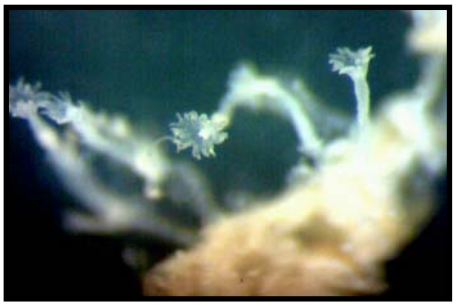

BIOFOULING ON THE VESSELS

PRIDE, SCAN, AND CAPE CHARLES

AT THE JAMES RIVER RESERVE FLEET AND

AFTER TOWING TO TEXAS

Prepared for

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Washington, D.C.

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EXECUTIVE SUMMARY

As part of its non-retention vessel disposal program, the U.S. Maritime Administration oversees transfers of ships from reserve fleet locations to ship-breaking facilities. These vessels may pose a high risk of hull-mediated invasions because their underwater surfaces can be heavily fouled by aquatic organisms, and many of the vessels have a long residence time at their destination ports before they are dismantled. As a result, the Maritime Administration has implemented in-water hull cleaning as one management option to reduce the risk of transferring nonindigenous species to new coastal regions where they may become established.

The extent of biofouling on the vessels PRIDE, SCAN and CAPE CHARLES was examined at the James River Reserve Fleet, Virginia. This study is one in a series that examines the biological growth on obsolete vessels and evaluates the effectiveness of in-water hull cleaning as a vector management option. The vessels were surveyed prior to hull cleaning, after hull cleaning, and after transit from the James River to Brownsville, Texas, where the ships were dismantled.

The sampling design for this study was similar to that implemented on previous biological surveys. Fifty samples per sampling iteration were collected by divers from the underwater surfaces of the vessels using a stratified random sampling design consisting of transects and starboard to port locations within transect. At each location, a 6-inch (15.2 cm) diameter PVC sampler was used to scrape approximately a 182 cm^2 area of the hull. Samples were stored in cloth bags, examined and photographed at the fleet, and transferred to the laboratory for sorting, enumeration, and identification of organisms.

The biological samples were accompanied by underwater photographs of the biota (photo-quadrats) and video. The system used for the photo quadrats consisted of an underwater camera and a “clear-water box” that provided a standard image area for all photographs. Samples were analyzed for differences in species abundance and composition across ships, surveys, transects, and locations within transect using univariate and multivariate analyses. Photo-quadrats were analyzed by the point-count method to determine percent cover of biofouling species (mussels, barnacles, hydroids, algae, etc.) and bare hull. Videos were examined to characterize type and extent of coverage.

Across all ships and surveys 112 taxa were found, 70 of which were identified to species level. Barnacles, mussels, amphipods, polychaete worms, mud crabs, sea squirts, hydroids, and bryozoans predominated and occurred frequently in the samples. The biofouling community was numerically dominated by the barnacle *Balanus improvisus* and the amphipod *Apocorophium lacustre*. These two species accounted, respectively, for 67% and 10% of total abundance. Other abundant species were the Hooked Mussel, *Ischadium recurvum*, the amphipods *Incisocalliope aestuarius* and *Melita nitida*, the polychaete worm *Neanthes succinea*, the sea squirt *Molgula manhattensis*, and the mud crab *Eurypanopeus depressus*. Among the colonial species, the hydroid *Blackfordia virginica* and the bryozoan *Conopeum chesapeakensis* were dominants.

Of all the taxa collected in the pre-cleaning and post-cleaning surveys, 39 were native to Chesapeake Bay, 4 were cryptogenic (i.e., of uncertain origin), and 4 were introduced. The introduced nonindigenous species were the hydroids *Blackfordia virginica* and *Garveia franciscana*, the isopod crustacean *Synidotea laevidorsalis*, and the Oriental Shrimp, *Palaemon macrodactylus*. This is the first record of *P. macrodactylus* in the Chesapeake Bay and the U.S. east coast. This notorious invasive species from east Asia was first noticed in San Francisco Bay in the 1950s, and has now spread in North America throughout much of the west coast.

The total number of species differed among surveys. In addition, there were significant differences in abundance among surveys, and significant differences in the mean number of species per sample among vessels and surveys. Densities and species numbers were lower in the post-cleaning surveys than in the pre-cleaning surveys. However, there was an increase in total species number, mean species numbers, and mean densities in the post-transit surveys of the SCAN and the CAPE CHARLES relative to the post-cleaning surveys. These increases were due to 'new' taxa found only in the post-transit surveys, suggesting attachment and growth of newly settled species while the ships were en route to Texas. Forty-two percent of all taxa representing at least 43 distinct species were 'new' taxa. This is a significant finding because species attachments on hulls augment the risk of species transfers and introductions.

In-water hull cleaning was successful at removing on average 91-97% of the biofouling cover, substantially reducing densities or organisms and species frequencies. However, hull cleaning was not particularly effective at removing species. Seventy-five percent of all the taxa found in the pre-cleaning surveys were found in the post-cleaning surveys, and 52% persisted on the hulls of the vessels during towing. Three species unknown to occur in Texas, the bryozoan *Alcyonidium* cf. *albescens*, the sea anemone *Diadumene leucolena*, and the amphipod *Incisocalliope aestuarius* were transferred on the hulls. None were invasive in the Chesapeake Bay.

There was evidence of large inter-annual variability in biofouling when the data were compared to those of the MISSISSINEWA, which was surveyed one year earlier in the same season (winter). The number of species collected from the PRIDE, SCAN, and CAPE CHARLES in the James River were about twice the number of species recorded from the MISSISSINEWA. Several common species in the present study, for example *Blackfordia virginica*, *Molgula manhattensis* and *Incisocalliope aestuarius*, were absent from the MISSISSINEWA. It is recommended that some characterization of extent and type of biofouling be conducted each year, and at different seasons, to capture the full range of biofouling and its potential to harbor invasive species.

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1.0 INTRODUCTION

As part of its non-retention vessel disposal program, the U.S. Maritime Administration oversees transfers of ships from National Defense Reserve Fleet (NDRF) locations to ship-breaking facilities. The vessels are towed from their fleet to other geographic locations where ship breaking takes place. Because many vessels have been laid up for long periods of time, their underwater surfaces can be heavily fouled by aquatic organisms and their transfer may create a risk of biological invasion at destination ports. This report is the fifth in a series that documents the biofouling of obsolete vessels, as well as the effectiveness of in-water hull cleaning as a vector management option.

In this study, the extent of biofouling on the hull of three James River Reserve Fleet (JRRF) vessels, PRIDE, SCAN, and CAPE CHARLES is examined. The vessels were transferred from the James River, Virginia, to Brownsville, Texas, where the ships were dismantled. Because of the inter-oceanic nature of the transfer, its biogeographical implications are significant. The biological characterization was conducted prior to hull cleaning, after hull cleaning, and after transit from Virginia to Texas. As in previous surveys, the objectives were to 1) identify and quantify the biota associated with the underwater surfaces of JRRF vessels, 2) describe differences in biofouling between the pre-cleaning, post-cleaning, and post-transit biological surveys, and (3) examine the biogeographic status and distribution of species with respect to their possible transfer from the JRRF to Texas.

The PRIDE and SCAN were built in 1960 and 1961, respectively, for the Moore-McCormack company as combination of freight and passenger vessels. They entered the JRRF in February 1977. The CAPE CHARLES was launched in 1963 as a freighter, and entered the JRRF in July 1984. All three vessels were scrapped at the Marine Metals, Inc., Brownsville facility in the Spring of 2008.

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2.0 METHODS

2.1 WATER CHARACTERISTICS

The salinity in the James River at Fort Eustis, Virginia, where the JRRF fleet is located (Figure 2-1), can fluctuate widely during the year and from year to year over the same season, depending on river flow. Salinity is usually in the tidal freshwater to mesohaline range (0-18 psu). To characterize the environment in the James River at the time of sampling, the salinity, conductivity, temperature, dissolved oxygen, and pH of the water column were recorded. A Yellow Springs Instrument (YSI Inc., Yellow Springs, Ohio) multiparameter probe with automatic temperature and salinity compensation was deployed at approximately 1 meter intervals from the water surface to the bottom of the hull over one tidal cycle. In Brownsville, water quality parameters were measured at the public pier basin, where the vessels were moored for sampling. These data characterized local conditions at the time of sampling, but did not provide information on exposure during the towing of the vessels to their final destination.

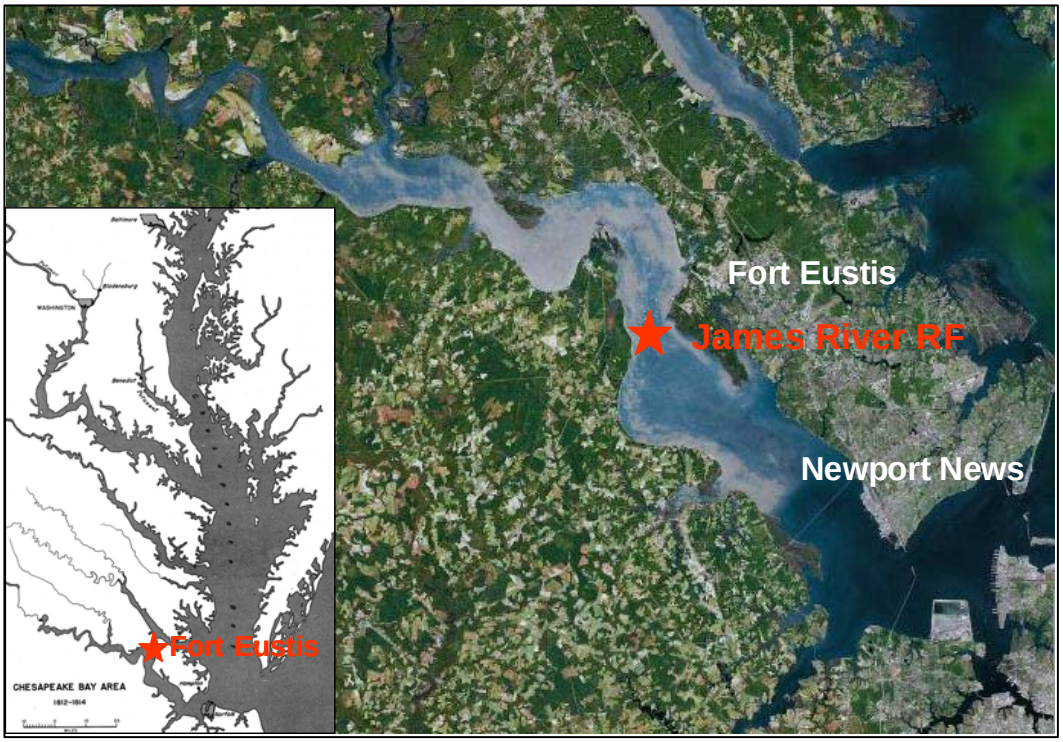

Figure 2-1. Location of the James River Reserve Fleet, Fort Eustis, Virginia

2.2 BIOLOGICAL SURVEYS

Each vessel was surveyed over three separate dives, two in the James River and one in Brownsville. The PRIDE was surveyed between January 4 and January 10, 2008, before and after hull cleaning, and on February 2 after transit from the James River. It departed the JRRF on January 18. The CAPE CHARLES was surveyed between January 12 and January 17, 2008, before and after hull cleaning, and on February 21 after transit. It departed the JRRF on February 5. Lastly, the SCAN was surveyed between February 24 and March 3, 2008, before and after hull cleaning, and on April 2 after transit. The SCAN did not leave the JRRF until March 13. All three vessels arrived in Brownsville in the late afternoon and were sampled early the next day.

Samples were collected with the help of professional divers. Diving was conducted using surface-supplied air and real-time audio and visual communications with the surface team. The surface team included a diver master and two scientists who directed two of the divers toward the locations where samples and photo-quadrats were to be taken. Diving services were provided by Underwater Services International (Gainesville, Florida). The sampling design was similar to that previously employed to survey other vessels (Davidson et al. 2006; Versar 2008a, b, c, d). Samples were taken at three depths (near the waterline, mid-depth, and bottom of the hull) along eight transects (Figure 2-2). The PRIDE and SCAN were 486 feet long, with a lightweight aft draft of 16 feet. The CAPE CHARLES was 495 feet long, with a lightweight draft of 19.6 feet. Transects in all three vessels were positioned 60 feet apart from each other from anchor chain to stern. Five samples were collected per transect: starboard upper, starboard lower, bottom, port lower, and port upper. The first transect near the bow did not have a flat bottom; therefore, only four samples were collected from this transect. Eleven additional samples were taken from the underwater appendages of each vessel, including the stern tube, rudder, propellers, and bilge keel.

At each sampling location, one diver positioned an underwater camera against the surface of the hull and photographed the biota covering the hull. The second diver then collected a sample from a random point within approximately a one-meter radius of the photo-quadrat location. A sampler constructed from a 6-inch (15.2 cm) diameter PVC pipe with a 4-inch adapter to attach the sample bag, was used to collect the biota (Figure 2-3). A diver placed the 6-inch end of the sampler against the hull of the ship and attached a numbered cloth bag to the opposite end. A 3-inch scraper applied between the hull and the sampler was used to remove the biological material from the hull, which was then collected in the sample bag. The PVC sampler was curved at a 45 degree angle, so that the sample would fall straight down into the bag. The bag was twisted closed and tied off before being removed from the sampler to minimize sample loss.

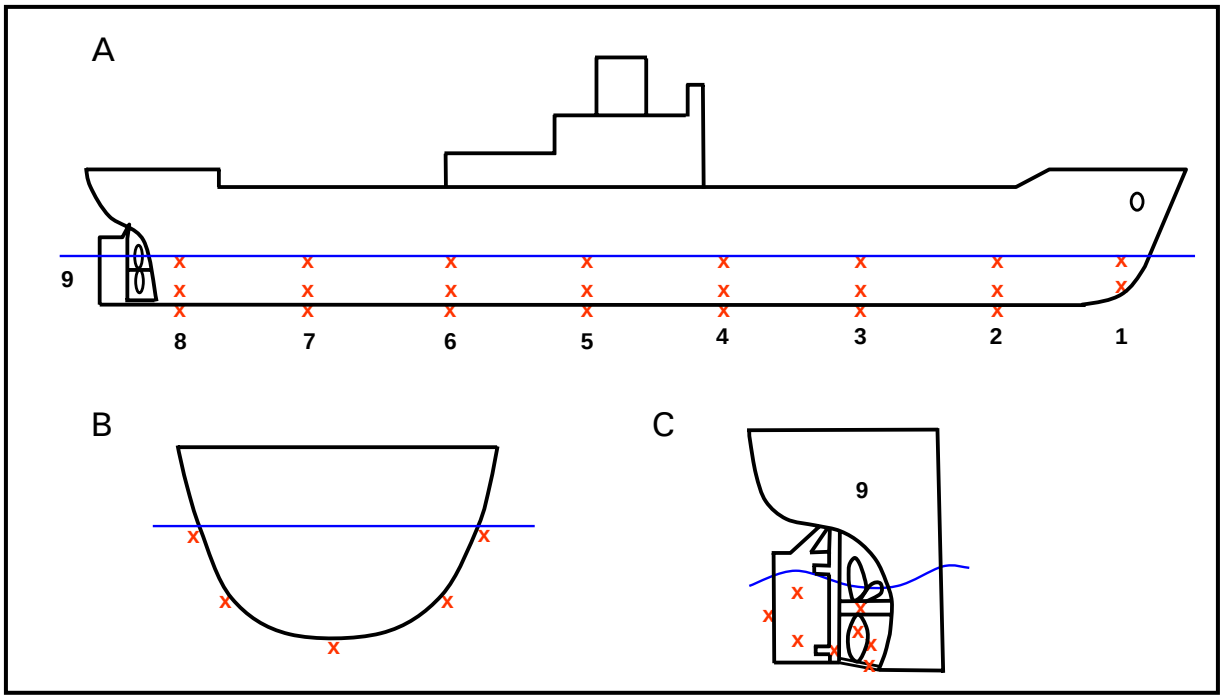

Figure 2-2. Sampling design. Samples and photo-quadrats were taken at 8 transects across the hull of the vessels (A). Five samples per transect were collected: starboard upper, starboard lower, bottom, port lower, and port upper (B). The first transect did not have a flat bottom; therefore, only four samples were collected from this transect. Additionally, samples were collected from the underwater appendages of the vessels (C), labeled as Transect 9.

