**Ballast Water Exchange: Efficacy of treating ships' ballast water
to reduce marine species transfers and invasion success?**

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INTRODUCTION

Marine Biological Invasions: Current State of Knowledge

Biological invasions are a potent force of ecological and evolutionary change, which also affects many dimensions of society. Invasions occur when species establish self-sustaining populations beyond their historical geographic ranges. Such nonindigenous species (NIS) have contributed to many conspicuous changes, including increased frequency of fires, declines in threatened and endangered species, loss of agricultural products and fisheries resources, outbreaks of emerging and reemerging diseases, and changes in food web structure and nutrient cycling (e.g., Wilcove et al. 1997, Mack et al. 2000, Pimentel et al. 2000). It is evident that biological invasions are transforming the structure and function of ecosystems throughout the world.

Marine invasions have received relatively little attention compared to terrestrial and freshwater communities, but the consequences of invasions are no less evident in marine systems than elsewhere (Carlton 1989, 2001, National Research Council 1995, Pew Ocean Commission 2003). Approximately 500 marine and estuarine NIS are known for the coastal U.S., and over 200 of these can occur in a single estuary (Cohen and Carlton 1995, Ruiz et al. 2000). Marine invasions are now known in most regions of the world; and where studied, NIS appear to be common (e.g., Australia: >100 species; Mediterranean Sea: > 200 species; Baltic Sea: 36 species; see reviews by Ruiz et al. 1997, 2000 and references therein; see also Hewitt et al. 1999). As in terrestrial and freshwater systems, many of these species become numerically dominant in invaded communities, where they can have significant impacts on population, community, and ecosystem-level processes (e.g., Cloern 1996, Ruiz et al. 1999, Grosholz et al. 2000, 2002).

The rate of new invasions appears to have increased dramatically in the past few centuries, often exhibiting an exponential increase across many geographic regions (Cohen and Carlton 1995, 1998, Reise et al. 1999, Hewitt et al. 1999, Ruiz et al. 2000). The observed increase is driven by human activities, which create a diverse array of transfer mechanisms (vectors) that move organisms throughout the world. The globalization of trade over the past century has resulted in an increasing number of vectors, transferring organisms among more source and recipient regions at increasing rates (Ruiz and Carlton 2003). Changes in recipient environments (e.g., human-mediated disturbances) also may have contributed to changes in susceptibility to invasions, which could operate to increase invasion rates (Carlton 1996, Simberloff and VonHolle 1999). The observed increase in invasion rate may be inflated somewhat by increase in search effort in recent years, whereby more effort and improved detection techniques have been applied over time. Nonetheless, it is evident that invasion rates are increasing for many conspicuous taxonomic groups (e.g., molluscs), which are least prone bias in search effort, and that human-mediated supply of organisms has increased greatly through time (Ruiz et al. 2000).

Historically, the introduction of marine NIS has resulted from (1) movement of organisms on the bottom of ships, (2) movement and/or intentional release of aquaculture and fisheries species along with their rich assemblage of associated organisms, (3) release of species associated with pet industries or management, and (4) release of organisms in the ballast materials of ships (Elton 1958, Carlton 1979, 1987, 1989, 1992). Although most of these transfer mechanisms (or vectors) remain active today, the relative importance of shipping appears to have increased and contributed most strongly to the overall increase in rate of known invasions (Carlton 1979, Carlton and Geller 1993, Mills et al. 1993, Cohen and Carlton 1995, Ruiz et al. 2000).

Today, the global movement of ballast water is considered the single largest transfer mechanism for marine NIS. Since the 19th century, ships have used ballast water for stability, discharging water both at ports of call and en route (Carlton 1985). Ports can receive relatively large volumes of ballast water, originating from source regions throughout the world. For example, the United States and Australia each receive annually >79 million metric tons of ballast water on ships arriving from foreign ports (Kerr 1994, Carlton et al. 1995). A taxonomically diverse community of organisms is entrained and transported within ballast tanks, resulting in many successful invasions of nonindigenous species at ports throughout the world (e.g., Carlton and Geller 1993, Cohen and Carlton 1995, Smith et al. 1999).

Mid-ocean exchange is currently the only treatment available for commercial and military ships to reduce the quantities of non-indigenous coastal plankton in ballast water (National Research Council 1996). Ballast water exchange consists of flushing coastal water from ballast tanks, replacing it with oceanic waters. This is intended to reduce the concentration of coastal organisms, which may become established in subsequent ports upon ballast discharge; in contrast, most oceanic organisms are considered unlikely to colonize coastal habitats, just as many coastal organisms cannot persist in open-ocean.

Despite the dominant role of shipping in the transfer and establishment of marine NIS, and rapidly emerging management strategies to reduce species transfers in ballast water, there remain many fundamental questions about ship-mediated invasions:

1. ***The relative contribution of ballast water versus hull fouling to invasions established by the shipping vector is unresolved***. Some invasions are clearly attributable to ballast water, and others are considered to result from hull fouling (for which transfer by ballast water is highly improbable). However, for a majority of shipping invasions, ballast water and hull fouling are both possible sources, because the life-cycle of many organisms include benthic and planktonic forms (Fofonoff et al. 2003). Ballast water clearly transfers and discharges many more organisms (in terms of sheer numbers) into recipient environments than associated hull communities. Since invasion success is density-dependent, ballast water is the likely source of most invasions for which both mechanisms are possible. Nonetheless, the relative contribution of each remains unresolved for a large majority of ship-mediated invasions.

