

Hull fouling is a risk factor for intercontinental species exchange in aquatic ecosystems

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Received 13 March 2007; accepted in revised form 25 May 2007

Abstract

Anthropogenic biological invasions are a leading threat to aquatic biodiversity in marine, estuarine, and freshwater ecosystems worldwide. Ballast water discharged from transoceanic ships is commonly believed to be the dominant pathway for species introduction and is therefore increasingly subject to domestic and international regulation. However, compared to species introductions from ballast, trans-location by biofouling of ships' exposed surfaces has been poorly quantified. We report translocation of species by a transoceanic bulk carrier intercepted in the North American Great Lakes in fall 2001. We collected 944 individuals of at least 74 distinct freshwater and marine taxa. Eight of 29 taxa identified to species have never been observed in the Great Lakes. Employing five different statistical techniques, we estimated that the biofouling community of this ship comprised from 100 to 200 species. These findings adjust upward by an order of magnitude the number of species collected from a single ship. Thus, overall invasion risk from biofouling may be comparable or exceed that of ballast water discharge.

Key words: biofouling, biological invasions, environmental policy, invasive species, risk analysis

Introduction

Invasions of non-indigenous species unintentionally introduced during commercial trade are an environmental concern, industrial hazard, and pathway for the spread of infectious diseases (Mooney et al. 2005). During the twenty years since zebra mussels (*Dreissena polymorpha*) were first introduced to the Great Lakes, most studies of global exchange of aquatic nuisance species have focused on ballast water (Grigorovich et al. 2003, Drake and Lodge 2004, Endresen et al. 2004) and to a lesser extent on the role of sediments that accumulate in ballast water tanks (Bailey et al. 2004a,b). National and international policies aimed at reducing these ship-related risks are now being implemented (e.g., United States National Invasive Species Act of 1996 and the International Convention for

the Control and Management of Ships' Ballast Water and Sediments). However, a third ship-related risk factor — biofouling on ships' hulls and other exterior surfaces — may also be important in causing invasions, but the composition and abundance of fouling communities on ships have seldom been quantified (but see Coutts 1999, Gollasch 2002, Coutts and Taylor 2004, Ruiz and Smith 2005).

Here we report on the biofouling community of a ship that entered dry-dock on Lake Ontario in the fall of 2001 after transoceanic passage from Algiers, Algeria in August and coastal passage from Saint Vincente, Chile in October. From samples scraped directly from the hull, we estimated the total species richness and average individual abundance of the biofouling community. Our results show that compared with ballast water discharge biofouling represents a

greater risk for species introduction (as indicated by the number of species potentially introduced) and a similar risk in terms of the abundance of individuals introduced. This finding implies that even if policies aimed at ballast water and sediment discharge are successful, future invasions may continue to occur at a high rate unless biofouling is also addressed. We urge caution therefore in the adoption and implementation of the International Convention of the Control of Harmful Anti-fouling Systems on Ships, which was passed by the International Maritime Organization in 2001 (cf. Floerl et al. 2005).

Materials and Methods

A bulk carrier with overall length of 164 m and breadth of 22 m was sampled on 20 October 2001 within 2 h of admission to dry-dock on the shore of Lake Ontario. According to the ship's records, the ship had experienced a prolonged stay in Algiers during summer 2001 and a subsequent stay on the coast of Chile before arrival in Lake Ontario via Callao, Peru. The hull was visibly fouled below the waterline (Annex 3). Samples were collected by scraping all material from c. 930 cm² areas at nine locations on the ship: Bilge Keel (port side), Sea Bay Grid (port side), Bottom Shelf (starboard side), Side Shell (port side forward), Side Shell (port side midship), Side Shell (port side aft), Lower Bilge (port side midship), Keel (port side aft), and Keel (starboard side aft).

