CATHELCO LTD
BALLAST WATER MANAGEMENT SYSTEM

PART 4 ANNEX 2b

MEA Report on Shipboard Tests

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MEA-NL
MARINE ECO ANALYTICS

MEA-nl B.V. is an independent research and test facility

Ship board testing of Cathelco's Ballast Water Treatment System on board MV "Eddystone"

– Final Report

Produced by MEA-nl B.V., The Netherlands
Date: August 2013
Version 1.1

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	Den Oever	02. 09. 2013
Signature of responsible scientist	Place	Date

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1. Summary

Over a period of six months, five independent test runs were performed to evaluate the performance of the Cathelco Ballast Water Treatment System. Performances of the system were according to specifications and meet the D2 standard of the IMO BWM Convention (IMO, 2004) for the organism size classes tested. Test run five was rejected based on an operator fault. Regarding biological efficiency, the tested system proved to be capable of operating reliable and consistently under the ship's operational conditions.

2. Introduction

Between November 2012 and April 2013 a total of five independent test runs were conducted on board the MV *Eddystone* (UK flag) in order to assess the performance of the installed Cathelco Ballast Water Treatment System (BWTS) against organisms. This time span is in accordance with the duration requirements as described in guideline G8 of IMO (Anon. 2008) and agreed upon with the certifying administration. The latter being the Bundesamt für Seeschifffahrt und Hydrographie (BSH), Germany. Tests were generally conducted according to the test protocol submitted to the BSH before starting the ship board test program. Deviations of the test protocol and an evaluation of the possible influence on the test results will be discussed as appropriate. However, this report does not document the technical performance of the system other than where it affected the analytical procedures performed by MEA-nl. The technical performance of the system during ship board tests is documented in another report.

For test run I and II MEA-nl submitted a preliminary data report each. Due to the short time span between test runs III to V, no preliminary reports on the latter were submitted. The preliminary data reports are not part of the certification process. Furthermore not all data were available at time of submission of the preliminary reports. Therefore all five test runs will be described in this document.

Intake conditions varied from open oceanic (test runs I and IV), intermediate (test run V) to coastal (test runs II and III). Therefore the five test runs covered all marine water types encountered by the ship during normal operation. Fresh-water has not been tested, since the MV *Eddystone* did not operate in fresh-water at all during the six month test period.

A ship under normal operating conditions is not a controlled test facility. In order to assess the system's performance rather than external factors, the impact of the ship's operation on the system should be taken into consideration. Furthermore does the test regime as mandated by Guideline G8 of IMO (Anon. 2008) have an impact on the system.

The most severe and most obvious negative effect on BWTS performance results from pumping untreated water. This is both due to the requirement of a control for the tests and the fact that the convention (IMO, 2004) is not into force yet. Under normal (future) operational conditions, no untreated water will be pumped through the ballast system behind the BWTS, thereby minimizing the risk of tank contamination. This technical and operational factor turned out to be the reason of the rejection of test run V.

Since the BWM Convention is still not into force, crewmembers have very different levels of knowledge and experience with the ballast-water treatment system installed. It is not part of the operational routine yet. The latter effected in particular test runs III and V.

Different experts of MEA-nl are responsible for the analysis and reporting of different size classes of organisms (table 1). As a result the report is dividing the results into different sections based on groups rather than presenting each test run separately. The report ends with a general conclusion.

Table 1: Scientists involved in ship board testing and their responsibilities

Frank Fuhr	Testing on board, Plankton larger than 50 micrometre
Marcel Veldhuis	Phytoplankton, Bacteria
Isabel van der Star	Microzooplankton smaller than 50 micrometre

3. General information on test runs

Test runs are numbered in roman numerals from I to V. Replicate samples are numbered 1 to 3. Ballast-tanks of the ship are numbered 1 to 16, but not all were used for testing. Ballast tanks 9 and 10 were used for the storage of treated water for all 5 test runs. Control tanks were assigned depending on ship's operational procedures.

3.1 Test run I

The first test was conducted from the13th until 16th of November 2012. At time of start of the actual testing procedure the ship was in transit, north of Gran Canaria (Canary Islands) heading towards Marchwood (UK). Intake was on November 13th P.M. and discharge in the afternoon of November 16th, resulting in an effective holding time of 3 days.

As can be seen in table A1, annex 1 during intake of the treated water approximately 23 kilometres were travelled. Intake of control water was done as soon as practically possible. Between the stop of the treated intake and the start of the control 37 kilometres were travelled. During the intake of control water approximately 18 kilometres were travelled. The whole test therefore covers a travelled distance of just under 80 kilometres. As can be derived from the physical parameters measured, the water quality itself did not change significantly over this distance. The water depth at the sampling transect varied between 4000 and 4200 metres.

Ballast water tanks used for the tests were flushed with oceanic water 4 times, prior to the test run and once more due to an aborted test run earlier on the 13th. Residual volumes that could not be discharged without stripping are given in table A1, annex 1. These residual volumes are in accordance with the ballasting practice on board MV *Eddystone*, their presence was acceptable. Residual volumes pose an extra challenge for pure in-line treatment systems (as Cathelco's UV system) since the residual water does contain organisms which will be treated only at discharge.

Sampling volume for the larger organisms during this test run deviated from the test protocol due to logistical reasons. This was reported in the preliminary data report (MEA-nl 2012b) and further analysed in two additional information papers (MEA-nl 2012c and 2012d). In terms of data quality and analysis of the samples the volume did not have adverse effects on the test run.

3.2 Test run II

From January 11th till 16th, 2013 the second ship board test was conducted. Ballast water intake was done in the harbour of Marchwood (UK). The ship left for Gibraltar, where it arrived early in the morning of the 16th. Intake was on January 11th P.M. and discharge four days later in the morning of January 15th, resulting in an effective holding time of 3,5 days. This is similar to test I. During intake of the control water, the ship moved 250 metres from its original pier to another. This movement did not have any influence on the tests conditions.

