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Recommended citation:

Delacroix S, Vogelsang C, Tobiesen A, Liltved H. 2013. Disinfection by-products and ecotoxicity of ballast water after oxidative treatment - Results and experiences from seven years of full-scale testing of ballast water management systems. *Marine Pollution Bulletin* 73(1):24-36.

Disinfection by-products and ecotoxicity of ballast water after oxidative treatment – results and experiences from seven years of full-scale testing of ballast water management systems

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Abstract

Since 2005, five different ballast water management systems (BWMS) based on chlorination treatment have been tested by Norwegian Institute for Water Research (NIVA) according to guidelines from the International Maritime Organization (IMO). 25 % and >50 % of all the tested discharge samples exhibited acute and chronic toxic effects on algae, respectively. In most cases this toxicity was plausibly caused by a high free residual oxidant (FRO) level (>0.08 mg Cl/l). Of the 22 disinfection by-products (DBPs) that were identified in treated water at discharge, four compounds were at times found at concentrations that may pose a risk to the local aquatic environment. However, there seemed to be no clear indication that the measured DBP concentrations contributed to the observed algal toxicity. The addition of methylcellulose instead of lignin in the test water to comply with IMO requirements seemed to limit the formation of DBP.

Keywords: Ballast water treatment, disinfection by-product, oxidant, toxicity, full-scale testing.

1. Introduction

More than 80 percent of world's commercial products are transported overseas by ships also carrying between 3 and 5 billion tons of ballast water around the world each year (Globallast, 2000). An estimated 7,000 different marine and coastal species are transported as stowaways across the world's oceans every day in ballast water (Carlton, 1999), and 84% of the world's marine ecoregions have already reported findings of so-called "invasive marine species" (WWF, 2009). The discharge of ship's ballast water has been recognized as a major vector for invasive aquatic species spreading into new environments (Ruiz et al., 1997). Invasive marine species discharged into a new environment may threaten the native ecological balance, affect local economic activities such as fisheries, and even cause human fatalities. For example, the European zebra mussel has infested over 40% of internal waterways in USA (Globallast, 2000). By invading and clogging water intake pipes, water filtration and electric generating plants, the mussel might cause 1 billion dollars cost per year (Pimentel, 2005).

In order to address this challenge, the International Maritime Organisation (IMO) has adopted the international convention for the control and management of ships ballast water and sediments (IMO, 2004). This convention requires that ballast water quality shall meet strict standards regarding number of viable organisms and residual toxicity at the time of discharge.

An estimated number of 57,000 maritime vessels will have to install a type-approved ballast water management system (BWMS) by the end of 2020 if the convention is finally ratified (Royan, 2010). The convention will come into force 12 months after 30 countries representing 35% of the world merchant shipping tonnage have ratified it. To date, 37 countries representing 29% of the world merchant tonnage have signed.

According to Lloyd's register, a total of 68 different ballast water treatment systems were available to serve this market in September 2012. Of these, 21 systems apply UV irradiation as the main disinfection process, 23 systems apply electrochlorination by electrolysis of saline water, 6 are based on ozonation, 5 apply deoxygenation, 3 apply chlorination using a chlorine containing solution and 10 other technologies are applying heating or non-chlorine chemical disinfection. This means that 30% of the technologies are based on UV treatment, while 45% are using chlorine as the active substance.

If active substances are used as disinfectant, IMO requires the manufacturers to document the potential toxicity risk of treated ballast water to the aquatic environment and to human health, including the ship crew and swimmers (IMO 2008c, IMO 2008d). According to the guidelines, all BWMS should be tested at a land-based testing facility by an independent part using at least two different test water types (seawater, brackish water and/or freshwater) with elevated content of dissolved and particulate organic matter, and a minimum of five test cycles should be conducted with each water type. Both ecotoxicological tests and chemical assessment of DBPs in ballast water at discharge have to be included.

The chemistry of chlorinated fresh water is very different to the chemistry of chlorinated seawater and brackish water. In fresh water, applied chlorine will be hydrolyzed into hypochlorous acid (HOCl) and hypochlorite ion (OCl⁻), which are the main active substances and will co-exist in a pH dependent equilibrium. By introducing chlorine to seawater systems, a series of redox-reactions take place, and several reactive intermediates are formed. Chlorine can rapidly oxidize bromide ion (Br⁻) and iodide ion (I⁻) to form aqueous bromine (HOBr/OBr⁻) and aqueous iodine (HOI/OI⁻), respectively (Westerhoff et al. 2004). The bromide ion, in concentrations of 60-70 mg/l in seawater, gives a high formation potential of bromine (HOBr/OBr⁻) which is the main active substances in chlorinated seawater. In seawater with a typical pH of 8, hypobromous acid (HOBr) will predominate and be the most important disinfectant with a half-life of hours to days dependent on light conditions and water quality characteristics (Liltved et al. 2006).

Ballast water contains various amounts of natural organic substances that, dependent on local conditions, may be oxidized to halogenated organic compounds such as trihalomethanes (THMs) and other disinfection by-products (DBPs). The DBPs most frequent found in chlorinated seawater are bromoform, dibromoacetic acid, bromoacetonitrile and traces of bromophenols (Fabbricino et al., 2005; Bowmer et al., 2010; Shi et al., 2013). Several DBPs might be harmful to aquatic animals and humans because of their potential carcinogenic and mutagenic effects (Richardson et al., 2007), and are regulated in drinking water and bathing water standards (WHO, 2011, WHO, 2003). Some DBPs can be persistent in the marine environment and may bio-accumulate in food chains (Gregg, 2009). The amount of organic DBPs in chlorinated ballast water is mainly dependent on the oxidant type and dosage and on the type and concentration of natural organic matter in the local ballast water (Gregg, 2009; Ichihashi et al., 1999). The high reactivity of hypobromous acid can create a variety of brominated DBP compounds in chlorinated and ozonated marine water (Werschun et al., 2012). In order to address this DBP formation potential when active substances are used for ballast water treatment, the joint Group of Experts on the Scientific Aspects of Marine Environmental Protection-Ballast Water Working Group GESAMP-BWWG has suggested a preliminary list of 18 compounds to be assessed in all BWMS tests before final approval (IMO, 2009a).

Several authors have addressed the formation potential of DBPs in chlorinated and ozonated ballast water (Gregg et al., 2009; Bowmer and Linders 2010 and Tsolaki and Diamadopoulos 2010). However, there is a lack of information about the causes and mechanisms of DBP formation, and the effects of different DBPs to the marine aquatic environment. These deficiencies have also been pointed out by Werschun et al. (2012). Previous work do not address the effect of concentration and nature of organic precursors on DBP formation potential, and do not compare observed toxicological effects to concentration levels of DBPs in an attempt to explain causes of toxicity. It is evident that the formation potential of different DBPs may vary considerably from test site to test site dependent of the nature of additives used to comply with the requirements regarding organic content of test water. Additives used include natural sediments from the seafloor, lignin, humic acids and a starch mixture. No previous published work has focused on the connection between the nature of the organic additive used and the abundance of various DBPs found in chlorinated ballast water.

Since 2005, five different ballast water management systems (BWMS) based on chlorination treatment have been tested at the Norwegian Institute for Water Research's (NIVA) test facility according to guidelines from the IMO. In this paper, the results from toxicity tests and DBP analyses of chlorinated

ballast water during the full scale testing of these BWMS are presented. The objective of the work was to evaluate a possible correlation between measured concentrations of various DBPs detected in treated ballast water and the oxidant dosage used or the oxidant consumption by the ballast water, as well as attempting to identify the main factor causing elevated levels of certain DBPs in ballast water at the time of discharge. A further objective was to find out if the DBPs found in treated ballast water could pose a risk to the marine aquatic environment.

2. Material and methods

2.1 Tested Ballast Water Management Systems

The five BWMS reported here all included treatment with active substances (e.g. hypochlorous acid, hydroxyl radicals) in combination with cavitation, ultrasonic treatment or similar, and always with filtration as pre-treatment. The active substances were introduced either by direct injection or by in-situ production. The latter was done by electro-chlorination or similar. During discharge, only physical treatment or neutralisation was applied, except for one BWMS that did use active substances, hence increasing the level of total residual oxidants (TRO) to a maximum of 2.0 mg/l at discharge. Each BWMS was operated by its vendor, but with inspection during operation by NIVA staff personnel to confirm and report operating parameters.

2.2 Test site facilities for full scale land-based tests

NIVA's test site facility located at Solbergstrand 20 km south of Oslo with direct access to seawater was used for the full scale land-based tests. The facility consists of four circular glass-fibre reinforced polyester tanks; one of 516 m³ for test water preparation, and three others of 231 m³ each for treated and control water (**Figure 1**). The surfaces of the tanks are coated with coatings for ships (Balloxy HB light, Jotun, Norway)

2.3 Chemical water quality of test waters used in full scale land-based tests

Test waters with high and medium range salinities were prepared following the requirements stated in the IMO G8 guidelines (IMO, 2008). The chemical requirements to the two test water types are shown in **Table 1**. Seawater for the tests was pumped up from 60 m depth in the Oslofjord. To obtain the required salinity content of the brackish water (20-22 PSU), water was prepared by mixing seawater from 1 m depth in the fjord with freshwater. Freshwater was pumped from ground water bore holes or from a local creek. To meet the required contents of dissolved organic carbon (DOC), particulate organic carbon (POC) and total suspended solids (TSS), soluble lignin or methylcellulose, starch and kaolin were added, respectively. Methylcellulose was used only for one of the BWMS fullscale testing.

Table 1 Required chemical water quality of test waters before any additional test organisms have been added. The difference between high and medium range salinities should be at least 10 PSU.

	Salinity (PSU)	DOC (mg/l)	POC (mg/l)	TSS (mg/l)
High Salinity				
Test water 1: IMO requirements	>32	>1	>1	>1
Test water 1: NIVA's test water	32 -33	1-5	1-5	10-15
Medium salinity				
Test water 2: IMO requirements	3-32	>5	>5	>50
Test water 2: NIVA's test water	20-22	5-8	5-8	50-70

A combination of indigenous harvested organisms and cultured surrogate species ($\geq 50 \mu\text{m}$: *Artemia franciscana*; $\geq 10\text{-}50 \mu\text{m}$: *Tetraselmis suecica*) were added to fulfil the biological water quality criteria stated in the IMO G8 guidelines (IMO, 2008). Indigenous algae and planktonic animal species were harvested from the sea outside Solbergstrand by pumping brackish water from 1 m depth to a rotating fine screen filter (20 μm mesh size, Unik Filter type 450, Unik Filtersystems, Os, Norway).

The chemical compounds were added only few hours prior to commencing the test. The homogeneity in the test water tank was assured by stirring, using propellers mounted near the bottom of the tank and close to the water surface. The homogeneity of the water was checked by on-site measurements of turbidity at different depths at the centre and at the periphery of the tanks.

2.4 Test cycle description

Each BWMS was subjected to at least 10 test cycles with a minimum of five test cycles for each water quality. Two of the three water qualities defined by IMO were used, which were seawater and brackish water (See **Table 1**). Before each test cycle, all tanks and piping system were cleaned by high pressure steam. Every test cycle included the ballasting operation, 5 days storage period of treated water and control water, and discharge operations (**Figure 1**). Before start-up of a test cycle, 500 m^3 of test water was prepared in the test water tank (WST). Ballasting operation consisted of pumping of at least 200 m^3 test water from the WST tank, through the treatment system, and over to a storage tank (TT2). For control water, at least 200 m^3 of test water was pumped to another storage tank (CT2), by-passing the treatment system. After five days storage in the dark, treated water was subjected to partial treatment or neutralisation during discharge pumping to another storage tank (TT1). The TT2 tank was then cleaned by high pressure steam, and control water was pumped in by-pass of the treatment unit to TT2.

