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Ballast water treatment by ozone nanobubbles

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Abstract

BACKGROUND: The uncontrolled discharge of untreated ballast water, which is essential for a ship's optimal operation, is one of the major causes of sea environmental pollution by shipping. Among the disinfection methods available for ballast water treatment, ozonation is a commonly used method, but its performance is limited by the rapid ozone auto-decomposition rate. Nanobubbles (NBs) technology has attracted much scientific interest as it is characterized by a long residence time in the aqueous phase and a high surface area; therefore, ozone nanobubbles (OzNBs) are expected to enhance disinfection capacity and residual activity. The present study was designed to provide further insight into the inactivation of heterotrophic bacteria in saline water and to examine the use of OzNBs for disinfection of saline water.

RESULTS: The survival rate of *Escherichia coli (E. coli),* which was used as indicator microorganism, along with the ozone consumption at different salinities (1.5, 4, 8 and 15 PSU) and bacterial concentrations (10^7, 10^6, and 10^5 CFU mL^{-1}) with and without supplementation of OzNBs were investigated. The results indicated a statistical difference in the residual concentration of total residual oxidants (TRO) with the presence of OzNBs at salinity level 1.5 PSU and at 4 PSU only at the lowest bacterial content. At a low salinity and high bacterial concentration, the concentration of TRO was 6-fold higher in the presence of OzNBs.

CONCLUSION: The salinity of water has a strong impact on the residual concentration of ozone. When salinity is increased, ozone reacts more rapidly with the bromide and chloride ions. The use of OzNBs exhibited a greater disinfection performance and higher residual activity.

Supporting information may be found in the online version of this article.

Keywords: ozone nanobubbles; seawater disinfection; *E. coli*

INTRODUCTION

The vast majority of world cargo handling takes place by sea with suitable ships. For decades, shipping has been the leading choice for the movement of goods around the world.[1] However, this inevitably leads to a large environmental footprint on the planet that has attracted increasing attention because international shipping is considered a significant source of pollution derived from the emissions of toxic pollutants from internal combustion engines[2] and the largely uncontrolled disposal of seawater that is used as ballast.[3] Ballast water is a large amount of seawater that is stored in ballast tanks and is essential to maintain ship buoyance and maneuverability. Ballast tanks on a ship fill and drain seawater to offset the weight reduction that occurs when loading and unloading goods. Therefore, from just one ship, thousands of cubic meters of seawater loaded from a potentially polluted marine area can be disposed of in another area with completely different quality characteristics.[4] In fact, the ballast water discharged into a new marine environment may include non-indigenous species (NIS) that can result in extensive ecological and economic damage due to the changes they cause to the structure and functioning of marine ecosystems.[5,6]

To reduce the global spread of these invasive aquatic species, international regulations have set environmental limits for the number of organisms present in the ballast water discharged by ships. Since 2018, the International Maritime Organization (IMO) has defined the D2 standard to determine the maximum amount of viable organisms that are allowed to be discharged, a limit that all ships must meet by 2024 (Table 1).[7]

In general, within the context of minimizing environmental risks, the implementation of ballast water treatment systems is

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Table 1. Acceptable limit of indicator microbes according to the D2 standard[7]

Indicator microbes	Acceptable limit
Escherichia coli (E. coli)	<250 CFU per 100 mL
Intestinal *enterococci*	<100 CFU per 100 mL
Toxicogenic *Vibrio cholerae*	<1 CFU per 100 mL
Size of microbes	
≥50 μm	<10 viable organisms per m^3
≥10 μm and <50 μm	<10 viable organisms per mL

Table 2. Reaction of bromide and chloride with ozone[17]

