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Woods Hole Oceanographic Institution April 21-22, 2005

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INTERNATIONAL INVASIVE SEA SQUIRT CONFERENCE

April 21-22, 2005 Woods Hole Oceanographic Institution Woods Hole, Massachusetts

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INTERNATIONAL INVASIVE SEA SQUIRT CONFERENCE April 21-22, 2005 Program Schedule

Dov 1	Program Schedule
0800-0900	Continental breakfast and registration
	<u>Dann Blackwood</u> Didemnum sp. – Coming to a Habitat Near You Underwater video in coastal and offshore habitats of New England
0900-0910	Laurence Madin Welcome and introductory remarks
0910-0940	Keynote speaker: <u>Gretchen Lambert</u> Invasive ascidians: a growing global problem
Invasive As	reidian Biology and Discussion and Discussion
0940-1000	<u>Gittenberger, Adriaan</u>
	Recent population expansions of non-native ascidians in the Netherlands
1000-1020	<u>Ruiz, Gregory M.</u> , Larson, K., Huber, T., Steves, B., Hines, A., and Lambert, G. Spatial analysis of assidion according to the Newtonian Structure of the second structure of
	of standardized field measures across bays, coasts and latitudes
1020-1040	Coffee break
1040-1100	<u>Fofonoff, Paul W.</u> , Ruiz, G.M., Lambert, G., and Carlton, J.T. Nonindigenous tunicates in North American continental waters: An overview of invasion patterns by time
1100-1120	Bourque, D., Davidson, J., Arsenault, G., LeBlanc, A.R., <u>Landry, T.</u> , and Miron, G.
	Gonadal development, larvae abundance and recruitment of an invasive ascidian <i>Styela clava</i> Herdman in Prince Edward Island, Canada
1120-1140	<u>Petersen, Jens K.</u> Feeding of ascidians: a comparison with commercial benthic suspension feeders
1140-1200	<u>Rocha, Rosana M.</u> and Kremer, L.P. Introduced ascidians in Paranaguá Bay, southern Brazil
1200 1200 1	

1200-1300 Lunch

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Invasive Ascidian Ecology - Session chair: Stefan Sievert

1300-1320	Dijkstra, J.A., <u>Harris, Larry G</u> ., and Westerman, E.L.
	Distribution and ecology of four colonial ascidians: Botryllus schlosseri,
	Botrylloides violaceus, Diplosoma listerianum and Didemnum sp. in the
	Gulf of Maine

 1320-1340.1 Locke, Andrea, Hanson, J.M., Thompson, J., and Ellis, K.M.
 Did green crabs facilitate the clubbed tunicate (Styela clava) invasion of Prince Edward Island?

1340-1400.1 <u>Rodriguez, Laura F.</u> Novel habitats created by non-indigenous species: The role of oysters and ascidians as biological substrata for fouling communities

- 1400-1420Bullard, Stephan G., Hamilton, J.F., and Whitlatch, R.B.Diurnal, lunar and tidal patterns of settlement in tunicate larvae
- 1420-1440 <u>Howes, Stephanie</u>, Herbinger, C.M., Darnell, P., and Vercaemer, B.
 Recruitment, settlement and mitigation of the tunicate *Ciona intestinalis* on a mussel farm in Nova Scotia, Canada
- 1440-1500 <u>Bourque, Daniel</u>, LeBlanc, A.R., Landry, T., Miron, G., and MacNair, N.G.
 Diel larval concentrations of an invasive ascidian *Styela clava* Herdman in Prince Edward Island, Canada

1500-1520 Coffee break

Poster session (complete listing at end of program schedule)

1800-1930 Banquet (additional fee)

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0800-0900	Continental breakfast
0900-0920	Keynote speaker: <u>James Carlton</u> Setting ascidian invasions on the global stage
Invasive As	cidian Ecology (cont.) – Session chair: Stephan Bullard
0920-0940	Osman, Richard W. and Whitlatch, R.B.
	Habitat variability in the invasion of Long Island Sound by <i>Didemnum</i> sp. and its interaction with the resident community
0940-1000	<u>Blum, Julia C.</u> , Liljesthröm, M., Schenk, M.E., Steinberg, M.K., Chang, A., and Ruiz, G.
	Does the non-native solitary ascidian <i>Ciona intestinalis</i> depress species
	richness?

1000-1020	<u>Altman, Safra</u> and Whitlatch, R.B. Space invaders: the effect of small-scale disturbance on invasion success in marine communities
1020-1040	Coffee break
1040-1100	<u>Byrnes, Jarrett E.</u> and Stachowicz, J.J. Diversity in fouling communities and resistance to invasive ascidians: the role of settlement complementarity and species interactions
1100-1120	<u>Agius, Brad P.</u> Facilitating invasions: The effects of temperature on the role of non- indigenous ascidians in fouling communities
1120-1140	Darbyson, Emily A., Hanson, J.M., Locke, A., and Willison, J.H.M. Assessment of boaters as a vector for the clubbed tunicate (Styela clava) in the southern Gulf of St. Lawrence
1140-1230	Lunch
Invasive Asc 1230-1250	cidian Impacts, Risks, and Management: Session chair – Robert Reid <u>Davidson, Jeff</u> and Arsenault, G. Risk factors and spread of Styela clava in Prince Edward Island, Canada
1250-1310	<u>Valentine, Page C.</u> , Carman, M.R., Blackwood, D.S., and Heffron, E.J. Ecological observations of the colonial tunicate <i>Didemnum</i> sp. in a New England tide pool habitat and strategies for managing invasive colonial ascidian species
1310-1340	<u>Coutts, Ashley D.M.</u> Dumb barge teaches important lessons: the arrival, spread and management of an undesirable sea squirt in the Marlborough Sounds, New Zealand
1340-1400	Coffee break
1400-1530	Expert Panel: Moderator – <u>James Carlton</u> Thomas Noji, Claire Carver, Gretchen Lambert, and Judith Pederson
	Mary Carman Closing remarks

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POSTERS: <u>Abdul, Jaffar Ali H.</u> and Sivakumar, V. Occurrence and distribution of ascidians in Vizhinjam Bay (southwest coast of India)

<u>Auker, Linda A.</u> An invasive tunicate, *Didemnum* sp. in Narragansett Bay, Rhode Island

Carman, Mary R. Benthic foraminifera on the invasive ascidian, *Didemnum* sp.

Carman, M.R., <u>Bullard, Stephan G</u>., Donnelly, J.P. Water quality, nitrogen pollution, and ascidian diversity in northwestern Atlantic coastal waters of Massachusetts, USA

<u>Davis, Marla D.</u> and Swalla, B.J. **Invasive tunicates in the Puget Sound, Washington, USA**

<u>Davis, Martin H.</u> and Davis, M.E. The distribution of *Styela clava* (*Tunicata, Ascidiacea*) in European waters

<u>Dijkstra</u>, Jennifer A., Sherman, H., and Harris, L.G. **Predicting the future of community development: a comparative approach**

Dupont, Lise, Viard, F., Bishop, J.D.D. The genetics of establishment and spread of three non-native ascidians in NW Europe

Grey, Erin K.

The ecology of two non-native ascidians in fouling communities along the Strait of Juan de Fuca

McCarthy, Anna

Effects of temperature on growth rates of colonial ascidians: A comparison of Didemnum sp. to Botryllus schlosseri and Botrylloides violaceus

<u>Nielsen, Signe E.</u>, Hawkins, S.J., Bishop, J.D.D. **Positive effects of non-native ascidians in Plymouth, England?**

<u>Nielsen, Signe E.</u>, Hawkins, S.J., Bishop, J.D.D. Localised diversity increases in the presence of a non-native species *Styela clava*

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Effect of acetic acid treatment on the epifaunal mussel sock species *Mitrella lunata* (lunar dove shells) and *Caprella* spp. (caprellid shrimp) in Prince Edward Island, Canada

<u>Pederson, Judith</u>, Bullock, R., Calder, D., Carlton, J., Chapman, J., Cohen, A., Dean, H., Drynda, P., Fisher, R., Harris, L., Hobbs, N., Lambert, C., Lambert, G., Laso Wasem, E., Mathieson, A., Miglietta, M., Smith, J., Tyler, S., and Winston, J. A preponderance of tunicates: observations from rapid assessment surveys

<u>Rajbanshi, Rubi</u> and Pederson, J. Competition among invading ascidians and a native mussel

<u>Simoncini, M.K.</u> and Miller, R.J. Botrylloides violaceus is less vulnerable to sea urchin predation than a dominant native ascidian, Aplidium glabrum

<u>Tait, E.</u>, Carman, M.R., and Sievert, S.M. A molecular approach to exploring bacterial associations with ascidian species of Woods Hole, Massachusetts

<u>Turon, Xavier</u> and Nishikawa, T. Spread of *Microcosmus squamiger* in the Mediterranean Sea

<u>Tyrrell, Megan C.</u> and Byers, J.E.

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Do invasive fouling organisms have greater recruitment and abundance on artificial substrates than on naturally occurring substrates?

<u>Valentine, Page C.</u>, Collie, J.S., Reid, R.N., Asch, R., and Guida, V.G. The invasive tunicate *Didemnum* sp. on Georges Bank – ecological observations and potential effects on groundfish and scallop fisheries

<u>Whitlatch, Robert B.</u> and Osman, R.W. Post-settlement predation on ascidian recruits: Predator responses to changing prey density

Invasive ascidians: a growing global problem.

Lambert, Gretchen. University of Washington Friday Harbor Laboratories, Friday Harbor, WA

What is a tunicate and why are they a problem? These primitive members of our own phylum (the chordates), all marine, are efficient filter feeders of tiny suspended matter. Large numbers of them are an indication of lots of available food in the water. The presence of nonindigenous ascidians (sedentary tunicates), often in huge numbers, has been recognized in many locations for over a century but usually confined to artificial substrates such as marina floats, pilings and boat hulls. A major increase in aquaculture facilities in the past 10-20 years has provided new surfaces (oyster and mussel shell, ropes, nets, etc.) for colonization by sedentary filter feeders. the most successful of which seem to be ascidians, resulting in overgrowth and smothering of the shellfish. Man-made substrates are still often the first sites of settlement for new arrivals, due to the available frequently cleaned or newly submerged surfaces and limited number of species in the fouling community. However, substantial areas of subtidal natural hard surfaces are now being colonized by invasive ascidians that grow quickly, overgrowing almost everything sessile in the process. Certain didemnid species are emerging worldwide as especially successful invaders with broad environmental tolerances and subject to little or no predation. It was formerly thought that the diversity and complexity of natural benthic communities would repel the establishment, overgrowth and ultimate dominance of nonindigenous species. We still do not know how resistant undisturbed, healthy, complex benthic communities are to invasion. If a given nonindigenous species becomes abundant, is this the result of a single successful invasion or multiple occurrences? Genetic analysis is necessary to determine the answer. Where did the invaders originate? Again, usually only genetics will give us the definitive answer.

