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Effects of seawater scrubbing on a microplanktonic community during a summer-bloom in the Baltic Sea

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ABSTRACT

The International Maritime Organization (IMO) has gradually applied stricter regulations on the maximum sulphur content permitted in marine fuels and from January 1, 2020, the global fuel sulphur limit was reduced from 3.5% to 0.5%. An attractive option for shipowners is to install exhaust gas cleaning systems, also known as scrubbers, and continue to use high sulphur fuel oil. In the scrubber, the exhausts are led through a fine spray of water, in which sulphur oxides are easily dissolved. The process results in large volumes of acidic discharge water, but while regulations are focused on sulphur oxides removal and acidification, other pollutants e.g. polycyclic aromatic hydrocarbons, metals and nitrogen oxides can be transferred from the exhausts to the washwater and discharged to the marine environment. The aim of the current study was to investigate how different treatments of scrubber discharge water (1, 3 and 10%) affect a natural Baltic Sea summer microplanktonic community. To resolve potential contribution of acidification from the total effect of the scrubber discharge water, “pH controls” were included where the pH of natural sea water was reduced to match the scrubber treatments. Biological effects (e.g. microplankton species composition, biovolume and primary productivity) and chemical parameters (e.g. pH and alkalinity) were monitored and analysed during 14 days of exposure. Significant effects were observed in the 3% scrubber treatment, with more than 20% increase in total biovolume of microplankton compared to the control group, and an even greater effect in the 10% scrubber treatment. Group-specific impacts were recorded where diatoms, flagellates *incertae sedis*, chlorophytes and ciliates increased in biovolume with increasing concentrations of scrubber water while no effect was recorded for cyanobacteria. In contrast, these effects were not observed in the “pH controls”, a suggestion that other parameters/stressors in the scrubber water were responsible for the observed effects.

1. Introduction

Shipping is emitting a vast number of harmful substances; primarily sulphur oxides (SO_x) and nitrogen oxides (NO_x) (Karl et al., 2019; Raudsepp et al., 2019) but also fine particulate matter (PM_{2.5}) (Jalkanen et al., 2014), polycyclic aromatic hydrocarbons (PAHs) (Teuchies et al., 2020) and metals (Turner et al., 2017). These emissions can result in severe health effects (SO_x, NO_x and PM_{2.5}) (Corbett et al., 2007; Sofiev et al., 2018), acidification (SO_x and NO_x) (Hassellöv et al., 2013), eutrophication (NO_x) (Raudsepp et al., 2019; Zhang et al., 2021), and

ecotoxicological responses in marine organisms (PAHs and metals) (Koski et al., 2017; Teuchies et al., 2020). To reduce the impacts of SO_x emissions on human health and acidification on primarily freshwater and terrestrial ecosystems, the International Maritime Organization (IMO) has gradually applied stricter regulations on the maximum permitted sulphur content in marine fuels. From January 1, 2020, the global fuel sulphur limit was reduced from 3.5% to 0.5% and in designated Sulphur Emission Control Areas (SECA), e.g. the Baltic Sea, the cap is even stricter, 0.1% as from 2015.

To comply with the stricter regulations, the ship owner can either

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switch from high sulphur fuel oil (HSFO) to distillates e.g. marine gas oil (MGO), retrofit vessel to use alternative fuels, such as liquified natural gas (LNG) and methanol, or install an exhaust gas cleaning system (EGCS), also known as a scrubber, and continue the use of HSFO. The scrubber removes SO_x , and to some, less investigated, extent also NO_x , from the exhausts. Since the HSFO is a residual product from the refineries and the cheapest of all marine fuels, installing a scrubber may pose an attractive solution for shipping companies. According to the latest global statistics from DNV GL (2020), 4549 vessels are in operation with a scrubber or on order to install a scrubber. This can be compared to the number of installations in 2018, which was at 731 (DNV GL, 2020). There are three types of scrubbers available on the market; open loop, closed loop and finally a hybrid type where the scrubber can be operated in either open or closed mode. The market is dominated by open loop installations (81% of market share), followed by hybrid systems (17%) and a small share of closed loop systems (1.5%) (DNV GL, 2020).

During the scrubbing process the exhausts are led through a fine spray of water, in which sulphur oxides (SO_x) are easily dissolved, resulting in large volumes of highly acidic water ($\sim\text{pH}3$). When operating in open loop mode, the acidic effluent is continuously discharged into the sea at a typical rate of $45 \text{ m}^3/\text{MWh}$ (IMO, 2008), causing a decrease in pH of the ambient seawater, which is of special concern in semi-enclosed areas with low water exchange. The extent of pH change is governed by the ambient waters' alkalinity but also temperature and salinity have shown to be important parameters (Karle and Turner, 2007).

In addition to acidification, the scrubber discharge water often contains elevated concentrations of metals such as Cu, Zn, Fe, Ni and Pb and polycyclic aromatic hydrocarbons (PAHs) (Endres et al., 2018; Hassellöv et al., 2020; Teuchies et al., 2020) that can pose a risk to the marine environment. Sources of PAHs includes combustion of fuel and lubricants, while emissions of metals besides combustion of fuels include leakage from the scrubber unit and piping resulting from corrosion due to the acidic scrubber washwater (Lunde Hermansson et al., 2021). Scrubber discharge water can also be a potential source of nutrients to the environment, which is why MARPOL guidelines on effluent discharge criteria state that a maximum of 12% of NO_x in the exhausts are allowed to be taken up by the washwater (IMO, 2008) to minimise discharge of nitrate into the sea.

The Baltic Sea is one of the largest brackish water bodies in the world, with surface water salinities ranging from 1 to 3 in the Bothnian Bay, around 7 in the Baltic Proper, to around 25 in the Kattegat. This gradient feature is also reflected in surface total alkalinity (A_T), since A_T is closely related to salinity. Kattegat has an A_T of $2055 \mu\text{mol kg}^{-1}$, which decreases to 1551 in the Baltic Proper and is $774 \mu\text{mol kg}^{-1}$ towards the Bothnian Bay (Hjalmarsson et al., 2008). Hence, compared to fully marine water bodies, the brackish Baltic Sea is more sensitive to acidification, as seawater's ability to withstand pH change is governed by the A_T of the water. A recent modeling study of the potential effect of large-scale use of scrubbers in the Baltic Sea, concluded that a reduction in A_T over time can be expected (Turner et al., 2018).

The Baltic Sea is also a heavily polluted sea where emissions of nutrients and contaminants from primarily agriculture and industries result in almost the entire Baltic Sea not fulfilling Good Environmental Status (GES), with respect to eutrophication and hazardous compounds, according the Marine Strategy Framework Directive (MSFD Directive, 2008/56/EC) (HELCOM, 2018b). The Baltic Sea is also one of the most heavily trafficked seas in the world, and both the number and sizes of ships are forecasted to increase (HELCOM, 2018a). The Baltic Sea is considered particularly sensitive to pollution due to its semi-enclosed character with low salinity, long retention time for contaminants, low biodiversity and limited food-web with few key-species (Kautsky and Svensson, 2003; Magnusson and Norén, 2012). In the Baltic Sea, there is a large seasonal fluctuation in phytoplankton populations, which usually consist of an early spring bloom dominated by diatoms, followed by

dinoflagellates in late spring, nitrogen-fixing cyanobacterial bloom in the summer and diatoms in early autumn (Bianchi et al., 2002). Phytoplankton communities are particularly suitable indicators to study how different human activities and emissions affect the aquatic environment as they constitute the base of the aquatic food web, and thus, any adverse effect on the phytoplankton community may affect organisms across multiple trophic levels. Previous mesocosm studies in the Baltic Sea on impacts of CO_2 -driven ocean acidification on plankton communities have shown large variation in responses. For example, Olofsson et al. (2019) found no effects of elevated $p\text{CO}_2$ on phytoplankton biovolumes while other studies have shown effects on plankton respiration (Spilling et al., 2016) as well as growth rate, biomass and composition (Horn et al., 2020).