$$O_3 + Br^- \xrightarrow{k_1} O_2 + BrO^- \quad [1]$$

$$O_3 + BrO^- \xrightarrow{k_2} O_2 + Br^- \quad [2]$$

$$2O_3 + BrO^- \xrightarrow{k_3} 2O_2 + BrO_3^- \quad [3]$$

$$H_2O + HBrO \overset{k_4}{\leftrightarrow} H_3O^+ + BrO^- \quad [4]$$

$$O_3 + Cl^- \xrightarrow{k_5} O_2 + ClO^- \quad [5]$$

$$O_3 + ClO^- \xrightarrow{k_6} O_2 + Cl^- \quad [6]$$

$$2O_3 + ClO^- \xrightarrow{k_7} 2O_2 + ClO_3^- \quad [7]$$

$$HClO + Br^- \xrightarrow{k_8} HBrO + Cl^- \quad [8]$$

addressed, with a view to providing discharged water quality according to the regulations. There are many systems that, through natural, chemical, and biological processes, eliminate microorganisms and suspend their growth. The most commonly employed methods for disinfection/sterilization are chlorination, ozonation, and ultraviolet (UV) irradiation.[8] Ozone is a strong oxidant and an efficient disinfectant against bacteria and viruses.[9] Several studies have been conducted on seawater ozonation and their findings confirmed that, when ozone is applied to seawater, secondary oxidants with disinfection capacity are formed that are expressed as total residual oxidants (TRO).[10–17] In conventional ozonation, ozone is directly bubbled into the aqueous solution and, since it has a low half-life, it decomposes rapidly, resulting in a low residual activity. An increasing number of studies that employ ozone delivery by nanobubbles (NBs) has been found to improve the disinfection capacity and residual concentration.[18] Nanobubbles are tiny spherical gas bodies sized at nanoscale (< 1 μm) that exhibit notable properties. First and foremost, the long residence time thanks to their stability is highlighted as a vital property.[19–21] Gaseous nanobubbles (NBs) have been found to remain stable in aqueous solutions for a long period of time due to their negligible buoyancy; this is in contrast to macro and microbubbles, both of which take a short period of time to rise to the liquid surface, where they burst out.[22,23] Considering their unique characteristics, NBs improve the mass transfer effect and oxidation ability due to the increased contact area of gas and water.[24] Moreover, the gas solubility and chemical reactions at the gas–liquid interface are remarkably enhanced.[20,23]

Although most studies have focused on water treatment and, particularly, on the inactivation of various microorganisms in the effluents by OzNBs,[25–29] research on their disinfection capacity in seawater remains limited. As mentioned by Meegoda *et al.*,[30] the generation of OzNBs under increasing salt concentrations by adding sodium chloride (NaCl) leads to a decrease in the magnitude of the zeta potential and a slight increase of the bubble size. Another experiment showed that the diameters of OzNBs under various salinity levels are stable, while a reduction of zeta potential in absolute value was reported.[31] The ozone demand in seawater ozonation is considerably influenced compared to freshwater ozonation, since the bromide Br^- and the chloride Cl^- (both of which are present in seawater) react with ozone very quickly, leading to the formation of oxidants that also have a disinfecting efficiency of their own, as shown in Table 2. Specifically, bromide is oxidized by ozone to secondary oxidants, such as hypobromous acid (HBrO) and the more stable forms, hypobromite ion (BrO^-) and bromate (BrO_3^-). The half-life of ozone in water is estimated to be only 5 s in the presence of high concentrations of bromide, while there is no hydroxyl radical formation observed in seawater.[32] In most oceanic waters, the pH is about 8 and, since the acid dissociation constant (pK_a) of $HBrO/BrO^-$ is approximately 9, the dominant bromine form will be HBrO.[33]

Despite the fact that the application of OzNBs technology has been widely investigated on wastewater, water disinfection, and degradation of organic contaminants, no studies have focused their application on ballast water treatment. The main objective of this research is to evaluate whether the use of OzNBs has any significant impact on microorganism inactivation and residual activity of ozone.

MATERIALS AND METHODS

Bacterial stock solution

E. coli was cultured in nutrient broth for 16 h and then the solution was centrifuged. The pellet of *E. coli* cells was washed by sterilized water 0.8% NaCl. Each sample was centrifuged at 3500 rpm for 15 min and resuspended in solution. The bacterial suspension reached an optical density equal to absorbance of 0.1 at 600 nm, as this corresponds to approximately 10^8 CFU mL^{-1} according to McFarland standard. The ozone disinfection of *E. coli* in three different bacterial concentrations (10^7, 10^6, and 10^5 CFU mL^{-1}) was added to the ozonated solution and samples were collected at predetermined time intervals (1, 5, and 10 min) into sterile tubes for further analysis. The initial bacterial concentration was confirmed in every disinfection experiment. In each sample, the residual ozone was immediately neutralized with excess sodium thiosulfate. After the ozone treatment, 0.1 mL of each sample in different dilutions was spread evenly over the nutrient agar-plate in triplicate. Following incubation at 37 °C for 24 h, the total viable count (CFU mL^{-1}) of the isolate was determined by the spread plate technique. All the disinfection experiments were conducted in terms of bacterial regrowth after 5 days, as suggested in the D2 standard.