2. ***The efficacy of ballast water exchange and other treatments to reduce the transfer of non-native organisms in ships' ballast is poorly quantified***. Although it is evident that flushing out ballast tanks in open-ocean will serve to reduce the initial concentration of coastal organisms, few data exist to evaluate the magnitude of this reduction (or efficacy of exchange). Further, of the limited data that does exist, there is a wide range of reported values, creating some confusion. This results in part from different methods, both for the exchange and measurement of effects, some of which do not control adequately for confounding variables.

3. ***The efficacy of ballast water exchange or other treatments to reduce the likelihood of new invasions remains unknown***. Although reductions in transfer of coastal organisms in ballast water will most certainly reduce the likelihood of invasions, the magnitude of this reduction is not clear. In other words, the quantitative relationship between dosage and invasion success is not known, so it is difficult to know where critical thresholds lie and to establish a scientifically-based "standard" for acceptable concentrations of residual organisms in treated ballast water. This issue is further complicated by the possible transfer of many organisms by ships hulls and some uncertainty about the relative contribution to hulls in the overall shipping vector (#1 above).

Ballast Water Delivery and Management for Prince William Sound, Alaska

Oil tankers arriving to Prince William Sound (PWS) have delivered approximately 17 million cubic meters of clean (i.e., non-oily) ballast water on an annual basis, making this port one of the largest recipients of ballast water in the nation (Carlton et al. 1995, Ruiz and Hines 1997, Hines and Ruiz 2000). Since 1997, Tankers arriving to PWS from foreign ports have been required to undertake mid-ocean ballast water exchange (BWE) prior to ballast discharge. However, most ballast water (>85%) is delivered from domestic ports in San Francisco Bay (CA), Long Beach (CA), and Puget Sound (WA); although these ships deliver relatively high concentrations of planktonic organisms, including a diverse collection of species (including several that are not native to North America), they are not presently required to exchange or otherwise treat this ballast water (Hines and Ruiz 2000).

More broadly, the National Invasive Species Act of 1996 (NISA) requested that vessels arriving from outside of the Exclusive Economic Zone (EEZ) voluntarily conduct open-ocean exchange of ballast tanks to be discharged in U.S. ports. In 2004, U.S. Coast Guard made BWE mandatory for such foreign arrivals. California has passed and implemented a separate law (Assembly Bill 703), requiring mid-ocean exchange or other treatments by all commercial vessels that arrive from outside the U.S. EEZ intending to discharge within the state. Several other states (Washington, Maryland, Virginia, and Michigan) have also passed legislation, and are implementing regulations, on the control and management of ballast water. Further, international standards for ballast water treatment have also advanced through a diplomatic convention at the International Maritime Organization in 2004, and this treaty is awaiting ratification my member countries.

State, federal, and international efforts are all clearly promoting technology development to treat ballast water, to overcome some of the logistical constraints of BWE that exist for some ship types, routes (especially coastal voyages of limited duration), and sea conditions. Such technology development and demonstration is still at an early stage. No technology is approved for use by U.S. Coast Guard to meet its requirements for ballast water management. It is likely to be several years before technologies are available and widely used. In the interim, BWE is viewed broadly as a stop-gap measure that is immediately available for use on most ships and that will be in use for the next decade, being gradually phased out by the world's fleet as technologies become available.

Ships practice two basic types of ballast water exchange that replaces coastal with oceanic water: Flow-Through Exchange, in which oceanic sea water is pumped continuously through a ballast tank to flush out coastal water of the source port; and Empty-Refill Exchange, in which a ballast tank is first emptied of coastal water and then refilled with oceanic water. Each method may vary in the efficiency of exchange due to amount of water pumped and to practical constraints in plumbing and tank configuration. For example, Flow-Through Exchange has the effect of dilution but not complete replacement of ballast water, and during Empty-Refill Exchange ballast tanks cannot be completely emptied so that a residual of coastal plankton may remain. To maximize the degree of exchange, multiple exchanges (300%) are recommended for Flow-Through Exchange.

Despite current public perception, and a rapidly advancing set of regulations and guidelines, the effects of BWE remain largely untested. In this study, we conducted a series of experiments aboard oil tankers arriving to Port Valdez, to quantify the efficacy of BWE in reducing transfer of coastal organisms (#2 above).

METHODS

Voyages

We conducted exchange experiments aboard commercial oil tankers en route to Valdez, Alaska from three different port systems in the western North America: Long Beach (California), San Francisco (California), and Puget Sound (Washington). These three port systems are the source ports for the majority of tanker arrivals and ballast water discharge to Valdez, representing approximately 88% of tanker arrivals and 87% of tankers' discharge volume (Hines and Ruiz 2000).

Tankers arriving to PWS from these ports generally arrive loaded with ballast water, as they carry cargo (oil) out of Port Valdez and return with ballast tanks relatively full. Most ballast water (about 67%) is carried in clean, dedicated ballast tanks, which are loaded at the last port of call prior to arrival in PWS.

For the experiments, we selected eight voyages to include these source ports and multiple vessels, in order to encompass a range of conditions that may influence BWE and the dynamics of plankton communities during transit. In particular, the size of vessels, configuration and volume of ballast tanks, and voyage duration differed among voyages (Table 1).

All experimental voyages occurred in the summer months of June and July (1998-1999) to coincide with a period of relatively high concentrations of zooplankton for western North America. Despite controlling for season, coastal plankton communities are highly variable in both space and time, such that concentrations and species composition differed among voyages.

