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ASCIDIANS OF SOUTH PADRE ISLAND, TEXAS, WITH A KEY TO SPECIES

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Abstract.–The ascidians of South Padre Island, Texas were surveyed in August 2004. Because the subtidal area is limited to soft sediments, the survey was restricted to marina floats and pilings, harbor buoys, boat hulls and other artificial substrates which offer suitable attachment surfaces for ascidians. Fifteen species were documented, with multiple species representing each of the three orders of ascidians. None of the species found in this survey are native, suggesting they were all introduced through boat traffic. About half the species were found in a reproductive state, however, indicating that they have established local breeding populations.

Ascidians are marine invertebrate chordates, some of which are classic model organisms for the study of development and evolution (Conklin 1905; Berrill 1932; Satoh 1994; Corbo et al. 2001). They are emerging model organisms for other fields, including genetics (Dehal et al. 2002; Satoh et al. 2003), immunology (Azumi et al. 2003; Khalturin et al. 2003; Du Pasquier 2004; Rinkevich 2004), and neurobiology (Meinertzhagen & Okamura 2001; Meinertzhagen et al. 2004). Ascidians are also attracting attention as potential bio-indicators of environmental health (Cima et al. 1995; Cima et al. 1997) and as seafood, particularly in Japan and Korea (Sawada et al. 2001). Ascidians are efficient filter feeders, and certain species with wide environmental tolerances have become highly invasive, especially in bays and harbors where they compete with and overgrow commercial shellfish (Lesser et al. 1992; Carver et al. 2003) and create a significant fouling community on boat hulls and marina floats (Teo & Ryland 1995; Hodson et al. 2000; Lambert 2001; 2002; Lambert & Lambert 2003). Thus, locales with high ascidian populations hold great potential for scientific and commercial research.

Most ascidian species require a hard substrate for attachment. The natural subtidal substrates along most of the Texas coast are composed of soft sediments. Thus, prior to the establishment of man-made substrates (marina floats, pilings, harbor buoys and boat hulls), few shallow-water ascidians were recorded from the Texas Gulf coast (Van Name 1945; Whitten et al. 1950; Van Name 1954). Informal observations indicate that the south Texas coast may support ascidians in greater abundance than the rest of the Texas coastline. This paper lists the 15 species observed during a recent survey around South Padre Island, their locations and abundance, and includes a taxonomic key to species.

METHODS

Individuals were collected from the waters of the Laguna Madre around the southern end of South Padre Island, Texas, on 7-8 August 2004. Collection locations were identified using the global positioning system (GPS). Figure 1 shows the six collection sites: (a) Sea Ranch marina (26° 4’ 33.4” N, 97° 9’ 52.8” W); (b) Parrot Eyes marina (26° 8’ 0.4” N, 97° 10’ 36.9” W); (c) Laguna Madre boat canal mid-channel buoy (26° 4’ 1.2” N, 97° 10’ 0.6” W); (d) the Coastal Studies Lab seawater intake support (26° 4’ 4.9” N, 97° 9’ 49.1” W); (e) Port Isabel deep water docks (26° 3’ 30.0” N, 97° 12’ 49.4” W), and; (f) Billy Kenan’s dock (26° 3’ 56.8” N, 97° 12’ 54.6” W).

Specimens were initially examined live under dissecting microscopes, with further examination of some species after preservation. Representative individuals were fixed either directly in 70% ethanol or relaxed in seawater containing a few drops of a concentrated menthol/ethanol solution, and then preserved in 10% seawater formalin buffered with sodium borate.

