FINAL REPORT

VIRGINIA FISHERY RESOURCE GRANT PROGRAM

PROJECT FRG-00-05

CONTROL OF MUD BLISTER FORMATION IN OYSTERS

Submitted By:
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Introduction

Oyster aquaculture, both commercial and recreational, continues to elicit interest, despite ongoing disease problems. One strategy for growing oysters in the presence of oyster diseases is to utilize off-bottom technology in order to "out-grow" the pathogens. It's acknowledged that oysters grown off-bottom grow quickly, but tend to have thinner, more brittle shells than those grown on-bottom. Due to the increased expenses of equipment and handling associated with growing oysters off-bottom, most oysters grown commercial in this manner are destined for the more lucrative half-shell market. In order to command higher prices, besides having a good flavor, half-shell oysters need to be visually appealing to the customer.

The shells of oysters provide suitable habitat for a variety of organisms. Some remain on the exterior of the shell; others, like *Polydora sp.* bore into the shell. *Polydora,* commonly known as mud worms, are responsible for the formation of mud-filled shell blisters on the inside wall of the oyster's shell. During the shucking process, these blisters can be broken, releasing mud into the oyster liquor and onto the meats. While not representing a health hazard, the mud detracts from the shucked product's appearance and can impart an off-flavor to the oyster. The presence of numerous *Polydora*-produced mud blisters can potentially reduce the marketability of oysters, especially those intended for the half-shell trade.

During the off-bottom oyster culture process, small seed oysters are confined in floats using appropriately-sized mesh liners or within cages in the floats. As the oysters grow, they are culled, separated, and put either into larger mesh cages or directly within the float with liners. While within the floats, conditions are conducive to attempt to control *Polydora* infestations by different methods. This project evaluated a dipping/drying protocol to combat *Polydora.*

Materials and Methods

Three small "Taylor" oyster floats, measuring 2-feet by 3-feet, each starting with 1,000 seed oysters were used. One float served as a control (Float 2), receiving no *Polydora* control treatments; the other two floats (Float 1 and Float 3) were used as experimental floats and received control treatments as described below. The floats were tethered to a private pier located on Lanes Creek, Mathews County, VA. The seed oysters were obtained from a commercial shellfish hatchery and were all from the same strain of oysters (DEBY's). Floats and oysters were initially deployed on June 16, 2001.

Beginning on June 25, 2001, the experimental floats were subjected to a 15-minute submerged dip in >70 parts per thousand salinity bath on a monthly basis. This salinity was adjusted starting with creek water and adding enough solar evaporated sea salt to raise the salinity to the desired level. Water temperatures varied with seasonal ambient temperatures. Following the dip, the oysters were removed and permitted to air dry for another 15-minutes before being returned to their respective floats and replaced in the natural environment. This dipping protocol was continued until December 2001. Beginning with the December 2001 dipping, only one experimental float (Float 1) was dipped monthly until the conclusion of the project. The final dipping procedure was conducted on June 19, 2002.

At each dipping event, a random grab sample of 25 oysters was taken from each float. Oysters were measured for growth to the nearest 0.1 millimeter, from the hinge to the farthest portion of the shell bill using vernier calipers. Each oyster was then opened and the number of mud blisters on the interior of both shell valves counted. Subjective information was recorded regarding the relative size of the blister. Additionally, the relative age ("new" versus "old") of mud blisters were recorded based upon the amount of shell material covering the blister.

Results

Refer to Appendix 1 for field notes which include dip salinities and temperatures, as well as provide additional observations. Some of these observations will be discussed more fully in the Discussion section.

Appendix 2 contains monthly growth measurements. While there was variability in the monthly measurements, most likely due to sampling error, mean size of oysters in all Floats increase over the project. By November 2001, mean size of oysters in Float 2 actually exceeded minimum market-size of 75 mm, and remained above this size until the end of the project. In Floats 1 and 3 mean size did not exceed minimum market-size until April 2002.

Appendix 3 contains monthly mean number of *Polydora* blisters per oyster sampled. Numbers of mud blisters generally increased as the oysters grew within all Floats. Initially Floats 1 and 3 had fewer blisters that the control Float 2. Following the cessation of dipping of Float 3, the number of blisters in those oysters were similar to the control Float 2. An apparent decrease in blister abundance in April 2002 was the result of a change in how blisters were identified and does not represent a true change in abundance. The dipped Float 1 continued to have fewer blisters than the control Float 2 or Float 3 following the cessation of it being dipped.