Experimental Design & Sample Collection

The experiment consisted of a paired design, which was replicated on the eight voyages. On each voyage, we compared changes in one or more ballast tanks that underwent experimental treatment (BWE) to a control tank that did not. The latter tank controlled for changes in the ballast water over time on each voyage, independent of exchange treatment. Six voyages included the two different types of treatment, both empty-refill and flow-through BWE, and the other two voyages included only Flow-Through BWE (see Table 1). For voyages with only one exchange treatment, the ships were routinely unable to conduct empty-refill BWE, due to safety concerns.

Ships were asked to nominate ballast tanks for the experimental voyages, and treatments were assigned randomly among tanks on each voyage. Tanks were selected to be similar in size and shape, including paired port and starboard wing tanks. Ships filled their ballast tanks at the departure port. During the filling process, rhodamine water-tracing dye (an inert and non-toxic dye) was added to each tank and allowed to mix, to achieve a concentration of 100 µg per liter.

Ships were asked to perform BWE during the voyage to Alaska. All exchange occurred at least 100 nautical miles from land. Experimental treatment tanks underwent either 100% empty-refill or 300% flow-through BWE. For empty-refill BWE, ships emptied tanks of coastal water and reballasted with oceanic water, constituting approximately 100% volumetric exchange. For flow-through BWE, ships pumped water into the tanks, in an estimated amount equivalent to 300% of the tank volume. The respective volumes for empty-refill and flow-through methods correspond to those recommended by U.S. Coast Guard and the International Maritime Organization, for ships arriving from overseas.

Table 1. Description of experimental BWE voyages.

Ship	Depart Date	Depart Port	Length of Voyage (days)	Control Tank Salinity		Port Salinity		Vessel Details			Type of BWE	
				Sfc.	Deep	0 m		Gross tons (LT)	Tank vol. (m³)	Tow Ht (m)	ER	FT
SR Baytown I	27/06/98	Benicia	5	1	1	0		32136	3112 – 4512	15.6	Y	Y
SR Benicia	01/07/98	Benicia	6	2	2	1		75272	11036	22	Y	Y
SR Long Beach 1	09/07/98	Benicia	4	3	3	0		94999	10863 – 18066	25	Y	Y
SR Long Beach 2	19/07/99	Benicia	4	10	10	5		94999	10863 - 18066	25	Y	Y
SR Baytown II	14/06/99	Anacortes	3	30		30		32136	3112 – 4512	15.6	Y	Y
SR Baytown III	10/07/99	Anacortes	1	30		29		32136	3112 – 4512	15.6	Y	Y
ARCO Independence	18/07/99	Long Beach	8	33	33	33		117515	13109 / 13109	25 / 25	N	Y
ARCO Spirit	16/06/99	Long Beach	6	35	35	36		117515			N	Y

We measured the effect of 100% empty-fill exchange and 100% flow-through exchange by sampling all tanks (including the control tank) before and after exchange. In addition, we measured the effect of 300% flow-through exchange by sampling the flow-through BWE and control tanks at a third time point, following this last treatment.

Rhodamine dye was used to measure removal of the original (coastal) water mass during BWE. For each time point, we collected replicate samples from at least two access points (deck hatches). For each access point, two replicates were collected from each of two depths (surface and 10m). Samples were collected with a Niskin bottle (1.5l) and stored in amber glass jars for analysis.

We also measured the effect of exchange on concentrations of coastal zooplankton. For each time point, we collected replicate samples from the same access points as above. For each access point, we sampled zooplankton with net tows (n=2 or 3), using a plankton net (30cm diameter, 80µ mesh) that was towed vertically from the bottom to top of the tank (15-20m; Table 1). Each replicate zooplankton sample was preserved in 5-10% buffered formalin for subsequent analysis.

Sample Analysis

We measured concentration of rhodamine dye for each sample using a flourometer (Turner Designs, Model 10AU), fitted with appropriate excitation and emission filters, employing standard methods. Concentrations were estimated to the nearest 0.2 µ/l.

We estimated concentrations of coastal zooplankton taxa by direct counts under a dissecting microscope. More specifically, we focused our analysis on "target" species or taxonomic groups, which met several criteria. First, we selected organisms that were known to be coastal in origin and that are considered unlikely to occur offshore (i.e., > 100 nautical miles). This was done to avoid any compensatory changes that could result from the influx of target organisms, with oceanic water, during BWE. Second, we further selected among this group only taxa that were relatively abundant (mean concentration > 25/m3) at the start of the voyage; this approach avoids the difficulty, or lack of sensitivity, in measuring changes over time for low concentrations.

Statistical Analysis

Removal of Coastal Water.
We estimated efficacy of BWE in removing the initial coastal water, comparing rhodamine concentrations between treatments within each voyage. For each voyage, efficacy was estimated as the % difference in rhodamine concentration between BWE and control tanks: [(Control Tank) – (Exchange Tank) / (Control Tank)] X 100.

All samples for each time point and tank were used to estimate the average concentration. We tested for differences by depth for time point and tank, using t-tests; in only two cases were there any differences by depth, and both were present following 100% flow-through BWE.

Removal of Zooplankton.
We estimated percent change in concentration of target zooplankton taxa over time for each tank. We tested for differences in percent change between treatments using Friedman ANOVA following 100% BWE (i.e., comparing control, empty-refill, and flow-through treatments). A separate analysis, using the same statistic, tested for differences among 300% flow-through BWE, 100% empty-refill BWE, and the control treatments; this was done to test whether any differences existed between the two different BWE treatments, which are considered to be roughly equivalent in terms of current management practices. Each test was followed by a multiple comparison among treatments.