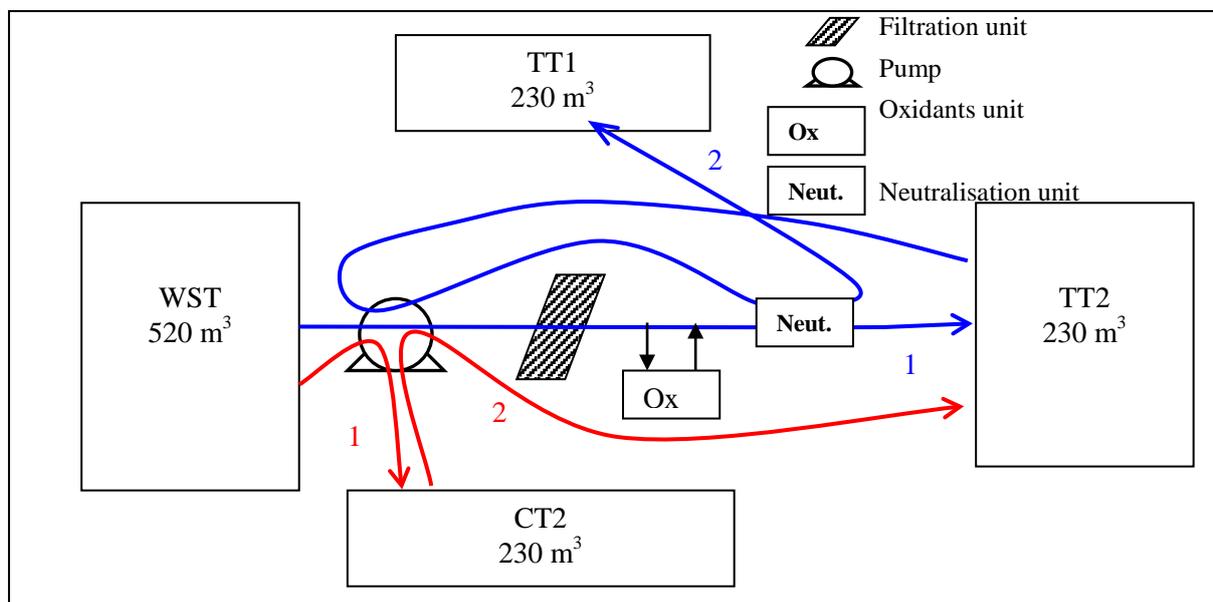


Figure 1. Transfer of test water during a test cycle with a BWMS including filtration, oxidation (chlorination) and neutralisation units. Blue line 1 indicates the day 0 ballasting operation of treated water, whilst blue line 2 indicates the day 5 discharge operation of treated water. Red line 1 indicates the day 0 ballasting operation of control water. Red line 2 indicates the day 5 discharge operation of control water.

2.5 Sampling and sample handling during full scale tests

All samples were collected directly from the tanks immediately after pumping and mixing operations. The homogeneity of the tank's water volume was checked before sampling by on-site measurements of turbidity at different depths at the centre and at the periphery of the tank. All samples were taken as grab samples from the periphery centre of the tanks using only gravity (no pumping) through a hose by the top of the tank. The samples for TRO and chemical water quality parameters analysis were collected as 3x 1 litres in clean glass and polyethylene bottles respectively. The samples for algal and crustacean toxicity tests were collected as 1x 5 litres in clean glass bottles and filtered (0.45µm cellulose membrane filter) before the tests were initiated. The samples for DBPs analyses were collected as one replicate in six 1L glass bottles which were top-filled and closed immediately after sampling. The DBPs samples were preserved with 150 mg/L sodium thiosulphate. For the fish toxicity test, more than 300 litres test water was collected in stainless steel containers. The test water was acclimatised to test temperature prior to exposing the fish to the test water. All analyses were performed at NIVA's laboratories immediately after sampling or within 24h after sampling, except for DBP samples which were analysed by external laboratories in Norway and Germany within 7 days after sampling. All samples were transported in cooling bags to the laboratories.

2.6 Laboratory tests with strong oxidants

Lignin could be an important precursor for some DBPs and especially trihalomethanes. To assess this under controlled conditions, laboratory tests were conducted. Sodium hypochlorite was added to brackish test water with the standard addition of lignin or methylcellulose together with kaolin and starch according to the G8 chemical requirements. The dosages of sodium hypochlorite were tuned in so that the TRO concentrations measured 30 minutes after treatment were 1 mg Cl/l, 5 mg Cl/l and 15 mg Cl/l. This procedure was followed to simulate the TRO measurement timing and TRO concentrations used during BWMS testing in full-scale.

The artificial brackish water was prepared in a 5 litres clean glass bottle by mixing 3.3 litres of 2µm filtered seawater and 1.7 litres distilled water to a salinity of 22 PSU. The seawater was collected from Oslofjord at 60 m depth with a salinity of 33PSU. The test water was added 5.0 mg Cl by adding 26mg/l of lignin (Borrebond FP-P53% Bulk, Norway) or 9.6mg/l methylcellulose (Metolose SM-25 from Shin-Etsu Chemical Co., Ltd, Japan). In addition, 40mg/l of kaolin (Kaolin china clay finest powder from Sigma-Aldrich®, Germany) and 20mg/l of corn starch flour (Maizena®, Norway) were also added. The artificial brackish water without any supplements was used as blank control. Only one replicate of each sample was prepared.

Sodium hypochlorite (GPR Rectapur, 4.5 % total Cl, 4.1 % free Cl) was added to the water in one-litre unused and annealed glass bottles and to additional 250-ml acid-washed glass bottles to obtain TRO concentrations of 1 mg Cl/l, 5 mg Cl/l and 15 mg Cl/l after a 30 min residence time at room temperature. Immediately after the sodium hypochlorite was added, the bottles were stoppered and quickly mixed. The one-litre bottles were then completely filled using a portion of the water from the associated small bottle, stoppered and then left to stand for five days in dark in room temperature. 150 mg/l sodium thiosulfate (Merck) was then added to each of the one-litre bottles to neutralise the residual oxidants and sent to an external laboratory for the quantification of trihalomethanes and bromate. The TRO concentrations after 30 min and after five days were measured in the small glass bottles, which also were stoppered and stored together with the larger bottles. The TRO consumption was calculated as the difference between the measured concentrations and the amounts added.

2.7 Acute and chronic toxicity test

Algal growth inhibition tests were done with samples from all test cycles after ballasting and at the time of discharge. The tests were performed according to International Standard ISO 10253 (2006) using the diatom *Skeletonema costatum*, NIVA-strain BAC 1 as test organism. The growth rate of each culture was calculated and expressed as percentage of the growth rate of control cultures in untreated ballast water. If a significant inhibition was observed in the treated ballast water, EC₅₀ (50 % growth inhibition) and EC₁₀ (10 % growth inhibition) concentrations were attempted estimated based on non-linear regression analysis of the growth rate against the concentration of ballast water. EC₅₀ is regarded an acute toxicity test end point, while EC₁₀ is regarded a chronic toxicity test end point.

In addition to the algal toxicity tests the following acute, chronic or sub-chronic toxicity effects were performed with treated ballast water at discharge from at least one test cycle of each water quality for each BWMS; acute toxicity to the marine crustacean *Acartia tonsa* according to ISO 14669 (1999), reproductive toxicity to the marine crustacean *Nitocra spinipes* (Bengtsson, 1987), the Oyster embryo bioassay according to the ASTM method (E724), chronic toxicity using rotatoria reproduction test with the marine species *Brachionus plicatilis* according to the standard ISO/FDIS 20666 adapted for marine species, acute toxicity towards the juvenile turbot *Scophthalmus maximus* according to OECD Guidelines for testing of chemicals No. 203 (OECD, 1992) and chronic toxicity towards the juvenile turbot *Scophthalmus maximus* according to OECD Guidelines for testing of chemicals No. 215 (OECD, 2000) adapted for marine species. The tests with copepods and fish involved long term exposure with frequent renewal of test water.

2.8 Chemical analysis

Total and free residual oxidants

Concentrations of total and free residual oxidants (TRO and FRO) were measured by the colorimetric DPD-method (American Public Health Association, 1995), which is currently the method recommended for measurement of TRO in seawater (Buchan et al., 2005). The method is based on the oxidation of N,N-diethyl-p-phenylendiamin (DPD) which turns to a pink Wurster-cation in the presence of strong oxidants. The intensity of the colour is proportional to the oxidants concentration. The colour intensity is measured by a Hach DR/2000 spectrophotometer (Hach Company, Loveland, CO, USA). The method requires 10 mL water samples. The detection range of this method is 0.02-0.20 mg/L Cl₂.

Disinfection by-products

All the DBPs that was analysed are listed in **Table 2**. Trihalomethanes were analysed by the purge and trap method according to US-EPA 524.2 (1995) with GC-MS detection. Bromate ions were measured by liquid ion chromatography according to NS-EN ISO 10304-1(1992). Haloacetic acids (HAA) were determined by GC-MS after liquid-liquid extraction and derivatisation according to NS-EN ISO 23631 (2006). Acetonitriles were analysed by liquid-liquid extraction and gas chromatography with electron-capture detection according to US EPA 551.1 (1990) method. Bromophenols were quantified by GC-MS after liquid-liquid extraction and derivatisation. Tribromobenzene, chlorotoluene and halogenated aliphates were analysed by purge and trap GC-MS according to US EPA 524.2 (1995). Whenever an individual DBP was found in the control test water at levels ≥ 10 % of what was measured in the treated test water, the concentration in the treated test water was adjusted by subtracting with the concentration found in the control test water.

Table 2 List of selected compounds analysed for in samples from full scale BWMS tests at NIVA in the period 2005-2011. Compounds included in the suggested DBP list from GESAMP-BWWG (IMO, 2009a) are shown in italics.

