

Early Life History of the Arrow Goby, *Clevelandia ios* (Jordan & Gilbert), Gobiidae¹

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Abstract—The arrow goby, *Clevelandia ios*, hatches over estuarine mud flats and remains entrained in shallow water until after caudal fin formation, when schooling behavior commences in the extreme nearshore. All larval stages are illustrated. Settlement is preceded by fusion of the pelvic fins and coincides with pigment pattern development, whereas burrow entry correlates most closely with completion of pigment pattern development. Laboratory tests indicate an experience component to latency times in burrow entry. Juveniles prefer sand substrate, but no preference for presence of burrows or ghost shrimp (*Callinassa californiensis*) in substrates was evident.

Introduction

The arrow goby, *Clevelandia ios*, inhabits estuarine mud flats from Southern California to British Columbia (Hart 1973) and often dwells in worm or shrimp burrows as a facultative commensal (Brothers 1975, MacGinitie 1935). Arrow gobies occur at the highest densities where burrowing hosts also occur (Prasad 1948). Spawning peaks in early summer in California (Macdonald 1972). Larvae hatch at 2.7–3.8 mm SL within 10–12 days at 15°C, and are found in surface waters of estuaries (Brothers 1975, Prasad 1948). Larval life history involves two habitat shifts: first, from planktonic distribution in surface waters to a schooling demersal habit, and second, settlement to a benthic juvenile habit (Brothers 1975). Around the time of this second habitat shift, fusion of pelvic fins and pigment deposition occur, along with initiation of burrow-entry by juveniles (Macdonald 1972). Gobies are absent from the intertidal during winter (Hoffman 1981).

The purpose of the present study was to determine the extent of drift dispersal of larvae, the distribution of schooling larvae and the timing of the transition from larval to juvenile behavior. Associations between the onset of burrow entry and morphology were examined, as well as substrate preferences of settling larvae and the role of learning in burrow-entry. Matarese et al. (1989) describe only the newly hatched larva and the settled juvenile of *C. ios*; the present paper provides the first complete larval description for the species.

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Materials and Methods

Schooling larval stages were collected from outlet streams draining from pools on Spanish Banks mudflats in English Bay (southern shore), Vancouver, B.C. (49°17'N, 123°14'W) on 25 July, 1990 by using fine meshed dip-nets while wading during low tides. Larvae were transported to the laboratory and reared in 100 L tanks with through-flowing seawater of 24‰ salinity and 10–13 C temperature, and were fed Selco-enriched *Artemia* nauplii daily. Larvae were reared to the juvenile stage as "naive" or "experienced" with respect to exposure to artificial burrows (Grossman 1980). The "experienced" larvae were reared in a 100 L glass tank with a false bottom with 13 mm ID tygon tubes in counter-sunk holes to create U-shaped burrows. Once gobies in these burrow tanks were well into the juvenile stage, they were transferred to plain 100 L tanks and held in the same manner as "naive" gobies until testing. Adult and juvenile gobies were collected on 3 October 1990 from *Callinassa californiensis* (ghost shrimp) burrows at Boundary Bay, Wash. (49°00'N, 123°10'W) using a 16 mm ID tygon siphon tube, and were held in the laboratory as "wild" gobies.

Clevelandia ios used for morphological comparison were collected from both sites from July to September 1990 and fixed in 3% phosphate buffered formalin. Burrow-entering gobies were siphoned from shrimp burrows in shallow pools, and schooling larvae were dip-netted from deeper pools. Juveniles that were chased with a dip-net for at least five seconds and passed within three body lengths of at least five burrow openings without entering were categorized as "non burrow-entering."

In 1994, plankton tows were conducted south of Spanish Banks, on Roberts Bank and Sturgeon Bank, the main mudflats of the Fraser River Delta. These tows were 5 min duration with a metered SCOR net (Marliave 1989) at depths within one meter of the bottom at 3, 6 and 10 m bottom depth, and 0.5 km offshore (from dropoff, or break-in-slope) over 100 m bottom at 5, ca. 10 and ca. 20 m towing depth. All larvae collected in plankton nets were fixed in 2% formalin in seawater, then sorted in the laboratory and preserved in 3% phosphate buffered formalin for later identification and analysis.

Positive identification of larvae was determined using characters described for preflexion larvae by Matarese et al. (1989). All developmental observations and standard length (SL) measurements were done under a dissecting microscope. Illustrations were drawn using a dissecting microscope and camera lucida.