We also estimated the efficacy of BWE in reducing concentrations of target zooplankton taxa, comparing changes in BWE tanks to those in control tanks over the same time periods (see Figure 1). For each taxon and voyage, we estimated efficacy separately for 100% empty-refill, 100% flow-through, and 300% flow-

through BWE. For each time period, % efficacy was estimated as:

   % Efficacy = (X-C)/(C+1), where
   (i) X is the percent change in the BWE tank expressed as $(x_0-x_1)/x_0$ X 100, and
   (ii) C is the percent change in the Control tank expressed as $(c_0-c_1)/c_0$ X 100.
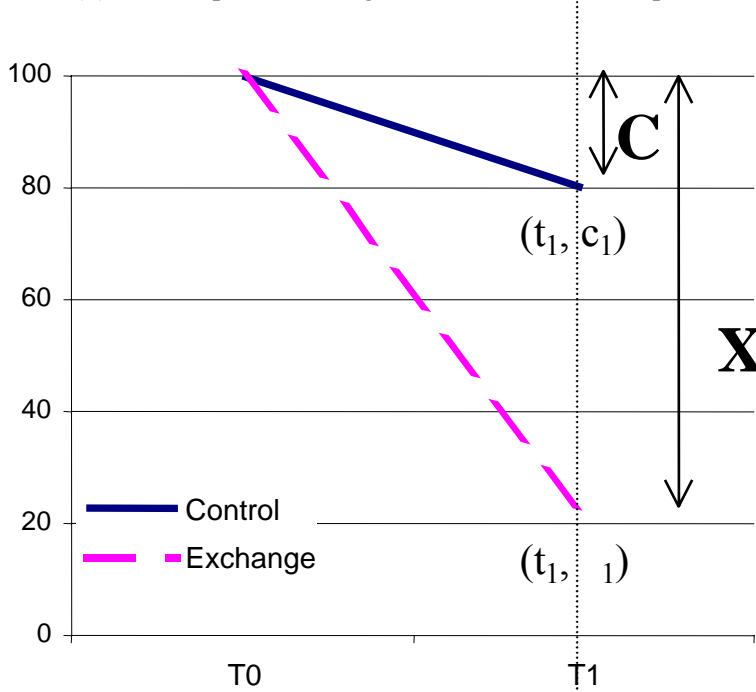

Figure 1. Calculation of BWE for Target Zooplankton. Efficacy is estimated as a function of change in concentration over time (T0 to T1) in exchanged tank (X) relative to those observed in the control tank (C). See text for further detail.

The efficacy of exchange for target zooplankton was compared to that for removal of initial coastal water for each time point by BWE method. More specifically, we tested the hypothesis that the rate of removal for zooplankton differed significantly from that of the water mass. We tested whether efficacy measures for zooplankton differed significantly from an expected value (mean value for efficacy of rhodamine), using a 1-tailed t-test. In addition, we also compared the efficacy for 100% empty-refill BWE to that of 300% flow-through BWE, using a 1-tailed t-test.

RESULTS

Removal of Coastal Water

The efficacy of BWE in removing original coastal water, using rhodamine dye tracer data, averaged 99% for each the empty-refill method (100% volume) and flow-through method (300% volume). Not surprisingly, the efficacy of 100% flow-through BWE was lower, exhibiting a wide range from 30% to 94% and an average of 66% among the 6 experimental tests (Figure 2).
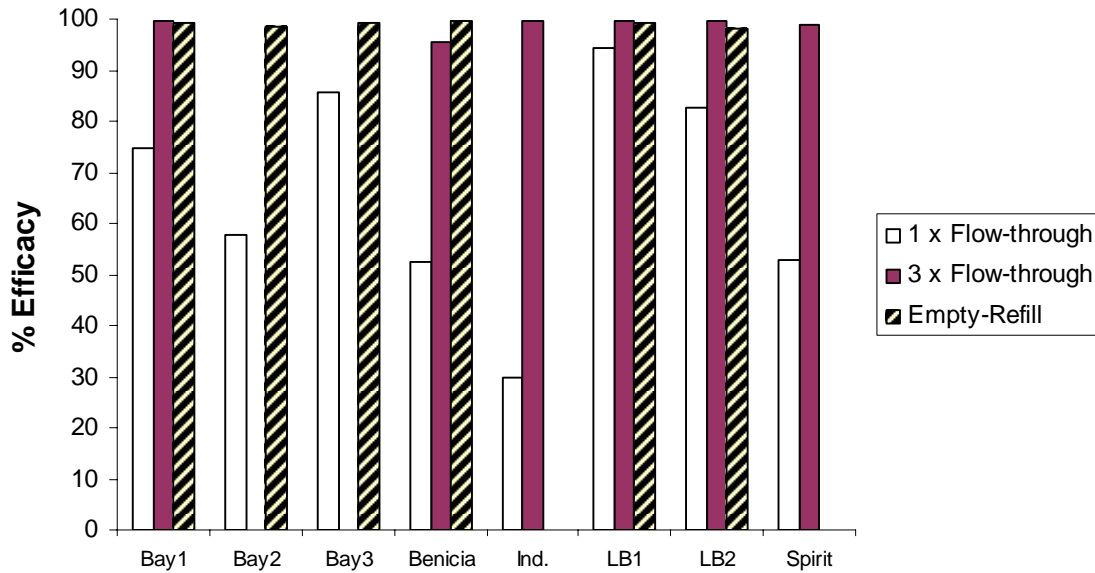
Figure 2. Efficacy of BWE for Removal of Coastal Water. Shown is the % efficacy by ship for each of 2 or 3 BWE treatments: 100% empty-refill, 100% flow-through, and 300% flow-through.