As scrubber discharge water, besides being acidic, contains a complex mixture of both nutrients and contaminants, it is difficult to predict environmental responses from large-scale discharges to the Baltic Sea, unless biological responses are studied in whole-effluent tests. At present, only a few studies have been conducted on biological impacts due to emissions of scrubber discharge water (Koski et al., 2017; Ytreberg et al., 2019). Koski et al. (2017) showed adverse effects on mortality and feeding behaviour on the copepod *Acartia tonsa* when exposed to scrubber discharge water containing metal concentrations which were orders of magnitude lower than expected to cause adverse effect from single compound studies. The result suggests scrubber water to have synergistic effects and points to the importance of more whole effluent studies to be carried out. The aim of the current study was to investigate short-term effects (weeks) of scrubber discharge water on a natural microplanktonic community from the Baltic Sea which was collected during the summer bloom. A second aim was to resolve the potential contribution of acidification from the total effect of the scrubber discharge water. Thus, "pH controls" were included where the pH of natural seawater was reduced to match the scrubber treatments. Biological effects (e.g. microplankton species composition, biovolume, primary productivity and photosynthetic activity) and chemical parameters (e.g. pH and alkalinity) were monitored and analysed after 14 days of exposure.

2. Material and methods

The preparation for the biological experiments was started on May 15th, 2014, by collecting seawater at Askö laboratory outside Stockholm, Sweden. The water was collected in four 20 L plastic bottles. The bottles were rinsed three times with the ambient water prior to the sampling and then brought by car to Gothenburg to produce scrubber discharge water in the engine lab at the Department of Mechanics and Maritime Sciences, Chalmers University of Technology.

2.1. Production of scrubber discharge water

Chalmers' engine lab is a four-cylinder 100 kW engine from Volvo Penta, equipped with a scrubber unit. Since most scrubbers on ships are made of stainless steel, the scrubber unit at Chalmers is also made of stainless steel with main constituents after iron, are chromium (16–18%), nickel (10–12%) and molybdenum (2–3%), with small (<1%) quantities of silicon, phosphorus and sulphur also present (SAE 316L grade). All sealing used are made from PTFE. The scrubber unit and the scrubbing procedure have recently been described in Ytreberg et al. (2019). The exhaust flow rate was kept at 15 L per minute (LPM) throughout the production of scrubber discharge water while the water flow rate was kept at 3.3 LPM yielding a water-to-air ratio of 0.22. The discharge water was immediately pumped out of the scrubber unit after the scrubbing process. A marine gas oil with 1.0% sulphur (analysed at Saybolt, Sweden according to EN-ISO 8754) was used in the experiment. The pH of the discharge water was 2.8, measured with a pH electrode (Aquatrode Plus Pt1000, Metrohm). Samples for nutrients and metals were collected and immediately sent for analysis according to the

descriptions in section 2.3.9. (pH) and 2.3.11. (metals).

2.2. Set-up of biological experiment

The biological experiment was conducted for 14 days between July 11 and 25, 2014, at Askö Laboratory, Sweden (58°49'N, 17°38'E) in the Baltic Sea. A natural community of Baltic Sea pelagic microplankton (i.e. phytoplankton, cyanobacteria and heterotrophic microplankton), dominated by the cyanobacteria *Aphanizomenon* sp., *Nodularia spumigena*, and *Dolichospermum* sp. was collected from the station B1 (58°48'28 N, 17°37'60 E). Surface seawater from 0 to 5 m water depth was collected and gently filtered through a 200 µm mesh to avoid larger grazers. In addition, microplankton were collected and concentrated through a plankton net (mesh size 25 µm).

21 transparent polyethylene bags (50.25 l) were placed two-by-three in four water filled basins filled with continuous flow-through seawater, with an in situ temperature of 17.3 °C (Std 0.94). The bags were filled by gently pouring a mixture of surface seawater (45 l), 250 ml microplankton-enriched water and either filtered seawater, scrubber discharge water or pH stock water (Table 1), creating seven different treatments in three replicates; control (Ctrl), 1% scrubber discharge water (1% scr), 3% scrubber discharge water (3% scr), 10% scrubber discharge water (10% scr), 1% pH control (1% pH), 3% pH control (3% pH) and 10% pH control (10% pH). The pH stock water was prepared by adding 1 M H₂SO₄ to 0.2 µm filtered seawater to reach pH 2.8, i.e. similar pH as the scrubber discharge water. Treatments called “pH control” were set up to examine possible treatment effects of the scrubber discharge water apart from acidification. To maintain homogeneity of the water mass inside, the bags were carefully stirred three times a day.

The basins were covered with green plastic mesh to reduce the irradiance, resulting in photosynthetically active radiation (PAR, 400–700 nm) of 155–255 µmol m⁻² s⁻¹ at midday, corresponding to water depths of 1–2.5 m at the sampling site, as measured with a LI-1000 datalogger equipped with a PAR sensor (Li-COR UWQ5201). The same sensor was used to monitor irradiances in the experimental container at 40 cm water depth, throughout the experiment.

2.3. Sampling and analysis of biological, physical and chemical parameters

Sampling for the different biological and physical/chemical parameters ($n = 1$) was performed in all bags immediately after experimental start (within 1h on day 0) and on day 14, while temperature and PAR were monitored continuously (Table 2). Metals were however only sampled from the ambient sea water ($n = 2$) and from the scrubber washwater ($n = 2$). The sampling procedure started with carefully stirring each bag and then siphoning 3 l water from 25 cm depth in the bag to pre-cleaned plastic buckets. Water for pH, alkalinity and dissolved inorganic carbon (DIC) analyses were sampled by gently overfilling twice the volume of separate borosilicate bottles, leaving no headspace. Subsampling for all other parameters were done from the buckets in the adjacent laboratory.

Table 1

Experimental set-up, initial biovolume concentration (standard deviation in brackets) and volumes of different waters added to the treatments and control bags.

Treatment	Net plankton (L)	0.2 µm -filtered seawater (L)	Scrubber discharge water (L)	pH stock (L)	Microplankton community (<200 µm) (L)	Total volume (L)	Initial biovolume concentration (mm ³ /L)
Control	0.25	5	0	0	45	50.25	3.3 (0.2)
1% pH	0.25	4.5	0	0.5	45	50.25	3.1 (0.5)
1% scr	0.25	4.5	0.5	0	45	50.25	3.1 (0.5)
3% pH	0.25	3.5	0	1.5	45	50.25	2.8 (0.4)
3% scr	0.25	3.5	1.5	0	45	50.25	3.0 (0.6)
10% pH	0.25	0	0	5	45	50.25	3.1 (0.7)
10% scr	0.25	0	5	0	45	50.25	3.1 (0.4)

Table 2

Investigated biological and physical/chemical parameters and sampling day.

	Parameters	Sampling day
Biological	Species composition	0, 14
	Biovolume	0, 14
	Photosynthetic activity	0, 14
	Primary productivity	0, 14
	Bacterial biomass	0, 14
	Bacterial productivity	0, 14
	Particulate organic carbon (POC)	0, 14
	Particulate organic nitrogen (PON)	0, 14
	Particulate organic phosphorous (POP)	0, 14
	pH	0, 14
Physical/ Chemical	Alkalinity	0, 14
	Metals	0, 14
	Dissolved inorganic nitrogen (DIN; NO ₂ ⁻ + NO ₃ ⁻ and NH ₄ ⁺)	0, 14
	Phosphate (DIP)	0, 14
	Silicate (Si(OH) ₄)	0, 14
	Dissolved inorganic carbon (DIC)	0, 14
	Temperature	Continuously
	Photosynthetically active radiation (PAR)	Continuously

2.3.1. Microplankton species composition and biovolume

Samples of 50 ml from each bag was preserved with acidic Lugol's iodine solution, stored in the dark and analysed within 12 months using the Utermöhl method according to HELCOM (2008). All cells, filaments and colonies were measured and grouped, either to species level or order, and biovolume (mm³ l⁻¹) was calculated according to Olenina et al. (2006). The natural community was comprised of cyanobacteria, ciliates, chlorophytes, flagellates *incertae sedis*, diatoms and dinoflagellates. Since the experiment was conducted during the summer, the majority of the biovolume within the group cyanobacteria consisted of the three bloom-forming species *Aphanizomenon* sp., *Dolichospermum* sp. and *Nodularia spumigena*, but there were also other filamentous species such as *Limnothrix* sp. and *Pseudanabaena* sp., as well as colony-forming species. The group flagellates *incertae sedis* is composed of cells with flagella that do not fit any of the previous mentioned groups, and may very well consist of several classes.