Causative factors for successful invasions are not well understood, though several have been implicated; well publicized vectors are ballast water transport and dumping, and transport via hull fouling. A gradual warming of temperate regions (where many of the invaders have successfully invaded) may be due to global changes or el niño conditions; the latter was implicated in the successful establishment of new invaders in southern California in the 1990's. Eutrophication of the water as a result of shoreline development leading to increased sediment runoff and sewage outfall can produce extra food for suspension feeders either directly, or indirectly as a result of large plankton blooms; bacterial increases were implicated in the rapid increase of *Trididemnum solidum* in the Caribbean. Disturbance of natural communities (possibly due to dredging, overfishing, or environmental stresses) that kill off some of the species and leave the community vulnerable to invasion may be another cause. What can be done to curb invasions or control populations already invaded? A number of approaches are necessary. Careful frequent monitoring and rapid response is key to control; educational outreach to both the public and the scientific community will be important. A knowledge of the environmental tolerances and possible predators will help determine successful responses, though it is well known that many ascidians can quickly acclimate to changed environmental conditions. Even a minor predator in the natural environment might be used successfully in an aquaculture system; e.g. certain species of sea urchins, gastropods or crabs could be placed in mussel or oyster cages to keep the cages and shells free of foulers. Our current knowledge of the basic biology of invaders, genetic fingerprinting to determine points of origin, and methods of control is still minimal. Increased governmental funding is crucial to winning the war against invaders.

Recent population expansions of non-native ascidians in the Netherlands. <u>Gittenberger</u>, Adriaan. National Museum of Natural History, Naturalis, P.O. Box 9517, 2300 RA Leiden. The Netherlands

Historically the Dutch coast has been an estuary consisting of mud and sand, with shells as the only hard substratum present. Still, almost the entire north-south running coast is one large sandy beach. Natural rocky coasts as in France, the United Kingdom, and Norway are missing. In the Netherlands, hard substrata are found mainly in the southwestern part of the country, bordering the waters of the Grevelingen, Oosterschelde and Westerschelde. These are about 500 km² large each and have artificially strengthened shores with rocks and dikes. The Westerschelde is not separated from the North Sea in any way, but the Oosterschelde is partly closed by a dike with large, usually open, corridors to the sea. The Grevelingen is isolated from the sea by a dike with only two, small, usually closed corridors. Therefore, the water conditions vary strongly in e.g. strength of tidal currents, ranges of temperatures, salinity gradients and murkiness. As a consequence, the species diversities and abundances are distinctly different in these waters. Over the past 20 years, SCUBA diving became very popular in the Netherlands and is practiced especially in the Oosterschelde and Grevelingen. Since 1994, the MOO-project of the ANEMOON-foundation has provided a few hundred volunteers with marine monitoring-forms on which they record all animal species that are observed during a dive. This has resulted now in about 10,000 completed forms and, as a consequence a far better knowledge of the species present, their habitat preferences and their population dynamics over the years and seasons. Several non-native ascidians have settled in the Netherlands since 1994, e.g. Botrylloides violaceus and Perophora japonica, but one species stood out because of its potential as an invasive ascidian with a devastating effect on the marine environment, i.e. Didemnum sp. This white didemnid was first recorded in the Netherlands in 1991. It remained rare until 1998, after which it dramatically expanded its population size and overgrew almost all hard substrata present, including organisms like algae, plants, bivalves, hydroids, sponges, sea anemones and other ascidians. Obviously mussel and ovster-farms are also bothered. Its impact over the years and seasons was additionally documented by photographs in the Grevelingen and Oosterschelde, shedding some light on its habitat preferences and survival abilities. Although most colonies die off during the winter when the water gets too cold, i.e. about 6 degrees Celsius, some seem to remain healthy and survive, expanding again when the water gets warmer in spring. The latter are usually found on living animals like hydroids and solitary ascidians, e.g. Ascidiella aspersa, Ciona intestinalis and Styela clava. It is hypothesized that Didemnum sp. could not expand its populations earlier than 1998 because of the minimum water temperature of -2degrees Celsius in 1996 and 1997. After 1997, for eight years now, the minimum temperature has remained about 4 degrees Celsius, which might be just warm enough for the didemnid to survive. The fact that cold winters, as in 1996 and 1997, were common before 1996-1997, but have not occurred since then, may have enabled the expansion of this didemnid and, more in general, may count for the unusual high number of non-native ascidian, molluscan, crustacean and fish species that have settled and expanded their populations in the Netherlands over the last eight years.

Spatial analysis of ascidian assemblages in North America: comparison of standardized field measures across bays, coasts and latitudes.

<u>Ruiz, Gregory M.</u>¹, Larson, K.¹, Huber T.¹, Steves B.¹, Hines A.¹ and Lambert G.². ¹Smithsonian Environmental Research Center, Edgewater, MD; ²University of Washington Friday Harbor Laboratories, Friday Harbor, WA

To examine spatial patterns for marine invasions, we conducted standardized surveys of fouling communities among 23 different bays in North America. In each bay, we measured species composition of the fouling community (sessile invertebrates) using fouling panels, which were deployed at 8-10 different sites per bay and retrieved after 3 months for analysis. For each panel (n>10 per site and >100 per bay), we collected sessile and mobile invertebrates for identification to species, and each species was classified by bay as native, non-native, or cryptogenic status. Our initial analyses show that tunicates dominate the fouling communities (percent cover) for many bays but also that strong geographic differences exist in percent cover, species richness, and species evenness among bays, coasts, and latitudes. Non-native tunicates contribute strongly to the observed geographic patterns, suggesting many changes in the structure of fouling communities over time.

In addition to examining such broad-scale diversity patterns, we detected several nonnative ascidian species that have yet to be reported for our survey sites. Two new introductions were of particular interest because they are the first to be found in U.S. coastal waters. *Perophora japonica*, originally from Japan, was found on panels deployed in Humboldt Bay, CA and *Didemnum perlucidum*, originally from the Indo-Pacific region, was found in Pensacola Bay, FL. *D. perlucidum* has previously been reported on off-shore oil rigs in Texas, but our survey was the first to see it in the coastal area. We found several range extensions along western North America for the non-native *Ascidia zara*, *Polyandrocarpa zorritensis*, *Styela plicata*, and *Didemnum* sp., which had previously been reported from Los Angeles area and occurred on our panels in San Francisco Bay. We also found a previously undescribed species of ascidian, *Distaplia alaskensis*, which occurred commonly at two sites in Alaska.

Overall, our field surveys are intended to provide spatially explicit and standardized baseline measures of community composition across sites, allowing us to test for changes over time and associated with particular invasion events and dynamics.

Nonindigenous tunicates in North American continental waters: An overview of invasion patterns by time.

<u>Fofonoff, Paul W.</u>¹, Ruiz, G.M.¹, Lambert, G.² and Carlton, J.T.^{3.1}Smithsonian Environmental Research Center, Edgewater MD.;² University of Washington Friday Harbor Laboratories, Friday Harbor, WA.; The Maritime Studies Program of Williams College and Mystic Seaport Mystic, CT

At least 21 species of tunicates have been introduced to continental North American coastal waters. Many additional species are cryptogenic and cannot yet be confirmed as introduced or native. Introduced tunicates are more numerous on the Pacific coast (17 species) than on the Atlantic coast (11 species) or the shores of the Gulf of Mexico (7 species). Overall, the Indo-West Pacific region appears to be the most frequent probable region of origin for tunicates invading US coastal waters (11 of 21 species), but many invaders are of unknown origins. At least 14 of the species have invaded the coastlines of more than one continent or island; many are successful worldwide invaders. A number of species which we consider nonindigenous were detected in the earliest collections of tunicates in the 19th century on the Atlantic and Gulf Coasts (and may have been invaders of the 1500s or 1600s)- their status as invaders is based on biogeographical patterns, and their preference for human-disturbed environments and artificial structures. However, since 1970, a new invading tunicate species has been reported every 5 years in Atlantic and/or Gulf waters, and every 3 years on the Pacific Coast. Our data are derived from a database maintained by the SERC Invasions Group on coastal biological invasions (National Exotic Marine and Estuarine Species Information System- NEMESIS) which incorporates extensive literature searches, searches of museum collections, interviews with taxonomic experts, and results of our ongoing fouling plate program.

Gonadal development, larval abundance and recruitment of an invasive ascidian *Styela clava* Herdman in Prince Edward Island, Canada.

Bourque, D.¹, Davidson, J.², Arsenault, G.², LeBlanc, A.R.¹, <u>Landry, T.¹</u>, Miron, G.³ and MacNair, N.G.⁴.¹Department of Fisheries and Oceans, Oceans and Science Branch, Gulf Fisheries Center, Moncton, NB, Canada, E1C 9B6; ²Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, PEI, Canada, C1A 4P3;

³Département de Biologie, Université de Moncton, Moncton, NB, Canada, E1A 3E9; ⁴ PEI Department of Agriculture, Fisheries and Aquaculture, Charlottetown, PEI, Canada, C1A 7N8

In the past decade, many aquatic invasive species have been introduced into Prince Edward Island coastal waters. They have had adverse effects on the aquaculture and fisheries industries. For instance, the mussel aquaculture industry has been overwhelmed by extremely high abundance of the invasive ascidian Styela clava in some areas. In order to understand and minimize the risk of spread of S. clava to other shellfish aquaculture sites, some basic information on the reproductive biology is needed. This study investigated gonadal development, larval abundance and recruitment in relation to time. The study was conducted in 2003 in an area of high S. clava abundance. Gonadal development was evaluated through histological examination of samples collected over the study period. Both ovaries and testes were found to be morphologically ripe from mid-July through early December. Water samples were collected weekly to evaluate larval concentrations in the water column. Larvae were present from 24 June to 29 October. A distinct peak in larval abundance was noted on 19 August. Instantaneous recruitment was evaluated by setting preconditioned collector plates in the water column every week during the study period. Recruitment on collector plates occurred from 24 June to 21 October. Results are discussed in relation to the environmental parameters measured. Overall, the reproductive phase of S. clava in PEI seems to extend from late June through late October.

Feeding of ascidians: a comparison with commercial benthic suspension feeders. <u>Petersen, Jens Kjerulf.</u> Department of Marine Ecology, National Environmental Research Institute, DK-4000 Roskilde, Denmark

Invasive and native fouling species of ascidians are suspension-feeders like key species in marine aquaculture. They thus compete for food with mussels, oysters and clams. Ascidians feed by pumping water through the gill sac. On the inside of the gill sac a mucus sheet is continuously produced and suspended particles in the water current are captured on the mucus sheet. This is different from the way bivalves feed on the same food source. In this presentation, a review of our current knowledge on ascidian suspension feeding is given in relation to rate and particles capture and how this may impact competition with bivalves for the same food resource. Since fouling ascidians are epibenthic, they may have a competitive advantage compared to the aquaculture bivalves they foul. The ecological impact of this competition in relation to feeding of both groups is discussed.

