae have a body depth of 18.5% SL and values increase to 25.2% SL in transforming larvae. Adult body depths are usually about 28% SL (Hart 1973).

Meristic Structures (Table 4)

The following discussion of the development of meristic structures describes only general trends, as specimens show much variation in the sequence of bone ossification and our collection does not include all size ranges. Variation occurs frequently in the size of larvae with respect to the development of meristic structures. In general, the development of meristic characters appears dependent on size rather than age. Different growth rates as seen in standard length differences among individuals and between tanks are also reflected in the development of meristic structures (Tables 1, 4).

Oral Region

Branchiostegals are the first meristic structures to develop as ossification occurs as early as 6.8 mm SL. The full complement of six branchiostegals (seven in a few specimens) is not consistently ossified until the larvae are 9.0 mm SL.

Gill arches are stained blue by 9.0 mm SL and most begin to ossify between 8.8 and 9.5 mm SL. Ossification of gill rakers is complete by 13.3 mm SL.

Axial Skeleton

Abdominal and caudal centra begin to form

between 8.8 and 9.0 mm SL, and development proceeds from anterior to posterior with the first signs of ossification occurring in larvae between 8.8 and 9.5 mm SL. Abdominal centra are completely ossified in 10.2 mm SL larvae. Caudal centra begin to ossify in 10.0 mm larvae and ossification of the completed vertebral column appears in 12.8 mm SL larvae.

Neural and haemal spines begin to ossify in larvae between 8.8 and 10.2 mm SL. All neural spines in the abdominal area are ossified by 10.2 mm SL, and the remaining neural spines in the caudal area are complete by 12.8-13.3 mm SL. Haemal spines took up red stain in our 10.2 mm SL larvae but are not completely ossified until 12.8-13.3 mm SL. Ossification of both neural and haemal spines proceeds anterior to posterior with the last neural and haemal spines associated with the caudal complex the last to ossify.

Fin Development

In general, all fins except caudal fin rays begin to ossify at 10.2 mm SL. Dorsal spines and pectoral fin rays are completely ossified by 12.8 mm SL, and dorsal and anal fin rays are fully ossified in 13.3 mm SL specimens.

The caudal complex begins to ossify with the hypural bones in larvae between 12.8 and 13.3 mm SL. The following description is based on our available specimens although our largest juvenile (17.6 mm SL) does not have the full complement of ossified caudal fin rays.

The caudal fin is associated with a complex of 4-5 centra (1 ural and 3-4 preural centra), 3-4 neural and 3-4 haemal spines, 3 epurals, 1 superior hypural (HY 4-5), 1 inferior hypural (HY 1-3), and 1 pair of uroneurals (Fig. 2). Caudal fin rays total 31-37 of which 10-13 are superior secondary fin rays and 8-11 are inferior secondary fin rays. Principal caudal fin rays supported by the hypural bones number 13 (6 are supported by the

TABLE 4.—Meristic features of larval and early juvenile *Ascelichthys rhodorus*. Approximate interval of notochord flexion is between dashed lines and interval of transformation is between solid lines.

