1983

# Chemical induction of spawning by serotonin in the ocean quahog Arctica islandica (Linne)

M. C. Gibbons
*Virginia Institute of Marine Science*

J. G. Goodsell

M. Castagna
*Virginia Institute of Marine Science*

R. Lutz

*Journal of Shellfish Research*, Vol. 3, No. 2, 203–205, 1983.

# RESEARCH NOTE

## CHEMICAL INDUCTION OF SPAWNING BY SEROTONIN IN THE OCEAN QUAHOG *ARCTICA ISLANDICA* (LINNÉ)

M. C. GIBBONS[1], J. G. GOODSELL[2], M. CASTAGNA[1] AND R. A. LUTZ[2]

[1] *Virginia Institute of Marine Science and School of Marine Science College of William and Mary Wachapreague, Virginia 23480*

[2] *Department of Oyster Culture New Jersey Agricultural Experiment Station Cook College, Rutgers University New Brunswick, New Jersey 08903*

***ABSTRACT*** Serotonin injected into the anterior adductor muscle induced spawning in the ocean quahog *Arctica islandica* (Linné) when using either individual or mass spawning techniques. This represents the first successful attempt to induce the release of gametes in this species which historically has been unresponsive to conventional spawning stimuli. The gametes released were competent and fertilization occurred without treating the encapsulated eggs with ammonium hydroxide or other chemicals. Larvae were reared through metamorphosis to early juvenile stage.

***KEY WORDS:*** Ocean quahog, *Arctica islandica*, spawning, serotonin

### INTRODUCTION

The ocean quahog *Arctica islandica* (Linné) spawns from August through November on the southern New England shelf and off New Jersey (Jones 1981, Mann 1982). Attempts to spawn the ocean quahog in the laboratory have been unsuccessful. Various combinations of stimuli such as thermal shock, addition of gonadal products, salinity and pH changes, and exposure to hydrogen peroxide, which are effective with many other bivalve species, have not induced spawning (Loosanoff 1953, Landers 1976, Lutz et al. 1982, Mann 1982). All larvae of ocean quahogs cultured to date under laboratory conditions have been reared from stripped gametes that had been fertilized after pretreatment of eggs with ammonium hydroxide (Landers 1976, Lutz et al. 1981).

Serotonin (5-hydroxytryptamine, creatinine sulfate complex) has proven to be an effective chemical inducer of spawning for many bivalve species (Matsutani and Nomura 1982, Gibbons and Castagna [in press]). The injection of serotonin into the anterior adductor muscle or gonad of certain bivalve species when ripe will induce spawning using individual spawning techniques without any additional stimuli. The present study describes the successful spawning of ocean quahogs in the laboratory using serotonin.

### MATERIALS AND METHODS

Sexually mature ocean quahogs, ranging in shell length from 8 to 13 cm, were obtained in October 1983 using a commercial hydraulic dredge in 50 to 80 m of water off Cape May, NJ. The specimens were kept on ice for approximately 12 hours during transport from the sampling site. Upon arrival at the Wachapreague Laboratory of the Virginia Institute of Marine Science, half of the ocean quahogs were immediately placed in individual dishes of seawater for spawning while the other half were held in a recirculating seawater table at 15–16°C.

A 2-mM solution of serotonin (Sigma Chemical Company, St. Louis, MO) was prepared by dissolving crystalline serotonin in 1-μm-filtered seawater. Each ocean quahog was washed and a small notch filed into the valve margin adjacent to the anterior adductor muscle. To induce spawning, 0.4 mℓ of the 2-mM serotonin solution was hypodermically injected into the anterior adductor muscle.