Discussion

There is no doubt that the high salinity dip and air drying was effective in killing existing *Polydora*. In subsequent samplings, mud blisters were observed that were considered to be "inactive", with the oyster continuing to cover over the blister with new shell. It was possible to separate "new" infestations from "old" blisters based upon the appearance of the blister. MacKenzie and Shearer (1959. Chemical Control of *Polydora websteri* and Other Annelids Inhabiting Oyster Shells. Proceeding of the National Shellfisheries Association, 50:105-111) investigated different chemicals to control *Polydora* infestations and found that sodium chloride (salt) was the most practical method for killing mud worms. They found that a 10 to 15-minute submersion in a saturated salt solution, followed by 15-minutes of air drying, killed 87 to 98% of *Polydora websteri*. In a related project conducted in North Carolina and funded by their Fishery Resource Grant Program, Kirby-Smith and Hooper reached a similar conclusion, stating "Immersion in saturated salt solution for 15 minutes followed by one hour of air drying significantly reduced the number of blisters...." (Kirby-Smith, William W. and Mark Hooper. 2000. Improving Quality of Farm Raised Oysters: Three Simple Treatments to Control the Boring Sponge [*Cliona sp.*] and Mud Blisters [*Polydora sp.*]. North Carolina Fishery Resource Grant Program, Project 00AM-02).

Unfortunately, the dipping protocol was not effective in "controlling" *Polydora* infestations. Once Floats 1 and 3 were returned to the natural environment and the effects of the salt dip were diluted, new infestations would occur between dip intervals. This is apparent in Appendix 3 where the number of blisters continued to increase in the dipped Floats, while still being fewer than the control Float. With *Polydora* being very active in the area and the close proximity to control Float 2, there was a continual "reservoir" of *Polydora* to continually re-infest the dipped floats. Given that the protocol was effective in killing *Polydora,* it is entirely plausible that the frequency of dipping was not sufficient to "control" subsequent infestations. In an area where *Polydora* is very prevalent, it may be necessary to dip oysters much more frequently in order to truly control infestations. This may be prohibitive when large numbers of oysters are being grown.

An unanticipated result of the dipping protocol was the impact on external appearance of the individual oysters. Besides killing *Polydora,* the dipping protocol also was effective in retarding the growth of other fouling organisms, in particular encrusting sponges, bryozoans, algae and to some extent barnacles. During the first portion of the project, oysters in Floats 1 and 3 were noticeable "cleaner" than those in control Float 2 (refer to field notes in Appendix 1). Once dipping of Float 3 was discontinued, oysters in that float had noticeably more surface fouling than Float 1 and was similar to Float 2 (refer to Appendix 1, beginning in March 2002). This was consistent with the Kirby-Smith and Hooper study, where they found no living boring sponges after saturated salt dips in their project. In areas where *Polydora* is not problematic, dipping may still be beneficial in controlling surface fouling to maintain external shell appearances.

Summary

The use of a salt dip protocol to "control" *Polydora* infestations while initially effective, did not succeed in "eradication" of the problem. Re-infestation following dip events resulted in continued mud blister presence. It may be that in areas with heavy *Polydora* infestations, more frequent dipping will ultimately result in control. However, the time and expense necessary to treat large numbers of oysters may preclude using this technique in large-scale oyster culture.

The dipping protocol and frequency of treatment was very effective in controlling other surface fouling organisms and may be useful in areas without heavy *Polydora* infestations.

APPENDIX 1

Field notes for Fishery Resource Grant Program, Project Number FRG-00-05, "Control of Mud Blister Formation in Oysters".

25 July 2001

1. Oysters went overboard on 16 June 2001. All animals DEBY's supposedly from the same batch, obtained from Middle Peninsula Aquaculture.
2. Oysters in 3 small Taylor floats. Floats numbered 1, 2, and 3, with float 1 being closest to shore and 3 being farthest from shore. Float 2 was designated as the Control float.
3. Dip initiated with 10 gallons of creek water: salinity = 17 ppt; temperature = 82° F. Five pounds of salt (water softener, evaporated sea salts) added and agitated by hand to dissolve, with a final salinity of 70 ppt.
4. Floats 1 and 3 dipped for 15 minutes and air dried for 15 minutes.

23 August 2001

1. River salinity = 19 ppt; temperature = 84° F. 7.5 pounds salt added to 15 gallons of river water for salinity = ~70 ppt.
2. Floats 1 and 3 dipped for 15 minutes and air dried for 15 minutes.
3. Oysters from Floats 1 and 3 exhibited "old" blisters.
4. Oysters from Float 2 had "dirtier" external shell surfaces, algae, sponge, etc.