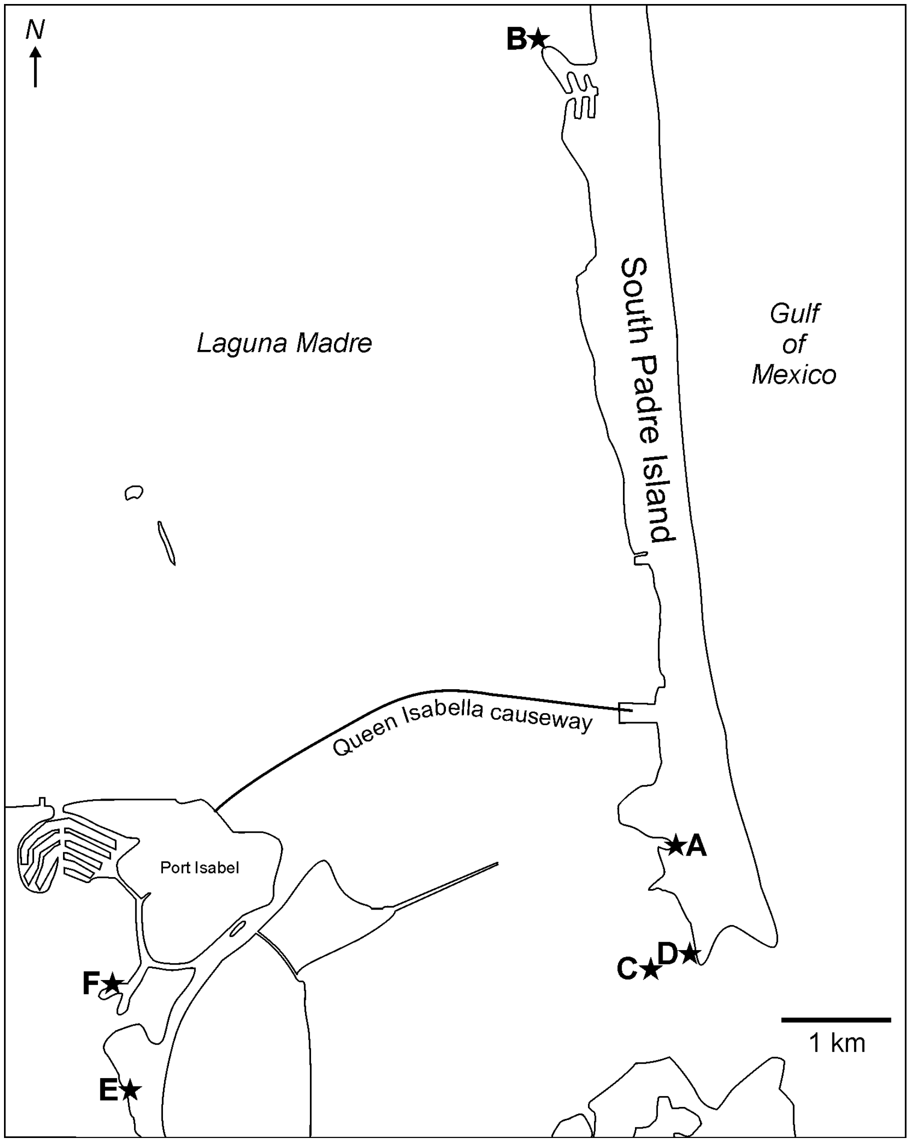

Figure 1. Map of South Padre Island area, Texas, showing collection sites of ascidians. a = Sea Ranch marina; b = Parrot Eyes marina; c = Laguna Madre boat canal mid-channel buoy; d = Coastal Studies Lab seawater intake support; e = Port Isabel deep water docks; f = Billy Kenan's dock (sites listed in order visited).

Specimens were identified at least to genus level. The primary sources used for identification were (Van Name 1945; Plough 1978); *Didemnum duplicatum* was identified from (Monniot 1983). Labelled voucher specimens were deposited in the Coastal Studies Laboratory on South Padre Island.

Table 1. Systematic listing of species collected. Locations as given in Methods and Figure 1.

Taxon	Location(s) collected	Solitary or Colonial	Reproductive Statis during Survey
Phylum Chordata			
Subphylum Tunicata			
Class Ascidiacea			
Order Aplousobranchia			
Family Didemnidae			
Didemnum psammathodes	b	Colonial	Not productive
Didemnum duplicatum	b, c	Colonial	Brooded larvae
Diplosoma listerianum	b	Colonial	Brooded larvae
Lissoclinum fragile	a, b	Colonial	Not productive
Family Polyclinidae			
Polyclinum constellatum	b, e	Colonial	Not productive
Family Clavelinidae			
Clavelina oblonga	d	Colonial	Brooded larvae
Order Phlebobranchia			
Family Perophoridae			
Perophora sp.	a, e	Colonial	Not productive
Family Ascidiidae			
Ascidia interrupta	a	Solitary	Not productive
Order Stolidobranchia			
Family Styelidae			
Botrylloides nigrum	b, e	Colonial	Not productive
Botrylloides sp.	c, e, f	Colonial	Brooded larvae
Polyandrocarpa zorritensis	e, f	Colonial	Not productive
Styela canopus	a, b, e	Solitary	Ripe gonads
Styela plicata	a, b, e, f	Solitary	Ripe gonads
Symplegma viride	e	Colonial	Not productive
Symplegma rubra	a, e, f	Colonial	Brooded larvae

