

# **Efficacy of hull cleaning operations in containing biological material**

## **II. Seasonal variability**

### **MAF Biosecurity New Zealand Project ZBS2005-22**

Chris Woods, Oliver Floerl, Isla Fitridge, Olivia Johnston, Karen Robinson, David Rupp, Niki Davey, Nicola Rush and Matt Smith

MAF Biosecurity New Zealand Technical Paper Series 08/11

Prepared for BNZ Pre-clearance Directorate by

The National Institute of Water and Atmospheric Research Ltd

ISBN 978-0-478-32191-3 (Online)

ISSN 1177-6412 (Online)

October 2007

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Requests for further copies should be directed to:

Border Standards  
MAF Biosecurity New Zealand  
Pastoral House  
25 The Terrace  
PO Box 2526  
Wellington 6140

Tel: 04 894 0100

Fax: 04 894 0733

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# Contents

<b>List of figures</b>	<b>5</b>
<b>List of tables</b>	<b>8</b>
<b>Executive summary</b>	<b>10</b>
Specific objective 1	10
Specific objective 2	11
Recommended guidelines for hull cleaning facilities	12
<b>Objectives</b>	<b>14</b>
Overall objective	14
Specific objectives	14
<b>1 Introduction</b>	<b>15</b>
<b>2 Methods</b>	<b>17</b>
2.1 Specific objective 1	17
2.1.1 Facilities studied	21
2.1.2 Definition of a viable organism	24
2.1.3 Sampling of hull-fouling assemblages before and after removal from hulls	25
2.1.4 Sampling of liquid effluent prior to discharge	28
2.1.5 Sampling of vessels cleaned in the water	30
2.1.6 Assessment of seasonality	30
2.1.7 Statistical analyses	30
2.2 Specific objective 2	31
2.2.1 Hydraulic residence time and sinking rate	31
2.2.2 Modelling water residence time and particle settling in treatment systems	36
<b>3 Results</b>	<b>47</b>
3.1 Specific objective 1	47
3.1.1 Solid fouling material	52
3.1.2 Sampling of liquid effluent	64
3.2 Specific objective 2	78
3.2.1 Modelling water residence time and particle settling in treatment systems	78
<b>4 Conclusions</b>	<b>97</b>

4.1 Specific objective 1	97
4.2 Specific objective 2	100
4.3 Consideration of recommendations for commercial hull cleaning facilities	101
4.4 Recommended guidelines	105
<b>Acknowledgements</b>	<b>107</b>
<b>References</b>	<b>108</b>
<b>Appendix</b>	<b>114</b>
Appendix 1: Guidelines used by NIWA field staff to assess viability of fouling biota removed from vessel hulls.	114

## List of figures

- Figure 1: Initial removal of fouling epibiota by water blasting at Tauranga Marina (A) and Lyttelton dry dock (B). 18
- Figure 2: Lyttelton dry dock: (A) Independent I about to be cleaned), (B) raw liquid effluent collecting on dock floor, (C) liquid effluent treatment system, and, (D) final settlement tanks in treatment system. 21
- Figure 3: Tauranga Marina Society travel lift: (A) Samovar being cleaned, (B) 1 mm primary screen before settlement tanks, (C) liquid effluent treatment system, and, (D) sand filter. In (A), the red arrow indicates the sump grill below Samovar. 23
- Figure 4: Orams Marine Maintenance travel lift: (A) Phase II about to be cleaned, (B) raw effluent collection area and sump grills, (C) liquid effluent settlement tanks, and, (D) sand filter. In (B), the red arrows indicate sump grills and the green arrows sealed area bunding. 24
- Figure 5: Visual check for signs of organism viability after immersion in seawater for 20-30 min (for example, barnacles, note feeding limbs indicated by red arrow). 26
- Figure 6: Various sampling stages for liquid effluent following hull cleaning. 28
- Figure 7: Water volume  $V(\tau)$  within a residence time interval  $\Delta \tau$  against residence time  $\tau$  for a given reservoir volume  $V$  and flow rate  $Q$ . 38
- Figure 8: Measured sink rates for various species and marine snow against characteristic length (from various sources, Table 3). Data show adults, larvae and eggs. Also shown is Stokes' law for an organism density of 1.2. 42
- Figure 9: Estimated biomass (a) and percent fouling cover (b) on vessels, cleaning duration (c) and exposure time (d) in winter (black bars) and summer (grey bars) at different hull cleaning facilities. DD = Lyttelton dry dock, OM = Orams Marine, TR = Tauranga Marina, and WP = Westpark Marina. Data are mean  $\pm$  1 s.e. 47
- Figure 10: Examples of still-wet fouling organisms at haul-out facilities (A and B) and desiccating organisms at Lyttelton dry dock (C and D). 54
- Figure 11: Percentage of hard-bodied and soft-bodied organisms undamaged (a) and alive (b) according to type of hull cleaning facility in winter (black bars) and summer (grey bars). Data are mean  $\pm$  1 s.e. 56
- Figure 12: Percentage of viable fouling organisms by taxonomic group according to type of hull cleaning facility in winter (black bars) and summer (grey bars). Data are mean  $\pm$  1 s.e. 59
- Figure 13: Percentage of viable fouling organisms by taxonomic group according to percentage of fouling cover on submerged hull surface. Unfilled symbols (winter), filled symbols (summer). Data are mean without s.e (to keep data points distinct) for individual boats in each facility and type of cleaning operation. 62
- Figure 14: Concentrations (no./10 L) of animals, propagules, unicellular organisms and filamentous algae collected in liquid effluent samples during winter (2003) and summer (2006) at various hull cleaning facilities. Data are counts of organisms and are not an indicator of viability. Data are mean  $\pm$  1 s.e. 73
- Figure 15: Representative images of material retained in a 60  $\mu$ m sieve from the final effluent at Lyttelton dry dock. Detrital aggregations (A) and propagule/zoospore (B). 75
- Figure 16: Representative images of material retained in a 60  $\mu$ m sieve from the final effluent at Tauranga Marina. Detrital aggregations and pollen grains (A-B) and stalked diatoms (C). 75

Figure 17: Modelled hydraulic residence time distribution for three flow rates at the Lyttelton dry dock liquid effluent treatment system (summer and winter). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 80

Figure 18: Modelled hydraulic residence time distribution for three flow rates at the Orams Marine liquid effluent treatment system (summer operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 81

Figure 19: Modelled hydraulic residence time distribution for three flow rates at the Orams Marine liquid effluent treatment system (winter operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 82

Figure 20: Modelled hydraulic residence time distribution for three flow rates at the Westpark Marina liquid effluent treatment system (summer operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 83

Figure 21: Modelled hydraulic residence time distribution for three flow rates at the Westpark Marina liquid effluent treatment system (winter operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 84

Figure 22: Modelled hydraulic residence time distribution for three flow rates at the Tauranga Marina liquid effluent treatment system (summer operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 85

Figure 23: Modelled hydraulic residence time distribution for three flow rates at the Tauranga Marina liquid effluent treatment system (winter operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility. 86

Figure 24: Relative modelled particle concentration in outflow with respect to inflow concentration for the Lyttelton dry dock liquid effluent treatment system (summer and winter). The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 87

Figure 25: Relative modelled particle concentration in outflow with respect to inflow concentration for the Orams Marine liquid effluent treatment system in summer. The lower

figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 88

Figure 26: Relative modelled particle concentration in outflow with respect to inflow concentration for the Orams Marine liquid effluent treatment system in winter. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 89

Figure 27: Relative modelled particle concentration in outflow with respect to inflow concentration for the Westpark Marina liquid effluent treatment system in summer. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 90

Figure 28: Relative modelled particle concentration in outflow with respect to inflow concentration for the Westpark Marina liquid effluent treatment system in winter. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 91

Figure 29: Relative modelled particle concentration in outflow with respect to inflow concentration for the Tauranga Marina liquid effluent treatment system in summer. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 92

Figure 30: Relative modelled particle concentration in outflow with respect to inflow concentration for the Tauranga Marina liquid effluent treatment system in winter. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility. 93

## List of tables

Table 1a: Hull cleaning facilities visited for seasonal assessment of hull cleaning efficacy during Research Project ZBS2002-04 (winter).	19
Table 1b: Hull cleaning facilities visited for seasonal assessment of hull cleaning efficacy sampled during Research Project ZBS2005-22 (summer).	20
Table 2: Biological data to be collected during visits to hull cleaning facilities.	27
Table 3: Sample collection of liquid effluent during winter (ZBS2002-04) and summer (ZBS2005-22) hull cleaning surveys. Samples taken or not taken are indicated by ‘+’ and ‘-’, respectively.	29
Table 4: Known sinking rates for various marine organisms and organic aggregates (marine snow).	33
Table 5: Model parameters for Lyttelton dry dock	40
Table 6: Model parameters for Orams Marine	40
Table 7: Model parameters for Westpark Marina	41
Table 8: Model parameters for Tauranga Marina	41
Table 9: Numbers of vessels cleaned per hull cleaning facility and related liquid effluent treatment system statistics.	43
Table 10: Salinity tolerances of various marine organisms that may be regarded as non-indigenous pest species (NIS) or encountered as hull-fouling. Minimum salinity tolerances are expressed as the minimum salinity (ppt) and the number of days for which the organism is able to survive at that salinity before death or severe damage occurs. Lower normal habitat salinity levels in which animals are found are in brackets ().	45
Table 11a: Vessels cleaned at different hull cleaning facilities during Research Project ZBS2002-04 (winter season).	48
Table 11b: Vessels cleaned at different hull cleaning facilities during Research Project ZBS2005-22 (summer season).	50
Table 12: ANOVA on percentage cover of fouling organisms on vessel hulls. Data were $\log(x+1)$ transformed.	52
Table 13: ANOVA on duration of hull cleaning at each facility. Data were $\log(x+1)$ transformed.	52
Table 14: Number of organisms or fragments of solid fouling material examined during the two hull cleaning projects (winter: 10 317; summer: 9070) in each facility.	53
Table 15: ANOVA on duration of exposure to air before cleaning (that is, time from removal from water to beginning of water blasting). Data were $\log(x+1)$ transformed.	55
Table 16: ANOVA on viability of fouling organisms sampled: Total Organisms (all groups pooled).	57
Table 17: ANOVA on viability of fouling organisms sampled: Total organisms excluding tubicolous polychaetes.	57
Table 18: Concentrations of organisms in the liquid waste sampled in dry dock and haul-out operations at various stages of treatment (ZBS2002-04, winter sampling). All concentrations are given as abundance/10 L (mean $\pm$ s.e.) except filamentous algae, for which a ranks scale of abundance (0–5) was used. Decimal places are removed to simplify table.	65



Table 19: Concentrations of organisms in the liquid waste sampled in dry dock and haul-out operations at various stages of treatment (ZBS2005-22, summer sampling). All concentrations are given as abundance/10 L (mean  $\pm$  s.e.) except filamentous algae, for which a ranks scale of abundance (0-5) was used. Decimal places are removed to simplify table. 67

Table 20: Reduction in abundance of animals, propagules, unicellular organisms and filamentous algae at the various stages of liquid effluent treatment. All percentage values represent reduction in abundance relative to the concentrations observed in the water blast runoff liquid. 70

Table 21: ANOVA on numbers of intact animals, propagules and unicellular organisms in the liquid effluent from the water blaster, that is, before entering settlement tanks. Data were analysed untransformed since  $\log(x+1)$  transformation did not remove heterogeneity of variances. 71

Table 22: Movement and mitochondria in liquid samples taken at the various stages of treatment. Presence and absence of mitochondrial stains and visible movement is indicated by '+' and '-', respectively. 76

Table 23: Modelled average number of hours liquid effluent resides in treatment systems at various facilities per cleaning season (summer or winter) and whether or not each facility's treatment system can theoretically allow particles or representative sinking rates to settle-out before filtering/screening and then discharge at typical flow rates. Figures in brackets are calculated average number of hours liquid effluent resides in treatment systems based on double and half input flow rates. 79

Table 24: Salinity tolerances of various marine organisms that may be regarded as non-indigenous pest species (NIS) or commonly encountered as hull-fouling. Minimum salinity tolerances are expressed as the minimum salinity (ppt) and the number of days for which the organism is able to survive at that salinity before death or severe damage occurs, or as lower normal habitat salinity level (ppt) in which animals are found (in brackets). For each organism, modelled hydraulic residence time was used to determine each facility's potential effectiveness at killing that organism: Lyttelton dry dock (LYT), Orams Marine (OMM), Westpark Marina (WPM), and Tauranga Marina (TRG). Facilities indicated with a question mark (?) are facilities that may be effective but the minimum salinity tolerance of the NIS in question does not allow certainty of mortality. 95

## Executive summary

Hull fouling is an important pathway or vector for the introduction and spread of marine non-indigenous species (NIS) in New Zealand. This project (ZBS2005-22, summer) sought to determine the seasonal risk to marine biosecurity of vessel hull cleaning (defouling) and assess the efficacy of hull cleaning methods and effluent treatments in reducing this risk, by comparison with an initial examination of the efficacy of various hull cleaning techniques and facilities carried out in the winter of 2003 (ZBS2002-04) (Floerl et al. 2003). The five facilities assessed in ZBS2002-04 were revisited during the 2005/06 summer season. These facilities were: Lyttelton Port, Orams Marine Maintenance, Westpark Marina, Tauranga Marina and Gulf Harbour Marina.

The project investigated the types, amounts and viability of fouling organisms discharged from these five different hull cleaning facilities, and evaluated the efficacy of waste treatment methods being used in reducing the amount of viable material reaching the coastal marine area.

Specific objectives to the project were:

1. Building on the outcomes of ZBS2002-04, investigate the types, amounts and viability of fouling organisms discharged from a variety of hull cleaning facilities, and evaluate the efficacy of waste treatment methods being used in reducing the amount of viable material reaching the coastal marine area for both winter and summer conditions and use.
2. Identify the critical requirements for hull cleaning and waste treatment that would most effectively minimise the release of viable organisms from hull cleaning situations.

### Specific objective 1

Comparison of the results obtained in this investigation (ZBS2005-22 summer) with those of ZBS2002-04 (winter) indicate a complex interaction of factors which complicate any attempt to discern seasonal differences in the efficacy of various hull cleaning methods. Hull cleaning method, rather than seasonality, was the main determinant in hull-fouling organism viability during the cleaning and post-treatment processes. Generally, dry dock and haul-out facilities and their associated hull cleaning methods (water blasting) result in fewer viable macro-fouling organisms in the solid debris than in solid debris removed from in-water cleaning (manual scraping).

The type and severity of physical damage to organisms removed from the hulls varied among vessels and operations, but not among season. In haul-out and dry dock operations, the pressure associated with the water blasting and trampling by cleaning operators had fragmented and crushed a large proportion of soft-bodied organisms. In-water removal of organisms from vessel hulls with a paint scraper or a soft cloth caused similar damage to hard-bodied taxa, but considerably less to soft-bodied taxa than water blasting in out-of-water facilities. Winter and summer sampling showed survival of soft-bodied organisms tended to be lower in haul-out and dry dock operations than following in-water cleaning. Rates of survival of hard-bodied organisms were generally low in all hull cleaning methods during both sampling seasons.

As system upgrades have been completed at all but Westpark Marina since ZBS2002-04 (winter), some seasonal comparisons of treatment stages were not valid. In both sampling seasons (winter and summer), average concentrations of intact animals, propagules and unicellular organisms were greatest in the initial runoff from the water blasting. Then, across all facilities, settlement and filtration progressively reduced the mean concentrations of organisms in the liquid effluent. In samples taken from the first chamber of the multi-chamber settlement tanks, concentrations of intact animals, propagules and unicellular organisms were reduced by between 20.5 and 99.5 percent, and the rank abundance of filamentous algae decreased by 10 to

60 percent. Concentrations of animals, propagules and unicellular organisms in samples taken from the final settlement chamber had been reduced by 39.7 to 100 percent and that of filamentous algae by 9.6 to 100 percent. In Lyttelton dry dock, Orams Marine and the Tauranga Marina, samples were taken of the final effluent during winter or summer. In all three locations, concentrations of animals, propagules and unicellular organisms had been reduced by  $\geq 98.5$  percent compared to concentrations observed in the runoff from water blasting, while filamentous algae had been reduced by 80 to 100 percent.

Rapid assessment of viability using the vital stain Janus Green and organism movement indicated that the majority of organisms discharged from hull cleaning effluent treatment systems are unlikely to be viable. However, true determination of whether organisms are actually alive following processing through the liquid effluent treatment can only be achieved through subsequent culture experimentation (for example, hatching of dinoflagellate cysts) which was not part of this project.

## Specific objective 2

As macro-fouling removed from vessel hulls in dry docks and haul-out facilities typically is collected and disposed of in land-fill it does not represent a non-indigenous species (NIS) invasion-risk. However, raw liquid effluent from water blasting carries a wide variety of marine organisms, which may subsequently pass back into the marine environment post-treatment, and therefore poses a quarantine risk. This risk varies according to the efficacy of any particular liquid effluent treatment system in question. This in turn depends upon a combination of factors such as effluent hydraulic residence time, sinking rates of various suspended organisms and their related tolerance to freshwater.

For each facility, the residence time of liquid effluent within each facility's treatment system was modelled as a function of the volume of liquid effluent entering each facility's treatment system in relation to the total volume of each treatment system. During summer, at typical flow rates the average residence times at Lyttelton dry dock, Orams Marine, Westpark Marina and Tauranga Marina were 7.4, 1296, 70.6, and 713 h respectively. During winter, at typical flow rates the average residence times at Lyttelton dry dock, Orams Marine, Westpark Marina and Tauranga Marina were 7.4, 2544, 156, and 1416 h respectively. Relative modelled particle concentration in outflow at typical flow rates revealed that 100 percent particle retention with particles of a fast sink rate of  $1.0 \text{ m min}^{-1}$  was achieved at all facilities. Orams Marine, Westpark Marina (winter only) and Tauranga Marina offered 100 percent particle retention with particles of a sink rate of  $0.1 \text{ m min}^{-1}$ . Orams Marine and Tauranga Marina offered 100 percent particle retention with particles of a sink rate of  $0.01 \text{ m min}^{-1}$ . No cleaning facility offered 100 percent particle retention for particles with very slow sink rates of  $0.001$  or  $0.0001 \text{ m min}^{-1}$ .

The larger the volume of the liquid effluent treatment system in relation to the volume of effluent input, the longer the residence time of effluent from each vessel in the system. For example, Lyttelton dry dock had a relatively small treatment system volume of  $16.7 \text{ m}^3$  and large volume of freshwater used per vessel ( $300 \text{ m}^3$ ). Thus the effluent residence time was only 7.4 h (summer and winter) before discharge. Conversely, at Orams Marine the treatment system volume was  $46 \text{ m}^3$  and smaller volumes of freshwater were used per vessel ( $0.34 \text{ m}^3$ ). Thus, a residence time of 1296 h in summer was calculated for this latter facility before discharge to the municipal sewage system. The more vessels cleaned per day (that is, summer versus winter cleaning rates) the lower the residence time of water from each vessel in the system. For example, at Westpark Marina the residence time of water from each vessel was 70.6 h in summer and 156 h in winter.

The liquid effluent treatment systems of most cleaning facilities studied would theoretically kill most pest NIS based on their known salinity tolerances in relation to modelled water residence

time distributions. Lyttelton dry dock and Westpark Marina, with their shorter residence times, were likely to be less effective. The longer residence times of effluent in the treatment systems of Orams Marine and Tauranga Marina meant that for species such as the crab *Eriocheir sinensis* and the bivalve *Perna viridis* which are tolerant of reduced salinity, these species would be killed by extended freshwater exposure. Seasonal differences in residence time had little influence on treatment efficacy. Certain freshwater-tolerant/osmoconforming organisms such as the bivalve *Potamocorbula amurensis* may be able to survive for long periods within all treatment systems on the basis of salinity alone.

The results obtained in this objective clearly indicate that to maximise the efficacy of treatment systems for hull cleaning effluent, treatment systems should aim to have as high a treatment system volume to input volume ratio as possible with a salinity of as close as possible to 0 ppt in order to maximise particle settling and/or mortality of marine fouling organisms that enter them. Because even facilities with high residence times (for example, Orams Marine with a residence time of 2544 h (106 d) in winter) do not offer 100 percent retention of particles with very slow sink rates (that is, 0.001 or 0.0001 m min<sup>-1</sup>), fine particle filtering/screening (preferably down to a size range of 10–20 µm) is required to minimise discharge of any surviving organism to the sea. However, discharge of treated effluent into municipal sewage systems (that is, Orams Marine and Tauranga Marina) or similar extensive freshwater treatment system may avoid the need for fine particle filtering/screening whilst reducing the risk of marine NIS introduction (although this depends upon the nature of the waste treatment system in question). Alternatively, storage and recycling of treated effluent (that is, Tauranga Marina) for use in water blasting without discharge to the marine area or sewage system may even further minimise the risk of marine NIS introduction.

## Recommended guidelines for hull cleaning facilities

1. Cleaning of vessels should be conducted out-of-water and in a facility where **all** fouling organisms removed are quarantined from the marine environment (that is, no material removed from vessel hulls should be allowed to aerosol-drift, drain or otherwise move back into the nearby marine environment). Where out-of-water cleaning is not practicable, in-water cleaning should be conducted in such a manner that **all** fouling material removed is collected (ideally down to a particle size of 50–60 µm) and disposed of in landfill as appropriate.
2. **All** macro (>1 mm) material from vessels cleaned out-of-water should be collected and disposed of in landfill as appropriate.
3. **All** liquid effluent (runoff) from out-of-water vessel water blasting/cleaning should be collected and treated in a liquid effluent treatment system prior to discharge or recycling for water blaster use.
4. This effluent should be coarse pre-screened (for example, to 1 mm) before entry into the liquid effluent treatment system. This will reduce inorganic and organic build-up within the treatment system and thus maintain system effectiveness (for example, removal of boundary layer acceleration of suspended particles caused by sediment bed build-up) and extend the period between maintenance sediment removals. Material caught on the pre-screen should be disposed of in landfill as appropriate.
5. **All** liquid effluent should be processed through multiple settlement tanks to facilitate settling-out of any marine organisms and particles (that is, vessel hull paint flakes). Where practicable, settlement tanks should be of large volume (hydraulic capacity) and of appropriate physical design (for example, use of weirs, baffles etc.) to maximise settlement and allow as long a possible residency time/exposure time of marine organisms to freshwater before progression to a discharge or fine filtering/screening stage. Residence time of effluent

water within the treatment system should be a minimum of 24 h, but preferably >48 h. Salinity should be as close to as possible to 0 ppt to achieve 100 percent mortality of most marine organisms. Sedimented material should be regularly removed from settlement tanks and disposed of in landfill as appropriate. Flocculating and precipitating agents which facilitate separation and removal of positively and negatively buoyant particles can be used if they improve the efficiency of the system. The use of diesel/oil absorbing mats may also be appropriate.

6. Following coarse screening and passage through settlement tanks, treated effluent may be wasted to a municipal sewage/wastewater system or similar extensive freshwater treatment system for additional treatment rather than direct discharge to sea. This wasting to a municipal sewage/wastewater system (dependant upon relevant council restrictions) should further reduce marine organism viability by increasing residence time within freshwater as well as exposing any organisms to other biological and physical treatment processes and contaminants which may kill them (depending upon the nature of the waste treatment system in question).
7. Where discharge of treated effluent will be directly to the sea, following processing in settlement tanks, **all** liquid effluent should be fine filtered/screened, preferably to a size range of 10–20 µm, but 50–60 µm is an acceptable minimum to remove the smallest of most types of marine organisms before discharge.
8. As an alternative to discharge of treated effluent to the sea or sewage system, treated liquid effluent could be stored and then recycled for water blasting other vessels rather than discharged. This theoretically increases the residence time of any remaining marine organisms in freshwater (and thereby reduces their chances of survival) and reduces total freshwater usage by the cleaning facility.

# Objectives

## Overall objective

1. To determine the risk to marine biosecurity of vessel hull cleaning (defouling) and assess the efficacy of hull cleaning methods and effluent treatments in reducing this risk.

## Specific objectives

1. Building on the outcomes of ZBS2002-04, investigate the types, amounts and viability of fouling organisms discharged from a variety of hull cleaning facilities, and evaluate the efficacy of waste treatment methods being used in reducing the amount of viable material reaching the coastal marine area. These investigations and evaluations are to include both winter and summer conditions and use.
2. Identify the critical requirements for hull cleaning and waste treatment that would most effectively minimise the release of viable organisms from hull cleaning situations.

# 1 Introduction

New Zealand's indigenous biodiversity – native species, their genetic diversity, and the habitats and ecosystems that support them – is essential to the quality of life of New Zealand's citizens and their sense of identity as a nation. New Zealand's high level of endemic biodiversity makes a unique contribution to global biodiversity and places an obligation on New Zealanders to ensure its continued existence.

Human-mediated biotic invasions consist of several successive stages: 1) engagement of organisms with a transport vector, 2) transport from source to recipient location, 3) establishment of a self-sustaining population, and 4) spread through the new habitat (Carlton 1989; Mack et al. 2000). If an invasive organism has been transported to a new habitat or location, then successful establishment depends upon a number of factors, such as: survival of the invasive organisms upon first introduction, adequate invasive organism density (that is, for propagation), adequate quality or condition of invasive organisms, appropriate end-habitat environmental conditions and ineffective biotic resistance etc. (Ruiz et al. 2000; Wonham et al. 2001; Hewitt 2002).

Hull fouling is an important vector for the introduction and spread of marine non-indigenous species (NIS) in New Zealand and worldwide (Cranfield et al. 1998; Coutts 1999; James and Hayden 2000; AMOG-Consulting 2002; Floerl 2002; Hewitt 2002; Floerl and Inglis 2003; Minchin and Gollasch 2003; Floerl et al. 2004; Floerl and Inglis 2005; Floerl et al. 2005; Hayes et al. 2005; Wonham and Carlton 2005; Lewis et al. 2006; Minchin et al. 2006). For example, the recent introduction into Cairns, Australia of the invasive pest serpulid *Hyroides sanctaerucis* (a fouling pest NIS) is thought to have most likely arrived through hull-fouling on an international vessel (Lewis et al. 2006).

Local establishment of NIS can occur following release of reproductive propagules from intact fouling assemblages on vessels' hulls, or by survival of organisms removed during vessel cleaning (Environment-and-Natural-Resources-Committee 1997; Minchin and Gollasch 2003). Around 30,000 vessels are removed from the sea each year in New Zealand for cleaning. This produces approximately 140 tonnes of biogenic fouling residue (McClary and Nelligan 2001). In addition, an unknown number of vessels are cleaned each year in tidal grids, careening bays, or by divers. The treatment and disposal of waste from cleaning activities varies widely among facilities and cleaning situations. Some facilities dispose of fouling waste to landfill and liquid waste to municipal water systems or recycle their treated liquid waste for water blasting. Others discharge solid waste and/or filtered or unfiltered liquid effluent, sometimes after other treatment, into the sea. Because of the variation in treatment and disposal methods it is largely unclear what risk hull cleaning facilities may pose to New Zealand's marine biosecurity.

In 2003 the Ministry of Fisheries (MFish) contracted NIWA (MFish Research Project ZBS2002-04) to assess the risk posed to marine biosecurity by New Zealand's commercial hull cleaning facilities (Floerl et al. 2003). The specific objectives of this project were:

1. To investigate for a variety of hull cleaning situations the types, amounts and viability of fouling organisms discharged and assess whether the effluent control methodology used is successful in reducing the amount of viable material reaching the coastal marine area.
2. To discuss and make recommendations on control methodologies that would be most effective in minimizing the release of viable organisms from hull cleaning situations taking into account the efficacy, practicality and cost of using effective methodologies in an existing or new hull cleaning situation.

Between May and August 2003, NIWA staff visited hull cleaning facilities in Lyttelton, Auckland, Whangaparaoa and Tauranga, where vessels are removed from the water for cleaning (dry dock and haul-out facilities) or where fouling organisms are removed *in situ* by divers (Floerl et al. 2003). Nineteen vessels ranging from 10–104 m in length were sampled. Samples of fouling organisms and liquid effluent were taken at all stages of the hull cleaning and waste treatment process. Macrofouling was sampled before, and immediately following its removal from vessel hulls by freshwater blasting or manual scraping (in-water cleaning). Liquid fouling waste was examined before it entered settlement tanks and any filters and following treatment prior to discharge into the sea.

In all cleaning operations examined, physical removal of fouling assemblages from vessel hulls did not result in mortality of all organisms (Floerl et al. 2003). The relative survival of organisms removed from hulls was lowest in haul-out (16 percent) and dry dock (43 percent) facilities, and highest (72 percent) following in-water cleaning by divers, which did not involve exposure to air or water blasting. According to Floerl et al. (2003), the multi-chamber settlement tanks used by the facilities examined to remove solid particles from liquids (water blast effluent) were effective at killing and removing biota suspended in the liquid effluent. No biological material occurred in the final effluent of facilities that filtered (sand filter) settlement tank contents prior to their discharge into the sea. Floerl et al. (2003) noted that the residence time of water blasting effluent in settlement tanks is likely to vary between seasons because far more vessels are cleaned per day during summer months than during winter at most hull cleaning facilities. Actual liquid effluent residency time within a treatment system will be a function of the capacity of the settlement tanks and the number of vessels cleaned per day (and, therefore, the volume of water entering the tanks). It is likely therefore, that organisms and propagules contained within the cleaning effluent are subject to freshwater exposure (settlement tanks) for shorter periods during summer than during winter (ZBS2002-04).

Other environmental influences that vary seasonally may also affect the survival and viability of organisms removed from the hulls. For example, organisms removed in dry dock or haul-out facilities may become desiccated faster in hotter, summer conditions than in winter. Conversely, greater proportions of the organisms may be carrying viable offspring or gametes during the summer months which may be released upon damage or shocking incurred during hull cleaning.

In 2005, Biosecurity New Zealand (then MFish Biosecurity) contracted NIWA (Research Project ZBS2005-22) to assess the seasonal risk posed to marine biosecurity by New Zealand's commercial hull cleaning facilities. The ZBS2005-22 project repeated over a single summer season the research undertaken in ZBS2002-04 over a single winter season, so that survival and viability of organisms cleaned from vessels during a summer event could be compared with the earlier survey (ZBS2002-04). Consequently, this study investigates the same hull cleaning facilities and methods of cleaning, collecting, treatment and disposal of waste that were evaluated in the earlier research (ZBS2004-02).



## 2 Methods

### 2.1 Specific objective 1

As this project is essentially an informed extension of ZBS2002-04, we used the same methods used in ZBS2002-04 to assess the efficacy of selected hull cleaning facilities and techniques.

The five facilities assessed in ZBS2002-04 were revisited during the 2005/06 summer season. These facilities were: the dry dock at Lyttelton Port (Banks Peninsula), Orams Marine Maintenance (Waitemata Harbour), Westpark Marina (upper Waitemata Harbour), Tauranga Marina Society (Tauranga Harbour) and Gulf Harbour Marina (Whangaparaoa Peninsula, Hauraki Gulf). Detailed descriptions of these cleaning facilities as they were operating in ZBS2002-04 are summarised in Table 1a and as they operated in 2006 (during this project) are summarised in Table 1b. The operations comprised three types of cleaning situations:

1. Dry dock;
2. Hardstand cleaning operations;
3. In-water hull cleaning.

and four methods for treating fouling waste:

1. No containment of fouling waste and discharge of all material into the sea (in-water hull cleaning).
2. Separation of solid and liquid waste and discharge of unfiltered liquids into the sea.
3. Separation of solid and liquid waste and discharge of filtered liquids into the sea.
4. Separation of solid and liquid waste and storage and recycling of treated effluent and/or discharge of filtered liquids into a municipal sewage system for further treatment prior to discharge to sea.

In each facility, fouling waste arising from hull cleaning may be subject to one, two or three successive treatment stages. Treatment stage 1 consists of the actual physical removal of fouling material from vessel hulls using water blasting (vessels removed from the water) (Figure 1A, B) or hand-held scrapers (in-water cleaning). No containment of fouling material occurs in the in-water cleaning operations investigated in this study; all material thus remains in the sea. In facilities where vessels are removed from the water for cleaning, solid waste is collected and disposed of as landfill. Liquid fouling waste is collected in multi-chamber settlement tanks, where finer solids are separated from the liquid (Treatment Stage 2). After the liquid has passed through all chambers of the settlement tanks it either passes through a filter (Treatment Stage 3) prior to discharge in the sea/municipal sewage system or is discharged without filtration.

Figure 1: Initial removal of fouling epibiota by water blasting at Tauranga Marina (A) and Lyttelton dry dock (B).



A



B

**Table 1a: Hull cleaning facilities visited for seasonal assessment of hull cleaning efficacy during Research Project ZBS2002-04 (winter).**

	Facility	Location	No. vessels cleaned p.a. (no. intern'l) <sup>a</sup>	Hull cleaning operations visited	Separation of solids and liquids	Filtration of liquids	Disposal Solids/Liquids
Vessels removed from water for cleaning	Lyttelton Port Co.	Lyttelton	70 (7)	Dry dock Slipway	Settling tanks & flocculating agent	No	Landfill/Sea
	Orams Marine Maintenance	Auckland	2000 (15)	Travel lift Slipway	Grit arrestors	Sand filter	Landfill/Sea
	Westpark Marina	Auckland	2000 (50)	Travel lift	Settling tanks	20 mm screen	Landfill/Sea
	Tauranga Marina Society	Tauranga	2000 (8)	Travel lift	Settling tanks	Sand filter 100 µm	Landfill/Sea
Vessels cleaned in the water	Gulf Harbour Marina	Whangaparaoa	Unknown	Diver services	n/a	n/a	Sea
	Orams Marine Maintenance	Auckland	Unknown	Diver services	n/a	n/a	Sea

<sup>a</sup> Source: McClary and Nelligan (2001). These figures refer to the sum of vessels cleaned in all available operations offered by the facilities. For example 2000 vessels are cleaned in the Tauranga Marina each year; 5 percent on the tidal grids and 95 percent on the hardstand (using a travel lift).

The number in brackets is the number of international vessels (those arriving from overseas).

**Table 1b: Hull cleaning facilities visited for seasonal assessment of hull cleaning efficacy sampled during Research Project ZBS2005-22 (summer).**

	Facility	Location	No. vessels cleaned p.a. (no. intern'l) <sup>b</sup>	No. vessels cleaned in summer/winter	Hull cleaning operations visited	Separation of solids and liquids	Filtration of liquids	Disposal Solids/Liquids
Vessels removed from water for cleaning	Lyttelton Port Co.	Lyttelton	87 (10)	22/22	Dry dock Slipway	Settling tanks & flocculating agent/Sand filter	Sand filter	Landfill/Sea
	Orams Marine Maintenance	Auckland	600 (0)	300/150	Travel lift Slipway	Settling tanks/Sand filter	Sand filter	Landfill/Sewage
	Westpark Marina	Auckland	1500 (6)	600/300	Travel lift	Settling tanks	20 mm	Landfill/Sea
	Tauranga Marina Society	Tauranga	1600 (15)	600/300	Travel lift	Settling tanks/Sand filter	Sand filter	Landfill/Sewage
Vessels cleaned in the water	Gulf Harbour Marina	Whangaparaoa	Unknown	Unknown	Diver services	n/a	n/a	Sea
	Orams Marine Maintenance	Auckland	Unknown	Unknown	Diver services	n/a	n/a	Sea

<sup>b</sup> Source: cleaning statistics and waste treatment methods for each facility are based on interviews with cleaning facility staff.

The number in brackets is the number of international vessels (those arriving from overseas).

## 2.1.1 Facilities studied

### 2.1.1.1 Lyttelton dry dock

The Lyttelton dry dock is operated by the Lyttelton Port Company (<http://www.lpc.co.nz>). It is the only dry dock in New Zealand's South Island and can accommodate vessels of up to 120 m in length. Ships are manoeuvred into the dock basin and the dock is then closed using sealed lock gates. The dock water is usually drained overnight. During draining, wooden beams are wedged between the dock walls and the vessels to keep the ships in an upright position (Figure 2). Services offered by the dry dock include mechanical repairs, removal of fouling and application of antifouling paint. Cleaning of vessels is carried out using up to four water blasters (freshwater; 12 000 psi).