Removal of Zooplankton

Percent Change in Concentration.

There was a significant difference among treatments in the reduction of target zooplankton, when comparing percent change in concentration between initial samples and time point 1 or following the first 100% BWE event (Figure 3; Freidman's Test, $p<0.001$). The majority of taxa declined in the control tank, but the pattern was highly variable among ships and taxonomic groups, exhibiting no obvious association to either variable; such variation is also evident in the 100% flow-through treatment. Even accounting for the decline in the control tanks for some taxa, the magnitude of decline for both the 100% empty-refill and the 100% flow-through BWE tanks was significantly greater from that observed in the control (Friedman's multiple comparison, $p<0.05$). In addition, the two BWE treatments also differed from each other in the magnitude of decline (Friedman's multiple comparison, $p<0.05$).

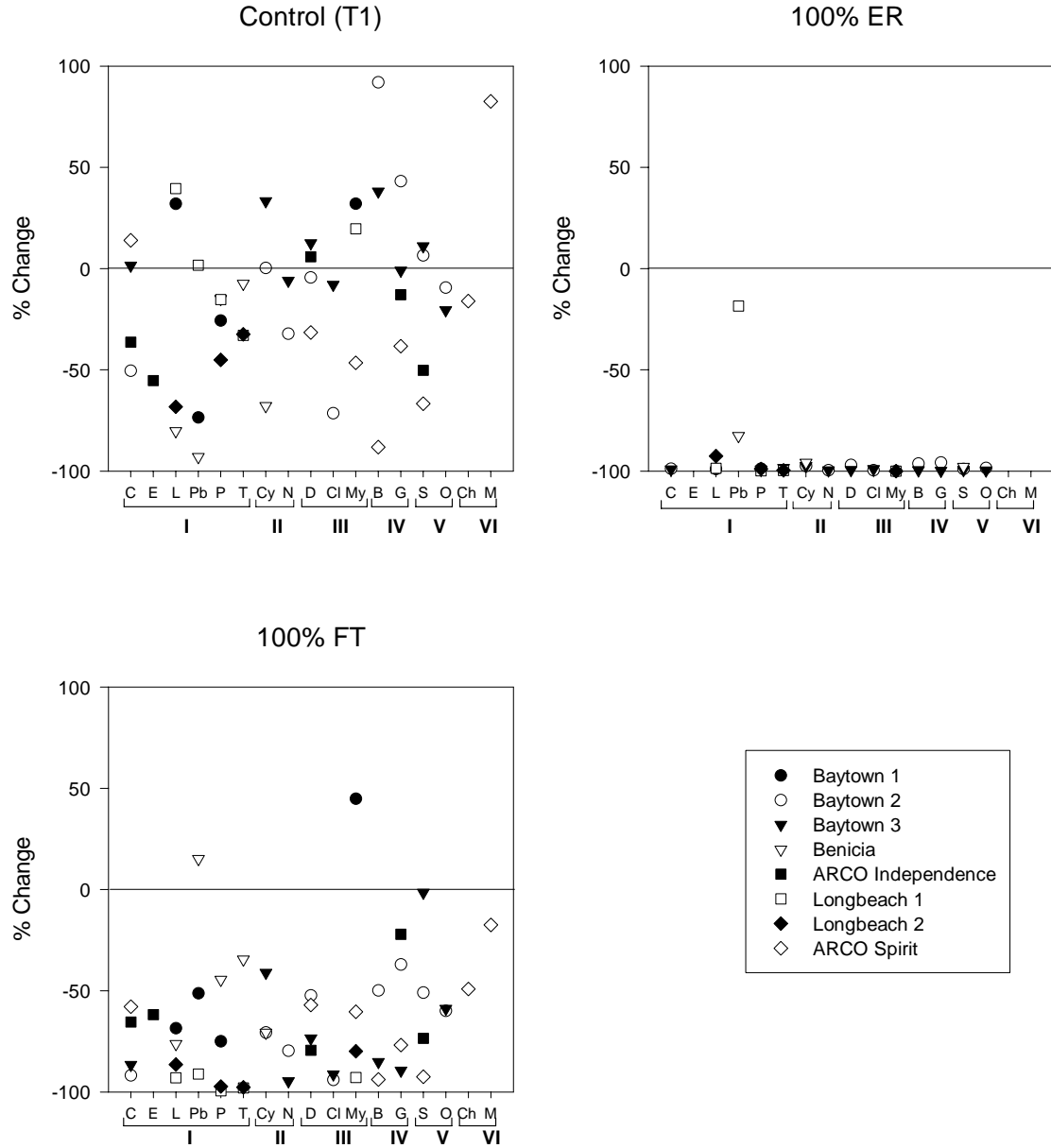## Control (T1)

100% ER

100% FT

Key:
- ● Baytown 1
- ○ Baytown 2
- ▼ Baytown 3
- ▽ Benicia
- ■ ARCO Independence
- □ Longbeach 1
- ◆ Longbeach 2
- ◇ ARCO Spirit

Figure 3. Percent Change in Concentrations of Target Zooplankton Following 100% BWE. Shown are the percent change for target zooplankton by taxonomic group (x axis) and ship (symbols as indicated in key). Taxonomic information along the x-axis are as follows: Group 1 -- copepods (C=*Corycaeus*, E=*Euterpina*, L=*Limnothoina*, Pb=*Pseudobradya*, P=Pseudodiaptomus, T=*Tortunas*); Group II – barnacles (Cy=cyprids, N=nauplii); Group III – Other crustaceans (D=decapod larvae, Cl=cladocerans, My=mysids); Group IV – molluscs (B=bivalve larvae; G=gastropod larvae); Group V – polychaetes (S=Spionidae larvae, O=other polychaete larvae); Group VI – other taxonomic groups (C=chaeteognaths, M=medusae).

