SRAC Publication No. 421

Southern
Regional
Aquaculture
Center

November 1991

Introduction to Hormone-Induced Spawning of Fish

R.W. Rottmann, J.V. Shireman, and F.A. Chapman*

The demand for fish for food, recreation, and ornamental aquariums is steadily increasing. Natural fish populations have declined during the last several decades because of environmental degradation and over-fishing. This has resulted in an increased effort in the development of techniques for hatchery production of fish. Traditional aquaculture species such as trout, catfish, common carp, golden shiner, and goldfish reach sexual maturity and spawn in hatcheries or ponds, when conditions are appropriate.

However, a number of fish species that have or potentially have great economic significance for aquaculture do not reproduce spontaneously in captivity. Many of these fish spawn in environments that are nearly impossible to simulate in a hatchery. Hormone-induced spawning is the only reliable method to induce reproduction in these fishes.

Hormone-induced spawning of fish has been used for almost 60 years. Surprisingly, the same procedures, with only minor modifications, have been used to spawn an entire range of fishes from the ancient sturgeon and paddlefish to carp, catfish, salmon, sea bass, redfish, snook, and mullet. In addition to breeding other desirable fish species, induced spawning can be used to:

- produce hybrids that are different from the parent species;
- produce sterile polyploid fish (for example, sterile triploid grass carp for aquatic weed control);
- synchronize reproduction of large numbers of fish for simultaneous spawning, thereby simplifying production and marketing of the fish;
- produce fry outside the normal spawning season for maximum hatchery production and to provide fish when the price and market demand is greatest; and
- maximize survival of fry under controlled hatchery conditions.

Proper fish handling

The physical injury and physiological stress of capturing, handling, transporting, injecting, and holding brood fish can have a greater detrimental effect on spawning success than almost any other factor. Fish must be handled carefully and optimum water conditions must be maintained to minimize stress. The importance of proper handling and water quality cannot be overemphasized. Female brood fish ready for spawning are in a particularly delicate condition. When female fish are stressed or injured, they may undergo rapid physiological changes that can result in the breakdown (resorption) of eggs in the ovary. Crowding, dissolved oxygen depletion, rapid changes in temperature, and osmotic imbalance are well known causes of stress and must be avoided. Suboptimum conditions, while not immediately lethal, may stress brood fish, resulting in delayed mortality or failure to spawn. Reducing stress and injury to brood fish can greatly increase the success of hormone-induced spawning.

Determine sexual maturity

The external appearance of brood fish has long been used to assess the stage of sexual development. In some species, males change in appearance during the spawning season. These physical changes make it relatively easy to identify sexually mature males. However, secondary female sex characteristics such as plumpness of the abdomen and redness of the vent are extremely subjective and can be misleading. Sampling the eggs and sperm of the brood fish eliminates the guesswork in determining the stage of sexual development.

Milt can usually be stripped from males of most species when they

* Institute of Food and Agricultural Sciences, University of Florida

are ready for spawning by applying gentle pressure to the abdomen between the pelvic fins and the vent. Sperm viability usually can be determined by observing motility with a microscope.

Several methods are available to determine the developmental stage of the eggs in the fish's ovary. The diameter and appearance of the egg and the position of the nucleus in the egg are visual indicators of development. The steroid assay procedure determines the physiological response of the eggs to hormones. Both require that an egg sample be collected. The ovary can be sampled with either a rigid or flexible tube (catheter). An egg sample can also be taken by making a small incision along the belly or side of the fish. This technique is commonly used for sturgeon and paddlefish. An understanding of sperm viability and egg stage development will greatly improve the success of hormone-induced spawning of fish.

Control of reproduction

Reproduction in fishes is regulated by external environmental factors that trigger internal mechanisms. The final event of the reproductive cycle, the release of eggs and sperm resulting in spawning, can be controlled by either placing the fish in an appropriate environment or by changing the fish's internal regulating factors with injected hormones or other substances. The internal mechanisms that regulate spawning are similar for most fishes. The external environmental factors that control reproduction, however, vary considerably among species.

For this reason, more is known about the internal regulatory mechanism of fish reproduction than the specific environmental requirements for spawning each species. Environmental factors that have been shown to play a significant role in the reproductive cycle are:

- photoperiod;
- water temperature;
- water quality (e.g., dissolved oxygen, pH, hardness, salinity, alkalinity);
- flooding and water current;
- tides and cycles of the moon;
- weather cycles (e.g., atmospheric pressure, rainfall);
- spawning substrate (e.g., aquatic plants, sticks, gravel, spawning caverns);
- nutrition;
- disease and parasites; and
- presence of other fish.

These factors do not function independently of each other, but are interrelated.

The internal mechanism that regulates the process of reproduction in fish is the brain-hypothalamus-pituitary-gonad chain (Figure 1). Hormone-induced spawning techniques influence this sequential mechanism at several levels, by either promoting or inhibiting the process. The primary substances used for hormone-induced spawning have been: (1) pituitary extracts and (2) purified gonadotropin to stimulate the ovaries and testes; (3) LHRH analogs (LHRHa) alone or in combination with (4) dopamine blockers which enhance the potency of LHRHa to stimulate the pituitary; or (5) steroids to stimulate the gametes directly. The appropriate hormone preparation should be selected on the basis of the species to be spawned and the availability y of the hormones.

Preparation of hormones

The hormones must be mixed and stored properly to prevent contamination and preserve potency. The proper dosage must be calculated for the brood fish, and the optimum injection schedule must be used for best results. To calculate the proper dosage, (1) the recommended dose, (2) approximate weight of the brood fish, and (3) the desired volume of the injection must be determined. The quantity of hormone to be injected can then be calculated from the weight of each individual brood fish and the appropriate injection schedule.