Group	Compounds
Trihalomethanes	<i>Trichloromethane, dichlorobromomethane, dibromochloromethane, tribromomethane</i>
Halomethanes	Dichloromethanes, dibromomethane
Haloethanes	1,2-dibromoethane, 1,2-dichloroethane, tetrachloroethane, 1,1,1-trichloroethane, 1,1,2-trichloroethane, 1,1-dichloroethane,
Halopropanes	1,1-dichloropropane, 1,3-dichloropropane, 2,2-dichloropropane, <i>1,2,3-trichloropropane</i> , 1,2-dibromo-3-chloropropane
Haloacetic acid	<i>Monochloroacetic acid, dichloroacetic acid, trichloroacetic acid, monobromoacetic acid, dibromoacetic acid, bromochloroacetic acid, dichlorobromoacetic acid, dibromochloroacetic acid, tribromoacetic acid</i>
Haloacetonitriles	Monobromoacetonitril, <i>dibromoacetonitril</i> , bromochloroacetonitril, chloroacetonitril, dichloroacetonitril, trichloroacetonitril
Haloethylenes	Trans-1,2-dichloroethylene, tetrachloroethylene, trichloroethylene, 1,1-dichloroethylene, cis-1,2-dichloroethylene
Halopropylenes	Trans-1,3-dichloropropylene, 1,1-dichloropropylene, cis-1,3-dichloropropylene
Halobenzenes	2-chlorotoluene, 3-chlorotoluene, 4-chlorotoluene, 1,2,3-tribromobenzene, 1,2,4-tribromobenzene, 1,3,5-tribromobenzene, chlorobenzene, 1-bromodimethylbenzene, 1,2-dichlorobenzene, 1,2,3-trimethylbenzene, 1,2,4-trimethylbenzene, 1,3,5-trimethylbenzene, 1,2-dimethylbenzene, 1,3-dichlorobenzene, 1,4-dichlorobenzene, bromobenzene
Haloamines	<i>Monochloroamine</i> , bromoamine
Halophenols	2,4-dibromophenol, 2,6-dibromophenol, 2,4,6-tribromophenol, 2-monochlorophenol, 3-monochlorophenol, 4-monochlorophenol, 2,3-dichlorophenol, 2,4+2,5-dichlorophenol, 2,6-dichlorophenol, 3,4-dichlorophenol, 3,5-dichlorophenol, 2,3,4-trichlorophenol, 2,3,5-trichlorophenol, 2,3,6-trichlorophenol, 2,4,5-trichlorophenol, 2,4,6-trichlorophenol, 3,4,5-trichlorophenol, 2,3,4,5-tetrachlorophenol, 2,3,4,6-tetrachlorophenol, 2,3,5,6-tetrachlorophenol, pentachlorophenol
Haloethandioles	Dichloroethanedirole, tribromoethanedirole
Other halogenated	<i>Bromate</i> , chlorate, vinyl chloride, carbon tetrachloride
Non-halogenated benzenes	Toluene, styrene, Tert-butylbenzene, dimethylbenzene (13+14), 1,2-dimethylbenzene, ethylbenzene, isopropylbenzene, n-butylbenzene, n-propylbenzene, sec-butylbenzene, 4-isopopyltoluene

2.9 Environmental risk assessment

A simple risk assessment of detected DBPs in the discharged waters was performed by comparing predicted environmental concentrations (PEC) with predicted no effect concentrations (PNEC) for the individual DBPs, and calculating the PEC/PNEC ratios. A ratio >1 indicate a potential risk in the environment. PEC values were calculated from the measured maximum concentration of the individual DBP for the five BWMS using active substances tested at NIVA and divided by a factor 100 to account for a realistic dilution in the recipient water based on a conservative MamPec v2.5 model output (MAMPEC, 2008). The PNEC value for the individual DBPs were estimated using available toxicity data and derivation rules according EU TGD (2003). Data for some compounds were often limited or completely lacking. When toxicity data was not available for the brominated compound, the toxicity data for the closest resembling chlorinated compound was used. According to Episuite 4.0, substituting Br with Cl in organic molecules reduces the aquatic toxicity only slightly.

3. Results and discussion

3.1 Correlation between observed toxicity and total residual oxidants

Of the 59 samples of treated water collected at the time of discharge, 32 and 15 samples exhibited chronic (EC_{10}) and acute (EC_{50}) algal toxicity, respectively. Acute crustacean and fish toxicity was observed in 1 and 2 of 13 and 11 samples, respectively, while chronic crustacean and fish toxicity was found in 5 and 2 out of 10 samples, respectively. In all samples where acute toxicity effect was observed on invertebrates and/or fish, the TRO values in these samples were above 1mg/L. All of the samples that exhibited acute or chronic crustacean and/or fish toxicity also exhibited algal toxicity; hence, the chronic toxicity end point of the algal growth inhibition test was the most sensitive of all the applied toxicity tests.

The main toxicity was expected to be caused by residual oxidants in the water. For instance, Gentile et al. (1976) found an EC_{50} value for sodium hypochlorite of 0.095 mg Cl/l in a study with *S. costatum* (i.e. the same algae as used in the applied algal toxicity tests here). **Fig. 2** shows the EC_{50} and EC_{10} values for algal toxicity relative to the measured FRO values in samples from the seawater and brackish water tests.

For seawater tests, the given correlation factors (R^2) for the exponential fits of the data points indicate a relatively good correlation; 0.81 for the acute toxicity tests (**Fig. 2A**) and 0.75 for the chronic toxicity tests (**Fig. 2B**). Of the 28 samples from the seawater tests, all 10 samples with EC_{50} values <100 % had FRO values >0.1 mg/l, hence an acute toxic effect was expected. However, one sample with FRO values >0.1mg/l (0.13mg/l) showed neither acute nor chronic toxicity effect. Eight samples with low FRO (<0.02-0.09 mg/l) showed chronic toxicity effect.

For the brackish water tests there seemed to be no correlation between observed acute (**Fig. 2C**) nor chronic (**Fig. 2D**) algal toxicity and measured FRO ($R^2 < 0.01$). Of the 31 samples from the brackish water tests, only one of the five samples with EC_{50} values <100 % had measured FRO values >0.1 Cl mg/l. And for six of the 26 samples that did not indicate any acute algal toxicity, the measured FRO values were above 0.1 mg Cl/l (0.11-0.37 mg Cl/l). Five of these latter samples came from the same BWMS that was using active substances during discharge. Chronic toxicity was observed with only two of these five samples. However, of the 26 brackish water samples with FRO values <0.1 mg Cl/l ten samples showed chronic toxicity effects. The FRO levels, which should better reflect the levels of the stronger oxidising agents, were ≤ 0.02 -0.06 mg Cl/l.

Though a poor correlation could be observed between toxicity effect and FRO results, especially for brackish water, approximately 75% of the samples in total did show expected results for toxicity effect related to the FRO measurements. However 18 (8 seawater and 10 brackish water samples) of 59 samples with low FRO values showed toxic effect and only five samples (1 seawater and 4 brackish water) with high FRO values showed none toxic effect. This may suggest that there may have been additional factors to residual oxidants that contributed to the observed algal growth inhibition in these samples. Disinfection by-products (DBPs) formed from the use of active substances, and which are not contributing to the TRO value, may potentially influence the algal toxicity. However, the almost absence of algal toxicity in one sample collected from one of the BWMS and in 11 samples from another BWMS that both applied active neutralisation prior to discharge, indicates that neutralisation was effective to minimise the observed toxicity in these cases. The fact that the chronic toxicity observed in only one sample, despite the TRO level was below the quantification limit (<0.02mg/l), of the 12 collected samples after neutralisation was removed after increasing of the neutralisation dosage, supports this hypothesis. Hence, post neutralisation may assure a non-toxic ballast water discharge also when toxicity effect was observed in samples where no residual oxidants could be detected prior to neutralisation. Nevertheless neutralisation only affects compounds with oxidising abilities. Hence, high levels of non-oxidising DBP may still affect the toxicity of the treated water after neutralisation.

The DBP formation during ballast water treatment and possible concomitant toxicity effects are assessed in more detail in chapter 3.2.

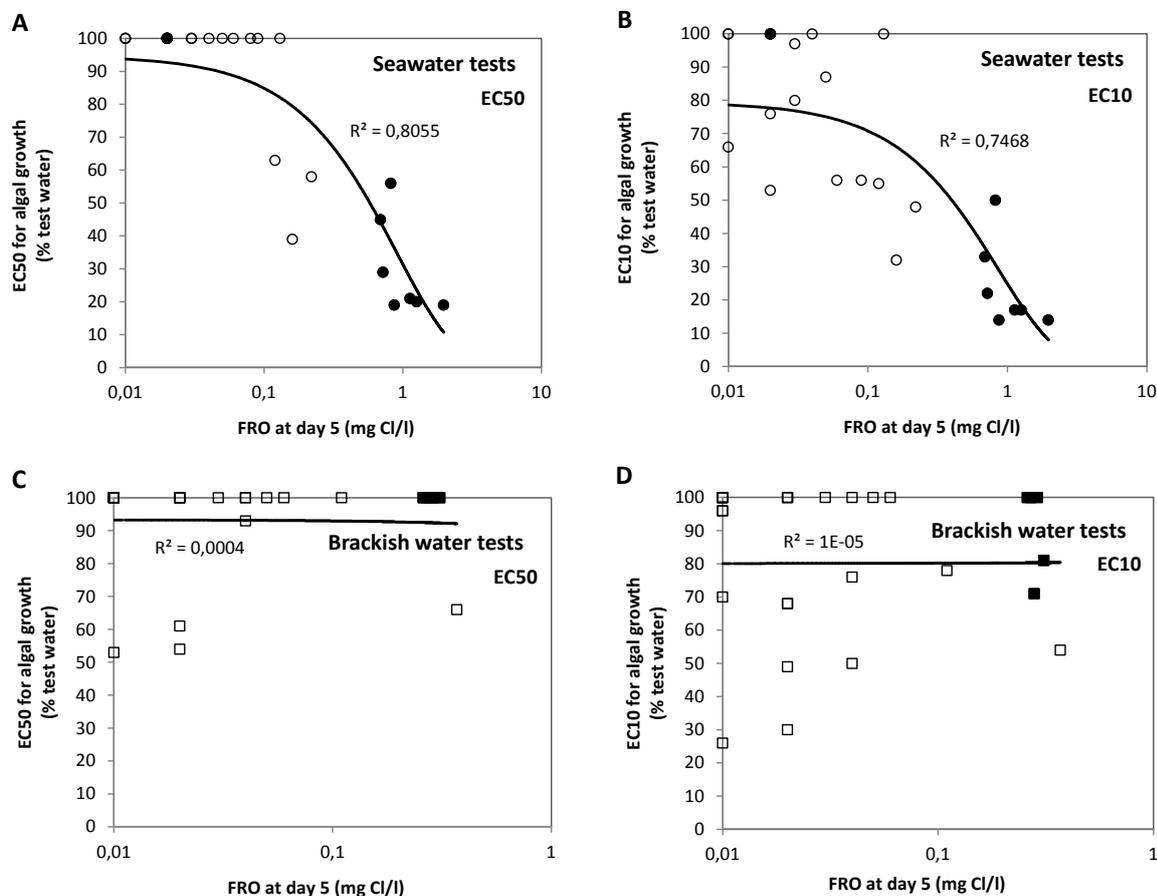


Fig. 2 Observed acute toxicity EC_{50} (A, C) and chronic toxicity EC_{10} (B, D) to the green algae *S. costatum* over 72 hours and total residual oxidants (TRO) in samples collected at the time of discharge after five days storage during full scale testing of BWMS using active substances during ballasting, with or without neutralisation. Results from seawater tests (A, B) and brackish water tests (C, D) are shown in separate figures. The values from the BWMS that were using active substances (AS) also during discharge, without neutralisation, are shown with filled squares (brackish water tests) and filled circles (seawater tests). The exponential fit is indicated with the regression line and its R^2 value.

3.2 Disinfection by-products detected in treated test water at the time of discharge

Of the close to 100 different potential DBP compounds that have been included by NIVA when analysing samples of treated ballast water (see **Table 2**), 22 compounds were detected above the detection limit in at least one sample collected at the time of discharge. See **Table 3**. The majority of the compounds on the DBP list suggested by GESAMP-BWWG for analysis in connection with risk assessment of treated ballast water (IMO, 2009a) are among these detected compounds (monochloroacetic acid was only detected above the detection limit in samples collected directly after treatment and monochloroamine was not analysed for). Notably, chlorate and dibromomethane are not included in the list from GESAMP-BWWG, but they were among the eight compounds that have been found in 50 % or more of all the treated discharge samples in which they have been analysed (see **Table 3**).