An area of approximately 182 cm^2 of hull was scraped for each sample. The bag number was relayed to the surface so that detailed notes could be taken on the location at which each sample was collected. Sample bags were stored in a mesh dive bag and returned to the surface, usually in groups of 10 bags corresponding to 2 sampling transects. Upon retrieval, all bags were immediately transferred to 5-gallon buckets with in situ marine water. Protexo bags manufactured by HUBCO (Hutchinson, Kansas) were used. Each bag was made of tightly woven white cotton cloth, and measured 10 x 17 inches (25.4 x 43.2 cm). Each bag included a drawstring that, in addition to a rubber band, kept the bag closed after sample collection. Fifty samples per survey were collected for a total of 150 samples per ship. Two samples were lost during collection and could not be re-taken. A total of 448 samples were processed for species abundance and composition.

The system used for the photo-quadrats consisted of an underwater camera with a "clear-water box" attached to the front of the lens and two strobe lights mounted above the box at 45 degree angles. This system provided a standard image area for all photographs. In addition, the divers carried a video camera that provided real-time visual communication with the surface and video footage of the hull and the associated biota.

Figure 2-3. Sampler constructed from a 6-inch (15.2 cm) diameter PVC pipe with a 4 inch adapter. A diver placed the 6-inch end of the sampler against the hull of the ship, and attached a numbered cloth bag to the 4-inch end. A scraper was used to remove the biological material from the hull, which was then collected in the cloth bag.

2.3 SAMPLE PROCESSING AND TAXONOMY

A visual examination of each sample was carried out in the field. Bags were opened, inverted, and rinsed into a plastic dissecting tray (12 x 18 inches, 2.5 inch deep), and the sample was examined and photographed. The condition of the biota in the samples (potential live versus dead material), and the general kinds and quantity of organisms, were documented. This general procedure was conducted on as many samples as possible. Some samples could not be photographed on site due to time constraints.

After examination, the contents of the tray were carefully poured back into the sample bag, and a label was added to the inside of the bag. Bags were then tightly closed with twist ties and rubber bands, and transferred to a propylene phenoxytol (POP) solution to relax the organisms for easier identification. A 0.15 % solution was made by adding 15 ml of POP to 1 L of warm tap water, and then mixing 9 L of in situ water into the solution (Green and Lambert 1994). After 30-60 min in the relaxant, bags were placed in 1-gallon plastic jars (3-5 bags per jar), and a buffered solution (10%) of formalin in seawater was added to preserve the organisms. In the laboratory, samples were stored in formalin until further processing and identification of organisms.

In the laboratory, samples were washed through nested 250-μm and 64-μm sieves. The finer 64-μm fraction of the sample was retained and stored for later examination. The 250-μm fraction was sorted under dissecting microscopes to separate organisms into major categories (i.e., mussels, barnacles, micro-crustaceans, etc.). Organisms in these major categories were identified to species level whenever possible and counted (non-colonial species only). Some organisms required further examination by specialist taxonomists for identification or confirmation. Voucher specimens of these organisms were placed in separate vials and sent to the specialists.

Due to time constraints, live and dead material were not separated in the field; however, the bulk component of each sample consisted of organisms that were alive at the time of collection. No obvious signs of dead material (e.g., exo-skeletons of crustaceans) were found in the samples upon examination in the field or in the laboratory, except for the empty tests of barnacles.

2.4 ANALYSIS

Samples were analyzed for differences in species numbers, composition, and abundance by transect and location (waterline, mid-depth, bottom, appendages) across the hull of the ship using multivariate analysis methods. Plots were constructed to examine sample configuration and to identify any tendency for samples to form groups according to the location from where they were taken from the hull. Species counts (square-root transformed) were subjected to non-metric multidimensional scaling (MDS) ordination on a Bray-Curtis similarity matrix using routines in the PRIMER (Plymouth Routines in Multivariate Ecological Research) v.6 statistical package (Clarke and Gorley 2006). The Group Average method was used to link samples in the analysis. Non-metric MDS constructs a plot in which samples are arranged in rank order according to their relative similarity. Samples that are similar in species composition and abundance are placed in close proximity to one another, whereas dissimilar samples are placed further apart. Because abundance for colonial species (bryozoans and hydroids) cannot be provided, the MDS analysis was repeated for presence/absence data using the full matrix of species and Sørensen's similarity index (Clarke and Gorley 2006). Multivariate and univariate (Analysis of Variance) analyses were also conducted on the post-cleaning and post-transit samples to identify gradients in species abundance and composition among surveys and ships.

Photo-quadrats were examined by quantifying the percent cover of eleven distinguishable categories of biofouling in each image: algae, barnacle, barnacle seat/ organism remnant, bryozoan, crustacean, hydroid, mussel, oyster, sea squirt, bare hull, and "other". Images were analyzed using the point count method to determine percentage cover of each category by superimposing a grid of 8 rows by 13 columns and populating each cell by 1 random point for a total of 104 random points. The area of hull analyzed from the image was approximately 204 cm^2 (11.5 x 17.8 cm), for a density of 1 point for every 2.0 cm^2 of hull (Figure 2-4). For the PRIDE the area analyzed was slightly

larger: approximately 240 cm^2 (13.5 x 17.8 cm) for a density of 1 point for every 2.2 cm^2 of hull corresponding to a grid of 9 rows x 12 columns. Points that were indistinguishable because the image was too dark were removed from the analysis. Thus the analysis provides percent cover of observable hull. Percent cover data (arcsine square-root transformed) were analyzed by MDS.

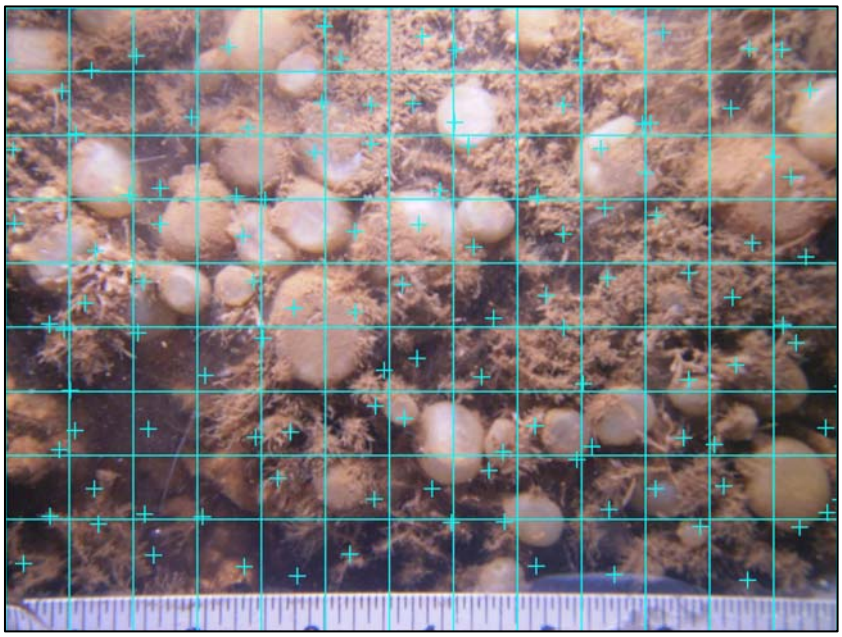

Figure 2-4. Grid of random points superimposed on an underwater photograph taken from the bottom of Transect 3 of the CAPE CHARLES prior to hull cleaning. Images were analyzed using the point count method to determine percentage cover of each of 11 categories of biofouling. Sea squirts and bryozoans can be observed in this image.

3.0 RESULTS AND DISCUSSION

3.1 WATER CHARACTERISTICS

The salinity of the James River at the time of sampling (January through early March) was in the low mesohaline range, between 9 and 15 psu, based on 6 YSI deployments over the tidal cycle (Figure 3-1, A-C). Water temperature ranged between 5.6 and 9.7 ºC, and dissolved oxygen was high near saturation, and ranged between 11.2 and 11.9 mg/L. Salinity in the mesohaline James River fluctuates widely depending on river flow, and inter-annual variability can be significant. For example, in January 2007, during the survey of the MISSISSINEWA, salinity ranged between 1.0 psu near the surface and 6.4 psu at 6 m depth. In Brownsville, Texas, salinity was 32 psu in February 2008, water temperature ranged between 16 and 17 ºC, and dissolved oxygen ranged between 6.4 and 6.9 mg/L (Figure 3-1, D).

3.2 SPECIES ASSEMBLAGES

One hundred and twelve taxa were recorded from the biological samples across all surveys and ships, of which 70 were identified to species level (Table 3-1). A total of 65 taxa were recorded in the James River whereas 47 taxa were recorded only in Brownsville. Appendix B provides species biogeographical and life history information.

The most common species in the surveys was the barnacle *Balanus improvisus*, which accounted for 67% of total abundance and 80,565 individuals. The next most common species was the amphipod *Apocorophium lacustre*, which accounted for 10% of total abundance and 11,573 individuals. Other numerically dominant species, in order from high (6.5%) to low (1.2%) abundance, were the Hooked Mussel, *Ischadium recurvum*, the amphipod *Incisocalliope aestuarius*, the polychaete worm *Neanthes succinea*, the sea squirt *Molgula manhattensis*, the amphipod *Melita nitida*, and the mud crab *Eurypanopeus depressus*. Among the colonial species, the nonindigenous small hydroid *Blackfordia virginica* and the bryozoan *Conopeum chesapeakensis* were abundant and covered mussels and barnacles.

In terms of frequency of occurrence among the pre-cleaning samples, *B. improvisus* was present in 100% of the samples, *A. lacustre* and *I. aestuarius* were present in 92-100% of the samples, *B. virginica* in 88-100% of the samples, and *I. recurvum* in 74-100% of the samples (Table 3-1). Other frequently occurring species were *N. succinea*, *E. depressus*, *C. chesapeakensis*, *M. manhattensis*, and the hydroid *Garveia franciscana*. These species occurred in 62-98% of the pre-cleaning samples (Table 3-1). Nematodes, or round worms, were also frequent and abundant in the samples, but they were not identified to species because of their taxonomic complexity.

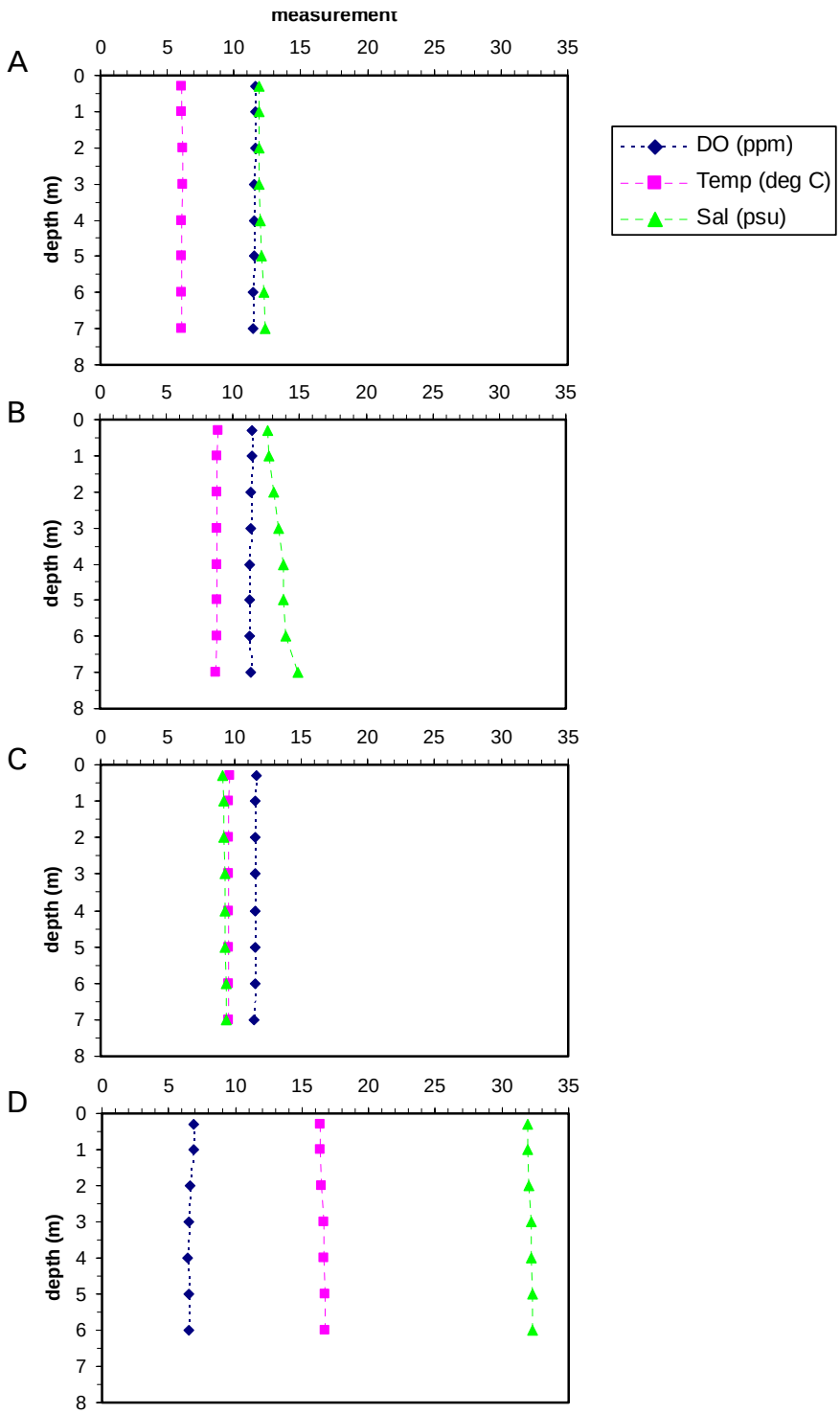

Figure 3-1. Salinity, temperature, and dissolved oxygen of the water in the James River at Fort Eustis, Virginia (A-C), and Brownsville, Texas (D), during the biological surveys of James River Reserve Fleet vessels PRIDE (A,D), CAPE CHARLES (B), and SCAN (C). Readings were taken on January 4, 1:30 p.m. (A), January 12, 3:00 p.m. (B), February 2, 9:15 a.m. (D), and March 5, 2008, 8:45 a.m. (C).

