Environmental Biology of Fishes **37**: 237–244, 1993.

The effect of light and darkness on hatching in the pomacentrid *Abudefduf saxatilis*

Florence A. McAlary & William N. McFarland
Wrigley Marine Science Center, Hancock Institute of Marine Studies, University of Southern California, P.O. Box 398, Avalon, CA 90704, U.S.A.

Received 4.2.1991 Accepted 4.2.1993

Key words: Fish hatching, Twilight effect, Demersal eggs, Antipredator adaptation, Hatching mechanism

Synopsis

Reports that pomacentrid embryos hatch after dusk are confirmed by photic manipulation of sergeant major eggs. Embryos placed in the dark for 20 minutes or longer prior to their normal hatching after sunset hatched, whereas controls held in light did not hatch. Percent of hatched embryos correlated with increasing exposure to darkness up to one hour after which no further improvement in hatching was observed. Embryos maintained in continuous light during their normal twilight hatching period did not hatch. Also, embryos exposed to 60 minutes of darkness, if interrupted by one minute of light every 10 minutes did not hatch. The percent hatch in dark treatments varied significantly between nests and, in some treatments, correlated negatively with the size of the egg clumps (number of eggs per clump) tested. To initiate hatching in the presence of light required intensities of 0.03 lux or less. These low intensities are not reached until about 20 minutes after sunset on the reef where the embryos occur. We conclude that hatching for some embryos occurs about 30 minutes after sunset but for most is not completed until at least one hour after sunset. Hatching therefore takes place at a time long after potential diurnal fish predators have refuged in the reef structure.

Introduction

Although extremely diverse, most coral reef fishes broadcast eggs and produce pelagic embryos and larvae (Johannes 1978, Barlow 1981, McFarland & Ogden 1985). In many species, however, eggs are demersal (Thresher 1984), a reproductive mode characteristic of the Pomacentridae. Male sergeant majors (*Abudefduf saxatilis* and *A. troschelli*) clear a surface on coral overhangs, actively court females to deposit eggs in nest sites, and vigorously defend the fertilized eggs against intruders (Shaw 1955, Foster 1987). On the fifth or sixth day of development the embryos hatch 30 to 70 minutes after sunset and disperse into the plankton (Foster 1987 and our observations). Similar reproductive behaviors have been demonstrated for a wide variety of pomacentrids (see Thresher 1984) and, significantly, in all pomacentrids where hatching has been described it occurs during the early evening (see Robertson et al. 1990). The timing of hatching during dusk is considered to remove the embryos from the threat of diurnal reef predators (Johannes 1978). Darkness has been shown to initiate and light to inhibit hatching in the temperate damselfish, *Stegastes clarki* (Kohda 1988). Reduced illumination at dusk, therefore, may act as a proximate cause of hatching among pomacentrids.

In this paper we report the effects of manipula-

tion of light and of darkness on hatching in the sergeant major, *Abudefduf saxatilis*.

Methods

During June and July 1989, 36 nests of *Abudefduf saxatilis* located on the Tague Bay Barrier forereef, St. Croix, U.S. Virgin Islands, were tagged with flagging. All nests occurred near the reef base at depths between 7–10 meters where temperatures varied between 27 and 28° C. Daily examination of the flagged nests provided a chronology of natural hatching. Eggs were removed from the nests with a dive knive by prying away a section of the underlying coral surface to which the eggs were attached. Using forceps, to minimize direct contact with eggs, the clumps were placed into small plastic bags expanded with sea water. The bags were placed in a plexiglass container at the collection sites to avoid crushing, brought to the surface, and transported to the laboratory in an open ice cooler.

Experiments were conducted before dusk (between 1400 and 1800 hours), except for tests of embryos held in light until 2100 hours or overnight in light or darkness. For photic manipulations nest samples were broken into small clumps (0.05–0.5 cm^{-2}) of less than 100 embryos and submerged in small bowls (3 cm diameter and 2 cm deep) of seawater 23–24° C. The number of eggs in each clump was counted using a dissecting microscope. To determine the effect of darkness, the bowls containing egg samples were placed in light-tight canisters (1.5 liter volume) for various time periods from 10 to 180 minutes, or overnight. One or more bowls with egg clumps from each sampled nest were held in continuous room light as controls. Hatchlings or unhatched embryos remaining at the end of each treatment period were counted immediately to determine the percent hatch. Room illumination from daylight fluorescent fixtures varied less than 10% from 260 lux. The chromatic sensitivity curve of *Abudefduf saxatilis* embryos is unknown. Many adult coral reef fish, however, have photopic sensitivity in the blue and green wavebands (Levine & MacNicol 1979, McFarland 1991). Because the embryos and larvae possess only cones, the use of fluorescent illumination, which is rich in the blue-green to yellow wavebands, was considered appropriate in the absence of specific information about the embryo's chromatic sensitivity. Light intensity was measured with a Luna Pro photometer, which has a peak sensitivity in the green-yellow wavebands and closely matches the chromatic peak of the fluorescent lamps.