Taking the spawn

The eggs and milt of fish can be taken by several different methods: (1) tank spawning; (2) hand stripping, and (3) surgically removing the eggs. The method of choice depends on the fish species,

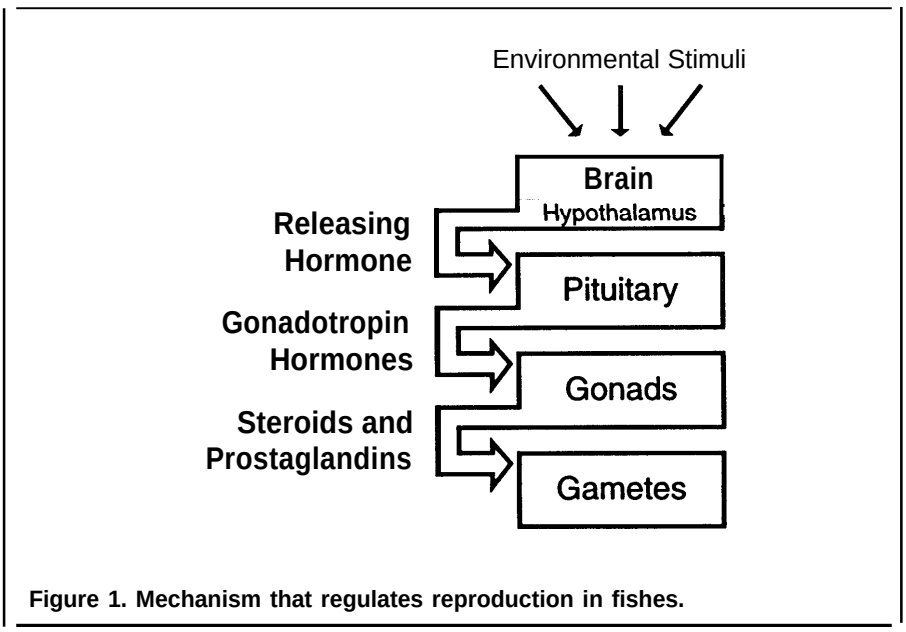

Figure 1. Mechanism that regulates reproduction in fishes.

hatchery facilities, experience and skill of the hatchery staff, and the desired manipulations of eggs, sperm, or fertilized eggs.

Tank spawning is the simplest method for obtaining a hormone-induced spawn. Brood fish of both sexes are placed together in the spawning tank following injection(s). The female ovulates when she is physiologically ready. The males stimulate the female to release the eggs and fertilize the spawn.

Hand stripping is commonly used for taking the spawn of many species of fish. Brood fish are separated by sex prior to hormone injection to prevent spawning in the holding tank. It is important to determine the exact time of ovulation when hand stripping. In many species, egg quality can deteriorate rapidly if the eggs are not taken shortly after ovulation. For most species, ovulation can best be verified by checking the female to determine when eggs flow freely from the vent. To strip the eggs, the fish is held slightly on her side, tail down; gentle hand pressure is applied to the abdomen, moving toward the vent. The stream of eggs is directed into a clean, dry bowl positioned so that water from the fish does not drip onto the eggs. It is important to insure that no water comes in contact with the eggs until after the milt is added and mixed. Water activates the sperm and also causes the opening through which the sperm enters the egg to close.

Because the internal anatomy of fish vary greatly, hand stripping may be difficult in some species. Sturgeon and paddlefish have no ovarian sac; the eggs are released into the abdominal cavity during ovulation. The best method for taking the spawn of these species is to surgically remove the eggs. For delicate species that seldom survive the rigors of hand stripping, humanely killing them and surgically removing the eggs may be the best option. In addition, more eggs can usually be obtained by this method than by hand stripping.

Fertilizing the spawn

The eggs obtained by hand stripping or surgical removal are usually fertilized with fresh milt. Males are captured, wiped off, and held belly down over the bowl containing the eggs. The portion of the abdomen posterior to the pelvic fins is gently massaged to extrude the milt onto the eggs. Milt can be collected from males and stored up to three weeks prior to stripping eggs.

Ovulated eggs of many species such as white bass, sturgeon, paddlefish, and common carp become sticky after water is added. During natural spawning, this stickiness causes the eggs to become attached to rocks, sticks, or aquatic plants. Catfish eggs are connected by a sticky matrix that holds the eggs together in a mass. In the hatchery, this stickiness causes problems during incubation. Siltclay, Fuller's earth, or bentonite suspension, urea and salt solution, and tannic acid solution are preparations commonly used to deactivate the sticky layer of fish eggs. In addition, the gelatinous matrix of catfish egg masses can be dissolved with sodium sulfite so the eggs can be incubated in hatching jars.

Induced hatchery spawning of fish requires a continuous series of decisions, any of which if improperly made, can diminish or completely obliterate the success of the project. There are many ways to fail at each step and only a very few that are productive. Consistent performance requires strict attention to detail.

Additional SRAC fact sheets on induced spawning

SRAC 422 *Capturing, Handling, Transporting, Injecting, and Holding Brood Fish for Induced Spawning*

SRAC 423 *Determining Sexual Maturity of Broodstock for Induced Spawning of Fish*

SRAC 424 *Hormonal Control of Reproduction in Fish for Induced Spawning*

SRAC 425 *Hormone Preparation, Dosage Calculation, and Injection Techniques for Induced Spawning* of *Fish*

SRAC 426 *Techniques for Taking and Fertilizing the Spawn of Fish*

SRAC 427 *Induction and Verification of Triploidy in Fish*

The work reported in this publication was supported in part by The Southern Regional Aquaculture Center through Grant No. 89-38500-4516 from the United States Department of Agriculture.