Both individual and mass spawning techniques as described by Castagna and Kraeuter (1981) were utilized without any thermal shock or other stimulation to spawn ocean quahogs. All spawning experiments were conducted at a salinity of 32 ppt and at a controlled temperature of 15–16°C. Ocean quahogs were spawned by placing single specimens in glass dishes containing 1ℓ of 1-μm-filtered seawater. Mass spawning was achieved by placing the quahogs in troughs containing 140 ℓ of static, 1-μm-filtered seawater. Equal numbers of quahogs in the control groups were treated in the same manner as the test groups except they were injected with 0.4 mℓ of 1-μm-filtered seawater instead of the serotonin solution. The control animals from trial 1 of the mass spawning were the test group for trial 2. The G-test of independence and Williams' correction for a 2 × 2 contingency table were used to statistically determine

Contribution No. 1220 from Virginia Institute of Marine Science. Publication No. D–32401–2–85, supported by state and various National Oceanic and Atmospheric Administration Sea Grant funds to Rutgers University.

whether spawning was independent of injection with the serotonin solution (Sokal and Rohlf 1981).

Eggs obtained from the serotonin-induced spawnings were fertilized using standard techniques developed for other bivalves (Loosanoff and Davis 1963, Castagna and Kraeuter 1981). Eggs were not pretreated with ammonium hydroxide or other chemicals prior to fertilization. The larvae were reared through settlement and metamorphosis to early post-set at 13.5°C.

## RESULTS AND DISCUSSION

Injection of the serotonin solution induced gamete release in both the individual and mass spawning trials, although greater percentages (35.5% and 37.1%) of ocean quahogs spawned using the mass spawning technique than for the individual method (17.1% and 22.5%) (Table 1). In each case larger numbers of quahog males spawned than females. This, however, may be a dose response. Ocean quahogs injected with serotonin extended their siphons, probed with their feet, and began spawning within 15 minutes. The control groups injected with filtered seawater did not exhibit any of these behavioral patterns and did not spawn.

The egg capsules of the ocean quahog are unlike any structures described for bivalves (Castagna et al. 1982). The encapsulated eggs were slightly ovoid and ranged from 75.0 to 85.0 $\mu$m in diameter ($\bar{X}$ = 79.9 $\mu$m; S.D. = 1.3 $\mu$m). Fertilization occurred in mass spawnings and similarly upon addition of sperm in individual spawnings without chemical pretreatment of the freshly spawned eggs. The egg capsules have been suggested as being responsible for the difficulty in spawning ripe ocean quahogs or in fertilizing stripped eggs (Lutz et al. 1982), but no difficulty was observed with this technique. Exposure of stripped eggs to ammonium hydroxide may result in a lower percentage of normally developing larvae compared to naturally spawned eggs (Loosanoff and Davis 1963). Serotonin-induced spawning appears to be a more effective means of obtaining gametes from ripe ocean quahogs than stripping gametes from mature individuals.

The development of larvae from the trochophore stage through metamorphosis was similar to that described for larvae of this species obtained from fertilization of stripped eggs (Landers 1976; Lutz et al. 1981, 1982). Developing eggs were encapsulated up to the gastrula stage, at which time the egg capsules were lost. Metamorphosis occurred at shell lengths of 170.6 to 266.7 $\mu$m ($\bar{X}$ = 220.5 $\mu$m; S.D. = 19.8 $\mu$m) between 37 and 62 days after natural fertilization, which was similar to results obtained by others for fertilized stripped eggs (Landers 1976, Lutz et al. 1982).

To date, serotonin has been effectively utilized to induce spawning in several species of bivalves (Matsutani and Nomura 1982, Gibbons and Castagna [in press]). It is a neurotransmitter that occurs naturally in the cerebropleural, pedal, and visceral ganglia of *Arctica islandica* at concentrations of 20 $\mu g \cdot g$ fresh tissue$^{-1}$ (Welsh and Moorhead 1960). In laboratory studies, serotonin has been found to excite excised hearts of ocean quahogs by stimulating the cardioregulatory nerves (Gaddum and Paasonen 1955, Leake and Walker 1980). The physiological role of serotonin as an inducer of spawning in bivalves is unknown.