To test the effect of intermittent light on hatching, we exposed eggs from the same nest to 60 minutes of darkness interrupted by one minute of light at intervals of 10, 20 or 30 minutes. During the one-minute period when the cans were open the unhatched embryos or free embryos were counted.

To evaluate the effect of varied light intensities on hatching, filters with different transmission values from 1.0–0.0001% were mounted over holes cut in the lids of the light tight metal cans, and exposed to an incident fluorescent flux of 300 lux for 60 minutes. Hatching responses of embryos tested at different light intensities were compared to embryos taken from the same nest that were exposed to continuous fluorescent light of 300 lux or to darkness for the same treatment period. The number of density filters available restricted experiments to two simultaneous exposures each afternoon; therefore, tests had to be executed over several days using eggs from different nests.

To determine the light responses of newly hatched embryos, 50 to 100 young were placed in a 13 cm diameter glass dish and exposed to laterally directed light from a tungsten bulb or placed in darkness under a light-tight canister. After five minutes exposure to the light source or darkness, the distribution and swimming behavior of the young were noted.

Results

The effect of light and darkness on hatching

Experiments in which embryos were exposed to constant light or held in darkness on the afternoon of natural hatching demonstrate significant positive correlations between hatching and increasing exposure to darkness (r1, Table 1). The regression slopes

of percent hatch calculated per nest, per egg clump and for total eggs per treatment were similar (mean slope = 1.86, 95% C.I. test). Because the percent hatch for additional embryos from these nests exposed to darkness from 70 to 180 minutes did not differ significantly from the 60 minute dark treatment (Rank sum test, $p > 0.95$), they are not considered further in our analysis. In three of the six dark treatments, the percent hatch decreased significantly as the number of eggs per clump increased (r2, Table 1). The distributions of egg clump sizes for different treatments, however, were similar (Kruskal-Wallis test corrected for ties, H = 7; d.f. = 5; $p > 0.9$). Embryos held in darkness for less than 20 minutes did not hatch. In all dark treatments, hatching declined abruptly and ceased within 20 to 60 minutes when embryos were returned to the light (e.g. Fig. 1). Control embryos held in continuous light did not hatch until exposed to darkness (40 minutes) at the conclusion of the experiments. The percent hatch for controls, respective to their source nests, was within the range of values for similar dark treatments. At the conclusion of each experiment, the hatchability of embryos from 10 and 20 minute dark treatments was tested by placing the embryos in darkness for 40 minutes or by leaving them overnight in natural light. In all instances, the percent hatch was within the range of 40–60 minute dark treatments. Embryos in eggs collected one day before the day of natural hatching did not hatch when tested in 40 minutes of darkness.

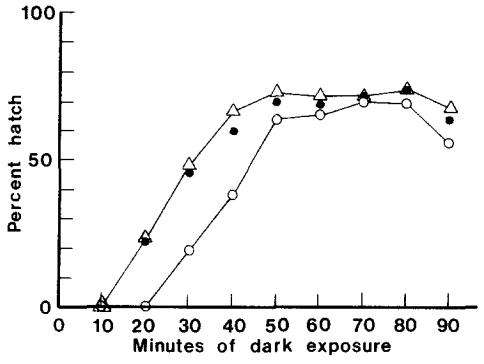

Fig. 1. Cessation of hatching for embryos from a nest of *A. saxatilis* when exposed to light following dark treatments of increasing duration. Open circles are the hatch immediately after each dark period, closed circles the hatch after 10 minutes in room light (260 lux), and open triangles the hatch after two hours in room light. Note that hatching continues in the light for a short period after a dark period and essentially ceases under continued illumination.