The observed median and maximum concentrations of the eight most often detected DBP compounds were in the range from low $\mu\text{g/l}$ to several hundred $\mu\text{g/l}$. In general, the brominated compounds were more predominant than the chlorinated counterparts with some important exceptions; chlorate and dichloromethane were found at higher concentrations than bromate and dibromomethane, respectively. Overall, the 5 most often detected compounds exhibited also the highest maximum concentration levels. Notably, tribromoacetic acid was not among the more frequently detected DBPs, however, when found the level was often relatively high (53-240 $\mu\text{g/l}$) as compared to the others DBPs. Nevertheless, some DBP can present high toxicity in low concentration therefore the toxicity effect of some DBP is studied further in the chapter 3.3.

Table 3 List of disinfection by-products sorted by their detection frequency in samples collected at discharge during BWMS testing at NIVA: Number of analyses (n), detection limit, detection frequency and median and maximum concentrations. The median values are presented only for DBP concentrations above the detection limit in at least 50 % of the samples (gray shading). DBPs included in the list suggested by GESAMP (IMO, 2009a) are shown in bold.

DBP compound	n	Detection limit ($\mu\text{g/l}$)	Detection frequency (%)	Concentration ($\mu\text{g/l}$)	
				Median	Maximum
Tribromomethane	43	<0.1	100	183	670
Chlorate	10	<3	100	330	370
Dibromoacetic acid	35	<0.1	100	5.9	120
Dibromochloromethane	43	<0.1-0.5	98	8.0	32
Bromate	39	<1-3	85	13	46
Dibromomethane	23	<0.1	74	0.40	7.8
Bromochloroacetic acid	31	<0.1	71	0.43	2.0
Dichlorobromomethane	43	<0.1-0.5	60	0.55	2.4
Vinyl Chloride	5	<0.1	40	-	0.30
Dichloromethane	9	<0.1	33	-	12.2
Tribromoacetic acid	34	<0.1	24	-	240
Trichloromethane	39	<0.1-0.5	23	-	2.5
Dichlorobromoacetic acid	34	<0.1	21	-	4.8
Dibromochloroacetic acid	34	<0.1	18	-	20
1,2-dibromoethane	33	<0.1-1	18	-	1.3
2,4,6-tribromophenol	34	<0.1	15	-	0.52
Monobromoacetic acid	34	<0.2	12	-	5.7
Bromochloroacetonitrile	14	<0.1-1	7.1	-	0.5
Dibromoacetonitrile	15	<0.1-1	6.7	-	0.80
1,2,3-trichloropropane	33	<0.1-1	6.1	-	2.3
Dichloroacetic acid	34	<0.3	2.9	-	4.8
Trichloroacetic acid	34	<0.2-1	2.9	-	0.24
Monochloroacetic acid	34	<0.5	0	-	<0.5

3.3 DBP formation and chronic algal toxicity

It is more likely to observe toxic effects from DBPs on the chronic toxicity end point (EC_{10}) of the algal growth inhibition test than on the acute toxicity end point (EC_{50}). However, to be able to distinguish between the toxic effects caused by DBPs rather than residual oxidants in the water, only samples with TRO values well below the expected no effect concentration (NOEC) of the residual oxidants should be used in such an assessment. Unfortunately, as discussed in section 3.1, the actual compounds that made up the TRO values at the time of discharge were not known. Though, assuming

an EC₁₀ value close to 0.08 mg Cl/l for sodium hypochlorite on the algae *S. costatum* (deducted from the fact that EC₁₀ would be even lower than 0.095 mg Cl/l EC₅₀ value reported by Gentile et al., 1976) and, hence, using only samples with TRO values ≤0.08 mg Cl/l, a simple qualitative assessment of the possible contribution from DBPs to the observed chronic toxicity was performed. As indicated in **Fig. 3**, there seemed to be no clear indication that the measured DBP concentrations affected the algal toxicity neither for individual DBPs (tribromoacetic acid, dibromochloromethane, chlorate, monobromoacetic acid and tribromomethane) frequently detected in the discharged water at levels of potential environmental concern (see section 3.4) nor for the sum of all DBPs. A direct comparison between the toxicity endpoints for the individual DBPs (see **Table 4**) and the highest levels at which they were found in the discharged waters (see **Table 3**) further substantiated this, as their concentrations were at least a factor 25 lower than the available PNEC value.

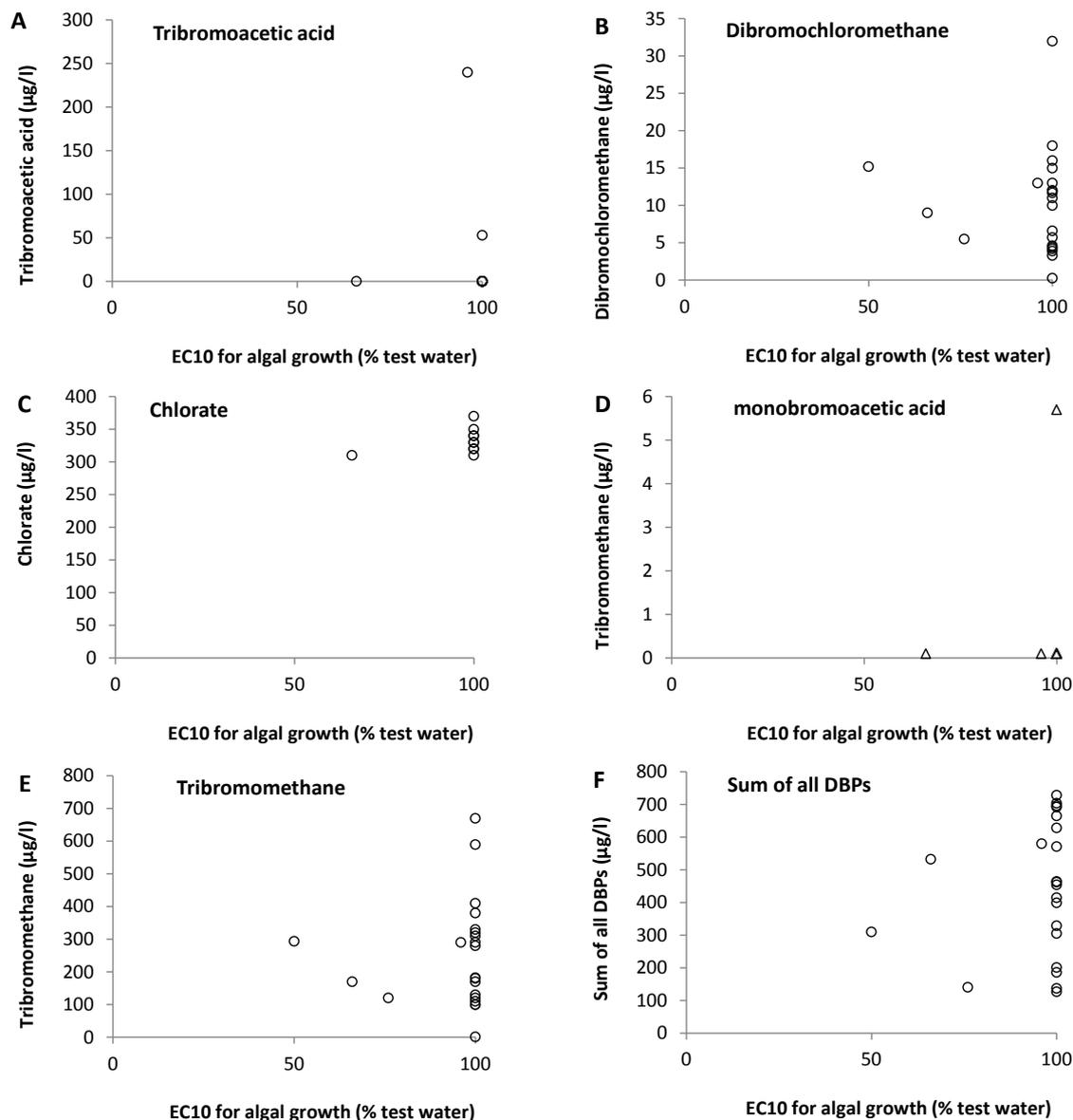


Fig. 3 Concentrations of tribromoacetic acid (A), dibromochloromethane (B), chlorate (C), monobromoacetic acid (D), tribromomethane (E) and the sum of all DBPs (F) and the measured EC₁₀ value for chronic algal toxicity in samples with TRO values ≤0.1 mg Cl/l at the time of deballasting.

3.4 Environmental risk from discharged DBPs

The results of a simple environmental risk assessment of the individual DBPs detected in the discharged waters are summarised in **Table 4**. Four of the compounds (tribromoacetic acid, dibromochloromethane, chlorate and monobromoacetic acid) were at times found at concentrations that may pose a risk to the local aquatic environment (PEC/PNEC value >1). It should be noted that one of these compounds, chlorate, is not on the DBP list suggested by GESAMP-BWWG for analysis in connection with risk assessment of treated ballast water (IMO, 2009a). We therefore recommend chlorate to be included in the environmental risk assessment.

The high assessment factors (AF) for many of the DBPs in the risk assessment (see **Table 4**) indicate that limited toxicity data are available. Hence, future toxicity tests on these compounds may alter the derived PNEC values considerably. Another important factor in the risk assessment is the cocktail effect from the simultaneous release of such a large range of DBPs. An environmental risk assessment should therefore include the summarised effect of all DBP compounds, at least those that have the same toxic mode of action, for example all the haloalkanes or haloacetic acids. Assessing the latter group, the PEC for tribromoacetic acid increased from 240 µg/l to 397 µg/l and the resulting calculated PEC/PNEC ratio increased from 4.0 to 6.6. However, when using the lowest available PNEC value within this group, namely that for monobromoacetic acid, gave a PEC/PNEC ratio of 99.

The second highest PEC/PNEC ratio shown in **Table 4** is for Dibromochloromethane. The study from which the PNEC value was derived is taken from an IMO MEPC document (Japanese ministry of environment, 2008), and the underlying study has not been evaluated. The test endpoint for this study is a factor 1000 lower than other toxicity studies for this compound, which seems unrealistic.

However PEC values are dependant of the technology tested and the PNEC values dependant of the available published data, therefore these results of PEC/PNEC are just an indication from the five BWMS using active substances tested at NIVA and available literature references. All BWMS has to be individually evaluated for each DBP and PEC/PNEC ratio calculation. Further comparison of BWMS testing results from NIVA to DBP results from other test facilities is presented in chapter 3.5.

Table 4 Risk assessment data for the individual DBP compounds: Maximum predicted environmental concentrations (PEC_{max}), toxicity data and assessment factors (AF) leading to predicted no effect concentrations (PNEC) in the environment and calculated PEC/PNEC ratios.