**Figure 2: Lyttelton dry dock: (A) Independent I about to be cleaned, (B) raw liquid effluent collecting on dock floor, (C) liquid effluent treatment system, and, (D) final settlement tanks in treatment system.**



**A**



**B**



**C**



**D**

Treatment of fouling waste (Figures 2A-D): Solid fouling waste is collected, lime-stabilised and disposed of as landfill. The liquid runoff from the water blasting is pumped from the dock floor and collected in an initial two-chambered settling tank. It then runs to a mixing tank where Alum (Aluminium Sulphate) is added to balance pH and help aid sedimentation. The liquid effluent then passes to a tank where the flocculating agent Magnafloc® is added to aid solids separation, and finer particulates are allowed to settle. Liquids then pass through two further settling tanks before being filtered through a sand filter. This sand filter has been added since the 2003 winter study (ZBS2002-04). Liquid contents from the sand filter pass back into a

chamber on the dock floor (separated from the raw effluent) before being discharged back into the sea. Solid waste is removed from the settlement tanks on average every three months for landfill disposal.

#### **2.1.1.2 Westpark Marina**

In the Westpark Marina (<http://www.westpark.co.nz>), vessels up to 25 m in length are removed from the water using two travel lifts. Services offered include mechanical repairs, removal of fouling and application of antifouling paint. Cleaning of vessels is carried out using a water blaster (freshwater; 4000 psi) on a bunded sealed area.

Treatment of fouling waste: Solid fouling waste is collected and disposed of as landfill. The liquid runoff from the water blasting is collected in a sump, then gravity-fed into a triple-chamber, in-ground Humes-type “oil and grit separator” (settling tank). Coarse solids settle out in the first chamber. The liquid stage then passes through another two chambers, separated by a weir that allows fine particulates to settle out and oils to separate (through addition of “Matasorb cushions”). The final tank has an overflow outlet from where the water is discharged through a 20 mm screen without filtration into an adjacent intertidal mangrove area via the local stormwater system. No flocculation agents are added to the settling tanks at any stage. Solid waste is removed from the tanks on average every three months or more frequently if required to maintain efficiency of the system.

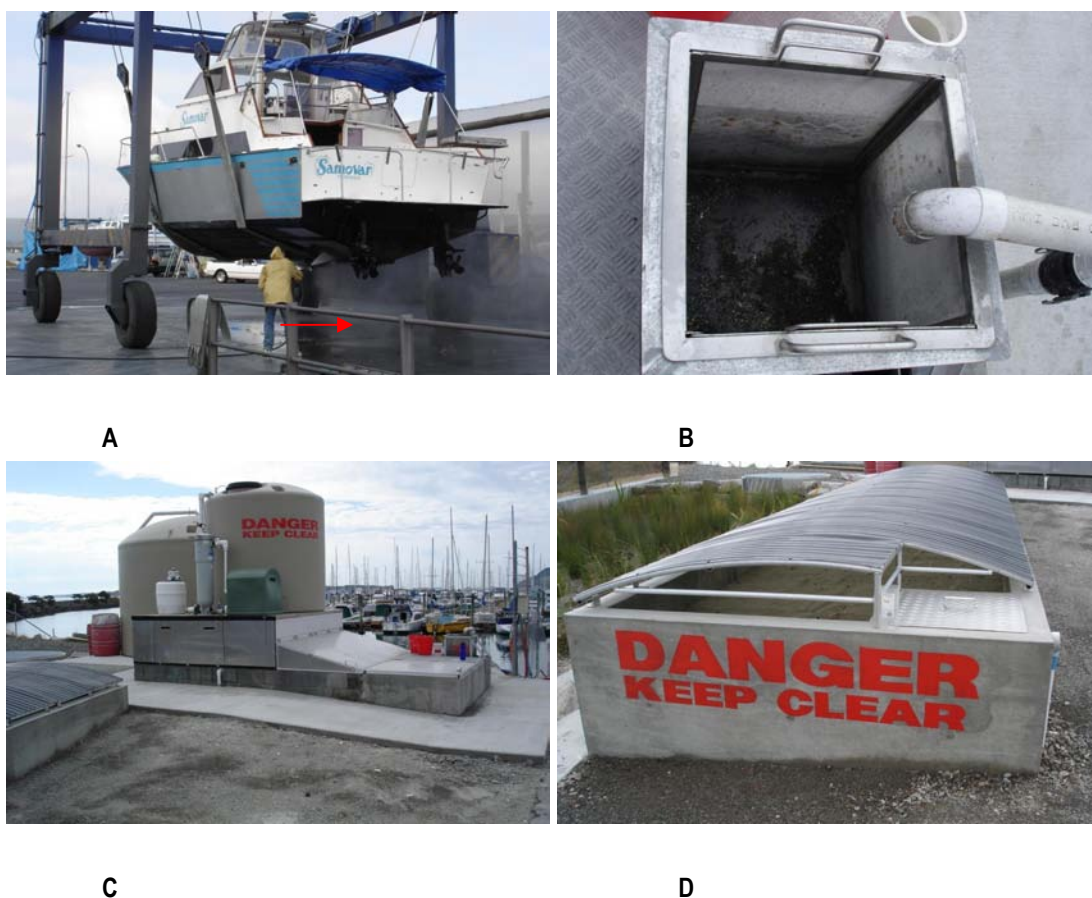
#### **2.1.1.3 Tauranga Marina Society**

The Tauranga Marina Society offers a travel lift and tidal grids for vessel maintenance. The travel lift can remove vessels up to 20 m in length from the water. Services offered include mechanical repairs, removal of fouling and application of antifouling paint. Cleaning of vessels is carried out using a water blaster (freshwater; 3500 psi) on a bunded sealed area.

Treatment of fouling waste (Figures 3A-D): Solid fouling waste is collected and disposed of as landfill. Liquid runoff from water blasting is collected in a sump tank, then pumped through a 1 mm pre-screen (screened material is disposed of as landfill) into the first of three settling tanks, where coarse solids settle out. The liquid stage then passes through another two settling tanks, which are separated by a weir allowing fine particulates to settle. The final tank has an overflow outlet from where the water is discharged into a large sand filter. After liquids have been filtered through the sand filter they are pumped to a bag filter (10 µm), then pass into a large storage tank. Liquids typically remain in the storage tank for up to 3 days, allowing further particulate settlement. This treated liquid is then recycled for use in water blasting. If excess treated liquids enter the storage tank, then the excess liquid overflows to a secondary storage tank which collects rainwater runoff from the bunded sealed area for settlement and eventual wasting to the municipal sewage system. No flocculation agents are added to the settling tanks at any stage. Solid waste is removed from the tanks on average every three months. The sand filter, bag filter and storage tanks for recycling treated liquid effluent are all new additions since the 2003 winter study (ZBS2002-04).



Figure 3: Tauranga Marina Society travel lift: (A) Samovar being cleaned, (B) 1 mm primary screen before settlement tanks, (C) liquid effluent treatment system, and, (D) sand filter. In (A), the red arrow indicates the sump grill below Samovar.

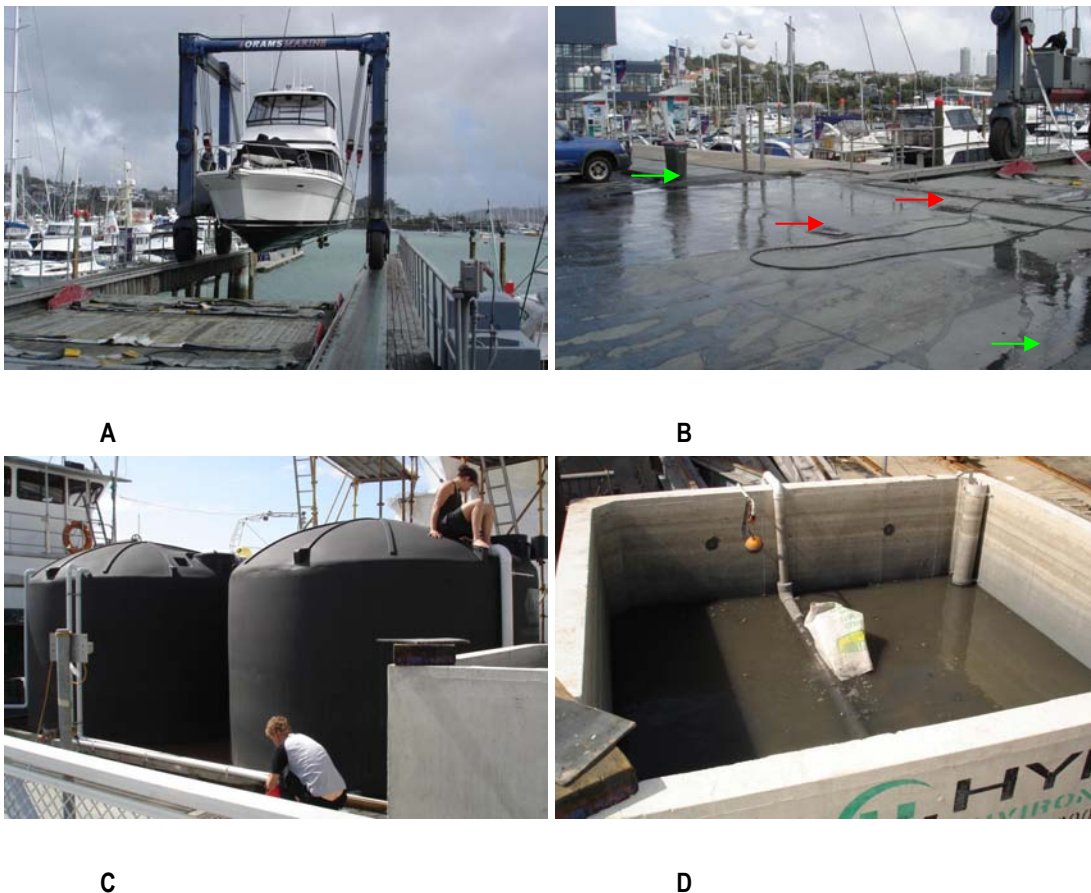


#### 2.1.1.4 Orams Marine Maintenance

At Orams Marine Maintenance (<http://www.orams.co.nz>), vessels up to 25 m in length are removed from the water using a travel lift. A slipway service is also operated for vessels up to 54 m in length. Services offered include mechanical repairs, removal of fouling and application of antifouling paint. Cleaning of vessels is carried out using a water blaster (freshwater; 2500 psi) on a bunded sealed area.

Treatment of fouling waste (Figures 4A-D): Solid fouling waste is collected and disposed of as landfill. The liquid runoff from the water blasting is collected in a sump tank then pumped through to the first of two successive large settlement tanks for coarse particulate settlement. This first tank has an overflow outlet that leads into a second tank, where fine particulates are allowed to settle. Overflow liquids from the second tank are then pumped to a sand filter. After liquids have been filtered through the sand filter they are pumped to waste in the municipal sewage system. This wasting to the municipal sewage system is a new addition since the 2003 winter study (ZBS2002-04). No flocculation agents are added to the settling tanks at any stage. Solid waste is removed from the tanks on average every three months.

Figure 4: Orams Marine Maintenance travel lift: (A) Phase II about to be cleaned, (B) raw effluent collection area and sump grills, (C) liquid effluent settlement tanks, and, (D) sand filter. In (B), the red arrows indicate sump grills and the green arrows sealed area bunding.



The methods for sampling and analysis to be employed follow those used in ZBS2002-04 and are described below.

### 2.1.2 Definition of a viable organism

For a fouling organism removed from a boat hull to establish a self-sustaining population in New Zealand waters, it must pass through two stages:

- survive the cleaning process and be returned to the sea, and
- be able to grow and produce offspring which are themselves capable of surviving and reproducing in New Zealand conditions.

In this project a viable organism (adult or propagule) is defined as being one that is “potentially capable of living and developing normally in the marine environment”. This simply means that the plant or animal has survived the cleaning process (Stage 1) and is in a condition that would *potentially* allow it to grow and produce offspring.

The likelihood of successful establishment of populations in New Zealand waters involves interactions between the organism, its local environment and present enemies (competitors, predators and parasites) and cannot be addressed without complex field experimentation and testing.

This simple definition of viability has some difficulties, as many marine species (particularly macroalgae and clonal invertebrates) are able to regenerate from very small fragments. The likely survival of these fragments can only be determined definitively by culturing them in the



laboratory. However, this project took a pragmatic approach that used field assessments of physiological condition (see Appendix 1) and best available knowledge as surrogates for more complex tests of viability.

### 2.1.3 Sampling of hull-fouling assemblages before and after removal from hulls

To obtain an estimate of the proportion of organisms that survive the hull cleaning (defouling) process and/or the subsequent treatment methods, the amount of fouling on a hull (expressed as wet weight) was determined before cleaning commenced. For each vessel, the “total wetted surface area (TWSA)” was determined as in ZBS2002-04 using formulae developed by the antifouling paint industry (Akzo Nobel, pers. comm. 2003). Separate formulae were used for different classes of vessel:

#### **Regular yachts:**

$$\text{TWSA} = 2 \times \text{Length} \times \text{Draft}$$

#### **Super yachts and trawlers:**

$$\text{TWSA} = (2 \times \text{Length} \times \text{Draft}) + (\text{beam} \times \text{draft})$$

#### **Large ships (>100 m length):**

$$\text{TWSA} = (\text{Length between perpendiculars} \times (\text{Beam} + (2 \times \text{light load draft}))) \times 0.72$$

Calculation of TWSA for each vessel used measurements provided from either the vessel owners or from technical plans provided by the operators of the cleaning facilities. An estimate of the proportion of the TWSA covered by fouling organisms was obtained by taking digital images of random areas around each hull (including main hull area, rudder and keel) (image size 20 x 20 cm; Sony Cyber Shot DSC-W5) and superimposing 60 random dots to calculate the average fouling cover per image. Ten randomly placed replicate images were taken for boats of 30 m length or less, while 20 images were taken of larger ships. The total biomass (weight) of fouling organisms on each hull was then determined by measuring the average fouling weight in three 400 cm<sup>2</sup> areas around the hull, and extrapolating the derived value to the estimated total hull area covered by fouling organisms. Fouling assemblages on vessel hulls are usually not distributed randomly (Coutts 1999; James and Hayden 2000). However, we did not use a stratified sampling approach to determine fouling cover in this study as this would have necessitated individual area calculations for the various strata, which was beyond the scope of this project. Instead, we used a randomised approach to obtain a broad estimate of fouling intensity on vessels and to ensure comparison between the projects.

After a vessel's hull was cleaned of fouling organisms by water blasting and/or scraping, samples of the removed biota were collected from the surrounding area by filling four replicate 1 L containers with fouling material collected haphazardly from the ground below and around the vessel and kept in a cool, shaded place. For each vessel, the times of its removal from the water, completion of the cleaning process and the collection of samples were recorded. Fouling cover and biomass varied among the vessels examined in this study. While in some cases enough fouling material was available to fill four 1 L jars, only one jar could be filled following the cleaning of other boats. Air temperature, sunlight intensity and approximate weather conditions at point of sample collection were also measured.

Following collection, each container was emptied into a sorting dish within one hour of collection and the types of organisms or fragments, their size and their state of structural damage (either completely intact or exhibiting some degree of damage) and dryness (desiccation recorded as either percentage wet/moist, percentage rather dry or percentage with complete lack of moisture) recorded. Organisms and fragments were separated by phylum or major taxonomic group (for example, barnacles, bivalves, colonial ascidians) into additional sorting dishes. These dishes were flooded with clean filtered (60 µm) seawater and left

undisturbed for 20–30 minutes to allow the organisms to recover. The organisms and fragments in each sorting dish were then examined under magnification using either a handheld magnifying glass (5x magnification) or a Wild M-7A dissecting microscope (31x magnification) for signs of active feeding and movement (Figure 5). Decisions as to whether an organism or fragment was viable were based on criteria developed with guidance from NIWA taxonomists (see Table 2 and Appendix I for detail). This sequence of activities was continued until all four 1 L containers have been emptied and examined.

**Figure 5: Visual check for signs of organism viability after immersion in seawater for 20-30 min (for example, barnacles, note feeding limbs indicated by red arrow).**



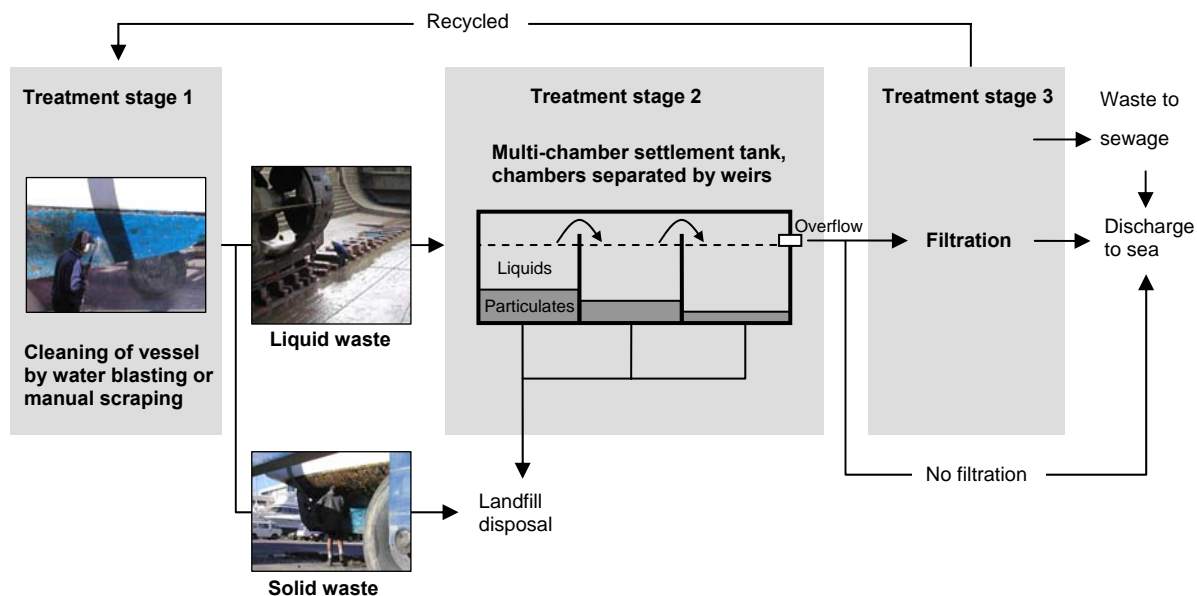
**Table 2: Biological data to be collected during visits to hull cleaning facilities.**

Group	Count	Size of organism or fragment	% intact alive (incl. weight)	% damaged/fragmented alive	% dead	Time out of sea	Degree of dryness
1. Organisms removed from hulls							
Barnacles	✓		✓	✓	✓	✓	
Bivalves	✓		✓	✓	✓	✓	
Encrusting bryozoans	✓	✓	✓	✓	✓	✓	✓
Erect bryozoans	✓	✓	✓	✓	✓	✓	✓
Hydroids	✓	✓	✓	✓	✓	✓	✓
Tubicolous polychaete worms	✓		✓	✓	✓	✓	✓
Sponges	✓	✓	✓	✓	✓	✓	✓
Colonial ascidians	✓	✓	✓	✓	✓	✓	✓
Solitary ascidians	✓	✓	✓	✓	✓	✓	✓
Macroalgae	✓	✓	✓	✓	✓	✓	✓
Other taxa	✓	✓	✓	✓	✓	✓	✓
2. Liquid effluent	Count	Mitochondria intact?	% damaged				
Larvae	✓	✓	✓				
Eggs	✓	✓	✓				
Spores	✓	✓	✓				

### 2.1.4 Sampling of liquid effluent prior to discharge

In the haul-out facilities four 10 L samples of liquid effluent were taken at up to four separate stages in the treatment process. The number of the stages sampled varied according to cleaning facility systems and between sampling season as some facilities had upgraded their liquid effluent treatment system since 2003. Wherever possible, and for each vessel sampled, liquid effluent was collected (1) from the water blast runoff before it enters the settlement tanks, (2) from the first chamber of the settlement tanks, (3) from the final chamber of the settlement tank prior to any filtering, and (4) from the discharge pipe following settlement and any filtration (Figure 6).

Figure 6: Various sampling stages for liquid effluent following hull cleaning.



In 2003 (ZBS2002-04, winter sampling) (Table 3), samples of liquid effluent were collected: (1) from the water blast runoff before it entered the settlement tanks (Lyttelton, Orams and Westpark: two vessels per facility; Tauranga: three vessels), (2) from the first chamber of the settlement tanks (all four facilities), (3) from the final chamber of the settlement tank (all four facilities), or (4) from the discharge pipe following settlement and filtration (sand filter) (Lyttelton and Orams). In the Tauranga Marina, sampling at (4) was attempted on various occasions but could not be achieved as, at the time of our visits, the settlement tanks were not sufficiently full to cause overflow of tank contents into the sand trap following cleaning of vessels. At Westpark Marina sampling at (4) was not possible as final treated water was discharged directly into the local stormwater system.

In 2006 (ZBS2005-22, summer) (Table 3), samples of liquid effluent were collected: (1) from the water blast runoff before it entered the settlement tanks (all four facilities: three vessels per facility), (2) from the first chamber of the settlement tanks (all four facilities), (3) from the final chamber of the settlement tank (all four facilities), or (4) from the discharge pipe following settlement and filtration (sand filter) (Lyttelton and Tauranga). At Orams, sampling at (4) was not possible as final treated water was discharged directly through a sealed pipe into the municipal sewage system. At Westpark Marina sampling at (4) was not possible as final treated water was discharged directly into the local stormwater system.

**Table 3: Sample collection of liquid effluent during winter (ZBS2002-04) and summer (ZBS2005-22) hull cleaning surveys. Samples taken or not taken are indicated by '+' and '-', respectively.**

		Winter	Summer
Lyttelton dry dock	Cleaning runoff	+	+
	First tank	+	+
	Final tank	+	+
	Final discharge	+	+
Orams Marine	Cleaning runoff	+	+
	First tank	+	+
	Final tank	+	+
	Final discharge	+	-
Westpark Marina	Cleaning runoff	+	+
	First tank	+	+
	Final tank	+	+
	Final discharge	-	-
Tauranga Marina	Cleaning runoff	+	+
	First tank	+	+
	Final tank	+	+
	Final discharge	-	+

All samples of effluent were filtered through a 60 µm mesh to retain the biological material within them. A combination of visual observations and vital staining analysis were performed on three replicate subsamples (2 ml) from each 10 L sample to determine the viability of small organisms or propagules. The samples were added to fresh seawater and left undisturbed for 20–30 minutes before examination under a microscope for movement and damage. The material was then filtered again and one drop of filtrate added to a cover slide together with one drop of the vital stain Janus Green B (made up from distilled water at 1:10,000). This was then examined under a compound microscope within 36 hours. Janus Green stains mitochondria in eukaryotic tissue samples (Clark 1973). Like all other available stains, Janus Green does not provide information on mitochondrial or cellular activity. However, as eukaryotic cells degenerate soon after they die the colour and intensity of the stain can provide an indication of whether the contents of a sample are in their original state (staining visible within the superstructure (for example, exoskeleton)) or whether mortality has occurred a considerable time ago (staining dispersed throughout the sample due to tissue degeneration). In this study, the combined use of visual observations (movement) and vital stains is useful for rapid assessment of viability of small organisms or propagules in liquid samples. The remaining filtrate from each 10 L sample was made up to 50 ml of 5 percent formaldehyde/seawater and transported to a laboratory for further analysis. Three subsamples (2 ml each) were then taken from each 50 ml filtrate sample and organisms, propagules and their fragments presented within them identified and enumerated using a Leitz Fluovert FS microscope (100x magnification). The abundance of all organisms and propagules was estimated using direct counts, with the exception of filamentous algae, for which a rank scale of abundance was used (0 –

absent; 1 – very low abundance; 2 – low abundance; 3 – moderate abundance; 4 – high abundance; 5 – very high abundance).

### **2.1.5 Sampling of vessels cleaned in the water**

An assessment of the viability of fouling organisms removed from vessel hulls during in-water cleaning operations was made in Gulf Harbour Marina and Orams Marine. In both June 2003 (winter survey) and March 2006 (summer survey), three vessels were sampled at each of these facilities. Before the cleaning, the amount of fouling on each vessel hull was determined using the techniques described above. NIWA divers then mimicked in-water cleaning by removing fouling organisms from randomly chosen locations on the hull of each vessel using the same method (a paint scraper) used by many hull cleaning facilities and private individuals. A soft cotton cloth was used to remove fouling organisms from vessel hulls in the Gulf Harbour Marina in 2006, where the use of scrapers for in-water hull cleaning is not allowed as per Auckland Regional Council (ARC) directive. Fouling organisms removed from hulls were collected using catch bags made from fine nylon mesh (0.2 mm) attached to the fouling assessment quadrats. The quantity of fouling organisms collected was standardised to the same volume used for vessels cleaned in haul-out facilities by transferring the contents of the mesh bags into four (or fewer, depending on the material available) replicate 1 L containers. Immediately after collection, all material was placed in sorting trays filled with clean filtered (60 µm) seawater, and the viability of organisms was assessed using the procedures described above (Section “Sampling of hull-fouling assemblages before and after removal from hulls”). Our approach did not include an assessment of propagules (eggs and larvae) released from adult organisms during in-water hull cleaning, as it was not possible to distinguish these from other sources of propagules in the water column or to tell if material removed from the hull had recently released gametes.

### **2.1.6 Assessment of seasonality**

ZBS2002-04 assessed survival and viability of organisms removed from vessel hulls exclusively during winter months (May–August 2003). In ZBS2005-22, sampling occurred from January to May 2006. Sampling locations and number of vessels sampled for ZBS2005-22 was the same as for ZBS2002-04. This provided a balanced dataset and allowed for basic statistical comparisons to be made between the condition (for example, degree of desiccation) and survival of de-fouled organisms in a single winter and a single summer period at a range of stages in the cleaning process (for example, just after removal from a hull versus upon entry into settlement tanks) and between shore-based and in-water cleaning facilities (see Tables 1a, b).

A rigorous assessment of the repeatability of any seasonal patterns in the survival of organisms in cleaning facilities would require sampling in more than a single summer and winter season. However, this was beyond the scope of this project.

### **2.1.7 Statistical analyses**

#### **2.1.7.1 Solid fouling samples**

Analysis of variance (ANOVA) was used to test for differences in: (1) fouling percentage cover, (2) fouling biomass, (3) period of exposure of fouling organisms to air prior to cleaning, (4) duration of the cleaning process, and (5) viability of organisms sampled from vessels cleaned in the various operations during winter and summer. No statistical analyses were carried out on viability of individual taxonomic groups (for example, barnacles, bryozoans, etc.) due to the large variation in the number of specimens examined on different vessels in different operations and seasons. For example, the number of bryozoans and tubicolous polychaetes examined on the 37 vessels sampled varied by a factor of 33.3 and 821.5 between cleaning operations, respectively. This unbalanced design does not allow for robust statistical comparisons. Statistical tests on

organism viability were only performed on the variable ‘total organisms’, that is, where all organisms or fragments of different taxonomic groups had been pooled. The linear model for this consisted of two factors, Season (fixed) and Operation (random), and the interaction of these factors. Because varying numbers (1 to 4) of replicate 1 L jars had been examined for the various vessels depending on fouling biomass, this factor was omitted from the design and individual vessels were used as the replicates in the model.

Data used for ANOVA were checked for normality and homogeneity of variances (Cochran’s C test). Dependent variables were  $\log(x+1)$  transformed if untransformed data had heterogeneous variances. However, if transformation did not remove heterogeneity of variances then untransformed data were used for the analysis (Underwood 1997).

Similarity Percentages analysis (SIMPER, Plymouth Marine Laboratories, Primer software v5.1) was used to determine the relative contribution of the various taxonomic groups to the differences in fouling percentage cover between summer and winter surveys determined by ANOVA.

#### **2.1.7.2 Liquid samples**

Analysis of variance (ANOVA) was used to test for differences in the numbers of animals, propagules and unicellular organisms in the raw cleaning effluent among different cleaning facilities and cleaning season. The linear model for this consisted of two factors, Season (fixed) and Facility (random), and the interaction of these factors. Because varying numbers (1 to 4) of replicate 1 L jars had been examined for the various vessels depending on fouling biomass, this factor was omitted from the design and individual vessels were used as the replicates in the model.

Data used for ANOVA were checked for normality and homogeneity of variances (Cochran’s C test). Dependent variables were  $\log(x+1)$  transformed if untransformed data had heterogeneous variances. However, if transformation did not remove heterogeneity of variances then untransformed data were used for the analysis (Underwood 1997).

## **2.2 Specific objective 2**

In specific objective 2, the likelihood of organisms with a range of sinking rates and freshwater tolerances to pass through settlement tanks during a summer and a winter period, where the frequency of vessel cleaning events – and the associated volume of water entering the tanks – is higher and lower, respectively was assessed. In each type of cleaning operation, we assessed the efficacy of each facility’s effluent treatment process in successfully killing and/or retaining organisms or their propagules removed from vessel hulls.

The potential for viable organisms to pass through one or several settlement tanks in an effluent treatment system and be returned back into the marine environment is a function of: (1) the residency time of liquids in the tank(s), (2) the ability of the organisms to tolerate low salinity conditions, and (3) the sinking rate of the organism. The latter is of importance, since even if an organism can tolerate exposure to low salinities for longer than the residency time of liquids in a settlement tank, the organism is unlikely to survive the treatment process if it quickly sinks out of suspension and is not discharged from the tanks in the final effluent.

### **2.2.1 Hydraulic residence time and sinking rate**

The residency time of liquid waste in the settlement tanks of shore-based hull cleaning facilities is determined by:

the total settlement tank volume;

1. the number of successive tanks comprising this volume (for example, single tank vs. multi-chamber tanks);

2. the average volume of water required to clean a vessel and rate of entry of this water into the treatment system;
3. the number of vessels cleaned per unit time (for example, per week); and
4. the average volume of any additional liquid entering the tanks per unit time (for example, average rainfall per week).

Information on the above variables was gathered through on-site interviews with facility operators and maintenance staff for each shore-based cleaning facility to determine the approximate residency time of liquid waste in settlement tanks during the summer and winter season.

The determination of sinking rates for various fouling organisms commonly found on vessel hulls without actual *in situ* or laboratory determination of sinking rates (as was recommended to BNZ in the original tender) is problematic. Derivation of sinking rates applicable to marine bio-foulers from available literature is difficult as most available literature concentrates on free-living organisms or detrital agglomerations collectively referred to as “Marine snow” (organic aggregates >0.5 mm in diameter). Organism-specific sinking rates may be temporally and spatially variable according to: cell nutrient status, light levels, dissolved gas concentrations, organism density, organism shape (for example, coefficient of form resistance), size, passive or active behaviour, phase of growth, physical mixing processes (for example, boundary layer effects, turbulence and shear stress), solitary or aggregating stage/state (for example, chain-forming diatoms or organisms trapped in organic aggregates), and salinity/temperature/viscosity of the medium (Foxon 1934; Smayda and Boleyn 1965; Prakash 1967; Winet 1973; Titman and Kilham 1976; Forward et al. 1984; Anderson et al. 1985; Asper 1987; Alldredge and Gotschalk 1988; Shanks and Edmondson 1990; van Leeuwen and Maly 1991; Luckenbach and Orth 1992; Riebsell 1992; Callieri 1997; Noji et al. 1997; Ghosal et al. 2000; Knutsen et al. 2001; McHenry 2001; Hamm 2002; Thornton 2002; Peperzak et al. 2003; Titelman and Kiorboe 2003; Fuchs et al. 2004; Waite et al. 2005; Wang et al. 2005; Ouellet and Allard 2006).

Estimates of sinking rates of organisms were derived from relevant literature (see Table 4) and simple mathematical models based on assumed density, volume and surface area in freshwater (given limitations outlined as above). The sinking rates given in Table 4 are for organisms and detrital aggregations that sink passively.



**Table 4: Known sinking rates for various marine organisms and organic aggregates (marine snow).**

Taxon/category	sub Taxa	Species	Life stage	Radius (mm)	Length (mm)	Sink rate (m min <sup>-1</sup> )	Reference
Annelida	Nematode					0.022	(Shanks and Edmondson 1990)
Annelida	Polychaete		Larva			0.025	(Shanks and Edmondson 1990)
Crustacea	Copepod	<i>Centropages tenuiremis</i>	Egg			0.016-0.022	(Wang et al. 2005)
Crustacea	Copepod	<i>Calanus finmarchicus</i>	Egg			0.016-0.025	(Knutsen et al. 2001)
Crustacea	Copepod	<i>Calanus glacialis</i>	Egg			0.018	(Knutsen et al. 2001)
Crustacea	Copepod					0.01	(Shanks and Edmondson 1990)
Crustacea	Copepod					0.009	(Shanks and Edmondson 1990)
Crustacea	Decapoda	<i>Rhithropanopeus harrisi</i>	Larva			0.163-0.443	(Forward et al. 1984)
Crustacea	Decapoda	<i>Porcellana sp.</i>	Larva			0.706	(Foxon 1934)
Crustacea	Decapoda		Larva			0.459-0.719	(Foxon 1934)
Crustacea	Copepod	<i>Acartia tonsa</i>	Adult		0.118-0.229	0.008-0.0198	(Titelman and Kiorboe 2003)
Crustacea	Copepod	<i>Calanus helgolandicus</i>	Adult		0.231-0.553	0.016-0.046	(Titelman and Kiorboe 2003)
Crustacea	Copepod	<i>Centropages typicus</i>	Adult		0.132-0.225	0.003-0.008	(Titelman and Kiorboe 2003)
Crustacea	Copepod	<i>Eurytemora affinis</i>	Adult		0.132-0.202	0.006-0.011	(Titelman and Kiorboe 2003)
Crustacea	Copepod	<i>Euterpina acutifrons</i>	Adult		0.112-0.200	0.005-0.016	(Titelman and Kiorboe 2003)
Crustacea	Copepod	<i>Temora longicornis</i>	Adult		0.138-0.308	0.005-0.014	(Titelman and Kiorboe 2003)

Crustacea	Decapoda	<i>Pandalus borealis</i>	Larva		6.4-13.2	0.18	(Ouellet and Allard 2006)
Echinoderm	Urchin	<i>Strongylocentrotus franciscanus</i>	Egg			0.024-0.031	(McDonald 2004)
Echinoderm	Urchin	<i>Strongylocentrotus pupuratus</i>	Egg			0.023-0.027	(McDonald 2004)
Echinoderm	Urchin	<i>Strongylocentrotus droebachiensis</i>	Egg			0.022-0.023	(McDonald 2004)
Echinoderm	Sand dollar	<i>Dendraster excentricus</i>	Egg			0.018-0.022	(McDonald 2004)
Mollusca	Gastropod	<i>Alderia modesta</i>	Larva		0.126-0.193	0.054-0.059	(Krug and Zimmer 2004)
Mollusca	Gastropod	<i>Ilyanassa obsoleta</i>	Larva		0.59-0.77	0.383-0.588	(Fuchs et al. 2004)
Mollusca	Bivalve	<i>Mytilus edulis</i>	Larva		0.5-2.0	0.6	(Lane et al. 1985)
Phycophyta	Phytoplankton	<i>Asterionella formosa</i>	Adult	0.001		0.00005-0.0002	(Titman and Kilham 1976)
Phycophyta	Phytoplankton	<i>Scenedesmus quadricauda</i>	Adult	0.004		0.0002-0.001	(Titman and Kilham 1976)
Phycophyta	Phytoplankton	<i>Cyclotella meneghiniana</i>	Adult	0.012		0.0001-0.001	(Titman and Kilham 1976)
Phycophyta	Phytoplankton	<i>Melosira agassizii</i>	Adult	0.027		0.0004-0.001	(Titman and Kilham 1976)
Phycophyta	Dinoflagellate	<i>Scrippsiella trochoidea</i>	Adult	0.014	0.029	0.008	(Anderson et al. 1985)
Phycophyta	Dinoflagellate	<i>Gyrodinium uncatenum</i>	Adult	0.017	0.048	0.005	(Anderson et al. 1985)
Phycophyta	Dinoflagellate	<i>Gonyaulax tamarensis</i>	Adult	0.021	0.047	0.007	(Anderson et al. 1985)
Phycophyta	Dinoflagellate	<i>Ceratocorys horrida</i>			0.048-0.064	0.017-0.02	(Zirbel et al. 2000)
Phycophyta	Diatom	<i>Coscinodiscus concinnus</i>	Adult		0.146	0.0004-0.0007	(Granata 1991)
Phycophyta	Diatom	<i>Thalassiosira cf. nana</i>	Adult			0.00003-0.0004	(Smayda and Boleyn 1965)

Phycophyta	Diatom	<i>Thalassiosira rotula</i>	Adult		0.012-0.018	0.0001-0.031	(Smayda and Boleyn 1965)
Phycophyta	Diatom	<i>Nitzschia seriata</i>	Adult			0.0001-0.002	(Smayda and Boleyn 1965)
Phycophyta	Diatom	<i>Rhizosolenia shrubsolei</i>	Adult		0.23	0.0003	(Peperzak et al. 2003)
Marine snow					>1	0.019	(Shanks and Edmondson 1990)
Marine snow					<0.01	0.00004-0.003	(Callieri 1997)
Marine snow					0.01-0.05	0.00004-0.008	(Callieri 1997)
Marine snow					>0.05	0.0007-0.026	(Callieri 1997)
Marine snow					>1	0.056-0.75	(Noji et al. 1997)
Marine snow					2.0-16	0.032-0.214	(Riebsell 1992)
Marine snow						0.028-0.035	(Asper 1987)
Marine snow					2.4-75	0.051	(Alldredge and Gotschalk 1988)

## 2.2.2 Modelling water residence time and particle settling in treatment systems

### 2.2.2.1 Water residence time

Let  $T$  be the residence time (the elapsed time from entrance to exit) of a water particle in a single reservoir (for example, a settling tank) within a water treatment system. The relative frequency distribution for all water particles passing through the reservoir is given by the probability density function (pdf)  $p(T)$ . We assume that  $p(T)$  takes the form of the exponential pdf:

$$p(T) = \frac{1}{\mu} e^{-T/\mu} \quad (1)$$

where  $\mu$  is the mean residence time inside the reservoir. The flow rate  $Q$  through the reservoir is assumed to be steady (and water homogeneously mixed) and  $\mu$  is estimated as  $V/Q$ , where  $V$  is the volume of water in the reservoir.