For four of the seven nests tested the percent hatch was available for all dark treatments. Comparison of their mean percent hatch revealed significant differences between nests as well as across dark treatments (2-factor ANOVA, $p < 0.01$ and p

Table 1. The mean percent hatch for embryos of *Abudefduf saxatilis* from different periods of dark exposure and the median of eggs per clump per dark treatment. Percentages are mean ±1 standard deviation, r_1 the regression correlation coefficient per nest, per clump, and for total eggs, r_2 the coefficient for egg clumps within each dark treatment. Symbols are * = $p \leq 0.05$, ** = $p \leq 0.01$, *** = $p \leq 0.001$.

Percent hatch	Minutes of dark exposure						r_1
	10	20	30	40	50	60	
Per nest	0	2.7± 6.5	19± 17.8	36± 8.5	61± 8.6	76± 21.4	0.90***
Range	0	(0–16)	(0–44)	(26–46)	(48–74)	(52–100)	
n	6	6	7	6	5	6	
Per clump	0	4.9± 13.5	34± 31	42± 17	69± 22	84± 21	0.82***
Range	0	(0–47)	(0–100)	(20–91)	(38–100)	(36–100)	
n	13	19	20	18	14	20	
r_2	0	−0.21	−0.48*	−0.36	−0.76**	−0.76**	
Total eggs	0	4	25	40	60	75	0.99***
Median eggs per clump	22	20	24.5	31	21	20	
Range	(7–58)	(6–47)	(6–76)	(9–53)	(11–52)	(7–50)	

<0.0001, respectively; Hartley's F_{max} test $p > 0.05$, Sokal & Rohlf 1981).

Eggs held in continuous light overnight (through the period of natural hatching during dusk) did not produce free embryos (n= 164 eggs). Most of the embryos in these eggs were dead by morning. In contrast, embryos from eggs in the same nests hatched when removed from the light as late as 2100h after 40 minutes of darkness (51 embryos from 115 eggs, 44%), when held in darkness overnight (114 embryos from 193 eggs, 59%), or when exposed to natural twilight (41 embryos from 62 eggs, 66%).

The effect of light interruptions on hatching

Interrupting a 60-minute dark period by exposing embryos to one-minute of light every 10 or 15 minutes inhibited hatching throughout the dark exposure (n= 80 and 79 embryos, respectively), whereas controls placed in continuous darkness for 60 minutes hatched (81 of 81 embryos). The percent hatch of embryos exposed to one-minute light interruptions at either 20 or 30 minute intervals during 60 minutes of darkness was not significantly different than the percent hatch of embryos exposed to 60 minutes of continuous darkness (83 of 97, respectively, versus 100%, proportions test). Although we did not pursue this experimental approach further (e.g. by examining light dosage effects), the results reinforce observations that eggs must be secluded from light for some minimum period of time before hatching is initiated.

The effect of varied intensities of light on hatching

Because *Abudefduf saxatilis* embryos are reported to hatch within the first 30 to 70 minutes after sunset (Foster 1987), the hatching process apparently is initiated during dusk as light intensity rapidly decreases. To evaluate intensity effects, 60 minutes was used as a test period, because that exposure to darkness produced a high percent hatch in the previous experiments. To adjust for differences in the hatching response of embryos from different nests, the percent hatch for each experimental clump was normalized against its respective control (i.e. the 60-minute dark treatment). Results indicate that hatching occurred when eggs were exposed to light intensities of 0.03 lux or less (≤1/1000 the intensity of laboratory light, Fig. 2). Thus, total darkness was not essential for hatching and light was inhibitory only above some specific intensity. As in the previous experiments, percent hatch tended to correlate negatively with the number of eggs per clump (this correlation was significant for 0.0003 lux and total darkness where $r = -0.5$, $p < 0.05$ and $r = -0.78$, $p < 0.001$, respectively); however, the distributions of egg clump sizes in different light intensity treatments were similar (Kruskal-Wallis test corrected for ties, $H = 0.21$; d.f. = 4; $p > 0.99$).

Hatching in field nests

To determine if natural hatching had occurred on a given night, we examined the nests from which the egg samples were collected on the following day. In all cases, when a 60-minute dark exposure did not promote hatching in the laboratory, eggs were still present in the sampled nests on the following morning. When hatching was induced by darkness in the laboratory, hatching was evident in the field nests and, usually, was complete. In a few instances hatching of all the embryos from a nest occurred over two consecutive nights.

