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BROOD STOCK CONTROL IN A LOBSTER CULTURE SYSTEM

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ABSTRACT

Brood stock now used in lobster culture systems is usually obtained as wild stock from the commercial fishery. Socio-legal problems in many areas prevent the purchase of ovigerous females from the fishery, but lack of understanding of the natural reproductive cycle and the mechanisms controlling it have resulted in low incidence of successful oviposition by wild females in captivity. Most of these problems can now be avoided.

Techniques are now available for recognizing preovigerous female American lobsters in the spring commercial catch, and a high percentage of such females will produce eggs in captivity if handled properly. Once eggs are obtained, they can be assigned to one of three temperature schedules that will permit larvae to be obtained in all months of the year.

Lobsters grown in a culture system at constant 20-22°C will mature rapidly, but few will extrude eggs on that temperature schedule. To obtain successful oviposition with such females, their approaching maturity must be recognized in advance so that they can be mated and transferred to a natural environmental regime until spawning occurs.

INTRODUCTION

In the last decade, researchers have eliminated many of the biological and engineering problems that originally confronted lobster culturists. However, in two critical areas - nutrition and brood stock management - progress has been distressingly slow. These two areas now are recognized as major impediments to further development of commercial lobster culture.

When lobster culture was first seriously considered, surprisingly little was known about the natural reproductive cycles and the mechanisms controlling them. A summary of the fragmentary and often inaccurate

information available at that time can be found in Aiken and Waddy (1976). Research since then has more clearly defined both the basic biology and the controlling mechanisms (Aiken and Waddy 1980a, b), and as a result, solutions are now available for some of the broodstock problems that have recently been described (Hedgecock et al. 1978; Richards and Wickins 1979; Schuur et al. 1976; Van Olst et al. 1980).

Lobster culturists have had difficulty obtaining successful extrusion and egg attachment with females maintained in captivity (Hedgecock et al. 1978; Van Olst et al. 1980), and have had to rely on wild-caught ovigerous females for their larval production. Since possession or sale of ovigerous American lobsters is prohibited in most areas of the fishery, the impracticality of this approach on a large scale is obvious.

We have maintained a pilot scale lobster culture facility for 4 years, and have hatched females year-round in an attempt to keep this facility in constant production. Larval production in the early stages of this project came exclusively from ovigerous females obtained under special permit from the fishery. With these females techniques were developed for accelerating and retarding egg development so that hatching could be obtained in all months of the year. Eventually this supply of ovigerous females was supplemented with repeat-spawners maintained in the system. A method for identifying mature, preovigerous females was also developed. This enabled us to purchase barren but preovigerous females from the commercial catch, and obtain new eggs with a minimum investment in care and feeding. It also terminated our dependence on the commercial fishery as a source of ovigerous females.

The smooth operation of a lobster culture facility requires that larvae be available not only every month of the year, but at consistent intervals and in uniform numbers during those months. This requires relatively precise control over egg extrusion and egg development. It also requires flexibility, since ovigerous females inevitably will die or eggs will be lost. The system must be capable of compensating for this without requiring an exceedingly large (and expensive) reserve brood stock.

The ultimate objective of brood stock management is the development of a completely closed system utilizing only cultured stock. In this way, such desirable culture traits as rapid growth, efficient food conversion, social tolerance and disease resistance can be genetically enhanced. Progress is being made in this direction as well, but the first step is the development of a system of brood stock management that will be independent of the commercial fishery as a source of ovigerous females, and which will permit both precision and flexibility in the scheduling of eggs and larvae. The following describes what we feel to be the basic elements in such a system.

BROOD STOCK MANAGEMENT STRATEGIES

Use of Wild Stock

For another few years at least, brood stock for new lobster culture projects will have to come from wild stocks simply because there are no domesticated stocks available. There are two basic strategies involved in the use of wild stock: obtain ovigerous females directly, or obtain preovigerous females and hold them until eggs are extruded. Each of these in turn yields a second pair of options: sell the female once her eggs are

hatched, or retain her for egg production in subsequent cycles. Each of these methods has legal, political and economic ramifications that will differ from one area to another and should therefore be carefully examined before a particular approach is adopted.