Table 3-1. Species recorded in biological samples of three James River Reserve Fleet vessels. The frequency of occurrence (percent of samples) in each survey and the biogeographic status of species in Chesapeake Bay is shown. Frequency of occurrence for copepods and ostracods is that of the 250-μm samples. The 64- μm samples were not examined. Species found in MISSISSINEWA surveys (Versar 2008b) are indicated in the last column.

	Pre-Cleaning			Post-Cleaning			Post-Transit				
Species	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Status Chesapeake**	**Miss**
Algae											
Algae sp. A	14	18	8	10	2	0	2	0	2	?	x
Algae sp. B	14	16	12	6	0	4	12	0	4	?	x
Amphipods											
Ampithoe valida	0	6	0	0	0	0	0	0	0	native	
Apocorophium lacustre	94	100	100	58	80	67	72	36	48	native	x
Caprella equilibria	0	0	0	0	0	0	0	2	0	native	
Caprella scaura	0	0	0	0	0	0	0	2	0	no record/not present	
Elasmopus cf. rapax	0	0	0	0	0	0	0	70	14	no record/not present	
Ericthonius brasiliensis	0	0	0	0	0	0	0	70	28	native	
Ericthonius sp.	0	0	0	0	0	0	6	0	0	native species present	
Gammarus daiberi	12	78	68	20	31	16	0	0	0	native	x
Grandidierella bonnieroides	0	0	0	0	0	0	0	2	0	no record/not present	
Incisocalliope aestuarius	92	96	100	52	55	43	22	6	32	native	
Jassa marmorata	0	0	0	0	0	0	0	4	0	native	
Laticorophium baconi	0	0	0	0	0	0	4	40	16	no record/not present	
Melita nitida	38	72	96	18	33	27	12	10	8	native	x
Monocorophium acherusicum	0	0	0	0	0	0	0	0	6	cryptogenic	
Paracaprella sp. (juv.)	0	0	0	0	0	0	0	2	0	native species present	
Podocerus brasiliensis	0	0	0	0	0	0	2	0	0	no record/not present	
Anthozoans (Sea Anemones)											
Diadumene leucolena	10	2	14	4	2	0	0	0	2	native	
Ascidians (Sea Squirts)											
Molgula manhattensis	72	66	80	18	18	18	2	0	0	native	
Bivalves											
Arcidae spp. Indeterminant	0	0	0	0	0	0	0	2	0	native species present	
Crassostrea virginica	22	34	70	16	33	20	16	38	40	native	x
Ischadium recurvum	74	80	100	50	86	59	44	60	78	cryptogenic	x
Macoma balthica	0	2	4	0	0	0	0	0	0	native	
Mya arenaria	16	6	34	2	0	0	0	0	0	native	
Mytilopsis leucophaeata	6	2	40	2	2	6	2	0	0	native	x
Tellina agilis	6	0	10	2	4	0	0	0	0	native	
Chaetognaths (Arrow Worms)											
Chaetognatha spp.	0	0	0	0	0	0	0	0	12	native species present	x

Table 3-1. Continued.

	Pre-Cleaning			Post-Cleaning			Post-Transit				
Species	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Status Chesapeake**	**Miss**
Cirripedia (Barnacles)											
Balanus amphitrite	0	0	0	0	0	0	0	10	0	introduced	
Balanus improvisus	100	100	100	74	98	90	96	96	96	native	x
Lepas anatifera	0	0	0	0	0	0	0	6	2	no record/not present	
Copepods											
Acartia sp. (copepodite)	8	0	4	6	2	10	0	0	0	native species present	
Acartia tonsa	8	0	22	8	20	4	0	2	70	native	
Calanoida spp. (copepodite)	0	0	0	0	0	2	2	0	10	native species present	
Calanus sp.	0	0	0	0	0	0	0	0	2	native species present	
Centropages hamatus	0	0	0	0	0	0	0	0	10	native	
Clausidiidae spp. (copepodite)	0	0	0	0	2	0	0	0	0	native species present	
Copepoda spp. 1 Indeterminant	0	0	0	0	0	0	0	0	2	?	
Corycaeus speciosus	0	0	0	0	0	0	0	0	4	native	
Coullana canadensis	0	0	16	0	0	0	0	0	0	native	
Cyclopoida spp. (copepodite)	2	0	4	0	0	0	0	0	0	native species present	
Dactylopusia sp.	0	0	0	0	0	0	0	2	0	native species present	
Ectinosomatidae spp.	0	0	4	0	0	0	0	0	0	native species present	
Eurytemora affinis	2	0	0	0	0	0	0	0	0	native	x
Eurytemora sp. (copepodite)	0	0	0	2	4	0	0	0	0	native species present	
Euterpina acutifrons	0	0	2	0	0	0	0	0	0	native	
Halicyclops sp.	2	10	20	4	4	2	0	0	0	native species present	x
Hemicyclops sp.	0	0	0	0	0	0	0	6	0	native species present	
Labidocera aestiva	0	0	0	0	0	0	0	4	4	native	
Miraciidae spp.	0	0	0	0	0	0	2	0	0	native species present	
Paracalanus sp.	0	0	0	0	0	0	2	0	4	native species present	
Paronychocamptus huntsmani	6	4	2	0	0	0	0	2	0	native	
Parvocalanus crassirostris	0	0	0	0	0	0	0	6	0	native	
Pseudocyclops sp.	0	0	0	0	0	0	0	2	0	native species present	
Pseudodiaptomus pelagicus	0	0	4	0	0	2	0	0	6	native	
Rhincalanus cornutus	0	0	0	0	0	0	0	2	0	native	
Schizopera sp.	4	0	0	0	0	0	0	0	0	native species present	
Tisbe sp.	0	0	0	0	0	0	0	2	2	native species present	
Decapods (Shrimps & Crabs)											
Alpheus sp. (juv.)	0	0	0	0	0	0	0	2	0	native species present	
Caridea spp. (juv.)	0	0	0	0	0	0	0	52	0	native species present	
Decapoda spp. Indeterminant	0	0	0	2	0	0	0	0	2	native species present	
Eurypanopeus depressus	82	80	98	26	31	16	20	20	36	native	x
Lucifer faxoni	0	0	0	0	0	0	0	0	2	native	

Table 3-1. Continued.

	Pre-Cleaning			Post-Cleaning			Post-Transit				
Species	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Status Chesapeake**	**Miss**
Pachygrapsus transversus?	0	0	0	0	0	0	4	0	0	no record/not present	
Palaemon macrodactylus	0	0	2	0	0	0	0	0	0	introduced	
Palaemonetes pugio	4	0	10	0	0	4	0	0	0	native	
Palaemonetes sp.	0	0	2	2	0	2	0	0	0	native species present	
Palaemonetes vulgaris	2	0	4	0	0	0	0	0	0	native	
Penaeidae spp.	0	0	0	0	0	0	0	2	0	native species present	
Pinnotheridae spp. (juv.)	0	0	0	0	0	0	0	2	0	native species present	
Porcellanidae spp. (juv.)	0	0	0	0	0	0	0	22	0	native species present	
Rhithropanopeus harrisii	22	24	30	4	2	2	4	2	12	native	x
Xanthidae spp. (juv.)	0	0	0	0	0	0	0	20	2	native species present	x
Ectoprocts (Bryozoans)											
Alcyonidium cf. albescens	16	2	2	2	2	2	6	2	2	native	
Bowerbankia gracilis	16	12	46	4	8	14	6	2	8	native	
Conopeum chesapeakensis	90	72	96	66	69	20	58	66	60	native	x
Conopeum tenuissimum	0	22	4	0	8	6	0	4	0	native	
Membranipora tuberculata	0	0	0	0	0	0	0	2	0	no record/not present	
Fishes											
Alosidae spp. (eggs)	0	0	0	0	0	0	0	0	10	native species present	
Gobiosoma bosc	4	2	0	0	0	0	0	0	0	native	
Gastropods											
Doridella obscura	0	0	4	0	0	2	0	0	0	native	
Naticidae spp. (juv.)	0	0	0	0	0	0	0	2	0	native species present	
Nudibranchia spp. (juv.)	4	4	8	4	0	2	0	0	0	native species present	
Hydroids											
Aglaophenia cf. latecarinata	0	0	0	0	0	0	0	2	0	no record/not present	
Blackfordia virginica	100	88	96	64	69	61	40	6	32	introduced	
Clytia sp.	0	0	0	0	0	0	2	6	8	native species present	
Garveia franciscana	66	62	82	34	73	43	44	72	78	introduced	x
Obelia geniculata	0	0	0	0	0	0	6	70	12	cryptogenic	
Insects											
Telmatogeton japonicus	0	0	0	0	2	0	4	0	0	cryptogenic	
Isopods											
Cirolana parva	0	0	0	0	0	0	0	0	4	no record/not present	
Edotea triloba	2	0	0	2	0	0	0	0	0	native	
Sphaeromatidae spp. (Dyameninae)	0	0	0	0	0	0	0	30	6	native species present	
Synidotea laevidorsalis	46	26	32	8	2	6	0	0	0	introduced	
Mysids (Fairy Shrimps)											
Mysidae spp.	0	0	0	0	0	0	2	0	0	native species present	

Table 3-1. Continued.

	Pre-Cleaning			Post-Cleaning			Post-Transit				
Species	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Pride**	**Scan**	**C. Charles**	**Status Chesapeake**	**Miss**
Nematodes (Roundworms)											
Nematoda spp.	88	48	98	56	80	100	92	84	92	native species present	x
Oligochaetes											
Tubificoides heterochaetus	0	0	2	0	0	0	0	0	0	cryptogenic	
Ostracods											
Ostracoda spp.	2	26	26	0	53	8	0	16	66	native species present	
Plants											
Distichlis spicata	0	0	0	0	0	20	0	0	0	native	
Sargassum sp.	0	0	0	0	0	0	0	2	0	no record/not present	
Polychaetes											
Boccardiella ligerica	12	6	6	2	0	2	2	0	0	cryptogenic	x
Neanthes succinea	96	98	98	52	80	57	46	76	88	native	x
Polydora cornuta	22	28	64	6	8	4	2	0	2	native	x
Polydora sp.	2	0	0	0	0	0	0	0	0	native species present	
Polydora websteri	52	24	76	30	27	12	12	10	26	native	
Proceraea cornuta	0	0	0	0	0	2	6	26	2	native	
Streblospio benedicti	2	0	20	0	2	2	0	0	0	native	
Porifera (Sponges)											
Halichondria bowerbanki?	0	0	0	0	2	0	0	0	0	native	
Sponge-like organism	2	0	0	10	0	2	10	0	0	native species present	
Tanaids											
Sinelobus stanfordi	0	0	0	0	0	0	0	4	0	no record/not present	
Turbellarians (Flatworms)											
Euplana gracilis	30	14	8	8	0	0	0	0	0	native	x
Stylochus ellipticus	68	62	48	30	29	10	28	18	32	native	x
Turbellaria spp. (juv.)	0	4	4	0	0	4	0	6	2	native species present	

By ship, 41 species and 32,000 individuals were collected in the James River from the PRIDE (pre-cleaning and post-cleaning samples), 43 species and 33,400 individuals from the SCAN, and 47 species and 36,400 individuals from the CAPE CHARLES, based on distinct species. The CAPE CHARLES therefore exhibited higher number of species and total abundance than the other two vessels. Most species were present in all ships, the ships differing only in the number of rare or infrequent species. Further comparisons among vessels are presented in Section 3.3 of this report.

Of the 65 taxa collected in the James River, 39 were native to Chesapeake Bay, 4 were cryptogenic (i.e., of uncertain origin), and 4 were introduced (Table 3-1). Of the remaining taxa, 16 were genus or higher level identifications with native species present in Chesapeake Bay, and 2 were of undetermined status. Eleven of the James River taxa had no record in Texas, 6 were cryptogenic in Texas, and 2 were introduced.

Species cryptogenic in Chesapeake Bay were the Hooked Mussel, *Ischadium recurvum*, the polychaete *Boccardiella ligerica*, the oligochaete *Tubificoides heterochaetus*, and the chironomid insect *Telmatogeton japonicus*. These species were also cryptogenic in Texas, except for the Hooked Mussel, which is native to the Gulf of Mexico and the Northwest Atlantic Ocean south of the Chesapeake Bay. Introduced species in Chesapeake Bay were the hydroid *Blackfordia virginica*, of Black Sea origin; the hydroid *Garveia franciscana*, which is possibly native to the Indian Ocean; the isopod crustacean *Synidotea laevidorsalis*, which is native to the Northwest Pacific Ocean; and the Oriental Shrimp, *Palaemon macrodactylus*, native to northeast Asia.

The Black Sea species *Blackfordia virginica* is best known for its medusa, which is often abundant in surface plankton in the regions from which it has been reported. The polyps are small and inconspicuous, and thus difficult to see in the field. We found dense colonies of the polyp covering barnacle and mussels in all James River ship hull surveys, which constitutes the first report of such densities in Chesapeake Bay. The species was likely transported to the east coast of North America prior to 1910 by shipping (Mills and Sommer 1995), and appeared in San Francisco Bay in the early 1970s. Medusae have been reported in coastal lagoons of Veracruz, Mexico, (Segura-Puertas et al. 2009) and although no *B. virginica* records exist for Texas, it is likely that it occurs there as well.

Garveia franciscana has been introduced in the east and west coasts of North America, including the Gulf of Mexico, and is reported as widespread in Texas, whereas *Synidotea laevidorsalis* is unknown to occur in Texas. *S. laevidorsalis* has invaded California, the Chesapeake Bay, and other mid-Atlantic U.S. coastal states, but there is no record of this invasive isopod in the Gulf of Mexico. The only member of the genus *Synidotea* known from the Gulf of Mexico is the recently described *Synidotea fosteri* (Schotte and Heard 2004), which ranges from Western Florida to Texas. *S. laevidorsalis* has been referred to as *Synidotea laticauda*, but this species is considered a junior synonym of *S. laevidorsalis* by Chapman and Carlton (1994), although Poore (1996) has

argued that *S. laticauda* is morphologically distinct from *S. laevidorsalis* and probably forms part of the native estuarine fauna of the U.S. Atlantic coast.

The specimen of *Palaemon macrodactylus* collected from the hull of the CAPE CHARLES constitutes the first record of this invasive species in the east coast of North America. It was first reported on the U.S. west coast in the 1950s (Newman 1963), and more recently it has been recorded from the Atlantic coast of Europe, Australia, and Argentina (Beguer et al. 2007). It resembles the Grass Shrimp, *Palaemonetes pugio*, with which it may compete for habitat and food resources. Nothing is known about the prevalence of this species in Chesapeake Bay, but the implications of its appearance in Chesapeake Bay may be significant.

In addition to *S. laevidorsalis* and *P. macrodactylus*, there were 8 other species in the James River samples, all native to the Chesapeake Bay, that are unknown to occur in the Gulf of Mexico. These species are the bryozoan *Alcyonidium* cf. *albescens* (previously known as *Alcyonidium polyoum*; Winston and Key 1999), the sea anemone *Diadumene leucolena*, the isopod *Edotea triloba*, the amphipods *Gammarus daiberi* and *Incisocalliope aestuarius*, and the infaunal bivalves *Macoma balthica*, *Mya arenaria* and *Tellina agilis*. Among the Chesapeake Bay native species, one recently described species, the bryozoan *Conopeum chesapeakensis*, has already been reported outside its native range in San Francisco Bay and in Beaumont (TX).