Behavioral testing involved a series of substrate-preference tests and some tests on latency of burrow entry. When larvae were starting to settle, plastic trays were introduced with different substrates, and visual counts of numbers settled on each tray were made at 24 hour intervals. Ambivalence over settlement permitted repeated observations without requiring removal of fish settled at the time of any one counting. Trays were rotated between counts to control for artifacts due to flow patterns in the tanks. In the first settlement experiment, sand, clay, and bare tray bottom were used as substrates. The sand was from a *Callinassa californiensis* population at Spanish Banks and the clay was from a *Upogebia pugettensis* (mud

shrimp) population at Boundary Bay. In the second substrate test, sand was compared to sand with artificial burrows (five U-tubes each of 4, 9, 13 mm ID; 15 tubes total). The third settlement experiment compared sand to sand with *C. californiensis* (four of various sizes from Boundary Bay). Totals of successive counts in each experiment were examined with Chi-square, using Yate's corrected Chi-square value where degrees of freedom equalled one.

Burrow-entry behavior was tested with an artificial burrow tank similar to that used for rearing the "experienced" group of gobies. A double, black plastic blind was constructed around the tank to allow screened observations from between the blinds. Black plastic also covered the tank sides below the level of the false bottom, so that observations of gobies in burrow tubes could be made by lifting a flap in the plastic. Fish were released into the tank from a clear plastic cylinder (45 mm ID) in the tank center, by means of lifting the cylinder via an overhead pulley cord after ten minutes acclimation for each single fish. Latency time was recorded for each goby from the time of its first movement until entry into a burrow. If, after ten minutes, a goby had failed to enter a burrow the trial was ended with no latency time recorded. Observations continued for five minutes following burrow entry. Results were examined with one-factor analysis of variance, using log transformation of latency time to accommodate skewed data. Post-hoc comparisons were carried out using the Scheffe F-test.

Results

Plankton tows yielded large catches at 3m bottom depth, with the net within one meter of the substrate. These larvae were in yolksac and preflexion stages (Fig. 1, a and b). The relative catches, illustrated in Figures 2 and 3, reveal that larvae associate with shallow bottom, with highest densities at the shallowest bottom depth sampled (3m). About 5 yolksac larvae per cubic meter were caught in bottom tows at 3m depth, versus $0.8/m^3$ for preflexion larvae. In midwater tows, catch rates were lower than for bottom tows except for catches of $0.07/m^3$ for preflexion larvae (Fig. 3) at both 10m bottom depth and midwater at 3m over the break-in-slope (ca. 100m bottom depth).

Larvae dipnetted in outlet streams in 1990 were all in the flexion stage illustrated in Figure 1 c. These larvae were schooling in the extreme nearshore, in less than half a meter depth (usually ca. 20–30 cm depth). Schooling behavior was not, however, pronounced in the confines of a laboratory tank without strong currents.

Schooling *Clevelandia ios* larvae dip-netted from pools under 1 m deep ranged from 9.1 to 13.0 mm SL ($n=35$) with an average length of 10.9 ± 0.8 mm SL (Fig. 1 c and d). All of these larvae from pools had completed notochord flexion. The majority (91%) of these schooling larvae possessed a characteristic larval pigment pattern (Fig. 1 d), consisting of ventral midline, ventral gut and dorsal swimbladder melanophore rows, and a single large melanophore on the posterior dorsal midline (Matarese et al. 1989). The other 9% were transitional between larval and juvenile pigment (Fig. 1 e); this transitional stage was characterized by several series of melanophore rows (internal dorsal notochord, mediolateral and

