

1 **An initial assessment of native and invasive tunicates in shellfish aquaculture of the North**
2 **American east coast**

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11
12 **Summary**

13 The objective of the study was to assess the distribution of native and invasive tunicates in the
14 fouling community of shellfish aquaculture gear along the U.S. east coast of the Atlantic. Since
15 the 1980s, several species of invasive tunicates have spread throughout the coastal waters of the
16 North American east coast and have become dominant fouling organisms on docks, boat hulls,
17 mooring lines, and in shellfish aquaculture. Invasive and native tunicates negatively impact
18 shellfish aquaculture through increased maintenance costs and reduced shellfish growth. While
19 the presence of alien tunicates has been well documented at piers, harbors, and marinas, there are
20 few published reports of invasive tunicate impacts to aquaculture. We surveyed shellfish
21 aquaculture operations at Martha's Vineyard, Massachusetts and shellfish aquaculturists in other
22 areas along the North American east coast and report high levels of fouling caused by seven
23 invasive, three native, and two cryptogenic species of tunicates. All study sites were fouled by
24 one or more tunicate species. Biofouling control treatments varied among aquaculture sites and
25 were effective in removing tunicates. Invasive and native tunicates should be considered when
26 assessing the economic impacts of fouling organisms to the aquaculture industry.

27

28

29 **Introduction**

30 Following a series of alien tunicate introductions in the 1980s, the tunicate fauna of New
31 England has become dominated by invasive colonial and solitary tunicates, including *Ascidella*
32 *aspersa* (D.F. Müller 1776), *Botrylloides violaceus* Okra 1927, *Didemnum vexillum* Kott 2002,
33 *Diplosoma listerianum* (Milne-Edwards 1841), *Styela clava* Herdman 1881, *Styela plicata*
34 (Lesueur 1823), and *Botryllus schlosseri* (Pallas 1774). *B. schlosseri* was introduced to New
35 England in the early 1800s; *B. violaceus*, *D. vexillum*, *S. clava* in the 1970s; *D. listerianum* and
36 *A. aspersa* in the 1980s; and *S. plicata* in 2000 (Pederson, 2005; J. Carlton, pers. comm.).
37 Invasive and native tunicates are an economic concern for shellfish aquaculturists because they
38 overgrow bivalves and foul gear, thereby adding weight and restricting water exchange and
39 nutrients (Kluza et al., 2006; Howes et al., 2007; Locke et al., 2007; Rajbanshi and Pederson,
40 2007; Lutz-Collins et al., 2009).

41 Removal and control methods for biofouling on oysters and mussels include exposure to
42 air (Katayama and Ikeda, 1987), plastic wrap (Sinner and Coutts, 2003; Coutts and Sinner,
43 2004), and applications of dilute bleach (Denny, 2008), vinegar (Carver et al., 2003), acetic acid,
44 or calcium hydroxide (Locke et al., 2009). These practices can account for up to 30% of the
45 operational expenses in bivalve farming (Claereboudt et al., 1994), although what proportion of
46 these costs are attributable solely to tunicates is unknown. The objective of this study was to
47 assess the distribution and prevalence of native and invasive tunicates associated with shellfish
48 aquaculture in North American east coast waters and to document tunicate removal and control
49 measures presently being employed in this region.

50

51 **Materials and Methods**

52 During summer 2008, we qualitatively surveyed U.S. east coast shellfish aquaculture operations
53 (n=24) for tunicate prevalence and control measures by written communication (n=15) and by
54 rapid assessment (n=19 around Martha's Vineyard, Massachusetts). A written questionnaire was
55 sent by email to shellfish farmers requesting information on economic losses owing to the
56 fouling of both native and non-native tunicates (sea squirts). Of particular interest were the costs
57 associated with the following: 1) maintenance of fouled gear, 2) costs of antifoulant coatings (if
58 used), 3) growth rate reductions of shellfish due to fouling of gear by tunicates, 4) shellfish
59 mortality that may be attributed to tunicate fouling, and 5) any other costs.