Half of the species found in the pre-cleaning or post-cleaning surveys were also found in the post-transit surveys, indicating active transfer by obsolete vessels of Chesapeake Bay species to Texas. Among these species, three (*A. albescens*, *D. leucolena*, and *I. aestuarius*) emerge as significant because they are not known to occur in the Gulf of Mexico and therefore there is potential for their dispersal outside of their native range. In addition, the widespread occurrence of *C. chesapeakensis* in hull samples of both the pre-cleaning and post-transit surveys is cause of concern, because the distribution of this species in the Gulf of Mexico may be restricted to Beaumont (TX), where specimens attributable to this species were collected from ship hulls (Versar 2008d), and we do not know whether it has been established in other areas of the Gulf region. Bryozoans are notorious invasive species. They reproduce quickly and can foul intake pipes of power plants and waste water treatment facilities at great economic loss.

Underwater photographs taken from the hull of the PRIDE, SCAN, and CAPE CHARLES show with great clarity many of the species discussed in this report (Figure 3-2). Mud crabs of the family Xanthidae, specifically *Rhithropanopeus harrisii* and *Eurypanopeus depressus*, are visible in the photographs, and the isopod *Synidotea laevidorsalis* also appears in some of the photographs. The hulls were mostly covered by a thick layer of mussels and barnacles, which were often overgrown by sea squirts, bryozoans (both the encrusting and branching forms), and hydroids. Most characteristic was *Blackfordia virginica*, which covered most other organisms in a dense layer reminiscent of turf.

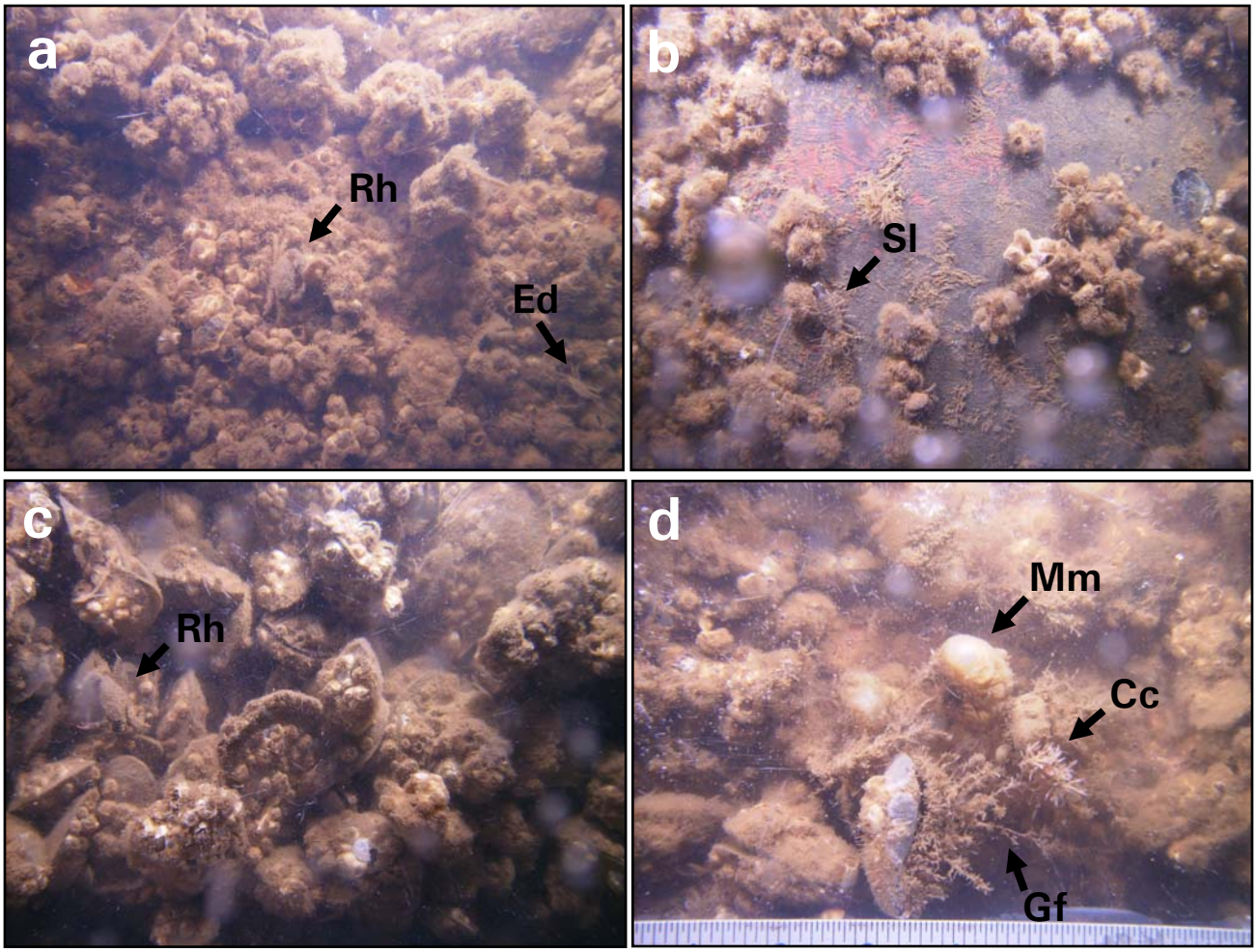

Figure 3-2. Underwater photographs taken from the hull of the PRIDE (a-c) and CAPE CHARLES (d) prior to hull cleaning showing abundant barnacle, mussel, and soft growth. Mussels and barnacles in the photographs are covered by a dense layer of *Blackfordia virginica*. Mussels and barnacles are prominent in photograph c, and soft growth is abundant in photograph d. The arrows point at white-fingered mud crabs, *Rhithropanopeus harrisii* (Rh); the flatback mud crab, *Eurypanopeus depressus* (Ed); an isopod, *Synidotea laevidorsalis* (Sl); the sea squirt *Molgula manhattensis* (Mm); the bryozoan *Conopeum chesapeakensis* (Cc), and the hydroid *Garveia franciscana* (Gf).

In another photograph, tubes of the tube building amphipod *Apocorophium lacustre* can be seen attached to the hull (Figure 3-3). *A. lacustre* is a common brackish water species that builds tubes from mud in marshy banks, pilings, and other substrates. The tubes often aggregate in mats consisting of hundreds or thousands of individuals. Some of these mats were observed on the SCAN.

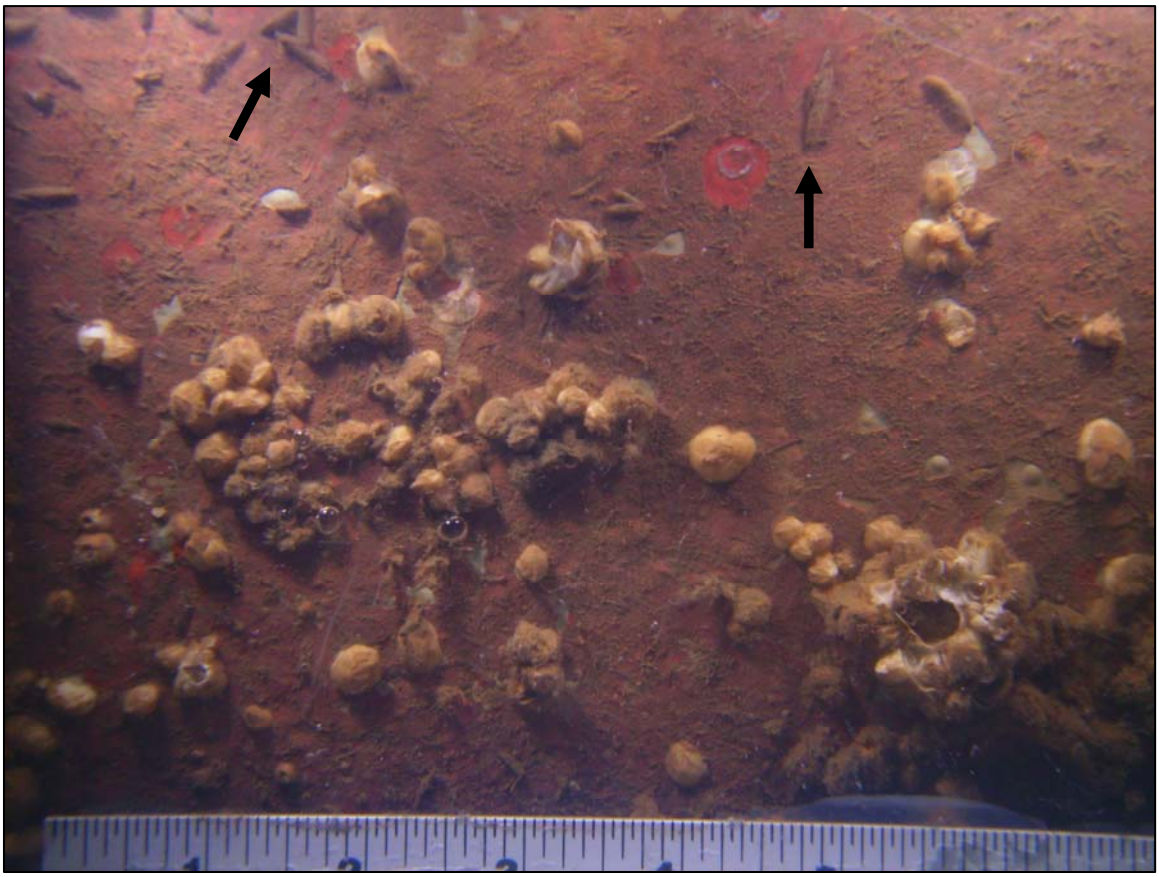

Figure 3-3. Underwater photograph taken from the hull of the SCAN prior to hull cleaning. Tubes of the amphipod *Apocorophium lacustre* can be observed in the upper part of the image (arrows).

3.3 DIFFERENCES BETWEEN SHIPS, SURVEYS, AND LOCATIONS ON THE HULL

The total number of species found in the samples differed among the surveys. There was a reduction in the number of species after hull cleaning, but there was also an increase in the total number of species in the post-transit surveys of the SCAN and CAPE CHARLES (Figure 3-4 A). Many of the post-transit species were species with limited or no distribution in the Chesapeake Bay, suggesting that they attached during transit.

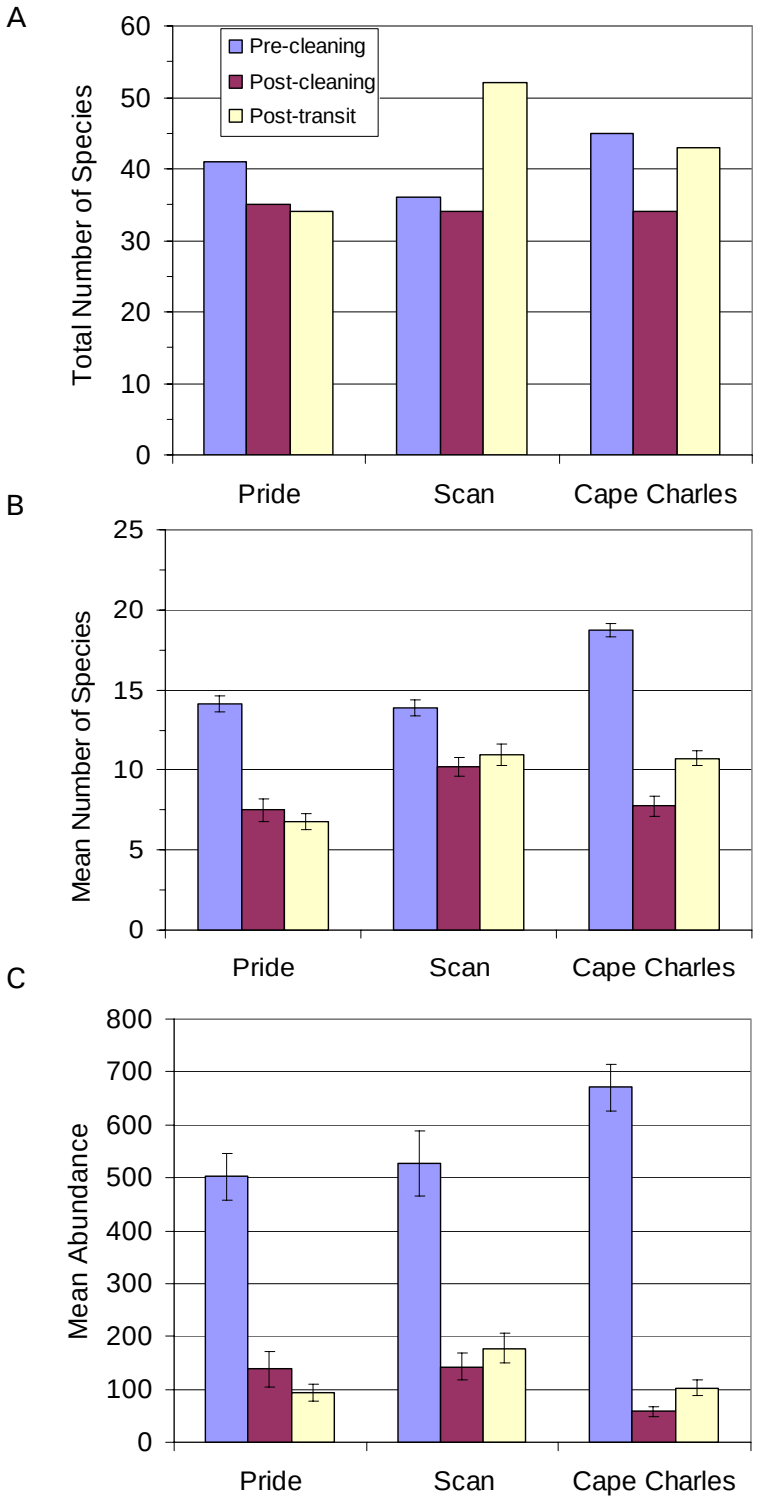

Figure 3-4. Differences in total number of species (A), mean number of species per sample (B), and mean abundance per sample (C) of biofouling organisms in three James River Reserve Fleet vessels, before and after hull cleaning, and after transit from Virginia to Texas. Distinct species only. Error bar = +/- one standard error.

There were significant differences in the mean number of species per sample between vessels and between surveys (Figure 3-4 B), but the interaction term was significant indicating that these differences depended on vessel and survey (2-way ANOVA; survey, $F = 143.8$, $p < 0.0001$; vessel, $F = 22.5$, $p < 0.0001$; interaction, $F = 13.2$, $p < 0.0001$). The pre-cleaning surveys of all vessels had significantly more species per sample than the post-cleaning surveys, and in the CAPE CHARLES the post-transit survey had significantly more species per sample than the post-cleaning survey (Figure 3-4 B).

Abundance (total counts per sample) differed significantly among surveys (Figure 3-4 C), but the differences depended on vessel (2-way ANOVA; survey, $F = 164.6$, $p < 0.0001$; vessel, $F = 1.04$, $p = 0.35$; interaction, $F = 4.63$, $p = 0.001$). As with the number of species, the pre-cleaning surveys had significantly greater abundance per sample than the post-cleaning surveys (Figure 3-4 C).