2.3.2. Photosynthetic activity

Photosynthetic activity was estimated by chlorophyll fluorescence measurements in photosystem II (PSII). A pulse amplitude modulation (PAM) fluorometer calibrated for cyanobacterial application was used (Water-PAM with Water ED unit, Walz Mess-und Regeltechnik, Germany). Immediately after sampling effective quantum yield ($\Delta F/F_m$) was measured and after 1.5-h dark adaptation in treatment temperature, maximum quantum yield (F_v/F_m) measured. The saturation pulse of measuring light was set to 4000 µmol photons m⁻² s⁻¹ for 0.6 s.

2.3.3. Primary productivity

Primary productivity was measured using the radiocarbon technique (Ærtebjerg-Nielsen and Bresta, 1984) using incorporation of H¹⁴CO₃⁻ (PerkinElmer, Waltham, MA, USA) as described in Torstensson et al.

(2013). Incubations were performed in a constant temperature room at 17 °C and 200 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ at day 0 and at 100 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ at day 14.

2.3.4. Particulate organic carbon, nitrogen and phosphorous

For analyses of POC, PON and POP, 250–350 ml from each bag were filtered onto pre-combusted (400 °C for 4 h) GF/F filters (Whatman). The filters for POP analyses were washed using 0.1 M HCl and rinsed with Milli-Q prior to combustion. POP filters were dried in room temperature and analysed at Tvärminne Zoological Station, Finland, according to Solórzano and Sharp (1980). Filters for POC and PON analysis were frozen at –20 °C, freeze-dried for 36 h (Heto Power Dry PL3000, Thermo Fisher Scientific), ground into a fine powder (MM301, Retsch) and analysed in an elemental analyser (EA 1108 CHNS–O, Fisons Instruments) applying 2,5-bis(5-tert-butyl-benzoaxazol-2-yl)thiophen as an internal standard.

2.3.5. Bacterial biomass and productivity

Bacterial biomass was determined by using a FACSCalibur flow cytometer (BD Biosciences). Samples (1.5 ml) were fixed with glutaraldehyde (Sigma-Aldrich, 1% final concentration), and stored at –80 °C until analysis. Cells were covered from light and stained with SYBR Green I Nucleic Acid Gel Stain (Invitrogen, Thermo Fisher Scientific) for 10 min. As internal standard in every sample, 1.0 μm green fluorescent polymer microspheres (CountBright absolute counting beads, Invitrogen, Thermo Fisher Scientific) were used.

Bacterial productivity was measured using ^3H thymidine incorporation (20 nM final concentration, NET355001MC, PerkinElmer) following Fuhram and Azam (1982). Samples were incubated at 17 °C in darkness for 1 h, and later analysed using liquid scintillation counting as described in (Torstensson et al., 2015). Thymidine incorporation was converted to bacterial carbon production using 1.4×10^{18} cells mol^{-1} thymidine incorporated (average calculated from published Baltic Sea data, $\text{SE} = 0.1 \times 10^{18}$ cells mol^{-1} thymidine, $n = 73$ (HELCOM, 2017), and 20 fg C cell $^{-1}$ (Lee and Fuhrman, 1987)). Saturation curves were made beforehand. Cell specific production was calculated by relating bacterial production to bacterial biomass.

2.3.6. Dissolved organic carbon

A volume of 7.5 ml was filtered immediately after sampling through a 0.2 μm syringe filter (Supor, PALL) to a 15 ml polypropylene Falcon® tube (VWR). 100 μl 1.2 M HCl was added and all samples were stored at 4 °C until analysis at ALS Scandinavia AB, Sweden, according to the CSN EN 1484 method. Syringes, filters and vials were washed with 0.2 μm filtered sample water prior to acidification.

2.3.7. Inorganic nutrients

Samples (12 ml) for determination of dissolved inorganic nitrogen (DIN; $\text{NO}_2^- + \text{NO}_3^-$ and NH_4^+), phosphate (DIP), and silicate ($\Sigma\text{Si}(\text{OH})_4 + \text{Si}(\text{OH})_3\text{O}^-$), onwards referred to as $\text{Si}(\text{OH})_4$ were filtered through 0.2 μm filters, stored frozen in –20 °C, and thereafter analysed using photometric determination (Hansen and Koroleff, 1999) performed on a QuAatro auto-analyser at Kristineberg Marine Research Station, Sweden.

2.3.8. Dissolved inorganic carbon

Samples for DIC were collected in 250 ml borosilicate bottles and preserved using 60 μl of saturated HgCl_2 . Samples were wrapped in parafilm and stored at 4 °C until analysis. DIC was determined using a coulometric titration method based on Johnson et al. (1987) with a modified Single Operator Multiparameter Metabolic Analyzer (SOMMA) system (coulometer type UIC 5012). The precision from replicate analyses was $\pm 2 \mu\text{mol kg}^{-1}$, with the accuracy set by routine analysis of Certified Reference Materials (CRM, Batch #137) provided by A.G. Dickson, Scripps Institution of Oceanography, USA.

2.3.9. pH

Samples for pH were gently siphoned in 100 ml borosilicate bottles. The sampling and analysis followed established protocols (Dickson et al., 2007). pH was measured in the total scale (pH_T) spectrophotometrically. A 2 mM solution of the sulphonaphthalein dye, m-cresol purple (Aldrich, lot MKBC2604V), dissolved in MilliQ water, was used as indicator. The measurement was performed using a spectrophotometer (HELIOS ZETA UV-VIS Thermo scientific). Prior to analysis, the samples were slowly heated to ~ 25 °C. Samples were measured in a 1 cm quartz cell, where the temperature was measured using a thermistor (Ama-Digit ad 15th, Amarell GmbH & Co. KG) with a precision of 0.1 °C. The pH perturbation due to the added indicator was corrected for by making 5 indicator additions of 20 μl to each sample (ca. 3 ml). The absorbance at the isosbestic point (488 nm) was used as a measure of the indicator concentration, and the pH calculated at each point was then extrapolated linearly to zero indicator concentration. pH_T was calculated from the absorbances at 434 nm and 578 nm according to Clayton and Byrne (1993) using the brackish water pK values for m-cresol purple determined by Mosley et al. (2004).

2.3.10. Alkalinity

Total alkalinity (A_T) was determined by automated titration of weighed samples against 0.5 M hydrochloric acid using a Metrohm 888 Titrando titration system (Metrohm). Since the evaluation routine in the titrator's firmware was found to be unsuitable for brackish water samples, the raw titration data were analysed using the VINDTA algorithm as described in Ulfsbo et al. (2015). Certified Reference Materials (CRM, Batch #137) provided by A.G. Dickson, Scripps Institution of Oceanography, USA were analysed together with each batch of samples, and the sample alkalinities corrected for the small differences between the measured and CRM values.

2.3.11. Metals

Samples for metal and element analysis (Ca, Fe, K, Mg, Na, Si, Al, Ba, Cd, Co, Cr, Cu, Hg, Mn, Mo, Ni, P, Pb, Sr, V, Zn and S) were collected from the scrubber discharge water and from the Baltic Sea water, filtered through acid-cleaned 0.45 μm filters (Supor, PALL) and collected in acid-cleaned 250 ml Polypropylene bottles. Dissolved metal analysis was performed by ALS Scandinavia AB, Sweden, using Inductively Coupled Plasma Sector Field Mass Spectrometry (ICP-SFMS) according to EPA method 200.8 rev.5.4 (1994) and SS EN ISO 17294–1 (2006). The metal concentrations in the treatments were estimated by considering Ctrl, 1% pH, 3% pH and 10% pH as 100% Baltic Sea water, while 1% scr was calculated as a mixture of 99% Baltic Sea water and 1% scrubber water. Analogously, metal concentrations in 3% scr and 10% scr were calculated.