Specimens were preserved with denatured ethanol, and processed under light microscopy. All specimens were identified to the lowest feasible taxonomic level. References consulted for taxonomic identification were Abé (1998), Ahlstrom (1940, 1943), Angel (1993), Athersuch et al. (1989), Balcer et al. (1984), Barnes (1968), Barr (1994), Boxshall (1977), Brooks (1957), Chu (1949), Comstock (1967), Coull (1977), Czaika (1982), Darwin (1854), Davis (1955), Davis (1968), De Melo and Hebert (1994), Dumont and Negrea (2000), Dussart and Defaye (2001), Edmondson (1959), Epler et al. (2000), Gonzalez and Bowman (1965), Green and Macquitty (1987), Hickman et al. (1984), Hudson et al. (1998), Huys et al. (1996), Korovchinsky (1992), Kranz (1970, 1978), Mason (1973), Newman and Ross (1976), Nishida (1985), Ohtsuka et al. (1996), Owre and

Foyo (1967), Parrish (1975), Pennak (1978, 1989), Rivier (1998), Rose (1933), Smirnov (1996), Taylor et al. (2002), and Thorp and Covich (2001). Because many specimens could not be identified to the species level, some species may be lumped. Our calculations therefore estimate a minimum bound on the true species richness.

We used five statistical estimators of species richness to extrapolate from the observed list of distinct taxa to the total richness of the community from which the specimens were sampled:

(1) *Homogeneous model*. As a baseline, we adopt a homogeneous model assuming equal abundance and equal catchability (eqn. 2.1 in Chao et al. 2000, cf. Chao and Lee 1992). This model tends to underestimate the true number of species (Chao and Lee 1992). The remaining models represent different approaches to relaxing this restrictive assumption.

(2) *Chao1*. This nonparametric estimator, which is based on the idea that the abundance of rare species contains most information about the number of species missing in the sample, was originally proposed as an estimator of the lower bound of the true species richness (Chao 1984). Recent results justify a more general interpretation, i.e. as a nearly unbiased estimator (Shen et al. 2003, cf. Chao 2005)

(3) *Abundance-based Coverage Estimator* (ACE). This estimator, based on the notion of sample coverage as the sum of the probabilities of the observed classes (Good 1953) was first proposed by Chao and Lee (1992) to relax the homogeneous assumption of the first model. Like the Chao1 estimator, this model exploits the information represented by the rare species and appears to perform well, even for species with rather heterogeneous (fat-tailed) species abundance distributions (Chao and Lee 1992).

(4) and (5) *First and Second Order Jackknife*: These estimators were derived by Burnham and Overton (1978, 1979) in the context of capture-recapture theory to reduce negative bias resulting from heterogeneity in individual catchability. In the context of species richness estimation, this corresponds to the fact that since some species are likely not to be collected, the total number of observed species is an underestimate of the true number of species. Numerical experiments failed to uncover a general rule for choosing between the different orders of the estimator (Burnham and Overton 1978), though there was a negative correlation between sample size and the order

resulting in minimum error (Burnham and Overton 1979).

See Chao (2005) for a more detailed comparison of these estimators. Estimation was performed using SPADE v. 2.1 (Chao and Shen 2003). Following Chao et al. (1993, 2003) we adopt 10 as a cutoff separating “rare” from “abundant” species. Heuristics for choosing among estimators include adopting a modification of ACE for highly heterogeneous communities (Chao and Lee 1992) in the event that the estimated coefficient of variation (c.v.) exceeds 0.8 (SPADE User’s Manual; Chao and Shen 2003), as in this case where the estimated c.v. was 1.02. Accordingly, the reported result corresponds to eqn 2.15 in Chao and Lee (1992) rather than eqn 2.14.

Results

We obtained 944 organisms from 9 samples covering a total of 0.836 m² resulting in an average density of 1129 individuals m⁻². The specimens represented 74 distinct taxa from seven phyla and at least 15 classes (Figure 1). The most common species belonged to the phylum Arthropoda (77%) followed by Cnidaria (17%). Within the arthropods (Figure 2), the class Maxillopoda dominated (56%), represented mostly by copepods, followed by Branchiopoda (39%).

Twenty-nine taxa were identified to species (Annex 1). The individuals identified to species accounted for 51% ($n = 482$) of all specimens. Of the 29 identified species, 23 inhabit fresh waters (Annex 1). Seven and 22 species, respectively, reproduce parthenogenetically and sexually (Annex 1). Eight of the 29 species have not been documented in the Great Lakes (Table 1 in Balcer et al. 1984; <http://www.glsr.usgs.gov/greatlakescopepods/>). Of these, two – *Acanthocyclops exilis* and *Oithona similis* – are freshwater species. Both are cyclopoid copepods. *A. exilis* belongs to the family Cyclopidae while *O. similis* is a member of the Oithonidae. The remaining specimens that could not be identified to species represented 7 phyla and at least 13 classes (Annex 2).

