PREMATURE EGG PROCUREMENT FROM STRIPED BASS

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ABSTRACT

Striped bass were successfully ovulated in Albemarle Sound, North Carolina some 150 miles from their natural spawning grounds and about one month before their normal spawning time. Brood fish were purchased from commercial fishermen, consequently the eggs so obtained were salvaged out of a sacrificial situation.

The fish were collected from pound nets following narcosis by electric shock; the narcosis being maintained by the addition of Quinaldine to the water in the boat holding tank until the female fish had been injected with human chorionic gonadotropin at the egg taking station.

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During egg maturation, the female striped bass were held individually in glass-fronted aquaria. Whenever a fish was transferred from one aquarium to another, or whenever egg samples needed to be taken, the fish normally were tranquilized by the addition of Quinaldine to the aquarium water before the fish were handled.

A myriad of problems were encountered during this experimental operation but, as a whole, it was successful. Of 243 striped bass brought to the egg taking station, 125 survived to ovulation and produced some 28,530,000 eggs.

Two hybridization experiments were successful: One male white perch x female striped bass cross produced 80,000 fry, and one male white bass x female striped bass cross produced 200,000 fry.

INTRODUCTION

During the period April 12 to May 4, 1966, an attempt was made to ovulate striped bass *Roccus saxatilis* (Walbaum) on an experimental basis using human chorionic gonadotropin hormone. The operation was conducted on the shore of Albemarle Sound near Roper, North Carolina. Fish purchased from local commercial fishermen were collected from any of 19-pound nets after being narcotized by electric shock. They were placed in a holding tank and transported by boat to the egg taking station where the female fish were injected with the hormone and held in an aquarium until ovulation. After fertilization, the eggs were incubated in standard McDonald hatching jars.

The primary purpose of the experiment was to determine if striped bass purchased from commercial fishermen could be induced to spawn successfully even though they were collected approximately 150 miles from their natural spawning grounds and up to one month prior to their normal spawning time. If successful, the experiment would open the way for the large-scale production of eggs and fry salvaged from fish otherwise lost via the commercial market.

Since the operation was conducted on an experimental basis, it was anticipated that many problems would need to be solved if success was to be obtained. An electro-fishing device was employed to facilitate handling and lessen the chance of injury to the fish but it was unknown whether the device would operate successfully in the waters of Albemarle Sound. In either case would high handling mortality develop as the result of collection procedures? Assuming that the initial collecting operations did not adversely affect the fish, could they withstand conditions in the boat holding tank enroute to the egg taking station? Having once arrived at the station, could they be held a week or more in outdoor holding tanks, or in aquaria, with a minimum of mortality from fungus, infectious diseases, dissolved oxygen deficiency, and other lethal factors? Should a significant mortality develop from any of these factors, could effective counter-measures be developed? Finally, and most important, could striped bass females 150 miles from their natural spawning grounds and one month prior to their normal spawning time be ovulated successfully using human chorionic gonadotropin and would healthy fry result from eggs fertilized by males equally far from their natural spawning grounds? All of these questions, as well as many others, materialized during the experimental operation.

FACILITIES AND EQUIPMENT

Egg Taking Station:

The station was established on the southern shore of Albemarle Sound adjacent to the NC-32 bridge. Albemarle Sound water was supplied to a 515-gallon reservoir tank by a 1½-HP centrifugal pump the intake for which was located 80 feet offshore at a normal depth of two feet.

Holding Facilities:

Boat Holding Tank: A 144-gallon tank equipped with a 12-volt

agitator mounted on the lid was used for holding striped bass during transport from the pound nets to the egg taking station.

Aquaria: Thirty-six 53-gallon glass-fronted aquaria (24" x 32" x 16") made of plywood or painted sheet metal were used to hold gravid female striped bass following hormone injection. A bottom draw-off was effected in each aquarium to promote efficient circulation.