60 Rapid assessment surveys were conducted by small boat and included examination of
61 submerged substrate at each aquaculture site to assess the level of tunicate fouling. The amount,
62 type, and age of substrate (cages, bags, ropes, floats, and cultured shellfish) varied at each site.
63 Tunicate species presence and absence was determined and no distinction was made between
64 juvenile or market size shellfish gear in this initial assessment. All tunicates were identified via
65 collection of specimens or photographs. Recent literature references of tunicate prevalence at
66 shellfish aquaculture sites in Massachusetts, Rhode Island, and Prince Edward Island, Canada,
67 were also included in this assessment.

68

69 **Results**

70 Twelve species of tunicates were found at shellfish aquaculture sites along the North American
71 east coast (Table 1). This list included seven invasive species: *A. aspersa*, *B. violaceus*, *B.*
72 *schlosseri*, *D. vexillum*, *D. listerianum*, *S. clava*, *S. plicata*; two cryptogenic species: *C.*

73 *intestinalis* and *Molgula citrina* (Alder and Hancock 1848); and three native species: *Aplidium*
74 *constellatum* (Verrill 1871), *Didemnum albidum* (Verrill 1871), and *Molgula manhattensis*
75 (Dekay 1843). The most common fouling tunicate was the invasive *B. violaceus*. Invasive and
76 native tunicates were found to be common fouling organisms in shellfish aquaculture,
77 comprising in some cases nearly 100% of the biofouling community (Fig. 1). The amount and
78 type of gear (cages, boxes, bags, ropes, and buoys) and number of bivalves examined varied at
79 surveyed sites. *Aplidium constellatum* and *C. intestinalis* were attached only to aquaculture gear,
80 whereas *A. aspersa*, *B. violaceus*, *B. schlosseri*, *D. albidum*, *D. vexillum*, *D. listerianum*, *M.*
81 *manhattensis*, *S. clava* were attached to gear and shellfish (Table 2). Tunicates fouled cultured
82 bay scallops *Argopecten irradians irradians* (Lamarck 1819), oysters *Crassostrea virginica*
83 (Gmelin 1791), and blue mussels *Mytilus edulis* Linnaeus 1758 at several locations. Quahogs
84 *Mercenaria mercenaria* (Linnaeus 1758) and steamer clams *Mya arenaria* Linnaeus 1758
85 contained in treated boxes and benthic cages were partially within bottom sediments so that the
86 shellfish were infaunal or epifaunal and were thus tunicate free. Up to seven species of tunicates
87 occurred in a single embayment at Martha's Vineyard (Fig. 2). Salinity at locations containing
88 tunicates ranged from 28 to 33.5 (Table 2).

89 Tunicates were absent on treated shellfish or aquaculture gear. Observed strategies for
90 fouling control on aquaculture gear include peroxide and anti-fouling paint applied to boxes,
91 freshwater sprays with a garden hose for five minutes, and air-drying on land for three days.
92 Treatments applied directly to oysters included five-minute freshwater sprays, exposure to air for
93 24 hours, tumbling for ten minutes, and salt brine dips for 10 minutes followed by air exposure
94 for two hours. Floating bags of oysters were flipped over every two weeks during spring,
95 summer, and fall. All treatments were effective in removing attached tunicates. Bay scallops

96 cannot tolerate these treatments and were routinely removed from fouled bags and placed in
97 clean bags every two to three months during spring, summer, and fall.