The rank-abundance distribution of all the specimens was extraordinarily fat-tailed (Figure 3), indicating that most groups are represented by only one (singletons) or two (doubletons) individuals. Estimates of the number of distinct

taxa in the community ranged from 94.6 to 196.8, depending on the estimator (Figure 4).

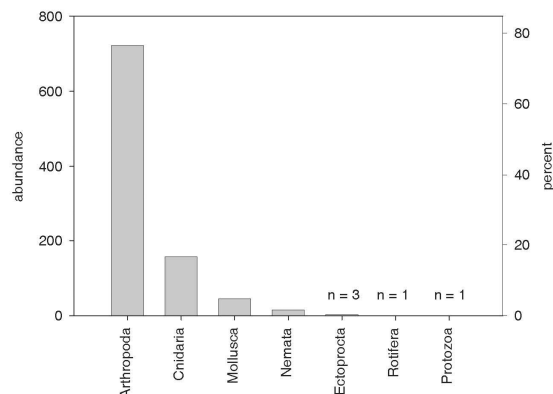


Figure 1. Relative abundance of phyla in 944 individuals collected from a biofouling community on a ship in the Great Lakes

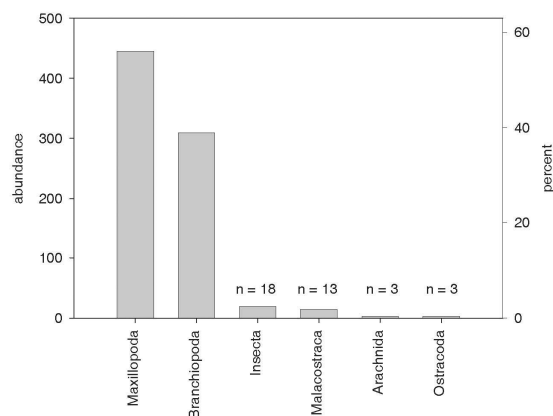


Figure 2. Relative abundance of arthropod classes collected from a biofouling community on a ship in the Great Lakes

The lowest estimate is from the homogeneous model, and is almost surely an underestimate. The abundance-based coverage model is probably the most reliable estimator and also makes the highest prediction. Comparing the results of the different estimators suggests that the total species richness of the community was likely greater than 100 and could plausibly be as large as 350. Estimates of the predicted number of new species that would be collected in a further survey of equal size (944 individuals) range from 11.5 for the estimator of Boneh et al. (1998) to 26.2 for the estimator of Shen et al.

(2003). A further survey increasing sampling effort 10-fold (9440 individuals), would find an estimated 16.6 (Boneh et al. 1998) to 62.2 (Shen et al. 2003) additional species.

Discussion

Our results suggest that biofouling communities represent a considerable risk factor for biological invasion. We collected 74 distinct taxa and estimate the total species richness on the hull of one ship as between 100 and 200 species. This finding adjusts upward by an order of magnitude the maximum number of species observed in a fouling community and therefore also the maximum potential risk posed by this pathway.

Although fouling communities are increasingly recognized as a continuing source of non-indigenous species (Fofonoff et al. 2003, Minchin and Gollasch 2003), few studies have quantified this risk (Coutts 1999, Gollasch 2002, Coutts and Taylor 2004, Ruiz and Smith 2005). Of those that have, Gollasch (2002) found between 1 and 15 species on each of 127 vessels calling on German ports between 1992 and 1995, while Coutts and Taylor (2004) found between 1 and 11 broad taxonomic groups on each of 30 vessels in New Zealand. In another study, Coutts (1999) found between 0 and 16 animal taxa (47 species total) on each of 21 vessels visiting Bell Bay and Long Reach in northern Tasmania. Finally, Ruiz and Smith (2005) found up to 20 animal taxa on 8 of 9 ships calling on Oakland, California, USA. Thus, our study finds more species by about an order of magnitude than previous studies.