3.3 Test run III

From February 28th till March 3rd, 2013 the third ship board test was conducted. During intake the ship remained in the harbour of Marchwood (UK). During this period the crew changed and the new crew was unaware of the status, procedures and implications of an on-going test. Therefore the two control tanks (nos. 3 and 4) were discharged without prior notice. A third tank (no. 16) was also filled during intake on the 28th. This tank had roughly 190 cubic meters in it, which subsequently were used as discharge control.

3.4 Test run IV

At intake the ship was anchored off Las Palmas de Gran Canaria (Canary Islands). Water conditions on the anchorage qualified as oceanic, rather than coastal, despite the shallow waters (fig. 1). Intake was on April 18th and discharge three days later on April 21st, resulting in an effective holding time of three days.

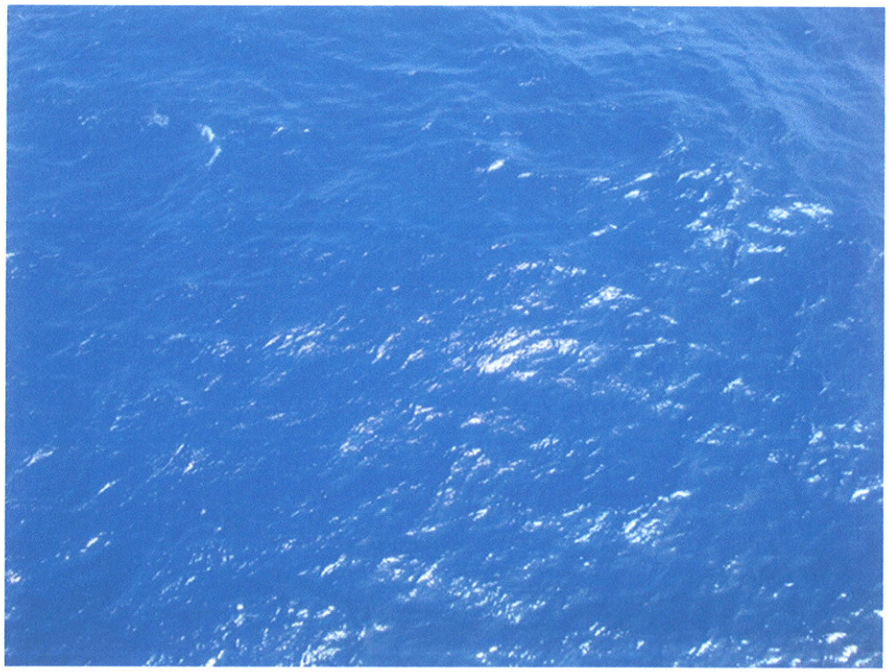

Figure 1: Water conditions at the anchorage of Las Palmas de Gran Canaria on April 18th. Water was classified as oceanic

3.5 Test run V

At intake the ship was in transit, at the southern entrance of the English Channel off the coast of France (near Brest), heading towards Marchwood (UK). Water conditions were assessed as intermediate between true coastal and oceanic (figure 2). Intake was on April 23rd and discharge five days later on April 28th.

As can be derived from table A5, annex 1, during intake of the treated water approximately 32 kilometres were travelled. Intake of the control water took place as soon as practically possible. Between the stop of the treated intake and the start of the control intake the ship sailed approximately 25 kilometres. During the actual intake of control water approximately 22 kilometres were travelled. The whole test therefore covers a travelled distance of just under 80 kilometres. As can be derived from the physical parameters measured, the water quality itself did not change significantly over this distance.

Due to communicational issues approximately 50 cubic meters of untreated water was pumped into tank 9, one of the two tanks used for treated water. This was done after the initial intake of treated water. This was only discovered after discharge, when the first sample for larger organisms had been analysed. A comparison of the laboratory log, the operators log and the ballast log of the bridge showed what happened. A malfunction of the system and direct contamination of the sample could be ruled out.

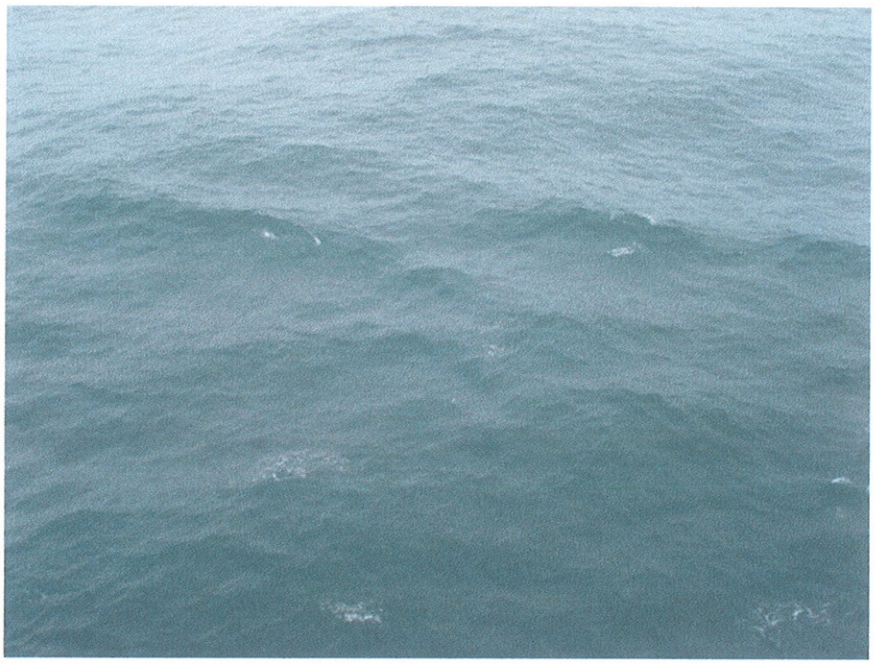

Figure 2: Water conditions at the entrance of the English Channel on April 23rd. Water was classified as intermediate

4. Results

4.1 Organisms larger than 50 micrometre

Averages of discharge results after treatment are given in table 1. A compilation of all results can be found in tables A6 through A10, annex 2. Test runs I to IV were in compliance with regulation D2 (IMO, 2004). Test run V was not compliant for the reason described in section 3.5 (accidental pumping of circa 50 cubic meters of untreated water into the treatment tank).