Outdoor Holding Tanks: Ten 720-gallon outdoor holding tanks (4' x 8' x 3') were constructed of reinforced 5/8" exterior grade plywood. A bottom draw-off was effected in each holding tank. The tanks were located adjacent to the egg taking station and were individually supplied with water from the station reservoir tank.

Stock Tanks: Standard galvanized stock tanks of 100- and 232-gallon capacity also were used for holding brood stock. Water was supplied to the stock tanks by direct and continuous pumping from Albemarle Sound. The oval shape and the bottom drain of these tanks greatly facilitated rapid water exchange.

Hatching Jars:

Standard McDonald hatching jars were used for incubating the eggs.

Shipping Containers:

All eggs were shipped in plastic bags charged with oxygen. The plastic bag was carried in a cardboard carton containing a styrofoam liner for insulation. For trips of eight hours or more, ice was usually placed inside the styrofoam liner.

Electro-Fishing Device:

The electro-fishing device was mounted in a 16-foot, flat bottom, Monark aluminum boat. The device consisted of a three-phase, 180-cycle, 230-volt A.C. generator with a capacity of 7.5 amperes. The generator supplied a multi-tap transformer with a voltage range between 190 and 520 volts, then a rectifier for producing, through half-wave rectification, pulsed direct current of 60 pulses per second on a 50 percent duty cycle. The pulsed D.C. current was supplied to a 14-inch circular electrode of ¼-inch aluminum tubing mounted on a 10-foot fiber glassed aluminum probe. The aluminum boat served as the negative electrode for the system. Safety measures included a "dead-man" switch, rubber gloves and boots worn by the crew, and complete insulation of the generator.

PROCEDURES

When collecting fish from the pound nets, the commercial fishermen wearing insulating rubber gloves and operating from a wooden scow, worked the fish into a small pocket (approximately 30' x 5') at the end of the trap section. The electro-fishing boat was moored alongside of the pocket for easy access to the fish. The electrode was placed in the pocket and current applied until all fish were narcotized. Selected fish were then removed by means of an insulated dip net of knotless ¼" mesh nylon. Sex of the striped bass was then determined by stroking the belly to establish the presence or absence of milt. Males were taken regardless of size if they produced milt. Females under three pounds in weight generally were rejected.

Accptable fish were held in the boat holding tank until they could be taken ashore. Seven ppm Quinaldine was used in the tank as a tranquilizer and 5 ppm acriflavine added as a bactericide. A maximum of 100 pounds of fish was held for not longer than two hours at a time and continuous aeration maintained by the agitator.

When the boat arrived at the station, those females for which there was space in the aquaria were removed immediately from the tank with a dip net and injected with human chorionic gonadotropin. The dosage rates employed were: 1,000 I.U. for fish weighing up to eight pounds; 1,500 I.U. for fish weighing eight to 12 pounds; and

2,000 I.U. for fish weighing over 12 pounds. The hormone was injected with a 2½ ml. disposable plastic syringe. A different syringe was used for each fish as a precaution against the spread of disease and the used syringes discarded. To facilitate the injection and further reduce the handling of females, the individual weights were estimated when determining the proper hormone dosage.

After injection, the fish were transferred by dip net into a No. 2 washtub containing 1:15,000 parts, by weight, malachite green in which they were submerged for 30 seconds as a precaution against fungus. They were then carried into the station and placed, individually, into vacant aquaria. The remainder of the fish in the boat holding tank were subjected to the malachite green dip and then segregated by sex in the outdoor holding tanks or the stock tanks where they remained until needed.

Initial egg samples were collected 24 or 48 hours following injection, depending upon water temperature. While the daily minimum water temperatures remained below 63° F., the initial egg sample was taken 48 hours following injection. However, when the daily minimum temperatures were 63° F. or higher, the initial egg sample had to be taken 24 hours after injection because of the accelerated egg maturation. The egg sample was extracted, and the degree of maturation determined, following the techniques described by Stevens (1964). All egg samples were collected with the aid of Quinaldine until the female fish became torpid just prior to ovulation. The females became sufficiently tranquilized for egg samples to be easily collected after exposure to 7 ppm Quinaldine for five to 10 minutes. When water temperatures were 70° F. or above, the same results were achieved using 3 ppm Quinaldine. As ovulation became imminent, the use of Quinaldine was discontinued to prevent the release of eggs into the aquarium.