Indeed, fouling may represent an even greater source of non-indigenous species than ballast water. For comparison, Carlton and Geller (1993) collected 369 taxa from the ballast water of 159 ships entering Coos Bay, Oregon from Japan and estimated typical species richness to be 20-30 species per ship. Similarly, Chu et al. (1997) collected between 2 and 42 species in each of 12 ballast water tanks from 5 container ships entering Hong Kong. A study of ballast water zooplankton in 34 ships entering three ports in Nova Scotia found between 0 and 16 taxa per ship (Carver and Mallett 2001). In the Great Lakes, Locke et al. (1991, 1993) found between 3 and 34 species (mean: 12.8, s.d.: 8.8) in ballast water collected from 15 ships entering the Great Lakes or upper St. Lawrence River in

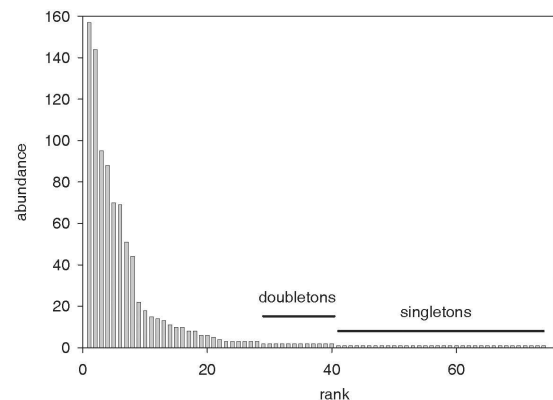


Figure 3. Species rank-abundance distribution of uniquely identified taxa collected from the biofouling community of a ship in the Great Lakes; x-axis: species rank, y-axis: abundance

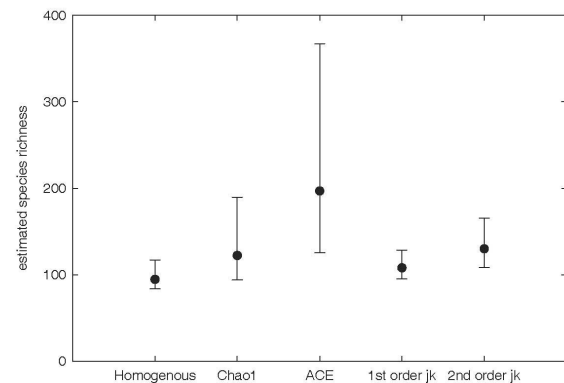


Figure 4. Species richness of a biofouling community on a ship in the Great Lakes estimated using five statistical estimators. Bars are 95% confidence intervals

1990, while a study commissioned by the Canada Coast Guard collected between 0 and 12 distinct taxa in 63 collections made from 58 ocean-going ships entering the Great Lakes (Aquatic Sciences, Inc., 1996). Most ocean-going ships calling on Great Lakes ports were estimated by Aquatic Sciences, Inc. (1996) to carry less than 8 species. Thus, on a per ship basis our data show that biofouling communities can represent an order of magnitude more taxa than ballast water.

Our study was not noticeably more thorough than these studies, though the methods of each were different. Thus, we suspect the different estimates of species richness represent the true range of richness in oceangoing vessels. Fouling is known to vary dramatically, depending on numerous factors including the condition of anti-fouling paint, the average speed of the vessel, the source of the vessel, and biological factors like

spawning and seasonal variation in species abundance (Minchin and Gollasch 2003, Lewis et al. 2004, Ruiz and Smith 2005). The ship sampled in this study was heavily fouled and is probably not representative. However, as with ballast water discharge (Drake et al. 2005) it is likely the atypical cases that introduce the most species and pose the greatest threat.