Table 2. Averages and standard deviations of the three samples taken of the treated water on discharge.

Test run	I	II	III	IV	V
Average of 3 samples upon discharge	2,00	2,67	0,67	0,67	25,33
Standard deviation	2,00	1,53	0,58	1,15	43,88

As can be seen by the comparatively high number of organisms on discharge from test V the BWT system could not cope with roughly one sixth of the total volume being pumped into the tank of untreated and therefore rich in viable organisms. The volumes of tank 9 and 10 were regarded as one for the purpose of testing and as such discharged together.

The accidental action did however allow to make an assumption of the mortality rate of a single passage of larger organisms, which were normally retained by the filter at intake, through an UV-reactor. The population encountered upon intake for test V was quite healthy. This can be seen from the survival rate of 62% (averaged) in the control after five days holding time. During discharge the individual counts were 76,0 and 0 per cubic meter, (table A10, annex 2). Assuming a survival rate equal to the control, that would mean, that a single pass through the UV treatment without filtration killed approximately 95% of the larger zooplankton.

In terms of certification, test run V was obviously invalid. However the data from this test show that the BWTS is quite robust and that the second treatment on discharge can contribute substantially to the performance even in case of a failure of the filter at intake. Supporting evidence can be derived from test runs I and II. In the discharge samples from those test runs many freshly killed Harpacticoida were found. These benthic organisms were absent at intake for test run I. In test run II upon intake some Harpacticoida were found, but in low numbers. Upon discharge they were the most common, identifiable remains (fig. 3). Therefore it can be concluded, that these organisms originated from a population living inside the ballast tank itself. These organisms were only treated by UV once, upon discharge. Still the BWTS performed well within the limits of regulation D2.

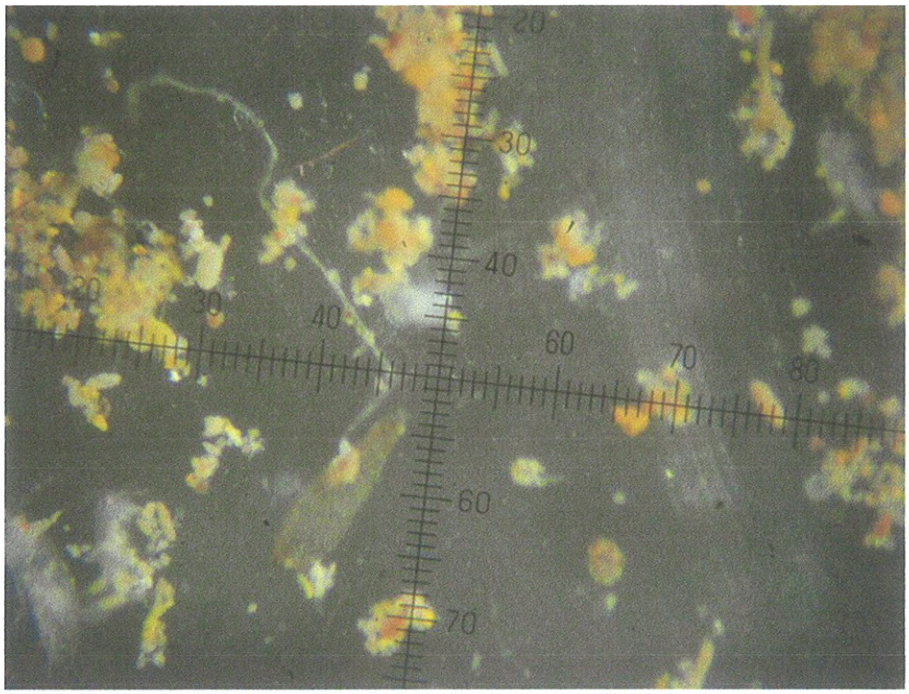

Figure 3: Fresh dead harpacticoid copepods from a treated discharge sample (test run I)

As mentioned earlier, pumping of untreated water will be phased out once the convention (IMO, 2004) comes into force. In the future pumping of untreated water should only occur as an emergency measure but no longer as part of the routine operation. This will minimize the contamination risk. The test runs performed on board the MV *Eddystone* show that the Cathelco BWTS is capable of dealing with residual volumes of untreated ballast water and the organisms of this size class therein which might be encountered on vessels undergoing a retro fit.

4.2 Organisms between 10 and 50 micrometre

4.2.1 Phytoplankton

Phytoplankton samples for total biomass and photosynthetic efficiency measurement (PAM) were taken at three different sampling times effectively covering the entire intake and discharge period. Analysis was usually (i.e. test runs I, II intake, IV and V) conducted immediately on board of the vessel. In case of test runs II and III this was not possible for logistical reasons. Analysis of discharge water for test run II was conducted two days after sampling. Intake water of test III was tested five and discharge water two days after sampling, respectively.

Besides single end-point measurement also a light response curve was taken of control and treated water to gain more insight in the kinetics and recovery of the photosynthetic apparatus.

Samples, varying in volume of 50 to 1000 mL, for flow cytometric analysis were stored in a refrigerator in the dark to prevent degradation and regrowth of the phytoplankton community. Analysis was usually within a few days with the exception of the intake samples of test run III and IV which were analysed after five and seven days respectively.

A long storage time resulted in a decline in total chlorophyll biomass as measurements with the PAM (up to 30%). The consequence of this is also a decline in total cell numbers of phytoplankton, including the fraction larger than 10 micron. Nevertheless,

in the first three test runs cell numbers of the larger fraction at intake were well above the required minimum (table 4).