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Introduced ascidians in Paranaguá Bay, southern Brazil.

Rocha, Rosana M. and Kremer, Laura P. Universidade Federal do Paraná, Departamento de Zoologia, CP 19020, 81.531-980, Curitiba, PR, Brazil

The port in Paranaguá Bay has the greatest volume of exports and imports in southern Brazil. The port, in a large estuarine bay, is a very important potential source for the introduction of non-native species due to the heavy ship traffic. In spite of that potential, only phytoplankton has recently been monitored. Besides the relatively well-known fish community, little is known of the bay's indigenous biodiversity. The goal of this study was to find and examine introduced species of ascidians in Paranaguá Bay using taxonomic and molecular techniques. Ascidians are very useful bioindicators of invasion due to the short larval stage. Samples were obtained at depths of 2-5 meters in four locations within the bay: Ilha das Cobras (Island of Snakes), Keys owned by the Tenenge Company, Ilha do Mel (Honey Island) and Ilha da Galheta (Galheta Island). Thirteen species were identified: Ascidia curvata, A. sydneiensis, Botryllus planus, B. tuberatus, Clavelina oblonga, Cystodytes dellechiajei, Didemnum granulatum, Diplosoma listerianum, Distaplia bermudensis, Eudistoma carolinense, Lissoclinum fragile, Perophora multiclathrata and Symplegma rubra. Only two of these 13 species are certainly introduced: Ascidia sydneiensis and Diplosoma listerianum. These species have been described from the Pacific, Indian and Atlantic Oceans, occurring in temperate and tropical regions, with disjunct populations along the Brazilian coast. The species Ascidia curvata, Botryllus planus and Eudistoma carolinense may be native, since their distributions are restricted to the western Atlantic, and colonization may have been natural. The species Lissoclinum fragile, Distaplia bermudensis, Clavelina oblonga, Botryllus tuberatus, Didemnum granulatum, Symplegma rubra, Cystodytes dellechiajei and Perophora multiclathrata all have wide-spread geographic distributions yet their origins are unknown. Thus, these species are tentatively better classified as cryptogenic (possibly introduced), pending DNA studies. Besides geographic distribution, historical records for species are important for determining residency status, yet these data were never collected for Paranaguá Bay. Hence, we are initiating a molecular study of C. oblonga with the objective of establishing the relationship between the populations in Paranguá Bay and those of other regions of coastal Brazil. We are sequencing a part of the COI gene for a phylogeographic reconstruction of the studied populations. At present, none of the species studied is sufficiently abundant to consider them invasive or pests, in contrast to other areas in which they may cause great environmental and economic damage. The possibility of such damage suggests that long-term study is needed in Paranaguá Bay.

Distribution and ecology of four colonial ascidians: Botryllus schlosseri, Botrylloides violaceus, Diplosoma listerianum and Didemnum sp. in the Gulf of Maine. Dijkstra, Jennifer A.¹, <u>Harris.L.G.¹</u> and Westerman. E.L.¹. ¹Department of Zoology, University of New Hampshire, Durham, NH

Exotic ascidians are a growing concern for both ecologists and natural resource managers as they are rapidly becoming dominant members in estuarine and coastal zone communities. Within these communities they disrupt the functional integrity of a system by altering local habitats and distribution patterns of resident species. More recently, attention has been directed towards the introduction of colonial ascidians in coastal and estuarine areas in the Gulf of Maine. This study focuses on the invasion of the Gulf of Maine by the colonial ascidians Botryllus schlosseri, Botrylloides violaceus, Diplosoma listerianum, and Didemnum sp. The goal of this study was to document the arrival, changes in abundance and space occupied throughout the year to compare seasonal variability among the four ascidian species. Distribution of species in New Hampshire and Maine coastal zones were documented by surveys and personal observations beginning in 1969. Seasonal variability of the area occupied per species was documented using $0.1m^2$ Plexiglas panels deployed underneath the pier at the University of New Hampshire's Coastal Marine Laboratory. Vertical and horizontal panels were deployed in February and July of 2003 and continue to be photographed on a monthly basis. Given the ubiquity of invasive species in coastal and estuarine systems, this study of colonial ascidian ecology and documentation of their arrival is essential to the management of benthic communities in the Gulf of Maine.

Did green crabs facilitate the clubbed tunicate (*Styela clava*) invasion of Prince Edward Island?

Locke, Andrea¹, Hanson, J.M.¹, Thompson, J.² and Ellis, K.M.².¹Gulf Fisheries Centre, Fisheries and Oceans Canada, P.O. Box 5030, Moncton, NB E1C 9B6, Canada; ²Department of Biology, University of New Brunswick, Saint John, NB, Canada

The initial establishment of clubbed tunicates (Styela clava) in Atlantic Canada occurred · in an estuary of eastern Prince Edward Island in 1998. The same estuary had served as the point of introduction of green crabs (Carcinus maenas) to Prince Edward Island in 1997. From this point source, both clubbed tunicates and green crabs have dispersed to other estuaries of Prince Edward Island, especially those on the eastern end of the Island. Curiously, clubbed tunicates have only flourished in estuaries that contain large populations of green crabs. Our studies of green crab distribution and ecology in the southern Gulf of St. Lawrence have shown green crab presence to be associated with significant losses of diversity and/or biomass of fishes and invertebrates. Did the green crab destabilize Prince Edward Island estuaries such that the clubbed tunicate could gain a foothold? We will discuss the effect of green crabs on predators of the clubbed tunicate. For example, the lunar dovesnail (Astyris lunata), one of the known predators of recently settled clubbed tunicates, was heavily preved upon during an experiment where densities of green crabs were manipulated (the snail was all but eliminated from high density enclosures). Other biotic and abiotic factors potentially affecting the establishment of clubbed tunicates in the region will also be discussed.

Novel habitats created by non-indigenous species: The role of oysters and ascidians as biological substrata for fouling communities.

Rodriguez, Laura F. Section of Evolution and Ecology, University of California - Davis, Davis, CA

The facilitative impacts of non-indigenous species were examined by comparing the fouling communities growing on two different biological substrata: the commercially grown non-indigenous oyster *Crassostrea gigas* and the invasive ascidian *Microcosmus squamiger*. In a system where space is a limiting resource, the introduction of habitat-creating species likely increases species diversity and abundance. This study examines the diversity dynamics at small scales, comparing fouling community development on two novel substrata. Results show that non-indigenous species can facilitate both native and invasive species by providing critical habitat. Further, results demonstrate that all substrata are not equal, and that there are species-specific distribution differences depending on substrata sampled.

The shallow bay of San Quintin (Baja California, Mexico) was historically dominated by soft sediment habitats. The only natural hard substrata available for the development of fouling communities were limited to small areas of volcanic rock debris. Currently, however, the bay supports a large oyster (*C. gigas*) aquaculture business. Oyster shells provide the primary source of space for the development of fouling communities in this system. Further, this available fouling space has facilitated the invasion of the ascidian, *Microcosmus squamiger*. This solitary ascidian (Order Stolidobranchia, Family Pyuridae), native to Australia, was first reported in 1986, and has since become a dominant member of the fouling communities in Southerm California harbors. *M. squamiger* is dependent upon the non-indigenous oyster as a source of available living surface. However, *M. squamiger* also provides a secondary source of space for other fouling species. Other species of the *Microcosmus* genus are known to be colonized by up to 200 epibiont species, a characteristic which is likely the namesake for the genus.

This study compares the fouling communities that develop on both of these biological substrata. Recruitment and succession of oyster fouling communities have been sampled monthly, and show patterns of seasonal settlement and growth of epibenthic organisms. The role of both oysters (*C. gigas*) and the invasive ascidian (*M. squamiger*) as biological substrata has also been examined by analyzing fouling species assemblages present on each. Results show that, while some species commonly foul both types of biological substrata, other species are found on only one or the other. Oysters are inherently important as the main source of hard substrata, without which fouling communities would be mostly absent in the system. This study finds that *M. squamiger* also plays a unique role by providing spece for competitively inferior species, or species that are recruiting when the primary oyster shell is already occupied. It is important to consider the community wide impacts that can arise when invasive species are functionally novel in a community, such as this study shows with the presence of two non-indigenous habitat modifiers.

Diurnal, lunar and tidal patterns of settlement in tunicate larvae.

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Gross patterns of settlement have been documented for numerous species of tunicates; however, few studies have examined fine-scale patterns of tunicate settlement. We used automated in situ sampling devices to monitor diurnal, lunar, and tidal patterns of settlement by tunicate larvae in Long Island Sound, USA. From July-October 2003, we assessed diurnal differences in tunicate settlement in seven separate, week-long assays; diurnal periods were defined as morning (0300-0900 h), day (0900-1500 h), evening (1500-2100 h) and night (2100-0300 h). To determine if lunar or tidal components affected settlement, we consistently sampled during the weeks of the full or last quarter moon; these lunar phases corresponded to morning high- or morning low-tides respectively. Different tunicates species exhibited different diurnal patterns of settlement. The colonial tunicates, Botryllus schlosseri and Botrylloides violaceus, had the highest levels of settlement during the day and lowest levels at night. The colonial tunicate Didemnum sp. had high levels of settlement during the day and evening, but significantly lower levels during the morning and night. The solitary tunicate *Ciona intestinalis* had the highest level of settlement during the morning. The solitary tunicates Ascidiella aspersa and Molgula manhattensis had similar levels of settlement across all diurnal periods. Settlement of tunicates did not appear to be affected by lunar or tidal factors. Differences in settlement patterns appear to reflect differences in life history strategies between colonial and solitary tunicates.

Recruitment, settlement and mitigation of the tunicate *Ciona intestinalis* on a mussel farm in Nova Scotia, Canada.

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The ascidian species, *Ciona intestinalis*, although indigenous to Nova Scotia, Canada has become a problem for local aquaculturists. This study consisted of two separate components. In the first part, the local recruitment ecology was studied by recording the depth, distribution and timing of settlement on four characteristically different aquaculture leases (high wave exposure, low wave exposure, fallowed, and historically heavily fouled with tunicates) at Indian Point, Nova Scotia. The collection of settled animals occurred weekly from 11 designated collectors deployed at a depth of 3.5 m over two seasons (June-December in both 2003 and 2004). The depth settlement was studied by deploying four separate lines in a lease devoid of culture gear and a lease with culture gear, with collectors at depths of 0.5, 4.5, 8.5, 12.5 and 16.6 m. Water chlorophyll content, temperature, and number of recruits were recorded. Settlement occurred in two peaks in both 2003 and 2004, with the first peak lasting from late June to late July, and the second peak lasting from early September to early November. Consistent settlement differences among the four leases were observed, with the lease with high wave exposure having the lowest number of settled tunicates, followed by the recently fallowed lease. Testing for the depth of settlement indicated that for both years, recruitment occurred at a preferred depth of 4.5 m, which is also the average depth of the mussel lines, and to a lesser extent at 8.5 m. Practically no recruitment was seen at the surface (0.5 m) or in deeper water (12.5 and 16.5 m). These results seem to indicate a patchy recruitment on a small spatial scale, where many new recruits appear to settle very close to pools of adult tunicates on mussel lines. This result is also supported by a preliminary experiment where higher settlement was observed on collectors in on-shore flow-through tanks containing a few adult tunicates than in similar control tanks devoid of adult tunicates. This appears to indicate that at least some of the larvae may have very limited dispersal. In the second component of this study, three mitigation strategies were evaluated. Ultraviolet radiation, ultrasonic pulsation, and predation treatments were tested on field gathered animals under laboratory conditions. Preliminary results show that ultrasonic and predation treatments were not successful, but 10minute exposure to either moderate or high level of UV radiation induced 100% mortality in both adult and juvenile tunicates. The population dynamics of this invasive ascidian and options for management will be discussed.