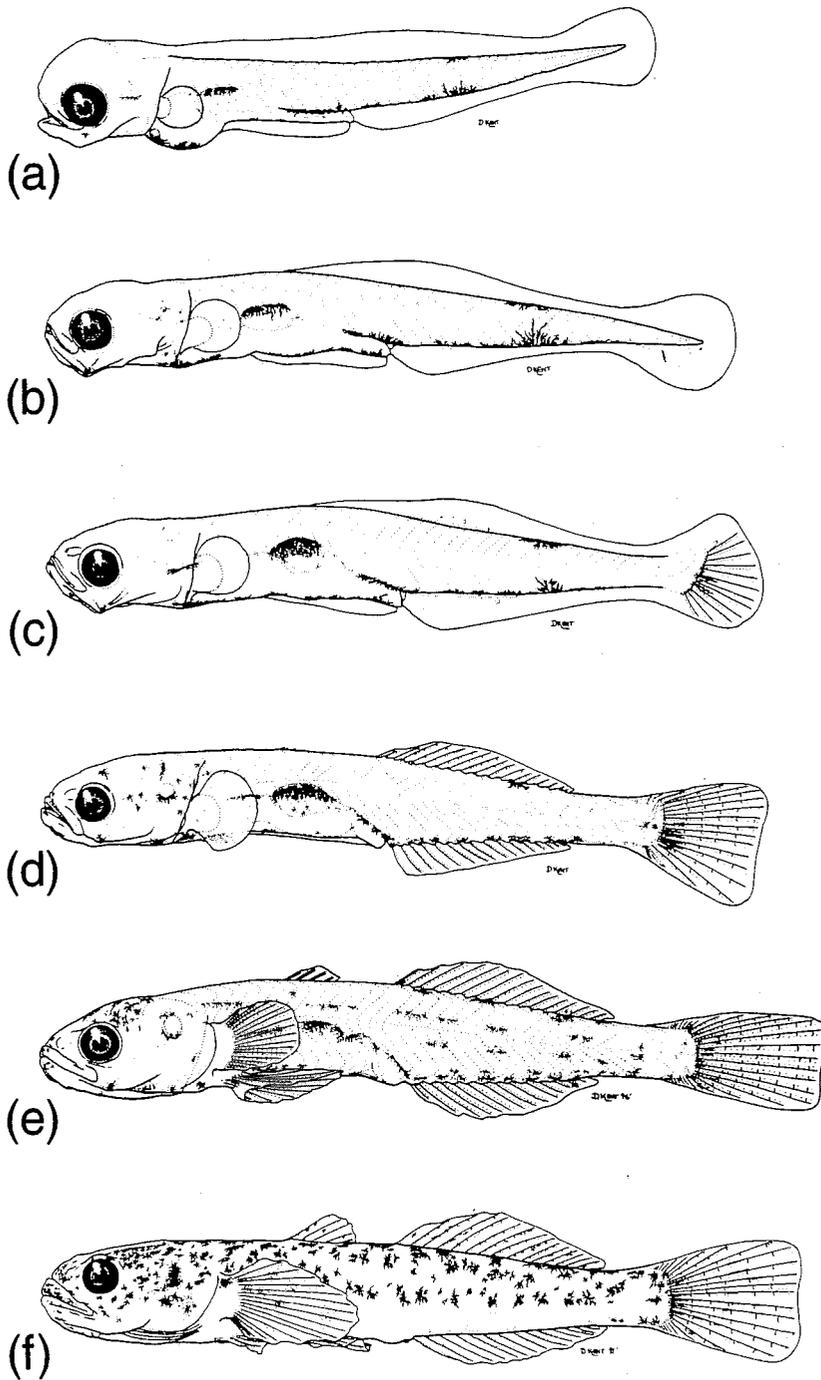


Figure 1. Larval development of *Clevelandia ios*: (a) 3.4 mm SL yolk sac larva; (b) 5.8 mm SL preflexion larva; (c) 7.0 mm SL flexion larva; (d) 10.2 mm SL postflexion larva; (e) 12.5 mm SL transitional juvenile; (f) 15.6 mm SL juvenile.

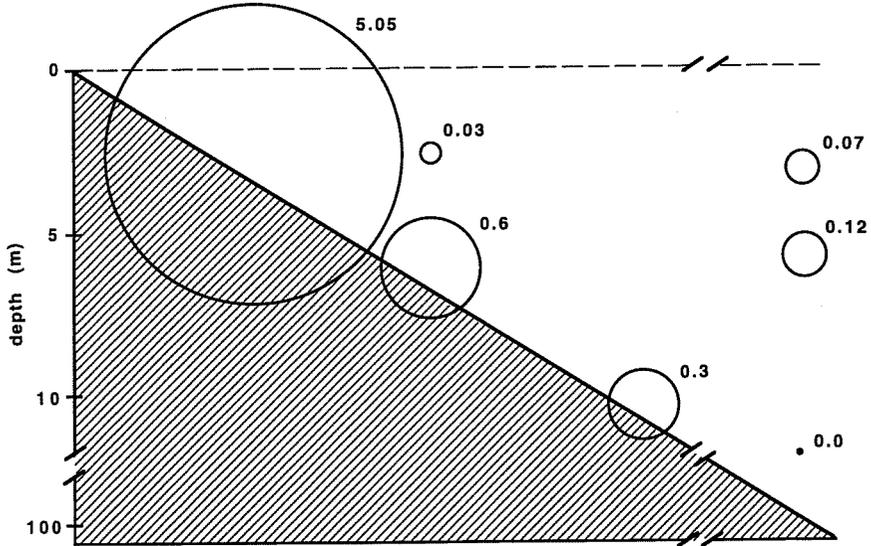


Figure 2. Catch densities at nearshore stations with respect to depth for yolk sac larvae of *Clevelandia ios*. Circle areas indicate catch densities, with average number of larvae per cubic meter indicated adjacent to each circle.