For a system comprised of  $N$  reservoirs connected in series, the cumulative total residence time  $T_{tot}$  of an arbitrary water particle passing through the entire system is given by:

$$T_{tot} = \sum_i^N T_i \quad (2)$$

where  $i$  denotes the  $i$ th reservoir with mean residence time  $\mu_i$ . Consequently, the system pdf of  $T_{tot}$  is:

$$p(T_{tot}) = p(T_1 + T_2 + \dots + T_N) \quad (3)$$

Equation (3) is solved by convolving the pdfs of all  $T_i$ :

$$p(T_{tot}) = p(T_1) * p(T_2) * \dots * p(T_N) \quad (4)$$

where  $*$  denotes the convolution integral:

$$f * g = \int_0^t f(\tau)g(t-\tau)d\tau \quad (5)$$

To simplify notation, let  $p_i = p(T_i)$  so (4) becomes  $P_{tot} = P_1 * P_2 * \dots * P_N$ . The first convolution in (4), denoted as  $p_{1*2}$ , is:

$$p_{1*2} = \int_0^{\tau} p_1(t)p_2(\tau-t)dt \quad (6)$$

The second convolution is thus carried out as:

$$p_{1*2*3} = \int_0^{\tau} p_{1*2}(t)p_3(\tau-t)dt \quad (7)$$

The process is repeated up to:

$$p_{total} = \int_0^{\tau} p_{1*2*...*N-1}(t)p_N(\tau-t)dt \quad (8)$$

The integrals (6) through (8) were solved numerically.

#### 2.2.2.2 Particle settling

Calculation of particle (organism) concentrations within a reservoir is based on the conservation of mass of water and of particles:

$$\frac{dV}{dt} = Q_{out} - Q_{in} \quad (9)$$

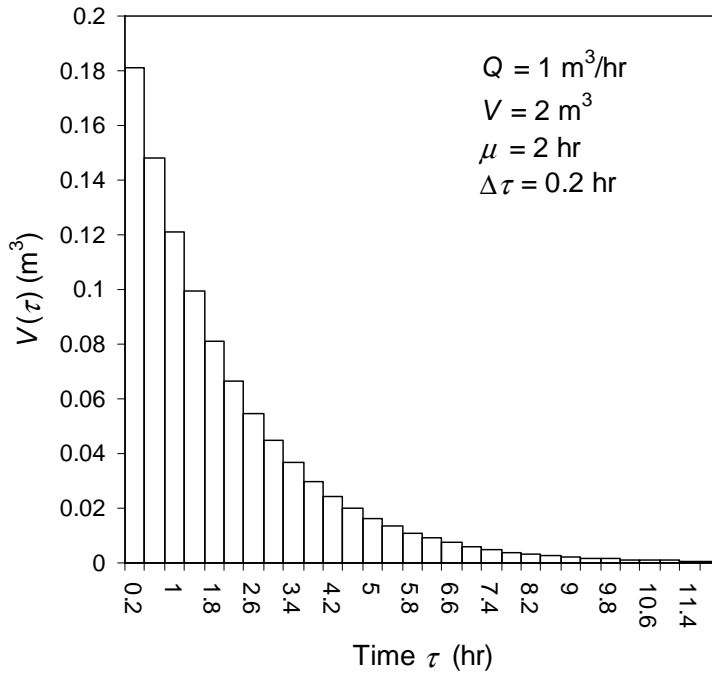
$$\frac{d(cV)}{dt} = (cQ)_{out} - (cQ)_{in} \quad (10)$$

where  $V$  is the total volume of water in the reservoir,  $c$  is the particle concentration, and  $Q_{in}$  and  $Q_{out}$  are the flow rates into and out of the reservoir, respectively. In the case of steady-state flow, which we assume from hereon, the change in volume  $V$  in (9) is zero. The volume of water that will reside within the reservoir for a time  $\tau$  before leaving, or  $V(\tau)$ , is then given by:

$$V(\tau) = Vp(\tau)d\tau \quad (11)$$

where  $p(\tau)$  is the residence time distribution of the water (see Equation (1)). An example of the equation (11) using a discrete exponential pdf is shown in Figure 7.

Figure 7: Water volume  $V(\tau)$  within a residence time interval  $\Delta\tau$  against residence time  $\tau$  for a given reservoir volume  $V$  and flow rate  $Q$ .



We assume that particles entering the reservoir that do not settle, would share the same residence time with the water, such that:

$$M_{in}(\tau) = c_{in}V(\tau) \tag{12}$$

where  $c_{in}$  is the constant particle concentration of the water entering the reservoir and  $M_{in}(\tau)$  is the mass of particles of residence time  $\tau$ . However, if particles are allowed to settle, the outgoing mass of particles that resides in the reservoir for time  $\tau$  will be less than the incoming mass by a fractional amount  $m(\tau)$ :

$$m(\tau) = M_{out}(\tau) / M_{in}(\tau) \tag{13}$$

where  $M_{out}(\tau)$  is the outgoing mass of particles at  $\tau$ .

We assume that particles of certain size, shape and density, sink at a constant rate  $s$ . Furthermore, assuming that the particles are uniformly vertically distributed upon entering the reservoir, the fractional particle mass that exits the reservoir is given by:

$$m(\tau) = \begin{cases} 1 - \frac{s\tau}{h} & s\tau < h \\ 0 & s\tau \geq h \end{cases} \tag{14}$$

where  $h$  is the height of the water in the reservoir. The total mass of particles leaving the reservoir,  $M_{out}$ , is the integration of outgoing particles over all residence times:

$$M_{out} = \int_{\tau} M_{out}(\tau) d\tau \quad (15)$$

Combining (11), (12), (13) and (15) yields:

$$c_{out} = c_{in} \int_{\tau} m(\tau) p(\tau) d\tau \quad (16)$$

where  $c_{out}$  is the outgoing particle concentration, or  $M_{out} / V$ . The outflow concentration  $c_{out}$  is delivered to the next reservoir in the series as the inflow concentration  $c_{in}$ . The integral in (16) with (14) is solved numerically.

### 2.2.2.3 Model parameterization

The parameters for the water residence time model are reservoir volume  $V$  and mean residence time  $\mu$ . The particle settling model requires both  $V$  and  $\mu$ , but also uses reservoir water level  $h$ , particle sink rate  $s$ , and particle inflow concentration  $c_{in}$ . Reservoir volumes and approximate operational water levels were acquired from system designs or on-site measurements. These are given in Tables 5 through 8 for each of the four out-of-water treatment systems under study. Each reservoir is a separate component of the treatment system (for example, for Orams Marine: collection sump = reservoir 1, settlement tank no. 1 = reservoir 2, settlement tank no. 2 = reservoir 3, and sand filter = reservoir 4).

The mean water residence time  $\mu$  is  $V/Q$ . Because direct measurements of flow rates through the system are not available, the flow rate  $Q$  was calculated from estimates of the total water volume used in hull cleaning and typical times spent cleaning (Table 9). The mean residence times are given in Tables 5 through 8. Note: flow rates as they appear for each facility in Tables 5 through 8 appear different than those in Table 9 due to model derivation, but do represent actual episodic flow events. As the typical flow rates are coarse estimates, the models were also run using twice and half these flow rates. For the calculation of mean water residence at the Tauranga Marina, treated water was assumed to be discharged from the system even though it is predominantly recycled for water blasting to allow for any excess wasting. Statistical comparison of modelled residence time of water for facilities where seasonal differences in the number of vessels are cleaned (Orams Marine, Westpark Marina and Tauranga Marina) was conducted with Student's  $t$ -test ( $P = 0.05$ ).

**Table 5: Model parameters for Lyttelton dry dock**

Reservoir	Height $h$ (m)	Horizontal Area $A$ (m <sup>2</sup> )	Volume $V$ (m <sup>3</sup> )	Mean residence time $\mu$ (hr)	
				Summer*	Winter*
1	1.0	1.8	1.8	0.8	0.8
2	1.0	3.8	3.8	1.7	1.7
3	1.0	1.8	1.8	0.8	0.8
4	1.0	3.8	3.8	1.7	1.7
5	1.0	3.8	3.8	1.7	1.7
6	2.0	0.5	1.0	0.4	0.4
7	1.0	1.0	1.0	0.4	0.4
<b>Total</b>			<b>16.7</b>	<b>7.4</b>	<b>7.4</b>

\*For flow rate of 38 L min<sup>-1</sup> (model derivation)

**Table 6: Model parameters for Orams Marine**

Reservoir	Height $h$ (m)	Horizontal Area $A$ (m <sup>2</sup> )	Volume $V$ (m <sup>3</sup> )	Mean residence time $\mu$ (hr)	
				Summer*	Winter**
1	0.6	0.6	0.4	10	20
2	1.5	9.6	14.4	406	798
3	1.5	9.6	14.4	406	798
4	1.5	11.2	16.8	473	929
<b>Total</b>			<b>46.0</b>	<b>1296</b>	<b>2544</b>

\*For flow rate of 0.6 L min<sup>-1</sup> (model derivation)

\*\*For flow rate of 0.3 L min<sup>-1</sup> (model derivation)



**Table 7: Model parameters for Westpark Marina**

Reservoir	Height $h$ (m)	Horizontal Area $A$ (m <sup>2</sup> )	Volume $V$ (m <sup>3</sup> )	Mean residence time $\mu$ (hr)	
				Summer*	Winter**
1	2.0	0.4	0.7	3	7
2	3.0	1.6	4.7	22	50
3	3.0	1.6	4.7	22	50
4	3.0	1.6	4.7	22	50
<b>Total</b>			<b>14.7</b>	<b>71</b>	<b>156</b>

\*For flow rate of 3.5 L min<sup>-1</sup> (model derivation)

\*\*For flow rate of 1.6 L min<sup>-1</sup> (model derivation)

**Table 8: Model parameters for Tauranga Marina**

Reservoir	Height $h$ (m)	Horizontal Area $A$ (m <sup>2</sup> )	Volume $V$ (m <sup>3</sup> )	Mean residence time $\mu$ (hr)	
				Summer*	Winter**
1	1.5	0.3	0.4	4	9
2	1.0	1.0	1.0	11	21
3	1.0	2.0	2.0	21	42
4	1.0	2.0	2.0	21	42
5	1.0	14.0	14.0	149	297
6	2.8	4.9	13.7	147	291
7	3.5	9.6	33.7	359	714
<b>Total</b>			<b>66.8</b>	<b>713</b>	<b>1416</b>

\*For flow rate of 1.6 L min<sup>-1</sup> (model derivation)

\*\*For flow rate of 0.8 L min<sup>-1</sup> (model derivation)

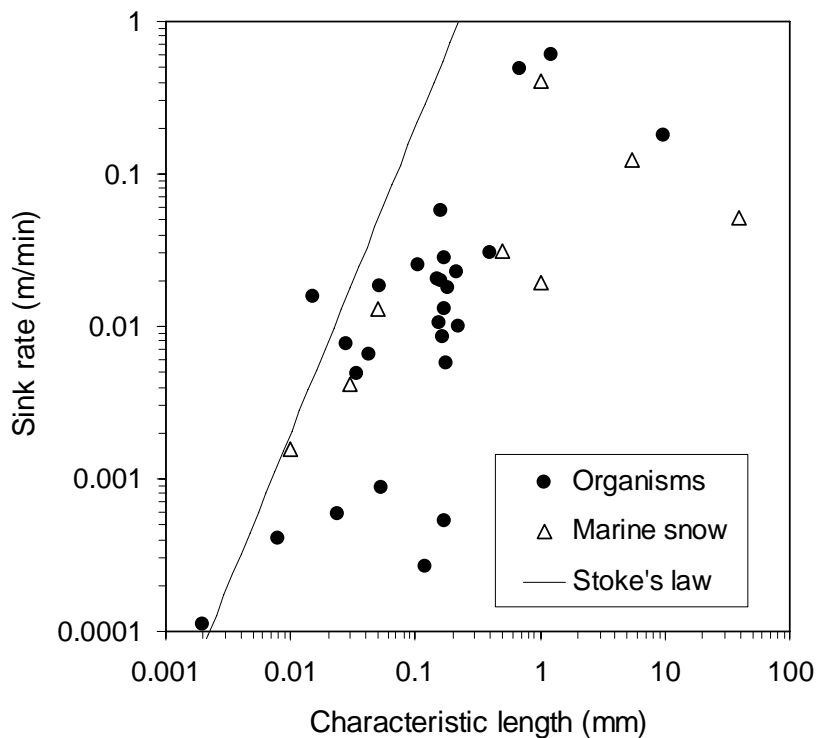
Because the types of organisms that may be present in the systems vary greatly, we ran the model using a wide range of sink rates. Based on existing data (see Figure 7), the sink rate  $s$  was varied from 0.0001 to 1 m min<sup>-1</sup>. As a reference, Stokes' law is widely used as a predictor of sinking rate and can be modified to take into account particle characteristics such as shape. Stokes' law for spherical objects is widely used where particular organism/particle shapes/densities are unqualified and is calculated as follows:

$$V_s = \frac{2grn^2(p-p_0)}{9\mu}$$

where  $V_s$  = sinking velocity (cm sec<sup>-1</sup>),  $g$  = acceleration due to gravity (980.7 cm

sec<sup>-2</sup>),  $\mu$  = viscosity of medium (poise),  $r$  = radius of particle (cm),  $p$  = density of particle (g cm<sup>-3</sup>) and  $p_0$  = density of medium (g cm<sup>-3</sup>). In Figure 8, Stokes' law is shown for a density of 1.2, which was the average specific density given for the limited number of organisms in Table 3 for which organism densities were provided.

Figure 8: Measured sink rates for various species and marine snow against characteristic length (from various sources, Table 3). Data show adults, larvae and eggs. Also shown is Stokes' law for an organism density of 1.2.



Measured inflow particle concentrations of different sink rates were not available. However, because sink rates are assumed to be independent of concentration, the choice of  $c_{in}$  has no effect on the modelled outflow concentration. For this reason,  $c_{in}$  was set to 1 for all runs, and the modelled outflow concentrations are reported as relative concentrations, that is,  $c_{out}/c_{in}$ .

Though some reservoirs within a treatment system are specifically settling tanks, others have different functions. For example, some may be filled with sand to serve as filters, while others may contain flocculating agents to promote the precipitation of particles. The purpose of these different treatments is to decrease the amount of undesirable particles leaving the system; in effect, to decrease  $m(\tau)$  in (13). As we do not consider these different functions, it might be expected that the estimates of outflow concentration given by the model will be higher than actual concentrations.

**Table 9: Numbers of vessels cleaned per hull cleaning facility and related liquid effluent treatment system statistics.**

Facility	No. vessels cleaned in summer d <sup>-1</sup>	No. vessels cleaned in winter d <sup>-1</sup>	Estimated water vol. used vessel <sup>-1</sup> (m <sup>3</sup> )	Vol. of treatment system (m <sup>3</sup> )	Typical flow rate into treatment system (L min <sup>-1</sup> )	Water vol. used d <sup>-1</sup> (m <sup>3</sup> ) in summer	Water vol. used d <sup>-1</sup> (m <sup>3</sup> ) in winter
Lyttelton dry dock	0.18	0.18	300	16.7	320	54	54
Orams Marine	2.5	1.25	0.34	46	11.4	0.85	0.38
Westpark Marina	5	2.5	0.90	14.7	30	4.5	2.25
Tauranga Marina	5	2.5	0.45	66.8	30	2.25	1.13

#### 2.2.2.4 Freshwater tolerance

For many potential marine NIS pest species there is inadequate or no information available on their minimum salinity range, tolerance to freshwater and duration for which they can tolerate freshwater exposure before severe damage or death occurs. In this study, representative organisms for which information on freshwater tolerance exists are organisms which are listed on the New Zealand register of unwanted organisms under the Biosecurity Act 1993 and the Australian Ballast Water Management Advisory Council's (ABWMAC) schedule of non-indigenous pest species (Table 10). It was decided to include some organisms on the latter schedule normally regarded as ballast water-transported, such as toxic dinoflagellates, as there is potential for them to be contained within interstices and sedimented habitat (Taylor and MacKenzie 2001) where extensive hull-fouling occurs. In addition, the freshwater tolerances of some marine organisms listed by Hayes et al. (2005) as being of medium-low to high priority as invasive species in Australia, and some fouling organisms actually encountered on vessel hulls during this study are also presented. Estimates of freshwater tolerances of these organisms were derived from existing literature. Freshwater tolerances must always be regarded with a degree of caution as they may be confounded by variables such as abruptness and magnitude of salinity change, initial condition of the organism, organism genetic strain, life stage, temperature etc. (Mills and Fish 1980; Anger 1991)

**Table 10: Salinity tolerances of various marine organisms that may be regarded as non-indigenous pest species (NIS) or encountered as hull-fouling. Minimum salinity tolerances are expressed as the minimum salinity (ppt) and the number of days for which the organism is able to survive at that salinity before death or severe damage occurs. Lower normal habitat salinity levels in which animals are found are in brackets ( ).**

Phylum	Class/Order	Genus and Species	Minimum salinity tolerance (ppt/days)	Reference
Annelida	Polychaeta	<i>Sabella spallanzanii</i> <sup>1,2,3</sup>	0.0/0.1 days, (26.0)	(Currie et al. 2000; Gunthorpe et al. 2001)
Arthropoda	Amphipoda	<i>Caprella mutica</i> <sup>4</sup>	20/2 days poss., (11)	(Oakley 2005)
Arthropoda	Amphipoda	<i>Monocorophium acherusicum</i> <sup>4</sup>	1.0/10 days, (15)	(Lee et al. 2005)
Arthropoda	Decapoda	<i>Carcinus maenus</i> <sup>1,2,3</sup>	1.0/2 days, (1.4)	(Cohen et al. 1995; Cieluch et al. 2004)
Arthropoda	Decapoda	<i>Eriocheir sinensis</i> <sup>1,3</sup>	0.0/7 days, (5.0)	(Anger 1991) (Noble, unpubl. Data)
Chordata	Ascidiacea	<i>Ciona intestinalis</i> <sup>3,4</sup>	(18)	(Jackson 2005)
Chordata	Ascidiacea	<i>Styela clava</i> <sup>1,3</sup>	0.0/1 day, (20.0)	(Lutzen 1999; Coutts and Forrest 2005)
Echinodermata	Asteroidea	<i>Asterias amurensis</i> <sup>1,2,3</sup>	0.0/0.01 days or 24/9 days, (26.0)	(McEnulty et al. 2001; NIMPIS 2002a)
Ectoprocta	Cheilostomata	<i>Bugula neritina</i> <sup>3,4</sup>	14.0, (18.0)	(Mawatari 1951)
Ectoprocta	Cheilostomata	<i>Watersipora subtorquata</i> <sup>3,4</sup>	(25.0)	(Cohen 2005)
Mollusca	Bivalvia	<i>Crassostrea gigas</i> <sup>2,3</sup>	3.0, (5.0)	(Hopkins 1936; Nell and Gibbs 1986; Chu et al. 1996)
Mollusca	Bivalvia	<i>Musculista senhousia</i> <sup>1,2,3</sup>	(6.6)	(Reusch and Williams 1998;

				Miyawaki and Sekiguchi 1999)
Mollusca	Bivalvia	<i>Perna viridis</i> <sup>3</sup>	0.0/14 days, (18.0)	(Shafee 1976; Segnini de Bravo et al. 1998)
Mollusca	Bivalvia	<i>Potamocorbula amurensis</i> <sup>1,3</sup>	<1.0/osmoconforms, (5)	(Nicolini and Penry 2000)
Phycophyta	Chlorophyta	<i>Caulerpa taxifolia</i> <sup>1</sup>	10.0/1–7 days	(NIMPIS 2002b; Creese et al. 2004)
Phycophyta	Chlorophyta	<i>Codium fragile</i> ssp <i>tomentosoides</i> <sup>3</sup>	12.5, (17.5)	(Trowbridge 1999)
Phycophyta	Chlorophyta	<i>Enteromorpha intestinalis</i> <sup>4</sup>	0.0/5 days, (5.0)	(Martins et al. 1999; Kamer and Fong 2000)
Phycophyta	Phaeophyceae	<i>Undaria pinnatifida</i> <sup>1,3</sup>	0.0/1 day, (20.0)	(Hayakawa 1987; Wallentius 1999; McEnnulty et al. 2001)
Phycophyta	Dinophyceae	<i>Alexandrium catenella</i> <sup>2</sup>	15.0/14 days	(Siu et al. 1997)
Phycophyta	Dinophyceae	<i>Alexandrium minutum</i> <sup>2,3</sup>	3.0	(Su et al. 1993)
Phycophyta	Dinophyceae	<i>Alexandrium tamarense</i> <sup>2</sup>	7.0	(Prakash 1967; Su et al. 1993)
Phycophyta	Dinophyceae	<i>Gymnodinium catenatum</i> <sup>2,3</sup>	10.0/2 days	(Band-Schmidt et al. 2004)

1 Organism is listed on the New Zealand Register of Unwanted Organisms under the Biosecurity Act 1993.

2 Organism is listed on the Australian Ballast Water Advisory Council's (ABWMAC) schedule of non-indigenous pest species.

3 Listed by Hayes et al. (2005) as being of medium-low to high priority as invasive marine species in Australia

4 Organisms encountered during ZBS2005-22 (summer).

## 3.0 Results

### 3.1 Specific objective 1

The 37 vessels compared during this study and previous study ZBS2002-04 (winter and summer sampling) included private sailing and motor yachts (31), fishing vessels (4), a tanker and a harbour tug ranging from 3.4–104.5 m in length (Table 11a, b). Fouling cover on vessel hulls ranged from  $2.8 \pm 1.2\%$  to  $67.5 \pm 9.2\%$  (mean  $\pm$  1 s.e.) of submerged hull surfaces, and from 0.1 kg to  $9.7 \text{ kg m}^{-2}$  in wet weight (Table 11a, b; Figure 9a, b). Fouling cover was generally higher on vessels sampled during summer ( $30.9 \pm 4.0\%$ ) than during winter ( $11.27 \pm 2.7\%$ ) (ANOVA: Season effect,  $P = 0.002$ ; Table 12, Figure 9b). Higher abundance of tubicolous amphipods and encrusting bryozoans in summer accounted for 43 percent of the multivariate dissimilarity in fouling assemblages between seasons (SIMPER). The average wetted surface area of vessel hulls cleaned in the dry dock ( $1027.8 \pm 259.14 \text{ m}^2$ ) was greater than of those cleaned in haul-out facilities ( $35.1 \pm 3.4 \text{ m}^2$ ). Consequently, the cleaning process took significantly longer for vessels in the dry dock, during both winter and summer sampling (dry dock:  $41.4 \pm 27.3 \text{ h}$ ; haul-out facilities:  $0.6 \pm 0.08 \text{ h}$ ; ANOVA: Operation effect,  $P = 0.007$ ; SNK pairwise comparisons,  $P < 0.05$ ; Table 13, Figure 9c).

**Figure 9:** Estimated biomass (a) and percent fouling cover (b) on vessels, cleaning duration (c) and exposure time (d) in winter (black bars) and summer (grey bars) at different hull cleaning facilities. DD = Lyttelton dry dock, OM = Orams Marine, TR = Tauranga Marina, and WP = Westpark Marina. Data are mean  $\pm$  1 s.e.

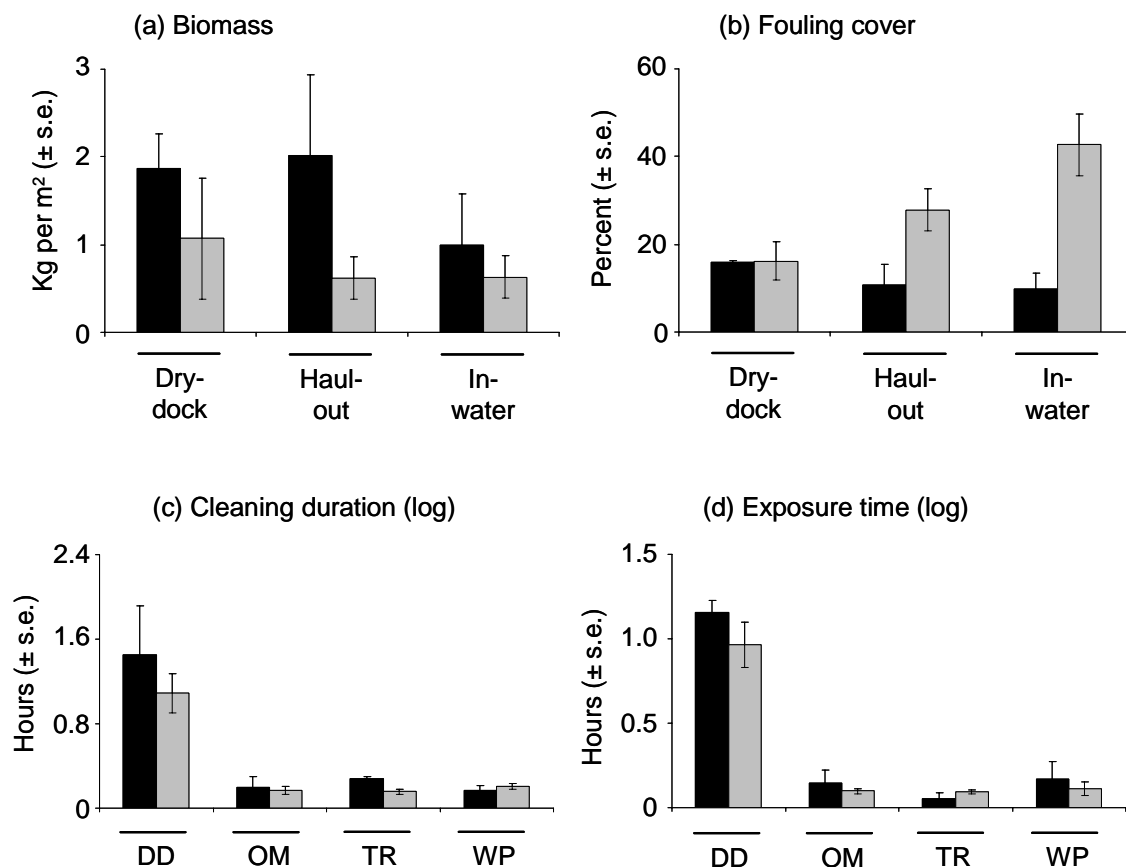


Table 11a: Vessels cleaned at different hull cleaning facilities during Research Project ZBS2002-04 (winter season).

	Operation	Facility	Season	Vessel name	Vessel type	Length	Total wetted surface area	Area fouled; fouling weight
1. Vessels removed from water for cleaning	Dry dock	Lyttelton Port	Winter	<i>Vessel 1</i>	Fishing trawler	104 m	1719.1 m <sup>2</sup>	263.2 m <sup>2</sup> ; 2.3 kg m <sup>2</sup>
			Winter	<i>Vessel 2</i>	Harbour tug	28.7 m	213.7 m <sup>2</sup>	36.1 m <sup>2</sup> ; 2.2 kg m <sup>2</sup>
			Winter	<i>Vessel 3</i>	LPG Tanker	76.4 m	715.6 m <sup>2</sup>	112.9 m <sup>2</sup> ; 1.1 kg m <sup>2</sup>
	Travel lift	Orams Marine	Winter	<i>Vessel 4</i>	Motor yacht	11.5 m	23 m <sup>2</sup>	1.2 m <sup>2</sup> ; 1.3 kg m <sup>2</sup>
			Winter	<i>Vessel 5</i>	Sailing yacht	16.2 m	36.5 m <sup>2</sup>	1.6 m <sup>2</sup> ; 0.4 kg m <sup>2</sup>
			Winter	<i>Vessel 6</i>	Sailing yacht	12.0 m	33.0 m <sup>2</sup>	15.5 m <sup>2</sup> ; 9.7 kg m <sup>2</sup>
		Westpark Marina	Winter	<i>Vessel 14</i>	Harbour tug	14.8 m	59.6 m <sup>2</sup>	17.2 m <sup>2</sup> ; 4 kg m <sup>2</sup>
			Winter	<i>Vessel 15</i>	Motor yacht	13.4 m	43.7 m <sup>2</sup>	1.6 m <sup>2</sup> ; 1.2 kg m <sup>2</sup>
			Winter	<i>Vessel 7</i>	Sailing yacht	12.5 m	50 m <sup>2</sup>	4.1 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
		Tauranga Marina	Winter	<i>Vessel 8</i>	Sailing yacht	12.2 m	18.3 m <sup>2</sup>	0.3 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
			Winter	<i>Vessel 9</i>	Launch	12.8 m	41.6 m <sup>2</sup>	1.3 m <sup>2</sup> ; 1.2 kg m <sup>2</sup>
	Winter	<i>Vessel 17</i>	Launch	12.8 m	23.8 m <sup>2</sup>	0.7 m <sup>2</sup> ; 1.3 kg m <sup>2</sup>		



			Winter	<i>Vessel 18</i>	Sailing yacht	11.9	42.8 m <sup>2</sup>	1.3 m <sup>2</sup> ; 0.6 kg m <sup>2</sup>
2. Vessels cleaned in the water	Cleaning by divers	Orams Marine	Winter	<i>Vessel 10</i>	Motor yacht	21.3 m	127.8 m <sup>2</sup>	4.8 m <sup>2</sup> ; 0.3 kg m <sup>2</sup>
			Winter	<i>Vessel 16</i>	Sailing yacht	15.2 m	48.8 m <sup>2</sup>	1.5 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
			Winter	<i>Vessel 11</i>	Motor yacht	13.7 m	71.2 m <sup>2</sup>	5.5 m <sup>2</sup> ; 0.1 kg m <sup>2</sup>
		Gulf Harbour Marina	Winter	<i>Vessel 12</i>	Sailing yacht	10 m	30 m <sup>2</sup>	1.2 m <sup>2</sup> ; 0.8 kg m <sup>2</sup>
			Winter	<i>Vessel 13</i>	Sailing yacht	10 m	30 m <sup>2</sup>	4.5 m <sup>2</sup> ; 0.8 kg m <sup>2</sup>
			Winter	<i>Vessel 19</i>	Sailing yacht	10.5 m	31.5 m <sup>2</sup>	8.1 m <sup>2</sup> ; 3.8 kg m <sup>2</sup>

**Table 11b: Vessels cleaned at different hull cleaning facilities during Research Project ZBS2005-22 (summer season).**

	Operation	Facility	Season	Vessel name	Vessel type	Length	Total wetted surface area	Area fouled; fouling weight
1. Vessels removed from water for cleaning	Dry dock	Lyttelton Port	Summer	<i>Vessel 20</i>	Fishing trawler	75.4 m	1096.8 m <sup>2</sup>	119.5 m <sup>2</sup> ; 0.4 kg m <sup>2</sup>
			Summer	<i>Vessel 21</i>	Fishing trawler	45 m	615.6 m <sup>2</sup>	78.2 m <sup>2</sup> ; 0.4 kg m <sup>2</sup>
			Summer	<i>Vessel 1</i>	Fishing trawler	104 m	1719.1 m <sup>2</sup>	429.8 m <sup>2</sup> ; 2.4 kg m <sup>2</sup>
	Travel lift	Orams Marine	Summer	<i>Vessel 22</i>	Motor yacht	14.5 m	37.7 m <sup>2</sup>	11.1 m <sup>2</sup> ; 0.4 kg m <sup>2</sup>
			Summer	<i>Vessel 23</i>	Sailing yacht	7.5 m	30 m <sup>2</sup>	12.4 m <sup>2</sup> ; 0.5 kg m <sup>2</sup>
			Summer	<i>Vessel 24</i>	Motor yacht	14.4 m	34.6 m <sup>2</sup>	9.6 m <sup>2</sup> ; 0.3 kg m <sup>2</sup>
		Westpark Marina	Summer	<i>Vessel 25</i>	Sailing yacht	12 m	38.4 m <sup>2</sup>	18.1 m <sup>2</sup> ; 2.4 kg m <sup>2</sup>
			Summer	<i>Vessel 26</i>	Sailing yacht	3.4 m	8.2 m <sup>2</sup>	2 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
			Summer	<i>Vessel 27</i>	Sailing yacht	17.8 m	71.2 m <sup>2</sup>	31.9 m <sup>2</sup> ; 0.7 kg m <sup>2</sup>
		Tauranga Marina	Summer	<i>Vessel 28</i>	Sailing yacht	10 m	30 m <sup>2</sup>	3.7 m <sup>2</sup> ; 0.9 kg m <sup>2</sup>

			Summer	<i>Vessel 29</i>	Motor yacht	10 m	20 m <sup>2</sup>	0.7 m <sup>2</sup> ; 0.1 kg m <sup>2</sup>
			Summer	<i>Vessel 30</i>	Motor yacht	12 m	24 m <sup>2</sup>	4.9 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
2. Vessels cleaned in the water	Cleaning by divers	Orams Marine	Summer	<i>Vessel 31</i>	Motor yacht	12 m	24 m <sup>2</sup>	6.2 m <sup>2</sup> ; 0.4 kg m <sup>2</sup>
			Summer	<i>Vessel 32</i>	Motor yacht	10 m	20 m <sup>2</sup>	6.6 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
			Summer	<i>Vessel 33</i>	Motor yacht	10 m	20 m <sup>2</sup>	7.2 m <sup>2</sup> ; 0.2 kg m <sup>2</sup>
		Gulf Harbour Marina	Summer	<i>Vessel 34</i>	Sailing yacht	15 m	60 m <sup>2</sup>	19.1 m <sup>2</sup> ; 0.4 kg m <sup>2</sup>
			Summer	<i>Vessel 35</i>	Sailing yacht	16 m	64 m <sup>2</sup>	38.1 m <sup>2</sup> ; 1 kg m <sup>2</sup>
			Summer	<i>Vessel 36</i>	Sailing yacht	15 m	60 m <sup>2</sup>	40.5 m <sup>2</sup> ; 1.7 kg m <sup>2</sup>

**Table 12: ANOVA on percentage cover of fouling organisms on vessel hulls. Data were log(x+1) transformed.**

	DF	SS	MS	F	P
Season <sup>1</sup>	1	1.193	1.193	11.750	<b>0.002</b>
Operation <sup>2</sup>	2	0.187	0.094	0.923	0.408
S x O	2	0.513	0.256	2.527	0.096
Residual	31	3.146			

1 Winter and summer surveys.

2 Dry dock, haul-out and in-water cleaning operations.

**Table 13: ANOVA on duration of hull cleaning at each facility. Data were log(x+1) transformed.**

	DF	SS	MS	F	P
Season <sup>1</sup>	1	0.089	0.089	1.834	0.268
Operation <sup>2</sup>	3	5.276	1.759	36.488	<b>0.007<sup>3</sup></b>
S x O	3	0.145	0.048	0.518	0.675
Residual	17	1.580	0.093		

1 Winter and summer surveys.

2 Lyttelton dry dock, Orams haul-out, Westpark haul-out and Tauranga haul-out operations.

3 Student Newman Keuls (SNK) post-hoc pairwise comparisons indicate that cleaning duration in the dry dock is significantly longer than in any of the haul-out facilities, with no differences between haul-out facilities.

### 3.1.1 Solid fouling material

A total of 19 387 organisms or fragments of solid fouling material were examined during the two projects (winter: 10 317; summer: 9070). These included species of barnacles, bivalves, bryozoans, ascidians, hydroids, polychaetes, sponges, algae, motile crustaceans and molluscs, flatworms, nemertean worms, anemones and fish (Table 14). For both winter and summer, the numerically most abundant taxa were tubicolous polychaetes (serpulids, sabellids and spirorbids), barnacles (goose and acorn barnacles) and bryozoans (encrusting and erect). The representation of some taxa varied between seasons. For example, barnacles comprised 24.2 percent of specimens encountered during winter sampling but only 9.9 percent during summer sampling. In contrast, motile crustaceans (amphipods, isopods, ostracods, and tanaid shrimp) comprised 1 percent and 16.6 percent of specimens examined during winter and summer, respectively. Similar variation occurred for a range of other taxa (Table 14).