Diel larval concentrations of an invasive ascidian *Styela clava* Herdman in Prince Edward Island, Canada.

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Ascidians generally have a very short larval stage. This makes the timing of larval sampling a crucial part of evaluating recruitment potential. A field experiment was carried out in Murray River, Prince Edward Island, to document *Styela clava* diel larval concentrations as well as various associated parameters. Data were gathered over 30 hour sampling periods conducted on 4 August and 14 September 2004. Water samples were taken at the surface, 2 m and 4 m simultaneously on an hourly schedule. Temperature, salinity and light intensity were measured at all depths at the time of larval sampling. Results indicated that peaks in larval abundance were in most cases limited to a 2-hour time interval in the early afternoon. Additionally, sampling depth had an effect on larval concentrations. Very few larvae were found at the surface, while abundance increased at greater depth. A better understanding of early life stage development was also achieved since abundance of a few embryonic stages was documented. Findings are discussed in relation to the environmental parameters measured. Information gathered in this study should lead to standardised sampling procedures for *S. clava*.

Setting ascidian invasions on the global stage.

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Maritime history provides a strong perspective on our state of knowledge of ascidian (and other) invasions both regionally and globally. Despite the fact that for thousands of years Scandinavians, Romans, Phoenicians and many others sailed around Europe, the Mediterranean, and Africa, we interpret the distribution of all ascidians within the Euro-African theater as natural. Although over 500 years ago Chinese vessels sailed over much of the Western Pacific and Indian Oceans, no Asian sea squirts are recognized as introduced anywhere until the 1800s. While European vessels began sailing around the world in the 1500s, no European ascidians are considered introduced anywhere until the 1800s, nor do we recognize any Pacific Ocean ascidians in the Atlantic until the 1800s. The potential scale of overlooked earlier invasions is critical to our understanding of what species we consider native, and thus to our understanding of evolved community structure. For example, the "classic" European ascidians Botryllus leachii (described in 1816 from the Mediterranean) and Botryllus schlosseri (described in 1766 from Falmouth harbor, England) may have been invasions as early as the 1500s from the Pacific Ocean, the center of botryllid diversity (and would thus be predicted to have lower genetic diversity in Europe than in the Pacific). The fact that ascidian invasions continue, despite centuries of shipping, suggests that new invasions serve as some of the clearest signals of environmental change.

Habitat variability in the invasion of Long Island Sound by *Didemnum* sp. and its interaction with the resident community.

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Over the past 30 years southern New England has been invaded by several species of ascidians, including *Botrylloides violaceus*, *Diplosoma listerianum*, *Styela clava* and *Ascidiella adspersa*. These species are dominate in coastal embayments and marinas, but are usually absent from more open water coastal areas. A species of colonial ascidian, *Didemnum* sp. has invaded southern New England over the past 10 years. and we first observed it in eastern Long Island Sound in 2000. Unlike the other ascidian invaders, *Didemnum* sp. seems to be more abundant in open coast and offshore deeper waters than in protected embayments, harbors and marinas. We conducted a field experiment to contrast the success of *Didemnum* sp. in open coast and protected harbor habitats and whether this success was affected by the community it invaded. We found that *Didemnum* sp. grew faster and was more dominant in open coast habitat. This success appears to be a consequence of the low recruitment rates of competing species in these habitats and the ability of *Didemnum* sp. to grow over and dominate available space quickly.

Does the non-native solitary ascidian Ciona intestinalis depress species richness?

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Non-native ascidians are a dominant feature of many sessile marine communities throughout the world and may have negative effects on species diversity. We tested the effect of the non-native *Ciona intestinalis* on the sessile invertebrate community in San Francisco Bay, where it occurs in dense aggregations. In particular, we compared species richness between PVC panels from which *C. intestinalis* were experimentally removed ("gardened") to panels with naturally dense *C. intestinalis* growth. Moreover, we used fouling panels of four sizes (between 49 cm² and 1177 cm²) to measure the effect of *C. intestinalis* recruitment on species-area relationships.

We initially deployed 120 fouling panels (30 of each size) at a site known to have dense populations of *C. intestinalis*, assigning these to three different treatments: (1) Experimental removal, whereby new recruits of *C. intestinalis* were removed on a weekly basis, pulling plates out of the water for a short time period to do so; (2) Manipulated control, whereby panels were removed from the water each week (as in the experimental removal) but were not gardened; and (3) Unmanipulated control, which remained in the water throughout the experiment. At a second site, where *C. intestinalis* occurred in relatively low abundance, we deployed 10 panels of each size (40 total) as unmanipulated controls, to measure inter-site differences in species richness associated with *C. intestinalis* density. After 4 months, all of the panels were collected and analyzed to estimate total species richness and abundance (percent cover) of *C. intestinalis*.

At our experimental site, C. intestinalis densities were greater than 20,000 individuals per m^2 . C. intestinalis abundance was negatively correlated with species richness, and the C. intestinalis removal treatment had a positive effect on species richness. Species richness at the non-experimental site was comparable to that of gardened panels. These data, combined with additional observations, suggest C. intestinalis depress local species diversity across many sites. Additional experiments are now underway to examine effects on both sessile and mobile biota.

Space invaders: the effect of small-scale disturbance on invasion success in marine communities.

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Introductions of non-indigenous species have resulted in ecological problems worldwide, though the link between disturbance and potential invasion has rarely been studied in the marine environment. In this study, hard substrate marine communities of Long Island Sound, USA were used as an experimental system to assess the effect of disturbance on 1) the resident species and recent invaders, 2) growth form (i.e. colonial and solitary growth forms of ascidians), and 3) the dominant species specific responses within the community. As different stages of community development may not respond to disturbance in the same manner, community age was an additional factor considered through manipulation of a "young" (5 wk) community and an "old" (1 yr) community. Disturbance treatments exposed primary substrate, a limited resource, and were characterized by frequency (single, biweekly, monthly) and magnitude (20%. 48%. 80%). In both communities, frequency of disturbance had a significant positive effect on space occupation of recent invaders and a significant negative effect on resident species. In the "young" community, magnitude of disturbance also had a significant effect. In both communities, colonial ascidians occupied more primary space than controls in response to increased disturbance frequency and magnitude. In contrast, solitary ascidians occupied significantly less space than controls. Species-specific responses were also similar in both "young" and "old" communities. The non-native colonial ascidian *Diplosoma listerianum* responded positively to increased disturbance frequency and magnitude, and occupied more primary space in treatments than in controls. The resident solitary ascidian Molgula manhattensis responded negatively to increased disturbance frequency and magnitude, and occupied less primary space in treatments than in controls. The amount of available secondary substrate (the surface of organisms attached to primary substrate) declined with increased disturbance frequency and magnitude. For the majority of organisms studied, disturbance treatments had negative effects on secondary space acquisition. The results from this study indicate that smallscale biological disturbance is an important factor in facilitating the success of invasive species and colonial organisms in subtidal hard substrate communities.

Diversity in fouling communities and resistance to invasive ascidians: the role of settlement complementarity and species interactions.

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New ascidian invaders commonly arrive via hull fouling and ballast transport into harbors, making them an excellent model system for understanding the role of native diversity in facilitating or repelling invasion. Interestingly, experimental manipulations and observational surveys often produce conflicting conclusions regarding the role of native diversity in community susceptibility to invasion. We propose to resolve this conflict by focusing on the mechanism by which communities may resist invasion, and then examine how processes later in life may mask this initial resistance. Settlement of larvae at complementary times of year reduces invasibility by ensuring consistent space occupancy and eliminating favorable times of year for invasive ascidian recruitment. We simulated recruitment in New England and northern California harbors with real data to examine the relationship between diversity and temporal complementarity. These simulations show that, as diversity declines, there are fewer days with high numbers of settlers, indicating more periods favorable to invasion. In surveys of adults in Bodega Harbor, however, we found a spurious (non-causal) positive correlation between native and invader diversity driven by similar effects of native predators on the diversity of both native and non-native species. Similarly, in a separate survey of adult ascidian communities that accounted for free space, plots with high amounts of free space showed a positive correlation between native and invader diversity, but not if they have low amounts of free space. However, in plots with a low abundance of the exotic mussel Mytilus galloprovincialis, which functions as a foundation species providing additional free space for settlement, we find that plots with a low amount of free space have a negative correlation between native and invader abundance. Biotic resistance shows a signature in adult communities only when space is limiting and foundation species are rare. These studies indicate that species loss within harbors due to pollution or other invaders may leave a potential port open to invasion by exotic ascidians. This increased susceptibility can be masked, however, if one does not account for other biological interactions occurring at the community level.

Facilitating invasions: The effects of temperature on the role of non-indigenous ascidians in fouling communities.

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Invasive species and climate change can drastically alter ecological processes in benthic marine communities. The subtidal benthos of Massachusetts has recently been invaded by five non-indigenous ascidian species, *Ascidiella aspersa, Botrylloides violaceus, Didemnum* sp., *Diplosoma listerianum*, and *Styela clava*. This study examines effects of seawater temperature, as a proxy for climate change, on *B. violaceus* and *D. listerianum* and the impact these ascidians have on native sessile fouling communities. Field experiments were conducted over a four month period at two locations (Lynn and Woods Hole, MA) to examine growth dynamics over the regional thermal and geographic range. Laboratory temperature growth experiments were conducted on *B. violaceus* and *D. listerianum* at Marine Science Center, Nahant, MA.

While *Diplosoma* and *Botrylloides* behaved similarly across this geographic and temperature range, temperature had a definite effect on colony growth. In laboratory growth experiments a 1.8°C increase in seawater temperature facilitated the proliferation of *D. listerianum* colonies by 53%, but elicited no response in *B. violaceus*. In the field, *B. violaceus* and *D. listerianum*, exhibited rapid two-week growth rates during the summer months (21 and 37-fold, respectively), with more rapid growth at the warmer Woods Hole site.