DBP compound	PEC_{max} ($\mu\text{g/l}$)	Toxicity data				AF	PNEC ($\mu\text{g/l}$)	PEC/ PNEC
		Test organism	End point	Conc. (mg/l)	Ref. .			
Tribromoacetic acid *	2.4	<i>Chlorella pyrenoidosa</i> Green algae	NOEC, 14d growth fresh water	0.3	⁸	500	0.6	4
Dibromochloromethane	0.32	Crustacea <i>Daphnia magna</i>	NOEC, 21d	0.063	¹	500	0.126	2.5
Chlorate	3.7	Growth reduction in several marine seaweeds	NOEC, 3 months	0.02	²	10	2	1.9
Monobromoacetic acid	0.06	<i>Scenedesmus subspicatus</i> Green algae	EC ₅₀ , 72 h	0.2	³	500	0.04	1.5
1,2,3-trichloropropane	0.02	<i>Chaetogammarus marinus</i> Amphipod	NOEC, 14 d Salt water	0.02	⁴	500	0.04	0.5
Dibromochloroacetic acid*	0.2	<i>Chlorella pyrenoidosa</i> Green algae	NOEC, 14d growth fresh water	0.3	⁸	500	0.6	0.33
Bromate *	0.46	Growth reduction in several marine seaweeds	NOEC, 3 months	0.02	²	10	2	0.23
Dibromoacetic acid	1.2	Fish <i>Pimephales promelas</i>	LC ₅₀ , 4 d	69	⁵	10000	6.9	0.17
Dibromoacetonitrile	0.008	Fish <i>Pimephales promelas</i>	EC ₅₀	550	⁵	10000	0.055	0.15
Tribromomethane	6.7	<i>C. variegatus</i> Sheepshead minnow	NOEC, 96h Mortality Salt water	2.9	⁶	50	58	0.11
Bromochloroacetic acid *	0.02	<i>Nitocra spinipes</i> Harpacticoid copepod	LC ₅₀ , 96h Salt water	23	⁹	10000	2.3	0.09
Dichlorobromoacetic acid*	0.05	<i>Chlorella pyrenoidosa</i> Green algae	NOEC, 14d growth fresh water	0.3	⁸	500	0.6	0.09
Bromochloroacetonitrile*	0.005	Fish <i>Pimephales promelas</i>	EC ₅₀	550	⁵	10000	0.055	0.09
2,4,6-tribromophenol	0.005	Bacciliariaphyceae Diatoms	NOEC, photosynthesis	0.5	⁷	1000	0.5	0.05
Trichloroacetic acid	0.002	<i>Chlorella pyrenoidosa</i> Green algae	NOEC, 14d growth fresh water	0.3	⁸	500	0.6	0.033
Dichloroacetic acid	0.05	<i>Nitocra spinipes</i> Harpacticoid copepod	LC ₅₀ , 96h salt water	23	⁹	10000	2.3	0.02
Dichlorobromomethane	0.02	Ciliate <i>Tetrahymena pyriformis</i>	EC ₅₀ , growth	240	¹⁰	10000	24	0.013
Dibromomethane	0.08	Acute fish test	EC ₅₀	10	¹¹	10000	1	0.0028
Dichloromethane	0.12	<i>Pseudokirchneriella subcapitata</i> Green algae	NOEC, 96h growth	56	¹²	1000	56	0.002
Trichloromethane	0.03	Crustacea <i>Cerodaphnia dubia</i>	NOEC, 10 d mortality	3.4	¹³	100	34	0.0009
1,2-dibromoethane	0.013	Rice fish (<i>Oryzias latipes</i>)	LC ₅₀ , 48h	183	¹⁴	10000	18.3	0.0007
Vinyl Chloride	0.003	Acute fish test (<i>Danio rerio</i>)	EC ₅₀ , 96h	210	¹⁵	10000	21	0.0001

¹⁾ Japanese ministry of environment (2008), ²⁾ Rosemarin et al. (1994), ³⁾ Kuhn and Pattard (1990), ⁴⁾ Kooijman (1981), ⁵⁾ Mayes et al. (1985), ⁶⁾ Heitmuller et al. (1981), ⁷⁾ Erickson and Hawkins (1980), ⁸⁾ Bednarz T. (1981), ⁹⁾ Linden et al. (1979), ¹⁰⁾ Yoshioka et al. (1985), ¹¹⁾ GESAMP (1989), ¹²⁾ U.S. Environmental Protection Agency (1978), ¹³⁾ Cowgill and Milazzo (1991), ¹⁴⁾ MITI (1992), ¹⁵⁾ Groeneveld et al. (1993).

* For brominated compounds lacking toxicity data, the PNEC data from the closest resembling chlorinated compound was used instead, which was; trichloroacetic acid for tribromoacetic acid, dibromochloroacetic acid for dichlorobromoacetic acid, chlorate for bromate, dichloroacetic acid for dibromoacetic acid and lastly dibromoacetonitrile for bromochloroacetonitrile.

3.5 Comparison of measured DBP concentrations with results from other test facilities

Fig. 4 presents a comparison between the maximum DBP concentrations observed in the full scale land-based tests done at NIVA's test facility and the maximum DBP concentrations reported from tests done at other test facilities (IMO 2008a-c, IMO 2009b-d, IMO 2010a-b and IMO 2011a-d). The average of the maximum levels found in discharged water from the five different BWMS tested at NIVA were well above the observed median of the maximum values reported by others for bromate (2.2 times higher), for all trihalomethanes (2.1-3.2 times higher), for tri- and di-brominated haloacetic acids (1.3-3.5 times higher) and for dichloroacetic acid (4.8 times higher). These results indicate that the DBP levels found at NIVA were, in general, relatively high. However, the average of the maximum levels found at NIVA were always within the 90 percentile of the maximum levels reported by others (percentile rank of 56-85 for the above), and the overall maximum level of any of the DBPs found at NIVA were below the highest reported level reported by others. This supports the risk assessment data presented in **Table 4** are relevant for these DBP concentrations measured.

These somewhat high levels of some of the brominated DBPs found during tests done at NIVA compared to tests done elsewhere may be coincidental, but they may also be caused by differences in the site-specific test waters that have been used. At NIVA, particularly the brackish test water may vary considerably due to seasonal variations in the composition of the water collected from 1 meter depth in the fjord. Though, there was no clear tendency of higher DBP levels in the samples collected during brackish water tests as compared to seawater tests (results not shown). However, the higher DBP formation may also be caused by for example lignin added to the test water to comply with the G8 requirements set by IMO.

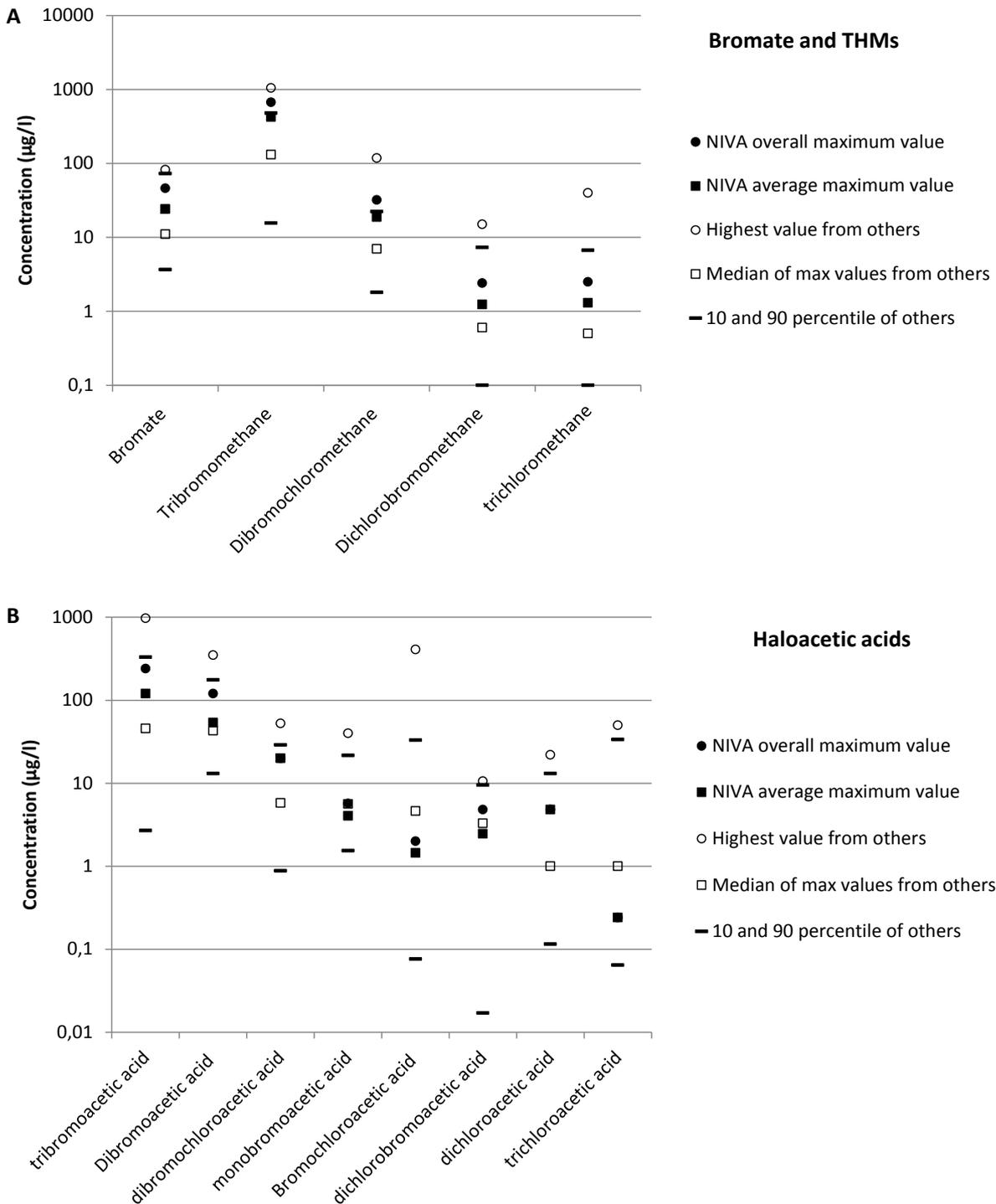


Fig. 4 Formation of bromate and trihalomethanes (A) and haloacetic acids (B) in high and medium salinity test waters in full scale land-based tests done at NIVA and at test facilities elsewhere; average of the maximum values and the overall maximum value found in discharged water for the five BWMS tested at NIVA since 2005 and the highest and the median value of the maximum values reported by others. The 10 and 90 percentile values summarize only the maximum values reported by others. (Sources: IMO 2008a-c, IMO 2009b-d, IMO 2010a-b and IMO 2011a-d)

3.6 Lignin as a precursor to selected DBPs

Both soluble and particulate organic matters have been added to comply with the required test water quality set by IMO. However, the soluble compounds (lignin and methylcellulose) are the most likely precursors to the formed DBPs. **Fig. 5** shows the consumed TRO over the five days storage period (i.e. the difference between in-tank TRO measurements at day 0 immediately after ballasting treatment and at day 5 immediately before discharge treatment) as a function of the initial TRO measured immediately after ballasting treatment for both brackish water tests (**Fig. 5A**) and seawater tests (**Fig. 5B**). When lignin had been added to the test water, the consumption of active substances strongly correlated with the TRO level in the water directly after ballasting treatment both for brackish water and seawater ($R^2 > 0.99$ and $R^2 > 0.96$, respectively for the linear fit), primarily due to the fact that most of the initial TRO had been consumed. Note also that the inclination lines for lignin in the brackish and seawater tests were both close to 1 (0.98 and 0.83, respectively), indicating that the TRO consumption was never limited by the presence of compounds that would be oxidised within the five days' timeframe. Though limited by the low number of tests, this did not appear to be the case when methylcellulose instead of lignin was added to the test water. However, if the stray point for methylcellulose in seawater is omitted (**Fig. 5B**), the linear fit is good ($R^2 > 0.99$). Nevertheless, the concomitant TRO consumption when the TRO dosage was increased was significantly lower for methylcellulose than for lignin, suggesting that lignin contributed more to the TRO consumption than methylcellulose. Note that the calculated TRO consumption does not take into account the immediate consumption occurring during ballasting treatment and until the sample was collected, at the latest 30 minutes after the treatment was finished, nor during discharge treatment (e.g. effect of neutralisation or additional active substances).