Extrapolating from the sampled abundance (1129 m^{-2}) to the total abundance of the fouling community requires estimating the percentage of the ship hull that is fouled and the surface area below the waterline. Coutts and Taylor (2004) found fouling (including both animals and algae) of 30 vessels photographed in New Zealand waters to range from 10.9% to 86.5% of the available surface. For typical vessel width of 30 m and lengths of 150-200 m (e.g., Coutts 1999), conservatively estimating the subsurface area as the product of length and width results in a range of 4500-6000 m^2 for the exposed underwater surface area. An estimate of the total abundance of the fouling community is the product of available area, percentage fouled, and average density. Using these minimum and maximum values to obtain lower and upper bounds, we estimate the minimum and maximum abundance of fouling organisms for a typical ship to be 5.85×10^5 and 6.19×10^6 , respectively. The ship sampled in this study had an underwater surface area of approximately 5200 m^2 , of which about 20% was fouled, for a total potential inoculation of 1.17×10^6 individuals. For comparison, volumes of ballast water actually discharged range from tens to thousands of metric tons (National Ballast Information Clearinghouse, <http://invasions.si.edu/nbic/>) and densities of organisms in untreated ballast water are typically less than 3,000 m^{-3} , but may be $>50,000 \text{ m}^{-3}$ (Minton et al. 2005). Using data on specimens collected from ballast tanks of ships arriving in four North American ports (Baltimore, MD; Coos Bay, OR; Norfolk, VA; and Valdez, AK), Minton et al. (2005) estimate the inter-quartile range in the abundance of organisms discharged in untreated ballast water to be 2.63×10^5 - 9.83×10^6 . Thus, it appears that biofouling presents a similar risk to ballast water discharge in terms of organism abundance. The chance of introduction (from release and/or reproduction) and the chance of survival may differ between organisms in ballast and those in the intact fouling community, but it is not clear that the overall per organism risk of establishment is

lower in fouling communities compared to organisms in ballast. Without further quantification of these later steps of invasion, the reasonable conclusion is that the two pose roughly similar risks.

We have shown that hull fouling is a significant risk factor for biological invasions. From the standpoint of management and policy, the problem is exacerbated by the relatively few options for control compared with ballast water (Claudi and Ravishankar 2005, Minton et al. 2005). Anti-fouling paint is currently the primary method of controlling fouling. Since the 1960s, tributyltin (TBT) has been the primary biocide in anti-fouling paint, and as of the year 2000 was used on greater than 70% of the world's ocean going fleet (Evans et al. 2000). However, because of its ecotoxicity, a global prohibition on the application of TBT is proposed by the International Convention of the Control of Harmful Anti-fouling Systems on Ships, which was passed by the International Maritime Organization in 2001. If ratified, the Convention would ban the use of TBT on ships after 2008.

The Convention has already motivated a search for alternative anti-fouling coatings (Yebra et al. 2004), but many substitutes also are toxic (Karlsson and Eklund 2004), and justifiable concern exists about economic losses (e.g., in decreased fuel efficiency, increased air pollution) if good alternatives cannot be applied (Champ 2000). In addition, costs of the introduction of invasive species if less effective anti-fouling coatings are used have been inadequately considered in the overall evaluation of the Convention (Nehring 2001, Lewis et al. 2004). To take legal effect, the Convention must be ratified by 25 countries that represent at least 25% of the world's merchant shipping tonnage, which has not yet occurred (<http://www.imo.org>). In combination with the few previous quantifications of fouling communities on ships, our results suggest that TBT should not be abandoned without a more careful consideration of the potential trade-offs, including the potential for increased harmful biological invasions.

Acknowledgements

Philip Jenkins and Aquatic Sciences Incorporated sampled the ship. James Vancil identified the specimens. With thank R. Schwartz and

A. Silletti for research and editorial assistance. Funding was provided by an EPA STAR award (to DML), the NSF-funded (DEB 02-13698) Integrated Systems for Invasive Species project (to DML), a Sabbatical Fellow-ship (to DML), and an EPA STAR graduate research fellowship (to JMD). This paper was written while Drake was a Postdoctoral Associate at the National Center for Ecological Analysis and Synthesis, a center funded by NSF (DEB-0072909), the University of California, and the Santa Barbara campus. Although the research described in this article has been funded in part by the United States Environmental Protection Agency through a STAR grant/cooperative agreement to DML, it has not been subjected to the Agency's peer and policy review and, therefore, does not necessarily reflect the views of the Agency and no official endorsement should be inferred.