In these fouling communities, invasive ascidians occupy as much as 80% of substrate primary space and rank relatively high in the species richness. In the warmest periods (late summer and early fall) invasive ascidians exhibit the highest percent cover, and thus have the greatest impact in a community. However, the presence or absence of a key species of the native community can affect the invasiveness of invaders as will the timing of recruitment. Stochastic recruitment processes enable native species to take hold when space becomes available over time, which in turn can decrease the percent cover of invaders as they lose their space holding. Observed over the four months, recruitment and growth of the native species influenced the structure of experimental communities more than did the pre-seeding with *Botrylloides* or *Diplosoma* colonies. However, with projected global climate change, a rise in sea surface temperatures could facilitate successful invasions by other ascidians, and exacerbate the cumulative impacts of invasions on benthic communities.

Assessment of boaters as a vector for the clubbed tunicate (*Styela clava*) in the southern Gulf of St. Lawrence.

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The clubbed tunicate (*Styela clava*), which is native to the Sea of Japan first became established in the southern Gulf of St. Lawrence (sGSL) in 1998 and has subsequently spread to several estuaries of Prince Edward Island (PEI). In PEI this species is causing major problems for the mussel aquaculture industry by overgrowing mussels, which have led to reduced yields and increased cost of harvesting and processing of mussels. To prevent the spread of this species in the sGSL, transfer of lines or spat from infected areas has been prohibited, but no measures have been put in place to prevent the spread of *S*. *clava* by boaters. To address this issue, I carried out a series of experiments to determine the preferential settlement patterns of *S*. *clava* on different types of boat-hull materials, and their ability to survive exposure to air (i.e., during trailering).

Settlement plates made of the most common types of boat-hull materials found in PEI were used to collect tunicates. These 10cm x 10cm plates were made of fiberglass, wood and aluminium. The aluminum plates were left untreated, while the fiberglass and wood plates were painted with either black or white anti-fouling paint, or black or white exterior (house) paint. Tunicates settled mainly on the aluminum plates and plates painted with exterior paint; very few settled on the plates treated with anti-fouling paint. There appeared to be a preference for the plates painted black rather than white and transport on boat hulls not treated with an anti-fouling paint appeared likely.

To determine the ability of *S. clava* to survive overland transport, another set of 10cm x 10cm settlement plates was constructed using aluminium and fiberglass. Again, in this experiment the aluminum plates were left untreated, while the fiberglass plates were painted with black exterior or anti-fouling paint. Tunicates were exposed to air during the first three days of September 2004 (for up to 48 hours). After exposure I assessed tunicate survival using vital dyes and physiological responses. I found that the majority of tunicates survived 48 hours of exposure. Their ability to survive this long out of water indicates that they could be transported, by normal boating activities anywhere in Atlantic Canada.

Risk factors and spread of Styela clava in Prince Edward Island, Canada. Davidson, Jeff and Arsenault, G. Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, Canada PE C1A 4P3

Styela clava (clubbed tunicate) was identified as an aquatic invasive species in a mussel (Mytilus edulis) growing area on Prince Edward Island, Canada in 1998. In affected areas, it has had a negative impact on mussel culture, attaching in high densities to mussel socks and equipment, competing for food resources and fouling equipment. It has since spread to other bays and estuaries. The impact of this spread has varied spatially and temporally. A three-year study to describe the spread of *S. clava* in newly introduced areas indicates that the intra-bay spread radiates from the point of where it was first identified and the intensity of recruitment has increased each subsequent year. In an area with minimal molluscan culture, recruitment and spread did increase in subsequent years.

In mussel growing areas with an established population of *S. clava*, risk factors were evaluated to assess an association with recruitment and growth. Risk factors considered were location in the estuary, depth of water, position in the water column, water flow, water temperature and salinity.

Ecological observations of the colonial tunicate *Didemnum* sp. in a New England tide pool habitat and strategies for managing invasive colonial ascidian species. <u>Valentine, Page C.¹</u>, Carman, Mary R.², Blackwood, Dann S.¹ and Heffron, Erin J.¹ ¹U.S. Geological Survey, Woods Hole, MA 02543; ²Woods Hole Oceanographic Institution, Woods Hole, MA 02543

The invasive colonial tunicate *Didemnum* sp. has colonized northwestern Atlantic coastal habitats from southern Long Island to Maine. It threatens to alter fisheries habitats and particularly shellfish aquaculture. New England tide pool habitats experience climates that are far harsher than those of subtidal habitats. Tide pool species are subject to air exposure, to daily changes in water depth and temperature, and to a broad annual temperature range. Stressful conditions and the accessibility of tide pools make them ideal sites for ecological studies of *Didemnum* sp. that can be a basis for its management and for predicting its distribution.

Observations at Sandwich, MA from December 2003 to January 2005 show that the species tolerates water temperatures ranging from -1.8 to 23.5 degrees C, with daily fluctuations of up to 9 degrees C. It attaches to pebbles, cobbles, and boulders, and it overgrows seaweeds, sponges, mussels, and other tunicates. In April and May, colonies are small olive green patches on the bottoms of rocks; they mature into colonies that range in color from pale pink to pale vellow to pale orange. Colonies grow rapidly from July through September. Colony health declines from October to March (presumably in response to low water temperature) and is manifested by color changes, by small dark spots that represent coalesced fecal pellets in individual zooids, and by a peeling-away of colonies from substrates. The species does not exhibit this seasonal cycle of growth and decline in subtidal habitats on the Georges Bank fishing grounds where the daily climate is relatively stable and annual temperatures range from 4 to 15 degrees C. At Sandwich, colonies died that were exposed to air at low tide for approximately three hours over a period of 28 days (this effect probably requires only a few days but remains to be documented at Sandwich). Experiments with colony fragments (5 to 9 sq cm) show they re-attach and grow rapidly by asexual budding, increasing 6- to 11- fold in size in the first 15 days. *Didemnum* sp. has no known predators except for common periwinkles (Littorina littorea) that graze on weak and dying colonies in the October to March time period and whenever colonies are stressed by desiccation.

The tendencies of the tunicate (1) to attach to firm substrates, (2) to rapidly overgrow other attached species, (3) to tolerate a wide temperature range, (4) to be free from predation, and (5) to spread by colony fragmentation combine to make it a potential threat to benthic marine habitats and aquaculture. *Didemnum* sp. is known to overgrow mussels and sea scallops, and it likely envelops oysters and other bivalves too. To address this threat, a management strategy for shellfish aquaculture could be based on the species' seasonal growth cycle, its temperature tolerances, and its vulnerability to desiccation. Induced environmental stress might deter the establishment of the tunicate, and it might weaken existing colonies, causing them to peel off the substrate and making them susceptible to predation by the common periwinkle. The species could be appropriate for the control of other colonial tunicate species such as *Botryllus schlosseri* and *Botrylloides violaceus*. Physical removal of living *Didemnum* sp. from shellfish and from aquaculture equipment must avoid the release of colony fragments to the habitat to prevent the spread of the species.

Dumb barge teaches important lessons: the arrival, spread and management of an undesirable sea squirt in the Marlborough Sounds, New Zealand.

<u>Coutts</u>, <u>Ashley D.M.</u> and Taylor, M.D. Biosecurity Group, Cawthron Institute, Nelson, New Zealand

This paper presents a case study about the arrival, spread (both natural and artificial) and management of an undesirable sea squirt, *Didemnum vexillum*, in the Marlborough Sounds, New Zealand. The species arrived in Picton, Marlborough Sounds via a large steel barge, the *Steel Mariner* on 23 April 2001. On 18 December 2001, during a routine biosecurity survey of Shakespeare Bay (Picton), Cawthron divers estimated that a total of 1,396 kg (wet biomass) of *D. vexillum* smothered the hull of the *Steel Mariner* and approximately 460 kg had established on the seabed below. The New Zealand Mussel Industry Council considered the species to be a serious biosecurity risk to the GreenshellTM mussel industry because of its smothering capabilities, rapid growth rates and preference for artificial structures.

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Initially there was controversy over whether the New Zealand Ministry of Fisheries (MFish) or Marlborough District Council (MDC) was responsible for managing the species. This resulted in valuable time lost, and the ascidian was able to spread naturally to surrounding artificial structures in Shakespeare Bay including other barges, recreational vessels, salmon cages and wharf piles making the likelihood of containment extremely challenging. Furthermore, some of the infected structures translocated the species to new locations throughout the Marlborough Sounds. Given the likelihood the species would spread to mussel farming areas, MFish and MDC have funded the development and application of various management strategies over the past two years in a hope to at least contain the spread, if not eradicate the species from the Marlborough Sounds (e.g. underwater vacuum and filtering system; various smothering techniques using dredge spoil, plastic wrapping and filter fabric; *in-situ* treatment of vessels using plastic and chlorine; re-antifouling vessels; water-blasting moorings lines; in-water scrubbing of artificial structures; scuttling and beaching of vessels).

While most of the management measures utilised over the past two years have been successful in reducing the biomass of *D. vexillum* in Shakespeare Bay, a successful eradication has not been achieved to date. Many lessons have been learned along the way, however, including the importance of early detection, positive identification and thorough knowledge of the species distribution, proven incursion response tools, clear identification of management responsibility, a large budget and stakeholder buy-in.

Occurrence and distribution of ascidians in Vizhinjam Bay (southwest coast of India). <u>Abdul Jaffar Ali H.</u>¹ and Sivakumar, V.². ¹Department of Zoology, Muslim Arts College, Thiruvithancode – 629 174, Kanyakumari District, India. ²V.O. Chidambaram College, Tuticorin – 628 008, India

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Occurrence and distribution of ascidians in the Vizhinjam Bay were studied in relation to three different habitats such as intertidal area (Station 1), harbour installation at a depth of 4 to 5 m (Station 2) and mussel bed at a depth of 15 to 20 m (Station 3). A total of 33 species of ascidians including 20 colonial and 13 simple ascidians fall under 6 families and 15 genera were recorded. Of the three stations, a maximum of 27 species were recorded at station 2 followed by 19 and 7 at station 1 and station 3 respectively. Four species namely *Styela canopus, Microcosmus exasperatus, Symplegma oceania and Didemnum candidum* were common in all stations studied. In the present account, the details of the occurrence and distribution of ascidians and also the probable reasons for the differences have been discussed.

An invasive tunicate, Didemnum sp., in Narragansett Bay, Rhode Island.

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There has been a recent appearance of a colonial invasive tunicate *Didemnum* sp. in Narragansett Bay, Rhode Island. Preliminary photographic observations made in the field in the late summer of 2004 indicated that there may be competition between the introduced *D*. cf. *lahillei* and the blue mussel *Mytilus edulis*. The proposed research will determine the extent of spatial competition between the tunicate and the mussel. Quantitative underwater quadrats using photography at the GSO dock and Fort Wetherill in Jamestown, RI will provide an overview of spatial competition in the field. If the tunicate shows a negative effect on the mussels and direct competition for food, growth will be monitored in several bay locations. Studying the effects of the invasive tunicate will provide information on the basic biology of this relatively unknown species, its effects on native species and possible environmental factors that control its distribution and growth.