However, the correlation between TRO consumption and formation of the individual DBPs after 5 days of storage after ballasting treatment were not that evident. Though, as can be seen from **Fig. 6**, the formation of tribromomethane and dibromochloromethane in seawater and brackish water tended to increase with increasing TRO consumption. The linear fits were slightly better for the individual DBP results from the seawater tests when they were plotted against the initial TRO concentration after ballasting treatment, but the linear fits for the results in brackish water were practically unchanged (results not shown). For DBPs such as dibromoacetic acid and bromate the tendencies were not evident (results not shown).

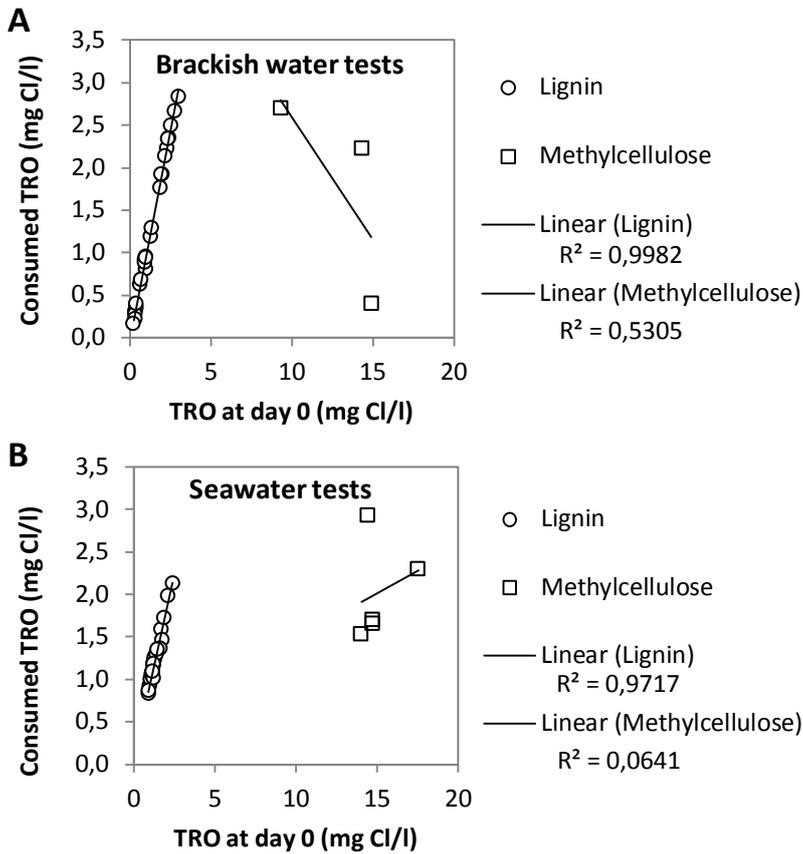


Fig. 5 Observed consumption of TRO at different initial TRO concentrations immediately after ballasting treatment in land-based full scale tests performed at NIVA where lignin or methylcellulose was added as supplemental soluble organic carbon; brackish water tests (A) and seawater tests (B). In the brackish water tests lignin and methylcellulose were added to 4.6 mg C/l and 6.9 mg C/l, respectively, while in the seawater they were added to 0.95 mg C/l and 1.14 mg C/l, respectively.

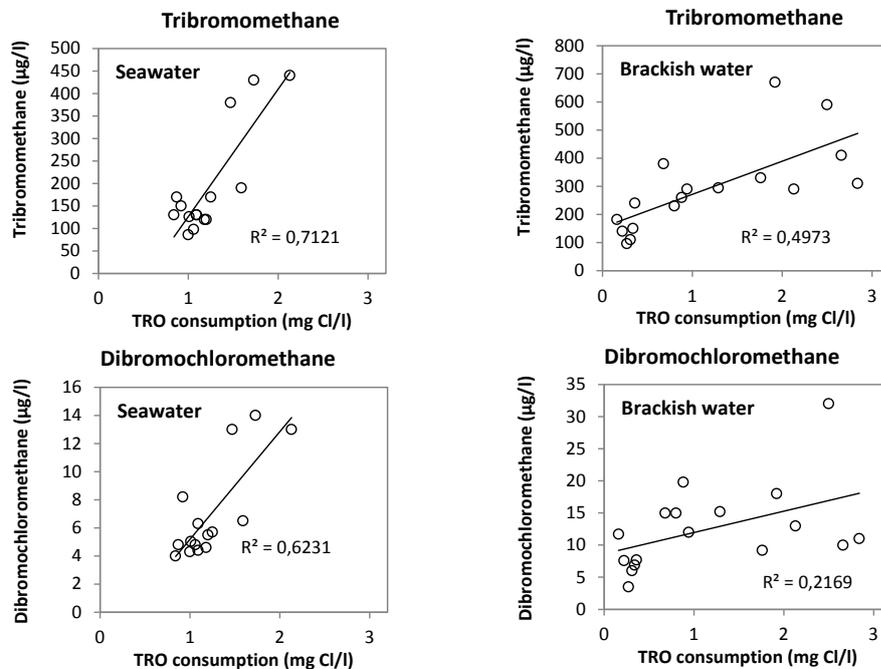


Fig. 6 Formation of tribromomethane and dibromochloromethane in seawater and brackish water plotted against the TRO consumption from right after treatment during ballasting to the time of discharge at day 5 in land-based full scale tests performed at NIVA.

3.7 Supportive laboratory scale tests with lignin and methylcellulose

These results from the full scale tests indicated that lignin could be an important precursor for some DBPs and especially trihalomethanes. To assess this under controlled conditions laboratory tests were conducted using sodium hypochlorite as oxidant in brackish test water added lignin or methylcellulose (to 5.0 mg C/l) together with kaolin and starch. Brackish water without any additions was used as blank control. The calculated TRO consumptions 30 min and five days after increasing doses of sodium hypochlorite had been added are shown in **Fig. 7**. The relative TRO consumption to the hypochlorite dosage was significantly higher in tests with lignin than in tests with methylcellulose, both the immediate consumption occurring the first 30 min as well as the consumption taking place the next five days. Actually, the observed consumption in the tests with methylcellulose was similar to what was observed in the control blank tests. The TRO consumption was 2.5 times higher for lignin than for methylcellulose over the five days of storage, as given by the increasing slope of the inclination line in **Fig. 7**. At the end of the five days there was an excess TRO in all methylcellulose and control blank tests, while in the lignin tests only in the test with the highest TRO dosage was there any distinct excess TRO at the end of the period.

Some studies from paper bleaching using hypochlorite or chlorine dioxide to degrade lignin compounds are available, but not relevant for ballast water due to the chemical differences between freshwater used in paper industry and seawater used for ballast water (Hamzeh et al., 2006). It has not been possible to find published TRO-data from other ballast water test facilities using lignin or other different additives, including micromate humates, instant tea or natural organics. In different natural seawaters and river waters without additives, great variability in TRO consumption was reported (Perrins et al., 2006). As indicated in our study, differences in initial TRO dose applied and differences in DOC composition will have significant influence on TRO-demand.

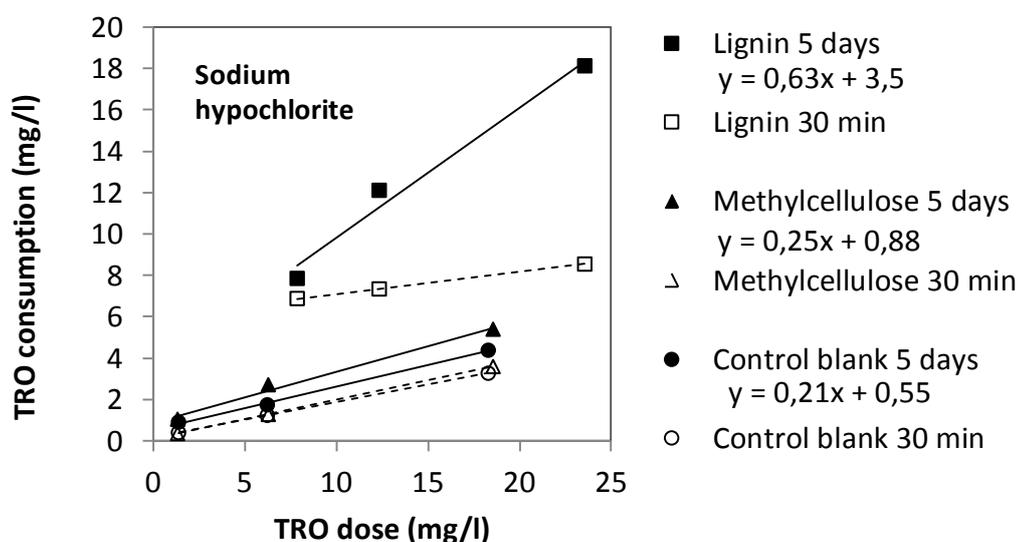


Fig. 7 Results from lab scale oxidation tests with sodium hypochlorite. TRO consumption after 30 min and five days when dosing sodium hypochlorite to obtain a TRO concentration of 1 mg Cl/l, 5 mg Cl/l and 15 mg Cl/l after 30 min in synthetic brackish test water with standard additions of kaolin, starch and lignin or methylcellulose. Brackish water without any other additives was used as blank control.

Fig. 8 shows the brackish control-corrected concentrations of tribromomethane, dibromochloromethane and dichlorobromomethane five days after sodium hypochlorite had been added. The linear fits for the correlation between the formation of these trihalomethanes and the concomitant TRO consumption in the test water with lignin were very good for all three compounds ($R^2 > 0.98$), clearly suggesting a relationship between the formation of these DBPs and the presence of lignin. In the test water with methylcellulose there was no indication of increased formation of trihalomethanes with increasing TRO consumption. The concentrations of trichloromethane were below the detection limit ($< 0.1 \mu\text{g/l}$) for all samples.