Intake of the control and treated water (t0) and at discharge of the whole phytoplankton community (PAM) and total phytoplankton as well as the phytoplankton fraction larger than 10 micron is presented in the following tables separated for each of the five test series.

4.2.1.1 Photosynthetic efficiency measurement; indicator of phytoplankton viability (PAM)

PAM measurements indicated that phytoplankton community at intake was in a healthy and viable condition. In all cases the values well exceeded the Fv/Fm value of 0.3 which is considered as an arbitrary boundary between healthy and viable (above) and phytoplankton experiencing stress (below, Table 3). A prolonged holding time in the dark, varying from 3 to 5 days negatively affected the viability of the phytoplankton community resulting in all 5 test runs in a decline in the Fv/Fm .The largest decline in viability was observed in the control tanks of test run II and III. But in each case the Fv/Fm value was still above critical value of 0.1, below which phytoplankton is considered to be a non-viable status of the phytoplankton cells.

The UV-disinfection showed an immediate response to the photosynthetic efficiency of the phytoplankton community resulting in an instantaneous decline in the Fv/Fm ratio close to or to a value well below 0.1 (table 3). The second disinfection step with the UV-reactor, at discharge, resulted in a further decline in the phytoplankton viability, often to values close to the detection limit (0). Based on the viability measurements of the PAM it can be concluded that not only the phytoplankton with a cell diameter larger than 10 micron but the whole community has effectively been treated and should be considered as non-viable.

Table 3: Phytoplankton photosynthetic efficiency at intake (t0) and discharge (number indicates days after intake) of the control and UV-disinfected water. Data separated for 5 test runs and averaged for total number of measurements (#), minimum (min) and maximum (max) values measured

I	**Fv/Fm**			
time (d)	C-t0	C-t3-dis	T-t0	T-t3-dis
average	0.580	0.263	0.006	0.004
stdev	0.047	0.090	0.007	0.011
#	27	27	27	27
min	0.440	0.168	0.000	0
max	0.644	0.542	0.024	0.047

II	**Fv/Fm**			
time (d)	C-t0	C-t4-dis	T-t0	T-t4-dis
average	0.336	0.137	0.0598	0.006
stdev	0.028	0.038	0.028	0.004
#	6	12	19	12
min	0.302	0.092	0.04	0
max	0.377	0.229	0.131	0.011

III	Fv/Fm			
time (d)	C-t0	C-t3-dis	T-t0	T-t3-dis
average	0.618	0.108	0.129	0.055
stdev	0.007	0.008	0.036	0.022
#	9	9	9	9
min	0.61	0.09	0.08	0.02
max	0.65	0.13	0.22	0.1

IV	Fv/Fm			
time (d)	C-t0	C-t3-dis	T-t0	T-t3-dis
average	0.764	0.315	0.047	0.066
stdev	0.059	0.058	0.059	0.066
#	9	9	9	9
min	0.646	0.242	0.000	0.000
max	0.841	0.390	0.164	0.150

V	Fv/Fm			
time (d)	C-t0	C-t5-dis	T-t0	T-t5-dis
average	0.618	0.431	0.001	0.009
stdev	0.009	0.016	0.002	0.008
#	9	9	9	9
min	0.606	0.405	0.000	0.000
max	0.633	0.455	0.005	0.024

Light induction and recovery experiments

Besides single end-point measurements of each sample an activity light-induction and recovery experiments was conducted to examine the phytoplankton photosynthetic response in more detail.

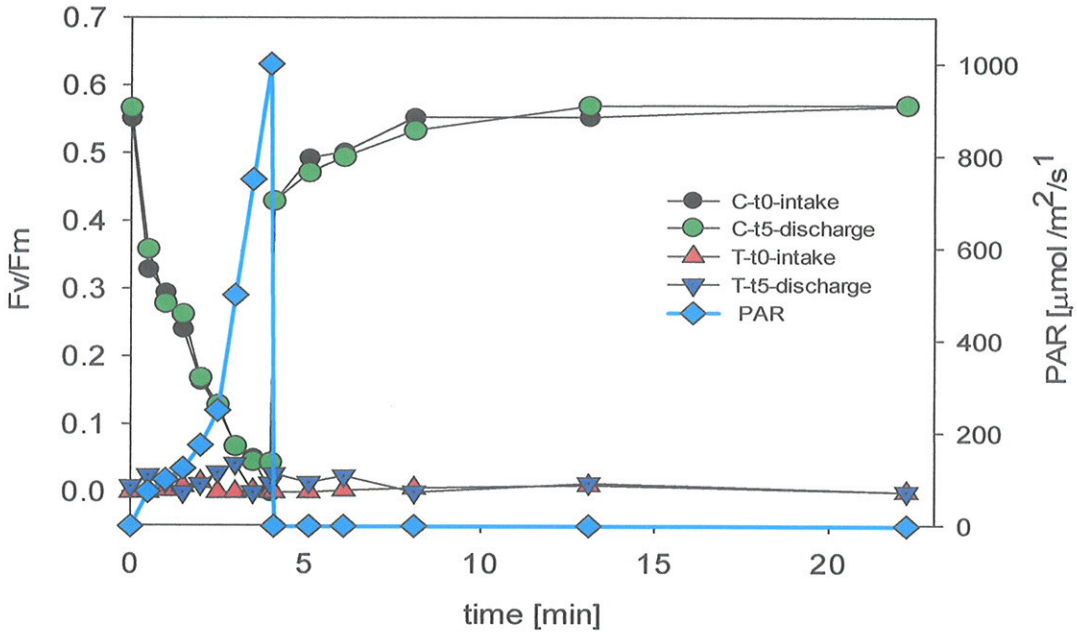

Figure 4: Light induction and recovery of the photosynthetic efficiency of the photosynthetic apparatus the phytoplankton of control, treated water at intake (t0) and discharger (t5) over time. Light (PAR) increases in intensity in the first 4 min and is followed by dark recovery of 18 min

Figure 4 is a typical light induction curve observed during all 5 test series and therefore representative of all test runs. The response of the phytoplankton in the control results in a typical decline with increasing incident irradiance (up to 1000 $\mu Mol.m^{-2}.s^{-1}$, first 4 min of the response curve). This was followed by a very sharp and full recovery after dark (period from 4 to 22 min). In contrast the UV-disinfection of the BWT system has damaged the photosynthetic apparatus to such and extend that there is no response to either the light or recovery in the subsequent dark phase. This typical pattern of response of the photosynthetic apparatus has been observed during all 5 test series.