RESULTS

Fifteen species of ascidians were identified in this survey (Table 1). *Styela plicata*, *S. canopus*, and *Lissoclinum fragile* were particularly abundant, with *S. plicata* being found in large numbers at four of the six collection sites. Several other species that are small or inconspicuous may also be more common or abundant than indicated by this survey. Seven species were reproductive at the time of the survey, indicating that these species have formed locally reproductive populations. *Ascidia interrupta*, though rare during this survey, is abundant in autumn and is reproductive during that time.

KEY TO SPECIES

"There are some groups of animals for which keys can be made that really work in a considerable number of instances, but the ascidians are not among them" (Van Name 1945). This key is specific for the organisms seen or previously collected in these bays but is not necessarily valid for other regions. It is based on a 7-9 August 2004 survey of South Padre Island; there may be additional species more abundant at other times of the year that are not included here. An asterisk (*) indicates species not found during this survey but which are expected due to their distribution: *Ciona intestinalis* has a cosmopolitan distribution, and *Molgula manhattensis* has been recorded elsewhere in Texas.

Explanations of terms, species descriptions, and illustrations can be found in Van Name (1945) or Plough (1978).

1. Solitary ascidians; each zooid enclosed in its own tunic......... 2
   Colonial ascidians; multiple zooids within a common tunic or connected by stolons... 6

2. Branchial sac without internal longitudinal folds.................... 3
   Branchial sac with four or more prominent internal longitudinal folds... 4

3. Body wall (easily visible inside smooth transparent tunic) with five to seven white wide longitudinal muscle bands on each side (often somewhat contracted in fixed animals); animal elongate, flaccid, attached basally .... *Ciona intestinalis**
   Body wall muscles in a meshlike pattern mostly on right (uppermost) side but not as above; animal attached broadly on left side, tunic semi-transparent, thin and not smooth .... *Ascidia interrupta*

4. Tunic thin, semi-transparent but usually muddy; body spherical, 2-4 cm in diameter; oral siphon with six lobes, atrial siphon with four lobes; six branchial folds per side ....*Molgula manhattensis**
Tunic leathery; four branchial folds per side........................... 5

5. Tunic brownish, furrowed; body usually 2-3 cm in height; siphon tips with numerous mottled reddish stripes; two long slender ovaries/side; testes large, white, often bifurcated, attached to posterior end of ovaries by long threadlike sperm ducts .... *Styela canopus*
Tunic white with large rounded soft lumps; body up to 10 cm in height; siphon tips with four black stripes; two gonads on left side, five on right; testes small and attached along most of the length of each ovary ....*Styela plicata*

6. Multiple zooids connected by stolons, each zooid enclosed by separate tunic 7
Multiple zooids all embedded in common tunic 9

7. Zooids spherical or up to twice as long as wide..................... 8
Zooids over four times as long as wide, transparent, colorless . ...*Clavelina oblonga*

8. Tunic soft and fragile, zooids globular, pale green, translucent, 2-4mm in height; branchial sac with four rows of stigmata ... . *Perophora* sp. (probably *P. viridis*)
Tunic tough and leathery, zooids elongate, dark brown or purple, up to 2 cm in height; stolons usually coalesced into a basal mat; branchial sac with more than four rows of stigmata ....*Polyandrocarpa zorritensis*

9. Zooids not divided into body regions; vascular ampullae present in tunic 10
Zooids divided into two or three distinct regions; vascular ampullae absent in tunic 13

10. Zooids (2.5-4 mm) flat, never organized in systems, widely spaced with clear tunic between, both siphons open at colony surface 11
Small zooids (<2 mm), organized in systems, only branchial siphon opens to surface of colony, densely spaced with little tunic between.. 12

11. Zooids red, tunic opaque ....*Symplegma rubra*
Zooids translucent with greenish or multicolored flecks of pigment..*Symplegma viride*

12. Zooids in elongate systems, colony a single color, usually purple or orange, vertically oriented in tunic, testis ventral (on side with incurrent siphon) and anterior to single ovary, stomach lobes bulbous at ends..
....*Botrylloides nigrum*
Zooids in elongate systems, two colors in colony, dark basic colony color, bright yellow around siphonal area..................
.... *Botrylloides* sp.