All glassware used for the experiment was washed with deionized distilled water and autoclaved at 121 °C for 15 min; the ozonated water before bacteria addition was also tested for any bacterial contamination. The disinfection experiments were conducted at a temperature of 18 °C throughout the experimental process.

Ozonation experiments

In this study, seawater collected from a small harbor located in Agios Onoufrios (Chania, Crete, Greece) was used to run the batch disinfection tests. The main water quality parameters are given in Table 3. The water quality parameters were measured using a calibrated HQ4300 Portable Multi-Meter (Hach).

Table 3. Fresh water and seawater content descriptions: physicochemical parameters

Measured parameter	Fresh water	Seawater	Unit
pH	8	8.28	-
T	17.5	17.5	°C
Conductivity	0.297	59.8	mS cm^{-1}
TDS		38.2	g L^{-1}
Salinity		39.9	PSU
ORP		221.8	mV
Chloride (Cl^-)		32 769.84	mg L^{-1}
Bromide (Br^-)		94.01	mg L^{-1}

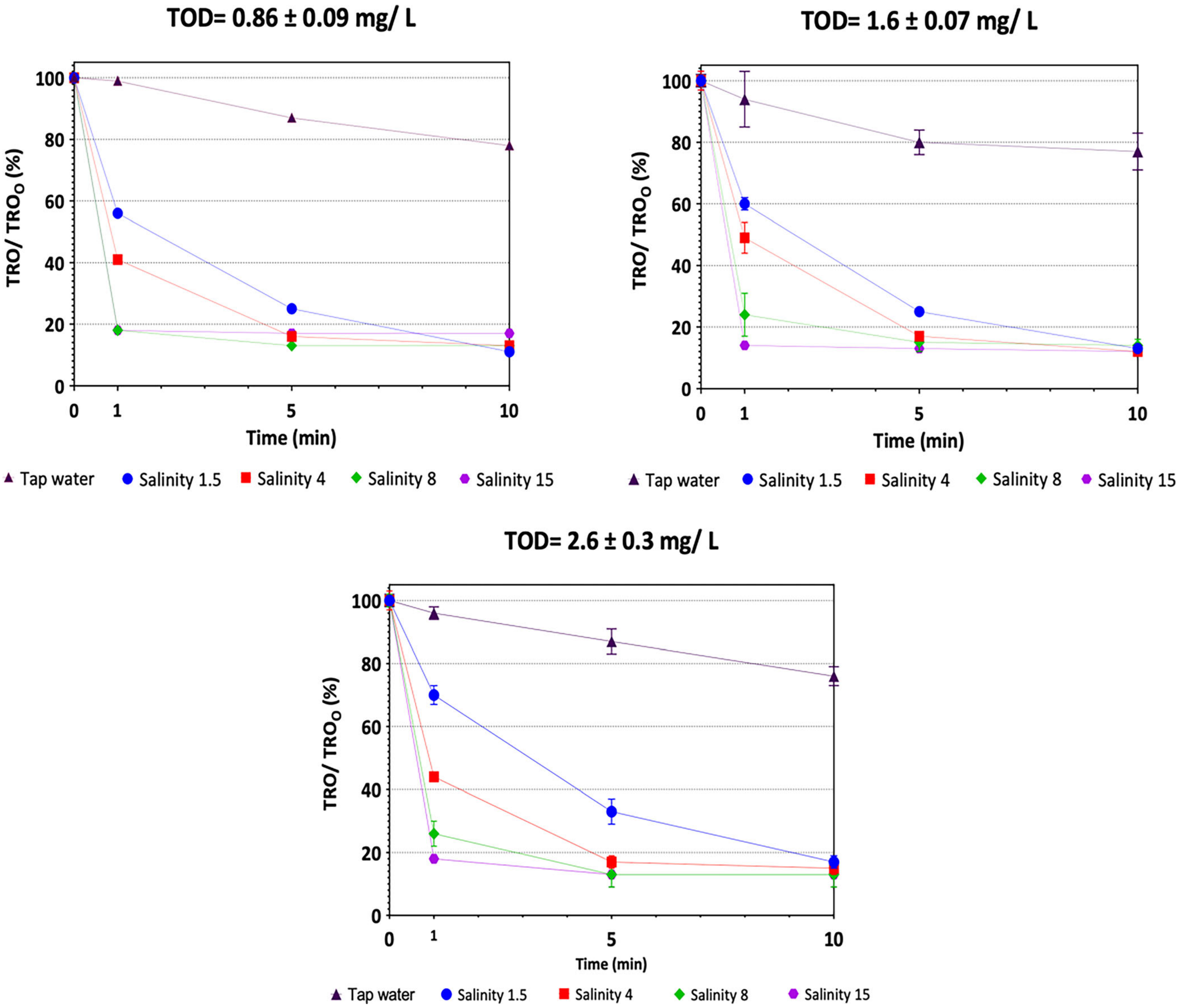

Figure 1. Effects of salinity on total residual oxidants (TRO) remaining, expressed as a percentage of different initial ozone concentrations (0.86 ppm, 1.6 ppm, and 2.6 ppm).

The experimental process used in this study is shown schematically in Supporting Information, Fig. S1. For OzNBs generation, the commercially available MK1 Nanobubbler™ (Fine Bubble Technologies (Pty) Ltd, Porterville, South Africa) was employed; this is a submersible device with the capability to generate nanobubbles at a concentration of approximately 10^8 bubbles/mL. The experimental set up consisted of a 350 L tank filled with tap water, in which the nanobubble generator was submerged (see graphical abstract). Ozone supplied by an ozone generator (Azure 2G, Wassertec, Cape Town, South Africa) was injected into