4.2.1.2 Phytoplankton cell numbers

Measurements of the phytoplankton cell counts (flow cytometric cell counts, table 4) showed that the total cell number declined substantial after the UV-disinfection step. Not only the fraction larger than 10 micrometre was affected but the entire phytoplankton community. With respect to the IMO relevant fraction (larger sized plankton) discharge numbers were in compliance for the test runs III, IV and V but slightly above the value of less than 10 per mL for the first 2 test runs. While these cells were identified as whole phytoplankton cells, containing chlorophyll, the PAM measurements indicated that the remaining phytoplankton cells at discharge should be identified as non-viable. Based on the combined observations it can be concluded that these cells should be classified as non-viable and therefore these test runs were also considered to be in compliance.

However, in terms of the necessary numbers in the control at intake phytoplankton numbers were too low in test runs IV and V and therefore in terms of numerical abundance do not meet the IMO requirements. Nevertheless, the UV-disinfection was similar effective as in the other test runs. For test run IV this was to be expected since the ballast water treatment procedure was conducted in spring in the open ocean.

Surface waters in the open oceans during that season are characterized by very low abundance of phytoplankton in the size class of a cell diameter larger than 10 micrometre.

Table 4: Average counted number, standard error and number of measurements, minimum (min) and maximum (max) counted value of total phytoplankton community and the fraction will cell diameter larger than 10 micrometre

I total phytoplankton number per mL					**phytoplankton number > 10 micron per mL**			
time (d)	C-t0	C-t3-dis	T-t0	T-t3-dis	C-t0	C-t3-dis	T-t0	T-t3-dis
average	1975	1743	717	229	248	178	63	17
stdev	140.9	41.3	280.8	52.2	36.1	44.2	31.1	9.9
#	3	3	3	3	3	3	3	3
min	1824	1695	519	171	207	136	31	10
max	2102	1769	1038	274	274	224	93	29

II total phytoplankton number per mL					**phytoplankton number > 10 micron per mL**			
time (d)	C-t0	C-t4-dis	T-t0	T-t4-dis	C-t0	C-t4-dis	T-t0	T-t4-dis
average	679	358	525	384	104	18.3	36	12.3
stdev	25.7	49.8	58.6	61.7	9	8.1	6.7	10.2
#	3	6	6	6	3	6	6	6
min	650	264	460	288	98	7	23	0
max	700	402	604	447	114	28.6	43	23.8

III total phytoplankton number per mL					**phytoplankton number > 10 micron per mL**			
time (d)	C-t0	C-t3-dis	T-t0	T-t3-dis	C-t0	C-t3-dis	T-t0	T-t3-dis
average	1390.3	84.5	755.9	509.5	100.9	12.0	66.2	4.9
stdev	68.9	109.4	86.8	106.4	27.0	4.0	42.1	4.3
#	9	9	9	9	9	9	9	9
min	1200	8	640	0	56	4	4	0
max	1500	564	1024	660	148	116	28	20

IV total phytoplankton number per mL					**phytoplankton number > 10 micron per mL**			
time (d)	C-t0	C-t3-dis	T-t0	T-t3-dis	C-t0	C-t3-dis	T-t0	T-t3-dis
average	9952	4010	14818	566	8.0	4.0	300.0	0.0
stdev								
#	1	2	1	2	1	2	1	2
min	-	3970	-	376	-	0	-	0
max	-	4050	-	756	-	8	-	0

V total phytoplankton number per mL					phytoplankton number > 10 micron per mL			
time (d)	C-t0	C-t5-dis	T-t0	T-t5-dis	C-t0	C-t5-dis	T-t0	T-t5-dis
average	24646	9938	6967	5969	63.4	25.3	42.0	5.3
stdev	1596.5	384.5	656.7	149.3	22.3	13.4	9.2	4.5
#	9	9	9	9	9	9	9	9
min	22439	9212	6116	5718	34	8	28	0
max	27263	10420	7929	6202	98	48	56	12

4.2.2 Micro zooplankton

Subsamples of the size class of organisms of 10 micron or bigger but smaller than 50 micron are taken as discrete samples (minimum 3) evenly distributed over the whole sampling period. This is done for both control and treated samples at intake and discharge. One-liter samples are taken for microscopic counts of total protists and preserved with Lugols solution. Concentrated samples were microscopically analyzed using the stain Bengal Rose.

Table 5: Micro zooplankton in control and treated samples at intake and discharge

SOP319	micro zooplankton / ml			
Test run	control intake	control discharge	treated intake	treated discharge
I	0,06	0,98	1,24	0,00
II	0,16	0,18	0,03	0,16
III	0,57	0,00	0,23	0,20
IV	0,17	0,04	0,00	0,00
V	4,43	0,40	4,74	0,09
average	1,08	0,32	1,25	0,09

Results indicate that this specific group in this size class has a low abundance in the challenge waters used. Although studies on regional protist abundance are comparatively sporadic, this result was expected. Maximum numbers for ciliates in the Southampton harbour area (intake site for tests II and III) were found to be 16 per mL in a one year monitoring program (Leaky et al., 1992). However, these maxima were not observed during winter and the high velocity and pressure caused by the ballast pump are likely to impact protists. The latter was not quantified in the scope of this study. Analysis after treatment at discharge show a substantially reduced number of micro zooplankton.