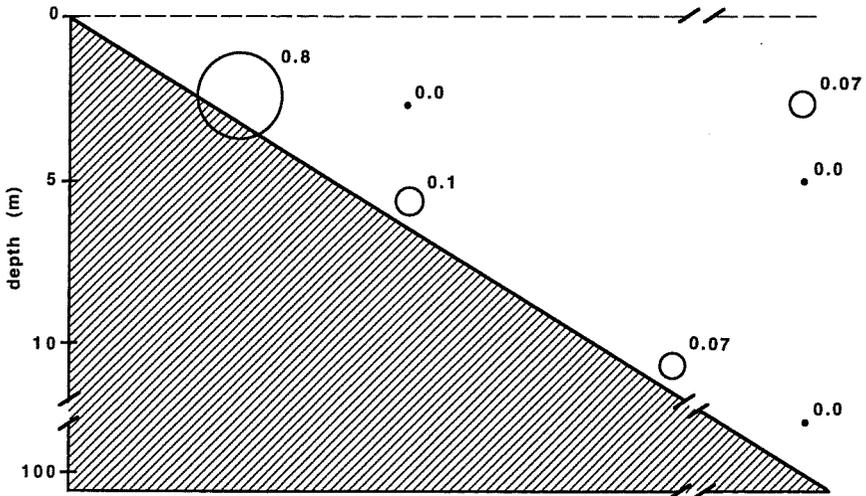


Figure 3. Catch densities at nearshore stations with respect to depth for preflexion larvae of *Clevelandia ios*. Circle areas indicate catch densities, with average number of larvae per cubic meter indicated adjacent to each circle.

dorsal midline) compared to the uniform diffuse melanophore distribution on juveniles and adults (Fig. 1 f). The first dorsal fin developed during this phase of transitional pigmentation. Pelvic fin buds were unfused in 72% of these schooling larvae. The remainder had fin buds contiguous but only partially fused (17%), or completely fused with a transverse membrane (11%). Although 11% of schooling larvae had fused pelvic fins, none of these fish had the adult pigment pattern; 9% showed the transitional pigment pattern characteristic of settled, non-burrowing fish.

Some settled gobies in tide pools (10.0–16.6 mm SL; avg. 13.1 ± 1.3 mm SL, $n=48$) would not enter burrows, even when pursued (classed as “non burrow-entering”). They had transitional (25%) or adult (75%) pigmentation, with melanophores on the entire dorsal surface and on sides (Fig. 1 f), and 98% had complete pelvic development (2% partially fused).

Burrowing *C. ios* collected during July–September ranged from 11.5 to 37.6 mm SL (avg. 18.7 ± 5.0 mm SL, $n=241$), with size modes at 17, 25 and 36 mm SL, which may correspond to year-classes (0-, 1- and 2-yr age). The adult pigment pattern was present on 97% of burrowing gobies (3% transitional pigment) and all had complete pelvic fins.

Settlement preference tests revealed a significant preference for sand or plastic over clay (Table 1), the settled *C. ios* in the laboratory averaging 13.1 ± 1.1 mm SL ($n=20$). In contrast, juveniles given a choice of sand with artificial burrows or plain sand showed no significant preference (8 counts, $\chi^2_{STAT}=0.012$, $\chi^2_{.05}=3.84$, 1 d.f., n.s.), as was the case for sand versus sand with ghost shrimps (4 counts, $\chi^2_{STAT}=0.595$, $\chi^2_{.05}=3.84$, 1 d.f., n.s.).

Clevelandia ios showed similar behaviors in burrow entry when observed in the laboratory and in the field. Movements across the substrate were in the form of short, zig-zagging, jumping motions with the goby resting only on its pelvic fin and lower edge of the caudal fin. If undisturbed, the goby would tilt its head down near the edge of a burrow opening and then enter the burrow head-first, immediately turning around to face out.

Mean latency time for burrow entry in 30 “naive,” 31 “experienced” and 25 “wild” fish were 54, 45 and 36 seconds, respectively. Two wild gobies entered a

Table 1. Chi-square analysis of numbers of *Clevelandia ios* larvae settled on sand, clay and plastic substrates ($\chi^2_{2d.f.} = 16.6$, $S_{.001}$).