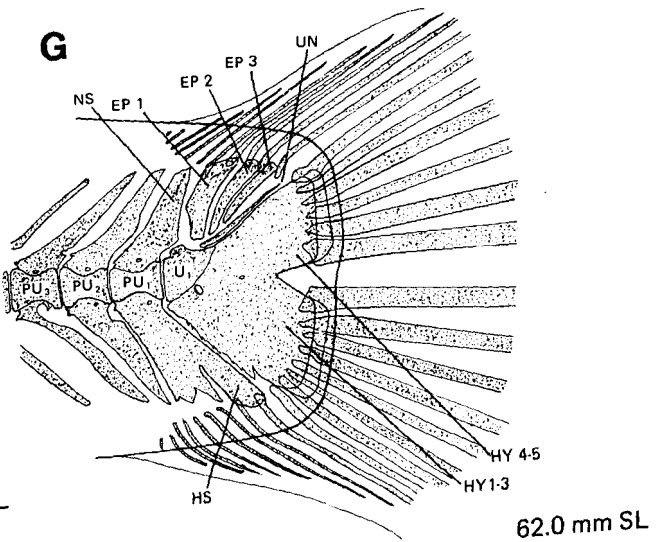
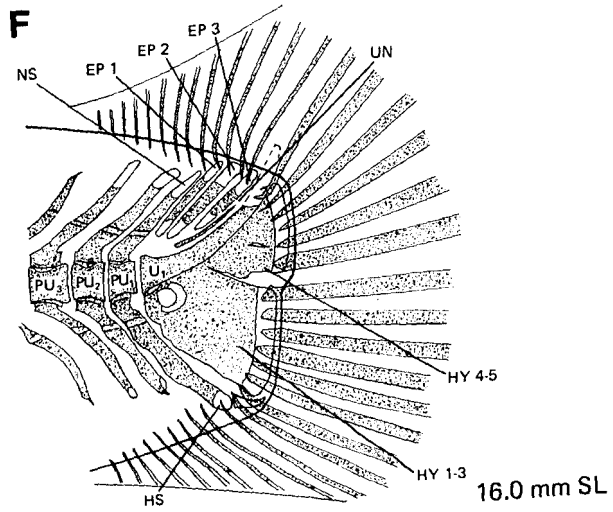
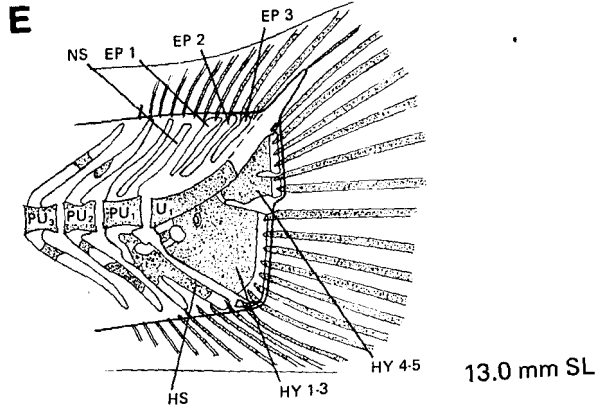
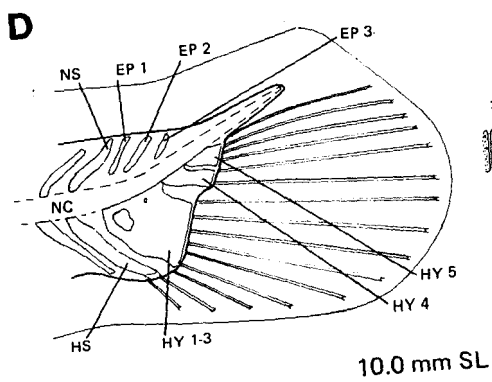
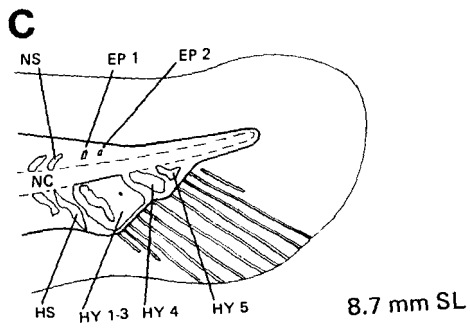
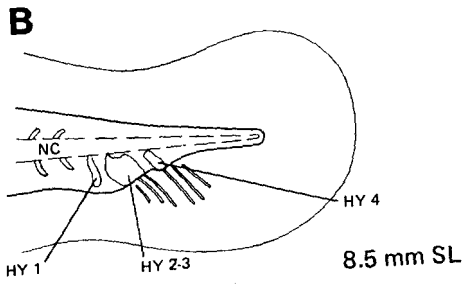
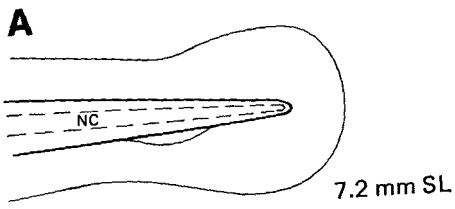
Standard length (mm)	Age ¹ (days)	Fins						Vertebrae			Branchio-stegals	Gills ²	Head spines ³					
		Dorsal		Anal rays	Pectoral rays	Caudal		Centra		Preopercular				Parietal	Nuchal			
		Spines	Rays			Principal	Total	Abdominal	Caudal	1st			2d			3d	4th	
6.1	8											-	-	-	-	-	-	
6.2	0																	
6.3	0																	
6.3	0																	
6.8	8																	
7.2	8										2							
7.7	14/17										3							
7.9	8																	
7.9	14/17										2							
7.9	14/17										2							
8.0	22										6							
8.2	14/17										4							
8.4	14/17										2							
8.5	22										6							
8.7	14/17										4							