4.3 Bacteria

Because of the often long timespan between sampling and transfer to the laboratory and the fact that in several cases water intake was in the open ocean it was decided to omit sampling for human pathogens (*E. coli* and enterococci) during this research. This was done in line with section 2.2.2.6.3.3 of guideline G8 (Anon. 2008; p.17) recognizing the possible limited applicability of testing for human pathogens.

However, total bacteria numbers were counted flow cytometrically using formaline preserved samples. It should be noted that bacteria were not discriminated as life or dead in this method. Measurements showed the presence of three distinct different subpopulations of bacteria based on their cellular DNA content. Based on the numerical distribution (table 6) and figure 5 it can be concluded that neither the UV-disinfection

treatment nor the holding period of the ballast water in the tanks affect bacteria numbers in any clear and significant positive or negative manner.

Table 6: Total bacteria numbers at intake and discharger in the control and treated water of the 5 test runs. Time of discharge varied (see section 3)

total bact/mL				
test run	C-t0	C-t-dis	T-t0	T-t-dis
I	1.1E+06	1.3E+06	1.0E+06	1.2E+06
II	5.7E+05	1.1E+06	6.1E+05	6.9E+05
III	9.2E+05	6.9E+05	1.0E+06	1.0E+06
IV	1.3E+06	1.6E+06	1.2E+06	1.3E+06
V	1.5E+06	2.1E+06	1.5E+06	2.6E+06

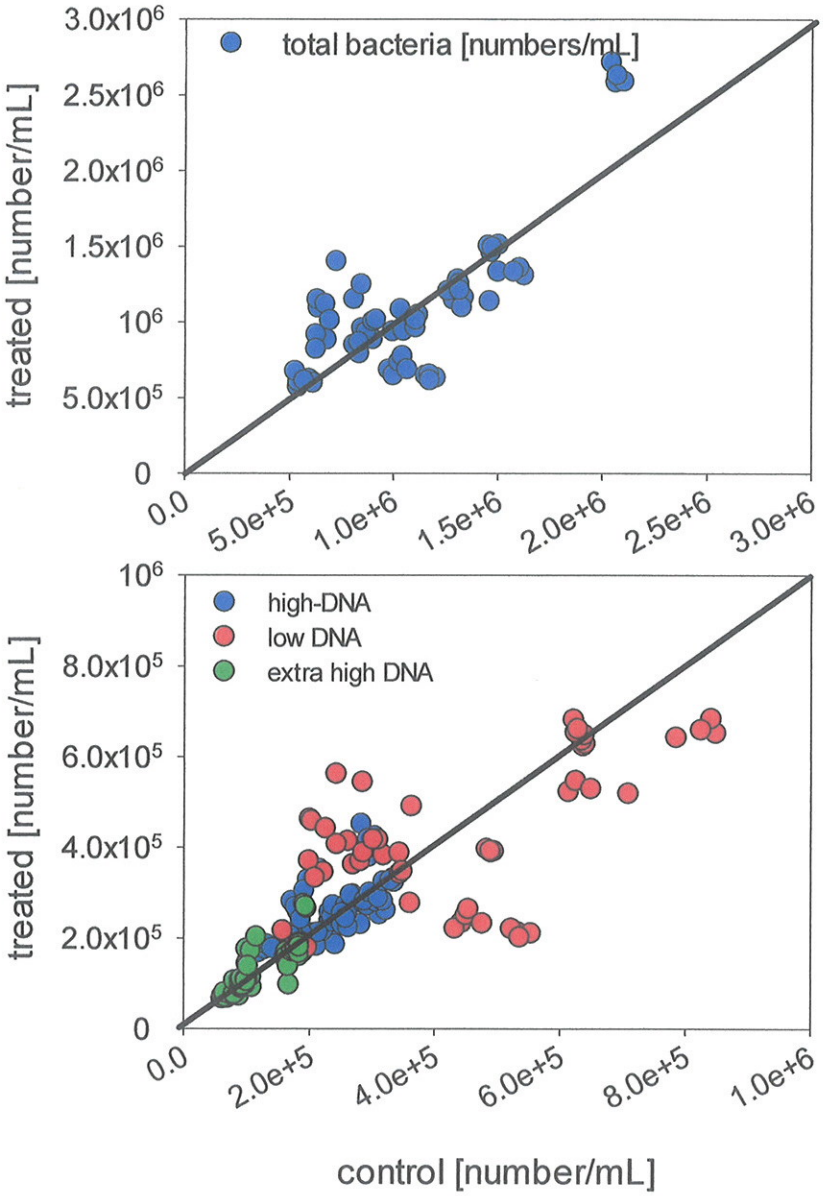

Figure 5: Total and three selected subpopulation based on their DNA content of bacteria in the control versus treated water samples. Lines indicate 1:1 relationship

4.4 Abiotic parameters

The parameters required by guideline G8 (Anon. 2008) to be reported are: salinity, temperature, total suspended solids (TSS) and particulate organic matter (POC). These were measured. The data can be found in annex 3. Within the encountered ranges, none of the parameters required did influence the performance of the system, sampling or sample analysis. They showed that the water-masses encountered during intake for test run I and V respectively did not change significantly during pumping.

Water temperature in the tanks changed according to the ship's location. It did not show any correlation with plankton survival.

5. Conclusion and recommendations

Even though some problems occurred during the campaign, the data collected are robust and cover all major parameters required according to the G8 guidelines. Sampling and analysis of samples could be performed up to standard at all times. In conclusion the results show that the Cathelco BWTS is capable of meeting Regulation D2 under operational conditions on board of a ship.