2.4. Statistical analyses

Statistical analyses were performed using linear mixed effects models (Harrison et al., 2018) with each of the biological, physical and chemical parameters (Section 2.3) at day 0 and 14 as response variables. The dose-response relationship with scrubber and pH treatments were modelled with linear, quadratic or categorical dose effects, choosing the best model in terms of Akaike Information Criterion (Akaike, 1974). Basin and bag effects were modelled as random effects, accounting for basin effects and correlations between repeated measurements (day 0 and 14) on the same bag (Harrison et al., 2018). Model fit and plausibility of normality assumptions were assessed by visual inspection of residual plots and normal probability plots of random effects and standardized residuals.

F-tests were used for evaluation of treatment effects at day 0 and 14. The effects of scrubber discharge water was first evaluated versus ordinary controls, with multiplicity adjustment using Holm's procedure (Holm, 1979). The effect of scrubber water was then further evaluated versus pH controls, again with multiplicity adjustment using Holm's

procedure. No p-value adjustments were made for day 0 comparisons and for comparisons between the two control groups.

Estimated dose effects and time trends are presented with 95% confidence intervals, without multiplicity adjustment. Microplanktonic biovolume (total and by microplanktonic group) was log-transformed prior to data analysis, assuming an exponential growth model. Statistical analyses were performed with R (R Core Team, 2016) version 3.5.14, and mixed effects models were fitted using the nlme package (Pinheiro et al., 2016), version 3.1–137.

3. Results and discussion

3.1. Microplankton species composition

At the start of the experiment (day 0), on average 54% of the biovolume consisted of filamentous cyanobacteria comprising the following species/genera *Aphanizomenon* sp. (41%), *Nodularia spumigena* (11%) and *Dolichospermum* sp. (2%) (Fig. 1). The ciliates accounted for on average 14%, dinoflagellates 7%, other cyanobacteria 5%, chlorophytes 2% and flagellates *incertae sedis* 17% of the total biovolume. The diatoms were less than 1% of the phytoplankton community. After 14 days, only 14% of the total biovolume consisted of cyanobacteria in the controls. Instead, diatoms were the most abundant group of microplankton in the controls, comprising on average 32% of the total biovolume. Bloom-forming cyanobacteria in the Baltic Sea have the ability to dominate, after the spring-bloom depletes the nutrients from the water column. However, as seen in this experiment, they cannot outcompete diatoms when the nitrate concentration is high. In addition, the total biovolume was on average higher in the scrubber treatments, as compared to the control. No difference was however observed between the pH treatments and control. The impact of scrubber discharge water on microplankton group level is discussed in detail in the next chapter.

3.2. Impacts of scrubber discharge water on biota

There were no significant differences in total biovolume of microalgal species between treatment groups at the start of the experiment (Fig. 2). However, at day 14 scrubber discharge water had a statistically significant effect on the biovolume of microplankton species as compared to the control, where the biovolume was on average 86% higher in the 10% scr treatment as compared to the control (Table 3 and Fig. S1). For the 3% scr treatment, the effect was lower with an average increase (95% CI) in biovolume of 21% (2%–42%), as compared to the control (Table 3 and Fig. S1). However, no significant treatment effect of pH on biovolume was observed (Table 3, Fig. 2, Fig. S1), indicating that

other factors than pH alone were responsible for the increased biovolume observed in the scrubber discharge water treatments. Nitrogen, in the forms of NO_3 , NO_2^- and NH_4^+ , is together with phosphorus two of the most important nutrients for microplankton to grow and chemical analyses of the scrubber treatments showed significantly higher concentrations of dissolved inorganic nitrogen (NO_3 , NO_2^- and NH_4^+) at the start of the experiment (day 0) as well as in the end (day 14) (Table S1) (Table 4). The increased nitrogen concentration can be explained by the uptake of NO_x and the formation of nitrogen enriched waters during the scrubber process. The uptake of NO_x has shown to vary substantially in onboard ship measurements. In a study submitted to MEPC in 2018, the concentration of nitrate was measured in inlet water to the scrubber and in the discharge water on nine ships equipped with an open-loop scrubber (MEPC 73/INF.5, 2018). The results showed no uptake of NO_x for five of the vessels. However, for the remaining four vessels an uptake corresponding to a net increase of 16.1, 19.4, 32.3 and 64.4 $\mu\text{mol l}^{-1}$ nitrate in the discharge water was observed. This can be compared with the 10% scr treatment which showed a nitrate concentration of on average 15.33 $\mu\text{mol l}^{-1}$. The scrubber discharge water used in the current study was not analysed for nitrate but the nominal concentration of nitrate in the scrubber discharge water was 150 $\mu\text{mol l}^{-1}$, i.e. roughly twofold higher as compared to the highest observed net increase of nitrate in the MEPC study. Since nitrogen is an essential element for all organisms and a potential limiting nutrient for primary production, the uptake capacity of NO_x in the scrubber could be the key parameter determining environmental impacts since this eutrophication response may mask any other short-term adverse effects due to e.g. acidification or increased concentrations of metals and PAHs. The scrubber water used in the current study was characterised for metals and other elements and in particular chromium, copper and zinc showed elevated, up to 400 times higher, concentrations as compared to the Baltic Sea water used in the scrubber process (Table S3 supporting material).

The effect of scrubber discharge water and pH treatments on total biovolume at day 14, by microplanktonic groups is shown in Fig. 3. No significant treatment effect of reduced pH on biovolume of diatoms, flagellates *incertae sedis*, chlorophytes or ciliates was observed. In contrast, diatoms, flagellates *incertae sedis*, chlorophytes and ciliates all showed a significant increase in biovolume with increasing scrubber discharge water, as compared to the control after 14 days exposure (Fig. 3). For flagellates *incertae sedis* and chlorophytes, the increase in biovolume in the 10% scr treatment was on average 69% and 127%, respectively, while silicates showed a 247% increase (Table 3 and Fig. S2 supporting material). The effect on biovolume was also significant for diatoms in the 10% scr treatment, which was on average 159% higher than the control (Table 3 and Fig. S2 supporting material). The diatoms were dominated by *Thalassiosira cf. pseudonana*, a species known to be sensitive to copper exposure. For example, Erickson (1972) showed inhibition in growth rate when *T. pseudonana* was exposed to copper in the range of 5–30 $\mu\text{g l}^{-1}$. In another more recent study, *T. pseudonana* was however shown to be less sensitive to copper exposure with a reported 96h-EC50 value of 970 $\mu\text{g l}^{-1}$ (Bao et al., 2008). These results were however based on nominal copper concentrations and neither the medium nor the treatments were characterized for dissolved organic carbon concentrations, making the comparison difficult. In the present study, filtered unenriched seawater holding a copper concentration of 0.68–1.14 $\mu\text{g l}^{-1}$ was used. The scrubber water used in the current study was also characterised for metals and other elements and in particular chromium (201 $\mu\text{g l}^{-1}$), nickel (141 $\mu\text{g l}^{-1}$), copper (87 $\mu\text{g l}^{-1}$) and zinc (210 $\mu\text{g l}^{-1}$) showed elevated, up to 400 times higher, concentrations as compared to pristine Baltic Sea water (Table S3 supporting material). One reason for the elevated concentrations is the leaching of metals from the scrubber unit which is made of stainless steel and holds high concentrations of chromium and nickel. Elevated concentrations of chromium, nickel, copper and zinc has also been reported by Lunde Hermansson et al. (2021), where measurements of scrubber discharge water from a total of 41 vessels equipped with scrubbers, were

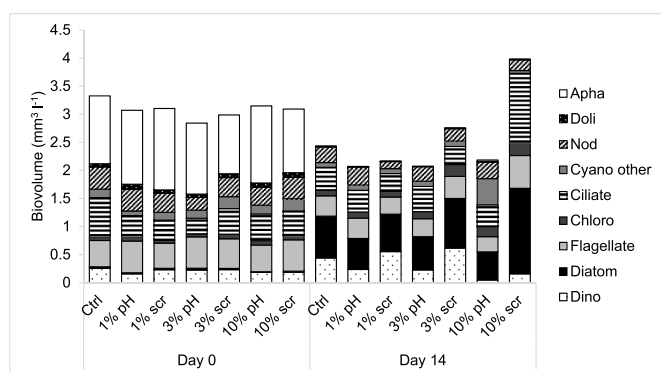


Fig. 1. Average biovolume ($\text{mm}^3 \text{l}^{-1}$) for all microplankton groups and species; *Aphanizomenon* sp., *Dolichospermum* sp., *Nodularia spumigena*, other cyanobacteria, ciliates, chlorophytes, flagellates *incertae sedis*, diatoms and dinoflagellates, at day 0 and day 14 in the untreated control (Ctrl), 1%, 3% and 10% added scrubber discharge water (1% scr, 3% scr and 10% scr) and the pH treatments (1% pH, 3% pH and 10% pH).