Response of newly hatched embryos to light and darkness

Under room illumination, newly hatched embryos swam actively about the volume of their containers. When exposed to directional light, embryos showed a strong positive phototaxis. Shifting the light source 90 or 180 degrees resulted in re-orientation of young toward the light source. After five minutes in darkness, embryos were dispersed throughout the volume of their container. In contrast to their active behavior in the light, dark adapted embryos were relatively inactive, more or less drifting in the

container. Embryos resumed active swimming within 30 seconds after return to room illumination.

Discussion

Few examples of environmental triggers for hatching in fishes exist (see Yamagami 1988 for review). Stranding of eggs and lowered oxygen levels have been implicated in species of *Fundulus* (Harrington 1959), and tidal efects in the stranded eggs of the grunion, *Leuresthes tenuis* (Walker 1952). Temperature is known to influence hatching in some species and the hatching patterns of the medaka, *Oryzias latipes,* are affected by day/night rhythms (Yamagami 1988). In the fourteen pomacentrid species reported in the literature, hatching occurs during dusk (Albrecht 1969, Emery 1973, Fricke 1973, MacDonald 1973, Moyer 1975, Moyer & Bell 1976, Ross 1978, Doherty 1983, Ochi 1985, 1986, Foster 1987, Kohda 1988). Embryos of the temperate damselfish *Hypsypops rubicundus* hatch after sunset (P. Sikkel personal communication). Embryos of *Stegastes clarki* (Kohda 1988) and *Dascyllus albisella* (J. Nunez personal communication) hatch in darkness in the laboratory. Hatching occurs during dusk in several blennies (Robertson et al. 1990). Our experiments with eggs of *Abudefduf saxatilis* extend these observations by demonstrating that (a) to induce hatching, embryos must be secluded from light for some minimum period of time (>15 min of uninterrupted darkness), (b) repeated exposure of embryos held in darkness to one-minute pulses of room light at intervals less than the critical minimum (<20 minutes) is sufficient to inhibit hatching, (c) most embryos hatch within one hour of exposure to favorable conditions, and (d) hatching can be initiated in dim light (≤0.03 lux).

At St. Croix (17 degrees North latitude) at 10 meters depth, where most of the sergeant major nests were located, downwelling light declines to 0.03 lux by 40 minutes, and to 0.003 lux at about 50 minutes after sunset. *Abudefduf* eggs, however, are usually deposited on the underside or downward slanted sides of coral rock. As a result intensities of 0.03 to 0.003 lux for horizontally directed light, to which the eggs are more likely exposed, are reached from 20 to 30 minutes post sunset (McFarland 1986). Actual light intensities at nest sites may be slightly higher during dusk because light during twilight is richer in the blue wavebands than in the green-yellow wavebands to which a lux meter is most sensitive (Fig. 4 in McFarland 1991). Therefore, we conclude that hatching is probably complete one hour or slightly longer after sunset. This chronology coincides with field descriptions of hatching in *A. saxatilis* (Foster 1987). The hatching process, therefore, allows embryos to depart the nest during darkness long after diurnal predators such as wrasses, hamlets and planktivorous pomacentrids are visually impaired and have taken refuge, and when nocturnal predators are emerging from the reef (Hobson 1972, Collette & Taylor 1972, Domm & Domm 1973). The transparency of *Abudefduf* embryos at hatching and, especially, their small size (<5 mm), must pro-