	Sand	Clay	Plastic
	6	5	9
	6	5	5
	9	3	11
	11	4	12
	4	4	7
	9	1	15
Observed	45	22	59
Expected	42	42	42

burrow in less than a second of their release, and wild gobies never left an entered burrow during the five minute observation periods. In contrast, many naive and lab-experienced fish exited burrows during observation. Latency time frequency distributions were skewed to the right, so a one factor ANOVA on \log_{10} transformed data revealed significant difference in mean latency times ($F=3.82$, $s_{.05}$). Further analysis of these transformed latency times with the Scheffe F-test showed the significant difference to be between wild and naive groups ($F=3.74$, $s_{.05}$). Latency time was lower for experienced than for naive fish, but not significantly.

Discussion

Larvae drifting offshore appear to rise toward the surface layer where they may be transported back to shallow water. Previous sampling (unpublished data) indicated that larval *Clevelandia ios* were not distributed in significant numbers beyond the break-in-slope of the estuarine delta bottom. They were not taken in nearshore tows along rocky shorelines in the same region. Thus, early larval drift dispersal tends not to remove this species from the region of soft-bottom estuarine waters where larvae will associate with substrate during later schooling stages and where settlement to the juvenile stage will occur.

Melanophore distribution on the larval stages of *C. ios* remained unchanged during larval development (Fig. 1). The diagnostic character of a single dorsal midline melanophore, listed by Matarese et al. (1989) for separating *C. ios* from *Coryphopterus nicholsi* (posterior dorsal midline melanophore row) and *Lepidogobius lepidus* (three spaced dorsal midline melanophores) was of diagnostic utility throughout the larval period.

In *C. ios*, the transition from demersal schooling larva to benthic juvenile accompanies morphological and behavioral changes. One change is the use of shrimp burrows as refuge. The initiation of this behavior may not occur immediately upon settling; the latency time differences found between naive, experienced, and wild fish suggest that learning may be involved in burrow-entry behavior. In field collections, a small proportion of newly settled fish would not enter burrows, but no developmental character absolutely distinguished burrow-entering from non-entering gobies.

The overlap in the size distributions of non-entering and burrow-entering gobies indicates that size is not the only factor involved, since gobies of 11–13 mm SL were found schooling, settled but not burrow-entering, as well as burrow-entering. Minimum sizes of various species of gobies found in burrows by other authors ranged from 8.7 to 10.00 mm SL, compared with a minimum 11.5 mm SL in this study (Yanagisawa, 1982, 1984, Karplus, 1987, Macdonald, 1972). The state of being settled but not entering burrows is probably a temporary condition, since attempts to sample gobies not entering burrows at the end of September were unsuccessful. Similarly, Yanagisawa (1982) found a portion of the gobies *Amblyeleotris japonica* did not occupy burrows as permanent residents, although he considered these "floaters" to represent a temporary condition.

Pelvic fin development and pigment pattern overlapped between non-burrow entering and burrow-entering gobies. In the schooling stage, pelvic fin development varied more than pigment pattern. The retention of a relatively transparent body during schooling, and the early completion of the fused pelvic fin prior to settlement, may be advantageous to the arrow goby. The transitional pigment pattern corresponded most to the settled, non-burrow entering behavior, whereas pelvic fusion was complete in nearly all settled fish. Macdonald (1972) described atrophy of the larval swimbladder at the time of pelvic fin fusion and pigment change, but dissection revealed a well developed swimbladder in all life history stages in the present study. Brothers (1975) mentioned presence of a relatively large swimbladder in adult *Clevelandia ios*.

Settling marine fish larvae often prefer substrates closely related to those of their adult habitat (Marliave 1977). The present study reveals that *C. ios* prefers to settle on sand rather than clay. Prasad (1948) states that adults are most abundant on clayed sand, so avoidance of clay may be specific to settlement. *Callianassa californiensis* prefers sand and *Upogebia pugettensis* prefers clay (MacGinitie 1934, 1935). No settlement preference was found for substrate with artificial burrows or substrates with burrowed *C. californiensis*, so that neither the burrows nor the ghost shrimp served as cues to stimulate settlement of gobies, compared to plain sand.

A significant difference in the mean latency times between "naive" and "wild" gobies suggests that experience has some influence on entry into burrows. Experience was shown to be a significant factor in the defense of burrows by a sympatric goby, *Lepidogobius lepidus* (Grossman 1980). The average length, however, of the "wild" gobies exceeded that of the "naive" and "experienced" fish, which may have affected burrow entry. Although experience influences burrow entry, it is not necessary for initiation of that behavior. Entry into burrows thus appears to have an innate component as well as a possible component of experience.

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