At time point 3, there was also a significant difference among the three treatments (control and flow-through tanks after 300% BWE, and empty-refill tank @ 100% BWE) in the reduction of target zooplankton. A comparison of the 300% flow-through BWE and control treatments are shown in Figure 4. Overall there was a significant difference among the three treatment (Friedman's Test, p<0.001), and a significant pairwise difference when comparing each of the BWE treatments individually to controls (Friedman's multiple comparison, p<0.05).

There was no longer a statistical difference between the empty-refill treatment (100%) and flow-through treatment (300%). However, an important characteristic of the overall pattern is the high variance in percent change among taxa and voyages for the 300% flow-through treatment (Figure 4). This high variation contrasts markedly with the more uniform reduction in most taxa for 100% empty-refill BWE (Figure 3). Thus, although the treatments did not differ statistically (on average), from a risk management perspective, the 300% flow-through treatment resulted in many instances (taxa and voyages) when percent change was relatively low (20-80% reduction).
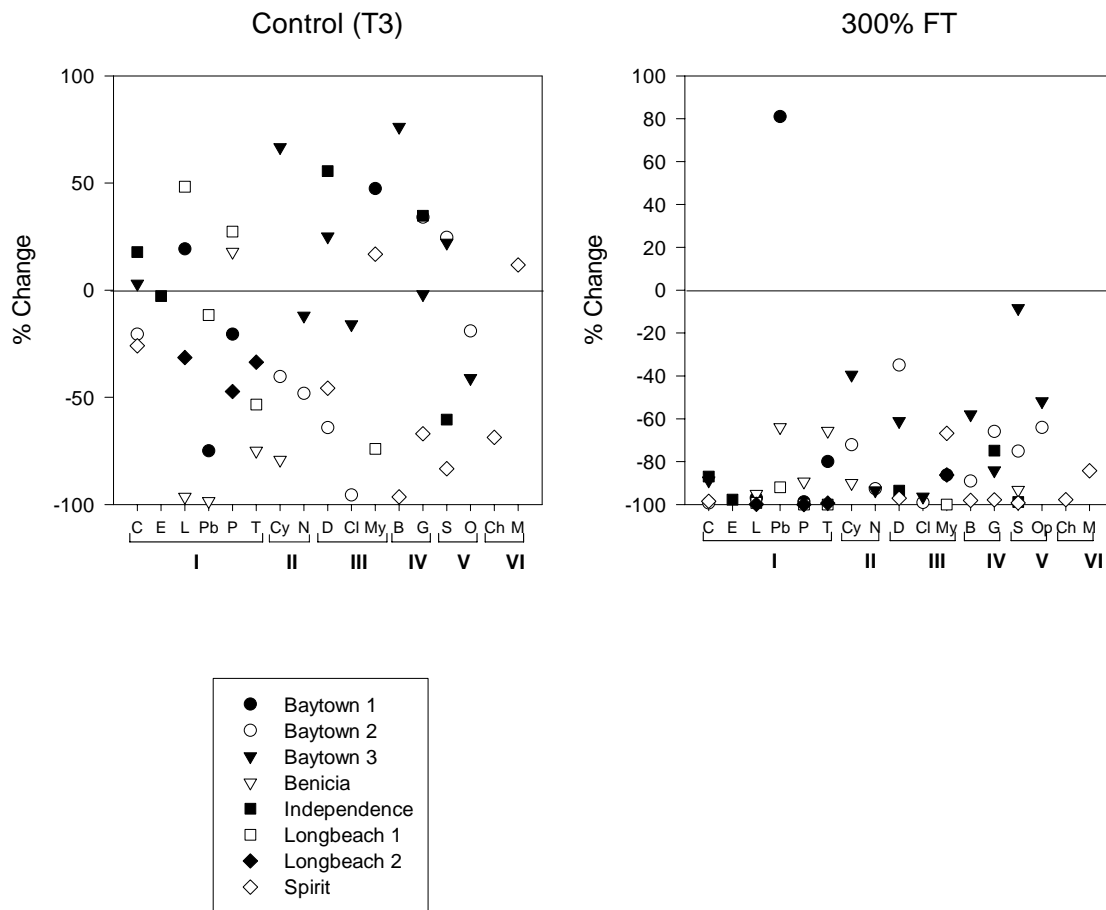

Figure 4. Percent Change in Concentrations of Target Zooplankton Following 300% BWE. Shown are the percent change for target zooplankton by taxonomic group (x axis) and ship (symbols as indicated in key). Taxonomic information along the x-axis are as follows: Group 1 -- copepods (C=*Corycaeus*, E=*Euterpina*, L=*Limnothoina*, Pb=*Pseudobradya*, P=Pseudodiaptomus, T=*Tortunas*); Group II – barnacles (Cy=cyprids, N=nauplii); Group III – Other crustaceans (D=decapod larvae, Cl=cladocerans, My=mysids); Group IV – molluscs (B=bivalve larvae; G=gastropod larvae); Group V – polychaetes (S=Spionidae larvae, O=other polychaete larvae); Group VI – other taxonomic groups (C=chaeteognaths, M=medusae).