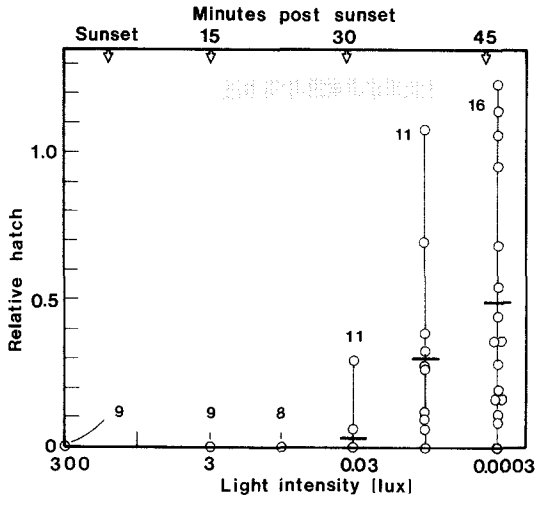

Fig. 2. The effect of lowered levels of room illumination on hatching of *A. saxatilis* eggs. Open circles are the relative hatch for individual clumps of eggs from three nests following exposure to different light intensities for 60 minutes. The relative hatches of test clumps were obtained by normalization against the mean percentage hatch per clump for each nest's dark control. Horizontal lines give the mean relative hatches. Values within the graph give the number of egg clumps tested at each intensity. Coincident data for zero relative hatch are not shown. Thus, at 0.03 lux, embryos hatched in two of the eleven egg clumps tested. The upper axis indicates the approximate time of sunset and the minutes after sunset at which the experimental light intensities would be reached at nest sites. The stippled area represents the light intensities over which most fishes undergo retinal dark adaptation and over which diurnal fishes lose visually guided behaviors (Blaxter 1988).

vide some protection from small nocturnal predators, such as cardinalfishes. In addition, the light responses of newly hatched embryos likely benefits dispersal. Relative inactivity in darkness should result in little motion disturbance and minimize the detection of embryos by predators. We could only observe the behavior of young at the conclusion of dark exposure; therefore, we cannot totally discount their inactivity as a response to the sudden onset of light. However, embryos of *Blennius pholis* when observed in dim red light were inactive (Qasim 1955). Quiescent behavior could explain, at least in part, the apparent lack of vertical stratification of *Abudefduf* embryos at night (Leis 1991a). The positive phototaxis that we observed is consistent with the peak concentrations of the young of *Abudefduf* sp. that Leis collected in near surface water during the day. Indeed, responses to light may have a central role in diel distributions of many species of tropical fish embryos and larvae (Leis 1991b), as demonstrated for many planktonic invertebrates (Kerfoot 1987, Forward 1991).

On two occasions we removed clumps of eggs from the nests of *Microspathodon chrysurus*. Unlike the embryos of *A. saxatilis*, which were virtually unaffected by collecting and transport disturbances, many embryos of *Microspathodon* hatched during transit. On a third attempt, we took precautions to minimize mechanical disturbances. These nest samples were carefully cut into small clumps and placed in a dark container or in continuous light. Thirty out of 100 embryos had hatched after 44 minutes in darkness. Hatching continued in the light in this sample to 44 percent after 16 minutes and stabilized at 46 percent a few minutes later. In continuous light, a small percentage of the embryos (11 out of 145, 8%) hatched immediately after being placed in the test containers, but ceased for the remainder of the experiment. We attribute this small initial hatch to disturbance. These data support the view that light inhibits hatching among fish with demersal eggs and indicate that, at least in some species, mechanical disturbance of the eggs may trigger hatching.

Although we found a consistent pattern of hatching in response to the use of darkness or light, there was significant variation in hatching between nests, yielding a higher hatch for some nests than others, and, in some treatments, a significant reduction in hatching as the number of eggs in a test clump increased (Table 1). Reduced hatching in some egg clumps probably resulted from the conditions of the experiment. Without continuous water circulation, oxygen may have depleted locally or carbon dioxide accumulated to a greater extent in the large as compared to the smaller egg clumps and, perhaps, have limited hatching. Embryos maintained for over three hours in room light, which did not hatch and presumably would accumulate a similar oxygen depletion and/or carbon dioxide accumulation, when placed in darkness for 30 or more minutes did hatch in all sizes of egg clumps. The canisters that we used for dark treatments were sufficiently large that volumes of oxygen in the dark environment were not limiting compared to bowls left in room light. Temperature effects due to transporting the eggs on ice or the slightly cooler temperatures on the laboratory seawater compared to the field may also have been a factor in hatching variability. Despite our care in handling nest samples, some of the eggs may have been damaged during manipulation.

Variation in percent hatch among egg clumps may, in part, be attributed to differences in the maturity of embryos at the time of testing. Females of *A. saxatilis* spawn in nests throughout the day and presumably also at night (Foster 1987, Robertson et al. 1990). As a result, in different nests and in a single nest the initiation of embryonic development might vary as much as 12 hours or more for eggs deposited and fertilized in the late afternoon as compared to eggs laid and fertilized in early morning. On the date of hatching, embryos we tested from different nests or different areas of the same nest sometimes had variable amounts of yolk on the afternoon of expected hatching. Although most eggs from nests with embryos that hatched had no visible yolk to about one-eighth that of an egg less than one day old, some eggs had considerably more. If embryos must achieve a level of developmental competence before hatching can be induced by reductions in light intensity, a batch of embryos that have not developed sufficiently by dusk may in fact not hatch until the next evening or, perhaps, hatch several hours or more after dusk (see Foster 1987).