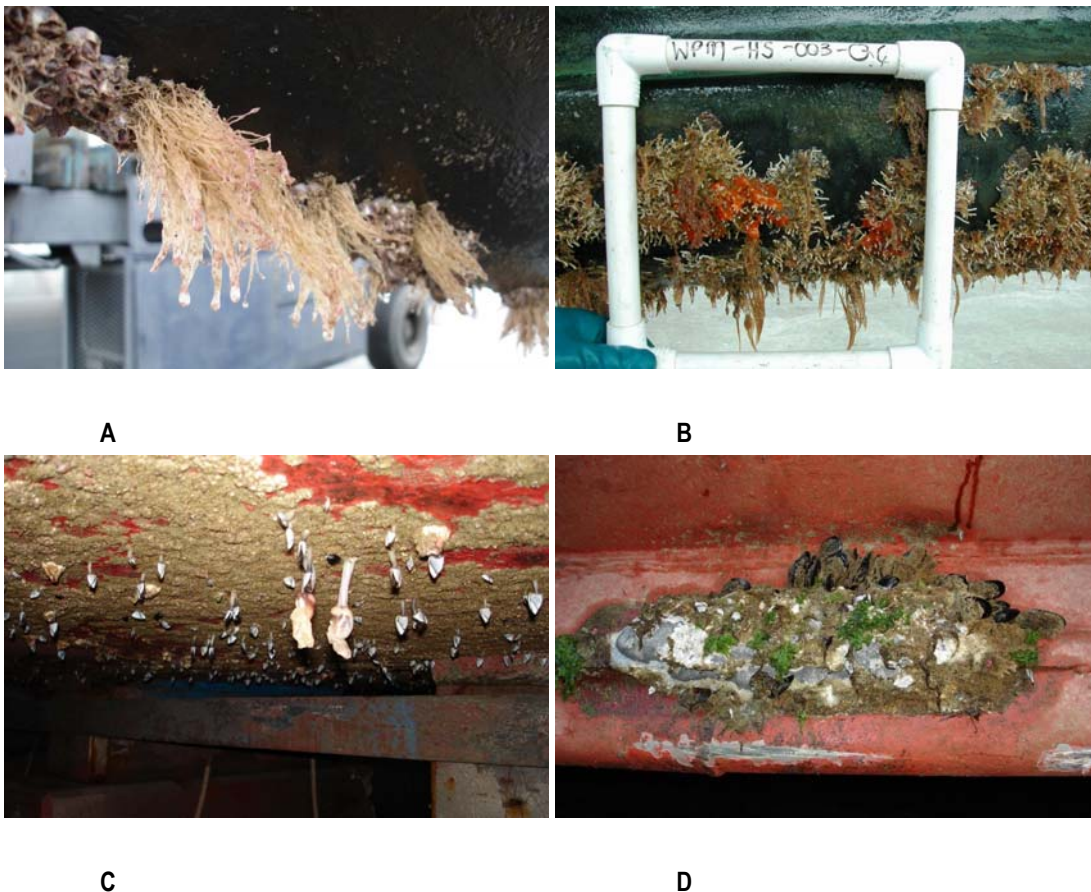
Table 14: Number of organisms or fragments of solid fouling material examined during the two hull cleaning projects (winter: 10 317; summer: 9070) in each facility.

Facility	Season	Algae	Anenomes	Ascidians	Barnacles	Bivalves	Bryozoans	Crustaceans (motile)	Fish	Flatworms/Nemertea ns	Hydroids	Molluscs (motile)	Polychaetes (errant)	Polychaetes (tubicolous)	Sponge	Total
Lyttelton dry dock	Winter	32		21	209	174	28	2			9		11	4	10	500
	Summer	43	204	22	89	529	34	1276		1	12	3	10	37	1	2261
Orams Marine	Winter	33		118	4	32	405	51		122		5	6	1332	14	2122
	Summer	1		30	29	9	985	31		1	11	1	3	1936		3039
Westpark Marina	Winter	19		50	154	35	254	1					17	79		609
	Summer			146	26	49	73	4			45		2	1037	1	1383
Tauranga Marina	Winter	2	3	72	1877	82	297	39			1		1	1814	5	4193
	Summer			8	841	2	20	2	1		131	1	2			1008
Gulf Harbour Marina	Winter		34	353	254	45	92	14				5	32	2055	12	2896
	Summer	2	8	52	7	263	263	288		2	1	2	2	751	1	1379
Total		132	249	872	3490	1220	2451	1708	1	126	210	17	86	9045	44	19 387

### 3.1.1.1 Degree of desiccation and types of damage to organisms examined

Because of the relatively short period between the removal of vessels from the water and the onset of cleaning in haul-out facilities ( $28 \pm 6$  min), most fouling organisms were still wet when fouling cover was examined (Figure 10a, b). In the Lyttelton dry dock, the time between removal from water and cleaning was significantly longer ( $11.3 \text{ h} \pm 1.85 \text{ h}$ ; ANOVA: Operation effect,  $P = 0.002$ ; SNK pairwise comparisons  $P < 0.05$ ; Table 15; Figure 9d). This meant that most soft-bodied organisms (especially ascidians, sponges and hydroids) had considerably dried out by the time water blasting commenced, whilst hard-bodied organisms such as barnacles and mussels had started to desiccate and exhibit shell-gape (Figure 10c, d). When the solid waste samples were collected all material from haul-out and dry docking facilities was re-hydrated and moist or wet from the freshwater used in the cleaning process.

Figure 10: Examples of still-wet fouling organisms at haul-out facilities (A and B) and desiccating organisms at Lyttelton dry dock (C and D).



**Table 15: ANOVA on duration of exposure to air before cleaning (that is, time from removal from water to beginning of water blasting). Data were log(x+1) transformed.**

	DF	SS	MS	F	P
Season <sup>1</sup>	1	0.025	0.025	1.781	0.274
Operation <sup>2</sup>	3	4.117	1.372	96.743	<b>0.002</b> <sup>3</sup>
S x O	3	0.043	0.014	0.759	0.533
Residual	17	0.318	0.019		

<sup>1</sup> Winter and summer surveys

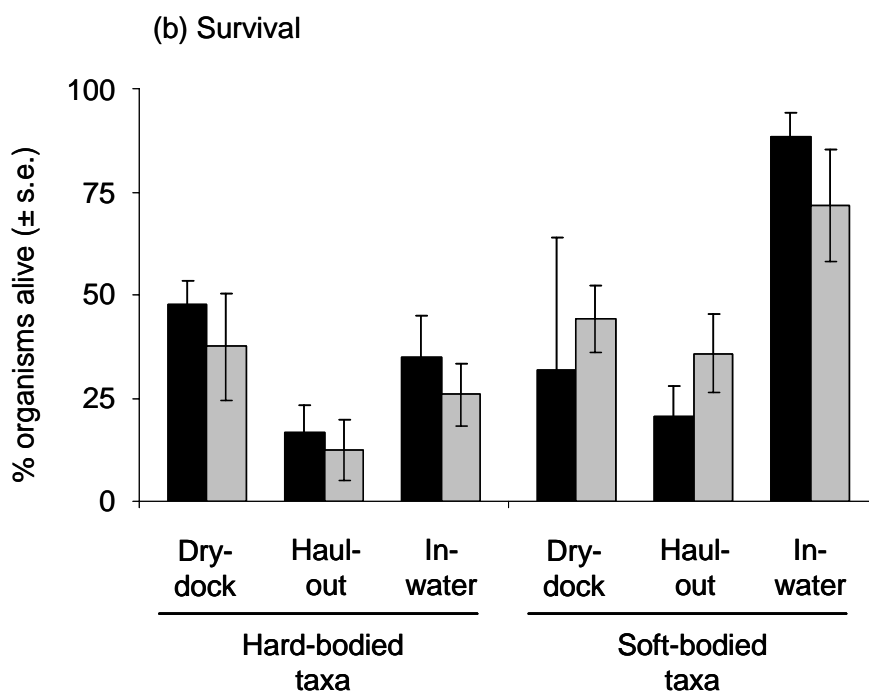
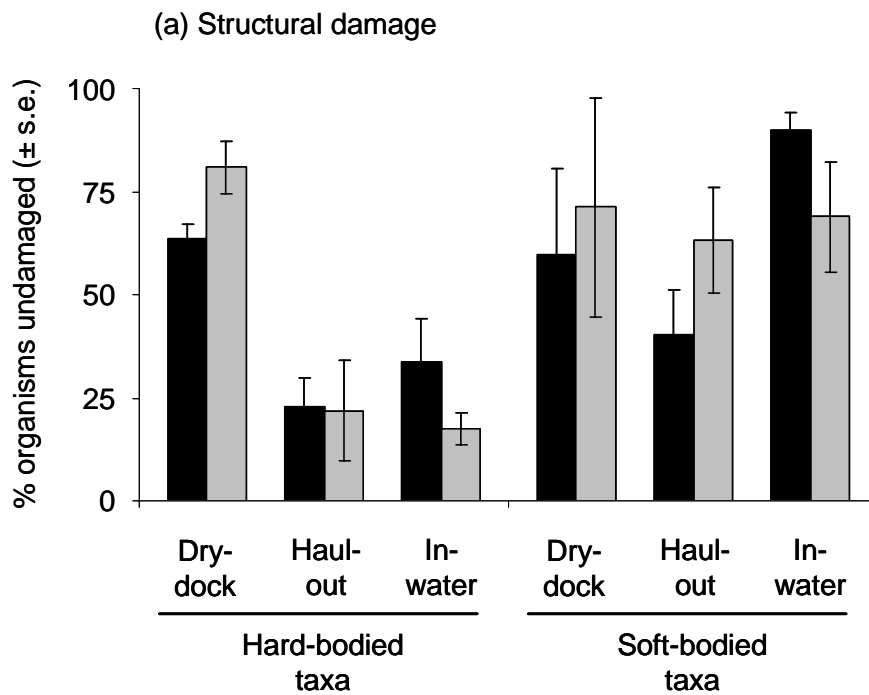
<sup>2</sup> Lyttelton dry dock, Orams haul-out, Westpark haul-out and Tauranga haul-out operations.

<sup>3</sup> Student Newman Keuls (SNK) post-hoc pairwise comparisons indicate that exposure period in the dry dock is significantly longer than in any of the haul-out facilities, with no differences between haul-out facilities.

The type and severity of physical damage to organisms removed from the hulls varied among vessels and operations. In haul-out and dry dock operations, the pressure associated with the water blasting and trampling by cleaning staff had fragmented and crushed a large proportion of soft-bodied organisms such as sponges, ascidians, flatworms and nudibranchs (percent undamaged in haul-out operations: 40 percent (winter), 63 percent (summer); percent undamaged in dry dock: 60 percent and 71 percent) and fragile or brittle hard-bodied organisms such as tubeworms and barnacles (percent undamaged in haul-out operations: 23 percent and 22 percent; percent undamaged in dry dock: 64 percent and 81 percent; Figure 11a). In-water removal of organisms from vessel hulls with a paint scraper or a soft cloth caused similar damage to hard-bodied taxa (34 percent (winter) and 17 percent (summer) undamaged), but considerably less to soft-bodied taxa (89 percent and 69 percent undamaged; Figure 11a).

Following prolonged exposure to air or high-pressure blasting with freshwater, patterns of mortality varied among operations. During both winter and summer sampling, survival of soft-bodied organisms tended to be lower in haul-out and dry dock operations (20.5 percent to 44 percent) than following in-water cleaning (72 percent to 88 percent, Figure 11b). Rates of survival of hard-bodied organisms were generally low in all hull cleaning methods during both sampling seasons and ranged from 12.5 percent (haul-out operations) to 47.6 percent (dry dock) (Figure 11b).

Figure 11: Percentage of hard-bodied and soft-bodied organisms undamaged (a) and alive (b) according to type of hull cleaning facility in winter (black bars) and summer (grey bars). Data are mean  $\pm$  1 s.e.





### 3.1.1.2 Viability of organisms

The proportion of organisms that remained viable following removal from vessel hulls varied considerably among broad taxonomic groups, cleaning operations and sampling season. In all three types of operations (dry dock, haul-out and in-water cleaning), 18–54.8 percent of the total number of organisms and fragments examined were viable, with no significant differences among seasons, operation type or their interaction (ANOVA, Season, Operation and Season x Operation effects,  $P > 0.05$ ; Table 16; Figure 12). However, tubicolous polychaetes comprised 71.2 percent (winter) and 76.4 percent (summer) of all organisms investigated in in-water cleaning operations, but only 0.8–38.7 percent of those examined in dry dock and haul-out operations, respectively. When tubicolous polychaetes were excluded from the data, the mean proportion of organisms that was alive and viable following in-water hull cleaning increased to  $72.3 \pm 8\%$  (winter) and  $66.2 \pm 5.1\%$  (summer) (Figure 12b). This exclusion resulted in a significant difference in the viability of organisms removed from vessel hulls between operation types (ANOVA, Operation effect,  $P = 0.001$ , Table 17), with viability being significantly higher for in-water cleaning operations than for dry dock and haul-out (SNK pairwise comparisons,  $P < 0.05$ ; Figure 12). There was no significant difference in viability between winter and summer sampling or the interaction of sampling season and operation type.

**Table 16: ANOVA on viability of fouling organisms sampled: Total Organisms (all groups pooled).**

	DF	SS	MS	F	P
Season <sup>1</sup>	1	1.210	1.210	0.710	0.405
Operation <sup>2</sup>	2	10.67	5.340	3.150	0.057
S x O	2	0.288	0.140	0.085	0.909
Residual	31	52.570	1.690		

<sup>1</sup> Winter and summer surveys.

<sup>2</sup> Dry dock, haul-out and in-water cleaning operations.

**Table 17: ANOVA on viability of fouling organisms sampled: Total organisms excluding tubicolous polychaetes.**

	DF	SS	MS	F	P
Season <sup>1</sup>	1	1.100	1.100	0.750	0.392
Operation <sup>2</sup>	2	25.700	12.880	8.820	0.001 <sup>3</sup>
S x O	2	0.710	0.350	0.240	0.787
Residual	31	45.280	1.460		

<sup>1</sup> Winter and summer surveys.

<sup>2</sup> Dry dock, haul-out and in-water cleaning operations.

<sup>3</sup> Student Newman Keuls (SNK) post-hoc pairwise comparisons indicate that viability following in-water cleaning is significantly greater than following cleaning in haul-out or dry dock facilities.

The proportion of viable organisms encountered in the various taxonomic groups examined within one hour after collection during winter and summer and in the various operation types varied considerably. We previously reported (final report for ZBS2002-04) that, during winter, much

greater average proportions of bivalves, ascidians, bryozoans, errant polychaetes and sponges remained viable after in-water hull cleaning (58.4–100 percent of specimens examined) than after cleaning in dry dock or haul-out operations (0–46 percent of specimens examined; Figure 12c, d, e, h and i). In addition, very large proportions (93–100 percent) of motile molluscs, nemerteans and flatworms – taxa that had not been encountered in dry dock and haul-out operations - were viable following in-water removal (Figure 12k, l). During summer sampling, this pattern of viability did hold for errant polychaetes, sponges, motile molluscs, flatworms and nemerteans (Figure 12h, i, k and l), but contrasting results were obtained for other taxa. For example, the mean viability of bivalves sampled in the Lyttelton dry dock during summer was  $79.7 \pm 2\%$ , compared to  $22.1 \pm 19.3\%$  during winter (Figure 12c). A similar trend was observed for bryozoans and ascidians. Viability of bryozoans examined in the Lyttelton dry dock during summer was  $58.3 \pm 20.9\%$  higher compared to that observed during winter (Figure 12d). Similarly, viability of ascidians during summer was  $58 \pm 20.9\%$  (dry dock) and  $36.1 \pm 13\%$  (haul-out facilities) higher than during winter (Figure 12e).

Figure 12: Percentage of viable fouling organisms by taxonomic group according to type of hull cleaning facility in winter (black bars) and summer (grey bars). Data are mean  $\pm$  1 s.e.

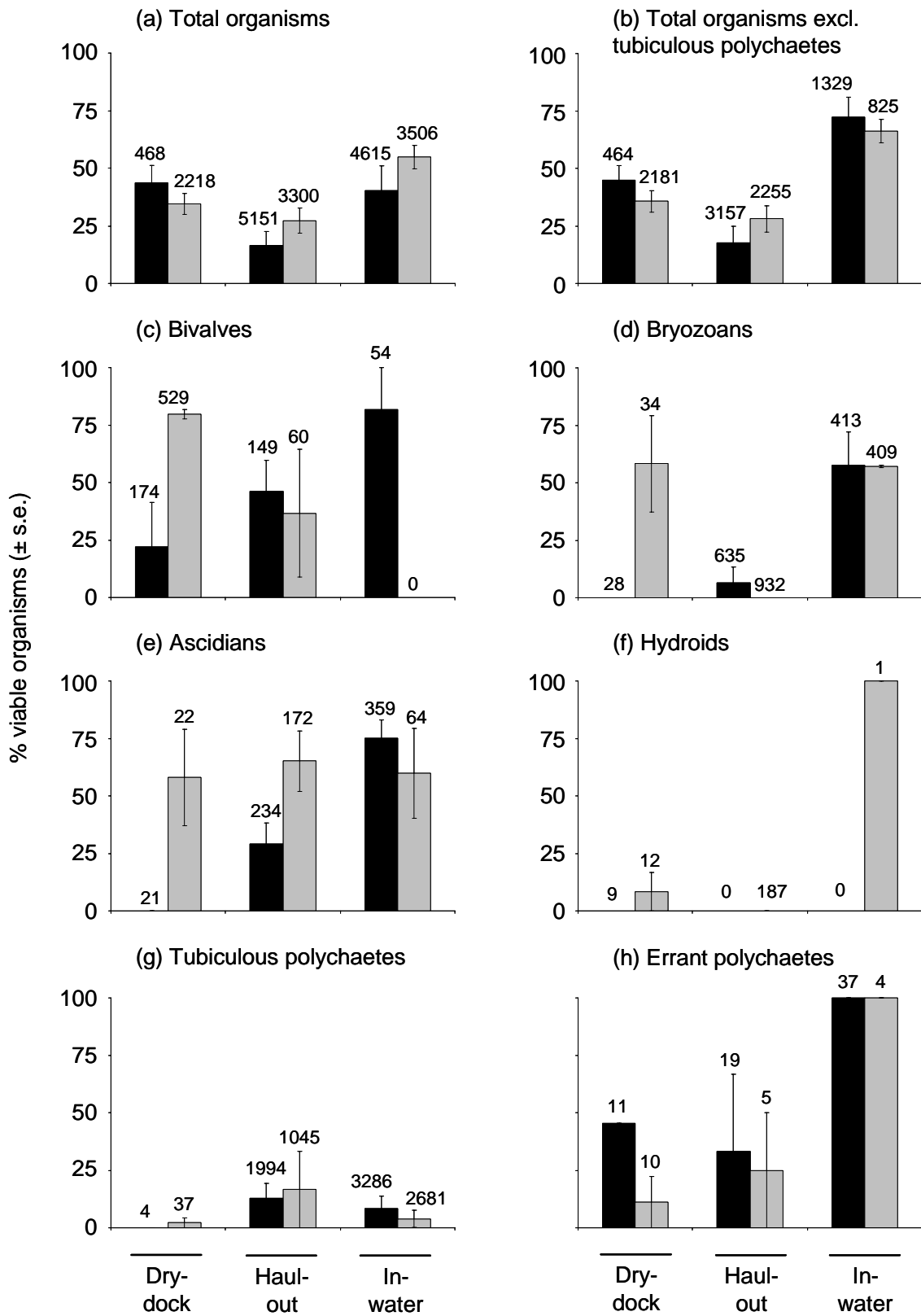
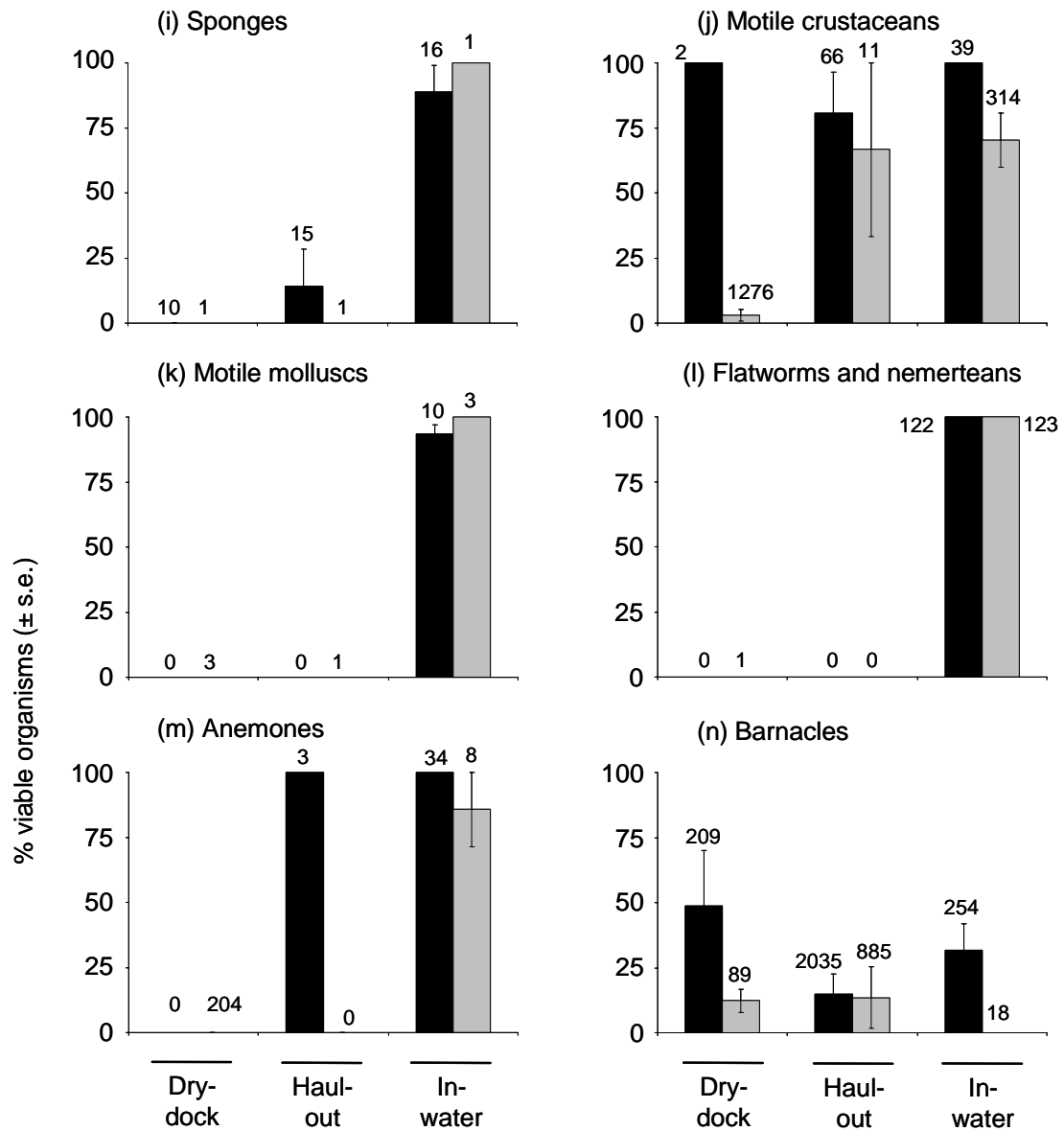


Figure 12 (continued): Percentage of viable fouling organisms by taxonomic group according to type of hull cleaning facility in winter (black bars) and summer (grey bars). Data are mean  $\pm$  1 s.e.



Considerable variability in the rates of survival of different organisms occurred associated with: 1) variation in fouling cover on individual vessels and 2) variation in the cleaning process. For any of the cleaning methods (water blasting and in-water removal using scrapers or cloths), survival and viability of fouling organisms was generally not a function of the amount of growth (percent cover) on the hulls (Figure 13a). This indicates that high levels of fouling did not protect individual organisms from being destroyed by the cleaning process. For either cleaning method, there was no apparent relationship between rates of viability and total amount of fouling cover for barnacles, bivalves, bryozoans, ascidians, hydroids and tubicolous polychaetes (Figure 13b, c, d, e, f, g). However, patterns observed for a range of taxa warrant discussion. For example, bryozoans (encrusting and erect colonies) did generally not survive the cleaning process in haul-out operations (water blasting), irrespective of the amount of fouling on the hulls (<1–76 percent of the submerged hull surface). Viability of bryozoans following in-water cleaning ranged from 0 to 100 percent and again was not related to total fouling cover on the hulls (Figure 13d). Interestingly, bryozoan viability was high (27–100 percent) on vessels cleaned in the Lyttelton dry dock during the summer season. Bivalve cover (mytilid mussels) on these vessels was up to 10.5 percent, and observations taken in the field suggest that bryozoans living amongst the bivalves were shielded from the water blasting and had a higher chance of survival (Figure 13d).

Survival of fragile and delicate organisms appeared affected by the destructiveness of the cleaning method. Removal of biota with a scraper or soft cloth is less destructive for non-brittle organisms than a water blaster, with correspondingly high survival of errant polychaetes, sponges, motile molluscs, flatworms, nematodes and anemones following in-water cleaning (Figure 13h, i, k, l, m). High anemone survival may have occurred because anemones were frequently encountered stuck to large barnacle tests or oyster valves. Few tubeworms living on the hull surfaces survived the cleaning process in any of the operations sampled. Of the 9047 serpulids, sabellids and spirorbids that were examined during winter and summer, 14.3 percent (winter) and 0.7 percent (summer) remained viable after cleaning (Figure 13g). In both summer and winter sampling, the most common forms of damage observed in this group were fragmentation of the tube and/or the worm inside it, and/or loss of the tentacular crown and feeding structure. In nearly all cases, the only living and viable individuals were growing epibiotically on other organisms such as barnacles and bivalves. Similarly, most barnacles growing directly on hull surfaces were also killed by cleaning, as their shell plates were detached from the basal plate and the animal inside from its test. However, in some cases – particularly *Vessel 2* and *Vessel 3* (winter) and *Vessel 1* (summer) sampled in the Lyttelton dry dock – barnacles also occurred as epibionts on bivalves or formed large clumps by growing on top of one another. When these vessels were cleaned, 61–82 percent of the barnacles examined were alive, compared with 0–28 percent survival per vessel in other operations (Figure 13b). Bivalves generally exhibited high mean rates of viability across all operation types. Their presence generally resulted in elevated viability of other taxa that lived on or amongst them and that were protected from the destructive cleaning action. For example, a large quantity of bivalves was removed from the sea chests of *Vessel 1* (Lyttelton dry dock, winter sampling), 61 percent of which remained viable. Sea chests are recesses built into the hull for ballast water intake of large ships. These recesses allow bivalves to persist in large clumps and also shield the organisms from the main force of water blasting during cleaning. The errant polychaetes and two motile crustaceans (all viable) that were collected following cleaning of this ship were encountered inside the clump of mussels removed from the sea chests. Most motile crustaceans (amphipods, isopods, ostracods, tanaids) examined in the dry dock and in haul-out and in-water cleaning operations during both winter and summer sampling were viable (Figure 13). They were generally encountered in protected micro-habitats such as empty barnacle tests or the internal cavities of sponges or solitary ascidians.

Figure 13: Percentage of viable fouling organisms by taxonomic group according to percentage of fouling cover on submerged hull surface. Unfilled symbols (winter), filled symbols (summer). Data are mean without s.e (to keep data points distinct) for individual boats in each facility and type of cleaning operation.

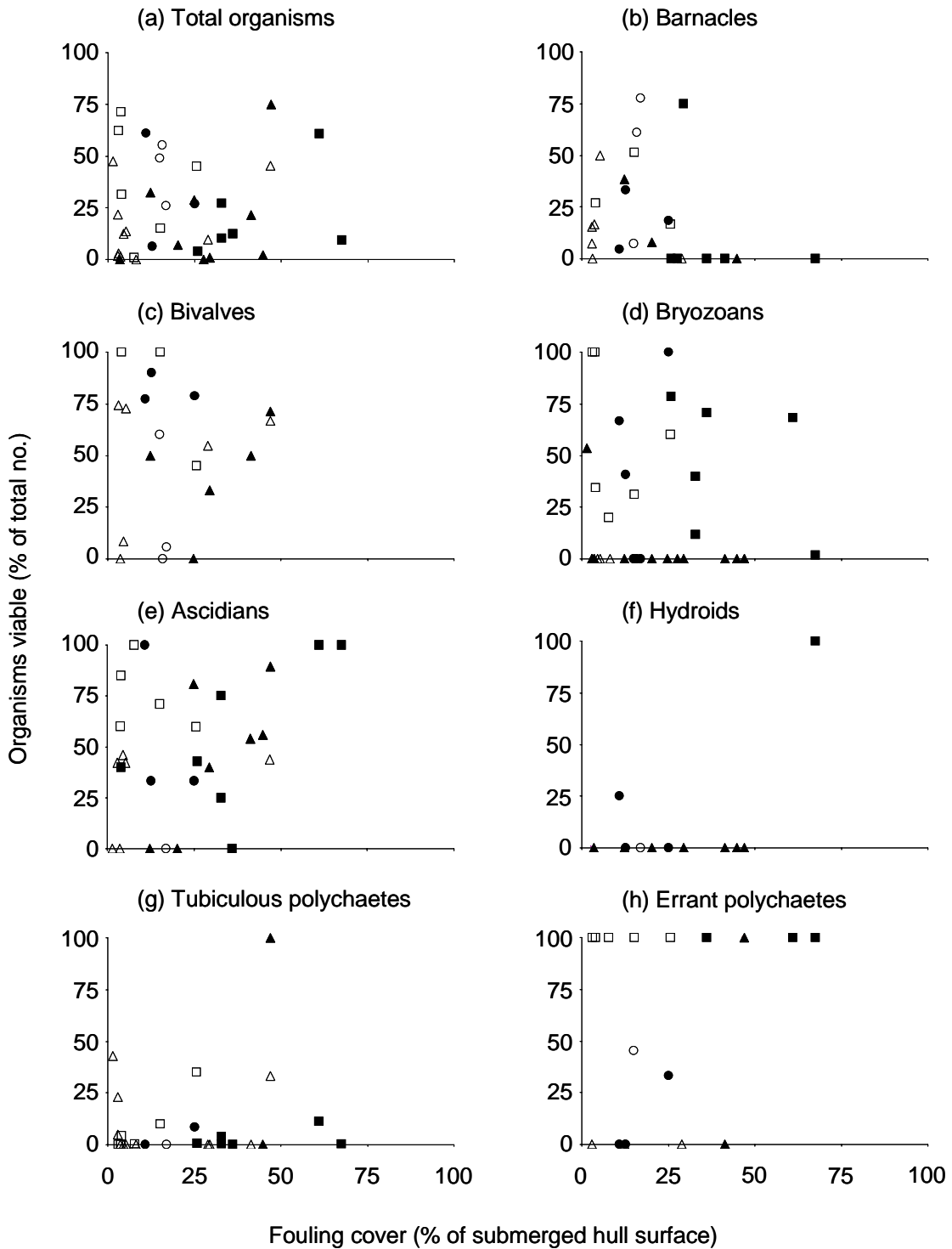
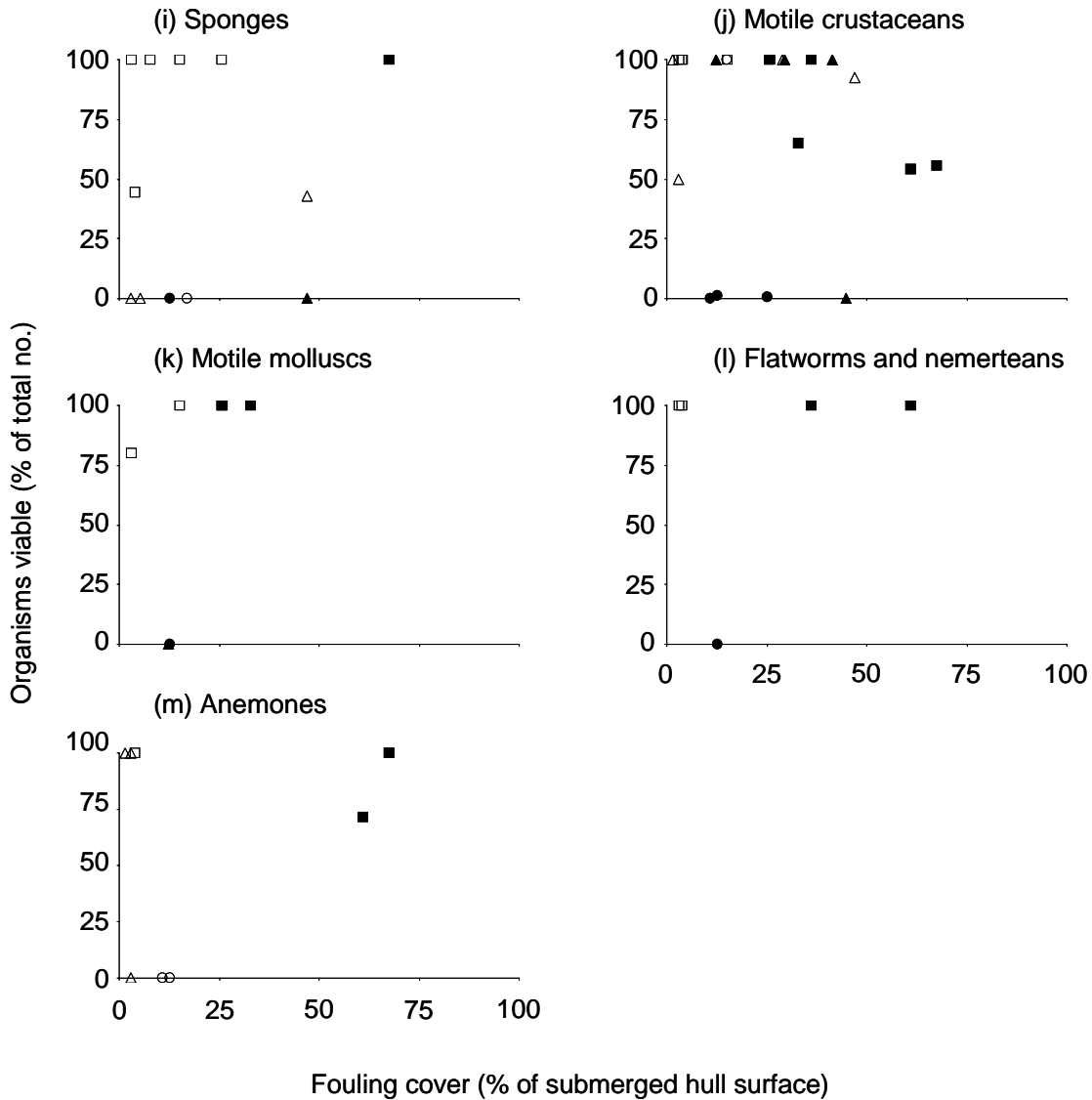


Figure 13 (continued): Percentage of viable fouling organisms by taxonomic group according to percentage of fouling cover on submerged hull surface. Unfilled symbols (winter), filled symbols (summer). Data are mean without s.e (to keep data points distinct) for individual boats in each facility and type of cleaning operation.



We were not able to reliably determine mortality or viability in marine macroalgae following their removal from vessel hulls. In total, 86 and 46 samples of algae were examined during winter and summer sampling respectively, most of which were *Enteromorpha/Ulva* sp. These algae grow as a double layer of cells, each of which, under certain conditions, could fragment from the main body of the alga and act as a colonising propagule (Adams 1994; Nelson, pers. comm. 2003). Fifty-one percent of all macroalgae collected were damaged and fragmented to varying degrees, and often faded due to a loss of pigment. More detail on this group is provided below (Sampling of liquid effluent).

### 3.1.2 Sampling of liquid effluent

#### 3.1.2.1 Abundance of organisms

A total of 223 samples of liquid effluent (3.5–10 L per sample; 64 taken in winter and 159 in summer samples) were taken in the four facilities from four different stages of treatment. These stages were: (1) the effluent from the water blast, (2) liquid from the first chamber or (3) liquid from the final chamber of the settlement tank system, and (4) liquid from the final discharge effluent (Table 3). Salinity at all stages of liquid effluent treatment was 0 to 0.5 ppt ( $\pm 0.5$  ppt). Intact specimens or fragments of nematodes, crustaceans (mainly copepods), gastropods, bivalves, rotifers, oligotrichous ciliates, diatoms, tintinnids, filamentous algae, spores, eggs and pollen (terrestrial plants) were encountered in liquid samples taken during both winter and summer. Hydroids, polychaetes, foraminiferans and dinoflagellates were encountered in summer but not in winter samples (Tables 18 and 19). In each sample, organisms were encountered at maximum concentrations of 63 100 and 259 400 organisms/10 L (winter and summer samples).