Table 4. ORP values in different salinities at ozone concentration 2.6 ppm

	1.5 PSU	4 PSU	8 PSU	15 PSU
ORP (mV)	883.7	806.0	795.0	779.8

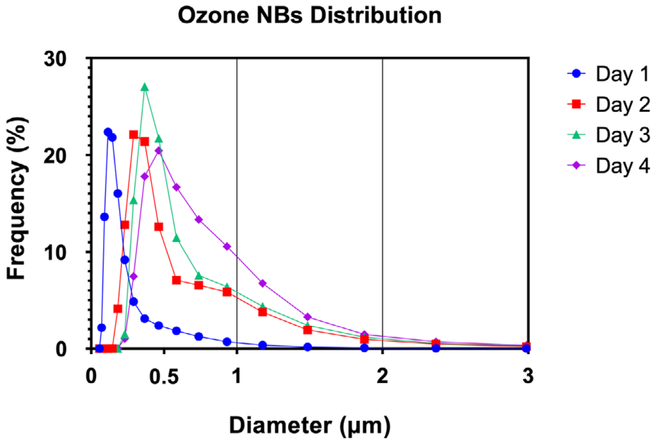

Figure 2. Effects of time on OzNBs diameter distribution.

the water through the gas intake of the MK1 Nanobubbler™. To increase the ozone concentration in the output of the ozone generator, high-purity oxygen from a compressed gas cylinder with a suitable pressure regulator was used as the feed gas. Ozone exhaust gas from the tank was carefully captured by ozone traps filled with 2% potassium iodide (KI) solution to ensure that no ozone gas escaped into the environment. The concentration of dissolved ozone was monitored directly from the tank during the generation of the ozone nanobubbles by an ozone amperometric sensor (Hach, Germany) with a working range of 0.005–2 mg-O_3/L and an accuracy of 3% or ± 10 ppb O_3.

The ozonation disinfection experiments were conducted in batch mode. Tap water containing ozone at the desired ozone level with and without the presence of OzNBs was added to unsterilized seawater in different ratios to obtain different salinities. The salinity in seawater samples was between 1.5 and 15 practical salinity units (PSU). The measurement of the ozone concentration (and the possible residual oxidants formed) in the aqueous solution used for disinfection was also tested by the Indigo Colorimetric Method.[34] For seawater treatment with dissolved ozone, the transferred ozone dose (TOD) was calculated according to the following equation:

$$TOD = \frac{C\,V_{ow}}{V_{sw} + V_{ow}}$$

where C is the ozone concentration in the ozonated water (mg L^{-1}); V_{OW} is the volume of ozonated water added in the flask (mL); and V_{SW} is the volume of seawater added in the flask (mL).