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Species exchange from biofouling

Annex 1

Hull fouling specimens identified to species. Species that have not been observed in the Laurentian Great Lakes are indicated with an asterisk. All species belong to phylum Arthropoda

| Class/Order/Family/Species | Abundance | Mating system | Habitat |
|---|-----------|-----------------|---------------------|
| Branchiopoda/Cladocera/Bosminidae | | | |
| <i>Bosmina liederi</i> De Melo and Hebert, 1994 | 69 | parthenogenesis | freshwater |
| <i>Eubosmina coregoni</i> Baird, 1857 | 95 | parthenogenesis | freshwater |
| Branchiopoda/Cladocera/Chydoridae | | | |
| <i>Monospilus dispar</i> Sars, 1862 | 1 | parthenogenesis | freshwater |
| Branchiopoda/Cladocera/Daphnidae | | | |
| <i>Ceriodaphnia pulchella</i> Sars, 1862 | 8 | parthenogenesis | freshwater |
| <i>Daphnia retrocurva</i> Forbes, 1882 | 88 | parthenogenesis | freshwater |
| Branchiopoda/Cladocera/Holopedidae | | | |
| <i>Holopedium gibberum</i> Zaddach, 1855 | 8 | parthenogenesis | freshwater/brackish |
| Branchiopoda/Cladocera/Leptodoridae | | | |
| <i>Leptodora kindtii</i> Focke, 1844 | 1 | parthenogenesis | freshwater |
| Maxillopoda/Calanoida/Diaptomidae | | | |
| <i>Leptodiaptomus siciloides</i> Lilljeborg, 1889 | 2 | sexual | freshwater |
| Maxillopoda/Calanoida/Parapontellidae | | | |
| <i>Parapontella brevicornis</i> * Lubbock, 1857 | 1 | sexual | marine |
| Maxillopoda/Calanoida/Pseudocyclopidae | | | |
| <i>Pseudocyclops crassiremis</i> * Brady, 1878 | 2 | sexual | marine |
| Maxillopoda/Calanoida/Ridgewayiidae | | | |
| <i>Placocalanus insularis</i> * Fosshagen, 1970 | 1 | sexual | marine |
| Maxillopoda/Calanoida/Temoridae | | | |
| <i>Eurytemora affinis</i> Poppe, 1880 | 144 | sexual | euryhaline |
| <i>Temora turbinata</i> * Dana, 1849 | 1 | sexual | marine |
| Maxillopoda/Cyclopoida/Cyclopidae | | | |
| <i>Tropocyclops prasinus</i> Fisher, 1860 | 1 | sexual | freshwater |
| <i>Acanthocyclops exilis</i> * Coker, 1934 | 2 | sexual | freshwater |
| <i>Diacyclops navus</i> Herrick, 1882 | 1 | sexual | freshwater |
| <i>Diacyclops thomasi</i> S.A. Forbes, 1882 | 22 | sexual | freshwater |
| <i>Diacyclops nanus</i> G.O. Sars, 1863 | 3 | sexual | freshwater |
| <i>Eucyclops prionophorus</i> Kiefer, 1931 | 2 | sexual | freshwater |
| <i>Macrocyclus albidus</i> Jurine, 1820 | 1 | sexual | freshwater |
| <i>Microcyclus rubellus</i> Lilljeborg, 1901 | 3 | sexual | freshwater |
| <i>Orthocyclops modestus</i> Herrick, 1883 | 1 | sexual | freshwater |
| Maxillopoda/Cyclopoida/Oithonidae | | | |
| <i>Oithona similis</i> * Claus, 1866 | 1 | sexual | euryhaline |
| Maxillopoda/Harpacticoida/Ameiridae | | | |
| <i>Nitokra hibernica</i> Brady, 1880 | 6 | sexual | euryhaline |
| Maxillopoda/Harpacticoida/Canthocamptidae | | | |
| <i>Canthocamptus robertcokeri</i> M.S. Wilson, 1958 | 3 | sexual | freshwater |
| <i>Mesochra alaskana</i> M.S. Wilson, 1958 | 11 | sexual | euryhaline |
| Maxillopoda/Harpacticoida/Harpacticidae | | | |
| <i>Harpacticus chelifer</i> * O.F. Müller, 1785 | 2 | sexual | marine |
| Maxillopoda/Poecilostomatoida/Corycaeidae | | | |
| <i>Farranula carinata</i> * Giesbrecht, 1891 | 1 | sexual | marine |
| Insecta/Diptera/Chironomidae | | | |
| <i>Cricotopus tricinctus</i> Epler, 2000 | 1 | sexual | freshwater |