Table 18: Concentrations of organisms in the liquid waste sampled in dry dock and haul-out operations at various stages of treatment (ZBS2002-04, winter sampling). All concentrations are given as abundance/10 L (mean ± s.e.) except filamentous algae, for which a ranks scale of abundance (0-5) was used. Decimal places are removed to simplify table.

		Nematodes	Crustaceans	Gastropods	Bivalves	Rotifers	Larvae	Ciliates	Diatoms	Tintinnids	Filamentous algae	Spores	Eggs
Lyttelton dry dock	Cleaning runoff	5343 (1883)	1275 (363)	43 (43)	8 (7)	0	6 (6)	1368 (499)	770 (375)	120 (88)	5 (0)	258 (205)	295 (106)
	Final tank	521 (504)	371 (337)	0	0	0	0	0	0	0	1 (0)	253 (253)	90 (73)
	Final discharge	2 (2)	0	0	0	0	0	0	20 (12)	0	1 (0)	0	5 (3)
Orams Marine	Cleaning runoff	36 727 (6357)	873 (629)	288 (104)	0	0	78 (78)	1939 (555)	0	1540 (663)	5 (0)	0	72 (47)
	Final tank	10 (10)	0	7 (7)	0	0	0	0	0	7 (7)	1 (0)	0	11 (11)
	Final discharge	0	0	0	0	0	0	0	0	0	1 (0)	0	1 (1)
Westpark Marina	Cleaning runoff	1595 (442)	43 (43)	0	0	0	0	1034 (351)	137 (137)	5016 (3036)	5 (0)	0	135 (889)
	Final tank	106 (51)	156 (90)	12 (12)	0	0	0	52 (30)	12 (12)	0	4	0	39 (10)

										(0)			
Tauranga Marina	Cleaning runoff	8628 (35)	1475 (476)	88 (41)	1 (1)	0	57 (40)	927 (520)	0	420 (133 )	4 (0)	175 (99)	253 (77)
	Final tank	6466 (238 3)	32 (32)	426 (146)	0	0	0	2152 (101 6)	0	2119 (129 5)	3 (1)	0	834 (328)
	Final discharge	35 (17)	30 (13)	0	0	56 (32)	0	23 (5)	0	0	1 (0)	0	0

Table 19: Concentrations of organisms in the liquid waste sampled in dry dock and haul-out operations at various stages of treatment (ZBS2005-22, summer sampling). All concentrations are given as abundance/10 L (mean ± s.e.) except filamentous algae, for which a ranks scale of abundance (0-5) was used. Decimal places are removed to simplify table.

		Nematodes	Crustaceans	Gastropods	Bivalves	Rotifers	Polychaetes	Hydroids	Larvae	Ciliates	Dinoflagellates	Diatoms	Foraminifera	Tintinnids	Filamentous algae	Spores	Eggs
Lyttelton dry dock	Cleaning runoff	1668 (276)	290 (71)	1 (1)	43 (24)	0	0	13 (9)	174 (28)	11 (11)	231 (58)	701 (183)	11 (5)	4972 (2375)	0	0	25 (11)
	First tank	152 (60)	10 (5)	0	4 (2)	0	0	0	9 (5)	0	23 (11)	172 (45)	5 (4)	496 (183)	0	5 (3)	0
	Final Tank	1 (1)	0	0	0	0	0	0	1 (1)	0	1 (1)	9 (5)	0	1 (1)	0	1 (1)	2 (2)
	Final discharge	0	8 (2)	0	0	0	1 (1)	0	4 (1)	0	1 (1)	4 (2)	0	1 (1)	0	0	0
Orams Marine	Cleaning runoff	47 646 (17960)	120 (54)	7 (6)	34 (19)	0	0	42 (42)	6 (3)	14 (14)	3 (3)	0	0	1672 (881)	0	0	1 (1)
	First tank	14 608 (6948)	17 (6)	29 (25)	29 (20)	0	0	0	1 (1)	1 (1)	0	1 (1)	0	51 (45)	1 (1)	0	13 (13)
	Final tank	12 (4)	1 (1)	0	0	0	0	0	1 (1)	0	0	1 (1)	0	0	0	0	0
Westpark Marina	Cleaning runoff	15 467 (7261)	72 (25)	2 (2)	407 (223)	0	7 (5)	0	48 (25)	24 (17)	0	7 (7)	6 (6)	30 (16)	1 (0)	40 (31)	96 (36)
	First tank	54 (15)	4 (2)	0	11 (6)	0	1 (1)	0	0	1 (1)	0	0	2 (2)	1 (1)	1 (0)	3 (2)	3 (2)
	Final tank	75 (42)	9 (8)	0	2 (2)	0	1 (1)	0	0	1 (1)	0	1 (1)	1 (1)	3 (3)	1 (0)	0	4 (2)

Tauranga Marina	Cleaning runoff	462 (115)	62 (21)	0	3 (2)	0	0	2 (2)	39 (15)	0	0	1 (1)	0	0	1 (0)	0	11 (5)
	First tank	3 (1)	1 (1)	0	0	0	0	0	1 (1)	0	0	0	0	0	1 (1)	0	2 (1)
	Final tank	0	1 (1)	0	0	0	0	0	1 (1)	0	0	0	0	0	0	0	0
	Final discharge	1 (1)	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

For both sampling seasons, average concentrations of intact animals, propagules (that is, eggs, spores and larvae) and unicellular organisms were greatest in the initial runoff from the water blasting (Figure 14). One exception was the Westpark Marina, where concentrations of propagules were higher in the final settlement tank than in the water blast runoff (Table 20). This is likely to have been caused by accidental stirring up of settled material from the tank floor. During both winter and summer sampling, the concentration of intact animals in water blast runoff was 2.3 to 11.7 times higher at Orams Marine ( $43\,372 \pm 10\,940/10\text{ L}$ , mean  $\pm$  s.e. for both seasons combined) and Westpark Marina ( $12\,322 \pm 5598$ ) than at the Lyttelton dry dock ( $3790 \pm 938$ ) or the Tauranga Marina ( $5301 \pm 1923$ ) (ANOVA: significant Facility effect, SNK pairwise comparisons  $P < 0.05$ ; Table 21; Figure 14) indicating a higher abundance of multicellular fouling organisms in untreated liquid effluent. Water blast runoff at Orams Marine and the Lyttelton dry dock had a higher concentration of unicellular organisms ( $1752 \pm 527$  and  $4318 \pm 1541/10\text{ L}$ , mean  $\pm$  s.e. for both seasons combined respectively) than that at the other facilities (approx.  $500/10\text{ L}$ ) (ANOVA, Table 21). Concentrations of intact propagules in water blast runoff varied significantly between facilities and sampling seasons and ranged from 0 to 1876 propagules/10 L) (Table 21; Figure 14).

**Table 20: Reduction in abundance of animals, propagules, unicellular organisms and filamentous algae at the various stages of liquid effluent treatment. All percentage values represent reduction in abundance relative to the concentrations observed in the water blast runoff liquid.**

	Lyttelton dry dock		Orams Marine		Westpark Marina		Tauranga Marina	
	Winter	Summer	Winter	Summer	Winter	Summer	Winter	Summer
<b>(a) Animals</b>								
1st tank	-	92.0 %	-	59.1 %	89.1 %	99.5 %	42.1 %	99.1 %
Final tank	87.4 %	99.9 %	99.9 %	100 %	-	99.5 %	-	99.7 %
Final discharge	99.7 %	99.4 %	100 %	-	-	-	98.9 %	99.9 %
<b>(b) Propagules</b>								
1st tank	-	79.7 %	-	80.9 %	76.5 %	95.38 %	49.5 %	20.5 %
Final tank	39.7 %	86.7 %	84.5 %	100 %	-	+229 %	-	75.6 %
Final discharge	99.1 %	98.5 %	100 %	-	-	-	100 %	100 %
<b>(c) Unicellular org.</b>								
1st tank	-	88.3 %	-	96.0 %	96.5 %	93.9 %	60.3 %	26.1 %
Final tank	98.9 %	99.8 %	100 %	98.9 %	-	92.2 %	-	100 %
Final discharge	100 %	99.9 %	100 %	-	-	-	99.7 %	100 %
<b>(d) Filament. algae</b>								
1st tank	-	n/a	-	24.4 %	10.0 %	39.8 %	23.1 %	66.0 %
Final tank	80.0 %	n/a	80.0 %	100 %	-	9.6 %	-	100 %
Final discharge	80.0 %	n/a	85.0 %	-	-	-	61.5 %	100 %

**Table 21: ANOVA on numbers of intact animals, propagules and unicellular organisms in the liquid effluent from the water blaster, that is, before entering settlement tanks. Data were analysed untransformed since log(x+1) transformation did not remove heterogeneity of variances.**

	DF	SS	MS	F	P
<b>(a) Animals</b>					
Season <sup>1</sup>	1	1.77 x 10 <sup>8</sup>	1.77 x 10 <sup>8</sup>	0.236	0.628
Facility <sup>2</sup>	3	1.92 x 10 <sup>10</sup>	6.41 x 10 <sup>9</sup>	8.521	<b>&lt; 0.001</b>
S x F	3	1.76 x 10 <sup>9</sup>	5.86 x 10 <sup>8</sup>	0.780	0.509
Residual	71	5.34 x 10 <sup>10</sup>	7.52 x 10 <sup>8</sup>		
<b>(b) Propagules</b>					
Season <sup>1</sup>	1	890 x 10 <sup>3</sup>	890 x 10 <sup>3</sup>	9.388	<b>0.003</b>
Facility <sup>2</sup>	3	503 x 10 <sup>3</sup>	167 x 10 <sup>3</sup>	1.770	0.161
S x F	3	100 x 10 <sup>4</sup>	334 x 10 <sup>3</sup>	3.529	<b>0.019</b>
Residual	71	673 x 10 <sup>4</sup>	94 x 10 <sup>3</sup>		
<b>(c) Unicellular organisms</b>					
Season <sup>1</sup>	1	990 x 10 <sup>3</sup>	990 x 10 <sup>3</sup>	0.073	0.787
Facility <sup>2</sup>	3	1.29 x 10 <sup>8</sup>	4.31 x 10 <sup>7</sup>	3.196	<b>0.029</b>
S x F	3	9.21 x 10 <sup>7</sup>	3.07 x 10 <sup>7</sup>	2.277	0.087
Residual	71	9.57 x 10 <sup>8</sup>	1.35 x 10 <sup>7</sup>		

1 Winter and summer surveys.

2 Lyttelton dry dock, and Tauranga Marina, Orams Marine and Westpark Marina haul-out operations.

The treatment stages in which liquid samples were taken varied between seasons and facilities and reflected the locations our field teams were able to access, or permitted access to (Table 3). Some differences in sampling also occurred because the Tauranga Marina and Orams Marine had received upgrades to their waste treatment system between our initial sampling in 2003 and samples taken in 2006. Across all facilities, settlement and filtration progressively reduced the mean concentrations of organisms in the liquid effluent. In samples taken from the first chamber of the multi-chamber settlement tanks, concentrations of intact animals, propagules and unicellular organisms were reduced by between 20.5 percent and 99.5 percent, and the rank abundance of filamentous algae decreased by a range of 10 to 60 percent (Table 20, Figure 14). Concentrations of animals, propagules and unicellular organisms in samples taken from the final settlement chamber had been reduced by a range of 39.7 to 100 percent and abundance of filamentous algae by a range of 9.6 to 100 percent. Where samples were taken of the final effluent (Table 3), concentrations of animals, propagules and unicellular organisms had been reduced by  $\geq 98.5$  percent compared to concentrations observed in the water blast runoff, while abundance of filamentous algae had been reduced by a range of 80 percent to 100 percent (Table 20; Figure 14). Material encountered in the final effluent included nematodes, copepod crustaceans, polychaetes, rotifers, ciliates, diatoms, dinoflagellates, tintinnids, filamentous algae, spores and eggs (Tables 18 and 19).



Figure 14: Concentrations (no./10 L) of animals, propagules, unicellular organisms and filamentous algae collected in liquid effluent samples during winter (2003) and summer (2006) at various hull cleaning facilities. Data are counts of organisms and are not an indicator of viability. Data are mean  $\pm$  1 s.e.

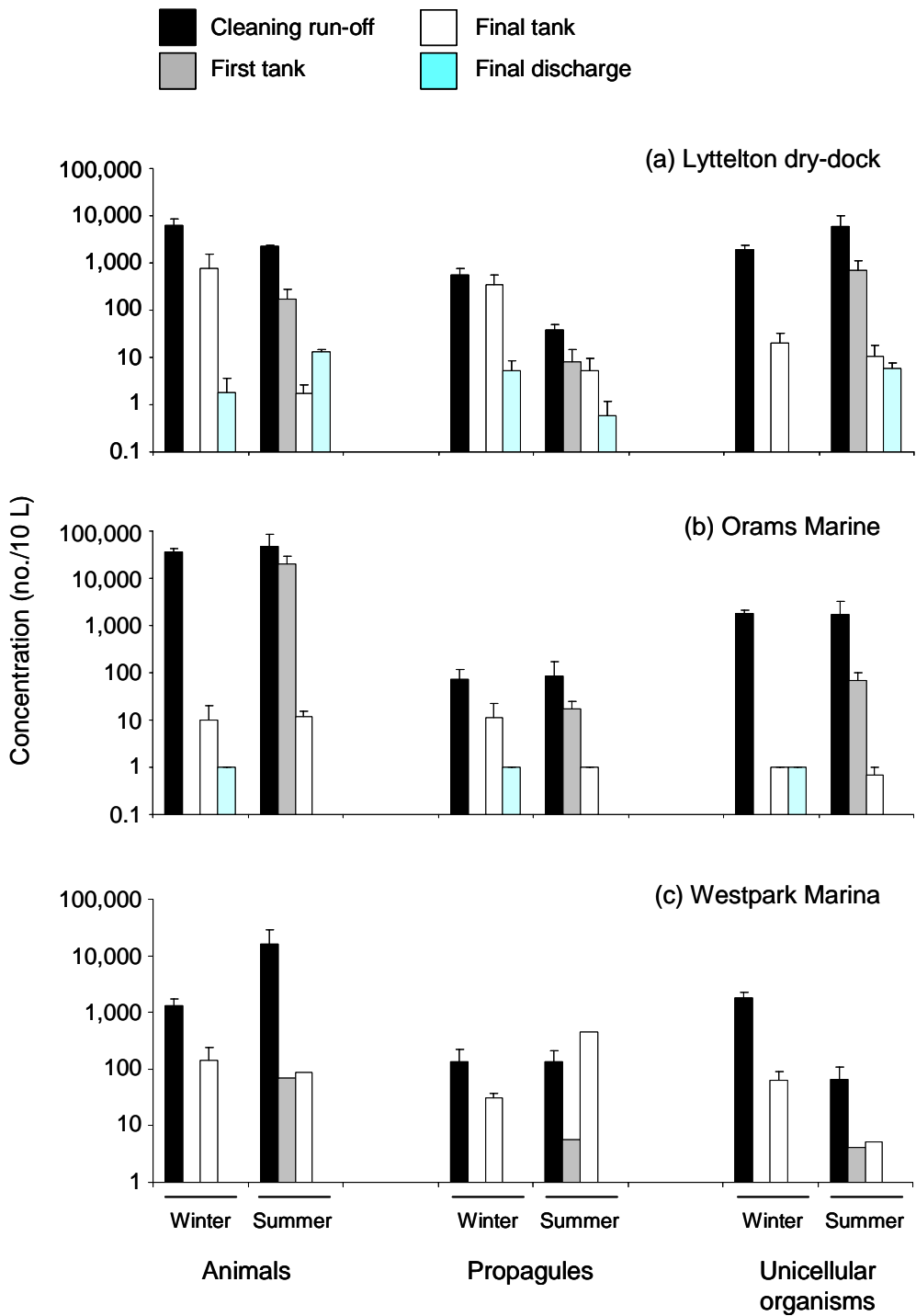
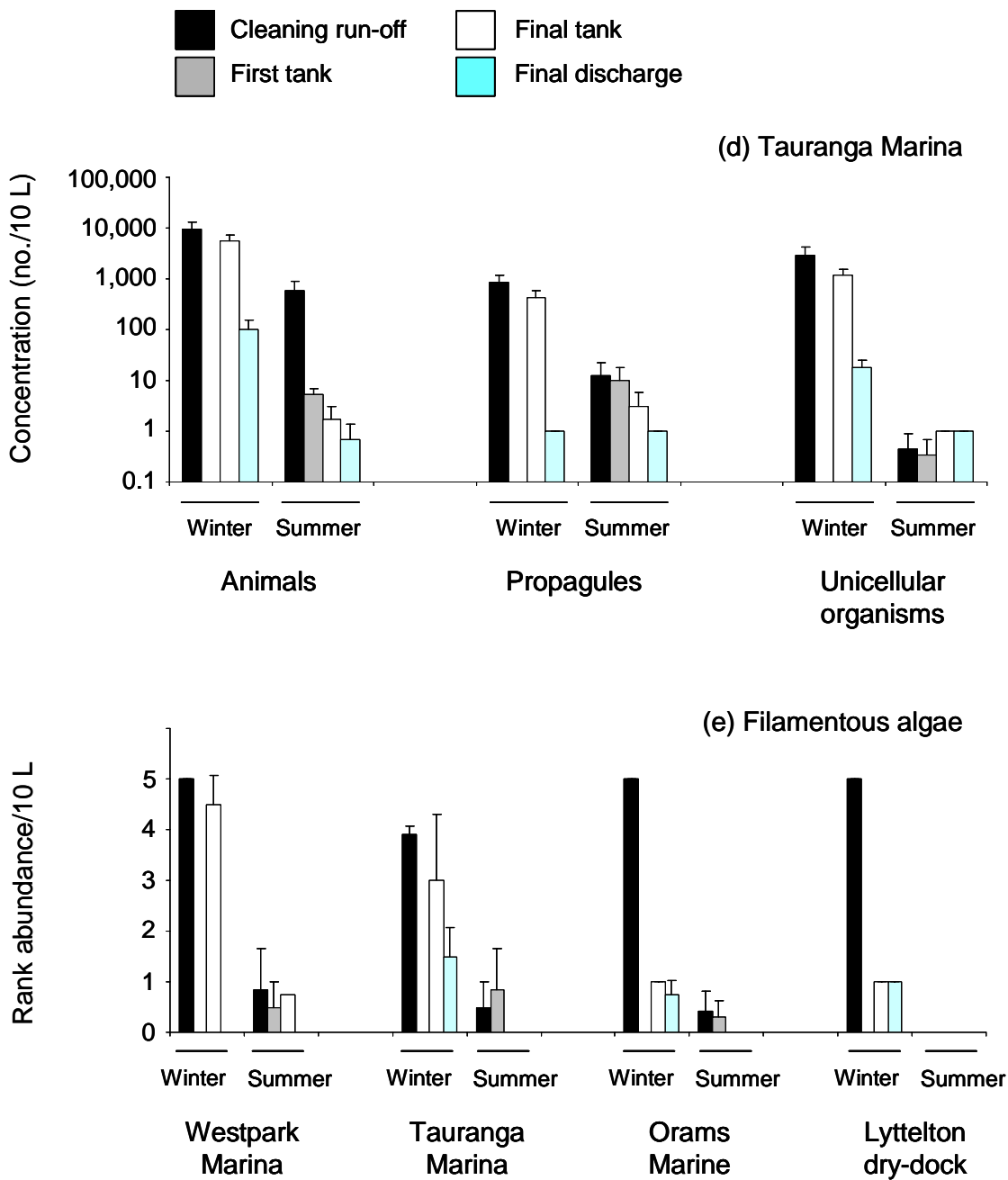
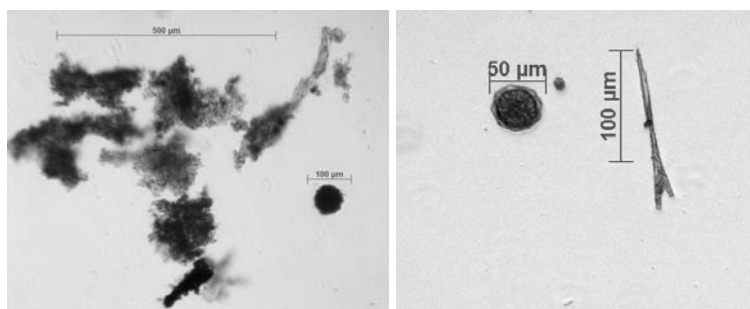


Figure 14 (continued): Concentrations (no./10 L) of animals, propagules, unicellular organisms and filamentous algae collected in liquid effluent samples during winter (2003) and summer (2006) at various hull cleaning facilities. Data are counts of organisms and are not an indicator of viability. Data are mean  $\pm$  1 s.e.



Where sampling of final liquid effluent was possible during summer sampling (Lyttelton dry dock and Tauranga Marina), representative images of material within the final effluent were taken using a Leica digital camera mounted on a compound microscope to obtain representative sizes retained by the 60 µm sieve. In Lyttelton dry dock, retained material ranged in size from detrital aggregations 50–500 µm in size (Figure 15A) down to what appeared to be propagules/zoospores around 50 µm in size (Figure 15B). At Tauranga Marina, retained material ranged in size from detrital aggregations 100–200 µm in size and pollen grains at 100 µm (Figure 16A-B) to stalked diatoms 100 µm in size with stalk sizes of 200 µm (Figure 16C).

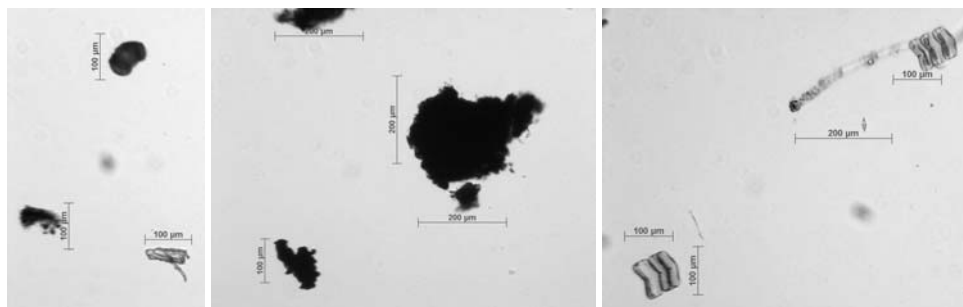
**Figure 15: Representative images of material retained in a 60 µm sieve from the final effluent at Lyttelton dry dock. Detrital aggregations (A) and propagule/zoospore (B).**



A

B

**Figure 16: Representative images of material retained in a 60 µm sieve from the final effluent at Tauranga Marina. Detrital aggregations and pollen grains (A-B) and stalked diatoms (C).**



A

B

C

### 3.1.2.2 Viability of organisms

Vital staining for mitochondria (Janus Green) returned positive results within liquid samples collected from the water blast runoff in all facilities during both winter and summer sampling (Table 22). Wherever staining was observed, it was clearly contained within the organisms, such as the exoskeleton of crustaceans, the bodies of nemertean worms or the cells of filamentous algae. In all runoff samples analysed, visible movement of organisms (mainly observed in nemerteans) occurred only in the initial runoff effluent from three vessels sampled during winter and four vessels sampled during summer. No movement of organisms was observed at any other stage of treatment during winter. During summer sampling, movement of rotifers was detected in the Lyttelton dry dock discharge effluent. However, this is likely to have been caused by sample contamination rather than the actual presence of live and moving material in the effluent as a small tear was observed in the seawater filtering sieve following the

processing of the final effluent samples for Lyttelton. During winter sampling, positive mitochondrial stains were obtained in samples from in the first settlement tank chamber of the Lyttelton dry dock and the Tauranga Marina. During summer, positive mitochondrial stains were obtained from the first and final settlement tank chambers in all facilities, and from the final effluent at the Lyttelton dry dock and the Tauranga Marina (Table 22). However, only in filamentous algae was the stain clearly retained within intact cell walls. For most other biota the staining was observed within fragments (for example, body parts) of organisms. Complete exoskeletons of crustaceans and bryozoans (and other taxa) were in most cases found to be empty and did not stain properly.

**Table 22: Movement and mitochondria in liquid samples taken at the various stages of treatment. Presence and absence of mitochondrial stains and visible movement is indicated by '+' and '-', respectively.**

		Winter		Summer	
		Movement	Mitochondria	Movement	Mitochondria
Lyttelton dry dock	Cleaning runoff	-	+	-	+
	<i>Vessel 1</i>	-	+	+	+
	<i>Vessel 2</i>			-	+
	<i>Vessel 3</i>				
	First tank			-	+
	Final tank	-	+	-	+
	Final discharge	-	-	+	+
Orams Marine	Cleaning runoff	+	+	-	+
	<i>Vessel 1</i>	+	+	+	+
	<i>Vessel 2</i>			-	+
	<i>Vessel 3</i>				
	First tank			-	+
	Final tank	-	-	-	+
	Final discharge	-	-		
Westpark Marina	Cleaning runoff	-	+	-	+
	<i>Vessel 1</i>	-	+	+	+
	<i>Vessel 2</i>			+	+
	<i>Vessel 3</i>				
	First tank			-	+
	Final tank	-	-	-	+

	Final discharge				
Tauranga Marina	Cleaning runoff	-	+	-	+
	<i>Vessel 1</i>	-	+	-	+
	<i>Vessel 2</i>	-	+	-	+
	<i>Vessel 3</i>	-	+	-	+
	First tank			-	+
	Final tank	-	+	-	+
	Final discharge	-	-	-	+

## 3.2 Specific objective 2

### 3.2.1 Modelling water residence time and particle settling in treatment systems

The total modelled hydraulic residence time distributions for the four treatment systems are illustrated in Figures 17 through 23 and Table 23 (see methods section “Water residence time” for modelling assumptions). Typically, as water inflow increases the effluent probability density function and cumulative probability residence times decrease (that is, liquid effluent spends less time in the treatment system as water inflow into the system increases). During summer, at typical flow rates used at each facility the residence times at Lyttelton dry dock, Orams Marine, Westpark Marina and Tauranga Marina were 7.4, 1296, 70.6, and 713 h respectively. During winter, the residence times at Lyttelton dry dock, Orams Marine, Westpark Marina and Tauranga Marina during winter were 7.4, 2544, 156, and 1416 h respectively.

The modelled concentrations of particles (or organisms) potentially leaving the treatment systems as a function of sink rate are shown in Figures 24 through 30 and Table 23 (see methods section “*Particle settling*” for modelling assumptions). Examination of log-transformed (this increases the y-axis scale for detailed examination) relative particle concentration in outflow at typical flow rates revealed that 100 percent particle retention with particles of a fast sink rate of  $1.0 \text{ m min}^{-1}$  was achieved at all facilities. Orams Marine, Westpark Marina (winter only) and Tauranga Marina offered 100 percent particle retention with particles of a sink rate of  $0.1 \text{ m min}^{-1}$ . Orams Marine and Tauranga Marina offered 100 percent particle retention with particles of a sink rate of  $0.01 \text{ m min}^{-1}$ . No cleaning facility offered 100 percent particle retention for particles with very slow sink rates of  $0.001$  or  $0.0001 \text{ m min}^{-1}$ .

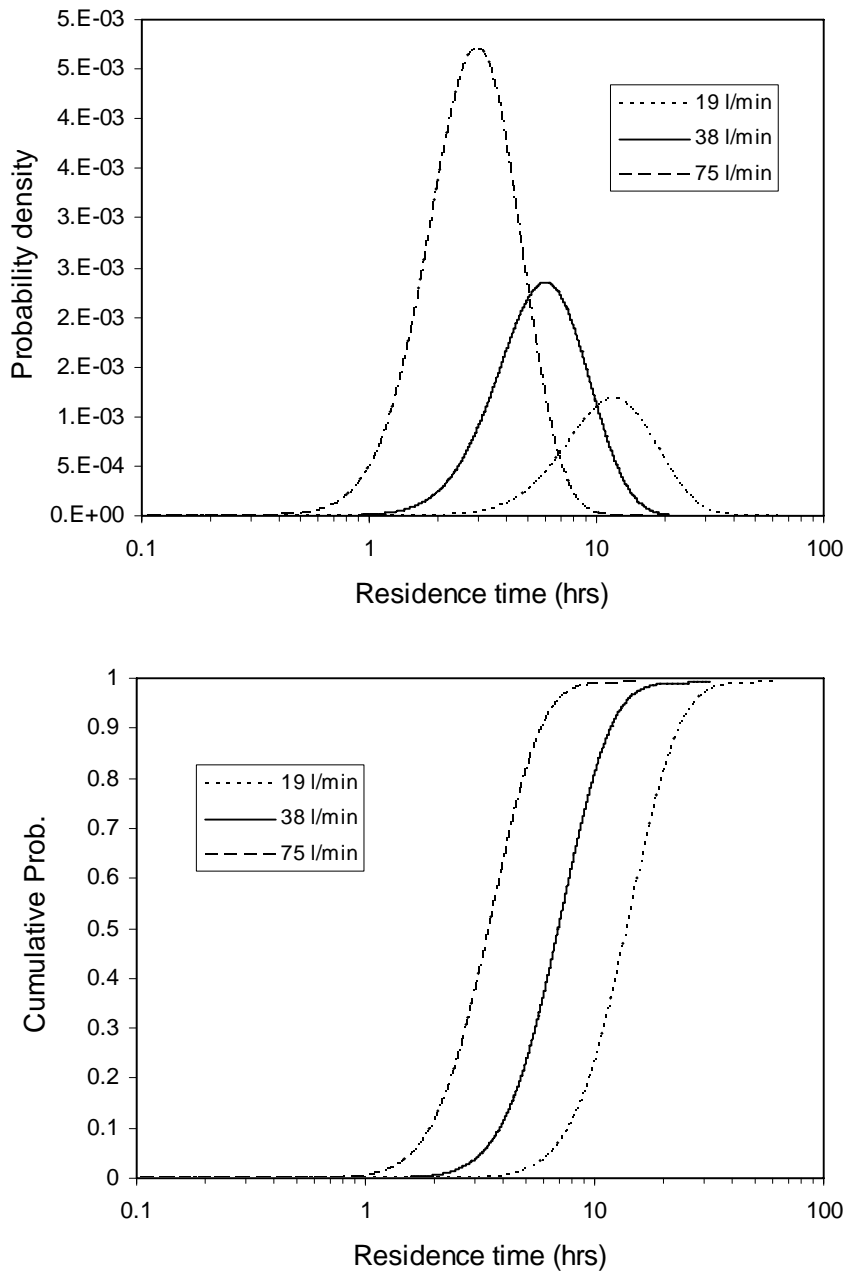
The larger the volume of the liquid effluent treatment system in relation to the volume of effluent input, the longer the residence time of effluent from each vessel in the system. For example, Lyttelton dry dock had a relatively small treatment system volume of  $16.7 \text{ m}^3$  and large volume of freshwater used per vessel ( $300 \text{ m}^3$ ). Thus the effluent residence time was only 7.4 h (summer and winter) before discharge. Conversely, at Orams Marine the treatment system volume was  $46 \text{ m}^3$  and smaller volumes of freshwater were used per vessel ( $0.34 \text{ m}^3$ ). Thus, a residence time of 1296 h in summer was calculated for this latter facility before discharge to the municipal sewage system.

The more vessels that were cleaned per day the lower the residence time of effluent from each vessel in the system. For example, at Westpark Marina the residence time of effluent from each vessel was 70.6 h in summer and 156 h in winter. The exception to any seasonal effect of residence time was the Lyttelton dry dock where hull cleaning is relatively constant throughout the year. Comparison of the residence time in the treatment systems for Orams Marine, Westpark Marina and Tauranga Marina revealed no statistical difference between season (Student’s t-test,  $P > 0.05$ ) due to low replication level and high data variability. Log-transformation of data did not result in any statistical difference either. Overall, mean  $\pm$  1 s.e. modelled residence time of water in the treatment systems for Orams Marine, Westpark Marina and Tauranga Marina was  $693.2 \pm 353.9 \text{ h}$  for summer and  $1372 \pm 689.7 \text{ h}$  for winter.

