INDUCED MATURATION AND SPAWNING OF MARINE FISH AT THE SOUTHWEST FISHERIES CENTER LA JOLLA, CALIFORNIA

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ABSTRACT

Technical details are given for spawning the following fish under controlled laboratory conditions: the northern anchovy (Engraulis mordax), Pacific mackerel (Scomber japonicus), croaker (Bairdiella icistia), and striped bass (Morone saxatilis).

INTRODUCTION

Recent successes in the sexual maturation and spawning of marine fish at the Southwest Fisheries Center, La Jolla, have been reported in the literature (Haydock, 1971; Leong, 1971). Continual refinement of maturation and spawning techniques has followed these published accounts. This paper provides an up-to-date progress report on these methods with emphasis on the northern anchovy, (Engraulis mordax). Preliminary details are given for other fish studied: Pacific mackerel (Scomber japonicus), croaker (Bairdiella icistia), and striped bass (Morone saxatilis). All of these studies have been performed to provide fish larvae for laboratory experiments (Lasker et al., 1970).

NORTHERN ANCHOVY

The spawning of anchovies in aquaria was achieved with a fixed photoperiod, 4 hours light and 20 hours darkness, and injection of hormones (Leong, 1971). Gradually, empirical changes in techniques

have permitted the establishment of anchovies which are perennially sexually mature and which spawn spontaneously every day of the year. The normal spawning cycle of the northern anchovy in the Pacific Ocean is protracted with some eggs off California found from April through December, but peak spawning occurs during January, February, and March.

Basic Laboratory Techniques

Anchovies of standard length greater than 110 mm are kept in plastic swimming pools, 4.6 m in diameter, 0.72 m deep. The sea water used in these experiments is sand-filtered and irradiated with ultraviolet light (for details see Lasker and Vlymen, 1969). Each pool receives 38 liters/min. The normal pass-through flow of sea water is augmented with a centrifugal pump outside the tank which recirculates the water at 290 liters/min and provides a jet current. Anchovies normally swim against this current. Light from a 200 watt incandescent bulb provides 32 foot-candles at the water surface for 4 hours (8 a.m.-12 noon) and two, three watt bulbs give 1-2 foot-candles at the surface for the "dark" period. This slight illumination is needed during the "dark" period to prevent the anchovies from colliding with the walls of the tank. Temperature is maintained throughout the year at 15 C with the maximum allowed variation from 14.5-15.5 C. Usually about 1,000 anchovies are kept in a single tank and mature sexually from the spent condition in 2-3 months. Oregon moist chow 1 (0.6 kg/day per 1,000 anchovies) is provided to the fish with an automatic feeder after a hand feeding of 0.34 kg of frozen brine shrimp. Minced squid (0.9 kg/day per 1,000 anchovies) can be substituted for the brine shrimp. The tank is cleaned by vacuuming once a week.

Under these conditions all of the <u>E. mordax</u> in our tanks achieve an average gonad index (GI = (gonad wet weight/total animal wet weight) x 100) of 5; yolked eggs are present in the females and there is active sperm in the males. We collect approximately 3,000 eggs per 34,000 liters of water filtered in 15 hours but we have not determined what full production is, because the anchovies actively filter out and ingest many of the eggs, which are also lost in the overflow when filtering is discontinued. Rarely are all eggs fertilized but we have never found all eggs to be unfertilized; occasionally 100% fertilization is achieved naturally. Hatching success varies from less than 1% to over 80% (average 20%). Spawning, in

Obtained from R. V. Moore, La Conner, Washington 98257. Reference to commercial products does not imply endorsement by the National Marine Fisheries Service.

Obtained from G-Z products, Inc., 2401 Gold River Road, Cordova, California 95670.

Obtained from Calimar, Building 10, Brown Field Airport, Chula Vista, California 92011.

our experience, occurs chiefly between 8 p.m. and midnight.

On occasion the number of eggs collected in a 15 hour period may drop off to less than 1,000 per day. Two methods usually can be used to restimulate production: a) either by cleaning the tank, or b) by raising the temperature of the sea water to 16 C overnight followed by a reduction to 15 C the following morning. Either method is effective and both have been tested on a number of occasions with duplicate test groups of anchovies.

From time to time a large number (100,000 or more) of fertilized eggs are needed. Leong (1971) described his technique for hormone injections into individual fish to insure mass spawning. Of the various hormones and injection routines tried he has adopted the following procedure: an intraperitoneal injection of 50 I.U. of human chorionic gonadotrophin (HCG)⁴ in 0.1 ml Holtfreter's solution (Emmel and Cowdry, 1964) using a no. 26 hypodermic needle followed 24 hours later by an injection of 5 mg of ground salmon pituitary in 0.1 ml Holtfreter's solution (Emmel and Cowdry, 1964) using a no. 24 needle. In less than 18 hours after the second injection the anchovies spawn spontaneously; stripping was found to be unnecessary. Each injected female produces, on the average, about 10,000 eggs.

PACIFIC MACKEREL

The Pacific mackerel is a commercially important fish off California whose natural maximum spawning period ranges from June to August (Knaggs and Parrish, 1973).