Ovigerous Females. In most areas it is extremely difficult to obtain ovigerous females from the commercial fishery. In reality, an ovigerous female taken from the grounds a week or a month after extrusion represents no greater loss to the fishery than a preovigerous female taken from the grounds a week or a month *before* extrusion. But in most places ovigerous females are protected by law, and permits to take them for commercial culture purposes are difficult to obtain. In areas such as the Bay of Fundy, where lobsters are impounded in large tidal enclosures, females captured in the preovigerous state may extrude eggs in the impoundment. These are a potential source of eggs for culture purposes, but we have found an extremely high incidence of disease among such animals and prefer not to have them in our system.

Ovigerous females can be obtained from the fishery with either new ("black") eggs or old ("brown") eggs, depending on locale and season of year. Old eggs can be used for larval production in the summer months. New eggs obtained during summer and fall can be accelerated or retarded with temperature according to the relationships developed by Perkins (1972) so that hatching occurs throughout the winter and following spring. The general technique for this is illustrated in Fig. 1 and described in the section on Techniques and Problems. As might be expected, the 4-month period from October through January is the most difficult in which to sustain production, and December is the month that consistently causes the most problems. This is illustrated in Fig. 2, where larval production over a 3-year period is expressed as a percentage of monthly target values. These failures occurred from a variety of factors (egg loss, disease, nemertean predation, premature molt) which seem to be exacerbated by the prolonged abnormal temperature exposure required to induce hatching during this period.

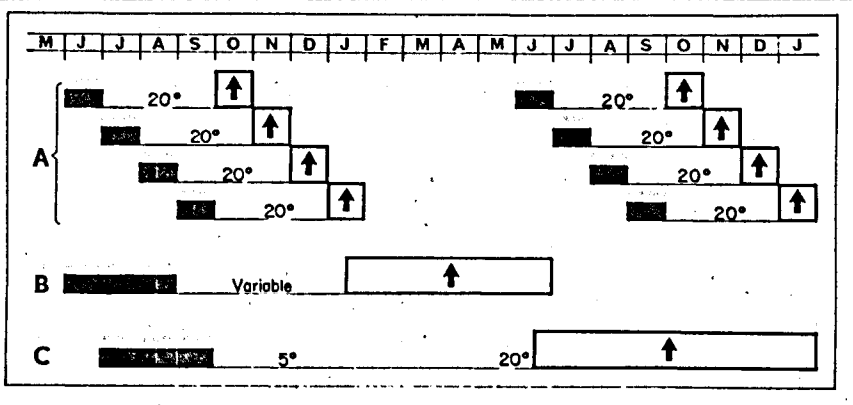

Figure 1. Procedure for producing Homarus larvae throughout the year. Solid blocks represent oviposition, blocks with arrows indicating hatching period. A- months during which larvae can be obtained by holding new eggs at constant 20°C. B- method for obtaining larvae during January through June. Required temperature is selected according to Perkins (1972). C- method for obtaining larvae during June through January. Temperature is raised to 20°C at appropriate time to induce hatching.