**Table 23: Modelled average number of hours liquid effluent resides in treatment systems at various facilities per cleaning season (summer or winter) and whether or not each facility's treatment system can theoretically allow particles or representative sinking rates to settle-out before filtering/screening and then discharge at typical flow rates. Figures in brackets are calculated average number of hours liquid effluent resides in treatment systems based on double and half input flow rates.**

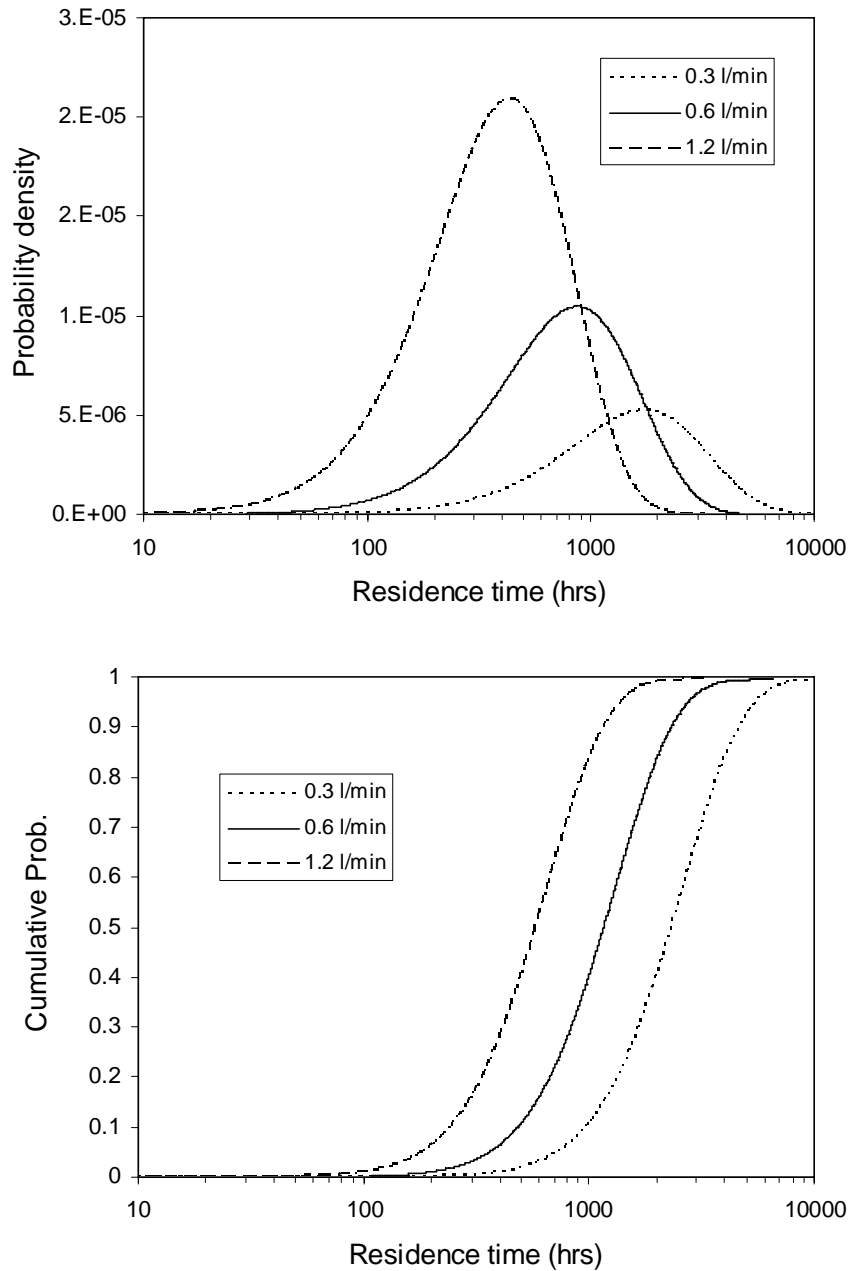
Facility	Season	Average no. hours effluent resides in system	Retains particles at sink rate of 1.0 m min <sup>-1</sup>	Retains particles at sink rate of 0.1 m min <sup>-1</sup>	Retains particles at sink rate of 0.01 m min <sup>-1</sup>	Retains particles at sink rate of 0.001 m min <sup>-1</sup>	Retains particles at sink rate of 0.001 m min <sup>-1</sup>
Lyttelton dry dock	Summer	7.4 (14.8, 3.7)	Yes	No	No	No	No
	Winter	7.4 (14.8, 3.7)	Yes	No	No	No	No
Orams Marine	Summer	1 296 (2592, 648)	Yes	Yes	Yes	No	No
	Winter	2 544 (5088, 1272)	Yes	Yes	Yes	No	No
Westpark Marina	Summer	70.6 (141.2, 35.3)	Yes	No	No	No	No
	Winter	156 (312, 78)	Yes	Yes	No	No	No
Tauranga Marina	Summer	713 (1426, 356.5)	Yes	Yes	Yes	No	No
	Winter	1 416 (2832, 708)	Yes	Yes	Yes	No	No

**Figure 17: Modelled hydraulic residence time distribution for three flow rates at the Lyttelton dry dock liquid effluent treatment system (summer and winter). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.**

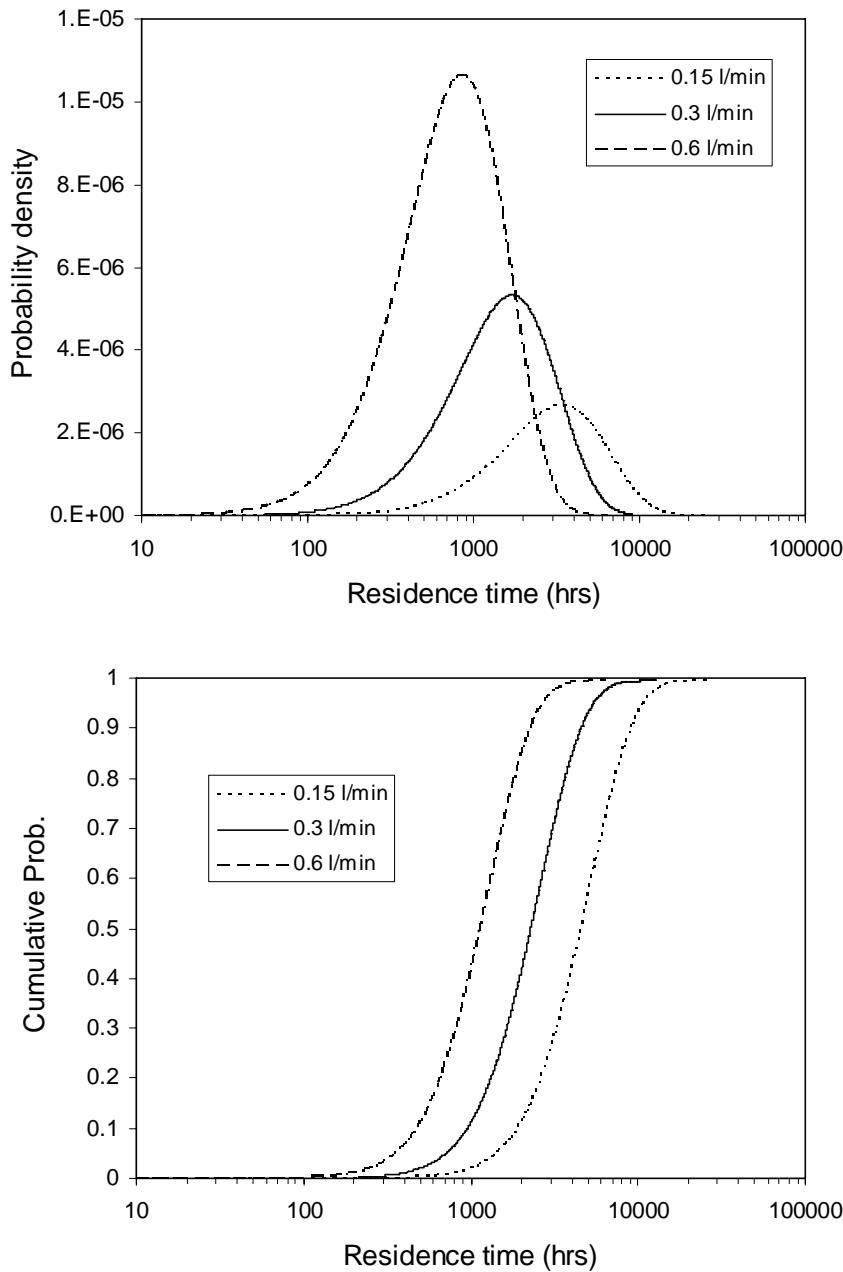




**Figure 18: Modelled hydraulic residence time distribution for three flow rates at the Orams Marine liquid effluent treatment system (summer operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.**



**Figure 19: Modelled hydraulic residence time distribution for three flow rates at the Orams Marine liquid effluent treatment system (winter operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.**



**Figure 20: Modelled hydraulic residence time distribution for three flow rates at the Westpark Marina liquid effluent treatment system (summer operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.**

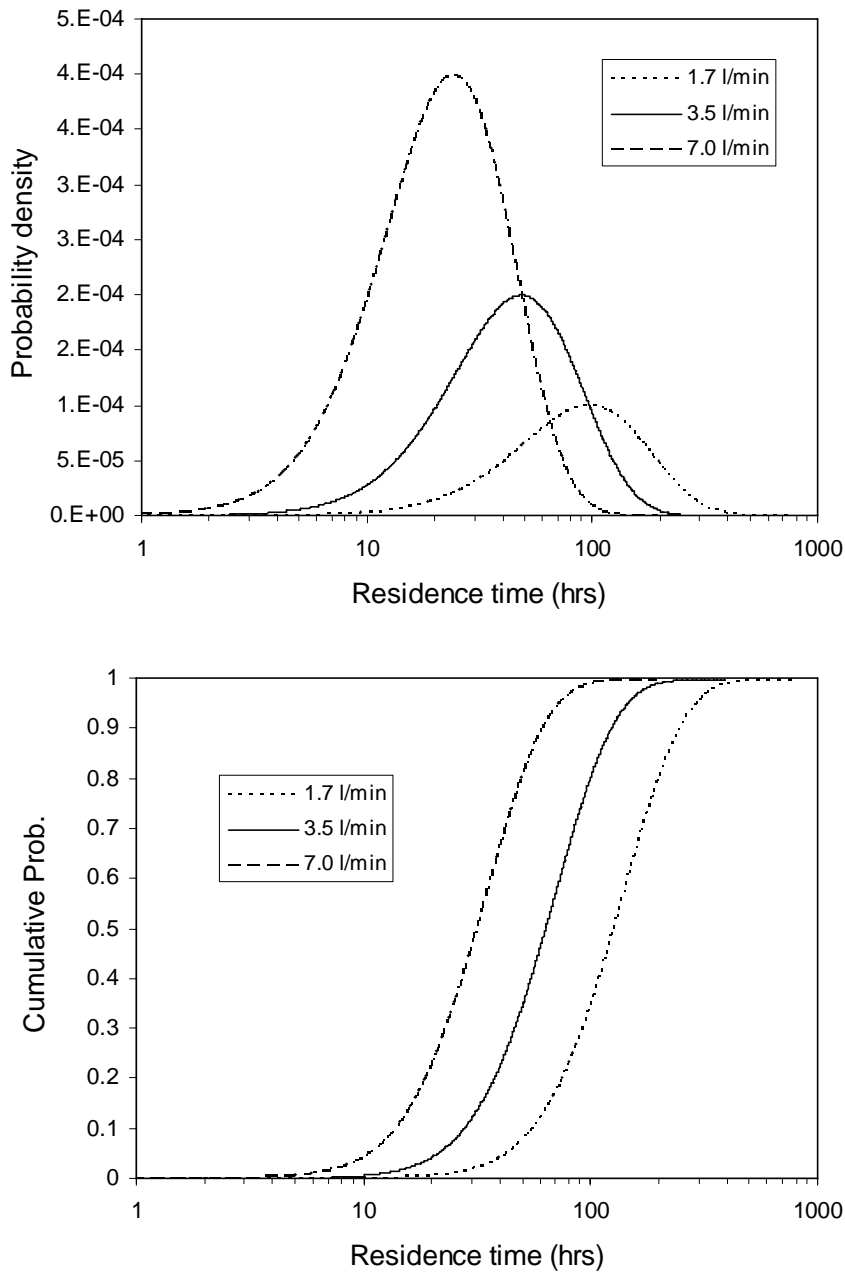


Figure 21: Modelled hydraulic residence time distribution for three flow rates at the Westpark Marina liquid effluent treatment system (winter operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.

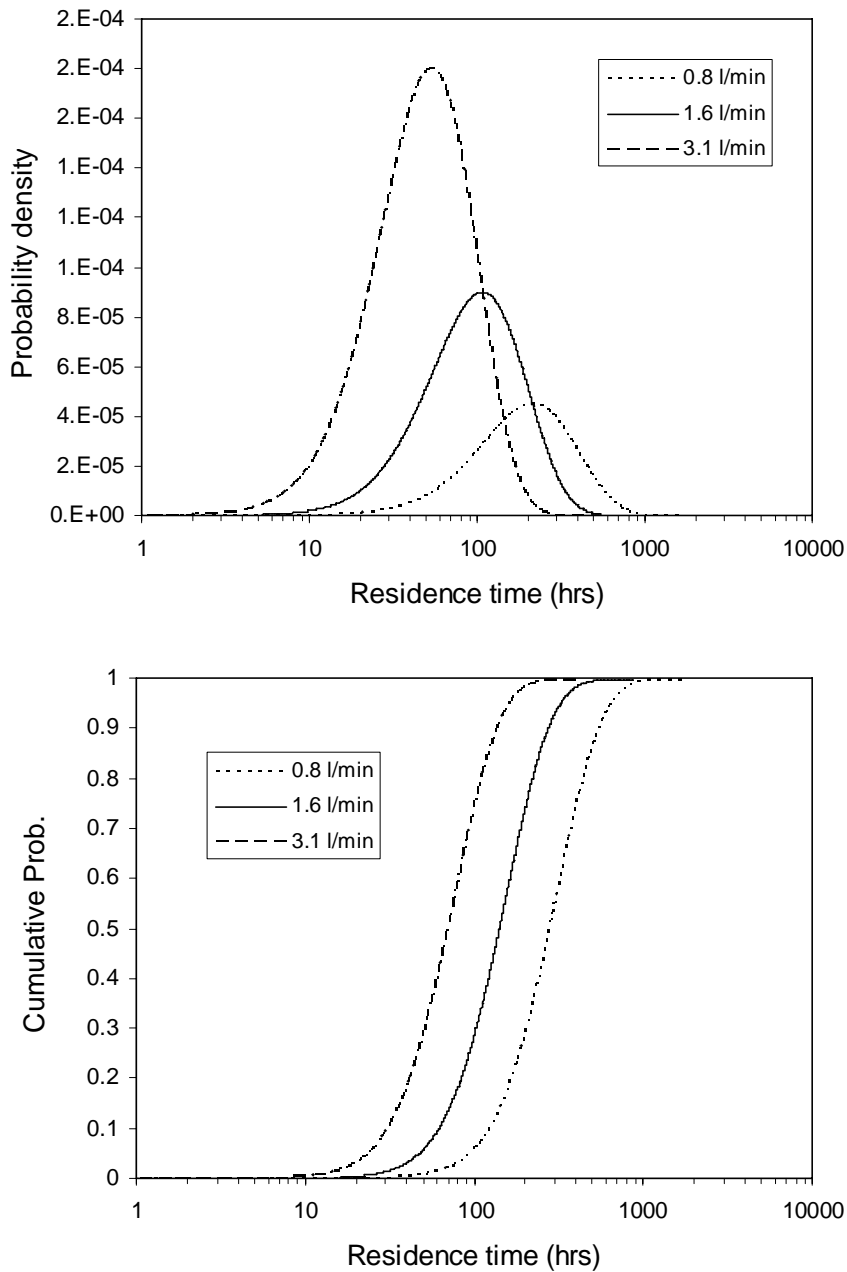
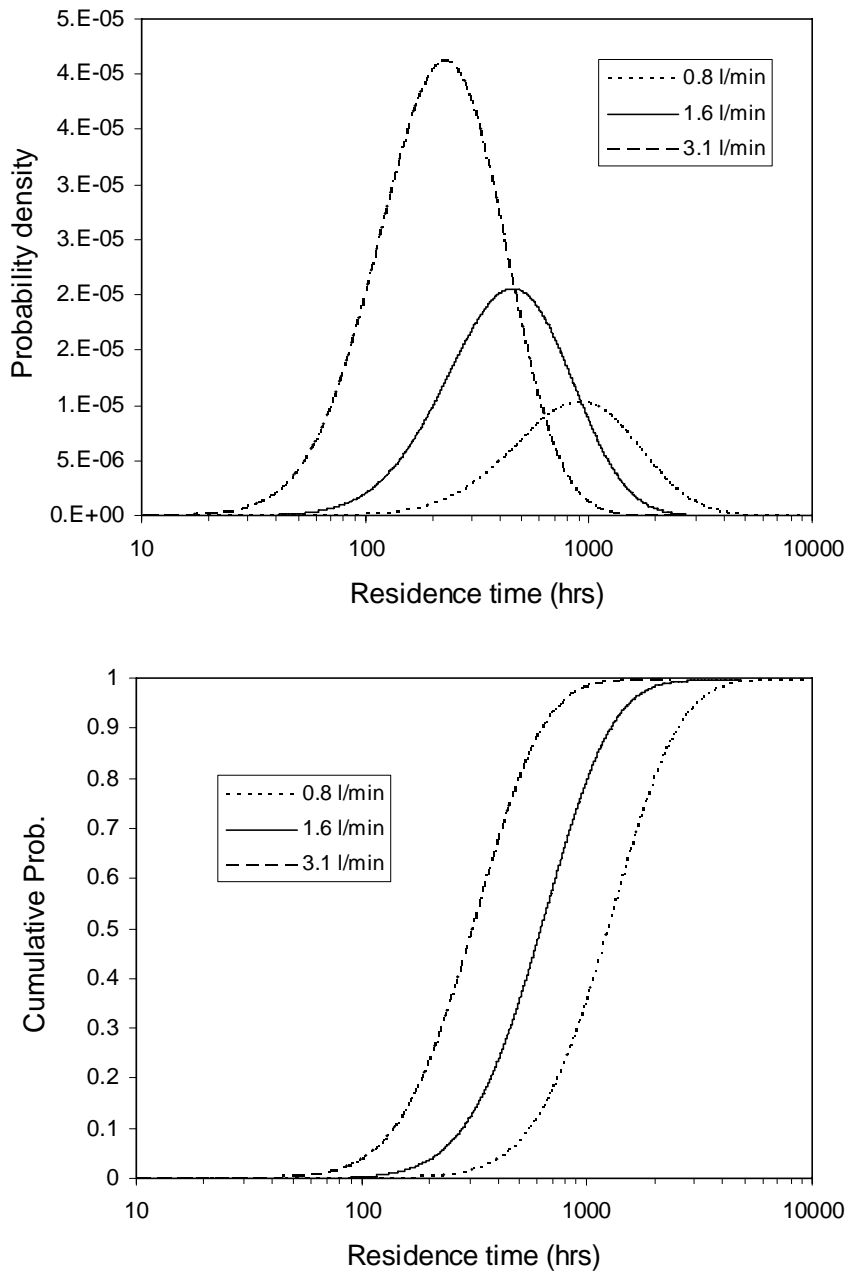


Figure 22: Modelled hydraulic residence time distribution for three flow rates at the Tauranga Marina liquid effluent treatment system (summer operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.



**Figure 23: Modelled hydraulic residence time distribution for three flow rates at the Tauranga Marina liquid effluent treatment system (winter operations). The top graph shows the relative frequency distribution for all water particles passing through a single reservoir in the treatment system as a probability density function. The bottom graph shows the cumulative residence time of an arbitrary water particle passing through the entire treatment system. Solid line indicates the usual inflow rate at this facility.**

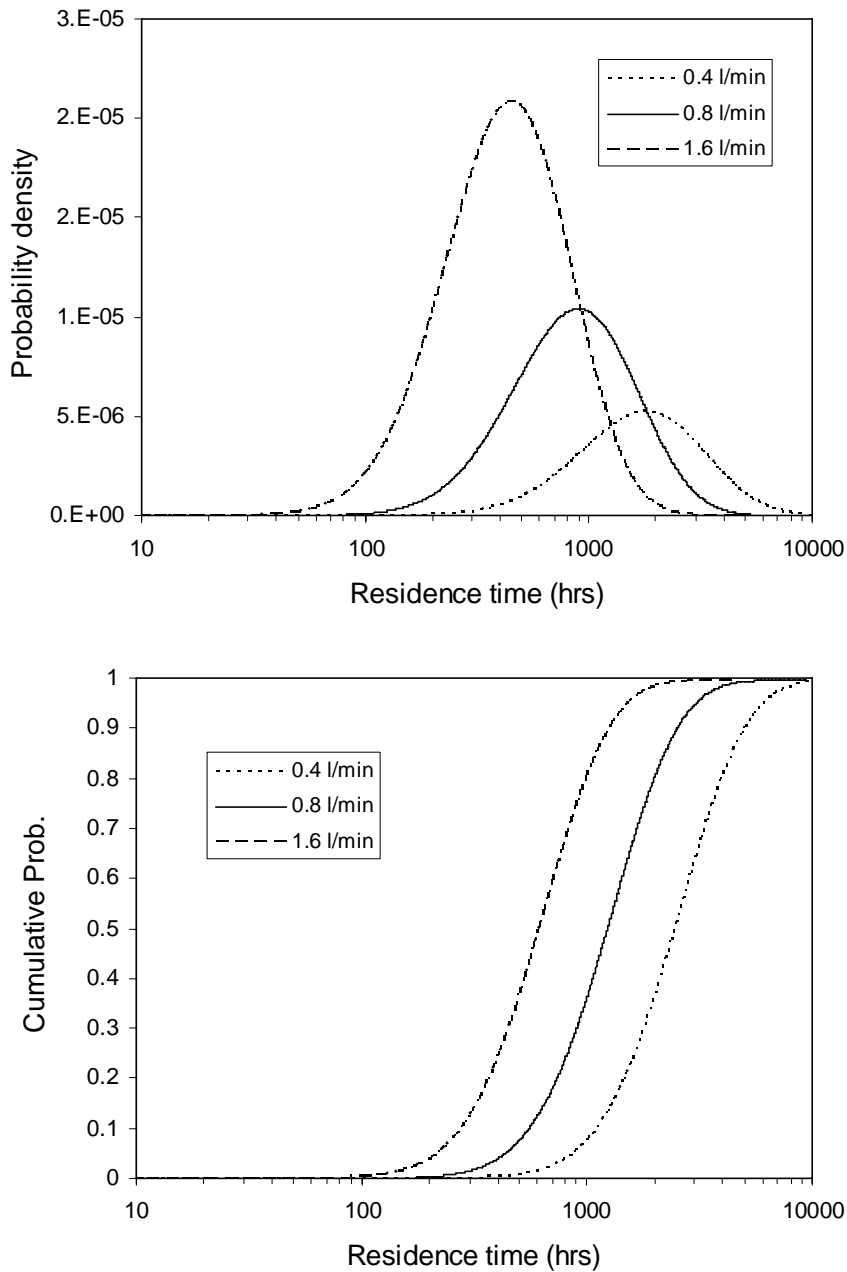


Figure 24: Relative modelled particle concentration in outflow with respect to inflow concentration for the Lyttelton dry dock liquid effluent treatment system (summer and winter). The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.

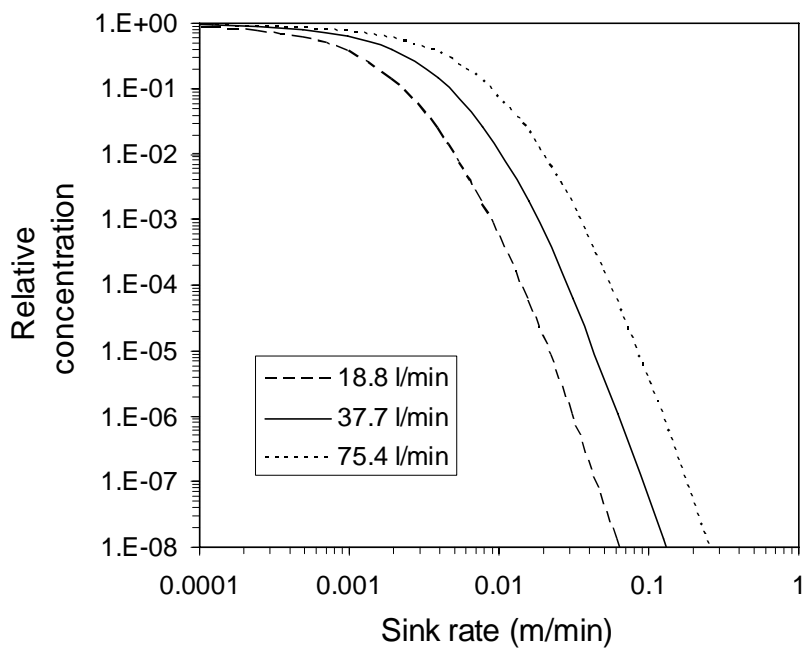
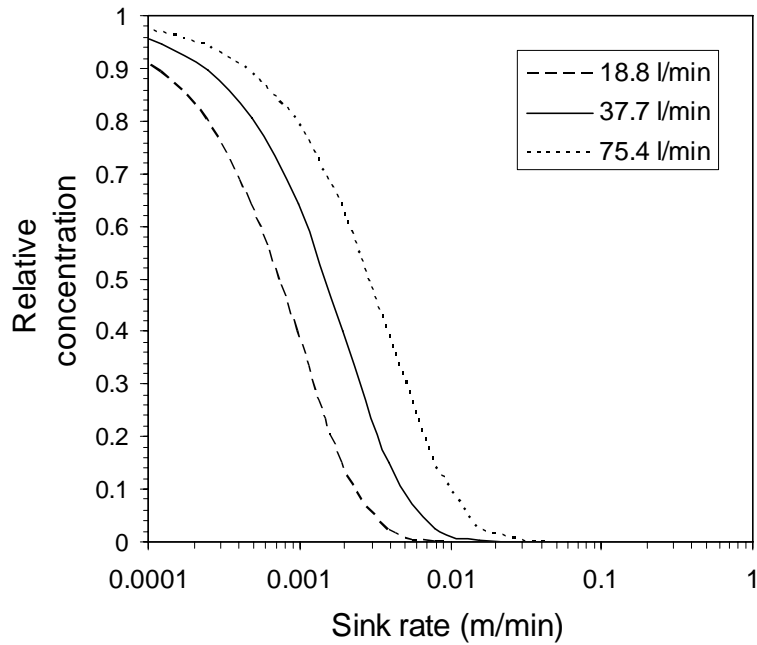


Figure 25: Relative modelled particle concentration in outflow with respect to inflow concentration for the Orams Marine liquid effluent treatment system in summer. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.

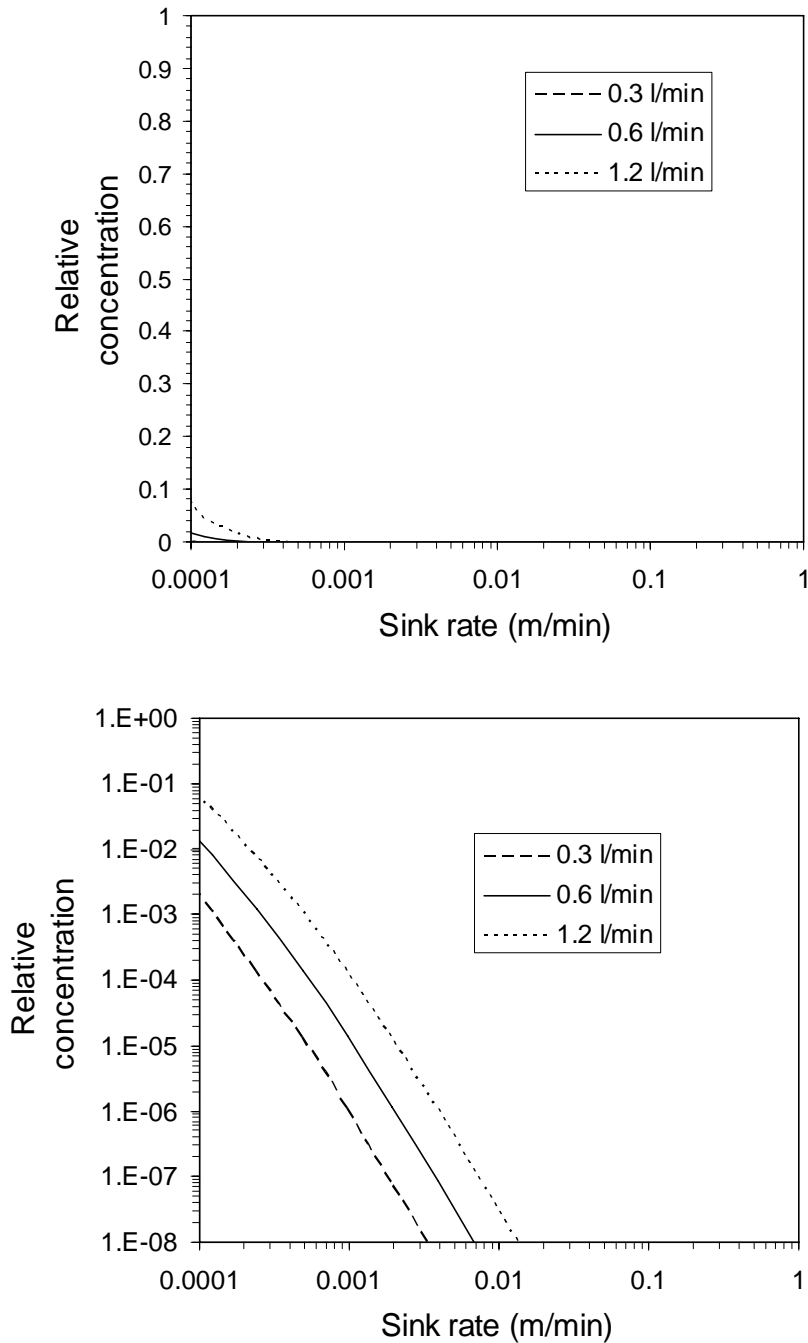




Figure 26: Relative modelled particle concentration in outflow with respect to inflow concentration for the Orams Marine liquid effluent treatment system in winter. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.

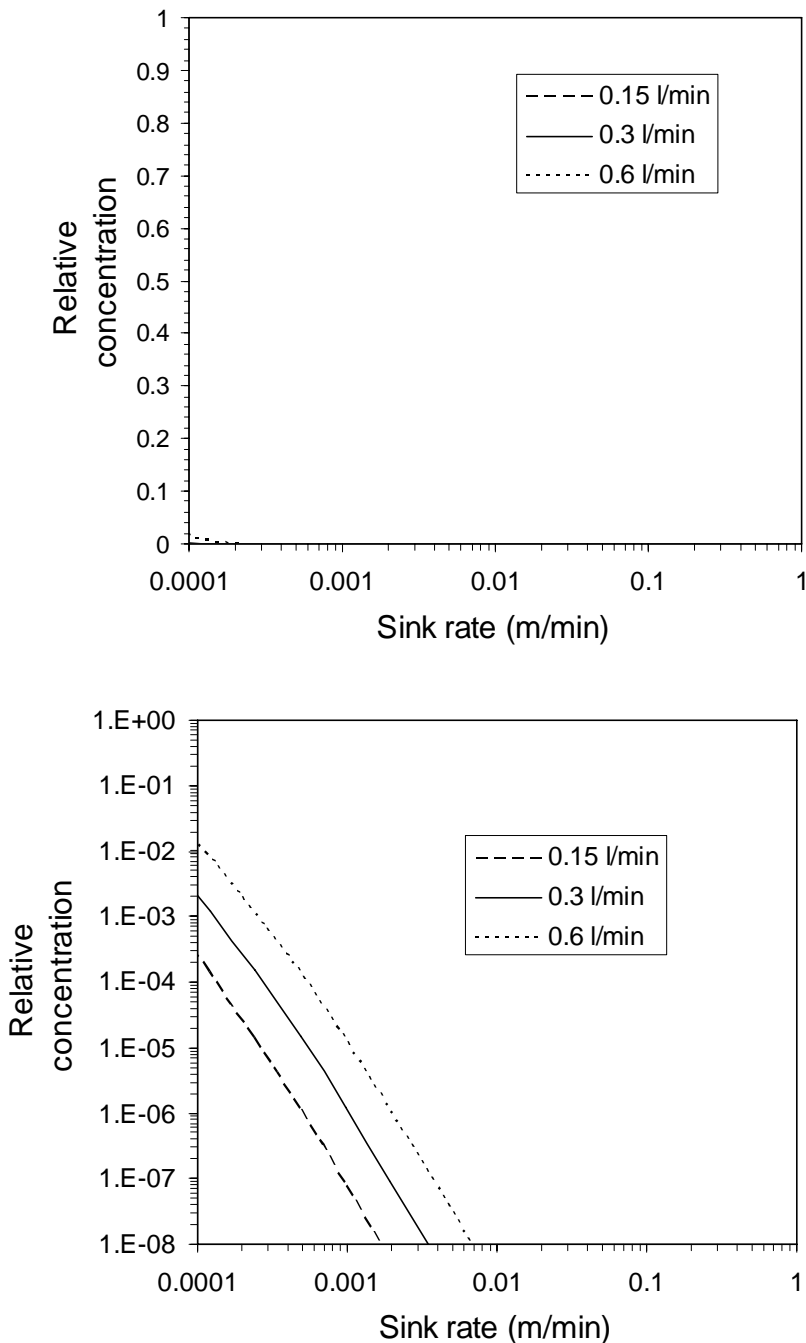


Figure 27: Relative modelled particle concentration in outflow with respect to inflow concentration for the Westpark Marina liquid effluent treatment system in summer. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.

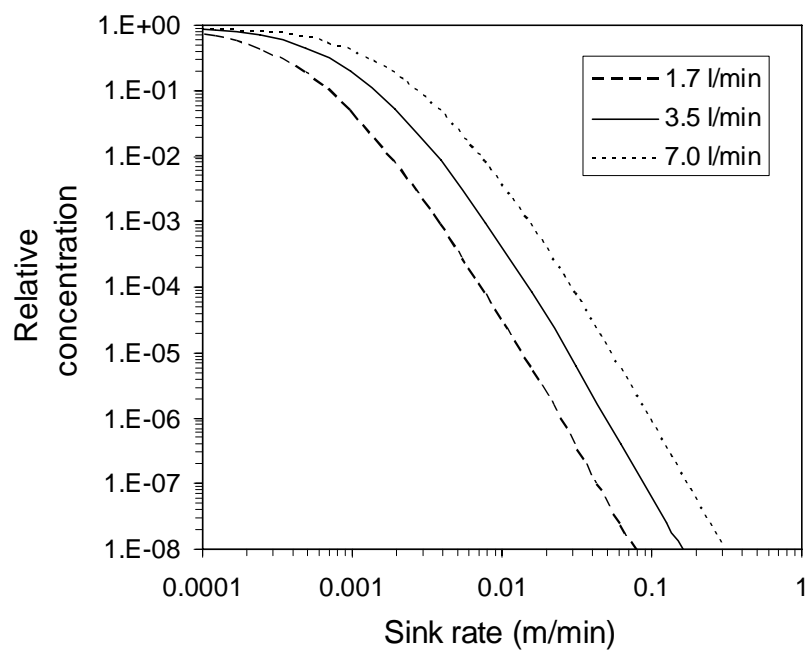
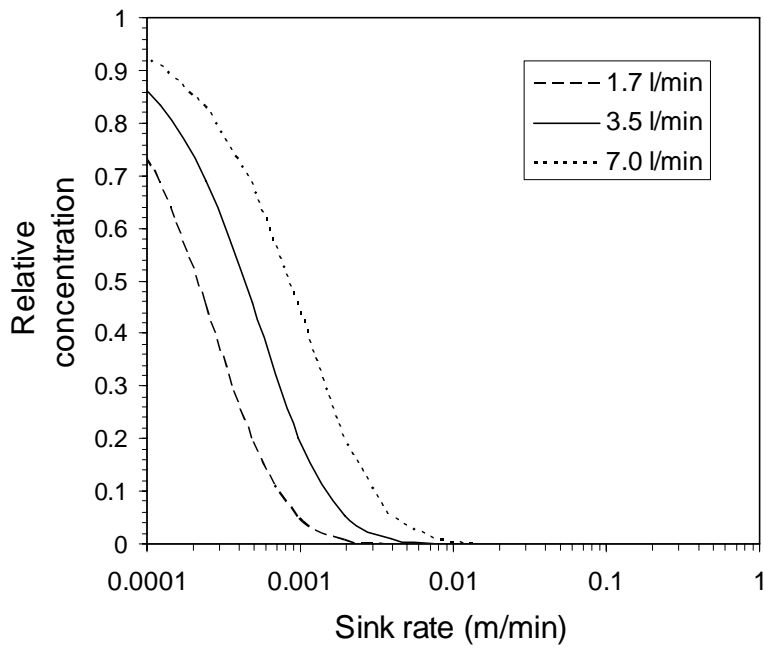


Figure 28: Relative modelled particle concentration in outflow with respect to inflow concentration for the Westpark Marina liquid effluent treatment system in winter. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.

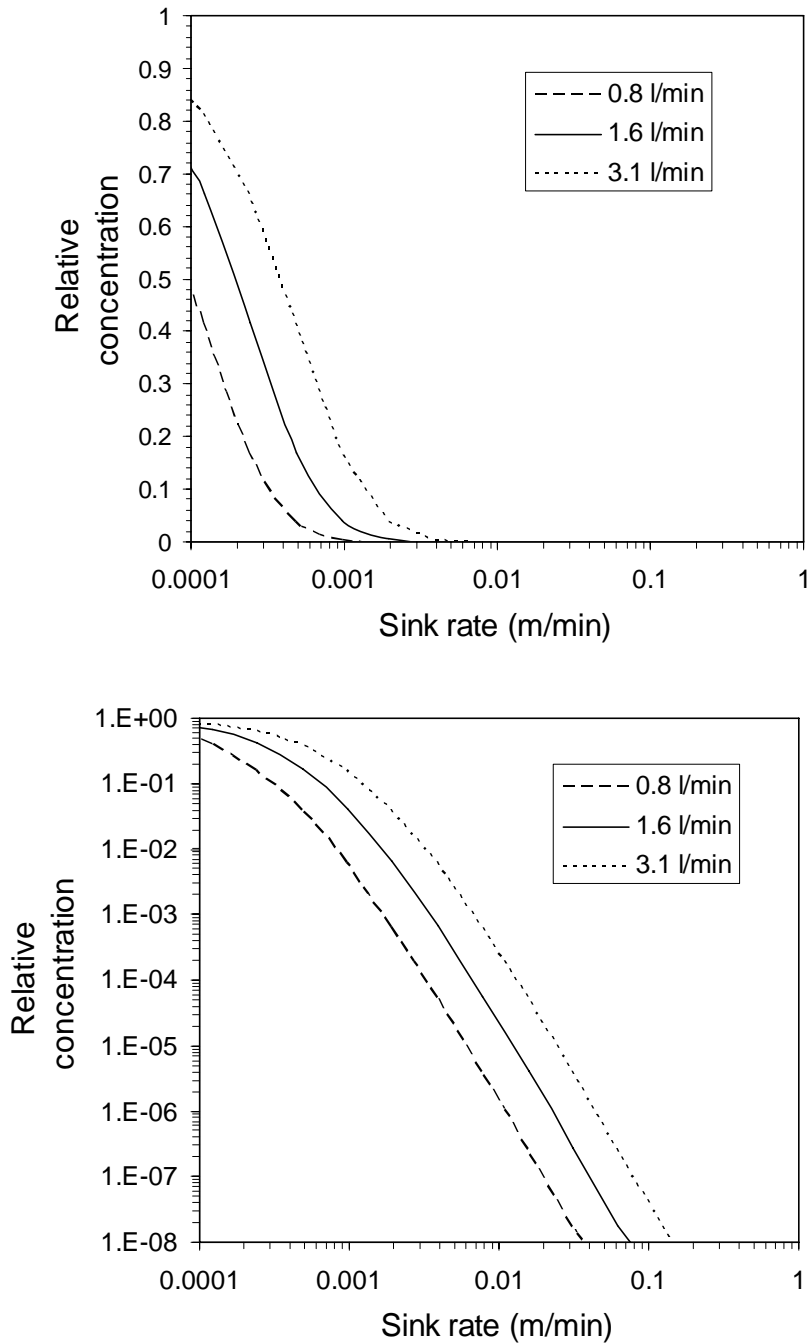


Figure 29: Relative modelled particle concentration in outflow with respect to inflow concentration for the Tauranga Marina liquid effluent treatment system in summer. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.