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Efficacy of BWE.
1. 100% Empty-Refill BWE. The efficacy of 100% empty-refill BWE in reducing concentrations of target zooplankton did not differ significantly from that observed for removal of the original water (Fig 5; t-test, p>0.05).
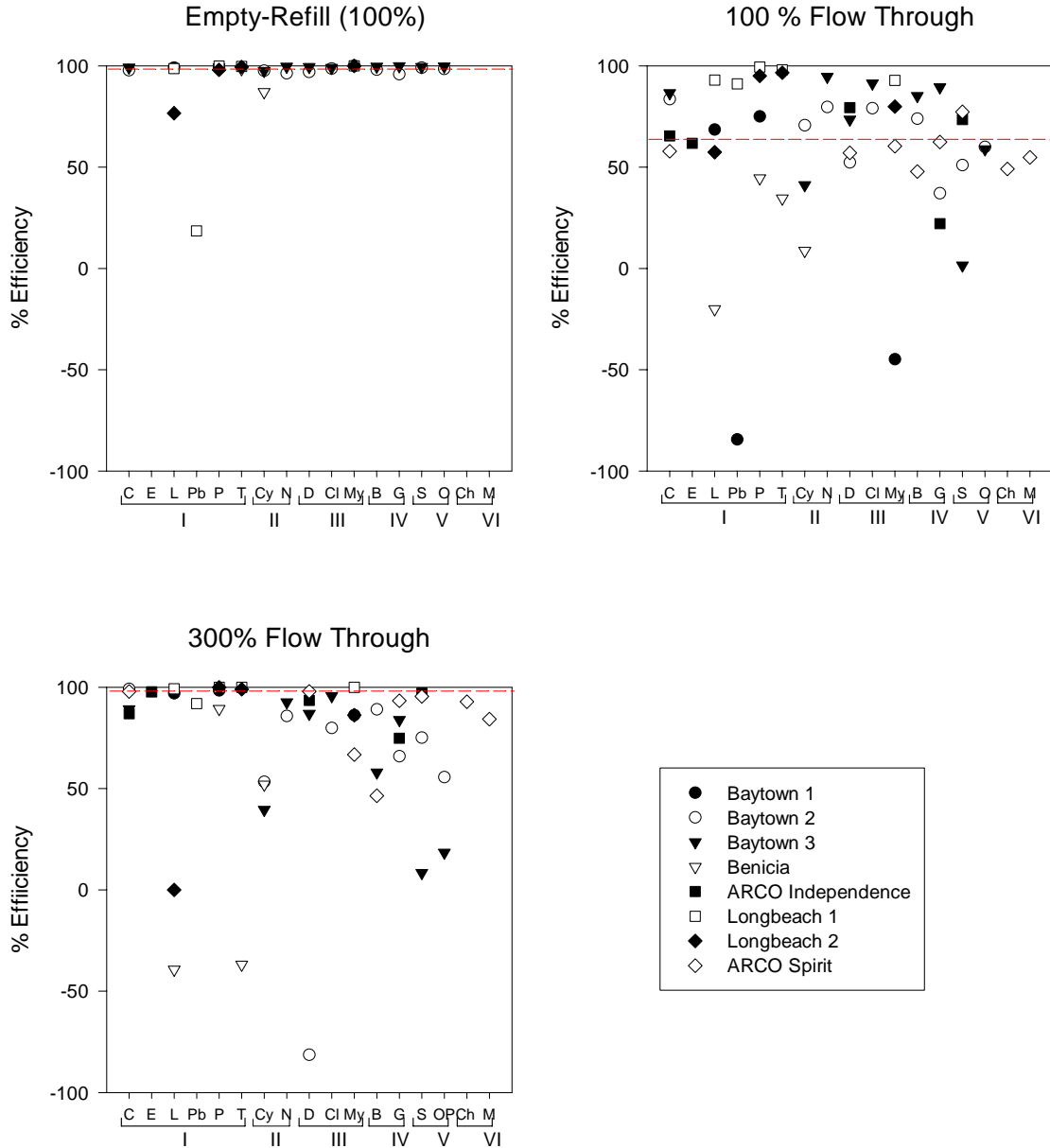

Figure 5. Efficacy of BWE for Removal of Target Zooplankton Taxa. Shown are the % efficacy for target zooplankton by taxonomic group (x axis) and ship (symbols as indicated in key). Dashed line indicated mean % efficacy for rhodamine dye removal. Taxonomic c information along the x-axis as follows: Group 1 -- copepods (C=*Corycaeus*, E=*Euterpina*, L=*Limnothoina*, Pb=*Pseudobradya*, P=Pseudodiaptomus, T=*Tortunas*); Group II – barnacles (Cy=cyprids, N=nauplii); Group III – Other crustaceans (D=decapod larvae, Cl=cladocerans, My=mysids); Group IV – molluscs (B=bivalve larvae; G=gastropod larvae); Group V – polychaetes (S=Spionidae larvae, O=other polychaete larvae); Group VI – other taxonomic groups (C=chaeteognaths, M=medusae).

In other words, the effect of this BWE treatment on average did not differ for biota and inert tracers. With two exceptions, the efficacy of 100% empty-refill BWE for target zooplankton ranged from 96-100% compared to 99% for the rhodamine dye tracer. The exceptions were the copepods *Limnothoina* (77% in one instance, but 99% for two other voyages) and *Pseudobradya*. The latter was consistently low across 3 different voyages, and we observed a sizable increase in concentrations in two voyages, possibly reflecting a benthic organism that was under-sampled initially and resuspended following BWE. Excluding *Pseudobradya*, the mean value across all target taxa and voyages was 97.9% efficacy (Appendix 1).