98

99 **Discussion**

100 This is the first descriptive assessment of invasive tunicate fouling and removal techniques in
101 shellfish aquaculture along the U.S. east coast. It is likely that other species of tunicates are
102 present at aquaculture sites in New Jersey, New York, North Carolina and Virginia (Table 1).
103 Our preliminary results suggest that site visits in these states would be warranted and will likely
104 add to the knowledge of the geographic distribution of tunicate species negatively impacting
105 shellfish aquaculture operations. Because the materials used at the farms are of different types
106 and age, studies where panels are exposed for a known exposure time to explore tunicate settling
107 behavior and panels of the identical material used at farms but of different ages (first time use,
108 second time use, or longer term use) to see whether settling characteristics are changing are
109 warranted.

110 Tunicates likely foul cultured shellfish and aquaculture gear because of the available hard
111 substrate they provide in the water column. It appears that the conditions suitable for shellfish
112 aquaculture are also highly conducive to tunicate growth. Antifouling procedures will likely be
113 different for each shellfish species and age of the shellfish. Tunicate removal requires additional
114 labor by aquaculturists, although cost estimates for this maintenance have not been calculated.
115 Future research is needed to determine the most efficient procedures for preventing and
116 removing fouling tunicates. The results of such studies could lead to recommendations that
117 would help environmental agencies and farm managers to optimize mitigation strategies when
118 trying to cope with tunicate fouling problems.

119

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129

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175 Fig. 1. Cage fouling by *Molgula manhattensis* in Virginia (photo credit, Tom Leggett)

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177 Fig. 2. Geographical distribution of tunicate species on Martha's Vineyard, Massachusetts, 2008

178

179 Table 1. Geographical distribution of tunicates at shellfish aquaculture sites along the North

180 American east coast, 2005-2008. Abbreviations: *Ac*=*Aplidium constellatum*; *Aa*=*Asciidiella*

181 *aspersa*; *Bv*=*Botrylloides violaceus*; *Bs*=*Botryllus schlosseri*; *Ci*=*Ciona intestinalis*;

182 *Da*=*Didemnum albidum*; *Dv*=*Didemnum vexillum*; *Dl*=*Diplosoma listerianum*; *Mc*=*Molgula*

183 *citrina*; *Mm*=*Molgula manhattensis*; *Msp*=*Molgula* sp.; *Sc*=*Styela clava*; *Sp*=*Styela plicata*.

184 The tunicate species at Maine (J. Dijkstra, pers. comm.), other sites in Massachusetts (Valentine

185 et al., 2007), Rhode Island (Carman et al., 2009), and PEI, Canada (A. Locke, pers. comm.) were

186 identified from live specimens and tunicate species at Maryland, New Jersey, New York, North

187 Carolina, and Virginia were identified from photographs.

188

189 Table 2. Shellfish aquaculture sites visited, 2008. Approximately one hour was spent looking

190 for tunicates at each of the 26 sites. Abbreviations (shellfish): *Aii*=*Argopecten irradians*

191 *irradians*; *Cv*=*Crassostrea virginica*; *Ma*=*Mya arenaria*; *Meme*=*Mercenaria mercenaria*.

192 Abbreviations (tunicates): *Ac*=*Aplidium constellatum*; *Aa*=*Asciidiella aspersa*; *Bv*=*Botrylloides*

193 *violaceus*; *Bs*=*Botryllus schlosseri*; *Ci*=*Ciona intestinalis*; *Da*=*Didemnum albidum*;

194 *Dv*=*Didemnum vexillum*; *Dl*=*Diplosoma listerianum*; *Mm*=*Molgula manhattensis*; *Sc*=*Styela*

195 *clava*.