Beginning one hour before the predicted ovulation time, and at 30-minute intervals thereafter, the belly of the female was palpated. At the first release of eggs, the female was carried by dip net to a table for artificial spawning. During the process, the female was kept ensnared in a fold of the net and held so that the vent was directly over a dishpan containing about ½ inch of water. The female was then rapped sharply on the head, just above the eyes, with a length of 1" diameter plastic pipe. This rendered the fish unconscious, whereupon the eggs were released immediately. The male, also "knocked-out" in like manner, was held over the dishpan and milt stripped concurrently with the eggs. The eggs and milt were then thoroughly mixed. After one minute, the excess water and milt were decanted. The eggs were then measured volumetrically and placed in the hatching jars at the rate of 100,000 per jar.

Flowing water kept the eggs in constant motion throughout incubation. Because of insufficient facilities at other hatcheries early in the season, a few fry were hatched at this station. Usually, however, the eggs were packaged for shipment after eight to 20 hours incubation at Albemarle Sound.

Prior to shipment, dead eggs were siphoned from the hatching jar and the viable eggs packaged 150,000 to 200,000 to the bag for trips of two to six hours, and from 50,000 to 100,000 for longer trips. On the longer trips, ice generally was placed inside the styrofoam liner of the shipping carton to prevent the water from becoming too warm.

DISCUSSION

A myriad of problems were encountered during this experimental operation—some of which were satisfactorily solved, some were not. Despite many adversities and many unanswered questions remaining, the experiment was successful.

The use of Albemarle Sound water for holding brood stock and for incubating eggs proved troublesome throughout the entire season. The greatest disadvantage was diurnal temperature fluctuations of 5° to 10° F., and occasionally as great as 15° F. Another major disadvantage

was the constant pumping of silt, sand, detritus and even aquatic invertebrates into the water system. Most of the larger items were removed by repeated screenings. The heavy silt load, however, could not be removed and frequently during periods of strong northerly winds, turbidity precluded observations of either fish in the aquaria or eggs in a hatching jar.

One of the major concerns of the experiment, while still in the planning stage, was the anticipated loss of brood fish from injuries sustained during capture. To minimize these losses, the fish were narcotized by electric shock while still in the pound net and then handled only while in a state of narcosis. Albemarle Sound water in the area was highly conducive to electro-fishing operations. The resistance of the water varied between 84 and 580 ohms and, as a result, sufficient amperage for narcotizing fish could be obtained at relatively low voltages—i.e., 190 volts produced from 0.5 to 3 amperes. Although the device proved highly effective in narcotizing the fish, a question arose whether the narcosis affected subsequent egg mortality rate. In an effort to evaluate the benefit derived from using the shocker at the pound net, the spawning results from 17 females collected on April 25 and 26 without benefit of the shocker were compared to those from a second group of 17 collected under narcosis on April 23 and 24 and with a third group of 17 also collected under narcosis on April 27. Of the 17 collected without the shocker, only three produced viable eggs; while in the other two groups, four and eight, respectively, produced viable eggs. These data indicate that narcotizing the fish by electric shock does result in a greater production of viable eggs.

Another practice followed in 1966 was the reinjection of all fish that evidenced no egg development within 48 hours following the first injection. In all cases, the quantity used in the reinjection was the same as in the first injection. The data gathered from 28 females thus reinjected clearly indicated that the practice should be discontinued: 13 of the females died before ovulation; nine produced eggs resulting in 100 percent mortality; two produced eggs resulting in 80 percent mortality; and a mortality of the eggs from the remaining four is unknown as they were stocked immediately after spawning because of a water supply failure.