How might the environmental control of embryo release relate to the hatching mechanism? Hatching in fishes is associated with two factors: the release of a hatching enzyme from embryonic glands which rapidly digests the inner hardened layers of the egg envelope; and sufficient movement by the embryo to break out of the weakened egg envelope (Armstrong 1936). The photic mechanism that controls hatching could be mediated by the eyes, and/or the pineal complex or, although we consider it less likely, by a direct inhibitory effect of light on the hatching glands. Intermediate processing that might inhibit release of the hatching enzyme involves the nervous system and the pituitary, but this is unclear for most fishes (Yamagami 1988). Because most fish embryos possess pure cone retinae (Blaxter 1988), the eyes of *Abudefduf* embryos presumably function only at photopic levels of illumination. A parsimonious hypothesis to explain hatching in darkness implies that light intensities near or below the embryos absolute light threshold lead to activation of hatching gland secretion and synchronize emergence behavior with weakening of the egg envelope. Repeated short pulses of light within a minimum dark period of 15 minutes or less inhibited hatching (i.e. dark exposures within this critical time period are not cumulative), suggest that the inhibiting effect of light on hatching has a long time constant. Although neural processes are likely involved in hatching, a long time constant implies that a humoral factor is important in the initial mechanisms that lead to release of the hatching enzyme. In this sense, pomacentrids offer a rare opportunity to unravel the physiological and molecular mechanisms that operate between a demonstrated environmental trigger and the hatching process itself.

Acknowledgements

We wish to thank Margaret McFall-Ngai for reviewing the manuscript and Christopher Petersen for guiding us to pertinent literature. This study was partly funded by Cornell University and is a contribution of the West Indies Laboratory.

References cited

Albrecht, H. 1969. Behavior of four species of Atlantic damselfishes from Columbia, South America (*Abudefduf saxatilis, A. taurus, Chromis multilineata, C. cyanea;* Pisces: Pomacentridae). Z. Tierpsychol. 26: 662–676.

Armstrong, P.B. 1936. Mechanism of hatching in *Fundulus heteroclitus*. Biol. Bull. 71: 407.

Barlow, G. 1981. Patterns of parental investment, dispersal and size among coral-reef fishes. Env. Biol. Fish. 6: 65–85.

Blaxter, J.H.S. 1988. Sensory performance, behavior, and ecology of fish. pp. 203–232. *In:* J. Atema, R.R. Fay, A.N. Popper & W.N. Tavolga (ed.) Sensory Biology of Aquatic Animals, Springer-Verlag, New York.

Collette, B.B. & F.H. Taylor. 1972. Activity patterns of coral reef fishes with emphasis on nocturnal-diurnal changeover. pp. 98–124. *In:* B.B. Collette & S. Earle (ed.) Results of the Tektite Program: Ecology of Coral Reef Fishes, Nat. Hist. Mus. Los Angeles, Sci. Bull. 14.

Doherty, P.J. 1983. Diel, lunar and seasonal rhythms in the reproduction of two tropical damselfishes: *Pomacentrus flavicauda* and *P. wardi*. Mar. Biol. 75: 215–224.

Domm, S.B. & A.J. Domm. 1973. The sequence of appearance at dawn and disappearance at dusk of some coral reef fishes. Pacific Sci. 27: 128–135.

Emery, A.R. 1973. Comparative ecology and functional osteology of fourteen species of damselfish (Pisces: Pomacentridae) at Alligator Reef, Florida Keys. Bull. Mar. Sci. 23: 649–770.

Forward, R.B. 1991. Behavioral responses of larvae of the crab *Rhithropanopeus harrisii* (Brachyura: Xanthidae) during diel vertical migration. Mar. Biol. 90: 9–18.

Foster, S.S. 1987. Diel and lunar patterns of reproduction in the Caribbean and Pacific sergeant major damselfishes *Abudefduf saxatilis* and *A. troschelii*. Mar. Biol. 95: 333–343.

Fricke, H.W. 1973. Okologie und Sozialverhalten des Korallenbarsches *Dasyllus trimaculatus* (Pisces, Pomacentridae). Z. Tierpsychol. 32: 225–256.