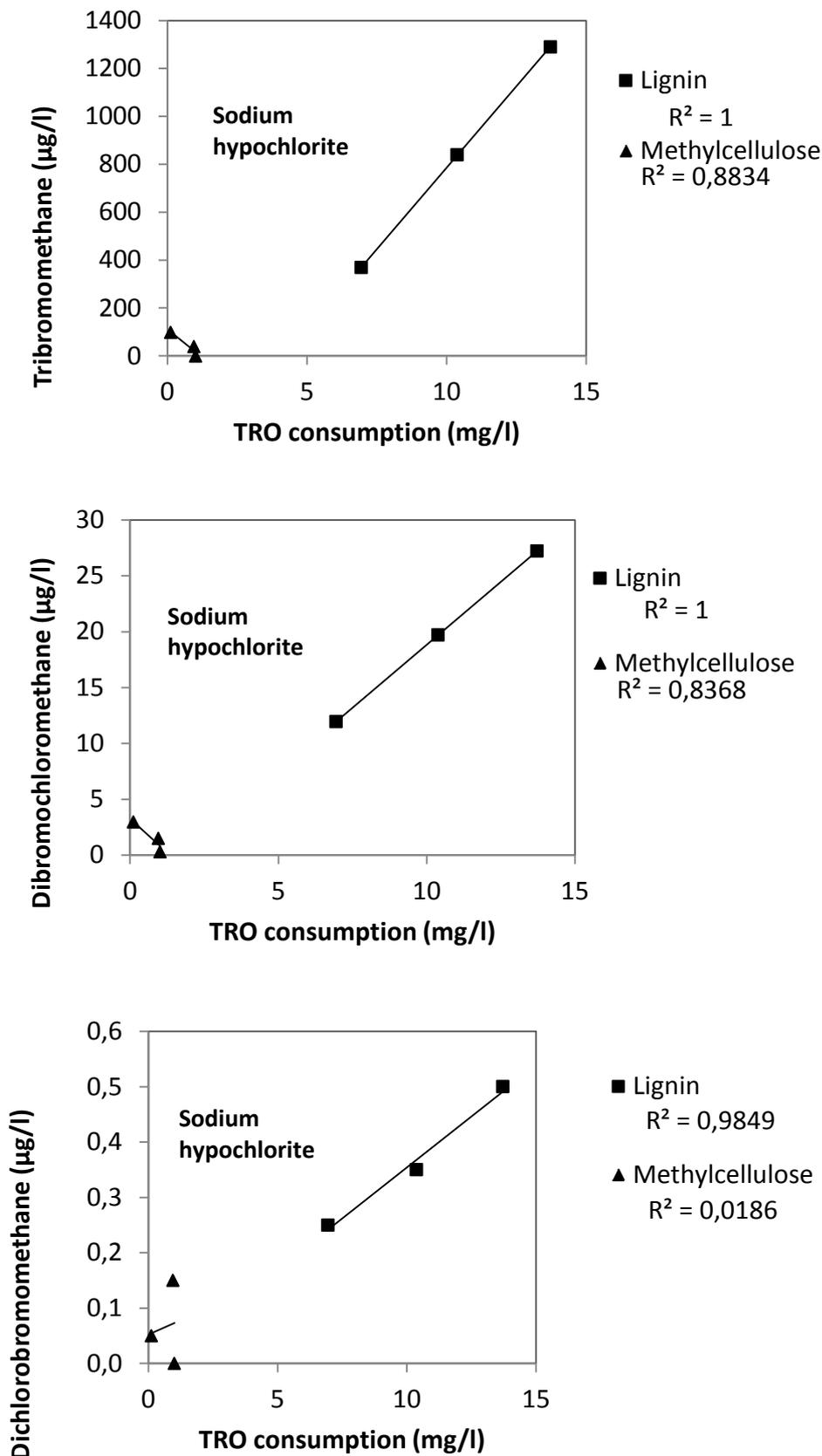


Fig. 8 Results from lab scale oxidation tests with sodium hypochlorite. Concentrations of tribromomethane, dibromochloromethane and dichlorobromomethane five days after the dosage of sodium hypochlorite to synthetic brackish test waters added lignin or methylcellulose and the concomitant TRO consumption. Sodium hypochlorite had been added to obtain TRO concentrations of 1 mg Cl/l, 5 mg Cl/l and 15 mg Cl/l after 30 min. The TRO consumptions and trihalomethanes concentrations have been blank control-corrected.

The concentrations of bromate five days after the dosage of sodium hypochlorite to the waters that contained lignin, methylcellulose or no additives (control blank) relative to the TRO consumptions are shown in **Fig. 9**. Interestingly, the presence of lignin appeared to limit the formation of bromate, whereas methylcellulose did not seem to affect the bromate formation potential. The good second order polynomial fits ($R^2=1.00$) suggest that the bromate formation was limited by the oxidation of other compounds at low sodium hypochlorite dosages.

Since lignin is one of the most common and naturally occurring compounds in the world, being a structural component in plants, its use as a dissolved organic carbon source in land-based full scale BWMS tests may be regarded as a realistic, but worst case, scenario. As these tests suggest, methylcellulose may be used as a less DBP producing alternative. In another study, the TRO consumption was compared to the formation of halogenated DBP in two different natural water sources, i.e. open seawater and costal seawater (Fabbicino and Korshin, 2005). In accordance with our study, the nature and the concentration of the DBP were different accordingly to the source water, with a predominance of tribromomethane.

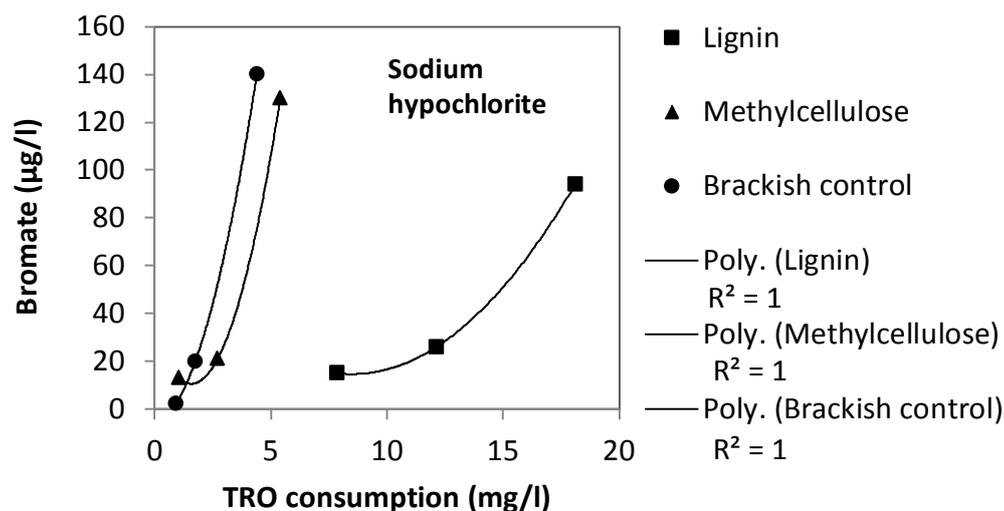


Fig. 9 Results from lab scale oxidation tests with sodium hypochlorite. Concentration of bromate five days after the dosage of sodium hypochlorite to synthetic brackish test waters added lignin, methylcellulose or none (brackish control) and the concomitant TRO consumption. Sodium hypochlorite had been added to obtain TRO concentrations of 1 mg Cl/l, 5 mg Cl/l and 15 mg Cl/l after 30 min.

4. Conclusion

In total, 75% of the samples showed toxicity effect for high FRO values as expected. Though there seemed to be a relatively good correlation for seawater tests between observed algal toxicity and measured FRO levels at the time of discharge, no such correlation was found for brackishwater tests. However, post neutralisation may assure a non-toxic ballast water discharge for oxidants removal but high DBP levels may still affect the toxicity of the treated water.

22 of the approximately 100 different potential DBP compounds that were analysed for at the time of discharge were detected above the detection limit. Two of these, chlorate and dibromomethane, are at the present time not on the DBP list from GESAMP-BWWG recommended for analysis in connection with risk assessment of treated ballast water. Chlorate was also among the four compounds (tribromoacetic acid, dibromochloromethane, chlorate and monobromoacetic acid) that at times were found at concentrations that may pose a risk to the local aquatic environment, and it is therefore proposed to be included on the list when the list is updated. The precision of the environmental risk assessment was limited by the lack of toxicity data; hence future toxicity tests on these compounds and the so called cocktail effects from the complex mixture in which these compounds are discharged may alter the derived PNEC values considerably. Nevertheless, there seemed to be no clear indication that the measured DBP concentrations affected the algal, crustacean or fish toxicity neither for individual DBPs nor for the sum of all DBPs.

The DBP levels found at NIVA were, in general, relatively high compared to DBP levels found in other land-based full scale BWMS tests. Lignin was identified as an important TRO consumer in the treated ballast water during storage and a possible precursor for the brominated trihalomethanes. As lignin is one of the most common and

naturally occurring compounds in the world, its use as a dissolved organic carbon source in the BWMS tests may be regarded as a realistic, but worst case, scenario. Methylcellulose could be a potential replacement for lignin as this compound did not seem to affect neither the TRO consumption nor the formation of trihalomethanes. Interestingly, the presence of lignin appeared to limit the formation of bromate, whereas methylcellulose did not seem to affect the bromate formation potential. Further research should be done with other substantives that are commonly used by test facilities worldwide. Only 5 different BWMS were studied here, all based on chlorination treatment, while IMO reported 34 basic approvals and 20 type approved BWMS using active substances (IMO, 2011e), therefore further study of the correlation between TRO, DBP and toxicity effect with other type of oxidation as ozonation for example should be performed.

Acknowledgements

The study was based on data obtained through land-based full scale testing at NIVAs test facility, Norway. The study was made possible by financial contributions from NIVA. We wish to thank the whole ballast team at NIVA, both technical staff at the test facility at Solbergstrand and the laboratory staff in Oslo for all contributions during the conducted land-based full scale tests.

References

- American Public Health Association (APHA), 1995. Standard Methods for the Examination of Water and Wastewater, 19th ed. American Public Health Association, Washington, DC.
- ASTM method E724 protocol, 2005. Conducting static toxicity tests starting with embryos of four species of saltwater bivalve molluscs. Environment Protection Agency.
- Bengtsson, B.-E. and Gergström, B., 1987. A flowthrough fecundity test with *Nitocra spinipes* (Harpacticoida Crustacea) for aquatic toxicity. *Ecotoxicity and Environmental Safety*. 14, 260-268.
- Bednarz, T., 1981. The effect of pesticides on the growth of green and blue-green algae cultures. *Acta Hydrobiol.* 23, 155–172.
- Bowmer T. and Linders Jan, 2010. A summary of findings from the first 25 ballast water treatment systems evaluated by GESAMP. *WMU Journal of Maritime Affairs*, Vol. 9 (2010), No.2, 223–230.
- Buchan, K.A.H., Martin-Robichaud, D.J., Benfey, T.J., 2005. Measurement of dissolved ozone in seawater: A comparison of methods. *Aquacult. Eng.* In press.
- Carlton J. T., 1999. The scale and ecological consequences of biological invasions in the world's oceans. In *Invasive species and biodiversity management*, edited by O. Sandlund, P. Schei & Å. Viken, pp. 195–212. Kluwer Academic Publishers.
- Cowgill, U.M., and Milazzo D.P., 1991. The Sensitivity of *Ceriodaphnia dubia* and *Daphnia magna* to Seven Chemicals Utilizing the Three-Brood Test. *Arch. Environ. Contam. Toxicol.* 20, 211-217.
- Environmental Protection Agency (EPA), 2010. Protocol for the Verification of Ballast Water Treatment Technologies. v5.1.
- Episuite 4.0 EPA toxicity estimation program. [http:// www.epa.gov/oppt/exposure/pubs/episuite.htm](http://www.epa.gov/oppt/exposure/pubs/episuite.htm)
- Erickson, S.J., and Hawkins C.E., 1980. Effects of Halogenated Organic Compounds on Photosynthesis in Estuarine Phytoplankton. *Bull. Environ. Contam. Toxicol.* 24, 910-915.
- EU TGD., 2003. European Technical Guidance Document on Risk Assessment in support of Commission Directive 93/67/EEC on Risk Assessment for new notified substances; Commission Regulation (EC) No 1488/94 on Risk Assessment for existing substances; Directive 98/8/EC of the European Parliament and of the Council concerning the placing of biocidal products on the market. http://ihcp.jrc.ec.europa.eu/our_activities/public-health/risk_assessment_of_Biocides/doc/tgd
- Fabbricino M. and Korshin G.V., 2005. Formation of disinfection by-products and applicability of differential absorbance spectroscopy to monitor halogenation in chlorinated coastal and deep ocean seawater. *Desalination*. 176, 57-69.
- Gentile J.H., Cardin J., Johnson M., and Sosnowski S., 1976. Power Plants, Chlorine and Estuaries. EPA-600/3-76-055, U.S.EPA, Narragansett, RI. 1-39.
- GESAMP, 1989. IMO/FAO/UNESCO/WHO/IAEA/UN/UNEP Joint Group of Experts on the Scientific aspects of Marine P, Reports and Studies No. 35.