Multivariate analyses of species abundance and composition data (colonial species excluded) and presence-absence data (all species included) revealed no differences in community organization among the pre-cleaning, post-cleaning, or post-transit samples based on transect or location on the hull, except for a group of stern appendage samples distinct in the MDS diagrams (diagrams not shown). Most of the post-cleaning and post-transit samples collected from the stern appendages of the PRIDE and CAPE CHARLES were characterized by fewer species, and had fewer mussels and barnacles, if any, than those remaining in other areas of the hull after hull cleaning. The running gear of the vessels (rudder, propellers, and shafts) is cleaned by hand because the SCAMP system cannot be applied to these surfaces. In the PRIDE and CAPE CHARLES hand cleaning was more effective at removing the hard shelled organisms and reducing the number of species harbored among them than the machine-operated brushes used on the hull. However, as we have observed in previous studies, the cleaning of vessel appendages by hand brushes is not always effective at removing species.

The multivariate analyses revealed no differences in community organization among ships for the pre-cleaning samples (Figure 3-5 A, B), but there were differences in the post-transit samples that became more clear when all species were included in the analysis of presence-absence data (Figure 3-5 C, D). There was some degree of overlap among the post-transit samples of the PRIDE, CAPE CHARLES, and SCAN in the MDS diagram, but distinct groups could be made, indicating differences in species composition among vessels after their arrival in Brownsville.

By ship, pre-cleaning, post-cleaning, and post-transit samples formed distinct groups in the MDS diagrams, indicating clear differences in species composition among surveys, with little overlap between surveys in the SCAN and CAPE CHARLES (Figure 3-6 A-C). These differences were consistent with those of the univariate analysis, and were due to (1) a substantial reduction in densities and species numbers in the post-cleaning samples, and (2) the appearance of 'new' species that colonized the ships while these were en route

from Chesapeake Bay to Texas. These species were more numerous (Figure 3-4 A) and of different kinds (Figure 3-6 B) in the SCAN than in the other two vessels. Note that although hull cleaning reduced densities and species frequencies, it failed to remove most species from the hulls (Table 3-1, Figure 3-4 A). Some species were presumably removed by the shear forces acting on the hull during the towing, or by changes in salinity, but numerous species (52% of the original assemblage) persisted on the hulls and were transferred by the ships to Texas.

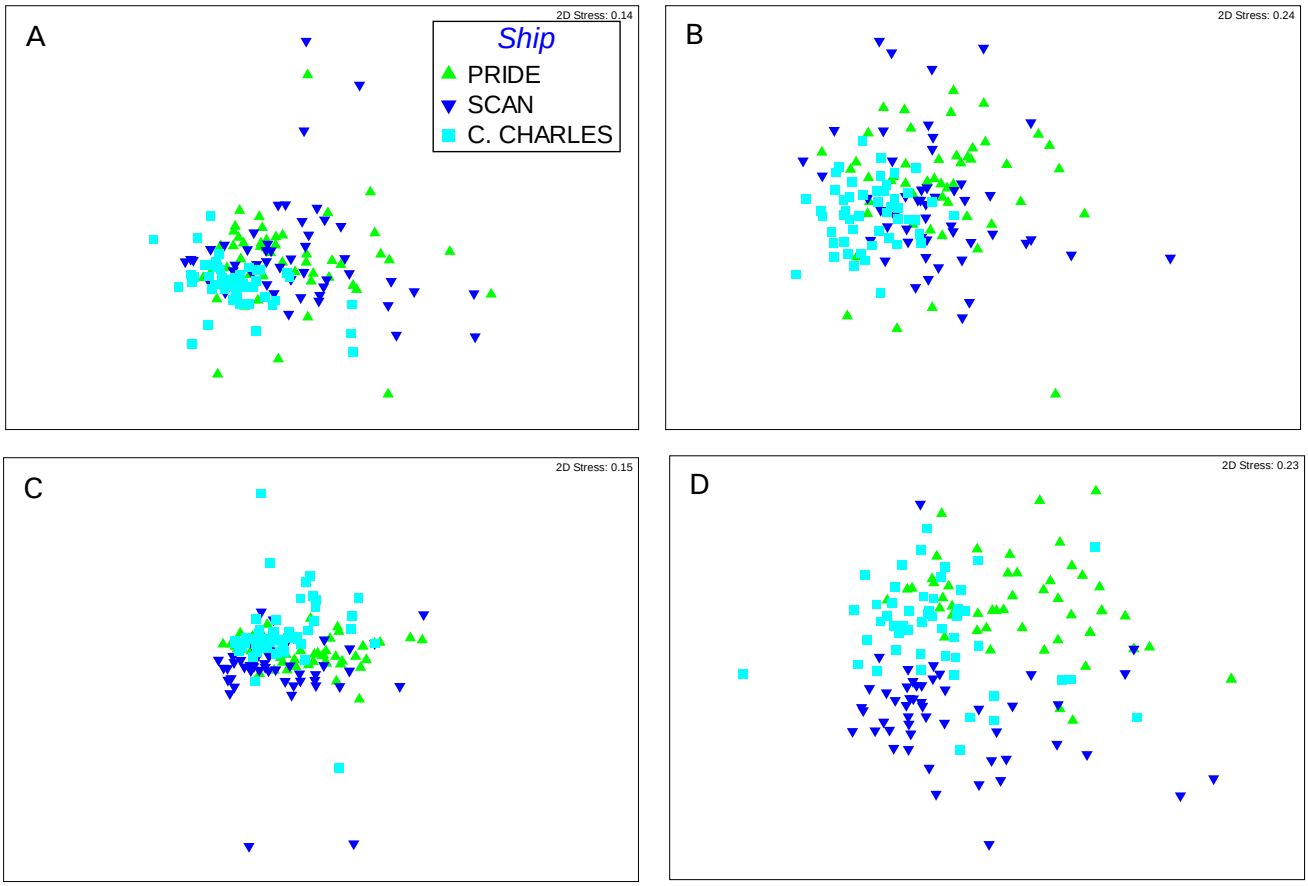

Figure 3-5. Differences between ships in the assemblage structure and species densities of pre-cleaning and post-transit samples. MDS diagrams of pre-cleaning species abundance (non-colonial species) (A), pre-cleaning species presence-absence data (all species) (B), post-transit species abundance (non-colonial species) (C), and post-transit species presence-absence data (all species) (D) shown.

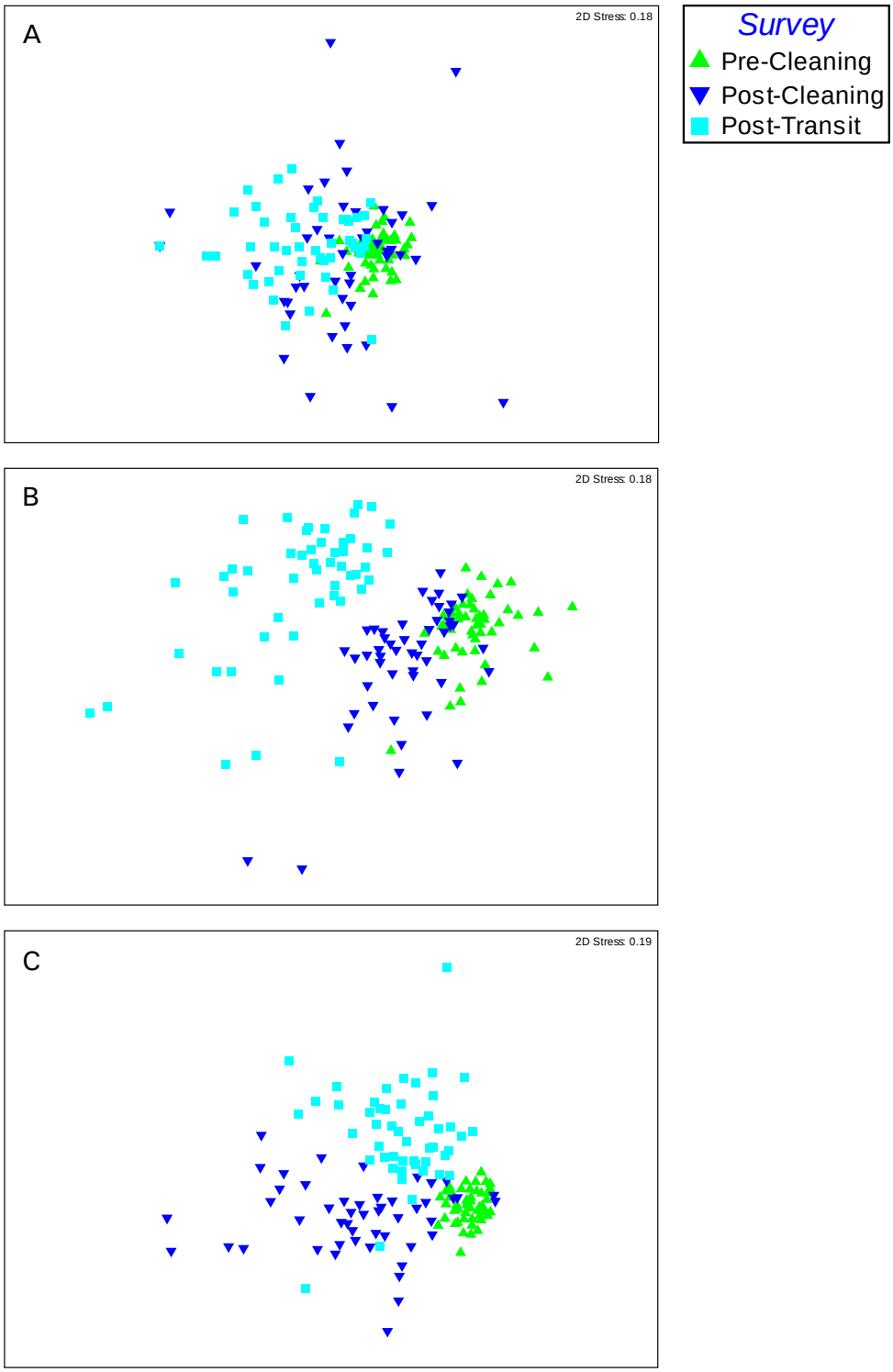

Figure 3-6. Differences in species assemblages among surveys. MDS diagrams of species presence-absence data of PRIDE (A), SCAN (B), and CAPE CHARLES (C) shown. Pre-cleaning, post-cleaning, and post-transit samples form distinct groups in the diagram, with little overlap in the SCAN.

3.4 ANALYSIS OF PERCENT COVER

The photo quadrat analysis revealed no consistent patterns in percent cover of organisms based on transect or location on the hull, although some differences within ship were noted. For example, hydroid cover on the PRIDE increased from the waterline to the bottom of the hull. This last pattern (higher percent cover of hydroids on the bottom of the hull) was also observed on the CAPE CHARLES. Mussels showed the opposite pattern, with higher percent cover near the waterline. The percent cover of mussels was also higher in the mid transects and stern appendages of the PRIDE. However, patterns in the distribution of hard-shelled species should be interpreted with caution because these species are often covered by soft growth which rend them invisible in many of the photographs.

Barnacles, hydroids, mussels, and sea squirts predominated in the pre-cleaning survey of all ships (Figure 3-7), but percent cover of hydroids, mussels, and sea squirts was higher in the CAPE CHARLES (7-28%) than in the other two ships (3-16%). Algae was a prominent feature of the SCAN (Figure 3-7 B).

In contrast with the pre-cleaning surveys, bare hull (65-76%) and barnacle seats (15-31%) predominated in the post-cleaning surveys (Figure 3-7). The CAPE CHARLES had a higher proportion of organism remnants (barnacle seats and the byssus threads of mussels) than the other two ships, and lowest percentage cover of other biofouling categories, indicating higher mussel cover prior to hull cleaning and more successful removal of biofouling organisms by hull cleaning (Figure 3-7 C).

Bare hull (61-73%) and barnacle seats (18-31%) were also prominent features of the post-transit surveys of all ships (Figure 3-7), but the SCAN exhibited considerable hydroid growth in the post-transit survey. On average, 10% of the hull of the SCAN was covered by hydroids after transit. This prominent, new hydroid growth was visible in many of the photos of the samples taken at dockside, and was due to *Obelia geniculata*, a species that also exhibited rapid growth on Suisun Bay Reserve Fleet vessels during towing from California to Texas.

Differences in percent cover of biofouling organisms among surveys are also indicated in the multivariate analysis of photo-quadrat data (Figure 3-8). Pre-cleaning and post-cleaning photo quadrats form distinct groups in the MDS diagrams, indicating differences in the relative proportions of bare hull, barnacles, mussels, hydroids, and barnacle seat/organism remnants. Note the spread of points in the diagram of the SCAN, revealing the presence of *Obelia geniculata* in many of the photo quadrats of the post-transit survey (Figure 3-8 B).

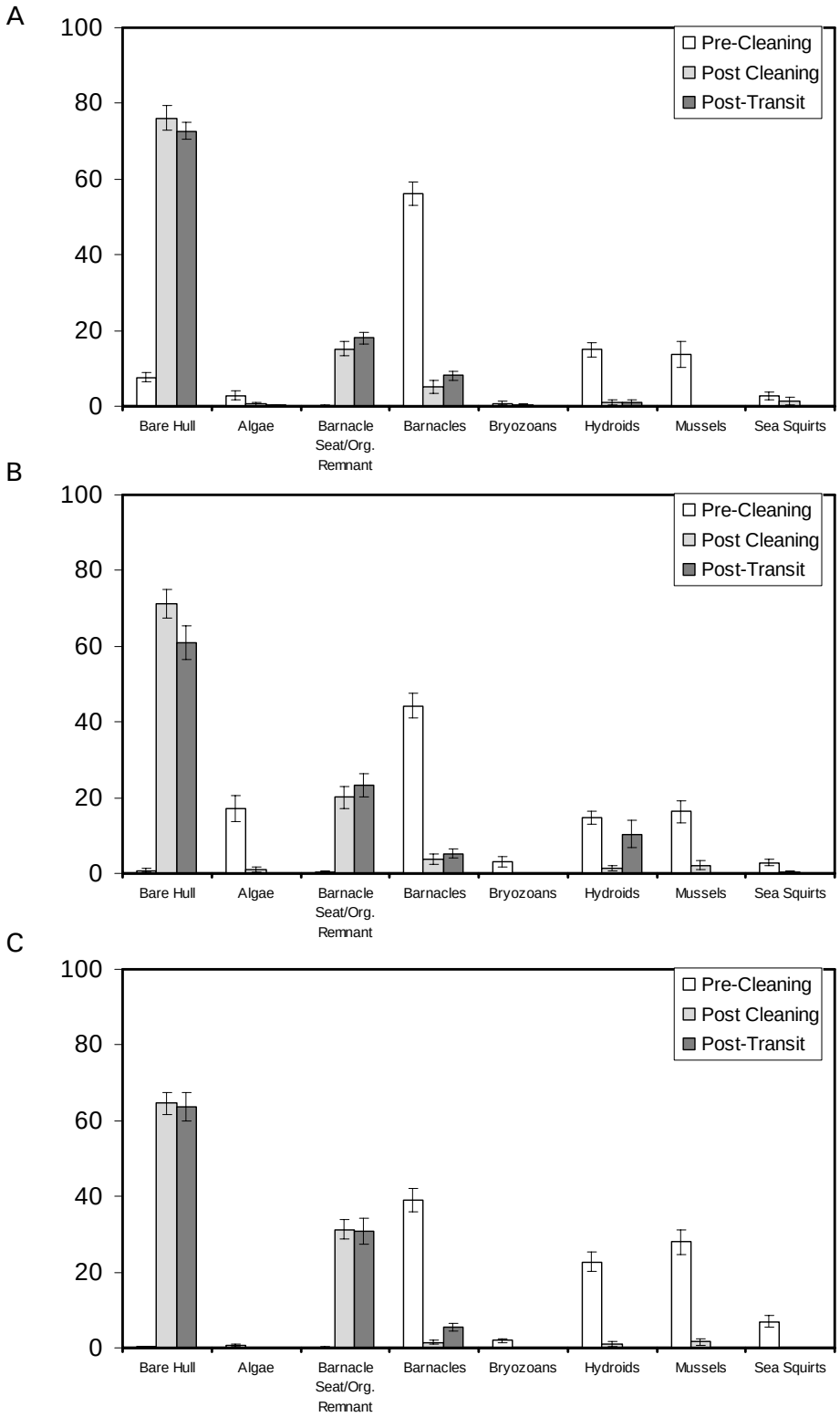

Figure 3-7. Biofouling percent cover of PRIDE (A), SCAN (B), and CAPE CHARLES (C), before hull cleaning, after hull cleaning, and after transit from Virginia to Texas. Shown is the mean +/- one standard error of 8 prominent categories of biofouling estimated from analysis of photo quadrats.