Annex 2

Hull-fouling specimens not identified to species

| Phylum/Class/Order/Family (Genus) | Abundance |
|---|-----------|
| Protozoa | |
| Granuloreticulosea/Foraminiferida | 1 |
| Rotifera | |
| Eurotatoria/Ploima/Brachionidae (<i>Keratella sp.</i>) | 1 |
| Nemata | 15 |
| Ectoprocta | 1 |
| Gymnolaemata/Ctenostomata | 2 |
| Cnidaria | |
| Anthozoa | 157 |
| Mollusca | |
| Bivalvia | 44 |
| Gastropoda | 1 |
| Arthropoda | |
| Arachnida/Oribatida | 1 |
| Arachnida/Trombidiformes/Halacaridae | 2 |
| Branchiopoda/Cladocera/Macrothricidae(<i>Discoidae</i> (sp.1)) | 1 |
| Branchiopoda/Cladocera/Macrothricidae(<i>Discoidae</i> (sp.2)) | 1 |
| Branchiopoda/Diplostraca | 1 |
| Branchiopoda/Diplostraca/Bosminidae (<i>Bosmina sp.</i>) | 1 |
| Branchiopoda/Diplostraca/Cercopagididae (<i>Cercopagis sp.</i>) | 5 |
| Branchiopoda/Diplostraca/Chydoridae | 2 |
| Maxillopoda (Infraclass Cirripedia) | 51 |
| Maxillopoda (Subclass Copepoda) | 4 |
| Maxillopoda/Calanoida | 70 |
| Maxillopoda/Calanoida/Aetideidae | 1 |
| Maxillopoda/Calanoida/Arietellidae | 1 |
| Maxillopoda/Calanoida/Clausocalanidae | 1 |
| Maxillopoda/Calanoida/Diaptomidae (<i>Diaptomus sp.</i>) | 1 |
| Maxillopoda/Calanoida/Paracalanidae | 1 |
| Maxillopoda/Calanoida/Scolecitrichidae | 1 |
| Maxillopoda/Cyclopoida | 10 |
| Maxillopoda/Cyclopoida/Cyclopidae (<i>Acanthocyclopsn sp.</i>) | 2 |
| Maxillopoda/Cyclopoida/Cyclopidae (<i>Diacyclops sp.</i>) | 3 |
| Maxillopoda/Cyclopoida/Cyclopidae (<i>Paracyclops sp.</i>) | 2 |
| Maxillopoda/Cyclopoida/Oithonidae (<i>Paroithona sp.</i>) | 1 |
| Maxillopoda/Harpacticoida/Ameiridae (<i>Ameira sp.</i>) | 3 |
| Maxillopoda/Harpacticoida/Ameiridae (<i>Nitokra sp.</i>) | 1 |
| Maxillopoda/Harpacticoida/Tisbidae | 2 |
| Maxillopoda/Sessilia | 3 |
| Maxillopoda/Sessilia/Balanidae | 6 |
| Maxillopoda/Sessilia/Chthamalidae | 1 |
| Maxillopoda/Sessilia/Chthamalidae (Subfamily Chthamalinae) | 18 |
| Maxillopoda/Sessilia/Chthamalidae (<i>Euraphia sp.</i>) | 10 |
| Ostracoda | 2 |
| Ostracoda/Podocopida | 1 |
| Malacostraca/Amphipoda/Lysianassidae | 13 |
| Insecta | 1 |
| Insecta/Diptera/Chironomidae | 1 |
| Insecta/Diptera/Chironomidae (Subfamily Chironominae) | 14 |
| Insecta/Diptera/Chironomidae (<i>Brillia sp.</i>) | 1 |

Species exchange from biofouling

Annex 3

Images of hull fouling: A, B - heavy fouling by algae and crustaceans was observed in a belt, approximately 2 m wide, below the waterline; C, D, E - the bottom shell was visibly encrusted with crustaceans in patches of varying sizes representing approximately twenty percent of the total surface; F - heavy fouling was observed in and around seabays (samples could not be collected from inside seabays but were taken from adjacent surfaces)