The problems encountered during test run III and V highlight the need to raise awareness among seamen about the ballast water issue. Furthermore it would be advisable to have the ballast water system running during all ballasting operations and not only during tests. That would reduce the risk of unintentional contamination. In this way one could also raise more acceptance for the new technology. Last but not least, more data concerning the technical performance of any system under operational conditions would become available much faster.

6. Acknowledgements

We would like to thank the captain and crew of MV *Eddystone* for their continued support despite our repeated disturbance of their working routine. We are very grateful for the hospitability and warm welcome we experienced during the five test runs.

7. Literature

7.1 Published documents cited in the report

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7.2 Published documents used in the preparation of the report but not cited

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7.3 Non-public documents

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- MEA-nl. 2012b. Preliminary data report: First ship board test for the verification of the UV ballast-water treatment system of Cathelco Ltd.

- MEA-nl. 2012c. Factors influencing sampling and sample volume for the group larger than 50 microns. Internal document compiled by MEA-nl for use by the BSH.

- MEA-nl. 2012d. Background information relevant for volume dependent organism counts. Internal document compiled by MEA-nl for use by the BSH.

- MEA-nl. 2013. Preliminary data report: Second ship board test for the verification of the UV ballast-water treatment system of Cathelco Ltd.

8. Annex 1: General information on test runs

Table A 1. Volumes, times and position information for the intake of test run I.

Tank	Type water	Tankvol. Start [m3]	Start time	Start position		Tankvol. end [m3]	End time	End position	
				LAT	LON			LAT	LON
9	Treated	27,68	16:36	32°44,2 N	13°44,6 W	170,36	17:40	32°57,9 N	13°37,9 W
10	Treated	56,63	16:36	32°44,2 N	13°44,6 W	161,64	17:40	32°57,9 N	13°37,9 W
5	Control	18,84	18:15	33°18,9 N	13°33,6 W	116	18:43	33°28,7 N	13°30,7 W
6	Control	30,1	18:15	33°18,9 N	13°33,6 W	110	18:43	33°28,7 N	13°30,7 W

Table A 2. Volumes and time information for the intake of test run II. Ship was in harbour during intake, therefore no position information is provided.

Tank	Type water	Tankvol. Start [m3]	Start time	Tankvol. end [m3]	End time
9	Treated	30,91	11:19	118,81	12:08
10	Treated	26,18	11:19	199,4	12:08
7	Control	41,5	12:50	167,91	13:27
8	Control	33,89	12:50	173,6	13:27

Table A 3. Volumes and time information for the intake of test run III. Ship was in harbour during intake, therefore no position information is provided. * = volume sensor malfunctioned. No reading. Due to accidental discharge, this did not further impact sampling (see section 2.3 for more details).

Tank	Type water	Tankvol. Start [m3]	Start time	Tankvol. end [m3]	End time
9	Treated	29,74	16:10	191,94	17:05
10	Treated	23,56	16:10	182,46	17:05
3	Control	15,50	18:09	69,60	18:50
4	Control	*	18:09	*	18:50
16	Control	23,50	18:09	214,66	18:50

Table A 4. Volumes and time information for the intake of test run IV. Ship was at anchorage during intake, therefore no position information is provided.

Tank	Type water	Tankvol. Start [m3]	Start time	Tankvol. end [m3]	End time
9	Treated	23,26	16:50	133,48	17:45
10	Treated	23,06	16:50	189,08	17:45
16	Control	64,92	18:48	216,55	19:27

Table A 5. Volumes, times and position information for the intake of test run V. Please note the end volume of tank 9. The first value is the volume recorded when ending the test. The volume in brackets is the volume in the tank the next morning at 06:00 as given in the ship's ballast log-book (see section 2.5 for more details).

Tank	Type water	Tankvol. Start [m3]	Start time	Start position		Tankvol. end [m3]	End time	End position	
				LAT	LON			LAT	LON
9	Treated	39,24	13:00	49°04,6 N	4°50,2 W	136,16 (189,44)	14:03	49°11,2 N	4°32,9 W
10	Treated	26,37	13:00	49°04,6 N	4°50,2 W	197,06	14:03	49°11,2 N	4°32,9 W
16	Control	13,87	14:46	49°16,6 N	4°18,8 W	193,32	15:20	49°21,2 N	4°06,6 W

9. Annex 2: Biological results

Table A 6. Plankton larger 50 micrometre in test run I.

	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
Treated	0	0	0	2	0	4
Control	700	725	750	675	475	500

Table A 7. Plankton larger 50 micrometre in test run II.

	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
Treated	4	2	2	4	3	1
Control	8.325	10.925	10.475	4.100	7.350	7.850

Table A 8. Plankton larger 50 micrometre in test run III.

	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
Treated	0	1	1	0	1	1
Control	5.175	3.150	2.950	250	375	425

Table A 9. Plankton larger 50 micrometre in test run IV.

	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
Treated	3	0	2	0	0	2
Control	2.025	1.875	2.525	325	675	1475

Table A 10. Plankton larger 50 micrometre in test run V.

	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
Treated	0	1	0	76	0	0
Control	6.400	6.750	2.875	1.775	3.750	4.400

10. Annex 3: Abiotic parameters measured

Test run I

Table A 11. TSS and POC data from test run I.

mg/L	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
TSS - C	3,54	4,24	4,38	5,60	4,52	6,03
TSS - T	3,56	3,20	3,32	5,19	5,17	6,28
POC - C	1,16	1,38	1,52	1,84	1,44	1,83
POC - T	1,08	1,12	1,26	1,47	1,46	1,85

Table A 12. Physical and chemical parameters measured during intake of test run I. * = salinity was determined from taken water-sample in the lab due to a sensor malfunction on board. Salinity: 34 PSU (average of three measurements in the same sample).

	Intake treated 16:52	Intake treated 17:20	Intake control 18:20	Intake control 18:41
Temperature [°C]	22	21,7	21,3	21,2
pH	8,46	8,40	8,40	8,47
Dissolved oxygen Saturation percentage [%]	104,1	101,7	100,3	99,2
Salinity [PSU]	*	*	*	*

Table A 13. Physical and chemical parameters measured during discharge of test run I. ** = value likely to high due to engine-room temperature.