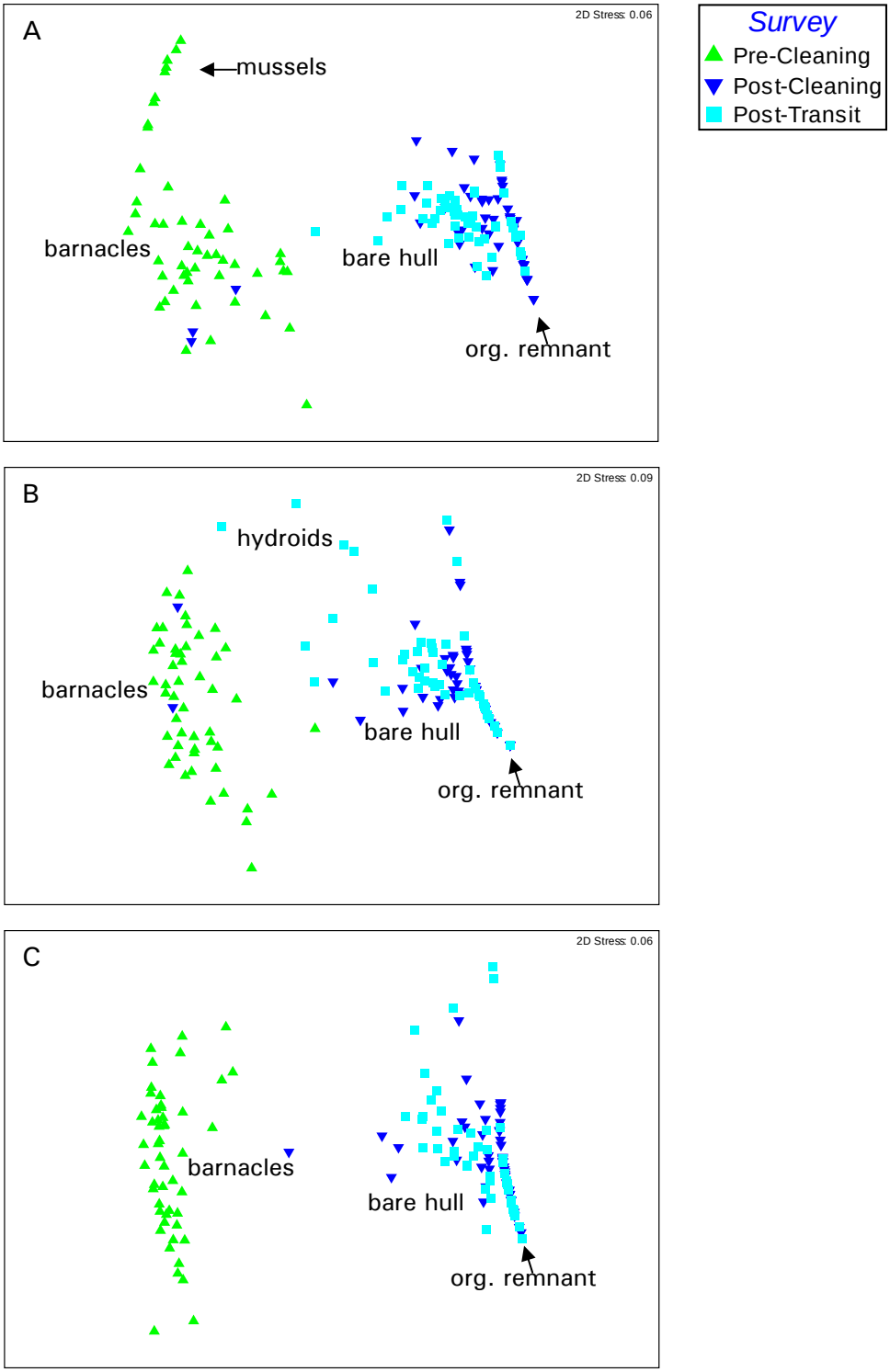

Figure 3-8. Differences in percent cover of biofouling organisms among surveys. MDS diagrams of photo quadrats of PRIDE (A), SCAN (B), and CAPE CHARLES (C) shown. Percent cover differed between pre-cleaning and post-cleaning surveys, indicating differences in the relative proportions of bare hull, hard-shelled organisms, hydroids, and barnacle seat/organism remnants.

3.5 RISK OF SPECIES INTRODUCTIONS

The cleaning of the hulls was successful at removing on average 91-97% of the biofouling cover, if the organism remnant and bare hull categories are summed. Yet, 75% of all the taxa found in the pre-cleaning surveys were found in the post-cleaning surveys, and 52% of these taxa persisted on the hulls of the vessels during their voyage from Virginia to Texas. Although hull cleaning was effective at removing most of the thick base layer of barnacle and mussels, and at reducing the densities of organisms (Figure 3-4 C), many species persisted on the hulls after hull cleaning and their overall frequency in the samples was high (Table 3-1, Figure 3-4 A, B).

Most of the Chesapeake Bay taxa found in the pre-cleaning and post-cleaning surveys were native or had native species present in Texas, but 10 species were unknown to occur in Texas. These last species were from a variety of taxonomic groups, including 3 infaunal bivalves, 2 amphipods, 2 isopods, one decapod, one bryozoan, and one sea anemone. All but two of these species were present in the post-cleaning samples, and 3, the bryozoan *Alcyonidium* cf. *albescens*, the sea anemone *Diadumene leucolena*, and the amphipod *Incisocalliope aestuarius*, were found in the post-transit samples, which indicates transfer on hulls of species nonindigenous to Texas. Two other species that were transferred to Texas on the hulls of JRRF ships are the hydroid *Blackfordia virginica* and the bryozoan *Conopeum chesapeakensis*, which are nonindigenous to Texas but are suspected to already occur in Texas (This report, Versar 2008d). Many other species were transferred on the hulls, some of which are of cryptogenic origin or introduced in both the Chesapeake Bay and the Gulf of Mexico. One invasive species, the isopod *Synidotea laevidorsalis*, was frequent in the pre-cleaning samples and infrequent in the post-cleaning samples, but was not detected in the post-transit surveys. Lastly, the finding of the Oriental Shrimp, *Palaemon macrodactylus*, on James River ship hulls is of concern because this is the first record for the U.S. east coast of this notorious invasive species and the implications of its appearance may be significant. We recommend that further surveys of JRRF vessels be conducted to determine if this shrimp occurs more frequently at other times of the year or in other vessels.

As we have observed in other surveys of Suisun Bay (CA) and Beaumont (TX) Reserve Fleet vessels, many species are able to attach to the hulls and grow during the voyage of the vessels to their final destination in Brownsville. In the present study, 47 taxa (42% of all the taxa found in the surveys) representing at least 43 distinct species, were 'new' taxa found only in the post-transit surveys. One of these species was the hydroid *Obelia geniculata*, which exhibited large, well-developed colonies in many of the samples collected from the SCAN in Brownsville. This same species was found growing on the hulls of Suisun Bay vessels. It is of wide distribution in the Pacific and Atlantic oceans, including limited distribution in the Chesapeake Bay, but was not found at the reserve fleet locations. This is one example of the capability of marine organisms to attach and grow on the hulls of vessels during relatively short inter-oceanic voyages. It augments the risk of species transfers and introductions worldwide. The SCAN had the largest

number of 'new' species, and there was a positive statistically significant relationship between the number of days the vessels were in transit and the number of 'new' species in the post-transit surveys (Table 3-2).

Table 3-2. Relationship between the number of days the vessel was in transit from Virginia to Texas and the number of species found only in the post-transit surveys.

Ship	No. of Days in Transit	No. of 'New' Species
PRIDE	15	9
CAPE CHARLES	16	19
SCAN	20	31

The PRIDE, SCAN, and CAPE CHARLES were surveyed in the James River in the same season (January-February 2008) than the MISSISSINEWA (January 2007), yet large differences between the 2008 and the 2007 surveys emerge. Mussels and barnacles formed a thick layer over most of the hull in all vessels, and percent cover of mussels was higher in the MISSISSINEWA than on the vessels surveyed in the present study. However, the MISSISSINEWA did not exhibit the more extensive hydroid and bryozoan growth observed in the present study, and did not carry sea squirts. The number of species recorded in 2007 from the MISSISSINEWA was 22, whereas the number of species recorded in 2008 from the PRIDE, SCAN, and CAPE CHARLES ranged between 41 and 47, about twice the number of species recorded from the MISSISSINEWA. Several common species recorded in the present study, for example *Blackfordia virginica*, *Molgula manhattensis* and *Incisocalliope aestuarius*, were absent from the 2007 survey. This is evidence of large inter-annual variability in biofouling, and seasonal variability is also likely to occur. Therefore, conclusions based on one study cannot be extended to other ships without considerations of seasonal and annual variability, and as we have seen in previous studies of Beaumont and Suisun Bay vessels, age of vessel and length of berthing. It is recommended that some characterization of extent and type of biofouling be conducted each year, and at different seasons, to capture the full range of biofouling and its potential to harbor invasive species.

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4.0 SUMMARY AND CONCLUSIONS

1. Pre-cleaning, post-cleaning, and post-transit surveys of biofouling were conducted on James River National Defense Reserve Fleet vessels PRIDE, SCAN, and CAPE CHARLES. The surveys yielded a total of 112 taxa, 70 of which were identified to species level. Among the species found in the pre-cleaning and post-cleaning surveys, 39 species were native to Chesapeake Bay, 4 were cryptogenic, and 4 were introduced.

2. The biofouling community was dominated by the barnacle *Balanus improvisus* and the amphipod *Apocorophium lacustre*, which accounted for 67% and 10% of total abundance, respectively. Secondary numerically dominant species were the Hooked Mussel, *Ischadium recurvum*, the amphipods *Incisocalliope aestuarius* and *Melita nitida*, the polychaete worm *Neanthes succinea*, the sea squirt *Molgula manhattensis*, and the mud crab *Eurypanopeus depressus*. Mussels, barnacles, amphipods, crabs, worms, sea squirts, and colonial species (hydroids and bryozoans) were common and occurred frequently in the samples.

3. There were no consistent patterns in abundance or species composition among transects or locations on the hull. However, the total number of species differed among surveys. There were significant differences in abundance among surveys, and significant differences in the mean number of species per sample among vessels and surveys. Densities and species numbers were lower in the post-cleaning surveys than in the pre-cleaning surveys. However, there was an increase in total species number, mean species numbers, and mean densities in the post-transit surveys of the SCAN and the CAPE CHARLES relative to the post-cleaning surveys.

4. In-water hull cleaning was successful at removing on average 91-97% of the biofouling cover, substantially reducing densities of organisms and species frequencies. However, hull cleaning was not particularly effective at removing species. Many of the species persisted on the hull after hull cleaning. Seventy-five percent of all the taxa found in the pre-cleaning surveys were found in the post-cleaning surveys, and 52% persisted on the hulls of the vessels during towing. Three species unknown to occur in Texas were transferred on the hulls. None were invasive in the Chesapeake Bay.

5. Forty-two percent of all taxa representing at least 43 distinct species were 'new' taxa found only in the post-transit surveys. The SCAN had the largest number of 'new' species. There was a positive significant relationship between the number of days the vessels were in transit and the number of 'new' species in the post-transit surveys. New growth and species attachments while the ships are en route augment the risk for species transfers and introductions.

6. There was evidence of large inter-annual variability in biofouling within seasons. It is recommended that some characterization of extent and type of biofouling be conducted each year, and at different seasons, to capture the full range of biofouling and its potential to harbor invasive species.

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APPENDIX A

SPECIES ABUNDANCES
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APPENDIX B