8.8	22										6			+	+	+	+	
8.8	22										6			+	+	+	+	
8.8	22							1			6			+	+	+	+	
8.9	14/17										3							
9.0	22										6		X	+	+	+	+	
9.0	22										6			+	+	+	+	
9.5	22									2	6			+	+	+	+	
9.5	22										6			+	+	+	+	
10.0	31/34							9	2		6+7							
10.0	31/34							3+3	6	11	5							
10.2	36/39							3+3	6	9	4							
10.2	36/39								11	4	7+7							
10.2	44	IX	19	16	16	4+4	8	11	11	11	6			+	+	+	+	
10.2	44	IX	18	15	16	5+6	11	10	10	10	7+6			+	+	+	+	
10.6	31/34					4+4	8	10	5	6	6							
10.8	31/34					5+4	9	10	8	6	6							
10.8	36/39					4+4	8	11	4	6	6							

11.1	31/34					4+5	9	11	8	6	6							
11.2	36/39					4+4	8	11	10	7+6	6							
11.3	36/39					3+3	6	10	4	6	6							

12.8	44/47	IX	13	13	18	6+7	20	10	24	6	6		XX	+	+	+	+	
13.3	44/47	IX	15	12	17	6+7	19	10	24	6	6			+	+	+	+	
13.3	58/61	IX	18	15	17	6+7	24	10	25	6	6			+	-	-	-	
13.8	58/61	IX	19	14	17	6+7	29	10	25	6	6			+	-	-	-	
16.8	58/61	IX	19	15	17	6+7	31	10	25	6+7	6			+	-	-	-	
17.6	58/61	IX	18	14	17	6+7	39	10	25	6	6			+	-	-	-	

¹Two ages separated by a slash indicates a mixed age group, different sibling groups (see Methods).²Gill rakers were not counted; X denotes onset of ossification of gill arches and XX denotes completed ossification of gill rakers.³+ denotes spine present, - denotes spine absent.



superior hypural and 7 are supported by the inferior hypural). Ahlstrom⁵ generalized that all members of the family Cottidae probably have a total of 12 principal caudal rays, 6 supported by the superior hypural and 6 supported by the inferior hypural. Verification of this principal fin ray count came from counts on 20 adult specimens acquired for this study, 19 of which actually had a 6+7 count. Richardson⁶ has also observed a number of exceptions to a 6+6 count in other members of the family Cottidae.