Most of the females brought to the station were injected immediately. To determine the efficacy of immediate injection, the following two experiments were conducted concurrently: (1) Ten females were held for six days before injection and then transferred to aquaria. One female died prior to injection, three prior to ovulation, and all eggs taken from the remaining six died; (2) Six females were held in aquaria prior to injection. Four of the females were accidentally killed during a prophylactic treatment for fungus. The two surviving females produced 75 percent viable eggs. These two experiments indicated that egg production decreased as the retention period of the brood stock increased.

Brood stock mortalities resulting from fungus infections were negligible. Fish exhibiting fungus responded favorably to a one-hour treatment with 10 ppm potassium permanganate. The practice of subjecting all brood stock to a 30-second dip in 1:15,000 parts, by weight, malachite green immediately upon their arrival at the egg taking station virtually eliminated all fungus.

During the latter part of the season, however, severe losses of brood stock exhibiting characteristic lesions of "redfin" disease developed. Treatments with formalin, salt, malachite green, and acriflavine, or the sterilization of aquaria with HTH had no apparent effect on this disease.

Determining the exact time of ovulation for females collected at Albemarle Sound proved quite difficult. Few of the criteria established at Weldon (1965) seemed applicable. At Weldon, the perivitelline fluid of eggs at ovulation was crystal clear, consolidation of oil globules had occurred and the eggs, when placed onto a microscope slide, readily assumed a hexagonal shape. The perivitelline fluid in eggs at Albemarle Sound always remained granular in appearance and consolida-

tion of the oil globules rarely had occurred in more than 50 percent of the eggs, and the eggs would assume a hexagonal shape on a microscope slide only after the excess water had been removed. The granular appearance of the eggs made it very difficult to distinguish between eggs some two hours before ovulation and eggs at ovulation—the only apparent difference being a slightly less granular appearance in the latter. Likewise, it was very difficult to distinguish between ripe and overripe eggs—the presence of free oil globules, which presumably came from deteriorating eggs, was the only indication of overripeness. Determining the proper time of ovulation was, in the final analysis, a decision based upon experience gained through repeated "trial and error" determinations.

Size of the brood fish was not infallible as a criterion for determining sexual maturity. Occasionally even eight- and 12-pound females were found to contain non-functioning ovaries and males weighing three to four pounds would yield no milt. The data indicated that a maximum, as well as a minimum, size should be placed upon the females brought to the hatchery (Table 1). Brood females weighing in excess

TABLE 1. STRIPED BASS EGG AND ADULT MORTALITIES, BY WEIGHT OF BROOD FEMALES.

Fish weight range (pounds)	Number of Specimens	Egg Mortality				Adult Mortality
		Unknown	0-49%	50-99%	100%	
3.0 - 4.9	68	10	5	10	13	30
5.0 - 6.9	110	15	15	6	16	58
7.0 - 8.9	35	1	3	5	6	20
9.0 - 18.9	30	3	1	—	6	20

of eight pounds produced very few viable eggs when subjected to the normal handling procedures.

Sperm viability was questionable in many cases. Normally, the milt obtained from Albemarle Sound males was little in quantity and of thick consistency. Some motile sperm were observed each time a sample was examined under a compound microscope, still it is believed that poor quality sperm was responsible for much of the egg mortality.

Brood stock were successfully transported from Albemarle Sound to the Weldon hatchery. The fish were narcotized at the pound net by electric shock, and were transported in water containing 7 ppm Quinaldine and 5 ppm acriflavine. Of 14 females thus transported, 11 survived to ovulation.

During the 1966 season, 243 female striped bass were processed through the Albemarle Sound station.

HYBRIDIZATION

While the primary purpose of the Albemarle Sound station was the experimental production of striped bass eggs, several hybrid crosses within the genus *Roccus* were also attempted. These crosses were made in the hope that a fish might result that would be more adaptable to North Carolina reservoirs than the striped bass with its specialized spawning requirements.