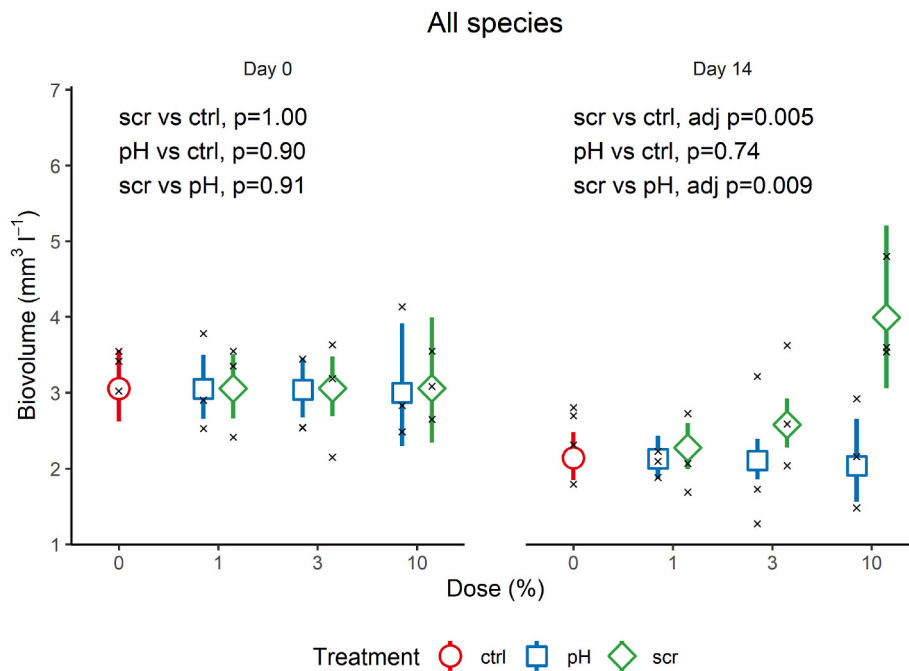


Fig. 2. Total biovolume of all microalgal species in the control (ctrl), scrubber treatments (scr) and pH treatments (pH) at day 0 and day 14. Crosses (x) show individual measurements. Circles, squares and diamonds with error bars represent geometric means with 95% confidence intervals.

Table 3

Effect of scrubber and pH treatments on biovolume at day 0 and 14. Treatment effects are shown in comparison to either the control (ctrl) or the pH control treatments (pH).

Group	Day	Ref	Ratio of geometric means (95% CI) vs control or pH reference					
			1% scr	3% scr	10% scr	1% pH	3% pH	10% pH
All species	0	ctrl	1.00 (0.97–1.03)	1.00 (0.91–1.10)	1.00 (0.73–1.38)	1.00 (0.97–1.03)	0.99 (0.90–1.09)	0.98 (0.71–1.35)
		pH	1.00 (0.97–1.04)	1.01 (0.90–1.12)	1.02 (0.71–1.47)			
		scr	1.06 (0.90–1.26)	1.21 (1.02–1.42)	1.86 (1.39–2.50)	1.00 (0.84–1.18)	0.99 (0.84–1.16)	0.95 (0.71–1.27)
Cyanobacteria	0	ctrl	1.00 (0.95–1.05)	1.00 (0.86–1.16)	1.00 (0.60–1.65)	1.00 (0.95–1.06)	1.01 (0.87–1.18)	1.04 (0.63–1.72)
		pH	1.00 (0.94–1.06)	0.99 (0.83–1.17)	0.96 (0.54–1.71)			
		scr	0.99 (0.74–1.31)	0.96 (0.73–1.27)	0.87 (0.55–1.39)	1.11 (0.83–1.47)	1.36 (1.03–1.79)	2.78 (1.76–4.41)
Diatoms	0	ctrl	0.89 (0.84–0.94)	0.71 (0.59–0.84)	0.31 (0.18–0.56)	0.89 (0.83–0.95)	0.70 (0.57–0.86)	0.31 (0.16–0.61)
		pH	0.95 (0.88–1.01)	0.85 (0.69–1.04)	0.58 (0.29–1.14)			
		scr	1.07 (0.98–1.15)	1.21 (0.95–1.53)	1.88 (0.86–4.13)	0.97 (0.66–1.42)	0.90 (0.62–1.31)	0.70 (0.38–1.32)
Dinoflagellates	0	ctrl	1.10 (0.75–1.61)	1.33 (0.92–1.93)	2.59 (1.39–4.83)	0.97 (0.66–1.44)	0.94 (0.37–2.40)	0.91 (0.37–2.22)
		pH	1.14 (1.05–1.23)	1.48 (1.17–1.87)	3.67 (1.67–8.06)			
		scr	1.08 (0.72–1.60)	1.17 (0.46–3.02)	0.87 (0.35–2.13)	0.97 (0.66–1.44)	0.94 (0.37–2.40)	0.91 (0.37–2.22)
Flagellates i.s.	0	ctrl	1.10 (0.74–1.64)	1.25 (0.50–3.17)	0.96 (0.35–2.63)	1.00 (0.96–1.03)	0.99 (0.89–1.10)	0.95 (0.67–1.36)
		pH	1.28 (0.73–2.22)	1.66 (0.76–3.62)	0.39 (0.17–0.88)	0.58 (0.33–1.00)	0.24 (0.11–0.51)	0.11 (0.05–0.25)
		scr	2.22 (1.49–3.30)	6.99 (2.77–17.67)	3.56 (1.30–9.76)	1.00 (0.96–1.03)	0.99 (0.89–1.10)	0.95 (0.67–1.36)
Chlorophytes	0	ctrl	1.01 (0.97–1.05)	1.03 (0.92–1.14)	1.09 (0.77–1.56)	1.00 (0.96–1.03)	0.99 (0.89–1.10)	0.95 (0.67–1.36)
		pH	1.01 (0.97–1.06)	1.04 (0.92–1.18)	1.15 (0.76–1.72)			
		scr	1.05 (0.88–1.26)	1.17 (0.98–1.40)	1.69 (1.22–2.35)	0.97 (0.82–1.16)	0.92 (0.78–1.10)	0.77 (0.56–1.06)
Ciliates	0	ctrl	1.08 (1.04–1.13)	1.27 (1.12–1.43)	2.20 (1.46–3.31)	1.00 (0.95–1.07)	1.02 (0.86–1.21)	1.06 (0.60–1.88)
		pH	0.98 (0.93–1.04)	0.95 (0.80–1.13)	0.85 (0.48–1.51)	1.01 (0.95–1.07)	1.02 (0.86–1.21)	1.06 (0.60–1.88)
		scr	0.98 (0.92–1.05)	0.94 (0.77–1.14)	0.80 (0.41–1.55)	1.04 (0.82–1.32)	1.13 (0.89–1.44)	1.52 (0.90–2.55)
Ciliates	14	ctrl	1.09 (0.85–1.38)	1.28 (0.99–1.65)	2.27 (1.33–3.89)	1.04 (0.82–1.32)	1.13 (0.89–1.44)	1.52 (0.90–2.55)
		pH	1.04 (0.97–1.11)	1.13 (0.93–1.38)	1.49 (0.77–2.90)			
		scr	1.04 (0.97–1.11)	1.13 (0.93–1.38)	1.49 (0.77–2.90)	1.19 (0.73–1.94)	1.48 (0.70–3.13)	0.96 (0.42–2.17)
Ciliates	14	ctrl	0.98 (0.90–1.07)	0.95 (0.72–1.24)	0.83 (0.34–2.06)	0.72 (0.50–1.03)	0.45 (0.19–1.09)	0.79 (0.34–1.86)
		pH	1.37 (0.97–1.92)	2.08 (0.94–4.62)	1.06 (0.38–2.90)			
		scr	1.13 (0.68–1.90)	1.45 (0.89–2.37)	3.47 (1.56–7.69)	1.19 (0.73–1.94)	1.48 (0.70–3.13)	0.96 (0.42–2.17)
Ciliates	14	ctrl	0.96 (0.68–1.33)	0.98 (0.45–2.15)	3.62 (1.32–9.95)			
		pH						
		scr						

CI, confidence interval; ctrl, control; i.s., *incertae sedis*; ref, reference; scr, scrubber water.

compiled. The copper concentration in the 10% scr treatment was calculated to be 11 µg l⁻¹ which is in the range of where Erickson (1972) observed adverse effects in growth rate. This potential adverse effect of elevated concentration of copper (and other metals) in the scrubber discharge water may have been counteracted by the elevated nitrate

concentration in the scrubber discharge water.