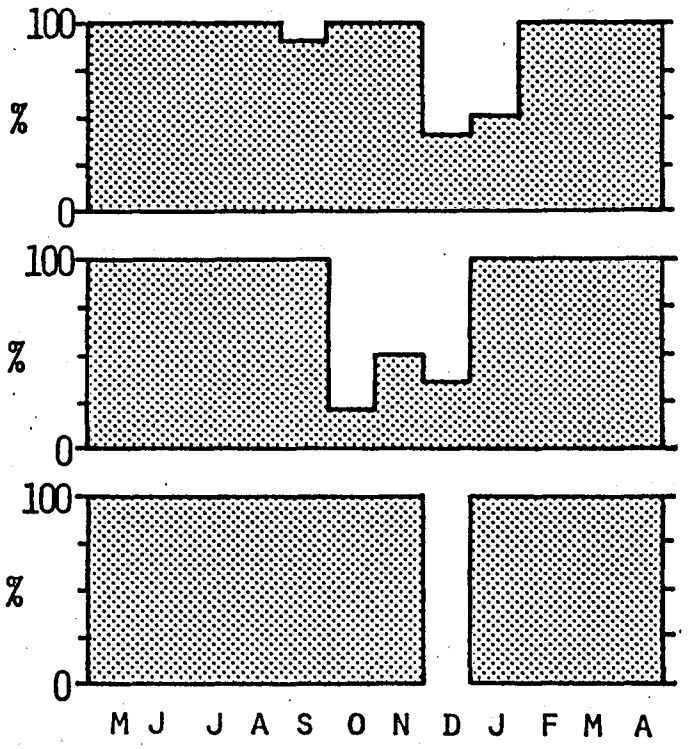

Figure 2. Monthly larval production in a culture facility over three successive years (May 1977-April 1980) expressed as percent of planned production level. Problem months are readily apparent.

Preovigerous Females. Barren mature females can be purchased from the commercial fishery with no legal complications, and held in a culture system until eggs are extruded. This approach has been attempted with mature females captured after mating in the autumn (Hedgecock et al. 1978; Schuur et al. 1976; Van Olst et al. 1980), but these females usually will not extrude eggs until the following summer, and must therefore be fed and maintained for several unproductive months.

An alternative is to purchase females in the spring, before the spawning season, on the assumption that those not carrying eggs are preovigerous and will spawn within a month or two. Females offered for sale in either spring or fall include a significant proportion of immatures and others from which the eggs have disappeared for a variety of reasons. Such females will not spawn for a year or more, and unless they can be identified and eliminated, this approach makes little economic sense.

Fortunately, the abdominal or cement glands are an excellent indicator of ovary condition (Aiken and Waddy 1980a, b). We have defined the relationship between abdominal gland development and time to extrusion (Waddy and Aiken 1980a) and, with this technique, preovigerous females can be selected from the commercial catch in the spring and need only be held about 2 months before eggs are produced. With this method we have obtained extrusion rates of 87% (Fig. 3). It should be noted that females very close to spawning will often resorb their ovary if exposed to the stress of

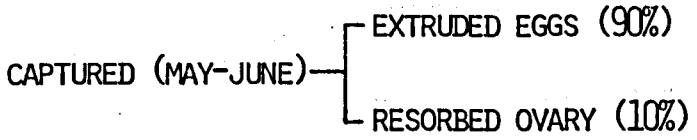

Figure 3. Egg extrusion obtained from 30 preovigerous females selected from commercial landings in May-June 1980. All of those that resorbed were captured within 2 weeks of projected spawning date.

handling and shipping. We have had no problem with females captured and shipped 2 months ahead of predicted extrusion time, whereas up to 30% of those shipped within 2 weeks of spawning have reacted with massive resorption. So the only constraint appears to be the requirement that females be captured and shipped at least a month before oviposition.

Repeat-Spawners. Because of the facilities and care required to maintain brood stock it appears to make more economic sense in the early stages of culture to dispose of females after their brood has hatched, and replace them with fresh stock the following spring. But occasionally a female, or her offspring, will appear particularly well adapted to culture conditions and therefore a likely candidate for continued production. With the present state of knowledge, the best way to accomplish this is to approximate the normal environmental cycle.

Female Adult-I American lobsters from the Gulf of St. Lawrence, and possibly other warm water areas as well, will molt and spawn in the same summer, but most females spawn between July and September of alternate years, with hatching, molting and mating occurring during the intervening summer (Aiken and Waddy 1976, 1980a, b; Waddy and Aiken 1979). The occasional female will extrude eggs in two consecutive summers without an intervening molt, but this is an unusual event that has occurred only three times in the hundreds of egg extrusions recorded at this laboratory.

It is significant that the reproductive cycles reported by Hedgecock et al. (1978) for American lobsters maintained under conditions north of San Francisco, California, differ from those described here. They report evidence for a cycle somewhat shorter than 2 years and a pronounced lack of seasonality in reproductive events, with extrusions occurring from February to December.