Globallast, 2000. The Problem. Available from:

<http://globallast.imo.org/index.asp?page=problem.htm&menu=true>

- Gregg M., Rigby G. and Hallegraef G.M., 2009. Review of two decades of progress in the development of management options for reducing or eradicating phytoplankton, zooplankton and bacteria in ship's ballast water. *Aquatic Invasions*, Volume 4, Issue 3: 521-565.
- Groeneveld A.H.C., Zijlstra A.G., Feenstra A.F.M. and Berends, A.G., 1993. The acute toxicity of vinyl chloride to the zebra fish. *Solvay-Duzphar report 56635/64/92*.
- Hamzeh Y., Mortha G., Lachenal D., Hostachy J.C. and Calais C., 2006. Comparative studies of chlorine dioxide reactions with muconic acid derivatives and lignin model compounds. *Journal of wood chemistry and technology*. 26, 153-164.
- Heitmuller P.T., Hollister T.A., and Parrish P.R., 1981. Acute Toxicity of 54 Industrial Chemicals to Sheepshead Minnows (*Cyprinodon variegatus*). *Bull. Environ. Contam. Toxicol.* 27, 596-604.
- International Maritime Organisation (IMO), 2004. International Convention for the Control and Management of Ships' Ballast Water and Sediments. BWM/CONF/36. 16 February 2004.
- International Maritime Organisation (IMO), 2008a. Harmful aquatic organisms in ballast water. Report of the sixth meeting of the GESAMP-Ballast Water Working Group. MEPC 58/2/7, 14 July 2008.
- International Maritime Organisation (IMO), 2008b. Harmful aquatic organisms in ballast water. Report of the seventh meeting of the GESAMP-Ballast Water Working Group. MEPC 58/2/8, 28 July 2008.
- International Maritime Organisation (IMO), 2008c. Guidelines for approval of ballast water management systems (G8). MEPC 58/23, Annexe 4 Resolution MEPC. 174(58) adopted in October 2008.
- International Maritime Organisation (IMO), 2008d. Procedure for approval of ballast water management systems that make use of active substances (G9). MEPC 57/21, Annexe 1 Resolution MEPC.169(57) adopted in April 2008.
- International Maritime Organisation (IMO), 2009a. List of chemicals, requested by the GESAMP-BWWG, to be evaluated by the GESAMP-EHS. MEPC 59/2/13. Annex 1. March 2009.
- International Maritime Organisation (IMO), 2009b. Harmful aquatic organisms in ballast water. Report of the eighth meeting of the GESAMP-Ballast Water Working Group. MEPC 59/2/16, 8 April 2009.
- International Maritime Organisation (IMO), 2009c. Harmful aquatic organisms in ballast water. Report of the tenth meeting of the GESAMP-Ballast Water Working Group. MEPC 60/2/11, 30 October 2009.
- International Maritime Organisation (IMO), 2009d. Harmful aquatic organisms in ballast water. Report of the eleventh meeting of the GESAMP-Ballast Water Working Group. MEPC 60/2/12, 1 December 2009.
- International Maritime Organisation (IMO), 2010a. Harmful aquatic organisms in ballast water. Report of the twelfth meeting of the GESAMP-Ballast Water Working Group. MEPC 60/2/16, 8 February 2010.
- International Maritime Organisation (IMO), 2010b. Harmful aquatic organisms in ballast water. Report of the thirteenth meeting of the GESAMP-Ballast Water Working Group. MEPC 61/2/15, 9 June 2010.
- International Maritime Organisation (IMO), 2011a. Harmful aquatic organisms in ballast water. Report of the fifteenth meeting of the GESAMP-Ballast Water Working Group. MEPC 62/2/11, 24 February 2011.
- International Maritime Organisation (IMO), 2011b. Harmful aquatic organisms in ballast water. Report of the sixteenth meeting of the GESAMP-Ballast Water Working Group. MEPC 62/2/12, 8 April 2011.
- International Maritime Organisation (IMO), 2011c. Harmful aquatic organisms in ballast water. MEPC 63/2/6, 19 August 2011.
- International Maritime Organisation (IMO), 2011d. Harmful aquatic organisms in ballast water. Report of the nineteenth meeting of the GESAMP-Ballast Water Working Group. MEPC 63/2/11, 14 December 2011.
- International Maritime Organisation (IMO), 2011e. List of ballast water management system that make use of active substances which received basic and final approval. BWM.2 Circ. 34. 09 August 2011.
- International Maritime Organisation (IMO), 2012. List of ballast water management systems that make use of Active Substances which received Basic and Final Approval. BWM.2 Circ. 34. November 2012.
- International Standard ISO 10253, 2006. Water Quality – Marine algal growth inhibition test with *Skeletonema costatum* and *Phaeodactylum tricorutum*.
- International Standard ISO 14669, 1999. Water quality -- Determination of acute lethal toxicity to marine copepods (Copepoda, Crustacea)
- International Standard EN ISO/FDIS 20666, 2008. Water quality– Determination of the chronic toxicity to *Brachionus calyciflorus*.
- International Maritime Organisation (IMO), 2011e. List of ballast water management system that make use of active substances which received basic and final approval. BWM.2 Circ. 34. 09 August 2011.
- Ichihashi K., Teranishi K. and Ichimura A., 1999. Brominated trihalomethane formation in halogenation of humic acid in the coexistence of hypochlorite and hypobromite ions. *Water Research* 33 (2), 477-483.
- Japanese ministry of environment, 2008. From the IMO Gesamp group as reported in IMO document MEPC58/2-7.
- Kooijman S.A.L.M., 1981. Parametric Analyses of Mortality Rates in Bioassays. *Water Res.* 15, 107-119.

- Kuhn R. and Pattard M., 1990. Results of the Harmful Effects of Water Pollutants to Green Algae (*Scenedesmus subspicatus*) in the Cell Multiplication Inhibition Test. *Water Res.* 24(1): 31-38
- Linden E., Bengtsson B.E., Svanberg O., and Sundstrom G. (1979) The Acute Toxicity of 78 Chemicals and Pesticide Formulations Against Two Brackish Water Organisms, the Bleak (*Alburnus alburnus*) and the Harpacticoid *Nitocra spinipes*. *Chemosphere* 8, 843-851.
- Liltved H., Vogelsang C., Modahl I. and Dannevig B, 2006. High resistance of fish pathogenic viruses to UV irradiation and ozonated seawater. *Aquacultural Engineering*, 34, 72-82.
- MAMPEC (2008) MAMPEC v2.5 Release Notes, November 2008. <http://www.deltares.nl/en/software/1039844/mampec/1232321>
- Mayes M.A., Blanchard F.A., Hopkins D.L., and Takahashi I.T., 1985. Static Acute Toxicity of Dibromonitripropionamide and Selected Degradation Products to the Fathead Minnow (*Pimephales promelas Rafinesque*). *Environ. Toxicol. Chem.* 4, 823-830.
- MITI, 1992. Ministry of International trade and Industry Japan. Ed. Chemicals inspection & testing institute Japan. Published Japan Chemical Industry Ecology-Toxicology & Information Center.
- Norwegian standard NS-EN ISO 10304-1, 2009. Water quality - Determination of dissolved anions by liquid chromatography of ions - Part 1: Determination of bromide, chloride, fluoride, nitrate, nitrite, phosphate and sulfate.
- Norwegian standard NS-EN ISO 23631, 2006. Water quality - Determination of dalapon, trichloroacetic acid and selected haloacetic acids - Method using gas chromatography (GC-ECD and/or GC-MS detection) after liquid-liquid extraction and derivatization.
- OECD, 1992. Guideline for Testing of Chemicals 203: Fish, Acute toxicity test.
- OECD, 2000. Guideline for Testing of Chemicals 215: Fish Juvenile Growth Test.
- Perrins J.C., Cooper W., van Leeuwen J.H and Herwig R.P., 2006. Ozonation of seawater from different locations: formation and decay of total residual oxidant-implications for ballast water treatment. *Marine Pollution Bulletin.* 52, 1023-1033.
- Rosemarin A., Lehtinen K.J., Notiti M., and Mattsson J., 1994. Effects of Pulp Mill Chlorate on Baltic Sea Algae. *Environ. Pollut.* 85, 3-13.
- Pimentel D., Zuniga R., Morrison D. 2005. Update on the environmental and economic costs associated with alien-invasive species in the United States. *Ecological Economics.* 52, 273.
- Richardson S.D., Plewa M.J., Wagner E.D., Schoeny R. and Demarini D.M., 2007. Occurrence, genotoxicity, and carcinogenicity of regulated and emerging disinfection byproducts in drinking water: a review and roadmap for research. *Mutation Research* 636 (1-3), 178-242.
- Royan F., 2010. Shipping Industry Sets Sail: Multi-billion Dollar Ballast Water Treatment System Market. <http://www.frost.com/prod/servlet/market-insight-top.pag?docid=203055815>.
- Ruiz G.M., Carlton J.T., Grosholz, E.D. and Hines A.H., 1997. Global invasions of marine and estuarine habitats by non-indigenous species: Mechanisms, extent and consequences. *Amer. Zool.*, 37, 621-632.
- Shi H., Qiang Z. and Adams C., 2013. Formation of haloacetic acids, halonitromethanes, bromate and iodate during chlorination and ozonation of seawater and saltwater of marine aquaria systems.
- U.S. Coast Guard, 2012. Standards for Living Organisms in Ships' Ballast Water Discharged in U.S. Waters; Final Rule. Federal register Part V. Vol 77, No. 57.
- U.S. Environmental Protection Agency, 1978. In-Depth Studies on Health and Environmental Impacts of Selected Water Pollutants. U.S.EPA Contract No.68-01-4646, Duluth, MN. 1-9.
- US EPA 524.2, 1995. Measurement of Purgeable Organic Compounds in Water by Capillary Column Gas Chromatography/Mass Spectrometry. Revision 4.1.
- US EPA 551.1, 1990. Determination of chlorination disinfection byproducts, chlorinated solvents, and halogenated pesticides/herbicides in drinking water by liquid-liquid extraction and gas chromatography with electron-capture detection.
- Tsolaki E. and Diamadopoulos E., 2009. Technologies for ballast water treatment: a review. *J Chem Technol Biotechnol*, 2010; 85: 19-32.
- Yoshioka Y., Ose Y. and Sato T., 1985. Testing for the Toxicity of Chemicals with *Tetrahymena pyriformis*. *Sci. Total Environ.* 43, 149-157.
- Werschkun B., Sommer Y. and Banerjee S., 2012. Disinfection by-products in ballast water treatment: An evaluation of regulatory data. *Water Research.* 46, 4884-4901.
- Westerhoff P., Chao P. and Mash H., 2004. Reactivity of natural organic matter with aqueous chlorine and bromide. *Water Research*, 38(6), 1502-1513.
- World Health Organisation, 2003. Guidelines for safe recreational water environments. Volume 1: Coastal and fresh waters.
- World Health Organisation, 2011. Guidelines for Drinking Water Quality, Fourth ed. Geneva.

World Wide Fund For Nature (WWF), 2009. Silent Invasion – The spread of marine invasive species via ships' ballast water. http://awsassets.panda.org/downloads/silent_invasion_briefing.pdf (last accessed 29 February 2012).