In contrast, cyanobacteria did not increase in biovolume in any of the scrubber treatments, presumably due to the addition of nitrogen enriched waters in the scrubber treatments and that cyanobacteria fix nitrogen only under nitrogen deficient conditions (Fig. 3 and Table 3).

Table 4
Concentrations of dissolved inorganic nitrogen (NH_4^+ , μM , and $\text{NO}_2^- + \text{NO}_3^-$, μM), silicate (Si(OH)_4 , μM), total alkalinity (A_T , $\mu\text{mol kg}^{-1}$), pH total scale at 17 °C (pH_T), dissolved inorganic carbon (DIC, $\mu\text{mol kg}^{-1}$) and partial pressure of carbon dioxide at 17 °C ($p\text{CO}_2$, atm) at Day 0 and Day 14 in the untreated control (Ctrl), 1%, 3% and 10% added scrubber discharge water (1% scr, 3% scr and 10% scr) and their respective pH treatments (1% pH, 3% pH and 10% pH). Mean values, $n = 3$ (SD).

	Day 0					Day 14								
	Ctrl	1% pH	1% scr	3% pH	3% scr	10% pH	10% scr	Ctrl	1% pH	1% scr	3% pH	3% scr	10% pH	10% scr
NH_4^+	0.39 (0.04)	0.13 (0.01)	0.37 (0.04)	0.36 (0.03)	0.47 (0.11)	0.57 (0.13)	0.76 (0.05)	0.45 (0.18)	0.26 (0.16)	0.45 (0.16)	0.64 (0.06)	0.44 (0.03)	0.63 (0.05)	0.58 (0.21)
$\text{NO}_2^- + \text{NO}_3^-$	0.99 (0.35)	2.48 (0.0)	4.04 (2.63)	2.51 (0.81)	7.10 (1.71)	1.54 (0.72)	15.33 (0.97)	1.03 (0.88)	1.55 (0.0)	1.14 (1.07)	0.54 (0.31)	1.21 (0.21)	0.64 (0.25)	12.20 (0.75)
Si(OH)_4	12.0 (0.15)	11.6 (0.0)	11.9 (0.48)	11.9 (0.25)	10.0 (1.38)	11.3 (0.62)	10.4 (0.74)	8.2 (1.43)	8.3 (0.0)	7.1 (0.43)	9.4 (1.21)	4.9 (1.04)	7.5 (1.22)	3.3 (0.11)
A_T	1460.8 (3.5)	1427.8 (1.7)	1420.0 (3.5)	1364.6 (1.7)	1341.4 (2.2)	1148.1 (0.6)	1056.6 (2.3)	1464.8 (0.9)	1432.0 (1.3)	1424.6 (5.3)	1366.0 (6.5)	1341.4 (2.0)	1152.9 (1.1)	1044.0 (5.6)
pH_T	8.51 (0.0)	8.42 (0.02)	8.37 (0.02)	8.15 (0.01)	8.02 (0.03)	7.12 (0.03)	6.89 (0.03)	8.64 (0.01)	8.58 (0.02)	8.59 (0.03)	8.38 (0.04)	8.51 (0.0)	7.58 (0.17)	7.88 (0.03)
DIC	1327.4 (2.0)	1319.3 (2.8)	1320.5 (0.1)	1307.6 (5.0)	1299.4 (3.0)	1251.8 (5.7)	1234.2 (4.1)	1295.7 (3.4)	1286.4 (7.0)	1276.3 (10.3)	1268.9 (8.3)	1216.5 (2.1)	1165.8 (22.1)	1015.3 (5.8)
$p\text{CO}_2$	121.6 (0.8)	152.1 (7.1)	172.2 (9.3)	290.4 (8.9)	392.6 (26.2)	2823.2 (223.8)	4432.6 (206.3)	86.3 (3.3)	100.9 (4.4)	97.3 (9.3)	162.2 (17.9)	112.6 (1.3)	760.8 (43.2)	421.2 (31.9)

This effect is consistent with the results of [Roleda et al. \(2008\)](#), who showed that the photosynthesis of *Nodularia spumigena* was reduced in nitrogen enriched medium. In the pH controls, the biovolume of cyanobacteria increased by on average 36% and 278% in the 3% pH and 10% pH treatment, respectively. Effects of pH, due to increased $p\text{CO}_2$, on cyanobacteria have been assessed in several studies (e.g. [Burford et al. \(2020\)](#)) and the results vary from no effects on Baltic filamentous cyanobacteria when exposed for 960 ppm $p\text{CO}_2$ ([Karlberg and Wulff, 2013](#); [Olofsson et al., 2019](#)) to higher biovolume of *Dolichospermum* spp. ([Wulff et al., 2018](#)). Responses to H_2SO_4 -induced acidification on cyanobacteria are however less studied. Dinoflagellates responded negatively in the 10% scr treatment after 14 days exposure; the biovolume was reduced by 61% compared to the control ([Table 3](#), [Fig. 3](#) and [Fig. S2](#) supplementary material). This negative response was even higher in the pH controls and significant effects were observed in all treatments as compared to the control.

The results also showed scrubber discharge water to have a significant effect on the biomass metric POC where an increased concentration was observed in the 3 and 10% scr treatments as compared to the control after 14 days exposure ([Fig. 4](#), [Table S2](#)). A similar pattern was observed for POP and PON, where the concentration increased with increasing scrubber discharge water. In addition, primary productivity was on average almost $9 \mu\text{g C l}^{-1} \text{h}^{-1}$ higher in the 10% scr treatment as compared to the control after 14 days ([Fig. 4](#) and [Table S2](#) supplementary material). Photosynthetic activity, determined as effective quantum yield ($\Delta F/F_m'$) and maximum quantum yield (F_v/F_m), was also significantly higher in the 10% scr treatment ([Fig. 4](#) and [Table S2](#) supplementary material).

In contrast, scrubber discharge water had a negative effect on bacterial biomass ([Fig. 4](#) and [Table S2](#) supplementary material). On the other hand, the cell-specific bacterial productivity was significantly higher in the 3 and 10% scr treatments as compared to the control after 14 days exposure ([Fig. 4](#) and [Table S2](#) supplementary material).

The very few studies of scrubber discharge water on single species have seen negative responses in photosynthetic activity for the cyanobacteria *Nodularia spumigena* ($\text{EC}_{10} = 8.6\%$) ([Ytreberg et al., 2019](#)), increased primary productivity for the diatom *Melosira cf. arctica* ($\text{EC}_{10} = 5.5\%$) ([Ytreberg et al., 2019](#)) and increased mortality and reduced feeding for the copepod *Acartia tonsa* when exposed to a concentration of 10% scrubber discharge water ([Koski et al., 2017](#)). Impacts of scrubber water on a microplanktonic community have to our knowledge only been assessed in one previous study ([Ytreberg et al., 2019](#)), where a microplankton community (dominated by the dinoflagellates *Peridiniella catenata* and *Gymnodinium* sp.) collected from the Baltic Sea during spring was exposed to scrubber discharge water. The results showed a significant increase in chlorophyll *a*, POP, POC and PON when the microplanktonic community was exposed to 10% scrubber discharge water. In the present study, significant increase in total biovolume of microplankton, bacterial productivity, POC and PON were detected in the 3% scr treatment ([Table 3](#) and [Table S2](#)) while a negative effect in bacterial biomass was detected in the 3% scr treatment as compared to the control.