Prespawning Pacific mackerel ranging in weight from 0.5-0.7 kg/fish have been subjected to a variety of photoperiods. Maturation is achieved with any light regime from a constant 4 hours of light per day to a constant 16 hours of light per day. Indoor tanks and intensity of illumination are the same as used for the anchovies (see above) but the temperature is maintained at 20 C (19.50-20.5 C). Two hundred and fifty fish are kept in each tank. Coarsely chopped anchovies are fed to the mackerel at 6% of the estimated mackerel live weight per day supplemented with 1% of the mackerel live weight per day in Oregon moist pellets. Under these conditions fish caught in March or April 1973 matured by June 1973 when they attained GIs of 7 or 8. Although there was spontaneous spawning in June this has not occurred again although at this writing (December 1973) the GIs are still high enough for spawning, i.e., 7 to 8. The spontaneous spawning produced eggs with low hatching success, about 5%. Approximately 200 mackerel caught in a spent condition were put into a larger outdoor tank (7.3 m diameter) and experienced seasonal changes in temperature (13 C in February and March, 22 C in July and August) and illumination. Gonads matured, spawning occurred spontaneously, but differed from laboratory maintained fish, in that the gonads decreased in size once spawning was over in August.

⁴ Purchased from Sigma Chemical Co., St. Louis, Missouri 63118.

When viable eggs and sperm are desired we catheterize 0.5-0.7 kg Pacific mackerel to determine if the maximum ovum diameter is at least 0.7 mm using the technique described by Stevens (1966) and modified by May (1972).

Intramuscular injections of females with 1 mg of ground salmon pituitary in 0.1 ml Holtfreter's solution followed by 500 I.U. of gonadotrophins from pregnant mare serum (PSM)⁵ 24 hours later stimulates hydration and ovulation. Eggs are obtained 12 hours later but must be stripped from the fish. HCG cannot be used with females because it causes overhydration of the ovaries and no release of eggs occurs. Males will respond to one injection of either 500 I.U. HCG or 1 mg of salmon pituitary. All of these results have been obtained by Roderick Leong of the National Marine Fisheries Service, La Jolla Laboratory, who is continuing to refine this technique with the ultimate goal of achieving spontaneously spawning animals throughout the year.

CROAKER

Haydock (1971) succeeded in maturing and spawning the Gulf Croaker at the National Marine Fisheries Service, La Jolla Laboratory, and we have used his technique at all times of the year with no extensive modifications for obtaining viable embryos. In the Salton Sea, this species usually spawns during April and May only. To summarize Haydock's work briefly: 1) To hold croakers in a sexually immature state requires 10 hours of light and 14 C temperature; 2) to mature fish in 1 month conditions must be changed to 16 hours of light and 22 C; 3) to keep mature fish at a GI of 5, maintain 16 hours of light and drop the temperature to 14 C; 4) to mature fish in 1 day after (3) above, increase the temperature to 22 C.

Continuous maintenance of fish at 16 hours light and 22 C causes eventual resorption of the gonads. Females will mature again in 2 weeks if exposed to 16 hours of light and a 22 C regime after spawning. Males usually remain running ripe under all these conditions once brought to ripeness.

The reader is referred to the doctoral thesis by May (1972) for his modifications of this technique, and for his method of assessing the state of maturity of the croakers. May spawned mature Bairdiella by using one intramuscular injection of 100 I.U. of PMS gonadotrophin in a carrier of Ringer's solution. Ovulation usually occurred 30-31 hours after the injection but eggs had to be obtained by stripping the fish.

May (personal communication) believes that not all mature Bairdiella eggs are ovulated with one injection. He says that the usefulness of the respawning method for the practical fish culturist would be very limited because the viability of the eggs on the second

¹ Purchased from Calbiochem, Los Angeles, California.

spawning is very low, sometimes almost nil. The best procedure, according to May, is to shift once-spawned fish to cold water and short days for several months to let them resorb the old batch of eggs completely, then gradually increase light and temperature and allow them to mature again. With adequate facilities and good planning, a continuous supply of eggs can be achieved as several batches of fish at different maturity stages could be maintained simultaneously.

STRIPED BASS

The striped bass is an anadromous fish introduced to the San Francisco area from the Atlantica coast in 1879 and 1882. It spawns in the lower reaches of rivers in the spring (Roedel, 1953). Thus far Messrs. Krishan Lal and Andrew Kuljis of the California Department of Fish and Game who are collaborating with NMFS on studies of striped bass have successfully matured only males in the Southwest Fisheries Center Laboratory. Females have showed increased GI and ova diameters, but an unexplained infection causing ulcers and abdominal perforations coincided with a decline in maturity. General laboratory conditions which have given us the best results thus far are as follows: tank size and water flow are the same as used for anchovies but only 50 fish over 1.5 kg each are kept in each tank. A photoperiod of 15 hours of light at an intensity of about 10 footcandles or less (60 watt incandescent bulb) is used and the temperature is maintained at a constant 19 C. A variety of foods have been used but chiefly whole anchovies are fed ad llbitum twice each day until the fish are satiated. Under this regime, males attained a GI of 10 in 6-8 weeks: females had a maximum ovum diameter of 0.75-0.8 mm and a GI of four under the same conditions. At this writing we are experimenting with suitable salinity alterations to simulate brackish conditions sought by the fish in nature. We expect this will stimulate maturation and subsequent ovulation.

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