2. 100 % Flow-Through BWE. The efficacy of 100% flow-through BWE in reducing concentrations of target zooplankton also did not differ significantly from that observed for removal of rhodamine dye (Fig. 5; p>0.05). There was a relatively high variance compared to the 100% empty-refill treatment, including a few cases of observed increases in concentration: (a) the copepod *Pseudobradya* on two voyages, (b) mysids on one voyage, and (c) decapod larvae on one voyage. These cases may result from either benthic habitat utilization, as mentioned above, or perhaps mobility. The efficacy for all other taxa ranged from 2%-99% (Appendix 1), with spionid polychaetes having the low value. Excluding *Pseudobradya*, the mean value across taxa of 59.8% efficacy.

3. 300% Flow-Through BWE. The efficacy of 300% flow-through BWE was significantly less effective in reducing target zooplankton than in removing rhodamine dye (Figure 5; one-tailed t-test, p<0.05). The variance in performance across taxa and voyages, as observed after 100% flow-through exchange, remained relatively high. In three cases, we observed an increase in concentrations, including the same taxa as previously discussed, including *Pseudobradya* and decapod larvae. The efficacy for other taxa ranged from 8%-100%, with spionid polychaetes remaining the lowest value for the same vessel as observed at 100% flow-through BWE (Appendix 1). Excluding *Pseudobradya*, the mean value across taxa was 70.1% efficacy.

DISCUSSION

To date, few data exist that begin to evaluate the efficacy of BWE, and there is considerable uncertainty about the effect of this treatment to reduce concentrations of coastal organisms in ballast tanks. This is reflected in the literature, for which a few scattered studies have reported a wide range of efficacy values. This has led to confusion about the relative value of BWE, both for reducing organism concentrations and reducing the subsequent risk of invasions.

Our study demonstrated that BWE on oil tankers arriving to PWS was highly effective. BWE removed 99% of the original water mass, following 100% empty-refill exchange or 300% flow-through exchange. Moreover, the performance of 100% empty-refill BWE had a similar efficacy in removing the target zooplankton taxa examined, with the exception of one taxon (*Pseudobradya*). In contrast, flow-through BWE (both 100% and 300%) had significantly lower efficacy that the empty-refill method, exhibiting a wide range of variation among taxa.

The low and variable efficacy of flow-through BWE is not particularly surprising. This may result from differences among vessels in tank configuration and hydrodynamics during BWE. Variable levels of flow and incomplete mixing (or 'dead zones'), both within and among tanks, is expected to result in variation in the removal of organisms by BWE. Variable flow rates may also serve to accentuate differences among taxa due to variable swimming speeds, habitat selection (i.e., shallow versus deep water), and responses to water motion.

In addition, the variable response of some zooplankton taxa may also be expected. For example, several organisms exhibited either an increase or only slight decrease in abundance, representing noticeable departures from the majority of organisms. These were all organisms that either are associated with benthic communities (e.g., *Pseudobradya*, mysids), exhibit relatively fast swimming speeds (e.g., decapod larvae, mysids), or may have been late-stage larvae that become benthic forms (e.g., spionid polychaetes).

Overall, the efficacy of BWE on these oil tankers is much greater than might be expected, based upon some previous studies. For example, the efficacy of BWE for container ships has been reported to be on the order of 40% for phytoplankton (Dickman and Zhang 1999, Zhang and Dickman 1999). However, the methods used in several previous studies did not adequately control for the effects of initial concentrations or time. In the container ship study, concentrations were compared for ships that underwent BWE versus different ships that did not. This approach does not (a) control for significant seasonal and spatial variation in initial plankton densities, or (b) control for the percentage of water exchanged. Thus, in addition to BWE, there are several uncontrolled sources of variation (or variables) that limit interpretation of the results.

In contrast, our current study uses a controlled experimental design, which includes measures before and after treatment. This approach compares changes of organism concentrations in ballast water of tanks that have undergone exchange to control tanks that have not undergone exchange, using identical measures for the experimental (exchanged) and control (unexchanged) tanks on the same ship, at the same times, and with the same source biota. This approach provides the highest quality data, controlling for effects of time and initial densities.

We predict the results from our analysis are generally representative of BWE efficacy for most ships. A great deal variation, and the sometimes low values, in the efficacy of BWE reported in previous studies may simply be artifact of differences in methodological approaches, including those with uncontrolled sources of variation (as discussed above). This view is supported by an additional study of container ships, measuring efficacy of BWE using controlled experiments similar to those in this study, in which efficacy is comparable to that reported herein (Ruiz et al., unpublished data).

Our analyses indicate that BWE has a clear and significant effect on reducing the concentrations of non-native organisms in the ballast tanks of oil tankers. On average, concentrations are reduced by 70 to 98%, for the flow-through (300%) and empty-refill (100%) method respectively. As expected there is considerable variation observed, both among taxonomic groups and across ships, in the efficacy of flow-through BWE. Nonetheless, our results indicate BWE is a valuable management tool that is presently available. Since invasions are density-dependent, we expect that BWE does indeed reduce the risk of future invasions.

Despite the demonstrable value of BWE, residual organisms are still present in the plankton (water column). In addition, our study has not examined effects of BWE on benthic (bottom-dwelling) organisms, which may be less effected by BWE. Both planktonic and benthic organisms pose some risk of invasions upon release. However, the risk associated with such residual organisms is poorly resolved, because the specific quantitative relationship between organism concentration (dose) and establishment (invasion) is not known (see discussion by Ruiz and Carlton 2003), representing a significant obstacle in identification of an acceptable organism discharge 'standard' for ballast water treatment. Understanding such this 'dose-response' relationship remains a major challenge and high priority for invasion ecology.

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