3.3. Chemical responses

A_T , pH_T and DIC were all significantly lower in all scrubber discharge water and pH control treatments at day 0 and day 14, as compared to the control ([Fig. 5](#) and [Table S1](#) supporting material). The pH_T and A_T were slightly lower in the scrubber discharge water treatments as compared to the corresponding pH control treatments at the start of the experiment. For A_T , this effect was also apparent at the end of the experiment. In contrast, pH_T was slightly higher in the scrubber treatments as compared with the pH controls. This result could be explained by the higher photosynthetic activity in the scrubber treatments as compared to the corresponding pH controls. This hypothesis is also supported by the significant lower DIC concentration in the scrubber treatments as

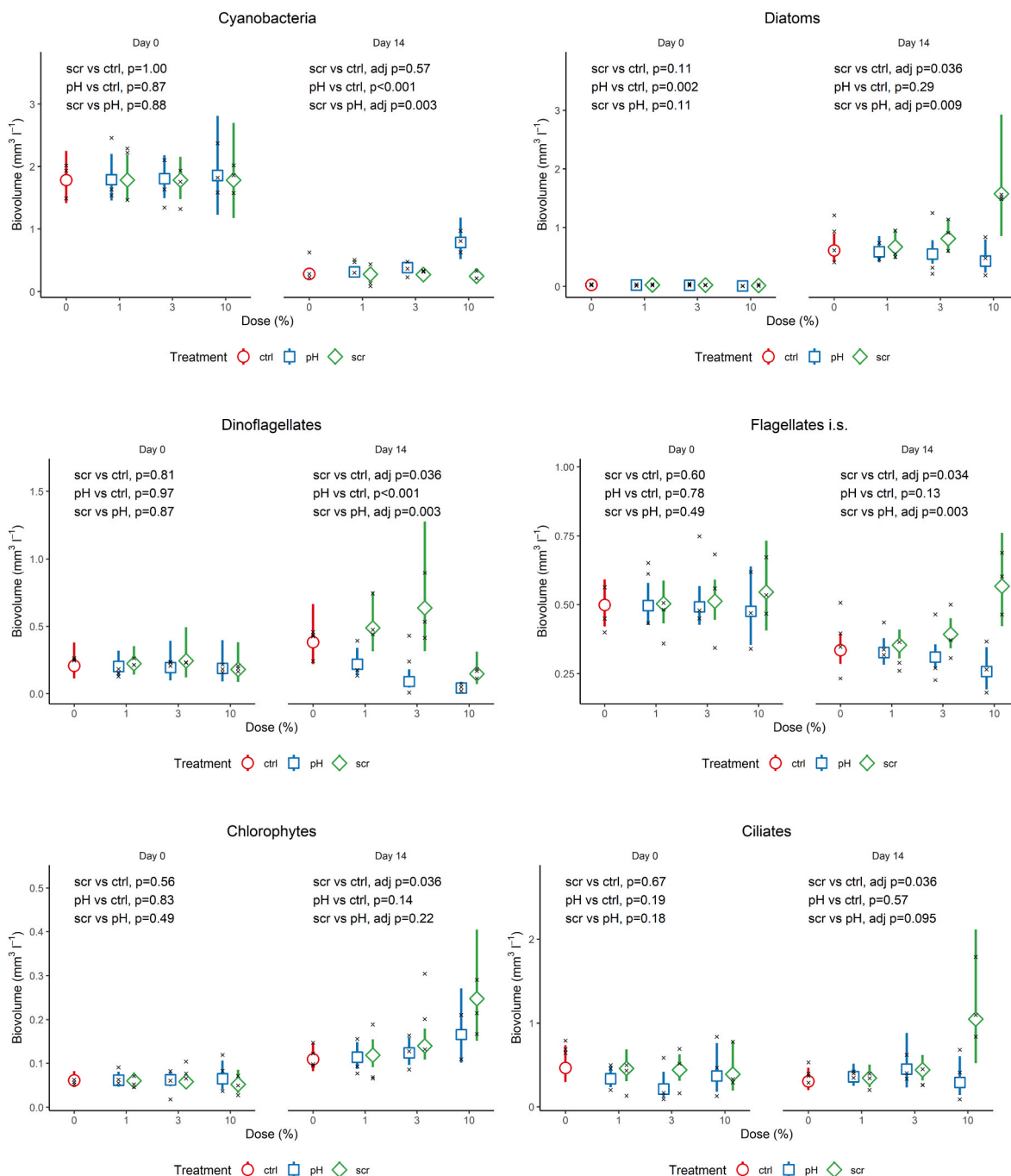


Fig. 3. Biovolume of microplanktonic groups in the control (ctrl), scrubber treatments (scr) and pH treatments (pH) at day 0 and day 14. Crosses (x) show individual measurements. Circles, squares and diamonds with error bars represent geometric means with 95% confidence intervals.

compared to the corresponding pH controls.

3.4. Environmental implication

The number of scrubber installations onboard ships has increased rapidly during the last years, where the vast majority (81%) are of open loop installations (DNV GL, 2020). These scrubber systems produce large volumes of discharge water which is continuously being discharged to the sea (typically 13,000 m³/day for a medium sized, 12 MW “roll on roll off” (RoRo) ship) (Turner et al., 2017), and there is growing concern regarding the environmental impact from wide-scale use of open-loop scrubbers (Hassellöv et al., 2020). Due to environmental concerns, restrictions or ban on using open-loop scrubbers have been

reported in an increasing number of ports, regions or territorial waters, e.g. China (territorial waters), Singapore (within port limits), Malaysia (territorial waters), Portugal (port waters), Belgium (ports and inland waters), Ireland (port waters in Cork, Dublin and Waterford), Germany (seaports adjacent to inland waterways and inland waterways), Bermuda (territorial waters), Panama (the Panama Canal) and the USA (Connecticut port waters and Californian waters) (IBIA, 2020).

The impacts of large-scale use of open loop scrubbers on the Baltic Sea ecosystems are difficult to assess. Presently, the species occupying the Baltic Sea are impacted by several other environmental and anthropogenic stressors, such as low (variable/brackish) salinity, stratified water and eutrophication and are therefore less resilient to further environmental changes. The impacts of additional stressors resulting