SPECIES BIOGEOGRAPHY AND LIFE HISTORY INFORMATION

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						Geographical Distribution		Salinity (psu)		Temperature (ºC)		
Phylum	**Class**	**Species/Taxon Name**	**Common Name**	**Chesapeake Bay Invasion Status**	**Status in Texas**	**Native Range**	**Invaded Range**	**Range**	**Optimum**	**Range**	**Optimum**	**Substrate Preference-adults**
Crustacea	Copepoda	Acartia sp.	calanoid copepod (copepodite)	native species present	native species present							
Crustacea	Copepoda	Acartia tonsa	calanoid copepod	native	native	Atlantic Ocean, Pacific Ocean, Indian Ocean	Caspian Sea; Baltic Sea; Black Sea, European brackish waters	freshwater to hypersaline	5-30			planktonic
Cnidaria	Hydrozoa	Aglaophenia cf. latecarinata	plume hydroid	no record/not present	native	Circumtropical epiphyte on pelagic Sargassum						epibenthic, epibiont
Ectropocta	Gymnolaemata	Alcyonidium cf. albescens	bryozoan or moss animal	native	no record/not present	Northwest Atlantic	Northeast Pacific (Oregon, CA), Gulf of Mexico?, possibly missidentified as Alcyonidium polyoum					epibenthic
Chlorophyta		Algae sp. A	algae	?	?							
Chlorophyta		Algae sp. B	algae	?	?							
Chordata	Osteichthyes	Alosidae spp. (eggs)	shads	native species present	native species present							
Crustacea	Decapoda	Alpheus sp. (juv.)	snapping shrimps	native species present	native species present			polyhaline to euhaline				
Crustacea	Amphipoda	Ampithoe valida	amphipod	native	native	Northwest Atlantic, Gulf of Mexico	Northeast Pacific		polyhaline to euhaline			epibenthic tube-building
Crustacea	Amphipoda	Apocorophium lacustre	tube-building amphipod	native	cryptogenic	Northwest Atlantic	Northeast Atlantic					epibenthic tube-building
Mollusca	Bivalvia	Arcidae spp. indeterminant	ark shells	native species present	native species present							
Crustacea	Cirripedia	Balanus amphitrite	acorn barnacle	introduced	introduced	Indo-West Pacific, but limits of native range are uncertain	North Atlantic, Southwest Atlantic, Western Pacific Ocean, Northeast Pacific (California to Panama)	52	10	1.5-40	15-32	epibenthic
Crustacea	Cirripedia	Balanus improvisus	Bay barnacle	native	native	Western Atlantic Ocean, Gulf of Mexico	Northeast Atlantic, Caspian Sea, North Pacific Ocean	0-?	5-25	-2-38	14-30	epibenthic
Cnidaria	Hydrozoa	Blackfordia virginica	hydroid	introduced	no record/not present	Black Sea	Caspian Sea, North Atlantic Ocean, Southwest Atlantic, Northeast Pacific, Indian Ocean	3-35		eury-thermal	15-23	epibenthic and planktonic
Annelida	Polychaeta	Boccardiella ligerica	polychaete or bristle worm	cryptogenic	cryptogenic	Northeast Atlantic	Baltic Sea, Northeast Pacific, South Atlantic Ocean, and possibly (cryptogenic range), Northwest Atlantic and Gulf of Mexico	0-30	2-20			infaunal
Ectoprocta	Gymnolaemata	Bowerbankia gracilis	creeping bryozoan	native	native	Western Atlantic Ocean	Northeast Atlantic, Northeast Pacific, Hawaii, Indian Ocean	3-30				epibenthic
Crustacea	Copepoda	Calanoida spp. indeterminant	calanoid copepod (copepodite)	native species present	native species present							
Crustacea	Copepoda	Calanus sp.	calanoid copepod	native species present	native species present							
Crustacea	Amphipoda	Caprella equilibra	skeleton shrimp	native	native	Cosmopolitan	Range possibly extended by shipping					epibiont
Crustacea	Amphipoda	Caprella scaura	skeleton shrimp	no record/not present	native	Cosmopolitan	Range possibly extended by shipping					epibiont
Crustacea	Decapoda	Caridea spp. (juv.)	caridean shrimps	native species present	native species present							
Crustacea	Copepoda	Centropages hamatus	calanoid copepod	native	native	North Atlantic Ocean, Gulf of Mexico, Mediterranean Sea			>20		<20	planktonic
Chaetognatha		Chaetognatha spp.	arrow worms	native species present	native species present							
Crustacea	Isopoda	Cirolana parva	isopod	no record/not present	native	Eastern Tropical Pacific, Tropical Atlantic, Gulf of Mexico, Caribbean Sea		euhaline				epibenthic, epibiont
Crustacea	Copepoda	Clausidiidae spp.	poecilostomatoid copepod (copepodite)	native species present	native species present							
Cnidaria	Hydrozoa	Clytia sp.	hydroid	native species present	native species present	Worlwide range in temperate waters	Range possibly extended by shipping					epibenthic
Ectoprocta	Gymnolaemata	Conopeum chesapeakensis	bryozoan or moss animal	native	introduced	Chesapeake Bay	San Francisco Bay (CA) and possibly Gulf of Mexico					epibenthic
Ectoprocta	Gymnolaemata	Conopeum tenuissimum	bryozoan or moss animal	native	native	Northwest Atlantic, Gulf of Mexico	Northeast Pacific					epibenthic
Crustacea	Copepoda	Copepoda spp. 1 indeterminant	copepods	?	?							
Crustacea	Copepoda	Corycaeus speciosus	poecilostomatoid copepod	native	native	Cosmopolitan						planktonic
Crustacea	Copepoda	Coullana canadensis	harpacticoid copepod	native	native	Northwest Atlantic, Gulf of Mexico	Northeast Pacific (cryptogenic)	0-15	5-10			epibenthic and planktonic
Mollusca	Bivalvia	Crassostrea virginica	Eastern oyster	native	native	Northwest Atlantic, Gulf of Mexico	NE Pacific (British Columbia), Hawaii	5-40	14-28	-2-36	15-30	epibenthic
Crustacea	Copepoda	Cyclopoida spp.	cyclopoid copepod (copepodite)	native species present	native species present							
Crustacea	Copepoda	Dactylopusia sp.	harpacticoid copepod	native species present	native species present							epibenthic, pelagic (rafting), and in submerged aquatic vegetation
Crustacea	Decapoda	Decapoda spp. indeterminant	crabs	native species present	native species present							
Cnidaria	Anthozoa	Diadumene leucolena	White anemone	native	no record/not present	Northwest Atlantic (Cape Cod to SC)	NE Pacific (CA and Oregon), Hawaii					epibenthic
Plantae	Liliopsida	Distichlis spicata	Saltgrass, Marsh Spikegrass	native	native	North, Central and South America		freshwater to euhaline				salt marshes
Mollusca	Gastropoda	Doridella obscura	sea slug	native	native	Northwest Atlantic, Gulf of Mexico	Northeast Atlantic, Black Sea, Southwest Atlantic (Brazil)					epibiont
Crustacea	Copepoda	Ectinosomatidae spp.	harpacticoid copepod	native species present	native species present							
Crustacea	Isopoda	Edotea triloba	Mounded-back isopod	native	no record/not present	Northwest Atlantic (Maine to SC)						epibenthic, epibiont
Crustacea	Amphipoda	Elasmopus cf. rapax	amphipod	no record/not present	native	Cosmopolitan in tropical and warm temperate waters	Northeast Pacific, Hawaii, range possibly extrended by shipping					epibenthic, epibiont
Crustacea	Amphipoda	Ericthonius brasiliensis	amphipod	native	native	Western Atlantic Ocean	Widely distributed, probably introduced in much of its range: Mediterranean Sea, Northeast and Northwest Pacific, Hawaii, Southeast Pacific, Indian Ocean	15-38	18-35			epibiont tube-building
Crustacea	Amphipoda	Ericthonius sp.	amphipod	native species present	native species present							epibiont tube-building
Platyhelminthes	Turbellaria	Euplana gracilis	flatworm	native	cryptogenic	Northwest Atlantic	Northeast Atlantic (The Netherlands)					
Crustacea	Decapoda	Eurypanopeus depressus	Flatback mud crab	native	native	Western Atlantic Ocean, Gulf of Mexico		4.5-20.4				epibenthic, among oysters
Crustacea	Copepoda	Eurytemora affinis	calanoid copepod	native	native	North Atlantic Ocean, Gulf of Mexico, Caspian Sea, Baltic Sea, North Pacific	Inland waters in Eastern and Southeastern North America, Great Lakes	0-40	5-15		10-15	planktonic
Crustacea	Copepoda	Eurytemora sp.	calanoid copepod (copepodite)	native species present	native species present							
Crustacea	Copepoda	Euterpina acutifrons	harpacticoid copepod	native	native	Cosmopolitan		>19				pelagic
Crustacea	Amphipoda	Gammarus daiberi	amphipod	native	no record/not present	Northwest Atlantic (Delaware Bay to SC)	San Francisco Bay, Hudson River estuary	1-15	1-5	?-32		epibenthic, pelagic
Cnidaria	Hydrozoa	Garveia franciscana	Rope Grass hydroid	introduced	introduced	Unknown, possibly Indian Ocean	Atlantic Ocean, Gulf of Mexico, NE Pacific, SW Pacific, Black Sea, Caspian Sea	1-35	5-25	0-37.5	9-34	epibenthic

						Geographical Distribution		Salinity (psu)		Temperature (ºC)		
Phylum	**Class**	**Species/Taxon Name**	**Common Name**	**Chesapeake Bay Invasion Status**	**Status in Texas**	**Native Range**	**Invaded Range**	**Range**	**Optimum**	**Range**	**Optimum**	**Substrate Preference-adults**
Chordata	Osteichthyes	Gobiosoma bosc	Naked gobi	native	native	Northwest Atlantic, Gulf of Mexico		0-45	0.3-22.3			demersal, among oyster reefs
Crustacea	Amphipoda	Grandidierella bonnieroides	amphipod	no record/not present	native	Cosmopolitan in tropical and warm temperate waters		1-40				epibenthic tube-building
Porifera		Halichondria bowerbanki?	Crumb-of-Bread sponge	native	native	North Atlantic Ocean, Gulf of Mexico	Northeast Pacific					epibenthic
Crustacea	Copepoda	Halicyclops sp.	cyclopoid copepod	native species present	native species present	Cosmopolitan		brackish to salt				epibenthic and planktonic
Crustacea	Copepoda	Hemicyclops sp.	poecilostomatoid copepod	native species present	native species present	Cosmopolitan						commensal with invertebrates
Crustacea	Amphipoda	Incisocalliope aestuarius	amphipod	native	no record/not present	Northwest Atlantic (Delaware Bay to Georgia, Florida?)	Northeast Atlantic	10-33		-2-29		epibiont
Mollusca	Bivalvia	Ischadium recurvum	Hooked mussel	cryptogenic	native	Northwest Atlantic south of Chesapeake Bay, Gulf of Mexico	Barnegat Bay, NJ, and northwards, with transplanted oysters	4.5-36	8-36		11.6-24.5	epibenthic, among oysters
Crustacea	Amphipoda	Jassa marmorata	amphipod	native	native	Northwest Atlantic (Newfounland to Texas)	Northeast Atlantic, Mediterranean, South Atlantic, Pacific Ocean, Indian Ocean					epibenthic tube-building
Crustacea	Copepoda	Labidocera aestiva	calanoid copepod	native	native	North Atlantic Ocean, Gulf of Mexico						planktonic
Crustacea	Amphipoda	Laticorophium baconi	amphipod	no record/not present	introduced	Possibly native to Northeast Pacific and Peru	Hawaii, Northwest Pacific, Southwest Pacific, Florida, and Gulf of Mexico	?-39	polyhaline to euhaline			epibenthic
Crustacea	Cirripedia	Lepas anatifera	pelagic goosneck barnacle	no record/not present	native	Cosmopolitan in tropical and temperate oceans (pelagic)					19-25	epibenthic, epibiont
Crustacea	Decapoda	Lucifer faxoni	shrimp	native	native	Warm waters of the Atlantic Ocean; Caribbean, Gulf of Mexico						planktonic
Mollusca	Bivalvia	Macoma balthica	Baltic macoma	native	no record/not present	Circumboreal in Northeast Pacific and Atlantic oceans, Baltic Sea. Temperate Northwest Atlantic populations may be a different species.	Northeast Pacific (California)					infaunal
Crustacea	Amphipoda	Melita nitida	amphipod	native	native	Northwest Altantic, Caribbean, Gulf of Mexico	Northeast Pacific, Northeast Altlantic	0-30	3-20	?-32		epibenthic, epibiont
Ectoprocta	Gymnolaemata	Membranipora tuberculata	bryozoan or moss animal	no record/not present	native	Circumtropical on pelagic Sargassum						epibenthic, epibiont
Crustacea	Copepoda	Miraciidae spp.	harpacticoid copepod	native species present	native species present							pelagic
Chordata	Ascidiacea	Molgula manhattensis	sae grape, sea squirt	native	native	North Atlantic Ocean, Gulf of Mexico	Mediterranean Sea, Northwestern Africa, North Pacific Ocean (WA to CA, Japan), Australia					epibenthic
Crustacea	Amphipoda	Monocorophium acherusicum	amphipod	cryptogenic	introduced	Unknown, possibly Northeast Atlantic from where it was originally described	Northwest Atlantic, Gulf of Mexico, Brazil, Northeast Pacific, Northwest Pacific, Hawaii, Southwest Pacific, Indian Ocean	0-38		-2 -30	10-30	epibenthic tube-building
Mollusca	Bivalvia	Mya arenaria	Soft-shell clam	native	no record/not present	Northwest Atlantic (Labrador to Cape Hatteras), Nortwest Pacific (Kurile Islands, Japan, Korea)	Northeast Atlantic, Baltic Sea, Mediterranean Sea, Black Sea, Northeast Pacific (Alaska to CA), with transplanted oysters	5-35	25-35	-2-28	6-14	infaunal
Crustacea	Mysidacea	Mysidae spp.	mysid or fairy shrimps	native species present	native species present							epibenthic, planktonic
Mollusca	Bivalvia	Mytilopsis leucophaeata	Dark False mussel	native	native	Chesapeake Bay to Gulf of Mexico	Hudson River, Europe, Baltic Sea, Black Sea	0.1-31	3-22	5-30	10-30	epibenthic
Mollusca	Gastropoda	Naticidae spp. (juv.)	moon snails	native species present	native species present							
Annelida	Polychaeta	Neanthes succinea	pile worm	native	native	Atlantic Ocean, Gulf of Mexico	Northeast Pacific, Southwest Pacific	2.5-65	5-50	-2-34		Infaunal and epibenthic
Nematoda		Nematoda spp.	nematodes or roundworms	native species present	native species present							
Mollusca	Gastropoda	Nudibranchia spp. (juv.)	sea slugs	native species present	native species present							
Cnidaria	Hydrozoa	Obelia geniculata	hydroid	cryptogenic	cryptogenic	Unknown, cited for the Pacific and Atlantic Oceans	Cryptogenic in all its range; molecular analyses suggest several cryptic species.					epibenthic and planktonic
Crustacea	Ostracoda	Ostracoda spp.	ostracods	native species present	native species present							
Crustacea	Decapoda	Pachygrapsus transversus?	Mottled shore crab	no record/not present	native	Atlantic Ocean (NC to Uruguay, Angola), Mediterranean, Eastern Pacific Ocean (CA to Peru)	Range possibly extended by shipping					epibenthic
Crustacea	Decapoda	Palaemon macrodactylus	Oriental shrimp	introduced	no record/not present	Northwest Pacific (Japan, Korea, China)	Northeast Atlantic, Northeast Pacific, Australia, Argentina	0-34		7-24		epibenthic, on submerged aquatic vegetation
Crustacea	Decapoda	Palaemonetes pugio	Marsh Grass shrimp	native	native	Northwest Atlantic, Gulf of Mexico		0-43	10-25	5-38	18-35	epibenthic, epibiont
Crustacea	Decapoda	Palaemonetes sp.	Grass shrimps	native species present	native species present							
Crustacea	Decapoda	Palaemonetes vulgaris	Daggerblade Grass shrimp	native	native	Northwest Atlantic, Gulf of Mexico		5-35	10-30	5-35	20	epibenthic, epibiont
Crustacea	Copepoda	Paracalanus sp.	calanoid copepod	native species present	native species present	Cosmopolitan		brackish to salt				planktonic
Crustacea	Amphipoda	Paracaprella sp. (juv.)	skeleton shrimp	native species present	native species present							
Crustacea	Copepoda	Paronychocamptus huntsmani	harpacticoid copepod	native	native	Northwest Atlantic						epibenthic, common in salt marshes
Crustacea	Copepoda	Parvocalanus crassirostris	calanoid copepod	native	native	Worlwide range in temperate and tropical waters						panktonic
Crustacea	Decapoda	Penaeidae spp.	prawns	native species present	native species present							
Crustacea	Decapoda	Pinnotheridae spp. (juv.)	pea crabs	native species present	native species present							
Crustacea	Amphipoda	Podocerus brasiliensis	amphipod	no record/not present	native	Cosmopolitan in tropical and warm temperate seas. Northwest Atlantic range: NC to FL	Hawaii, possibly introduced in California harbors					epibiont, tube-building
Annelida	Polychaeta	Polydora cornuta	mud worm	native	native	North Atlantic, Gulf of Mexico	Possibly introduced with oyster culture, ballast water and on hulls in the NE Pacific (BC to CA), NW Pacific (Russia, Japan, Korea), SW Pacific (Australia, New Zealand), and SE Atlantic (Argentina, Brazil)	brackish to salt, 0-30		0-25		epibenthic tube-building, epibiont
Annelida	Polychaeta	Polydora sp.	mud worm	native species present	native species present							
Annelida	Polychaeta	Polydora websteri	shell worm	native	native	Unknown, cited for Northwest Atlantic, Gulf of Mexico, and North Pacific Ocean	New Zealand, Australia, Hawaii; range possibly extended by osyster culture					epibenthic, boring in oysters
Crustacea	Decapoda	Porcellanidae spp. (juv.)	porcelain crabs	native species present	native species present							
Porifera		Porifera spp. indeterminant	sponge	native species present	native species present							
Annelida	Polychaeta	Proceraea cornuta	polychaete or bristle worm	native	cryptogenic	Arctic, North Atlantic (Labrador to NC, Norway to English Channel)	Cryptogenic in the North Pacific (Japan, Washington, California) and the Gulf of Mexico (West Florida Shelf)					epibenthic tube-building, epibiont
Crustacea	Copepoda	Pseudocyclops sp.	calanoid copepod	native	native							epibenthic
Crustacea	Copepoda	Pseudodiaptomus pelagicus	calanoid copepod	native	native	Northwest Atlantic (Nova Scotia to FL), Gulf of Mexico		brackish to euhaline				epibenthic and planktonic
Crustacea	Copepoda	Rhincalanus cornutus	calanoid copepod	native	native	Worlwide range in temperate and tropical waters						planktonic