13. Zooids with two body regions (thorax, abdomen), colony thin and encrusting, zooids with four rows of stigmata 14
Zooids with three body regions (thorax, abdomen, post-abdomen), colony dark, thick and encrusting, may be dome shaped, zooids in circular systems, each zooid with 14-18 rows of stigmata ....*Polyclinum constellatum*

14. Tunic with tiny (visible with compound microscope) white spherical calcareous spicules with many short pointed rays, mostly in surface layer of colony...................................... 15
Colony lacking calcareous spicules though there may be considerable white pigment granules; tunic transparent, very flaccid, zooids tiny (2-3 mm in length) usually with black pigment on thorax and abdomen.........*Diplosoma listerianum*

15. Atrial opening small or moderate size; sperm duct spirally coiled, colony not white and easily torn 16
    Atrial opening large, exposing most of branchial walls; sperm duct not spirally coiled, colony white, tunic very fragile and easily torn ....*Lissoclinum fragile*

16. Colony distinctly muddy gray colored due to numerous fecal pellets stored in the tunic .... *Didemnum psammathodes*
    Colony salmon colored, leathery, with meandering dark lines . ...*Didemnum duplicatum*

DISCUSSION

A diverse assemblage of ascidian species is present in considerable abundance along the southern Texas coastline. All of the species found in this survey are apparently non-native and have most likely been introduced on boat hulls. All have been recorded elsewhere in the Gulf of Mexico, on the Atlantic side of Florida, or various regions of the Caribbean as well as other warm water regions of the world (Lambert 2001; 2002). All are shallow-water species not recorded in the survey of (presumably native) deep-water ascidians of the Gulf of Mexico (Monniot & Monniot 1987), though a few were recorded from continental shelf depths of the Gulf (Plough 1978). Given that five colonial species contained brooded larvae, and two of the three solitary species had ripe gonads, it seems likely that many or most of the species found have formed breeding populations in the local waters.

The species sampled include more than one member of each of the three orders in class Ascidiacea, providing substantial diversity for comparative research. Indeed, the prospects for future research on ascidians in this area are extremely good. Many of the genera found on South Padre Island have been the focus of substantial research. For example, the natural pigmentation of *Styela* embryos enabled classic studies of chordate development (Conklin 1905; Gehring 2004). Colonial tunicates like *Botrylloides* are now model organisms for allorecognition and the evolution of immune responses (Scofield et al. 1982; Scofield & Nagashima 1983; Rinkevich 1995; Hirose et al. 1997; Paz & Rinkevich 2002; Rinkevich 2004). Several of the ascidian genera on South Padre Island have been the source of many novel chemical compounds, including some with possible therapeutic properties, including *Didemnum* (Kang & Fenical 1997; Smith et al. 1997; Davis et al. 1999; Mitchell et al. 2000; Oku et al. 2003), *Lissoclinum* (Badre et al. 1994), *Styela* (Lee et al. 1997a; Lee et al. 1997b; Zhao et al. 1997) and *Symplegma* (Lindsay et al. 1999).

The collecting sites are conveniently located near a well-equipped research and teaching laboratory (Coastal Studies Laboratory, University of Texas-Pan American). All the species described here should be easily maintained alive in the large seawater tanks, especially if placed in floating plastic sieves or grown on glass plates, or easily collected for same-day use. Most of the species have long breeding seasons and are easy to remove gametes from (for solitary species) or brooded embryos (for colonial species). Development of solitary species is very rapid (less than 24 hours to hatching) and the larvae of all ascidians are short-lived and non-feeding, allowing metamorphosis and post-metamorphic events to be followed easily. The readily available ascidians of South Padre Island also provide highly suitable material for classroom use in a number of teaching areas.

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