	Discharge treated 13:30	Discharge treated 13:55	Discharge control 14:53	Discharge control 15:16
Temperature [°C]	14,8	14,1	(16)**	14,4
pH	8,43	8,50	8,47	8,47
Dissolved oxygen Saturation percentage [%]	96,3	99,4	98,9	102,2
Salinity [PSU]	34,63	33,94	38,97	37,99

Test run II

Table A 14. TSS and POC data from test run II.

mg/L	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
TSS - C	8,70	30,60	19,60	3,50	4,90	1,70
TSS - T	10,50	8,30	8,80	4,30	6,30	6,20
POC - C	2,80	7,10	5,00	2,10	3,10	1,50
POC - T	3,30	2,40	3,00	2,10	3,40	2,80

Table A 15. Physical and chemical parameters measured during intake of test run II.

	Intake treated 11:34	Intake treated 12:06	Intake control 12:55	Intake control 13:14
Temperature [°C]	9,3	9,9	9,0	8,9
pH	8,22	8,27	8,30	8,28
Dissolved oxygen Saturation percentage [%]	90,4	93,0	92,3	92,8
Salinity [PSU]	30,96	30,36	30,64	30,64

Table A 16. Physical and chemical parameters measured during discharge of test run II.

	Discharge treated 09:16	Discharge treated 09:30	Discharge control 10:54	Discharge control 11:15
Temperature [°C]	16,3	16,4	15,8	15,8
pH	8,03	8,15	8,06	8,11
Dissolved oxygen Saturation percentage [%]	95,1	97,7	103,6	97,3
Salinity [PSU]	30,04	31,50	32,55	32,48

Test run III

Table A 17. TSS and POC data from test run III.

mg/L	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
TSS - C	9,30	13,20	10,00	16,20	4,10	3,20
TSS - T	14,40	11,10	10,60	7,20	8,10	7,80
POC - C	3,00	5,40	4,60	5,00	2,60	2,40
POC - T	3,70	3,10	3,40	2,80	3,50	3,20

Table A 18. Physical and chemical parameters measured during intake of test run III.

	Intake treated 16:43	Intake treated 16:59	Intake control 18:23	Intake control 18:43
Temperature [°C]	5,7	5,5	5,4	5,1
pH	8,31	8,37	8,36	(7,81)
Dissolved oxygen Saturation percentage [%]	91,3	91,9	91,1	92,4
Salinity [PSU]	32,38	32,85	33,86	34,38

Table A 19. Physical and chemical parameters measured during discharge of test run III.

	Discharge treated 12:18	Discharge treated 12:40	Discharge control 13:20	Discharge control 13:50
Temperature [°C]	7,7	6,6	8,5	9,3
pH	8,30	8,34	8,05	8,35
Dissolved oxygen Saturation percentage [%]	95,4	97,3	91,3	92,8
Salinity [PSU]	31,30	32,92	36,36	35,6

Test run IV

Table A 20. TSS and POC data from test run IV.

mg/L	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
TSS - C	1,95	2,50	2,45	2,55	3,20	6,05
TSS - T	2,20	1,95	0,95	1,15	12,10	10,00
POC - C	1,45	1,60	1,70	1,60	1,85	2,00
POC - T	1,60	1,45	1,00	1,10	3,50	3,40

Table A 21. Physical and chemical parameters measured during intake of test run IV.

	Intake treated 17:14	Intake treated 17:33	Intake control 18:53	Intake control 19:22
Temperature [°C]	20,0	20,0	19,7	19,6
pH	8,44	8,44	7,86	8,21
Dissolved oxygen Saturation percentage [%]	105,8	107,0	106,6	105,6
Salinity [PSU]	37,13	36,55	37,44	35,22

Table A 22. Physical and chemical parameters measured during discharge of test run IV.

	Discharge treated 09:35	Discharge treated 10:02	Discharge control 11:08	Discharge control 11:35
Temperature [°C]	16,6	17,0	16,5	16,3
pH	8,60	8,59	8,34	(7,08)
Dissolved oxygen Saturation percentage [%]	105,6	105,0	104,6	108,1
Salinity [PSU]	35,63	35,83	37,86	37,81

Test run V

Table A 23. TSS and POC data from test run V.

mg/L	Intake replicate 1	Intake replicate 2	Intake replicate 3	Discharge replicate 1	Discharge replicate 2	Discharge replicate 3
TSS - C	2,27	3,13	3,20	1,70	1,85	3,80
TSS - T	2,60	3,20	3,20	2,75	1,75	1,85
POC - C	1,87	2,27	2,27	1,30	1,40	1,90
POC - T	1,67	2,07	2,27	1,80	1,35	1,40

Table A 24. Physical and chemical parameters measured during intake of test run V.

	Intake treated 13:28	Intake treated 13:50	Intake control 14:50	Intake control 15:17
Temperature [°C]	11,2	10,6	10,5	10,1
pH	8,25	8,21	8,20	(7,59)
Dissolved oxygen Saturation percentage [%]	105,9	109,2	105,9	106,9
Salinity [PSU]	36,67	36,08	35,61	36,01

Table A 25. Physical and chemical parameters measured during discharge of test run V.

	Discharge treated 09:28	Discharge treated 09:56	Discharge control 11:29	Discharge control 11:55
Temperature [°C]	11,6	11,1	10,6	10,5
pH	8,21	8,17	8,27	(6,96)
Dissolved oxygen Saturation percentage [%]	107,5	106,5	105,8	107,2
Salinity [PSU]	37,14	39,12	37,97	38,11

END PAGE

Revision	Date	Description	Author	Checked	Approved
01	18/09/13	Reviewed	PH	SRE	RF
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