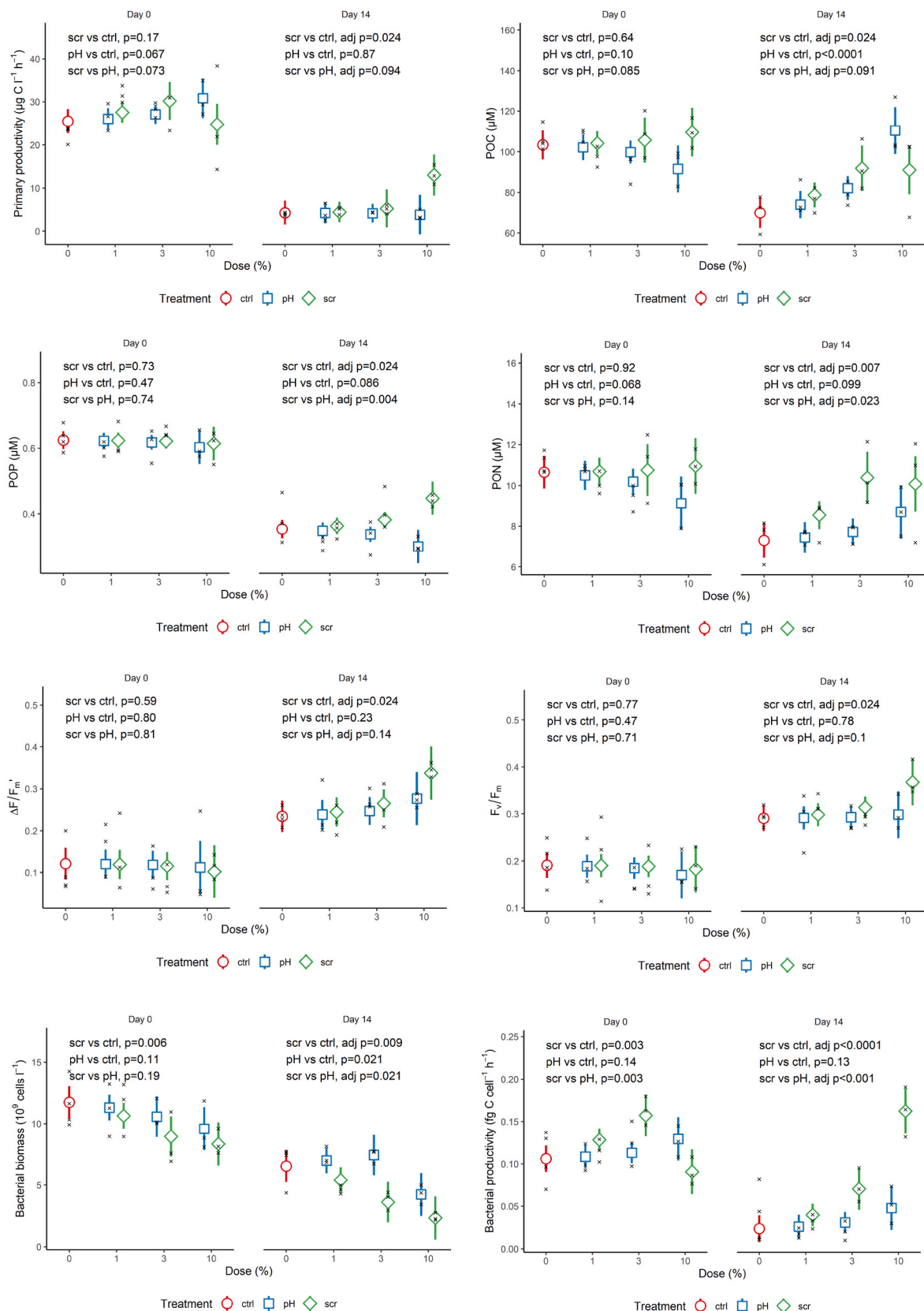


Fig. 4. Primary productivity, particulate organic carbon (POC), particulate organic phosphorous (POP), particulate organic nitrogen (PON) effective quantum yield ($\Delta F/F_m'$), maximum quantum yield (F_v/F_m), bacterial biomass and bacterial productivity in the control (ctrl), scrubber treatments (scr) and pH treatments (pH) at day 0 and day 14. Crosses (x) show individual measurements. Circles, squares and diamonds with error bars represent treatment means with 95% confidence intervals.

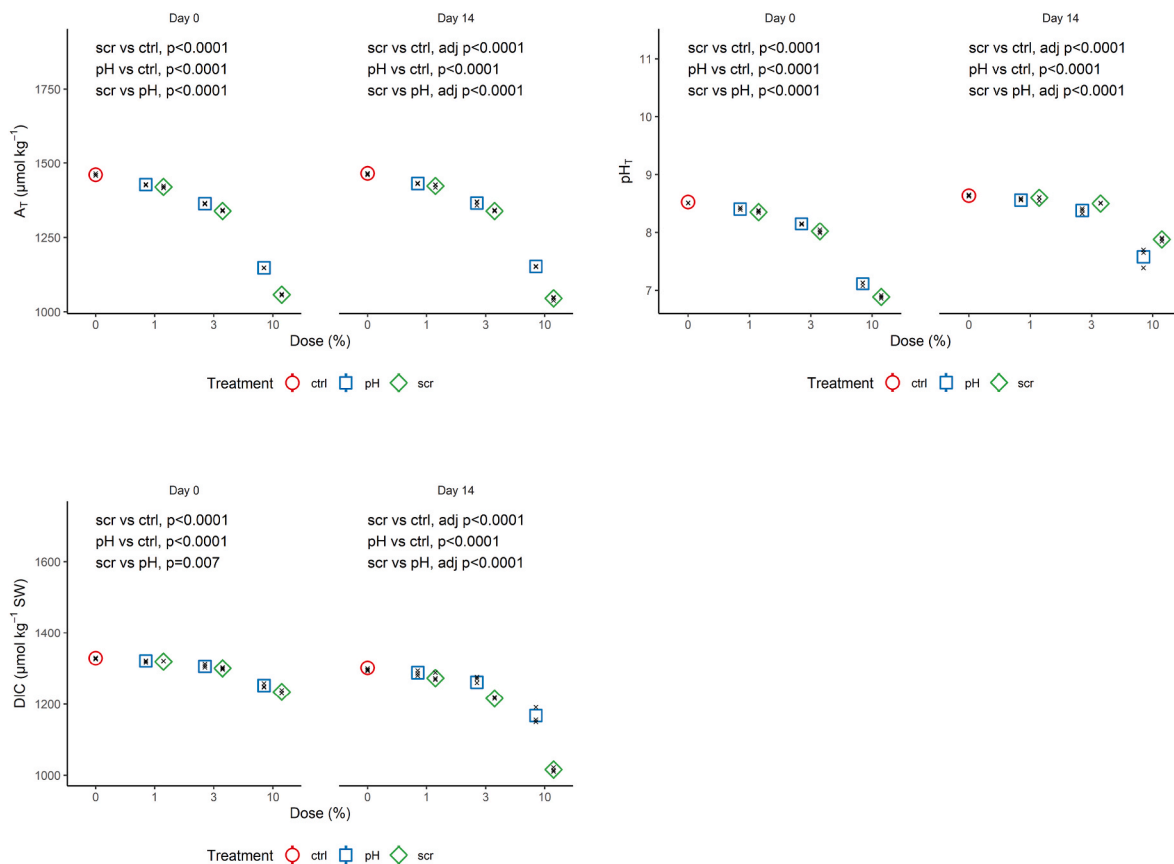


Fig. 5. Total alkalinity (A_T), pH_T and dissolved organic carbon (DIC) in the control (ctrl), scrubber treatments (scr) and pH treatments (pH) at day 0 and day 14. Crosses (x) show individual measurements. Circles, squares and diamonds with represent treatment means.

from scrubber discharge water, i.e. decreased pH, decreased alkalinity and increased concentrations of trace metals may be detrimental to the microplankton community of the Baltic Sea. There might be a shift in microplankton dominance towards groups or species more tolerable to low pH and high trace metal concentrations.

4. Conclusions

The use of open loop scrubbers on ships has created a new direct waste stream to the marine environment containing a complex mixture of metals, PAHs, acidifying pollutants and nutrients. To what extent a specific scrubber discharge water will impact the marine environment will be dependent on the concentrations and distribution of these stressors. In the current study, a discharge water with elevated nitrate concentration was assessed. While changes in pH alone had a minor impact on the microplanktonic community, a significant increase in total biovolume of microplankton was observed with increasing proportions of scrubber discharge water. Group-specific impacts were also observed with diatoms, flagellates *incertae sedis*, chlorophytes and ciliates increasing in biovolume while no effect was recorded for cyanobacteria. The different responses can likely be explained by the elevated nitrate concentration in the scrubber discharge water. It is not clear to what extent other discharge water components such as metals contributed to these Group-specific effects. In future research it is recommended that discharge water with other characteristics, e.g. with lower nitrate concentrations, are used to investigate how metals and PAHs from scrubber discharge water may impact aquatic ecosystems.

Credit author statement

Erik Ytreberg: writing – original draft preparation, Funding

acquisition, Data curation. Maria Karlberg: Writing- Reviewing and Editing, Methodology, Data curation. Ida-Maja Hassellöv: Conceptualization, Funding acquisition, Data curation, Writing- Reviewing and Editing. Mikael Hedblom: Writing- Reviewing and Editing, Methodology, Data curation. Amanda T. Nylund: Data curation, Writing-Reviewing and Editing, Visualization. Kent Salo: Data curation, Writing-Reviewing and Editing. Henrik Imberg: Formal analysis, Visualization; Writing- Reviewing and Editing. David Turner: Funding acquisition, Data curation Writing- Reviewing and Editing. Lucy Tripp: Data curation. Writing- Reviewing and Editing Joanne Yong: Data curation, Writing- Reviewing and Editing. Angela Wulff: Conceptualization, Funding acquisition, Data curation, Writing- Reviewing and Editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.envpol.2021.118251>.

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