Benthic foraminifera on the invasive ascidian, Didemnum sp. Carman, Mary R. Geology and Geophysics Department, Woods Hole Oceanographic Institution, Woods Hole, MA 02543

An invasive and unusually aggressive ascidian, *Didemnum* sp., first appeared in New England bays and harbors in the early 1990s, and in the waters around Cape Cod in 1993. The shipping industry probably introduced most of the New England ascidians, but the origin of *Didemnum* sp. and the precise date and location of its introduction there is unknown. *Didemnum* sp. is colonial, and forms large mats capable of biofouling artificial and natural substrates and of posing a threat to benthic habitats and aquaculture. *Didemnum* sp. has no known predators other than the common periwinkle (*Littorina littorea*), which grazes only on dying or dead colonies.

To the naked eye, the colony surfaces of *Didemnum* sp. appear very clean and not a favorable substrate for epibiota. However, careful washing of colony surfaces revealed the presence of benthic foraminifera. During 2003 and 2004, I collected 52 samples of *Didemnum* sp. and other ascidians for foraminifera. The purpose of the collection was to determine whether or not the foraminiferal assemblage might also be invasive and thus might provide a clue to the place of origin of *Didemnum* sp. Sample locations include the New England coast from Connecticut to Maine (with a concentration in the Cape Cod area), northern California, Zeeland in the Netherlands, and Shakespeare Bay, New Zealand.

From the New England samples, I have identified specimens of eighteen species of benthic foraminifera. The most common species of foraminifera represented were *Rosalina floridana*, *Cornuspira involvens*, *C. planorbis*, *Quinqueloculina bicornis*, *Miliolinella subrotunda*, *Glabratellina lauriei*, *Elphidium galvestonense*, and *E. margaritaceum*. Foraminiferal assemblages on *Didemnum* sp. from other regions sampled were composed of the same cosmopolitan species that are in New England, plus other species indigenous to each region. Because I observed no exotic foraminifera species, I conclude that *Didemnum* sp. did not introduce non-native foraminifera originating from their native habitats.

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The New England foraminiferal assemblages were relatively diverse on *Didemnum* sp. attached to natural substrates in the nearshore and offshore, however, the didemnid's surface and interior layers did not appear to contain any living foraminifera. The lack of living organisms attached to *Didemnum* sp. is probably attributable to the acidic tunic. Empty tests of benthic foraminifera, along with quartz grains, pieces of seaweed, bits of bivalve shell, and other detrital matter in the water column probably settled on or became stuck to the ascidian's surface and were overgrown and incorporated into the tunic and spicule layers of *Didemnum* sp.

Water quality, nitrogen pollution and ascidian diversity in northwestern Atlantic coastal waters of Massachusetts, USA.

Carman, Mary R.¹, <u>Bullard, S. G.²</u> and Donnelly, J. P.¹. ¹Geology and Geophysics Department, Woods Hole Oceanographic Institution, Woods Hole, MA; ²University of Hartford, Hillyer College, West Hartford, CT

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Nitrogen is the main contaminant causing degradation of water quality in the temperate coastal waters of Massachusetts (Buzzards Bay, Pleasant Bay, Vineyard Sound, northern Rhode Island Sound, and southern Cape Cod Bay). Nitrogen is an essential part of marine nutrient supply, but in excess, can lead to a decline in bio-diversity and water quality and in severe cases, eutrophication. The effect of nitrogen loading on ascidians that inhabit New England coastal waters was previously unknown. In a survey of coastal waters during 2003 and 2004, we correlated the presence of nine species of ascidians (three native and six nonindigenous) with water quality (good, fair, and poor) as reported in the literature and compared ascidian species diversity with water quality and nitrogen loading data.

We found that the biodiversity response of ascidians to nitrogen is different from most other organisms. Many faunal groups exhibit high biodiversity at low nitrogen levels, with diversity declining as nitrogen loading increases. However, the greatest diversity of ascidians (generally five, but up to seven species) was present in areas with fair water quality and intermediate levels of nitrogen, the most common water conditions in our region. A lower diversity of ascidians (two or three species) occurred in areas with good water quality and low excess nitrogen levels and in areas with poor water quality and high nitrogen (up to three species, but often no species surviving).

Both native and non-native ascidians were most diverse in areas with fair water quality. *Molgula manhattensis*, a native species, was tolerant of good, fair, and poor water quality, while the other two native species, *Ciona intestinalis* and *Aplidium stellatum*, were observed only in areas of fair water quality. Of the invasive, nonindigenous species, *Botryllus schlosseri* and *Styela clava* inhabited good, fair, and poor water quality sites; *Botrylloides violaceus* was found at sites of good and fair water quality; *Diplosoma listerianum*, *Ascidiella aspersa*, and *Didemnum* sp., were present in fair water quality only. Nonindigenous species dominated the ascidian population at good, fair, and poor water quality sites whether attached to artificial or natural substrates.

Overall, ascidian diversity was highest in areas with fair water quality and lower in areas with good or poor water quality. Given their unusual bio-diversity response, ascidians may be useful bio-indicators of nitrogen content in coastal waters.

Invasive tunicates in the Puget Sound, Washington, USA.

<u>Davis, Marla D.</u> and Swalla, B.J. Department of Biology, University of Washington, Box 351800 Seattle, WA

Invasive tunicates can be a menace to both ecosystems and marine aquaculture in the coastal United States. There are six species of non-native ascidians invasive in the Puget Sound, the majority of which appear to be relatively recent colonizations. One of the most recent invasives appears to be the same *Didemnum* sp. that has proven to be a significant worldwide economic and environmental hazard in the Atlantic along the coast of New England, in the Gulf of Mexico, France and Australia. In light of such grave threats, identifying the vectors and sources of introduced organisms in order to prevent and possibly combat invasions is crucial. Distinguishing between primary invasion (source population from native range) and secondary invasion (source population from a previously invaded site) is fundamental to identifying pathways of invasion. Toward this end, the distributions of invasive ascidians present in the Puget Sound were mapped and compared with distribution maps of the same six species along the Pacific Coast, from Southern California, USA to Vancouver Island, Vancouver B.C, Canada. Included among the Puget Sound sites are several new locations. Ongoing work utilizes molecular phylogenies to identify invasive populations relative to samples from both throughout native ranges and the US Pacific Coast, in order to pinpoint the source population and thereby yield further clues as to the probable vectors for the introduction of invasive ascidians in the Puget Sound.

The distribution of Styela clava (Tunicata, Ascidiacea) in European waters. <u>Davis, Martin H.</u> and Davis, Mary E. Fawley Biofouling Services, 270, Burgess Road, Swaythling, Southampton, Hampshire. SO16 3BE, UK

The introduction of the immigrant ascidian *Styela clava* Herdman, 1882 into European waters can be dated quite precisely; it was first recorded in British waters in 1953. The chronology of its spread along the west coast of Europe over fifty years is described, and the current distribution is reviewed with reference to physical constraints and the dispersal methods available.

Styela. clava provides a good model for the dispersion of organisms with short-lived larvae. It is oviparous; the eggs hatch after 12 to 15 hours and the negatively geotactic larvae are active for approximately 12 hours. Natural dispersal methods involve the drifting of larvae and of adults attached to flotsam. Suggested man-aided methods involve transport of larvae in ballast water, or of juveniles and adults either attached to oyster shells or to ships' hulls. The problems associated with these proposed dispersal methods are examined. Larval dispersion may explain the spread to neighbouring sites but, since larvae would not survive long journeys, isolated populations must have been established by mature adults. Adults may be transported attached to ships' hulls; however, they are rheophobic and hulls are anti-fouled, so dispersal of significant numbers is unlikely.

For successful establishment, the number of larvae settling must exceed a viable minimum determined by demographic stochasticity, and the receiving habitat must be suitable, a function of environmental stochasticity. Probabilistic modelling indicates that a translocated adult colony has a greater probability of establishing a new population than a single inoculum of larvae. Ships' sea-chests offer a suitable transport method for established colonies; these seawater intake chambers provide a relatively sheltered environment for the organisms to grow to maturity. Sea-chests provide a mobile substratum for reproductively mature adults, which may then spawn in any harbour visited with suitable water quality. The temperature shock on entering a harbour may initiate synchronised spawning, further increasing the probability of successful colonisation. This dispersal method would explain the current heterogeneous distribution of this species.

The ultimate distribution limits of *S. clava* can be predicted from knowledge of its temperature and salinity tolerance. Consideration of the available dispersal methods enables the identification of likely colonisation sites, leading to more selective sampling to determine the present limit of distribution. Consequently, our present search technique for *S. clava* focuses on commercial ports or adjacent marinas, and this has proved very successful.

Predicting the future of community development: a comparative approach. <u>Dijkstra, Jennifer A</u>.¹, Sherman, H.¹ and Harris, L.G. ¹Department of Zoology, University of New Hampshire, Durham NH

In the past 25 years, three invasive colonial ascidians (Botrylloides violaceus, Diplosoma listerianum and Didemnum sp.) have become dominant members of fouling and subtidal communities in New England. While it is clear these species have affected communities, it is unclear how these species influence succession in comparison to other functional groups. The purpose of this study is two-fold: 1) Compare historical and present-day changes in abundance and area occupied by colonial ascidians; 2) Examine short-term recruitment of individuals and species among functional groups. Long-term changes in spatial dominance of colonial ascidians were determined through the analysis of 8 vertical and 8 horizontal Plexiglas panels deployed underneath a pier in Newcastle, New Hampshire in July 2003. These panels were photographed on a monthly basis and compared to photographs, also taken on a monthly basis over three years, of similar Plexiglass panels deployed in Porstmouth Harbor, in July 1979. Differences in recruitment of species into functional groups were examined in 2 two-week experiments. Each experiment consisted of 3 replicates of two control and six functional groups (sponges, encrusting bryozoan, hydroids, solitary tunicates and colonial tunicates). Experiments were performed in July and August using 70-100% cover and 20-50% cover treatments on 100cm² Plexiglas panels. Preliminary results indicate that ascidians currently occupy more space when compared to historical data. Additionally, the observed recruitment patterns in colonial ascidians resemble those of sponges and not other functional groups. Initiating studies examining the role ascidians play on community development is crucial to evaluate the response of communities to invasions.

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The genetics of establishment and spread of three non-native ascidians in NW Europe. <u>Dupont, Lise¹</u>, Viard, F.² and Bishop, J.D.D.^{1,3}. ¹Marine Biological Association of the United Kingdom, Citadel Hill Laboratory, Plymouth PL1 2PB, UK; ²Station Biologique de Roscoff, 29682 Roscoff, France; ³School of Biological Sciences, University of Plymouth, Drake Circus, Plymouth PL4 8AA, UK

The English Channel appears to be a hotspot for marine invasions in general (although this impression may partially reflect the vigilance of local marine laboratories on both sides of the water). However, until relatively recently only one non-native ascidian was recognised in the region: *Styela clava* became established at Plymouth in the early 1950s and has since spread as far as northern Denmark and southern Portugal. A further three non-native species have now been found, two of which were reported in NW France prior to their discovery in the UK.