They have also found the production of eggs and larvae to be extremely low. Of 135 controlled matings, only 11% extruded eggs. This contrasts with the 90% rate of successful hatching we have obtained from 319 controlled matings in our facility (Fig. 4). The reasons for these differences are not easily determined as there are a number of possibilities. One is temperature: minimum winter temperature along the central California coast

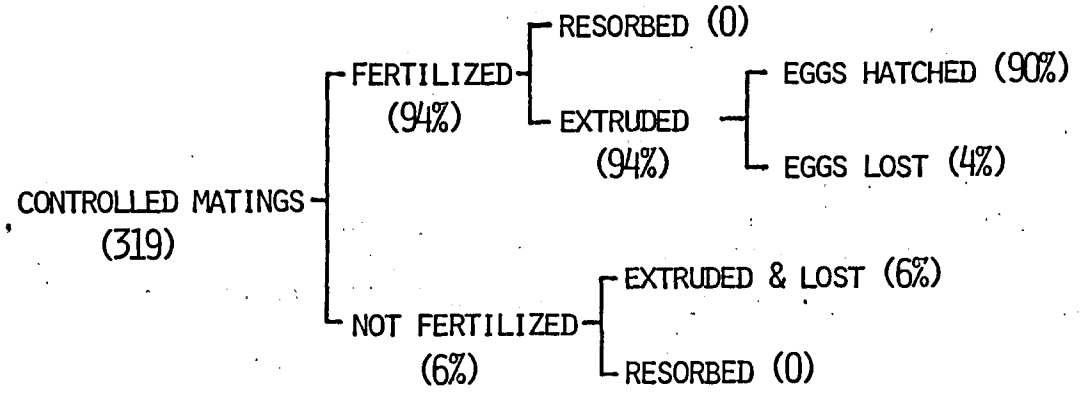

Figure 4. Results from the controlled mating of 319 females in a culture system. Sperm transfer was unsuccessful in 6% of the matings, but oviposition, egg retention and subsequent hatching success were normal when insemination was successful.

is about 8-9°C higher than the 0°C reached in our facility in late winter. In controlled temperature studies with mature females with an established reproductive cycle, we found incidence of extrusion to be reduced by almost 40% when winter water temperature was not allowed to go below 12°C, and reduced about 10% when the minimum temperature was 7°C.

For these reasons, and the fact that little is known about the relationships between temperature and ovary development, we recommend that valuable wild brood stock be returned to a natural photoperiod and temperature regime after hatching, and that winter temperature be reduced to 5°C or less. In our area this period of low temperature lasts for about 4.5 months (mid-December to mid-April). It would be beneficial if the length of the cold period could be reduced somewhat with a slightly accelerated year, but the effects of this on the reproductive cycle have not been determined. Females of moderate size handled in this way will usually molt the following summer. They can be mated at that time and held on the natural regime until the next extrusion.

At this point we would not recommend this as a method for supplying all the larvae required by a facility because the alternate-year reproductive cycle requires that more than twice as many females be maintained as would otherwise be required, and an additional stock of males must also be maintained for mating. At this time the most economically viable method is to purchase healthy preovigerous females directly from the fishery in spring, obtain extrusion and hatching in the facility, and then sell the females. Assuming approximately 10% of the preovigerous females selected from the fishery will fail to spawn successfully (Fig. 3), and approximately 25% of the resulting ovigerous stock will not hatch on schedule (Fig. 5), a culture facility using this system should obtain approximately 35% more than the calculated number of brood females required to sustain the facility, and schedule these for the difficult period between September and February (Fig. 2).