20 September 2001

1. 15 gallons of river water, 7.5 pounds of salt = salinity of 70 ppt; dip temperature = 23° C
2. Floats 1 and 3 dipped for 15 minutes and air dried for 15 minutes.
3. Control oysters (Float 2) covered with sponge, little or no sponge on oysters in Floats 1 and 3 (although some sponge on float liner).
4. Control oysters (Float 2) very "snappy" (long and slender), more so that other floats.

25 October 2001

1. 20 gallons of water (salinity = 21 ppt; temperature = 66°F), 10 pounds of salt, resultant salinity = 75 ppt, dip water temperature = 67° F.
2. Floats 1 and 3 dipped for 15 minutes, air dried for 15 minutes.
3. Control (Float 2) animals with more surface fouling, primarily sponge, than 1 and 3.
4. Thought: Control Float 2 could be serving as a "reservoir" for *Polydora*, constantly providing for re-infection. Perhaps other oysters in area also acting as reservoir.

29 November 2001

1. 20 gallons of water, 10 pounds of salt, resultant salinity = 76 ppt, dip water temperature = 58° F.

2. Floats 1 and 3 dipped for 15 minutes, air dried for 15 minutes.

28 December 2001

1. Dip water salinity = 71+ ppt, dip water temperature = 35° F.
2. Only Float 1 was dipped, 15 minutes, air dried for 15 minutes. [This is in accordance with the sampling protocol.]

24 January 2002

1. Dip water salinity = 80 ppt, dip water temperature = 46° F.
2. Float 1 dipped for 15 minutes, air dried for 15 minutes.
3. Many *Polydora* blisters have been covered by sufficient shell deposit to not present any "mud" problem when shucked (i.e., thick enough shell layer not to break).
4. Oysters in Float 3 appear to have more "new" blisters.

26 February 2002

1. Dip water salinity = 90 ppt, dip water temperature = 48° F.
2. Float 1 dipped for 15 minutes, air dried for 15 minutes.
3. Very few "new" worms; older worms well covered with shell.

26 March 2002

1. Dip water salinity = 85 ppt; dip water temperature = 52° F.
2. Float 1 dipped for 15 minutes, air dried for 15 minutes.
3. Float 3 oysters covered with "significantly" more green algae than other floats. Float 1 oysters looked the cleanest of all oysters.
4. "Old" blisters are well covered with shell, would not be a shucking problem.
5. Oysters show good growth, with nice bills.

23 April 2002

1. Dip water salinity = 75 ppt, dip water temperature = 56° F.
2. Float 1 dipped for 15 minutes, air dried for 15 minutes.
3. Creek appeared more silty/dirty. All oysters had more silt on their surfaces.
4. There had been a mussel and barnacle set since the last sampling.
5. Float 1 oysters looked real good; interior of shells well developed.
6. A decision was made to consider very old, well-covered blisters as "non-existent". Only blisters that could be broken in the shucking process will be counted.

24 May 2002

1. Dip water salinity = 75 ppt, dip water temperature = 68° F.

2. Float 1 dipped for 15 minutes, air dried for 15 minutes.
3. Float 2 oysters with noticeable encrusting sponges, Float 3 oysters encrusting sponge beginning but not as extensive as Float 2.
4. There has been a significant barnacle set since the last sampling.

19 June 2002

1. Dip water salinity = 70 ppt, dip water temperature = 77° F.
2. Float 1 dipped for 15 minutes, air dried for 15 minutes.
3. Oysters in Floats 2 and 3 heavily fouled with sponge and algae; Float 1 oysters clean.
4. A decision was made to terminate field data collection. After a year of sampling, it was decided that no new information would be obtained and that sufficient data was available to provide a report on the project.

APPENDIX 3

MEAN NUMBER OF *POLYDORA* BLISTERS PER OYSTER

DATE	FLOAT 1	FLOAT 2	FLOAT 3
25 JULY 2001	No measurements	2.3	No measurements
23 AUG 2001	2.1	4.6	3.0
20 SEPT 2001	3.9	4.7	4.2
25 OCT 2001	5.1	6.8	5.3
29 NOV 2001	4.1	6.1	3.8
28 DEC 2001	5.3	5.6	5.0
24 JAN 2002	6.4	7.5	7.0
26 FEB 2002	5.0	7.4	7.6
26 MAR 2002	5.2	7.6	8.0
23 APR 2002	3.0	4.2	4.6
24 MAY 2002	2.4	4.9	5.0
19 JUNE 2002	3.7	5.8	6.6