We have initiated a study, mediated by the EU Marine Genomics Europe network, of genetic patterns in three species present in both France and the UK, using microsatellites and mitochondrial DNA sequence information. *S. clava* has been established for over 50 years, while *Perophora japonica* (France 1982, UK 1999) and *Corella eumyota* (France 2002, UK 2004) are more recent arrivals. We hope to use molecular information to infer the origin and extent of the initial introductions, patterns of subsequent spread, and trends in genetic diversity with time and distance from the point of introduction for each of these species.

The ecology of two non-native ascidians in fouling communities along the Strait of Juan de Fuca.

Grey, Erin K. Department of Ecology and Evolution, University of Chicago, Chicago IL

Two ascidians of northwestern Pacific origin, the colonial *Botrylloides violaceous* and the solitary Styela clava, have recently established themselves in fouling communities in northeastern Pacific marinas. These communities vary in abiotic factors such as productivity and temperature and biotic factors such as species diversity and predation pressure. I am proposing to use both observational and experimental methods to understand how these variables contribute to spatial and temporal population dynamics of *B. violaceous* and *S. clava* in different fouling communities. Regular measures of productivity, temperature and salinity will be coupled with detailed, spatially-explicit observations of the dynamics of fouling communities (species richness, relative abundance, space availability, turnover rates, etc.) and sea squirt populations (densities, recruitment rates, growth rates, size/age at first reproduction, fecundity, survival probabilities, crowding intensity, etc.). Predator exclusion experiments will be performed in order to understand how predation on recruits and/or adults influences community structure and invasive sea squirt abundance and distribution in each community. Reciprocal transplants of the squirts will be employed to estimate the extent to which populations have adapted to their local habitats. I also plan to measure the extent to which the invasive ascidians cause declines in species richness, and to determine if it is mainly through exploitative (reduction in colonization rates) or interference competition (overgrowth). Possible facultative effects by S. clava, through providing a settling surface for a diversity of epibionts, will be investigated, too. Comments and suggestions for this study are very welcome.

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Effects of temperature on growth rates of colonial ascidians: A comparison of Didemnum sp. to Botryllus schlosseri and Botrylloides violaceus. McCarthy, Anna. Smithsonian Environmental Research Center, Edgewater, MD

The occupation of unusually large areas of seafloor and rapid spread of a previously undescribed species of colonial ascidian, provisionally identified as *Didemnum* sp., have recently been observed in areas of Georges Bank (Massachusetts). Over the past 2 years it has also been found to cover large areas of rocky and cobbley bottoms in eastern Long Island Sound. Preliminary observations suggest that it was most abundant at deeper water sites with slightly cooler summer water temperatures. In this study, I examined how growth rates of *Didemnum* sp. colonies varied with temperature and contrasted its growth rate to two other colonial ascidians residing in eastern Long Island Sound, *Botryllus schlosseri* and *Botrylloides violaceous*. Temperature data collected in the field were used to set temperature conditions for laboratory experiments. The three species were exposed to four temperature treatments: ambient conditions, 4-5° C below ambient, 2° C above ambient, and 4-5° C above ambient. *Didemnum* sp. had a faster growth rate in cooler conditions (ambient or below) and exceeded the growth rates of both *Botryllus* and *Botrylloides*. The ability of *Didemnum* sp. to spread rapidly may be the result of its comparatively fast growth rate and its ability to grow in cooler, deeper waters.

Positive effects of non-native ascidians in Plymouth, England?

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Nielsen, Signe E.^{1,2}, Hawkins, S. J.¹ and <u>Bishop, J.D.D</u>.^{1,2}.¹Marine Biological Association of the United Kingdom, Citadel Hill Laboratory, Plymouth, PL1 2PB, UK; ² School of Biological Sciences, University of Plymouth, Drake Circus, Plymouth, PL4 8AA, UK

Biological invasions may considerably alter community composition and ecosystem functioning and can result in considerable economic loss. Different ecosystems and communities, however, appear to vary in their susceptibility to invasion, and differ in how they are affected by the invaders. This study uses sessile epifaunal suspension-feeding invertebrates as tractable test systems to look at the effect of non-native suspension-feeder diversity on ecosystem functioning. Potentially important non-native ascidians in Plymouth, England include *Styela clava*, *Botrylloides violaceus* and *Perophora japonica*.

Naturally settled suspension-feeding communities were obtained on settlement tiles and communities were weeded to create communities with and without non-native species. Differences in particle clearance rate (ecosystem functioning) between the different communities were measured using flow cytometry to determine the impacts of non-native species on ecosystem functioning. Results appear to show that non-native species did not significantly alter ecosystem functioning and did not appear to displace other species. In addition, *Styela clava* was found to aid localised increases in diversity by providing additional settlement space for epibionts.

Localised diversity increases in the presence of a non-native species Styela clava.

Nielsen, S.E.^{1, 2}, Hawkins, S.J.¹and Bishop, J.D.D.^{1, 2}. ¹Marine Biological Association of the UK, Citadel Hill, Plymouth, PL1 2PB, UK; ²University of Plymouth, School of Biological Sciences, Drake Circus, Plymouth, PL4 8AA, UK

Biological invasions may considerably alter community composition and ecosystem functioning as well as result in considerable economic loss. Different ecosystems and communities however appear to vary in their susceptibility to invasion, as well differ in how they are affected by the invaders.

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Effect of acetic acid treatment on the epifaunal mussel sock species Mitrella lunata (lunar dove shells) and Caprella spp. (caprellid shrimp) in Prince Edward Island, Canada. Paetzold, S. Christine¹, <u>Davidson, J.</u>² and Giberson, D.¹.¹Department of Biology, ²Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, Prince Edward Island, C1A 4P3 Canada

Acetic acid treatments that are used to control the clubbed tunicate (Styela clava, Ascidiaceae), a fouling pest of mussel lines, may also affect other epifaunal mussel sock species, including potential tunicate predators. In this study, the effect of acetic acid treatment on potential predators of the tunicates, the snail Mitrella lunata (lunar dove shell) and the shrimp Caprella spp. (caprellid shrimp), was investigated. Snail and shrimp population sizes on mussel socks in the field were compared over the short (5d) and long term (5-6wk) before and after treatment and between control socks and treatment socks. Treatment consisted of either lifting the socks or lifting and spraying them with 5% acetic acid. The direct effect of acetic acid on both species was also studied in the laboratory. Snail populations were not affected by acid treatment in the short term, but in the long term, acid-sprayed populations were significantly lower than untouched control populations. Lifting of the mussel socks decreased snail populations, but this effect was not statistically significant. Acid treated shrimp populations were lower than those on socks that were simply lifted from the water in both the short term and the long term. In the laboratory, acetic acid spray killed all shrimp whereas snails were mostly unaffected by the treatment. Snails were more affected by the lifting process than shrimp. Further studies are necessary to evaluate the timing of acetic acid treatment with respect to the life cycle of the two species.

A preponderance of tunicates: observations from rapid assessment surveys. <u>Pederson. Judith</u>¹, Bullock. R², Calder, D.³, Carlton, J.⁴, Chapman, J.⁵, Cohen, A.⁶, Dean, H.⁷, Drynda, P.⁸, Fisher, R.⁹, Harris, L.¹⁰, Hobbs, N.², Lambert, C.¹¹, Lambert, G.¹¹, Laso-Wasem, E.¹², Mathieson, A.¹⁰, Miglietta, M.¹³, Smith, J.¹⁴, Tyler, S.¹⁵ and Winston, J.¹⁶ ¹M I T Sea Grant College Program, Cambridge, Massachusetts; ²University of Rhode Island, Kingston, Rhode Island; ³Royal Ontario Museum, Toronto, Canada, ⁴Williams College-Mystic Seaport, Mystic, Connecticut; ⁵Oregon State University, Newport, Oregon; ⁶San Francisco Estuary Institute, San Francisco, California; ⁷Museum of Comparative Zoology, Cambridge, Massachusetts; ⁸University of Wales, Swansea, Wales, UK; ⁹University of Massachusetts, Dartmouth, Massachusetts; ¹⁰University of New Hampshire, Durham, New Hampshire; ¹¹Friday Harbor Laboratory, Friday Harbor, Washington; ¹²Yale University, New Haven, Connecticut; ¹³Duke University, Durham, North Carolina; ¹⁴Winthrop University, Rock Hill, South Carolina; ¹⁵University of Maine, Orono, Maine; ¹⁶Virginia Museum of Natural History, Virginia

Rapid assessment surveys of fouling communities on floating docks and piers were conducted during August, 2000 and 2003 in the northeast from Maine through New York City. Taxonomic experts identified native and non-native major macro-alga and macro-invertebrate species at marine locations throughout the coast. In the 2003 New England survey a total of 340 species was identified of which 28 were introduced and 33 were cryptogenic species. For the 2000 Massachusetts and Rhode Island survey, a total of 302 species was identified, of which 32 were introduced and 37 were cryptogenic. A total of 13 ascidians was found with only the cryptogenic species, *Molgula provisionalis* being absent south of New Hampshire. Comparison of tunicate distribution data from locations sampled in Massachusetts in 2000 and 2003 were remarkably similar among all the sites for both years.

Spatial distributions of tunicates in marinas are compared to those in major ports of the Northeast region and discussed in the context of probable vectors influencing these distributions. Preliminary information on tunicates reported from an August 2004 survey of southern England highlight geographic differences between the east and west coasts of the North Atlantic. Despite the long history of marine transport between southern England and the northeast, far fewer species are found in the northeast even though ascidians have been successful invaders.

Competition among invading ascidians and a native mussel. <u>Rajbanshi, Rubi</u>¹ and Pederson, J.². ¹475 Park Avenue South, New York, NY; ²MIT Sea Grant College Program, Cambridge, MA

For sessile organisms, space is a limiting factor in marine ecosystems where dominance of competing species is related to temporal, physical and biological factors. Gaining insights into how introduced species use space may lead to predicting which species can successfully establish, persist, and expand to new areas. Competition among the introduced compound ascidian. Botrylloides violaceous and the native blue mussel. Mytilus edulis, and two solitary introduced ascidians, Ascidiella aspersa and Ciona intestinalis, were followed on deployed PVC plates in Boston Harbor, Massachusetts. The results indicate that the species competed directly with each other using different strategies. Botrylloides violaceous had a significant advantage in using space due to its consistent growth and ability to overgrow the other three species. Mytilus edulis also had an advantage because it was able to move around on the substrata, appeared to recruit multiple times during the year, and grew quickly in size. The solitary tunicates, A. aspersa and C. intestinalis, differed in their recruitment period, growth patterns, and space utilization from the *B. violaceous* and *M. edulis*, indicating that the strategies they used to occupy space were not as effective in the presence of other competitors. In the short term, they appear to have recruitment advantages. For the solitary ascidians, the number of surviving adults is less important to space occupation than the time of recruitment. Improved understanding of invasion processes can assist with managing introductions in marine ecosystems.