A symmetrical fin fold surrounds the tip of the notochord in newly hatched specimens 6.0 mm SL. In 7.2 mm SL larvae, a thickening is visible ventral to the notochord (Fig. 2A). By 8.5 mm SL, the ventral thickening is differentiated into three cartilaginous plates (Fig. 2B). The anterior plate represents hypural 1 (parhypural) followed by a larger plate presumably representing the fusion of hypurals 2 and 3. Posterior to hypurals 2 and 3, a third plate represents hypural 4. A few caudal fin rays are also visible by 8.5 mm SL. In slightly larger larvae of 8.7 mm SL the urostyle is just beginning to undergo notochord flexion, and the unossified hypural 1 has fused with hypurals 2 and 3 forming the inferior hypural plate (Fig. 2C). Also in 8.7 mm SL larvae, differentiation of hypural 5, and epurals 1 and 2 is visible (Fig. 2C). In larvae undergoing notochord flexion (10.0 mm SL) unossified hypurals 4 and 5 have begun fusing to form the superior hypural plate (Fig. 2D). We did not detect fusion of a sixth hypural bone during the formation of the superior hypural plate. If a sixth hypural bone develops late in the larval period as it does in the phylogenetically related blackgill rockfish, *Sebastes melanostomus*, (Moser and Ahlstrom 1978), it was not evident in the juveniles or adults we examined. The first appearance of unossified epural 3 also occurs in specimens about 10.0 mm SL (Fig. 2D). Ossification proceeds rapidly once the larvae have undergone notochord flexion. By

13.0 mm SL, the ural centrum and all preural centra are ossified (Fig. 2E). The single ural centrum is not fused to the first preural centrum. Hypural bones, neural and haemal spines, and caudal fin rays have also begun ossifying in 13.0 mm SL specimens (Fig. 2E). By 16.0 mm SL, a completely ossified pair of uroneurals is visible dorsad to the urostyle (Fig. 2F). All three epurals have begun to ossify, thus completing the caudal complex except for a few unossified secondary caudal fin rays. This caudal complex of a 16.0 mm SL early juvenile resembles in all details that of a 62.0 mm SL adult (Fig. 2G). In a number of specimens abnormalities of the last neural and haemal spines were observed, e.g., double neural spines from the first preural centra (Fig. 2E) and a large flattened haemal spine (Fig. 2G).

Spination

Four similar-sized preopercular spines are ossified on specimens 8.8-9.0 mm SL (Fig. 1B). In 10.2 mm SL larvae the upper preopercular spine is larger than the lower three (Fig. 1D). After transformation the three lower spines are no longer visible, leaving only the prominent upper spine (Fig. 1F). The single hook shaped spine is visible on our largest specimens, appearing very similar to the single, recurved spine for which the adults are commonly known.

On specimens 8.8-9.0 mm SL, one small, parietal spine is evident (Fig. 1B). This spine remains prominent and is joined by a nuchal spine in 10.2 mm SL larvae (Fig. 1D). The parietal and nuchal spines are no longer visible in larvae >12.8 mm SL (Fig. 1F).

REPRODUCTIVE BEHAVIOR AND LARVAL REARING

Egg masses of *A. rhodorus* were found wedged in irregular spaces among rocks under larger boulders, but the eggs adhered only to other eggs, not to rock surfaces. No egg masses were found under boulders lying on sand, shell, gravel, or solid rock surfaces. The egg masses were taken only in a narrow band at the low water level, which was the lowest tidal level during March and early April 1979. This cobble beach had been repeatedly searched for fish eggs during lower tides before and after the period of the vernal equinox, i.e., in December, January, April, May, and June of previous years, but this kind of egg was only found during the moderate

⁵E. H. Ahlstrom, Southwest Fisheries Center, National Marine Fisheries Service, NOAA, La Jolla, CA 92038, pers. commun., class notes, 1971. (Deceased.)

⁶S. L. Richardson, Gulf Coast Research Laboratory, East Beach Drive, Ocean Springs, MS 39564, pers. commun. February 1981.