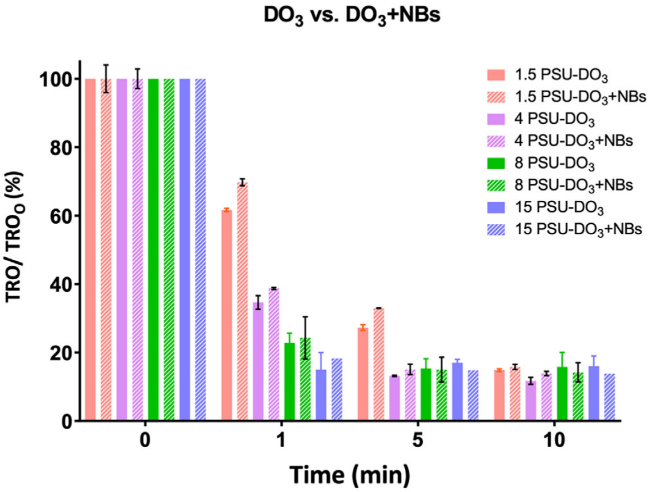

Figure 3. Total residual oxidants (TRO) remaining in different salinities (1.5, 4, 8, and 15 PSU) with and without the presence of OzNBs.

The reaction of indigo trisulfonate with the bromine formed in seawater is slow and is further inhibited by the addition of malonic acid (Supporting Information, Fig. S2 and Table S1). Hence, it is considered that the indigo decolorization is exclusively due to the ozone concentration and will be expressed in the results as TRO (Total Residual Oxidants) (mg-O_3/L).

To determine the difference in the efficacy between the treatments with dissolved ozone and those with dissolved ozone supplemented with OzNBs, a conventional ozonation system was also set up. The dissolved ozone was obtained by bubbling ozone gas into 1 L of tap water using a ceramic diffuser and the aforementioned experimental procedure was followed here, too.

Data were plotted as mean ± SD from at least three independent experiments. In every experiment, duplicate samples were taken at each time interval to estimate the microbial count and to measure the residual dissolved ozone. The temperature and salinity did not change in all experiments.

Dynamic light scatter analyzer

The nanobubble size distribution was measured by a SALD 7500nano nanoparticle size analyzer (Shimadzu, Japan), which uses the dynamic light scattering (DLS) method and is applicable to particle diameters between 7 nm and 800 µm. The refractive index of the material was chosen to be 1, since the refractive index of the air is less than that of water. After the generation of the nanobubbles and before the onset of the experiment, OzNB tap water samples were collected in triplicate from the upper and

Table 5. Effects of time on OzNBs median diameter as measured by DSL

Days	1	2	3	4
Median bubble diameter (µm)	0.136 ± 0.036	0.328 ± 0.059	0.388 ± 0.011	0.484 ± 0.087