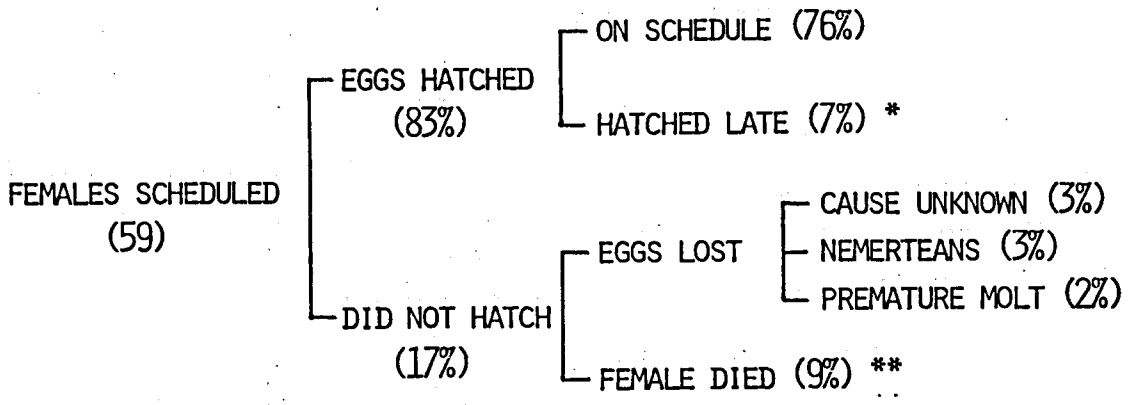

Figure 5. Summary of results from 59 ovigerous females scheduled to hatch larvae over a 12-month period in a culture facility. The data encompass two consecutive years. *Delayed hatch caused by Leucothrix infection. **Three deaths due to equipment malfunction; two attributed to failure of renal function.

Use of Cultured Stock

Wild stock hatched and reared in a culture system is subject to severe selection pressures. Those that survive in the system and grow rapidly to maturity can be considered better adapted to culture conditions than those that either do not survive or survive but do not grow. It seems reasonalbe to expect some of this tendency toward survival and growth under culture conditions to be heritable (Hedgecock et al. 1976; Hedgecock and Nelson 1978), and it is therefore important that methods be developed for obtaining offspring from males and females that have reached maturity in a culture system.

Recognizing Maturity. Females must be mated if their first egg mass is to be fertile, and most successful mating occurs at the molt. Since lobsters of this size are usually held individually, it is important that onset of maturity be recognized so that arrangements can be made for mating.

Body size is not a reliable indicator of maturity since females mature over a range of sizes. However, the "Maturity Index," or ratio of abdominal width to carapace length, is reasonably useful. The female AW:CL ratio diverges from that of the male (0.55-0.56) at carapace lengths of only 40-45 mm. In wild populations approximately 50% of females with an AW:CL ratio of 0.65, and 97% of those with a ratio of 0.67, are mature (Aiken and Waddy 1980a, b; Waddy and Aiken 1980b), and this same relationship exists in cultured females. With this simple determination, maturing females in the culture system can be identified and moved to conditions suitable for mating and final ovary development.

Intermolt mating does occasionally occur (Aiken and Waddy 1980a, b; Dunham and Skinner-Jacobs 1978; Waddy and Aiken 1979), but the stimulus for it is not known, and results have been unpredictable. To ensure successful mating the male and female should be together during or shortly after the female molt. Since molts are difficult to detect in individual containers in a large culture facility, communal holding with mature males should be considered.

Influence of Temperature. The culture temperature in most facilities is maintained at 20-22°C to promote rapid growth. At these temperatures females will mature at the maximum physiological rate, and many will reach full maturity at less than 60 mm carapace length and approximately 18 months of age. Our smallest ovigerous female was only 56 mm carapace length.

Unfortunately, about 90% of those held at constant 20°C resorb the ovary instead of extruding eggs. Other work indicates a period of "winter" temperature is essential for normal ovary development, at least in wild stock. Until the relationships between temperature, photoperiod, ovary development and oviposition are known, it would be a good idea to return potential culture brood stock (both male and female) to a normal environmental cycle after the pubertal molt.

TECHNIQUES AND PROBLEMS

Control of Hatching

Egg development rates vary according to temperature, salinity, age of the eggs, prior temperature history, health of the eggs, and other conditions. Perkins (1972) gives the number of weeks required for embryo development from fertilization to hatching at temperatures between 5 and 25°C, and provides an equation for calculating time to hatching where size of embryonic eye and holding temperature are known. We have found this so-called Perkins Index can be used to predict hatching time within ± 4 days as much as 6 weeks in advance when healthy eggs are exposed to normal temperature cycles. Eggs that are held for extended periods at less than 5°C and then brought up to 20°C will routinely hatch 1 week earlier than predicted from the Perkins Index. Eggs that become infected with microbial epibionts will often hatch considerably later than predicted from the Perkins Index.