The use of serotonin has induced spawning in the ocean quahog, a bivalve that historically has been difficult to spawn in the laboratory. Serotonin has potential value to induce spawning in other bivalves which are resistant to conventional spawning stimuli. The advantages of this technique include ease of use and rapid and synchronous spawning of ripe individuals.

TABLE 1.

Numbers of ocean quahogs induced to spawn by injection of serotonin.

| Spawning Technique | Treatment | Number Tested | Number Spawned | Percentage Spawned | Number Males | Number Females |
|---|---|---|---|---|---|---|
| Individual – trial 1 | Serotonin | 35 | 6* | 17.1 | 5 | 1 |
| | Control | 35 | 0 | 0 | 0 | 0 |
| Individual – trial 2 | Serotonin | 40 | 9* | 22.5 | 7 | 2 |
| | Control | 40 | 0 | 0 | 0 | 0 |
| Mass – trial 1 | Serotonin | 35 | 13* | 37.1 | 10 | 3 |
| | Control | 35 | 0 | 0 | 0 | 0 |
| Mass – trial 2 | Serotonin | 31 | 11 | 35.5 | 10 | 1 |

*significant at $P < 0.005$.

## REFERENCES CITED

Castagna, M., J. Goodsell, R. Lutz & R. Mann. 1982. The egg capsule of *Arctica islandica. J. Shellfish Res.* 2:91–92.

Castagna, M. & J. N. Kraeuter. 1981. Manual for growing the hard clam *Mercenaria. Va. Inst. Mar. Sci. Spec. Rep. Appl. Mar. Sci. Ocean Eng.* 249: 110 p.

Gaddum, J. H. & M. K. Paasonen. 1955. The use of some molluscan hearts for the estimation of 5-hydroxytryptamine. *Br. J. Pharmacol.* 10:474–483.

Gibbons, M. C. & M. Castagna. (in press) Serotonin as an inducer of spawning in six bivalve species. *Aquaculture*.

Jones, D. S. 1981. Reproductive cycles of the Atlantic surf clam, *Spisula solidissima,* and the ocean quahog, *Arctica islandica,* off New Jersey. *J. Shellfish Res.* 1:23–32.

Landers, W.S. 1976. Reproduction and early development of the ocean quahog, *Arctica islandica,* in the laboratory. *Nautilus* 90:88–92.

Leake, L. D. & R. J. Walker. 1980. *Invertebrate Neuropharmacology*. New York, NY: John Wiley and Sons. 102–143.

Loosanoff, V. L. 1953. Reproductive cycle in *Cyprina islandica. Biol. Bull. (Woods Hole)* 104:146–155.

_____ & H. C. Davis. 1963. Rearing of bivalve mollusks. *Adv. Mar. Biol.* 1:1–136.

Lutz, R. A., J. G. Goodsell, R. Mann & M. Castagna. 1981. Experimental culture of the ocean quahog, *Arctica islandica. J. World Maricul. Soc.* 12:196–205.

Lutz, R. A., R. Mann, J. G. Goodsell & M. Castagna. 1982. Larval and early development of *Arctica islandica. J. Mar. Biol. Assoc. U.K.* 62:745–769.

Mann, R. 1982. The seasonal cycle of gonadal development in *Arctica islandica* from the Southern New England Shelf. *U.S. Natl. Mar. Fish. Serv. Fish. Bull.* 80:315–326.

Matsutani, T. & T. Nomura. 1982. Induction of spawning by serotonin in the scallop, *Patinopecten yessoensis* (Jay). *Mar. Biol. Lett.* 3:353–358.

Sokal, R. R. & F. J. Rohlf. 1981. *Biometry.* 2nd ed. San Francisco, CA: W. H. Freeman & Co. 859 p.

Welsh, J. H. & M. Moorhead. 1960. The quantitative distribution of 5-hydroxytryptamine in the invertebrates, especially in their nervous systems. *J. Neurochem.* 6:146–169.