Five attempts were made to hybridize male white bass *Roccus chrysops* (Rafinesque) with female striped bass. The white bass had been collected with gill nets from Fontana Reservoir and by electrofishing in the tailrace of Tuckertown Powerhouse. Although the males were ripe when captured, they were relatively "spent" by the time they reached Albemarle Sound. This fact may have significantly affected the success of some attempted crosses. One attempt was highly successful and yielded an estimated 225,000 fry from 250,000 eggs. The other four attempted crosses proved unsuccessful although in one instance apparently viable eggs were obtained that died during incubation. Eggs from the one successful experiment were hatched at Albemarle Sound

and the fry equally divided between the State hatcheries at Fayetteville and Table Rock and the National Fish Hatchery at Edenton. A complete loss was reported from the rearing attempt at Table Rock but the hybrid fry currently are growing rapidly at both the Edenton and the Fayetteville hatcheries. No attempt was made to hybridize male striped bass with female white bass.

Three attempts were made to hybridize male white perch *Roccus americanus* (Gmelin) and female striped bass. All white perch were collected from commercial pound nets in Albemarle Sound and the males were sexually mature when collected. Two of the three attempts failed, but one was highly successful with 100,000 eggs yielding some 80,000 fry. As an experimental control, 400,000 eggs from the same female striped bass were fertilized with a male striped bass. These 400,000 eggs yielded approximately 300,000 striped bass fry. The hybrid fry were divided between the Fayetteville and Table Rock hatcheries. They suffered a 100 percent mortality at Table Rock, and only 18 fry survived to the two-inch fingerling stage at Fayetteville.

Considerable difficulty was experienced in determining the stage of egg development in the female white perch, and only one survived to what was thought to be ovulation. An attempt to cross this fish with a male striped bass resulted in 100 percent egg mortality.

It is believed that the Albemarle Sound hybridization experiments resulted in the first successful cross of the white perch and the striped bass.

CONCLUSIONS

1. Striped bass purchased from commercial fishermen in Albemarle Sound can be successfully ovulated using human chorionic gonadotropin hormone.
2. Striped bass were easily narcotized using an electro-fishing device thereby reducing the handling and subsequent strain upon the fish during collection and transportation.
3. Fish remained anesthetized in 7 ppm Quinaldine for periods up to two hours and evidenced no adverse effects.
4. Milt from male striped bass collected in Albemarle Sound, though thick in consistency and available only in small quantities, did contain motile sperm. Sperm viability, however, frequently appeared questionable.
5. Female striped bass should be injected with the hormone as soon as possible after reaching the holding site.
6. A 30-second dip in 1:15,000 parts, by weight, of malachite green virtually eliminated fungus infection.
7. A bottom draw-off was required in aquaria and holding tanks to obtain adequate water circulation.
8. Fish failing to evidence egg development by 48 hours following the initial hormone injection rarely produced viable eggs following a second hormone injection.
9. Females, when exposed to 3-7 ppm Quinaldine, were subdued within five minutes to an extent that an egg sample could be collected without unduly exciting the fish.
10. When ovulation is imminent, Quinaldine should not be used since the anesthetizing effect may cause the fish to release eggs into the aquarium.
11. Size cannot be used as the sole criterion for judging the sexual maturity of striped bass in Albemarle Sound.
12. Turbidity, associated with unfiltered water taken from Albemarle Sound, proved a major problem at the egg taking station.
13. Extreme diurnal water temperature fluctuations adversely affected both egg maturation and incubation.
14. Gravid females collected from Albemarle Sound were successfully transported to the Weldon Hatchery, a distance of some 125 miles, then ovulated with human chorionic gonadotropin.
15. Hybrid fry can be produced by crossing male white bass and female striped bass.
16. Hybrid fry can be produced by crossing male white perch with female striped bass.