Harrington, R.W., Jr. 1959. Delayed hatching in stranded eggs of marsh killifish, *Fundulus confluentus*. Ecology 40: 430–437.

Hobson, E.S. 1972. Activity of Hawaiian reef fishes during the evening and morning transitions between daylight and darkness. U.S. Fish. Bull. 70: 715–740.

Johannes, R.E. 1978. Reproductive strategies of coastal marine fishes in the tropics. Env. Biol. Fish. 3: 65–84.

Kerfoot, W.C. 1987. Adaptive value of vertical migration: comments on the predation hypothesis and some alternatives. pp. 91–113. *In:* W.C. Kerfoot & A. Sih (ed.) Predation: Direct and Indirect Impacts on Aquatic Communities, Univ. Press of New Hampshire, Hanover.

Kohda, M. 1988. Diurnal periodicity of spawning activity of permanently territorial damselfishes (Teleostei: Pomacentridae). Env. Biol. Fish. 21: 91–100.

Leis, J.M. 1991a. Vertical distribution of fish larvae in the Great Barrier Reef lagoon, Australia. Mar. Biol. 109: 157–166.

Leis, J.M. 1991b. The pelagic stage of reef fishes: the larval biol-

ogy of coral reef fishes. pp.183–230. *In:* P.F. Sale (ed.) The Ecology of Fishes on Coral Reefs, Academic Press, San Diego.

Levine, J.S. & E.F. MacNichol. 1979. Visual pigments in teleost fishes: effects of habitat, microhabitat, and behavior on visual system evolution. Sens. Processes 3: 95–131.

MacDonald, C.D. 1973. Reproductive strategies and social organization in damselfishes. Ph.D. Dissertation, University of Hawaii, Honolulu. 226pp.

McFarland, W.N. 1986. Light in the sea – Correlations with behaviors of fishes and invertebrates. Amer. Zool. 26: 389–401.

McFarland, W.N. 1991. The visual world of coral reef fishes. pp.16–32. *In:* P.F. Sale (ed.) The Ecology of Fishes on Coral Reefs, Academic Press, San Diego.

McFarland, W.N. & J. Ogden. 1985. Recruitment of young coral reef fishes from the plankton. pp.37–51. *In:* M.L. Reaka (ed.) The Ecology of Coral Reefs, Symp. Ser. Undersea Res., Vol. 3 (1), NOAA Undersea Res. Progr., Rockville.

Moyer, J.T. 1975. Reproductive behavior of the damselfish, *Pomacentrus nagasakiensis* at Miyakejima, Japan. Jap. J. Ichthyol. 22: 151–163.

Moyer, J.T. & L.J. Bell. 1976. Reproductive behavior of the anemone fish *Amphiprion clarkii* at Miyake-jima. Jap. J. Ichthyol. 23: 23–32.

Ochi, H. 1985. Temporal patterns of breeding and larval settlement in a temperate population of the tropical anemonefish, *Amphiprion clarkii.* Jap. J. Ichthyol. 32: 248–257.

Ochi, H. 1986. Breeding synchrony and spawning intervals in the temperate damselfish *Chromis notata.* Env. Biol. Fish. 17: 117–123.

Qasim, S.Z. 1955. Rearing experiments on marine teleost larvae and evidence of their need for sleep. Nature 4448: 216–217.

Robertson, D.R., C.W. Petersen & J.D. Brawn. 1990. Lunar reproductive cycles of benthic-brooding reef fishes: reflections of larval-biology or adult-biology? Ecol. Monogr. 60: 311–329.

Ross, R.M. 1978. Reproductive behavior of the anemonefish *Amphiprion melanopus* on Guam. Copeia 1978: 103–107.

Shaw, E.S. 1955. The embryology of the sergeant major, *Abudefduf saxatilis.* Copeia 2: 85–89.

Sokal, R.R. & F.J. Rohlf. 1981. Biometry. 2nd Edition. W.H. Freeman & Co., San Francisco. 859pp.

Thresher, R.E. 1984. Reproduction in fishes. T.F.H. Publications, Neptune City. 399pp.

Walker, B.W. 1952. A guide to the grunion. Calif. Fish Game 38: 409–420.

Yamagami, K. 1988. Mechanisms of hatching in fish. pp.447–499. *In:* W.S. Hoar & D.J. Randall (ed.) Fish Physiology, Vol. 11, The Physiology of Developing Fish, Part A: Eggs and Larvae, Academic Press, San Diego.