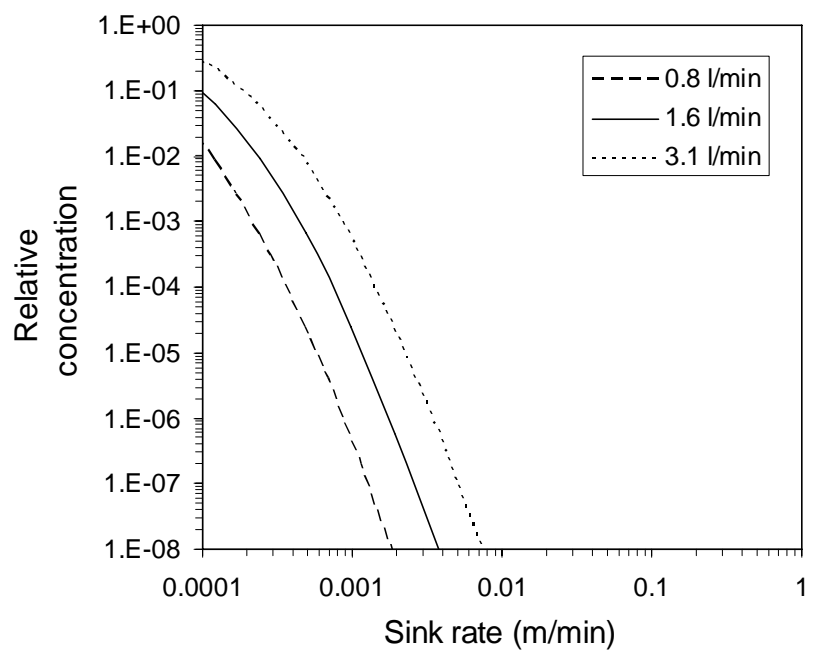
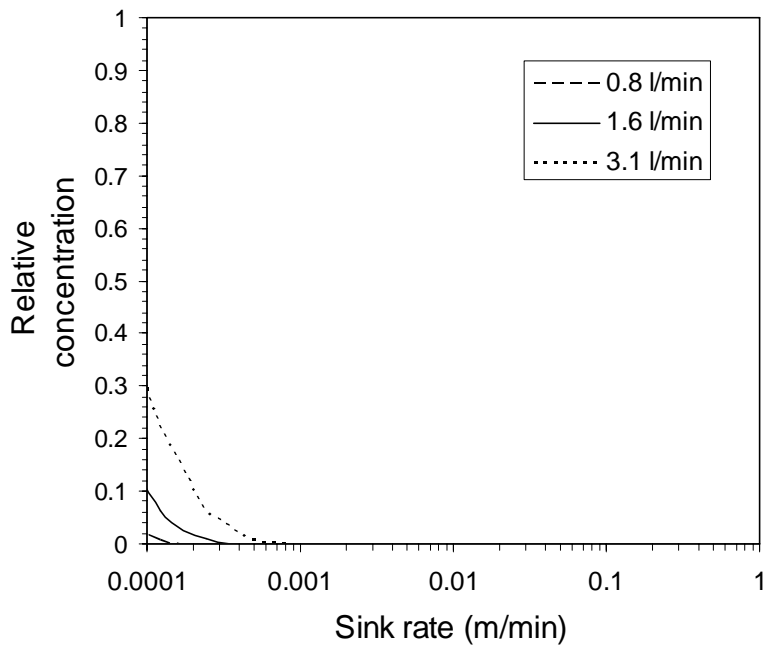
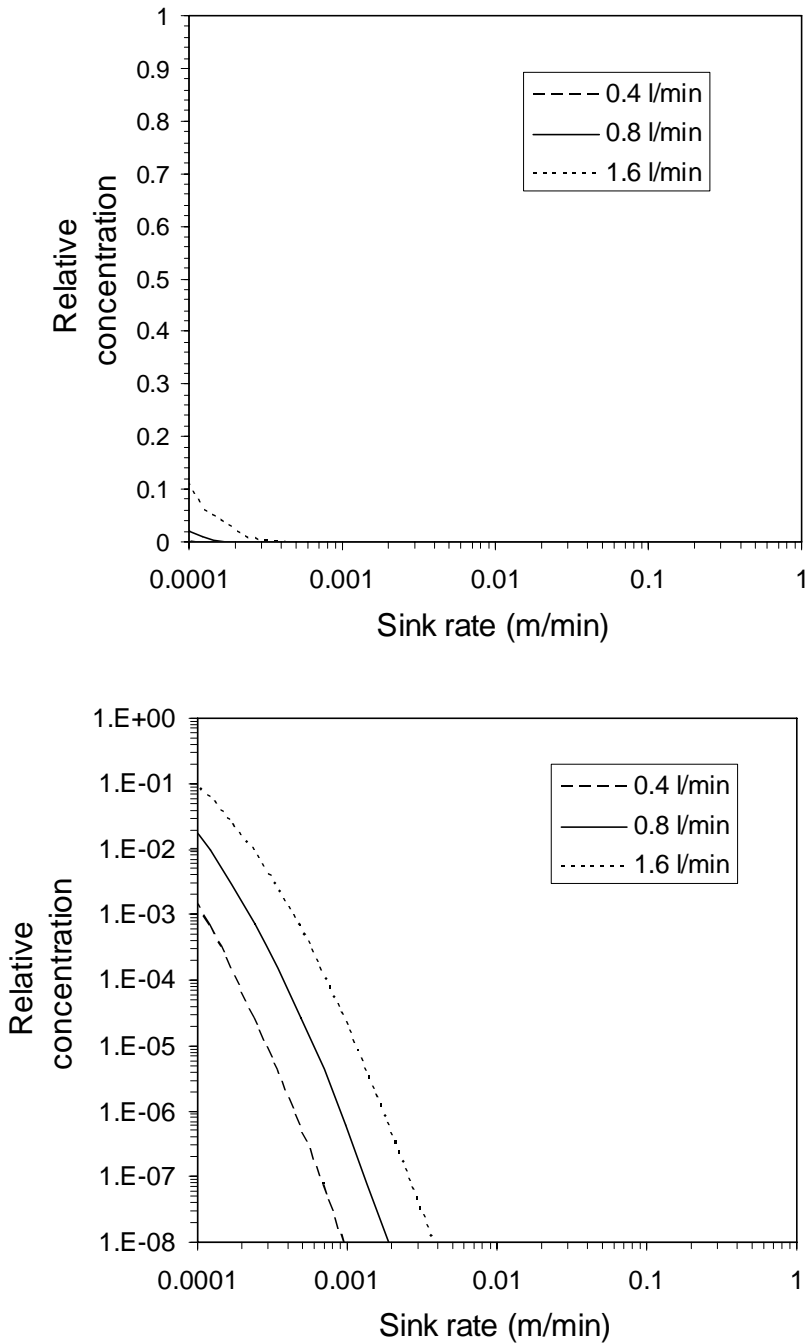


Figure 30: Relative modelled particle concentration in outflow with respect to inflow concentration for the Tauranga Marina liquid effluent treatment system in winter. The lower figure is the same as the upper figure except for the log-transformation of the y-axis to allow for more detailed scrutiny of particle concentration. Solid line indicates the usual inflow rate at this facility.



For residence time in the liquid effluent treatment systems of each facility, the marine NIS each facility could potentially be effective in killing are given in Table 24. Most facilities studied are theoretically effective in killing most NIS based on known salinity tolerances, although the Lyttelton dry dock and Westpark Marina with their shorter residence times were theoretically less effective. The longer residence times meant that species such as the invasive crab *Eriocheir sinensis* and the green mussel *Perna viridis*, which are tolerant of reduced salinity, are theoretically able to be killed within the Orams Marine and Tauranga Marina systems based solely on extended freshwater exposure provided by their systems. Seasonal differences in residence time only had an effect with the alga *Enteromorpha intestinalis* (not an NIS in New Zealand but which was found as hull-fouling and included as an example of a freshwater-tolerant alga) which could theoretically survive freshwater exposure in Westpark Marina's system in summer but not winter.

Certain freshwater-tolerant organisms such as the bivalve *Potamocorbula amurensis* may be able to survive for long periods within all treatment systems on the basis of salinity alone.

**Table 24: Salinity tolerances of various marine organisms that may be regarded as non-indigenous pest species (NIS) or commonly encountered as hull-fouling. Minimum salinity tolerances are expressed as the minimum salinity (ppt) and the number of days for which the organism is able to survive at that salinity before death or severe damage occurs, or as lower normal habitat salinity level (ppt) in which animals are found (in brackets). For each organism, modelled hydraulic residence time was used to determine each facility's potential effectiveness at killing that organism: Lyttelton dry dock (LYT), Orams Marine (OMM), Westpark Marina (WPM), and Tauranga Marina (TRG). Facilities indicated with a question mark (?) are facilities that may be effective but the minimum salinity tolerance of the NIS in question does not allow certainty of mortality.**

Genus and Species	Minimum salinity tolerance (ppt/days)	Effective facility based on hydraulic residence time in summer	Effective facility based on hydraulic residence time in winter
<i>Sabella spallanzanii</i> <sup>1,2,3</sup>	0.0/0.1 days, (26.0)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Caprella mutica</i> <sup>4</sup>	20/2 days poss., (11)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Monocorophium acherusicum</i> <sup>4</sup>	1.0/10 days, (15)	LYT, OMM, WPM, TRG?	LYT, OMM, WPM, TRG?
<i>Carcinus maenus</i> <sup>1,2,3</sup>	1.0/2 days, (1.4)	LYT, OMM, WPM, TRG?	LYT, OMM, WPM, TRG?
<i>Eriocheir sinensis</i> <sup>1,3</sup>	0.0/7 days, (5.0)	OMM, TRG	OMM, TRG
<i>Ciona intestinalis</i> <sup>3,4</sup>	(18)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Styela clava</i> <sup>1,3</sup>	0.0/1 day, (20.0)	OMM, WPM, TRG	OMM, WPM, TRG
<i>Asterias amurensis</i> <sup>1,2,3</sup>	0.0/0.01 days or 24/9 days, (26.0)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Bugula neritina</i> <sup>3,4</sup>	14.0, (18.0)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Watersipora subtorquata</i> <sup>3,4</sup>	(25.0)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Crassostrea gigas</i> <sup>2,3</sup>	3.0, (5.0)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Musculista senhousia</i> <sup>1,2,3</sup>	(6.6)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Perna viridis</i> <sup>3</sup>	0.0/14 days, (18.0)	OMM; TRG	OMM; TRG
<i>Potamocorbula amurensis</i> <sup>1,3</sup>	0.0/osmoconforms		
<i>Caulerpa taxifolia</i> <sup>1</sup>	10.0/1–7 days	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG

<i>Codium fragile ssp tomentosoides</i> <sup>3</sup>	12.5, (17.5)	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Enteromorpha intestinalis</i> <sup>4</sup>	0.0/5 days, (5.0)	OMM, TRG	OMM, WPM, TRG
<i>Undaria pinnatifida</i> <sup>1,3</sup>	0.0/1 day, (20.0)	OMM, WPM, TRG	OMM, WPM, TRG
<i>Alexandrium catenella</i> <sup>2</sup>	15.0/14 days	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Alexandrium minutum</i> <sup>2,3</sup>	3.0	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Alexandrium tamarense</i> <sup>2</sup>	7.0	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG
<i>Gymnodinium catenatum</i> <sup>2,3</sup>	10.0/2 days	LYT, OMM, WPM, TRG	LYT, OMM, WPM, TRG

1 Organism is listed on the New Zealand Register of Unwanted Organisms under the Biosecurity Act 1993.

2 Organism is listed on the Australian Ballast Water Advisory Council's (ABWMAC) schedule of non-indigenous pest species.

3 Listed by Hayes et al (2005) as being of medium-low to high priority as invasive species in Australia

4 Organism encountered during ZBS2005-22 (summer).



## 4.0 Conclusions

In relation to the spread of invasive marine organisms, vector management involves four common approaches: 1) to prevent exposure of vectors to invasive species, 2) to enhance resistance of vectors to colonization by the species, 3) to control the movement of infested vectors (quarantine of vectors) and, 4) to remove and quarantine infestations from affected vectors (sanitation) (Floerl et al. 2005). Hull cleaning is one effective method of sanitation through not only the removal of fouling organisms (which may be NIS), but also the quarantine and destruction of invasive marine organisms subsequent to and following their removal from vessel hulls. Secondly, hull cleaning plays an important role in vector protection by ensuring the proper removal of hull-fouling organisms in preparation for protective measures such as antifouling painting — failure to effectively remove existing fouling may actually even enhance subsequent recruitment by some fouling organisms (Floerl et al. 2005).

Because it is not standard practice for vessel owners/companies to examine the hull fouling on their vessels to determine whether NIS are present, a precautionary approach could be adopted assuming that all hull-fouling material removed during hull cleaning may contain NIS and that every NIS should be treated as a potential invasive species unless known as otherwise. To assist in the prevention and/or minimisation of the introduction and spread of hull-fouling marine NIS in New Zealand, facilities are needed which reflect this precautionary principle through providing for hull cleaning to be undertaken in a way that is effective at vessel sanitation, and provide for the quarantine and destruction of marine NIS organisms removed from vessel hulls.

### 4.1 Specific objective 1

Comparison of the results obtained in this investigation (ZBS2005-22 summer) with those of ZBS2002-04 (winter) indicate a complex interaction of factors which complicate any attempt to discern seasonal differences in the efficacy of various hull cleaning methods. However, some small seasonal trends were observed. For example, the concentration of propagules (that is, eggs, larvae and spores) in final discharge effluent was lower in the winter 2003 study (ZBS2002-04) than the summer 2006 study (ZBS2005-22), particularly at the Lyttelton dry dock.

Seasonal comparison of the number and viability of any organisms being discharged from the facilities examined is complicated by the fact that some facilities had improved their liquid effluent treatment systems between investigations. For example, improvements at the Tauranga Marina resulted in much lower concentrations of organisms in final discharge effluent in the summer 2006 study than the winter 2003 study. Comparison between summer and winter studies also revealed that although average fouling cover was generally higher on vessels in summer, average fouling biomass was higher in winter. This appears to be due to the seasonal differences in fouling organism representation; there was a higher abundance of barnacles during winter and higher abundance of small crustaceans (for example, amphipods) in summer. These seasonal patterns generally had little influence on the efficacy of hull cleaning. Seasonal differences in hull-fouling biomass may warrant further investigation in relation to determining whether there is a time period when minimising NIS invasion risk through hull cleaning activities is more effective (although this may be complicated if the vessels being cleaned are of international origin coming from a summer season). For example, if fouling biomass is higher in winter, and if fouling organisms are generally not reproductively active at this time, then winter may be a more efficacious time for minimising NIS invasion risk through hull cleaning.

Hull cleaning method, rather than seasonality, was the main determinant in hull-fouling organism viability during the cleaning and post-cleaning processes. Generally, dry dock and haul-out facilities and their associated hull cleaning methods result in fewer viable macro-fouling organisms collected as solid samples from beneath vessels. However, the results obtained in this summer assessment (ZBS2005-22) when compared with the original winter assessment

(ZBS2002-04) should only be regarded as a “snap-shot” assessment, and therefore treated with some caution. A rigorous assessment of the repeatability of any seasonal patterns in the survival of organisms in cleaning facilities would require sampling in more than a single summer and winter season (Hurlbert 1984; Underwood 1997) with comprehensive associated environmental monitoring.

The type and severity of physical damage to organisms removed from the hulls varied among vessels and operations, but not among season. In haul-out and dry dock operations, the pressure associated with the water blasting and the trampling of cleaning debris by cleaning staff had fragmented and crushed a large proportion of the soft-bodied organisms such as sponges, ascidians, flatworms and nudibranchs. In-water removal of organisms from vessel hulls with a paint scraper or a soft cloth caused similar damage to hard-bodied taxa, but considerably less to soft-bodied taxa. Patterns of mortality varied among operations following prolonged exposure to air (dry dock) or high-pressure blasting with freshwater (dry dock and haul-out operations). During both winter and summer sampling, survival of soft-bodied organisms tended to be lower in haul-out and dry dock operations than following in-water cleaning. Rates of survival of hard-bodied organisms were generally low in all hull cleaning methods during both sampling seasons.

The proportion of organisms that remained viable following removal from vessel hulls varied considerably among broad taxonomic groups, cleaning operations and sampling season. In all three types of operation (dry dock, haul-out and in-water cleaning), 18–54.8 percent of the total number of organisms and fragments examined as solid samples were viable. No statistically significant differences existed among seasons or operation type. However, when tubicolous polychaetes were excluded from analyses (due to their disproportionate representation in in-water sampling), viability was significantly higher for in-water cleaning operations than for dry dock and haul-out. There was no significant difference in viability between winter and summer sampling with or without inclusion of tubicolous polychaetes.

The proportion of viable organisms in various taxonomic groups examined during winter and summer and in the various operation types varied considerably. It was previously reported (ZBS2002-04) that, during winter, much greater average proportions of bivalves, ascidians, bryozoans, errant polychaetes and sponges remained viable after in-water hull cleaning than after cleaning in dry dock or haul-out operations. In addition, very large proportions of motile molluscs, nemerteans and flatworms were viable following in-water removal. During summer sampling, this pattern of viability did hold for errant polychaetes, sponges, motile molluscs, flatworms and nemerteans, but contrasting results were obtained for other taxa. For example, the mean viability of bivalves sampled in the Lyttelton dry dock was  $79.7 \pm 2\%$  during summer, compared to  $22.1 \pm 19.3\%$  during winter. A similar trend was observed for bryozoans and ascidians. The reasons for this are unknown. Further investigation to elucidate seasonal trends in fouling organism cover and related viability following hull cleaning (for example, time since and season of last clean, time spent in various ports, voyage history, environmental parameters during hull cleaning etc.) may be warranted. It might reasonably be expected that higher air temperatures in summer might reduce bivalve viability, not enhance it. However, there are many environmental- (for example, relative humidity), facility- (for example, degree of trampling caused by different foot traffic underneath vessels which is dependant upon nature of associated vessel maintenance work required), and biotic-related (for example, initial condition of organisms prior to hull cleaning or shielding effects from other organisms) interacting variables which are unknown. For example, for any of the cleaning methods, survival and viability of fouling organisms was generally not a function of the amount of growth (percent cover) on the hulls, indicating that high levels of fouling did not protect individual organisms from being destroyed by the cleaning process. However, there were exceptions as in the case of bryozoan viability which was high on vessels cleaned in the Lyttelton dry dock during the summer season due to a cover of bivalves which shielded the bryozoans from water blasting.

Survival of fragile and delicate organisms appeared affected by the destructiveness of the cleaning method. Removal of biota with a scraper or soft cloth in-water is less destructive for soft-bodied organisms than a water blaster out-of-water, consequently this study found high survival of errant polychaetes, sponges, motile molluscs, flatworms, nematodes and anemones following in-water cleaning. However, for brittle organisms such as tubeworms living on the hull surfaces, few of these organisms survived the cleaning process in any of the operations sampled both in summer and winter sampling. In nearly all cases of viable brittle organisms found following hull cleaning, the only living and viable individuals were growing on other organisms such as barnacles and bivalves. Bivalves generally exhibited high mean rates of viability across all operation types and their presence generally resulted in elevated viability of other taxa that lived on or amongst them and that were protected from the destructive cleaning action. Most motile crustaceans examined in the dry dock, haul-out and in-water cleaning operations during both winter and summer sampling were viable and were generally encountered in protected micro-habitats. The protection of brittle and motile organisms within certain hull structures (for example, sea chests) and associated with other fouling organisms from destructive hull cleaning techniques is of concern and clearly demonstrates the need to access all hull structures that potentially possess fouling organisms and collect **all** fouling material removed during hull cleaning for correct disposal and/or treatment.

In both sampling seasons (winter and summer), average concentrations of intact animals, propagules (that is, eggs, spores and larvae) and unicellular organisms were greatest in the initial runoff from the water blasting (apart from Westpark Marina), which is to be expected. Concentrations of intact propagules in water blast runoff varied significantly between facilities and sampling seasons. For example, during both winter and summer sampling, the concentration of intact animals in water blast runoff was 2.3 (Orams Marine) to 11.7 times higher (at Westpark Marina) than at the Lyttelton dry dock or the Tauranga Marina. Water blast runoff at Orams Marine and the Lyttelton dry dock had a higher concentration of unicellular organisms than that at the other facilities. This may be the result of possible differences in fouling biota between areas around New Zealand where the vessels being cleaned operate in or are maintained in (for example, Auckland compared with Tauranga or Christchurch) as well as possible seasonal differences in fouling composition (for example, hydroids, polychaetes, foraminiferans and dinoflagellates were encountered in summer but not in winter liquid effluent samples) and the physical design of the cleaning areas. For example, at Lyttelton dry dock vessels are floated into the dry dock and the accompanying seawater is then pumped out. Organisms brought in with the seawater which are not also pumped out could then settle on the dry dock floor to be incorporated into the runoff effluent from water blasting.

As system upgrades have been completed at all but Westpark Marina since ZBS2002-04 (winter), some seasonal comparisons of treatment stages were not valid. However, across all facilities, settlement and filtration progressively reduced the mean concentrations of organisms in the liquid effluent. For example, in Lyttelton dry dock, Orams Marine and the Tauranga Marina, samples were taken of the final effluent during winter or summer (it was not possible to sample the Westpark Marina at this effluent treatment stage). In all three locations, concentrations of animals, propagules and unicellular organisms had been reduced by  $\geq 98.5$  percent compared to concentrations observed in the water blast runoff, while filamentous algae had been reduced by a range of 80 to 100 percent. These results demonstrate the benefit in having treatment systems in hull cleaning facilities for removing hull-fouling organisms contained within liquid effluent derived from water blasting vessel hulls. However, variability of treatment effectiveness between facilities was also demonstrated. For example, during winter sampling, the mean concentration of organisms (all types combined) encountered in the final effluent was  $17.1 \pm 6.4/10$  L in the Lyttelton dry dock, and nil at Orams Marine (mean  $\pm$  s.e.).

In this study, the combined use of visual observations (movement) and vital stains (Janus Green) as a proxy for viability where organism movement was not observed, was used for rapid assessment of viability of small organisms or propagules in liquid samples. These techniques

indicated that the majority of organisms discharged from the land-based hull cleaning facilities examined are unlikely to be viable following liquid effluent treatment. However, true determination of whether organisms are actually alive following processing through the liquid effluent treatment can only be achieved through subsequent culture experimentation (for example, hatching of dinoflagellate cysts).

## 4.2 Specific objective 2

As discussed earlier, hull cleaning is an effective method of vector sanitation through the removal of fouling organisms (which may be NIS). As macro-fouling removed from vessel hulls in dry docks and haul-out facilities typically is collected from the ground and disposed of in land-fill it is effectively quarantined. Therefore, it should not represent a NIS invasion-risk. However, raw liquid effluent from water blasting carries a wide variety of marine organisms, which may subsequently pass back into the marine environment post-treatment if the final effluent is discharged to sea, and therefore does pose a quarantine risk. This risk will vary according to the efficacy of any particular liquid effluent treatment system in question.

This efficacy may vary between hull cleaning facilities based on their cleaning practices and work loads. For example, for haul-out facilities, the residency time of effluent passing through settlement tanks is likely to vary between winter and summer because more vessels are cleaned per day during summer than during winter months. Greater volumes of effluent may affect the mortality rate and settling-out of propagules and small organisms as they pass through the treatment system. In this investigation (ZBS2005-22, summer) the relative efficacy of the liquid effluent treatment system at the dry dock and haul-out facilities was assessed based on modelled hydraulic residence times, particle sinking rates and simple salinity tolerances.

The results derived from the modelling demonstrate that the larger the volume of the liquid effluent treatment system in relation to the volume of effluent entering the system, the longer the residence time of liquid effluent from each vessel cleaned. Consequently, the chances of suspended organisms settling are increased, particularly with particles of slower sinking rates. For example, Orams Marine was theoretically more effective than Lyttelton dry dock at settling out organisms/particles with slower sink rates due to its much longer residence time of liquid effluent in its treatment system. Modelling revealed that the hydraulic residence time of effluent treatment systems is also determined by the relationship between total system volume and facility workload. For North Island facilities this equated to a seasonal difference in residence time due to increased workload over the summer period while Lyttelton dry dock maintained a relatively constant workload. Ideally, the liquid effluent treatment systems of hull cleaning facilities should be designed such that residence times are sufficient to settle-out particles with very slow sink rates (that is,  $0.001$  or  $0.0001 \text{ m min}^{-1}$ ).

Most cleaning facilities studied would theoretically kill most pest NIS based on their known salinity tolerances and modelled residence time distributions. However, the Lyttelton dry dock and Westpark Marina with their shorter residence times were likely to be less effective. The longer residence times of effluent in the treatment systems of Orams Marine and Tauranga Marina meant that species, such as the invasive crab *Eriocheir sinensis* and the invasive green mussel *Perna viridis*, which are tolerant to reduced salinity, would theoretically be killed by extended freshwater exposure. Certain freshwater-tolerant organisms such as the bivalve *Potamocorbula amurensis* may be able to survive for long periods within all treatment systems on the basis of salinity alone. There were no seasonal differences in residence time effects with regards to the pest NIS examined.

Our methods for this specific objective were based on modelling and calculations in relation to poorly understood sinking rates and salinity tolerances for various pest NIS. Therefore, the results should be regarded with caution. However, the results obtained in this objective clearly indicate that to maximise the efficacy of hull cleaning effluent treatment systems, treatment systems should

aim to have as high a residence time as possible to facilitate particle settlement. However, within this general recommendation there are confounding treatment system design variables which must be considered. For example, vertical depth of the settling tanks, the use of baffles and weirs, and the use of flocculating or precipitating agents can influence the effectiveness of treatment systems for settling out particles in suspension. In terms of known salinity tolerances of various pest marine NIS, 24–48 hours exposure to a salinity of 0 ppt would appear to an acceptable minimum residence time for killing most species on the basis of simple salinity alone.

Because even facilities with high residence times may not offer 100 percent retention of particles with very slow sink rates (that is, 0.001 or 0.0001 m min<sup>-1</sup>), final fine particle filtering/screening preferably down to a size range of 10–20 µm, but down to 50–60 µm as an acceptable minimum (McClary and Nelligan 2001) is required to minimise the discharge of any surviving organism particularly if final discharge is to the sea.

### **4.3 Consideration of recommendations for commercial hull cleaning facilities**

To assist in determining critical elements for effective hull cleaning and appropriate recommendations for hull cleaning facilities in New Zealand, the findings from Specific Objectives 1 and 2 in this study must be contextualised with regards to what is currently known on the effective treatment of hull-fouling marine NIS through hull cleaning. There is insufficient knowledge of the species-specific freshwater tolerances (and their relationship with other environmental variables) of most marine NIS of concern to be able to provide an informed minimum freshwater exposure time pertinent to hull cleaning facilities that will ensure that 100 percent of marine fouling NIS are killed. As shown in Table 9, some marine NIS are vulnerable to short duration freshwater exposure, while others are extremely resistant over a long period and can even osmoconform. An exposure time of 24–48 hours would appear to be a minimum exposure time to kill most, but not all, of the marine pest NIS considered in this study.

There is also insufficient knowledge of the species-specific sinking rates of part/whole marine NIS of concern to be able to provide an informed safe residence duration or optimal settlement tank design to ensure that 100 percent of marine fouling organisms are settled-out in liquid effluent treatment. If marine hull-fouling NIS survive the initial removal from vessel hulls and can then survive >24–48 hours exposure to freshwater in liquid effluent treatment systems, then either the residence time of the treatment system must be high enough that they are settled-out (that is, designing treatment systems to allow settling-out of particles at a sink rate of 0.001 or 0.0001 m min<sup>-1</sup>), and consequently disposed of in landfill during periodic system cleaning, or they are prevented from being discharged from the treatment system by fine screening. Ensuring adequate residence time is particularly important during periods when cleaning activity is greater (that is, summer when more recreational vessels are typically cleaned per day than in winter) and therefore the residence time within non-recycling treatment systems is reduced. Further research into treatment system design appropriate to hull cleaning facilities, and the marine organisms they are dealing with, to ensure high residence time and optimal particle settling may be warranted.

Freshwater exposure and associated mechanical damage during hull cleaning in out-of-water cleaning, the use of settlement tanks for organism/particle collection, and physical screening are valuable tools for reducing marine hull-fouling organism viability, but each does not stand alone as an effective treatment option. Complete collection and proper disposal of all hull-fouling material through sequential treatment techniques is required to minimise the risk of marine NIS introduction and spread through New Zealand via hull-fouling removed during cleaning in a practicable manner.

Final filtering of treated effluent is desirable because of the ability of some marine organisms to reproduce by very small propagules. For fouling organisms, the physically disruptive nature of water blasting to fouling organisms and the associated freshwater exposure, the exposure to air and trampling by cleaning staff during and following removal, and the further exposure to

freshwater and settlement/treatment processes during the liquid effluent treatment stage all combine to reduce the chances of survival. However, if treated liquid effluent is subsequently discharged directly to the sea then physical screening down to a certain minimum size is still required to prevent the release of intact or fragmented organisms and their gametes/spores from the cleaning facility should they survive the treatment system.

For example, clonal organisms such as the invasive pest *macroalgae Caulerpa fragile* ssp. *tomentosoides* and *Caulerpa taxifolia* can successfully survive fragmentation and re-grow in new suitable habitat (Neill et al. 2006; Wright and Davis 2006), although Schaffelke (pers. comm. in McClary and Nelligan, 2001) considered that 1 mm was the minimum regenerative fragment size for *C. taxifolia*. This regenerative capacity also applies to other clonal/colonial organisms such as bryozoans, echinoderms, hydroids and sponges (Strathmann 1987; Harvell and Helling 1993; Murate et al. 1997; Knott et al. 2006). For example, colonial bryozoans can potentially regenerate an entire colony from a single zooid. As an example of colonial bryozoan zooid size, the colonial bryozoan *Membraniporopsis tubigera*, a recent marine NIS to New Zealand which is capable of creating extensive fouling encrustations, has encrusting zooids 0.34–0.51 mm long and 0.12–0.30 mm wide and erect zooids 0.36–0.58 mm long and 0.15–0.26 mm wide (Gordon et al. 2006).

In a study of hull cleaning facilities in New Zealand, McClary and Nelligan (2001) developed some preliminary technical guidelines for the collection and disposal of the solid matter from vessel hull cleaning. McClary and Nelligan (2001) examined the appropriate and relevant size (for example, adult, spore or gamete size) threshold for 43 provisional hull-fouling target species that could be considered first-order risks of introduction. McClary and Nelligan (2001) visited 37 cleaning facilities around New Zealand, including the five facilities investigated in this study (ZBS2005-22, summer) and that of Floerl et al. (2003) (ZBS2002-04, winter).

McClary and Nelligan (2001) recommended the capture of particles above an average size threshold of 60 µm diameter as an acceptable level of security from marine NIS. For example, this threshold would contain all the mature stages of the 43 target species examined by McClary and Nelligan (2001) as well as the majority of their propagules. However, it would not contain propagules of the pest alga *Undaria pinnatifida* at a size of 10 µm (McClary and Nelligan 2001). McClary and Nelligan (2001) identified gamete/spore size range for their identified risk species as 10–700 µm. McClary and Nelligan (2001) provided tentative costings for various facilities to upgrade their treatment systems to meet the particle capture average size threshold of 60 µm diameter

Filtration with screens or media down to a size of 50 µm should remove most, if not all, zooplankton and smaller screens or media down to a size of 20 µm should remove the hypnocyts of toxic dinoflagellate algae (Oemcke 1999). Fine filtering/screening down to a nominal size of 10–20 µm may be accomplished through a variety of different techniques such as sand filtration, mixed media filters, self-screening screens or cyclonic separators. Removal of particles between 1–10 µm often involves the use of cartridge or diatomaceous earth (DE) filters. According to McClary and Nelligan (2001), the Devonport dry dock's treatment system is capable of reducing particle size to 1 µm.

For each filtering/screening technique there are advantages/disadvantages which impact upon their applicability to particular hull cleaning facilities (Oemcke 1999). Appropriate wastewater-treatment and engineering expertise should be engaged in the design of any new, or modification of any existing liquid effluent treatment systems used in hull cleaning facilities appropriate to the volume of liquid effluent derived at each particular facility to maximise treatment effectiveness. Floerl et al. (2003) reviewed some of the wastewater treatment technology and their associated costs that could be used for treating hull cleaning waste in New Zealand.

At present the evidence for the spread of diseases (viral or bacterial) from small craft is inconclusive but does represent a potential risk (Minchin et al. 2006). If elimination of smaller organisms such as viruses (55–200 nm), bacteria (0.2–5 µm) and some protozoa (2–100 µm) is

desired, then physical disinfection treatment options with broad potential for secondary disinfection are considered to be ultraviolet (UV) irradiation and high power ultrasound and possibly ozonation (Oemcke 1999). For example, UV irradiation is an effective disinfection technique for a wide range of microbial organisms producing significant residual toxicants, although its efficacy relies on relatively low water turbidity (Oemcke 1999).

Various chemical and biocide disinfection treatments could also be used to treat final effluent water before discharge directly to the sea (for example, bromine, copper sulphate, ozone, and sodium hypochlorite). For example, bromine disinfection has been applied to secondary wastewater effluent where it is more effective than chlorine at high pH, and ozone (which can be generated on-site) is considered an excellent disinfectant for the control of resistant organisms (Oemcke 1999). The use of biocides may have potential detrimental effects on other organisms and ecosystems when they are discharged into the marine environment unless applied and managed properly (McClary and Nelligan 2001). Elevated salinity (hypersalinity) is another potential method worthy of consideration for killing marine organisms, but as for hyposalinity the upper salinity threshold that causes damage and death varies considerably for different organisms. For example, *Asterias amurensis* larvae cannot tolerate >42 ppt salinity but the toxic dinoflagellate *Gymnodinium catenatum* is tolerant up to 100 ppt (Oemcke 1999). However, it may be worth investigating these secondary treatment options further, not only for microbial NIS but also for larger NIS contained within liquid effluent treatment systems at hull cleaning facilities.

In this investigation (ZBS2005-22, summer), hull-fouling organisms such as chain diatoms (for example, *Melosira*) with individual cell widths <5 µm, zoospores/propagules around 50 µm along with miscellaneous organic aggregations and inorganic material 50–300 µm were found in final effluent samples in facilities (with multiple settlement tanks and filter systems) whose reduction percentages of hull-fouling organisms was 98–100 percent. Given that there is potential for small marine NIS organisms or propagules to pass through treatment systems of hull cleaning facilities with a number of settlement tanks and filtering/screening mechanisms as we observed, and that our knowledge of salinity tolerances of many marine NIS is poor or unknown, it would seem sensible either not to discharge the treated effluent back into the marine environment at best, or subject the liquid to further treatment such as discharge to a municipal sewage system or by fine screening of particles. Non-discharge of treated effluent could be facilitated by the storage and recycling of treated liquid effluent as the water source for the water blasters used in hull cleaning. The recycling of liquid effluent for reuse (following treatment and removal of suspended materials) in water blasting other vessels, such as is currently carried out by the Tauranga Marina Society, would be appropriate for many recreational marinas and boating clubs. Recycling of treated effluent would also have the benefit of reducing overall facility freshwater consumption. However, it may not be appropriate for very large facilities such as the Lyttelton dry dock or Devonport where large amounts of blasting water are required. For example, the Lyttelton dry dock uses an average 300 m<sup>3</sup> of freshwater for each vessel being cleaned, so unless water used for blasting could be reduced, capital costs required to purchase the required storage tanks may be prohibitive in addition to potentially being limited by available space.

Discharge of treated liquid effluent into a municipal sewage system (as occurs at Orams Marine and Tauranga Marina), specific oxidation ponds or even wetland “polishing” systems would facilitate the further treatment of any discharged treated effluent and may obviate the need for final fine screening of particles down to very small sizes. This currently occurs at some facilities and is largely necessitated by compliance with RMA requirements concerned primarily with containment of heavy metals from anti-fouling and hull painting. This discharge of treated hull cleaning facility effluent into the local sewage system could considerably decrease the *post-hoc* likelihood of hull-fouling organism/propagule survival. This is because any organism/propagule not killed or removed by the hull cleaning facility’s effluent treatment system is further exposed to freshwater, and other, harmful/stressful biological and environmental processes. As the liquid effluent is virtually pure freshwater, discharge of treated liquid effluent (with most or virtually all

suspended material removed) into a municipal sewage system should not disrupt the biological processes of the sewage system. There is the potential that discharge of liquid effluent into a municipal sewage systems, specific oxidation ponds or wetland “polishing” systems may actually remove the need for liquid effluent treatment processes at the hull cleaning facility other than coarse preliminary screening (that is, remove the need for settling tanks and fine filtration/screening). This would be beneficial in reducing plant costs incurred by hull cleaning facilities that need to put in place or upgrade existing liquid effluent treatment to reduce the risk of marine fouling NIS spread. However, to what extent such discharge impacts upon hull-fouling organisms/propagules depends upon the volume and nature of the effluent discharged, the nature of the sewage and wastewater treatment system involved, the residency time within that system before discharge, and where final effluent discharge takes place.