						Geographical Distribution		Salinity (psu)		Temperature (ºC)		
Phylum	**Class**	**Species/Taxon Name**	**Common Name**	**Chesapeake Bay Invasion Status**	**Status in Texas**	**Native Range**	**Invaded Range**	**Range**	**Optimum**	**Range**	**Optimum**	**Substrate Preference-adults**
Crustacea	Decapoda	Rhithropanopeus harrisii	White-fingered mud crab	native	native	Northwest Atlantic, Gulf of Mexico	Oregon, Northern California coast, San Francisco Bay, Northeast Atlantic, Black Sea, Caspian Sea; Inland lakes of Texas	0-40	0-20		20-31	epibenthic, among oysters
Plantae	Phaeophyceae	Sargassum sp.	Gulfweed	no record/not present	native species present							
Crustacea	Copepoda	Schizopera sp.	harpacticoid copepod	native species present	native species present	Cosmopolitan		brackish to euhaline				infaunal, epibiont
Crustacea	Tanaidacea	Sinelobus stanfordi	tanaid	no record/not present	cryptogenic	Unknown, cited for the Pacific Ocean, Northwest Atlantic, Caribbean, Gulf of Mexico (but not Texas), Southwest Atlantic and Southeast Atlantic	Possibly Northeast Pacific, Southwest Pacific	0-45+	0.5-30			epibenthic
Crustacea	Isopoda	Sphaeromatidae spp. (Dyameninae) (juv.)	marine pillbugs	native species present	native species present							
Annelida	Polychaeta	Streblospio benedicti	polychaete or bristle worm	native	native	Northwest Atlantic, Gulf of Mexico	Northeast Atlantic, Mediterranean Sea, Black Sea, Northeast Pacific	brackish to euhaline				infaunal tube-building
Platyhelminthes	Turbellaria	Stylochus ellipticus	flatworm	native	native	Northwest Atlantic, Gulf of Mexico, Caribbean	Northeast Atlantic	2.5-27	<20			epibenthic, commensal
Crustacea	Isopoda	Synidotea laevidorsalis	isopod	introduced	no record/not present	Northwest Pacific	Northeast Pacific? (SF Bay, California Coast, Willapa Bay), Southwest Pacific, North Atlantic (Europe, US Mid-Atlantic States - Delaware Bay, Chesapeake Bay, SC)	1-35				epibenthic, planktonic
Mollusca	Bivalvia	Tellina agilis	Northern dwarf tellin	native	no record/not present	Nortwest Atlantic (Gulf of St. Lawrence to Georgia)						infaunal
Hexapoda	Insecta	Telmatogeton japonicus	non-biting midge larvae	cryptogenic	cryptogenic	North Pacific Ocean	Northeast Atlantic, Baltic Sea, Australia, and possibly Northwest Atlantic (Newfoundland to Florida), and Gulf of Mexico (Panama City rock jetties)	freshwater to euhaline				Larvae: epibenthic tube-builders on splash zone in rocky intertidal shores
Crustacea	Copepoda	Tisbe sp.	harpacticoid copepod	native species present	native species present	Cosmopolitan						epibenthic
Annelida	Oligochaeta	Tubificoides heterochaetus	tubificid marine worm	cryptogenic	crypotogenic	Northeast Atlantic	Northwest Atlantic, Gulf of Mexico (cryptogenic range)	<0.5-20				infaunal
Platyhelminthes	Turbellaria	Turbellaria spp. (juv.)	flatworms	native species present	native species present							
Crustacea	Decapoda	Xanthidae spp. (juv.)	mud crabs	native species present	native species present							

Species/Taxon Name	Developmental Mode	Feeding Mode	Reference
Acartia sp.			
Acartia tonsa	eggs released; planktonic larvae	omnivore; suspension feeder	Johnson and Allen 2005
Aglaophenia cf. latecarinata	brooder?, planktonic larvae	suspension feeder?	
Alcyonidium cf. albescens	planktonic larvae	suspension feeder	Cohen and Carlton 1995, Winston and Key 1999, Boyd et al. 2002
Algae sp. A			
Algae sp. B			
Alosidae spp. (eggs)			
Alpheus sp. (juv.)			
Ampithoe valida	brooder	herbivore; detritus feeder	Cohen et al. 2002, LeCroy 2002, Cohen and Carlton 1995
Apocorophium lacustre	brooder	detritus feeder; suspension feeder	Bousfield 1973, USGS Nonindigenous Aquatic Species database: http://nas.er.usgs.gov/
Arcidae spp. indeterminant			
Balanus amphitrite	planktonic larvae	suspension feeder	Davidson et al. 2007
Balanus improvisus	planktonic larvae	suspension feeder	Davidson et al. 2006
Blackfordia virginica	asexual reproduction and planktonic medusa	carnivore	Mills and Sommer 1995
Boccardiella ligerica	demersal eggs laid in strings in burrows; planktonic larvae	interface feeder	NEMESIS database: http://invasions.si.edu/nemesis/chesapeake.html
Bowerbankia gracilis	brief planktonic larvae	suspension feeder	Winston 1977, Cohen and Carlton 1995
Calanoida spp. indeterminant			
Calanus sp.	eggs released; planktonic larvae		
Caprella equilibra	brooder	carnivore; omnivore	McCain 1968, CA Fish & Game 2002
Caprella scaura	brooder	carnivore; omnivore	McCain 1968, CA Fish & Game 2002
Caridea spp. (juv.)			
Centropages hamatus	eggs released; planktonic larvae	carnivore; omnivore	Johnson and Allen 2005
Chaetognatha spp.			
Cirolana parva	brooder	herbivore; detritus feeder	
Clausidiidae spp.			
Clytia sp.	asexual reproduction and planktonic medusa	carnivore	Davidson et al. 2007, Cohen and Carlton 1995
Conopeum chesapeakensis	brooder, planktonic larvae (inferred)	suspension feeder	Banta et al. 1995, Davidson et al. 2008, Versar 2008d
Conopeum tenuissimum	planktonic larvae	suspension feeder	Winston 1982
Copepoda spp. 1 indeterminant			
Corycaeus speciosus		herbivore; carnivore (inferred)	
Coullana canadensis		herbivore; detritus feeder; suspension feeder	Johnson and Allen 2005
Crassostrea virginica	planktonic larvae	suspension feeder	Kennedy et al. 1996
Cyclopoida spp.			
Dactylopusia sp.			
Decapoda spp. indeterminant			
Diadumene leucolena	asexual budding and planktonic larvae	carnivore	Cohen and Carlton 1995, CA Fish & Game 2002
Distichlis spicata			
Doridella obscura	egg masses attached to substrate, planktonic larvae	predators of bryozoans	Abbott 1974
Ectinosomatidae spp.			
Edotea triloba	brooder	herbivore	Kunkel 1918
Elasmopus cf. rapax	brooder	herbivore; detritus feeder	LeCroy 2000
Ericthonius brasiliensis	brooder	herbivore; detritus feeder; suspension feeder	Davidson et al. 2007
Ericthonius sp.	brooder	herbivore; detritus feeder; suspension feeder	
Euplana gracilis	demersal eggs, planktonic larvae	carnivore	Hyman 1940, Wolff 2005
Eurypanopeus depressus	brooder, planktonic and benthic larvae	omnivore	Williams 1984
Eurytemora affinis	brooder	filter feeder	Balcer et al. 1984, Saunders 1993, USGS Nonindigenous Aquatic Species database: http://nas.er.usgs.gov/
Eurytemora sp.			
Euterpina acutifrons	brooder	herbivore; suspension feeder	Johnson and Allen 2005
Gammarus daiberi	brooder	detritus feeder; omnivore	Bousfield 1973, Cohen and Carlton 1995, USGS Nonindigenous Aquatic Species database: http://nas.er.usgs.gov/
Garveia franciscana	brooder, planktonic larvae	suspension feeder	Cohen and Carlton 1995, NEMESIS database: http://invasions.si.edu/nemesis/chesapeake.html

Species/Taxon Name	Developmental Mode	Feeding Mode	Reference
Gobiosoma bosc	demersal eggs, planktonic larvae	carnivore (predator of annelids and small crustacea)	Dawson 1969; FishBase: www.fishbase.org
Grandidierella bonnieroides	brooder	herbivore; detritus feeder; omnivore	LeCroy 2002
Halichondria bowerbanki?	planktonic larvae	suspension feeder	Cohen and Carlton 1995
Halicyclops sp.	brooder	herbivore; detritus feeder; suspension feeder	
Hemicyclops sp.		parasitic, commensal	
Incisocalliope aestuarius	brooder	herbivore; detritus feeder; omnivore	Watling and Maurer 1973, Faasse and van Moorsel 2003
Ischadium recurvum	planktonic larvae	suspension feeder	NEMESIS database: http://invasions.si.edu/nemesis/chesapeake.html
Jassa marmorata	brooder	suspension feeder; carnivore	Cohen and Carlton 1995
Labidocera aestiva		carnivore; omnivore	Johnson and Allen 2005
Laticorophium baconi	brooder	herbivore; detritus feeder; suspension feeder	LeCroy 2004, Davidson et al. 2007
Lepas anatifera	planktonic larvae	suspension feeder	
Lucifer faxoni	planktonic larvae		Williams 1984
Macoma balthica	planktonic larvae	interface feeder	Väinölä 2003
Melita nitida	brooder	herbivore; detritus feeder; omnivore	Davidson et al. 2006, Cohen and Carlton 1995, Faasse and van Moorsel 2003
Membranipora tuberculata	planktonic larvae	suspension feeder	Winston 1982
Miraciidae spp.			
Molgula manhattensis	brief planktonic larvae	suspension feeder	Van Name 1945, Cohen and Carlton 1995
Monocorophium acherusicum	brooder	herbivore; detritus feeder; suspension feeder	Cohen and Carlton 1995, LeCroy 2004, Davidson et al. 2007
Mya arenaria	planktonic larvae	suspension feeder	Cohen and Carlton 1995, Global Invasive Species Database: www.invasivespecies.net/database
Mysidae spp.	brooder		
Mytilopsis leucophaeata	planktonic larvae	suspension feeder	Mills et al. 1996, Verween et al. 2007 and references herein
Naticidae spp. (juv.)			
Neanthes succinea	planktonic eggs; planktonic larvae	carnivore; detritus feeder; omnivore	Davidson et al. 2006, Cohen and Carlton 1995
Nematoda spp.			
Nudibranchia spp. (juv.)			
Obelia geniculata	asexual reproduction and planktonic medusa	suspension feeder; carnivore	Davidson et al. 2007
Ostracoda spp.			
Pachygrapsus transversus?	planktonic larvae	carnivore; omnivore	Williams 1984
Palaemon macrodactylus	brooder, planktonic larvae	omnivore; carnivore	Hieb 1999
Palaemonetes pugio	brooder, planktonic larvae	omnivore; carnivore	Williams 1984, Anderson 1985
Palaemonetes sp.			
Palaemonetes vulgaris	brooder, planktonic larvae	omnivore; carnivore	Williams 1984, Anderson 1985
Paracalanus sp.		herbivore; detritus feeder; suspension feeder	Johnson and Allen 2005
Paracaprella sp. (juv.)			
Paronychocamptus huntsmani	brooder		Jeff Cordell (pers. comm.)
Parvocalanus crassirostris		herbivore	Johnson and Allen 2005
Penaeidae spp.			
Pinnotheridae spp. (juv.)			
Podocerus brasiliensis	brooder	herbivore; detritus feeder	Cohen and Chapman 2005, Chapman 2007
Polydora cornuta	brooder, planktonic larvae	interface feeder	Cohen and Carlton 1995 (as Polydora ligni), Zajac and Whitlatch 1982
Polydora sp.			
Polydora websteri	brooder, planktonic larvae	interface feeder	Blake 1971
Porcellanidae spp. (juv.)			
Porifera spp. indeterminant			
Proceraea cornuta	schizogamy, planktonic larvae	carnivore	Uebelacker and Johnson 1984, CA Fish & Game 2002
Pseudocyclops sp.			
Pseudodiaptomus pelagicus	brooder	herbivore; detritus feeder; suspension feeder	Johnson and Allen 2005
Rhincalanus cornutus		herbivore; detritus feeder; suspension feeder	

Species/Taxon Name	Developmental Mode	Feeding Mode	Reference
Rhithropanopeus harrisii	brooder, planktonic and benthic larvae	omnivore	Williams 1984, Carlton 1979, Cohen and Carlton 1995, CA Fish & Game 2002, USGS Nonindigenous Aquatic Species database: http://nas.er.usgs.gov/
Sargassum sp.			
Schizopera sp.	brooder	herbivore; detritus feeder; suspension feeder	
Sinelobus stanfordi	brooder	suspension feeder; detritus feeder	Cohen and Carlton 1995, Davidson et al. 2007
Sphaeromatidae spp. (Dyameninae) (juv.)			
Streblospio benedicti	planktonic larvae	interface feeder	Cohen and Carlton 1995
Stylochus ellipticus	demersal eggs, planktonic larvae	carnivore	Hyman 1940, Kennedy et al. 1996
Synidotea laevidorsalis	brooder	carnivore; omnivore	Chapman and Carlton 1994, Bushek and Boyd 2006, NEMESIS database: http://invasions.si.edu/nemesis/chesapeake.html, but see Poore 1996
Tellina agilis	planktonic larvae	interface feeder	Abbott 1954
Telmatogeton japonicus	aquatic larvae and pupae, adult terrestrial stage	herbivore	Epler 1995, Colbo 1996
Tisbe sp.	brooder	herbivore; detritus feeder; suspension feeder	
Tubificoides heterochaetus	eggs released, direct development	deposit feeder	Harrel 2004, NEMESIS database: http://invasions.si.edu/nemesis/chesapeake.html
Turbellaria spp. (juv.)			
Xanthidae spp. (juv.)			