196



Figure 1

197
198
199

200

	<i>Ac</i>	<i>Aa</i>	<i>Bv</i>	<i>Bs</i>	<i>Ci</i>	<i>Da</i>	<i>Dv</i>	<i>DI</i>	<i>Mc</i>	<i>Mm</i>	<i>Msp</i>	<i>Sc</i>	<i>Sp</i>
Maine			X				X	X	X				
Maryland										X			
Massachusetts	X	X	X	X		X	X			X		X	
New Jersey										X			
New York			X	X							X	X	
North Carolina					X						X		X
Rhode Island			X		X		X				X	X	
Virginia										X			
PEI, Canada			X	X	X				X	X		X	

201

202 Table 1

Site #	SITE VISITED	DATE	TUNICATES	TUNICATE SUBSTRATE	SALINITY
1	Sengekontacket Pond	24-Jul-08	<i>Bs,Bv,Mm</i> <i>Mm</i> none	gear cultured <i>Aii</i> treated gear,cultured <i>Meme</i>	31
2	Sengekontacket Pond	24-Jul-08	none	treated dock	
3	Lagoon Pond	29-Jul-08	<i>Aa,Bv,Bs,Dv,Sc</i> <i>Aa,Bv,Bs,Dv,Sc</i>	cultured <i>Aii</i> dock,gear	31
4	Katama Bay	30-Jul-08	<i>Bs,Bv,Ci,Dv,Sc</i> <i>Dv,Sc</i>	gear cultured <i>Aii</i>	33.5
5	Katama Bay	30-Jul-08	none	gear,cultured <i>Aii</i>	31
6	Caleb's Pond	30-Jul-08	<i>Bv,Dv</i>	gear	29.5
7	Katama Bay	30-Jul-08	<i>Dv,Sc</i>	dock	30
8	Katama Bay	30-Jul-08	<i>Bv,Dv,Sc</i> none	gear wild <i>Meme</i>	30
9	Pocha Pond	30-Jul-08	<i>Aa,Sc</i> none	gear cultured <i>Aii</i>	28.5
10	Pocha Pond	30-Jul-08	<i>Aa,Bv,Da,Sc</i> <i>Da</i>	gear cultured <i>Aii</i>	28.5
11	Pocha Pond	30-Jul-08	<i>Ci,Mm,Sc</i> none	gear wild <i>Aii</i>	28
12	Edgartown Great Pond	30-Jul-08	none none	gear,cultured <i>Cv</i> wild <i>Ma</i>	14.5
13	Edgartown Great Pond	30-Jul-08	none	gear	14
14	Menemsha Pond	14-Aug-08	<i>Aa,Bv,Bs,DI,Dv,Mm,Sc</i> none	gear gear	33.5
15	Menemsha Pond	14-Aug-08	none <i>Bv,Bs,Dv,DI,Mm</i>	gear,cultured <i>Cv</i> gear	31
16	Menemsha Pond	14-Aug-08	<i>Aa,Bv,Bs,Dv,DI</i> <i>Aa,Bv,Dv</i>	gear cultured <i>Aii</i>	31
17	Menemsha Pond	14-Aug-08	none	crab trap	31
18	Lagoon Pond	14-Aug-08	none	wild <i>Aii</i>	31
19	Major's Cove	28-Aug-08	<i>Bv,Bs,Mm</i> none	eelgrass wild <i>Aii</i>	29.5
20	Major's Cove	2-Sep-08	<i>Bv,Bs</i> <i>Bv</i>	eelgrass wild <i>Aii</i> on eelgrass	30
21	Oak Bluffs Harbor	2-Sep-08	none <i>Aa,Bv,Bs,Dv,DI,Sc</i>	treated gear,cultured <i>Ma</i> dock	33
22	Lagoon Pond	5-Sep-08	<i>Aa,Ac,Bv,Bs,Dv,Sc</i>	gear	31
23	Lake Tashmoo	5-Sep-08	<i>Bs,Bv,DI,Mm</i> <i>Aa,Bv,Bs,DI,Sc,Dv</i>	gear gear	33
24	Lake Tashmoo	5-Sep-08	none	wild <i>Meme</i>	31
25	Lake Tashmoo	5-Sep-08	<i>Aa,Bv,Bs,Dv,DI,Sc</i>	gear	30
26	Lake Tashmoo	5-Sep-08	<i>Aa,Bs,Bv,DI,Dv,Sc</i>	gear	30

Table 2