The Tauranga Marina Society haul-out facility, for example, discharges any excess treated effluent to the Chapel Street Wastewater Treatment Plant (WWTP) (20 000 m<sup>3</sup> d<sup>-1</sup> capacity), where it is treated along with other industrial and domestic sewage and wastewater to:

- Pre-treatment: fine-screening, grit/sand removal and pre-aeration of incoming water,
- Primary treatment: separation of floatable and heavy solids for removal,
- Secondary treatment: mechanical aeration to provide oxygen to bacteria and other micro-organisms to break down organic compounds (contact stabilisation). Dead microbial cell material is then settled out in circular clarifier tanks and conveyed to a sludge system,
- Disinfection: using ultra-violet (UV) light irradiation to kill bacteria and other micro-organisms,
- Sludge treatment: using an anaerobic digestion system where organic matter in the sludge is broken down (stabilised) and methane (biogas) is produced.

From the Chapel Street WWTP the processed water is conveyed to a further treatment facility at Te Maunga, passing through two wetlands before it reaches this second facility. At the Te Maunga WWTP (8000 m<sup>3</sup> d<sup>-1</sup> capacity), the processed water is subjected to further treatment as follows:

- Pre-treatment: fine-screening, grit/sand removal and pre-aeration of incoming water,
- Secondary treatment: activated sludge aeration basin and a circular clarifier tank. The aerated activated sludge basin allows beneficial bacteria and other micro-organisms to further purify the wastewater and the resulting biological sludge is settled-out in the clarifier tank (the sludge is collected, de-watered and then disposed of on land),
- Oxidation Pond: in this pond (with around 20 days residency time) the treated wastewater is further purified by natural UV exposure and other natural decay processes,
- Wetland: treated wastewater then passes through a wetland (with around two days residency time),
- Discharge: after the final treatment in wetlands, located at the Te Maunga site, the treated wastewater from both catchments is discharged to the Pacific Ocean through a 950 m long ocean outfall into approximately 12 m of water depth.  
(<http://www.wastewater.tauranga.govt.nz>; accessed 22/10/2006)

Such further treatment of liquid effluent from hull cleaning facilities can only further reduce the chances of hull-fouling organisms surviving.

As found in this investigation (ZBS2005-22, summer) and in ZBS2002-04 (winter), manual in-water hull cleaning represents the highest risk of marine NIS introduction of all the hull cleaning techniques to New Zealand. The attraction of manual in-water hull cleaning to small vessel owners is that it is simple, of minimal or no cost, vessel owners may think that it will extend the lifetime of their antifouling paints, and is often conducted by the vessel owner themselves at their leisure.



However, in both this study (ZBS2005-22, summer) and that of Floerl et al. (2003) (ZBS2002-04, winter), manual in-water hull cleaning was the least effective hull cleaning method for killing or seriously damaging hull-fouling organisms. This has serious implications for the spread of invasive marine organisms as manual in-water hull cleaning typically does not involve the collection and destruction of organisms dislodged from the vessel hull. Invasive organisms dislodged from the hull in a careening bay can potentially survive and establish within the local careening area (Floerl et al. 2005). Mature organisms injured or shocked by the cleaning process may also be induced to release gametes and/or competent larvae through damage or shocking (Environment-and-Natural-Resources-Committee 1997).

Manual cleaning of vessels with scrapers, brushes or cloths, whether in-water or out-of-water may not remove all fouling organisms. However, identifying and removing fouling organisms is inherently easier out-of-water than in-water and fouling organisms (both removed and still attached) are exposed to greater environmental stress (for example, desiccation) during out-of-water manual cleaning. Out-of-water cleaning with water blasters typically results in virtually all fouling organisms being removed from accessible vessel surfaces below the waterline (Woods, pers. obs.). Fouling organisms removed from vessels out-of-water are significantly easier to collect and dispose of compared to most in-water cleaning procedures, particularly manual scraping or underwater blasting using water lances.

Mechanical systems have been developed that will collect the removed organisms. These reduce the risk of organism release where in-water hull cleaning occurs. These systems have been tested on hull-fouling pest species such as *Didemnum vexillum* in New Zealand (Coutts 2002). Such systems may involve vacuum cutting head configurations with filter bags of varying filter sizes for collecting and retaining cleaned solid material. Mechanical systems would be preferable to manual in-water cleaning but may still represent a potentially higher risk of hull-fouling NIS release during cleaning than out-of-water systems. For example, improper suction head adhesion caused by large macro-fouling or vessel hull shape/structures, or suction pipe blockages requiring reverse-flow clearing may allow cleaned organism escape. However, mechanical in-water cleaning systems are worthy of further investigation and improvement.

Based on the results of this study (ZBS2005-22, summer) and that of Floerl et al. (2003) (ZBS2002-04, winter, it is possible to make some recommendations as to appropriate treatment guidelines for the collection and treatment of fouling waste in order to minimise the risk of marine hull-fouling NIS release into the marine environment.

#### 4.4 Recommended guidelines

1. Cleaning of vessels should be conducted out-of-water and in a facility where **all** fouling organisms removed are quarantined from the marine environment (that is, no material removed from vessel hulls should be allowed to aerosol-drift, drain or otherwise move back into the nearby marine environment). Where out-of-water cleaning is not practicable, in-water cleaning should be conducted in such a manner that **all** fouling material removed is collected (ideally down to a particle size of 50–60 µm) and disposed of in landfill as appropriate.
2. **All** macro (>1 mm) material from vessels cleaned out-of-water should be collected and disposed of in landfill as appropriate.
3. **All** liquid effluent (runoff) from out-of-water vessel water blasting/cleaning should be collected and treated in a liquid effluent treatment system prior to discharge or recycling for water blaster use.
4. This effluent should be coarse pre-screened (for example, to 1 mm) before entry into the liquid effluent treatment system. This will reduce inorganic and organic build-up within the treatment system and thus maintain system effectiveness (for example, removal of boundary layer acceleration of suspended particles caused by sediment bed build-up) and extend the period

between maintenance sediment removals. Material caught on the pre-screen should be disposed of in landfill as appropriate.

5. **All** liquid effluent should be processed through multiple settlement tanks to facilitate settling-out of any marine organisms and particles (that is, vessel hull paint flakes). Where practicable, settlement tanks should be of large volume (hydraulic capacity) and of appropriate physical design (for example, use of weirs, baffles etc.) to maximise settlement and allow as long a possible residency time/exposure time of marine organisms to freshwater before progression to a discharge or fine filtering/screening stage. Residence time of effluent water within the treatment system should be a minimum of 24 h, but preferably >48 h. Salinity should be as close to as possible to 0 ppt to achieve 100 percent mortality of most marine organisms. Sedimented material should be regularly removed from settlement tanks and disposed of in landfill as appropriate. Flocculating and precipitating agents which facilitate separation and removal of positively and negatively buoyant particles can be used if they improve the efficiency of the system. The use of diesel/oil absorbing mats may also be appropriate.
6. Following coarse screening and passage through settlement tanks, treated effluent may be wasted to a municipal sewage/wastewater system or similar extensive freshwater treatment system for additional treatment rather than direct discharge to sea. This wasting to a municipal sewage/wastewater system (dependant upon relevant council restrictions) should further reduce marine organism viability by increasing residence time within freshwater as well as exposing any organisms to other biological and physical treatment processes and contaminants which may kill them (depending upon the nature of the waste treatment system in question).
7. Where discharge of treated effluent will be directly to the sea, following processing in settlement tanks, **all** liquid effluent should be fine filtered/screened, preferably to a size range of 10–20  $\mu\text{m}$ , but 50–60  $\mu\text{m}$  is an acceptable minimum to remove the smallest of most types of marine organisms before discharge.
8. As an alternative to discharge of treated effluent to the sea or sewage system, treated liquid effluent could be stored and then recycled for water blasting other vessels rather than discharged. This theoretically increases the residence time of any remaining marine organisms in freshwater (and thereby reduces their chances of survival) and reduces total freshwater usage by the cleaning facility.

## Acknowledgements

This study would not have been possible without the willing co-operation of the staff from the cleaning facilities involved in the study. We thank the following people for their time, patience and assistance: Hal Upton (Lyttelton dry dock), Craig Park (Orams Marine Maintenance), Kevin Lidgard (Westpark Marina), Bob Ellis and Andrew (Tauranga Marina Society), and Tom Warren (Gulf Harbour Marina). All these people recognise the potential impacts associated with their activities and work hard to mitigate these potential impacts. We also thank Barb Hayden, Don Robertson and John Zeldis (NIWA), Liz Jones and Andrew Bell (MAF Biosecurity New Zealand) for their reviews of this report.

## References

- Allredge, A.L.; Gotschalk, C. (1988). In situ settling behaviour of marine snow. *Limnology and Oceanography* 33(3): 339-351.
- AMOG-Consulting. (2002). Hull fouling as a vector for the translocation of marine organisms. Phase I: Hull fouling research. *Ballast Water Research Series*, Report No. 14. Department of Agriculture, Fisheries and Forestry Australia, Canberra.
- Anderson, D.M.; Lively, J.J.; Reardon, E.M.; Price, C.A. (1985). Sinking characteristics of dinoflagellate cysts. *Limnology and Oceanography* 30(5): 1000-1009.
- Anger, K. (1991). Effects of temperature and salinity on the larval development of the Chinese mitten crab *Eriocheir sinensis* (Decapoda: Grapsidae). *Marine Ecology Progress Series* 72: 103-110.
- Asper, V.L. (1987). Measuring the flux and sinking speed of marine snow aggregates. *Deep-Sea Research* 34: 1-17.
- Band-Schmidt, C.J.; Morquecho, L.; Lechuga-Deveze, C.H.; Anderson, D.M. (2004). Effects of growth medium, temperature, salinity and seawater source on the growth of *Gymnodinium catenatum* (Dinophyceae) from Bahia Concepcion, Gulf of California, Mexico. *Journal of Plankton Research* 26(12): 1459-1470.
- Callieri, C. (1997). Sedimentation and aggregate dynamics in Lake Maggiore, a large, deep lake in Northern Italy. *Mem. Ist. ital. Idrobiol.* 56: 37-50.
- Carlton, J.T. (1989). Man's role in changing the face of the ocean: biological invasions and implications for conservation of nearshore environments. *Conservation Biology* 3: 265-273.
- Chu, F.L.E.; Volety, A.; Constantin, G. (1996). A comparison of *Crassostrea gigas* and *Crassostrea virginica* - effects of temperature and salinity on susceptibility to the protozoan parasite, *Perkinsus marinus*. *Journal of Shellfish Research* 15: 375-380.
- Cieluch, U.; Anger, K.; Aujoulat, F.; Buchholz, F.; Charmienter-Daures, M.; Charmienter, G. (2004). Ontogeny of osmoregulatory structures and function in the green crab *Carcinus maenus* (Crustacea, Decapoda). *The Journal of Experimental Biology* 207: 325-336.
- Clark, C. (1973). Staining procedures. Williams and Wilkins, Baltimore, USA.
- Cohen, A.N. (2005). *Watersipora subtorquata*. Guide to the Exotic Species of San Francisco Bay. San Francisco Estuary Institute, Oakland, CA. <<http://www.exoticguide.org>> accessed 13/10/2006.
- Cohen, A.N.; Carlton, J.T.; Fountain, M.C. (1995). Introduction, dispersal and potential impacts of the green crab *Carcinus maenus* in San Francisco Bay, California. *Marine Biology* 122: 225-237.
- Coutts, A. (2002). The development of incursion response tools - underwater vacuum and filter system trials. Cawthron Report No. 755. Report prepared for New Zealand Diving and Salvage Ltd. 27p.
- Coutts, A.; Forrest, B. (2005). Evaluation of eradication tools for the clubbed Tunicate *Styela clava*. Cawthron Report No. 110. 48p.
- Coutts, A.D.M. (1999). Hull fouling as a modern vector for marine biological invasions: investigation of merchant vessels visiting northern Tasmania. M.App.Sc thesis, Australian Maritime College, Tasmania. 283p.
- Cranfield, H.J.; Gordon, D.J.; Willan, R.C.; Battershill, C.N.; Francis, M.P.; Nelson, W.A.; Glasby, C.J.; Read, G.B. (1998). Adventive marine species in New Zealand. NIWA Technical Report No. 34. 56p.

- Creese, R.G.; Davis, A.R.; Glasby, T.M. (2004). Eradicating and preventing the spread of the invasive alga *Caulerpa taxifolia* in NSW. *NSW Fisheries Final Report Series*. 64. 124p.
- Currie, D.R.; McArthur, M.A.; Cohen, B.F. (2000). Reproduction and distribution of the invasive European fanworm *Sabella spallanzanii* in Port Phillip Bay, Australia. *Marine Biology* 136: 645-656.
- Environment-and-Natural-Resources-Committee. (1997). Report on Ballast Water and Hull Fouling in Victoria. Victorian Government Printer, Melbourne, Australia. 220p.
- Floerl, O. (2002). Intracoastal spread of fouling organisms by recreational vessels. PhD thesis, James Cook University, Townsville. 295p.
- Floerl, O.; Inglis, G. (2003). Boat harbour design can exacerbate hull fouling. *Austral Ecology* 28: 116-127.
- Floerl, O.; Inglis, G. (2005). Starting the invasion pathway: the interaction between source populations and human transport vectors. *Biological Invasions* 7: 589-606.
- Floerl, O.; Inglis, G.J.; Marsh, H.M. (2005). Selectivity in vector management: an investigation of the effectiveness of measures used to prevent transport of non-indigenous species. *Biological Invasions* 7: 459-475.
- Floerl, O.; Norton, N.; Inglis, G.; Hayden, B.; Middleton, C.; Smith, M.; Alcock, N. (2003). An investigation of hull cleaning and associated waste treatment options for preventing the spread of non-indigenous marine species. NIWA Client Report for Ministry of Fisheries Project ZBS2002-04. 90p.
- Floerl, O.; Pool, T.K.; Inglis, G.J. (2004). Positive interactions between nonindigenous species facilitate transport by human vectors. *Ecological Applications* 14(6): 1724-1736.
- Forward, R.B.J.; Cronin, T.W.; Stearns, D.E. (1984). Control of diel vertical migration: photoreponses of a larval crustacean. *Limnology and Oceanography* 29(1): 146-154.
- Foxon, G.E.H. (1934). Notes on the swimming methods and habits of certain crustacean larvae. *Journal of the Marine Biological Association of the United Kingdom* 19(2): 829-850.
- Fuchs, H.L.; Mullineaux, L.S.; Solow, A.R. (2004). Sinking behaviour of gastropod larvae (*Ilyanassa obsoleta*) in turbulence. *Limnology and Oceanography* 49(6): 1937-1948.
- Ghosal, S.; Rogers, M.; Wray, A. (2000). The turbulent life of phytoplankton. Center for Turbulence Research. Proceedings of the Summer Program 2000: 31-45.
- Gordon, D.J.; Ramalho, L.V.; Taylor, P.D. (2006). An unreported invasive bryozoan that can affect livelihoods - *Membraniporopsis tubigera* in New Zealand and Brazil. *Bulletin of Marine Science* 78(2): 331-342.
- Granata, T.C. (1991). Diel periodicity in growth and sinking rates of the centric diatom *Coscinodiscus concinnus*. *Limnology and Oceanography* 36(1): 132-139.
- Gunthorpe, L.; Mercer, J.; Rees, C.; Theodoropoulos, T. (2001). Best practices for sterilisation of aquaculture farming equipment: a case study for mussel ropes. *Marine and Freshwater Resources Institute Report* No. 41. 48p.
- Hamm, C.E. (2002). Interactive aggregation and sedimentation of diatoms and clay-sized lithogenic material. *Limnology and Oceanography* 47(6): 1790-1795.
- Harvell, C.D.; Helling, R. (1993). Experimental induction of localized reproduction in a marine bryozoan. *Biological Bulletin* 184: 286-295.
- Hayakawa, Y. (1987). Deterministic model for marine algal population density. *Nippon Suisan Gakkaishi* 53(7): 1159-1166.

- Hayes, K.; Sliwa, C.; Migus, S.; McEnulty, F.; Dunstan, P. (2005). *National priority pests: Part II. Ranking of Australian marine pests*. Australian Government Department of Environment and Heritage. 102p.
- Hewitt, C.L. (2002). Distribution and biodiversity of Australian tropical marine bioinvasions. *Pacific Science* 56(2): 213-222.
- Hopkins, A.E. (1936). Adaptation of the feeding mechanism of the oyster (*Ostrea gigas*) to changes in salinity. *Bulletin of the Bureau of Fisheries* 48: 345-364.
- Hurlbert, S.H. (1984). Pseudoreplication and the design of ecological field experiments. *Ecological Monographs* 54(2): 187-211.
- Jackson, A. (2005). *Ciona intestinalis*. A sea squirt. Marine Life Information Network: Biology and Sensitivity Key Information Sub-Programme [online]. <<http://www.marlin.ac.uk/species/Cionaintestinalis.htm>> accessed 13/10/2006.
- James, P.; Hayden, B. (2000). The potential for the introduction of exotic species by vessel hull fouling: a preliminary study. NIWA Client Report No. WLG00/51. 66p.
- Kamer, K.; Fong, P. (2000). A fluctuating salinity regime mitigates the negative effects of reduced salinity on the estuarine macroalga, *Enteromorpha intestinalis* (L.) Link. *Journal of Experimental Marine Biology and Ecology* 254(1): 53-69.
- Knott, N.A.; Underwood, A.J.; Chapman, M.G.; Glasby, T.M. (2006). Growth of the encrusting sponge *Tedania anhelans* (Lieberkuhn) on vertical and on horizontal surfaces of temperate subtidal reefs. *Marine and Freshwater Research* 57: 95-104.
- Knutsen, T.; Melle, W.; Calise, L. (2001). Determining the mass density of marine copepods and their eggs with a critical focus on some of the previously used methods. *Journal of Plankton Research* 23(8): 859-873.
- Krug, P.J.; Zimmer, R.K. (2004). Developmental Dimorphism: consequences for larval behaviour and dispersal potential in a marine gastropod. *Biological Bulletin* 207: 233-246.
- Lane, D.J.W.; Beaumont, A.R.; Hunter, J.R. (1985). Byssus drifting and the drifting threads of the young post-larval mussel *Mytilus edulis*. *Marine Biology* 84: 301-308.
- Lee, J.-S.; Lee, K.-T.; Kim, C.-K.; Lee, J.-H.; Park, K.H.; Park, G.-S. (2005). Applications of indigenous benthic amphipods as sediment toxicity testing organisms. *Ocean Science Journal* 40(1): 17-24.
- Lewis, J.A.; Watson, C.; ten Hove, H.A. (2006). Establishment of the Caribbean serpulid tubeworm *Hydroides sanctaecrucis* Kroyer (in) Morch, 1863, in northern Australia. *Biological Invasions* 8: 665-671.
- Luckenbach, M.W.; Orth, R.J. (1992). Swimming velocities and behavior of Blue crab (*Callinectes sapidus* Rathbun) megalopae in still and flowing water. *Estuaries* 15(2): 186-192.
- Lutzen, J. (1999). *Styela clava* Herdman (Urochordata, Ascidiacea), a successful immigrant to North-west Europe: ecology, propagation and chronology of spread. *Helgolander Meeresuntersuchungen* 52: 383-391.
- Mack, R.N.; Simberloff, D.; Lonsdale, W.M.; Evans, H.; Clout, M.; Bazzaz, F.A. (2000). Biotic invasions: causes, epidemiology, global consequences, and control. *Ecological Applications* 10: 689-710.
- Martins, I.; Oliveira, J.M.; Flindt, M.R.; Marques, J.C. (1999). The effect of salinity on the growth rate of the macroalgae *Enteromorpha intestinalis* (Chlorophyta) in the Mondego estuary (west Portugal). *Acta Oecologica*. 20(4): 259-265.

- Mawatari, S. (1951). The natural history of a common fouling bryozoan, *Bugula neritina*. *Miscellaneous Reports of the Research Institute for Natural Resources Tokyo* 19-21.: 47-54.
- McClary, D.J.; Nelligan, R.J. (2001). Alternate biosecurity management tools for vector threats: technical guidelines for acceptable hull cleaning facilities. Kingett Mitchell & Associates Ltd. Report prepared for the New Zealand Ministry of Fisheries (Project ZBS2000/03). 29p.
- McDonald, K. (2004). Patterns in early embryonic motility: effects of size and environmental temperature on vertical velocities of sinking and swimming echinoid blastulae. *Biological Bulletin* 207: 93-102.
- McEnnulty, F.; Jones, T.E.; Bax, N.J. (2001). The Web-based rapid response toolbox. Web Publication: <<http://crimp.marine.csiro.au/NIMPIS/controls.htm>>. Date of release: June 2001, Date of access: 13/06/2002.
- McHenry, M.J. (2001). Mechanisms of helical swimming: asymmetries in the morphology, movement and mechanics of larvae of the ascidian *Distaplia occidentalis*. *The Journal of Experimental Biology* 204: 2959-2973.
- Mills, A.; Fish, J.D. (1980). Effects of salinity and temperature on *Corophium volutator* and *C. arenarium* (Crustacea: Amphipoda), with particular reference to distribution. *Marine Biology* 58: 153-161.
- Minchin, D.; Floerl, O.; Savini, D.; Occhinipint-Ambrogi, A. (2006). Small craft and the spread of exotic organisms. In: *The ecology of transportation: managing mobility for the environment* (eds. Davenport, J.; Davenport, J.L.). Springer, The Netherlands: 99-118.
- Minchin, D.; Gollasch, S. (2003). Fouling and ships' hulls: how changing circumstances and spawning events may result in the spread of exotic species. *Biofouling* 19 (Supplement): 111-122.
- Miyawaki, D.; Sekiguchi, H. (1999). Interannual variation of bivalve populations on temperate tidal flats. *Fisheries Science* 65(6): 817-829.
- Murate, M.; Kishimoto, Y.; Sugiyama, T.; Fujisawa, T.; Takahashi-Iwanaga, H.; Iwanaga, T. (1997). Hydra regeneration from recombined ectodermal and endodermal tissue. II. Differential stability in the ectodermal and endodermal epithelial organization. *Journal of Cell Science* 110: 1919-1934.
- Neill, P.E.; Alcalde, O.; Faugeron, S.; Navarrete, S.A.; Correa, J.A. (2006). Invasion of *Codium fragile* ssp. *tomentosoides* in northern Chile: a new threat for *Gracilaria* farming. *Aquaculture* 259: 202-210.
- Nell, J.A.; Gibbs, P.J. (1986). Salinity tolerance and absorption of L-Methionine by some Australian bivalve molluscs. *Australian Journal of Marine and Freshwater Research* 37: 721-727.
- Nicolini, M.H.; Penry, D.L. (2000). Spawning, fertilization, and larval development of *Potamocorbula amurensis* (Mollusca: Bivalvia) from San Francisco Bay, California. *Pacific Science* 54(4): 377-388.
- NIMPIS. (2002a). *Asterias amurensis* habitat and survival. National Introduced Marine Pest Information Service (eds. Hewitt, C.L.; Martin, R.B. Sliwa, C.; McEnnulty, F.R.; Murphy, N.E.; Jones, T.; Cooper, S.). Web publication <<http://crimp.marine.csiro.au/nimpis>>, date of access 19/9/2006.
- NIMPIS. (2002b). *Caulerpa taxifolia* habitat and survival. National Introduced Marine Pest Information Service (eds. Hewitt, C.L.; Martin, R.B. Sliwa, C.; McEnnulty, F.R.; Murphy, N.E.; Jones, T.; Cooper, S.). Web publication <<http://crimp.marine.csiro.au/nimpis>>, date of access 19/9/2006.
- Noji, T.T.; Bathmann, U.V.; von Bodungen, B.; Voss, M.; Antia, A.; Krumbholz, M.; Klein, B.; Peecken, I.; Noji, C.I.-M.; Rey, F. (1997). Clearance of picoplankton-sized particles and formation of



- rapidly sinking aggregates by the pteropod, *Limacina retroversa*. *Journal of Plankton Research* 19(7): 863-875.
- Oakley, J.A. (2005). *Caprella mutica*. Japanese skeleton shrimp. Marine Life Information Network: Biology and Sensitivity Key Information Sub-Programme [on-line]. <[http://www.marlin.ac.uk/marine\\_allies/species/Caprellamutica.htm](http://www.marlin.ac.uk/marine_allies/species/Caprellamutica.htm)> accessed 13/10/2006.
- Oemcke, D. (1999). The treatment of ships' ballast water. EcoPorts Monograph Series No. 18 (Ports Corporation of Queensland, Brisbane). 102p.
- Ouellet, P.; Allard, J.-P. (2006). Vertical distribution and behaviour of shrimp *Pandalus borealis* larval stages in thermally stratified water columns: laboratory experiments and field observations. *Fisheries Oceanography* 15(5): 373-389.
- Peperzak, L.; Colijn, F.; Koeman, R.; Gieskes, W.W.C.; Joordens, J.C.A. (2003). Phytoplankton sinking rates in the Rhine region of fresh water influence. *Journal of Plankton Research* 25(4): 365-383.
- Prakash, A. (1967). Growth and toxicity of a marine dinoflagellate, *Gonyaulax tamarensis*. *Journal of the Fisheries Research Board of Canada* 24(7): 1589-1606.
- Reusch, T.B.H.; Williams, S.L. (1998). Variable responses of native eelgrass *Zostera marina* to a non-indigenous bivalve *Musculista senhousia*. *Oecologia* 113: 428-441.
- Riebsell, U. (1992). The formation of large marine snow and its sustained residence in surface waters. *Limnology and Oceanography* 37(1): 63-76.
- Ruiz, G.M.; Fofonoff, P.F.; Carlton, J.T.; Wonham, M.J.; Hines, A.H.; Cohen, A.N. (2000). Invasion of coastal marine communities in North America: patterns and processes. *Annual Review of Ecology and Systematics* 31: 481-531.
- Segnini de Bravo, M.I.; Chung, K.S.; Perez, J.E. (1998). Salinity and temperature tolerances of the green and brown mussels, *Perna viridis* and *Perna perna* (Bivalvia, Mytilidae). *Revista de Biologia Tropical* 46(5): 121-126.
- Shafee, M.S. (1976). Effect of salinity and time of exposure to air on the metabolism of green mussel, *Mytilus viridis* L. *Indian Journal of Marine Sciences* 5(1): 130-132.
- Shanks, A.L.; Edmondson, E.W. (1990). The vertical flux of metazoans (holoplankton, meiofauna, and larval invertebrates) due to their association with marine snow. *Limnology and Oceanography* 35(2): 455-463.
- Siu, G.K.; Young, M.L.C.; Chan, D.K.O. (1997). Environmental and nutritional factors which regulate population dynamics and toxin production in the dinoflagellate *Alexandrium catenella*. *Hydrobiologia* 352: 117-140.
- Smayda, T.J.; Boleyn, B.J. (1965). Experimental observations on the flotation of marine diatoms. I. *Thalassiosira cf. nana*, *Thalassiosira rotula* and *Nitzschia seriata*. *Limnology and Oceanography* 10(4): 499-509.
- Strathmann, M.F. (1987). Reproduction and development of marine invertebrates of the Northern Pacific coast. University of Washington Press, USA: 670p.
- Su, H.-M.; Chiang, Y.M.; Liao, I.C. (1993). Role of temperature, salinity and ammonia on the occurrence of the Taiwanese strain of *Alexandrium tamarensis*. In: *Toxic phytoplankton in the sea* (eds. Smayda, T.J.; Shimizu, Y.). Elsevier Science Publishers, B.V. Amsterdam: 837-842.
- Taylor, M.D.; MacKenzie, L.A. (2001). Delimitation survey of the toxic dinoflagellate *Gymnodinium catenatum* in New Zealand. Cawthron Report no. 661. Prepared for Ministry of Fisheries. 12p.



- Thornton, D.C.O. (2002). Diatom aggregation in the sea: mechanisms and ecological implications. *European Journal of Phycology* 37: 149-161.
- Titelman, J.; Kiorboe, T. (2003). Motility of copepod nauplii and implications for food encounter. *Marine Ecology Progress Series* 247: 123-135.
- Titman, D.; Kilham, P. (1976). Sinking in freshwater phytoplankton: some ecological implications of cell nutrient status and physical mixing process. *Limnology and Oceanography* 21(3): 409-417.
- Trowbridge, C.D. (1999). An assessment of the potential spread and options for control of the introduced green macroalga *Codium fragile* ssp. *tomentosoides* on Australian shores. CSIRO Marine Research Report. 42p.
- Underwood, A.J. (1997). Experiments in ecology: their design and interpretation using analysis of variance. Cambridge, UK, Cambridge University Press.
- van Leeuwen, H.C.; Maly, E.J. (1991). Changes in swimming behavior of male *Diaptomus leptopus* (Copepoda: Calanoida) in response to gravid females. *Limnology and Oceanography* 36(6): 1188-1195.
- Waite, A.M.; Safi, K.A.; Hall, J.A.; Nodder, S.D. (2005). Mass sedimentation of picoplankton embedded in organic aggregates. *Limnology and Oceanography* 45(1): 87-97.
- Wallentius, I. (1999). Exotics of the ocean - *Undaria pinnatifida* (Harvey) Suringar, 1872. In: Exotics across the oceans: Case histories on introduced species: their biology, distribution, range expansion and impact (eds. Gollasch, S.; Minchin, D.; Rosenthal, H.; Voigt, M.). Department of Fisheries Biology, Institute for Marine Science, University of Kiel, Germany.: 11-19.
- Wang, G.; Jiang, X.; Wu, L.; Li, S. (2005). Differences in the density, sinking rate and biochemical composition of *Centropages tenuiremis* (Copepoda: Calanoida) subitaneous and diapause eggs. *Marine Ecology Progress Series* 288: 165-171.
- Winet, H. (1973). Wall drag on free-moving ciliated micro-organisms. *Journal of Experimental Biology* 59: 753-766.
- Wonham, M.J.; Carlton, J.T. (2005). Trends in marine biological invasions at local and regional scales: the Northeast Pacific Ocean as a model system. *Biological Invasions* 7: 369-392.
- Wonham, M.J.; Walton, W.C.; Ruiz, G.M.; Frese, A.M.; Galil, B.S. (2001). Going to the source: role of the invasion pathway in determining potential invaders. *Marine Ecology Progress Series* 215: 1-12.
- Wright, J.T.; Davis, A.R. (2006). Demographic feedback between clonal growth and fragmentation in an invasive seaweed. *Ecology* 87(7): 1744-1754.
- Zirbel, M.J.; Veron, F.; Latz, M.I. (2000). The reversible effect of flow on the morphology of *Ceratocorys horrida* (Peridinales, Dinophyta). *Journal of Phycology* 36: 46-58.

# Appendix

## Appendix 1: Guidelines used by NIWA field staff to assess viability of fouling biota removed from vessel hulls.

	Indicators for live and viable individuals/colonies	Indicators for non-viability of individuals/colonies
1. SESSILE TAXA		
Barnacles	<p>Structure: All shell plates present and intact, opercular plates present (acorn barnacles only – gooseneck barnacles have no opercular plates).</p> <p>Feeding/movement: Feeding structures (cirri) protrude out of the test and perform sweeping feeding movements. Or opercular shells closed by muscular action.</p>	<p>Structure: Shell/opercular plates and/or feeding structures (cirri) broken or missing.</p> <p>Feeding/movement: Feeding structures visible but motionless and slack and/or no reaction when poked.</p>
Bivalves	<p>Structure: Both shells present and intact.</p> <p>Feeding/movement: Shells may be locked by muscular action (that is, this bivalve lives). Shells may also be open (feeding), exposing mantle tissue and siphons (or gaps in mantle), but will close when poked (reaction).</p>	<p>Structure: One shell missing or one/both shells cracked or fragmented.</p> <p>Feeding/movement: Shells open but no reaction to touch.</p>
Encrusting bryozoans	<p>Structure: Colony/fragment contains several intact zooids (check for animal inside against light).</p> <p>Feeding/movement: Filtering apparatus (lophophore) protrude through opening in zooid.</p>	<p>Structure: All zooids damaged/smashed, no soft tissues visible. And/or: all colonies dried out, loss of all moisture. And/or loss of pigmentation.</p> <p>Feeding/movement: Zooids' soft tissues and/or feeding structures may be visible but no movement or reaction to touch.</p>
Erect bryozoans	<p>Structure: Colony/fragment contains several intact zooids (check for animal inside against light).</p> <p>Feeding/movement: Filtering apparatus (lophophore) protrude through opening in zooid.</p>	<p>Structure: All zooids damaged/smashed, no soft tissues visible. And/or: all colonies dried out, loss of all moisture.</p> <p>Feeding/movement: Feeding structures may be visible but no movement or reaction to touch.</p>
Colonial ascidians	<p>Structure: Colony/fragment in reasonable 'shape', moist to the touch (not dried) and not entirely</p>	<p>Structure: Shredded or crushed so that badly damaged. No polyps visible (polyps may have</p>

	<p>crushed. Several polyps intact.</p> <p>Feeding/movement: Inhalant and/or exhalant siphons open but close when poked.</p>	<p>‘popped out’ from mechanical pressure on colony). And/or colony dried out, loss of all moisture.</p> <p>Feeding/movement: Siphons open but no reaction to touch.</p>
Solitary ascidians	<p>Structure: Test (body) intact, no holes or gashes, not crushed flat or severely deformed. Moist, not dried.</p> <p>Feeding/movement: Inhalant and/or exhalant siphons open but close when poked (reaction).</p>	<p>Structure: Test badly damaged, crushed or deformed. Branchial basket exposed and/or damaged, guts hanging out. And/or colony dried out, loss of all moisture.</p> <p>Feeding/movement: Siphons open but no reaction to touch.</p>
Hydroids	<p>Structure: Body reasonably intact, feeding polyps (often at distal ends of braches) present.</p> <p>Feeding/movement: Feeding tentacles exposed.</p>	<p>Structure: All polyps damaged/smashed. And/or colony dried out, loss of all moisture.</p> <p>Feeding/movement: Feeding structures may be visible but no movement or reaction to touch.</p>
Tubicolous polychaetes	<p>Structure: Intact (body within tube), not crushed, no holes or gashes.</p> <p>Feeding/movement: Worm retracts into tube when poked (reaction), and/or feeding structures (tentacular crown) visible and moving.</p>	<p>Structure: Tube missing, loss of tentacular crown, body badly crushed or lacerated. And/or dried out, loss of all moisture.</p> <p>Feeding/movement: Feeding structures may be visible but no movement or reaction to touch.</p>
Sponges (assessment of viability very difficult or impossible)	<p>Structure: Fragments retain natural colour, firm texture (don’t fall apart). Sponges retain a “fleshy/translucent/shiny” appearance. Look for “translucent” tissue between fibres</p> <p>Feeding/movement: Impossible to observe.</p>	<p>Structure: Colony/fragment faded and bleached, falling apart. Sponge a mass of golden fibres/hair-like structures without “translucent fleshy tissue” between the fibres. And/or colony dried out, loss of all moisture. Usually no chance for survival if removed from water for more than 3 hours.</p> <p>Feeding/movement: Impossible to observe.</p>
Macroalgae	<p>Structure: Contain pigment and have natural colour. Dryness often not a good indicator as some species are intertidal. Look out for and preserve reproductive structures.</p> <p>Feeding/movement: n/a</p>	<p>Structure: Badly crushed, fragmented, or faded (loss of pigments).</p> <p>Feeding/movement: n/a</p>

2. MOTILE TAXA		
Crabs	Visible movement / reaction. Eyes/sensory organs in head region moving. Missing limbs no problem unless all are gone. Carapace intact.	All limbs or both pincers missing. Carapace damaged (e.g. large holes or parts missing). No movement / reaction to touch. Loss of moisture - dried out.
Molluscs (gastropods, sea slugs, chitons)	Body intact (gastropod snails: shell present), reaction to touch.	Body damaged, crushed or lacerated. No movement/reaction to touch. Loss of moisture - dried out.
Seastars/brittlestars	Basal disc or parts of it present (can regenerate from that), body (or whatever's present) has natural shape, not crushed.	Arm only without part of basal disc (can't regenerate), body damaged, crushed or lacerated. No movement/reaction to touch.
Amphipods/Isopods	Visible movement/reaction, especially feeding limbs will beat if submerged and alive. Missing limbs no problem unless all are gone. Carapace intact.	All limbs or feeding structures missing. Carapace damaged (e.g. large holes or parts missing). No movement/reaction to touch. Loss of moisture - dried out.