The procedure for regulating time of hatch is summarized in Fig. 1. Eggs extruded between June and September are handled in three different ways:

Fig. 1A - eggs maintained in constant 20°C from extrusion to hatch produce larvae from October through January;

Fig. 1B - eggs maintained at an appropriate temperature selected from Perkins' (1972) data to produce larvae from January through June; and

Fig. 1C - eggs held at constant 5°C from oviposition and transferred to 20°C at appropriate time to produce larvae from June through the following January.

Larvae can be obtained from October through January only by maximum acceleration or maximum retardation of embryonic development. Both are extreme conditions that may contribute to the reduced success in producing larvae during these months. Development is most easily retarded in new eggs, as those that are close to hatching will continue to develop slowly even in temperatures below 5°C.

Estimating Fecundity

Continuous culture requires that larvae be produced not only at specific intervals, but in specific quantities as well. Production that exceeds the carrying capacity of the larval system is wasted. Insufficient production results in poor utilization of expensive facilities and an uneven flow of lobsters through the system. Therefore, it is important to control not only the time of hatch, but the size of hatch as well.

Time of hatch, as noted above, can be controlled by temperature, but size of hatch is a function of body size of the female. Because of the logarithmic relationship of size and fecundity, one female American lobster of 160 mm carapace length has the potential to produce as many eggs as ten females of 80 mm carapace length. Since one large female can be housed in a smaller area than ten small females, and will require less food, it might appear sensible to rely on a small number of very large females for larval production. Unfortunately, the loss is also proportionally greater if a large female dies or loses her eggs because of disease, parasitism, or perversity.

Thus it seems more prudent to maintain an assortment of sizes. If a disaster kills off several small females scheduled to produce in subsequent months, it is possible to fill the breach by accelerating a very large female. If the normal capacity of the larval system is temporarily reduced, larval output can be adjusted by accelerating small females and retarding larger ones. The secret to this is a proper blend of temperature manipulation and fecundity.

A variety of values exist for fecundity of homarid lobsters (see Aiken and Waddy 1980a, b for review). We have found that Perkins' (1971) values generally are within 10-20% of actual first stage production of *H. americanus* in our system, and this is sufficient to permit the type of manipulation described here.

Loss of Eggs

Extruded eggs are attached by some ill-defined process to the non-plumose setae of the pleopods and ventral abdominal sterna. This attachment is usually secure, but occasionally an entire egg mass will disappear before hatching. While there are many possible causes of such loss, a few are relatively common and can upset a production schedule unless recognized and controlled.

Unfertilized Eggs. Unfertilized eggs either will not attach or will attach and be lost in succeeding weeks. This can be a problem when preovigerous females are obtained from stocks with a significant proportion of unmated females (see Krouse 1973). This condition can be identified in

preovigerous females by gently probing the seminal receptacle: an inseminated female has a sperm plug that blocks the entrance to the receptacle, an uninseminated female does not. Extruded eggs that are insecurely attached can be examined for the cleavage planes indicative of embryonic development.

Micropredators. In 1978, nemertean worms were found on the egg mass of a wild female in our culture facility (Aiken et al. 1980). This worm, which appears to be new to science, is capable of causing extensive destruction to an egg mass and fosters secondary infections by microbial and fungal organisms. It is capable of remaining in a culture system indefinitely, pervading all quarters, and is easily transferred to previously uninfested females. At this point there is no known treatment that will kill the worms without destroying the eggs as well. This worm is a destructive pest and an important consideration for culturists obtaining wild stock from an area where this nemertean has been found.

Diseases. A number of different diseases are known to affect lobster eggs (for reviews see Sindermann 1977; Fisher et al. 1978). Microbial epibionts such as Leucothrix are particularly troublesome because development of the infection is enhanced at the culture temperatures used to accelerate embryo development, and can transfer to the larva at hatching. We have found that Leucothrix infections can delay embryo development by as long as 6 weeks (Perkins Index), and severe infections can cause total egg mortality. Leucothrix can be controlled with neomycin (Fisher et al. 1978), and we have had success with a 1-3 min dip of the female abdomen and attached eggs in a 5-10 ppt solution of a commercial iodophore (Wescodyne). This treatment, applied once or twice a week at 20°C and less frequently at lower temperatures, restores a normal rate of embryo development and permits hatch with normal larval viability.

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