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Botrylloides violaceus is less vulnerable to sea urchin predation than a dominant native ascidian, *Aplidium glabrum*.

Simoncini, M.K. and Miller, R.J. Biology Department, University of Massachusetts, Boston, MA

Subtidal benthic communities typically show distinct patterns of community structure related to substrate angle. Suspension feeding invertebrate communities often dominate vertical and undercut rock surfaces, while macroalgae dominate horizontal surfaces. One factor that may shape this pattern is sea urchin grazing, which can be more intense on horizontal surfaces. The native Gulf of Maine (GOM) ascidian *Aplidium glabrum*, like other native ascidians, is generally restricted to vertical and undercut surfaces, whereas the introduced ascidian, *Botrylloides violaceus*, is often abundant on horizontal surfaces. We tested the hypothesis that this pattern is partly due to differing predation intensity on these two ascidians by *Strongylocentrotus droebachiensis*, an important omnivore in the GOM. Feeding preference of *S. droebachiensis* on the native *A. glabrum* vs. *B. violaceus* was estimated in the laboratory and in field experiments. Results show that *S. droebachiensis* prefer to feed on the native ascidian distribution in the GOM, and ultimately help explain observed patterns in ascidian distribution in the GOM, and ultimately help explain the overall success of *B. violaceus* as a major invader in New England.

A molecular approach to exploring bacterial associations with ascidian species of Woods Hole, Massachusetts.

Elia Tait^{1,3}, Carman, M.² and Sievert, S.M¹. ¹Biology Department, and ²Department for Geology and Geophysics, Woods Hole Oceanographic Institution, Woods Hole, MA³ Drew University, Madison, NJ

Recent studies have revealed that many marine invertebrates are closely associated with diverse microorganisms, frequently resulting in the production of compounds of biomedical interest. Thus far, ascidians have not been widely examined for the presence of bacterial associations, although the production of secondary metabolites is well documented. In this study, we examined the gonad tissue of *Molgula manhattensis* and the surface tissue of *Botryllus schlosseri*, *Didemnum* sp., and *Botrylloides violaceus* for the presence of associated bacteria. We used denaturing gradient gel electrophoresis (DGGE) as well as 16S rDNA cloning and sequencing to analyze the microbial communities. A single spiroplasma-like phylotype was found to dominate the community in the *M. manhattensis* gonads. The communities of the external tissue samples were found to be much more diverse, with each ascidian species having a unique population profile, predominated by alpha-proteobacteria. This study demonstrated that ascidian species are associated with diverse bacterial populations and future studies will aim to elucidate the precise relationships between bacteria and ascidians and to identify bioactive compounds that might be produced as a result of these relationships.

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Spread of *Microcosmus squamiger* in the Mediterranean Sea. <u>Turon, Xavier</u>¹ and Nishikawa, T². ¹ Dept. of Animal Biology, University of Barcelona, 645, Diagonal Ave., 08028 Barcelona, Spain; ² Nagoya University Museum, Chikusa-Ku, Nagoya 464-8601, Japan

Microcosmus squamiger Michaelsen, 1927 is a species that inhabits shallow rocky littoral habitats, particularly in bays and harbours. It is probably of Australian origin. This solitary ascidian has a great invasive potential, and its expansion in southern California harbours and marinas since the eighties is well documented. In the Mediterranean, its presence has been noted since the seventies, but it has been repeatedly confused with *Microcosmus exasperatus* Heller, 1878 with the result that, to our knowledge, it has been formally cited only once in the Mediterranean.

To ascertain the spread of Microcosmus squamiger and M. exasperatus in the Mediterranean, we have revised the material in the collection of the Museum National d'Histoire Naturelle, Paris, we have sampled several points in the western Mediterranean, and we have checked the true identity of specimens classified as M. exasperatus by several authors. The results show that specimens unambiguously attributable to M. squamiger are common in Spain, France, Italy and Morocco. They can form dense aggregations in harbours, bays, and shallow littoral habitats. In contrast, we have identified only a few specimens of *M. exasperatus* from the eastern Mediterranean, in the Lebanese coast. Moreover, we also found *M. sauamiger* for the first time for the Atlantic shores of Spain and identified specimens of this species from Madeira. It seems, therefore, that *M. squamiger* has greatly spread throughout the western Mediterranean, and there are also well-established Atlantic populations by now. *M. exasperatus*, on the other hand, has a much more restricted distribution in the Mediterranean, and is probably a Lessepsian migrant, as we have identified material of this species from Suez and the Gulf of Aden. The ability of *M. squamiger* to form dense, monospecific crusts that outcompete native species, and the finding that the species can be abundant in some habitats outside harbours and marinas indicate that this species is a potential threat to Mediterranean littoral communities. This instance illustrates the crucial importance of taxonomy in studies of invasive species.

Do invasive fouling organisms have greater recruitment and abundance on artificial substrates than on naturally occurring substrates?

<u>Tyrrell, Megan C.</u>¹ and Byers, J.E.². 'Wells National Estuarine Research Reserve, Wells, ME; ²Zoology Department, University of New Hampshire, Durham, NH

Invasive species are prominent constituents of fouling communities. If invasive fouling organisms are able to colonize a wider range of substrate types than native species, it may partially explain their high abundance in fouling communities. We used four natural and four artificial substrates to investigate how substrate type influences the recruitment and abundance (percent cover) of fouling organisms in the Gulf of Maine. There was a pattern of higher richness of introduced and cryptogenic species on the artificial substrates than on the natural substrates, but this difference was not statistically significant. A total of four tunicate species were found during the course of the experiment. Two of these, *Botrylloides violaceus* and *Botryllus schlosseri* are introduced. These two tunicate species recruited to all substrate types and, after the first analysis in July, they consistently ranked second and third in percent cover over all substrate types. A repeated measures analysis of variance indicated that there was no difference in the percent cover of either *B. violaceus* or *B. schlosseri* on natural versus artificial substrates for the time periods analyzed. Nevertheless, by the last analysis in November, the abundance of *B. violaceus* was three times higher on polyvinychloride than on the any other substrate type.

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The invasive tunicate *Didemnum* sp. on Georges Bank – ecological observations and potential effects on groundfish and scallop fisheries.

Valentine, Page C.¹, Collie, J.S.², Reid, R.N.³, Asch, R.² and Guida, V.G.³.¹U.S. Geological Survey, Woods Hole, MA; ²University of Rhode Island, Narragansett, RI; ³NOAA Fisheries, Sandy Hook, NJ

Colonization of a part of the Georges Bank fishing grounds by the invasive colonial tunicate *Didemnum* sp. first became apparent in 2002 in an area open to groundfish trawling and scallop dredging on the Northern Edge of the bank near the U.S./Canada boundary. This is the first offshore record of a species that has colonized the U.S. east coast from New York to Maine. The benthic fauna of this area has been monitored since 1994. Video and sampling surveys in 2003 and 2004 documented the tunicate over an area of more than 40 square miles. At some sites colonies coalesce to form large mats that cover more than 50 percent of the seabed. The affected seabed is a pebble and cobble pavement that lies at water depths of 45 to 60 meters. The water column is mixed year round, ensuring a constant supply of nutrients; annual temperatures range from 4 to 15 degrees C. The gravel is bounded on the east, west, and south by sand ridges of mobile sediment that move daily in strong tidal currents that exceed 1 knot. Local distribution of *Didemnum* sp. indicates that it cannot survive in a mobile sand habitat.

It is possible that the mats of *Didemnum* sp. colonies affect the gravel habitat and species that are adapted to it. The tunicates grow over sea scallops, mussels and attached epifauna such as hydrozoa and bryozoa. The mats cover infaunal bivalves and worms. A sample containing 420 g of *Didemnum* sp. contained 15 polychaete species and 7 bivalve species living beneath the tunicate mat. Upper surfaces of tunicate colonies appear to be very clean, with no attachment by other species; and it is possible that larvae of benthic species, including sea scallops, will not be able to settle there. Further study is required to determine if tunicate mats can reduce bottom roughness, accessibility to prey by fish, and space available for larval attachment.

Compositional analyses of the benthic community in the affected area from 1994 to 2003, compared with non-affected control sites, show that the polychaete *Nereis zonata* increased at the affected site in 2003. In contrast, the hermit crab, *Pagurus acadianus*, increased at non-affected sites while decreasing slightly at sites with high tunicate cover. Stomach contents of three groundfish species (haddock *Melanogrammus aeglefinus*, winter flounder *Pseudopleuronectes americanus*, and longhorn sculpin *Myoxocephalus octodecemspinosus*) caught in the affected area in 2004 contained undigested fragments of *Didemnum* sp. Analyses of colony tissue suggest it has poor nutritional value. Gravimetric and biochemical analyses of a single colony (20 x 30 cm) yielded 37% tissue ash, 30% sand and silt, and 5% tunicin (cellulose) by dry weight, leaving a maximum of 28% with potential energetic value: 3% lipids, <0.5% non-cellulose carbohydrates, and the remaining 25% presumably protein.

Colonial tunicates reproduce sexually with larvae that live only a few hours. After larval attachment to a firm substrate, colonies enlarge by asexual budding. Experiments in a Cape Cod tide pool with *Didemnum* sp. show that fragments of colonies re-attach and grow rapidly by budding. It is likely that the species will spread on Georges Bank most rapidly by the transport of colony fragments that are ripped from the seabed by fishing trawls and dredges and natural processes. Habitats of firm substrates are most vulnerable to colonization, while habitats of moving sediment will be unaffected.

Post-settlement predation on ascidian recruits: Predator responses to changing prey density.

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In order to better understand the nature of factors contributing to sessile marine species mortality, we assessed the role that two species of small predaceous neogastropods (Mitrella lunata and Anachis lafresnavi) have on mortality patterns of different species of ascidian recruits. By using the functional response approach (i.e., predator responds to variations in prey density) we were able to obtain comparative information on the survival and potential local persistence patterns of several species solitary and colonial ascidians. In addition, we were able to compare ascidian species which were relatively recent invaders (e.g. last ~30 yrs) into southern New England to those species which have been resident in the region for $\sim 150 + \text{ vrs}$. Ascidian recruits examined included both solitary (Ciona intestinalis, Styela clava, Molgula manhattensis, Ascidiella aspersa) and colonial (Botryllus schlosseri, Botrylloides violaceus and Diplosoma listerianum) forms. These studies indicate that both species of snails displayed fairly generalized prey preferences and the only recruits that they did not readily consume were *Botrylloides*. When recruit densities were manipulated over a 1 to 2 order range of abundance, predator consumption rates differed between prey species. Proportional mortality did not significantly differ with any prey density for Styela and Botrylloides. For most of the other species, proportional mortality varied inversely with increasing prey abundance, indicating Type II functional responses. The experiments suggest that the predators are capable of effectively eliminating the prey regardless of naturally occurring variations in ascidian recruitment densities. Collectively, these results indicate that the snails can be critical in regulating ascidian recruitment dynamics in southern New England.

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