FIGURE 2.—Development of the caudal fin of *Ascelichthys rhodorus*: A. 7.2 mm SL; B. 8.5 mm SL; C. 8.7 mm SL; D. 10.0 mm SL; E. 13.0 mm SL; F. 16.0 mm SL; G. 62.0 mm SL. EP = epural; HS = haemal spine; HY = hypural; NC = notochord; NS = neural spine; PU = preural centrum; U = ural centrum; UR = uroneural. Ossified elements are stippled.

low tides of the vernal equinox. In April 1981, *A. rhodorus* eggs were found in the same area, but about half were dead while the remainder all hatched upon return to the laboratory. Perhaps the protracted exposure to air in April killed many of the earlier embryonic stages; an increase in embryonic temperature tolerance with development has been documented for another intertidal cottid, *Clinocottus acuticeps* (Marliave 1981a). Considering a relatively dense spawning of about one mass/3 m² found in March 1979, and the lack of egg masses at other times, this species might be characterized by a brief spawning season.

Superficially, *A. rhodorus* eggs resembled *Hexagrammos* spp. eggs in size (about 2.0 mm) and color, although there were far fewer eggs per mass. As with *Hexagrammos* spp., newly spawned eggs were a semitranslucent blue to purple, grading toward opaque white toward the egg center (personal observation by J. B. Marliave). Eggs with advanced embryos, showing guanine eye pigment, appeared brown overall, due to melanophores overlying dark olive yolk material. All egg masses were incubated and hatched in the laboratory; none were used for egg counts but some egg diameter measurements were taken. Hatching occurred on March 23 (1 mass) and 26 (1 mass) and on April 9 (3 masses) and 17 (4 masses). This range of hatching dates indicated a minimum spawning period of 25 d. The collection and final hatch dates indicated a minimum egg incubation period of 24 d at 10°C.

During the planktonic larval stage, larvae of *A. rhodorus* displayed relatively high-speed schooling behavior and a marked tendency toward startle responses. It is of note that both this species and another common northeastern Pacific Ocean fish, *Trichodon trichodon* (Marliave 1981b), are rare or unknown from plankton samples and school soon after hatching in the confines of a tank. Unlike *T. trichodon*, however, *A. rhodorus* larvae do not school immediately upon hatching but develop schooling behavior within 2 wk of hatching. *Ascelichthys rhodorus* larvae do not swim as fast as those of *T. trichodon*; *A. rhodorus* cruised at 2.5-7.5 body lengths/s at 2 wk of age, at 3-10 body lengths/s at 4 wk, and at 2.5-9.0 body lengths/s at 6 wk, with usual speeds close to 5 body lengths/s. From hatching onward, the *A. rhodorus* larvae were very easily disturbed, either by physical interference from other types of zooplankton, by movements of observers, or by abrupt changes in lighting. Startle responses

were characterized by rapid bursts of undirected swimming which, in older larvae, effected the breakdown of schools.

After 2 wk, larval *A. rhodorus* schooled near the surface at all ages except for those larvae in tanks with larval shrimp, *Pandalus danae*. The *P. danae* occupied the surface layers and *A. rhodorus* schooled off the tank bottom until the *P. danae* settled from the plankton, after which *A. rhodorus* schooled near the surface. This pattern occurred successively in two separate tanks; no shrimp were present in the third tank. Thus, the vertical distribution of the *A. rhodorus* larvae was modified by the presence of other planktonic organisms.

Settlement to the bottom started at 55-60 d of age (14-18 mm SL) and schooling generally ceased. However, for unknown reasons all larvae in a tank would temporarily resume schooling from time to time. Between 60 and 90 d, there was a gradual increase in the proportion of settled larvae with no observed difference in feeding behavior between settled and schooling fish. By 90 d of age, the majority of juveniles were permanently settled and no further schooling was noted. Among cottids, protracted ambivalence about settlement from the plankton has been observed in *Gilbertidia sigalutes* (Marliave 1981c).

After initial settling was observed, substrate trays containing sand, gravel, and pebbles were placed in the tanks to determine substrate preferences of the larvae (Marliave 1977), but the trays were avoided. Larval *A. rhodorus* never settled against vertical surfaces, as is typical of a variety of other cottids which lack discreet substrate preferences (personal observation by J. B. Marliave). Settlement was typically on open bottom throughout the month of ambivalence between settlement and reentry to the plankton.

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