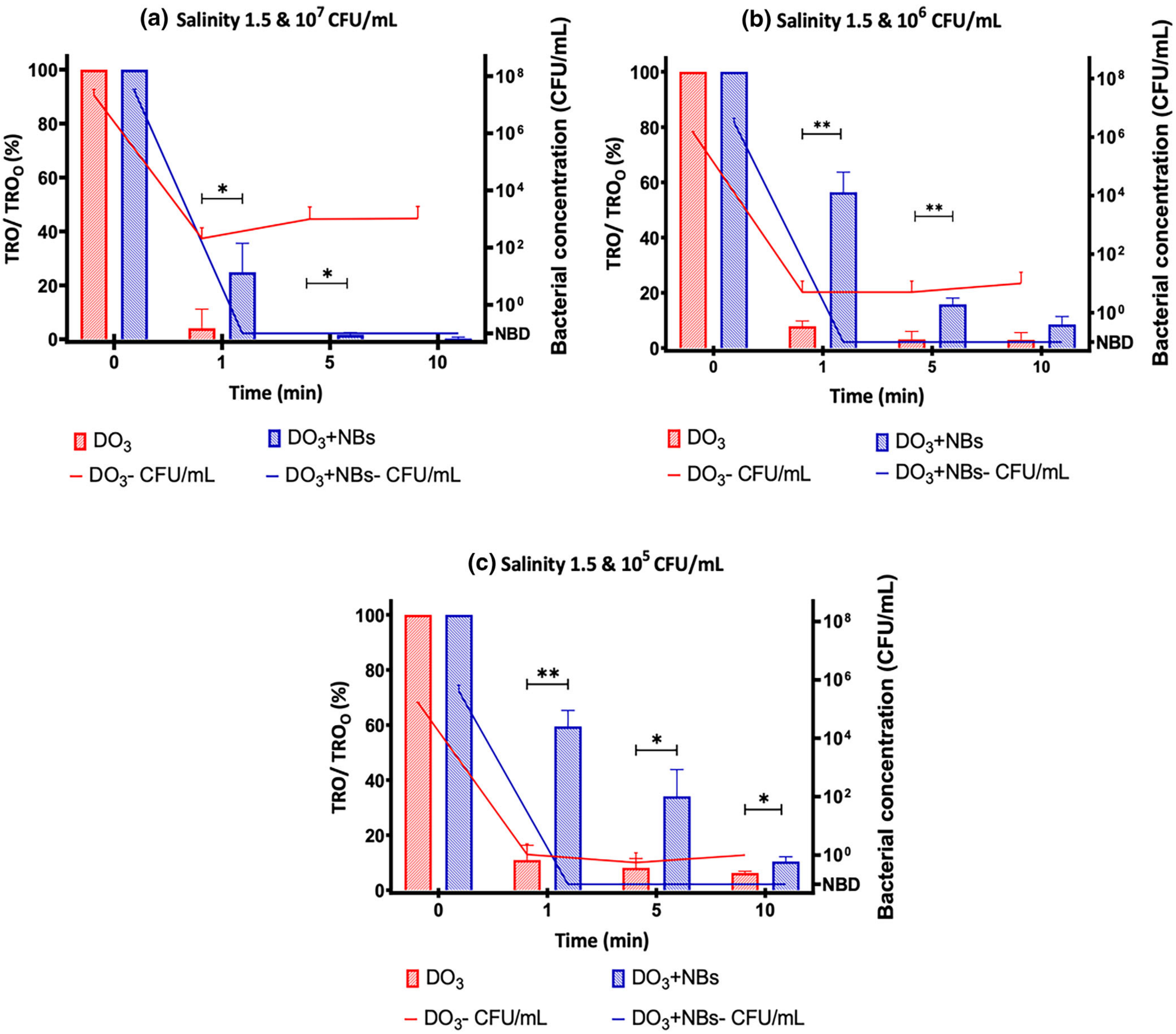

Figure 4. Total residual oxidants (TRO) remaining and bacterial concentrations after disinfection experiments with and without the presence of OzNBs in salinity 1.5 PSU and at bacterial concentrations: (a) 10^7, (b) 10^6, and (c) 10^5 CFU mL^{-1} (NBD, No bacteria detected).

lower parts of the tank and were analyzed immediately. To investigate the stability of the formed OzNBs with respect to time, samples after generation were sealed in glass vials, stored at ambient temperature, and measured every 24 h over the next 3 days.

Ion chromatography

The bromide, chloride, bromate, and chlorate concentrations were analyzed by ion chromatography. The detection limits of the analytical technique are 1 and 20 $\mu g\ L^{-1}$ for bromate and chlorate, respectively. A 10-fold dilution was applied to all samples to avoid chloride interference.

RESULTS AND DISCUSSION

Effects of ozone concentration and salinity on ozone reaction

The half-life of dissolved ozone in seawater is complicated compared to that in fresh water. To have a direct comparison, we performed a series of batch experiments identical to those conducted for disinfection without the presence of microbes. In Fig. 1, the ozone reduction over time is shown for four salinities (1.5, 4, 8, and 15 PSU) and at different initial ozone concentrations 0.86, 1.6, and 2.6 ppm. In the same figure, the ozone decay in tap water is also shown. As seen, the residual TRO was attenuated in both tap and saline water. In tap water, the residual TRO declined steadily, reaching a remaining percentage of approximately 77% in all initial ozone concentrations at a reaction time of 10 min. This is consistent with first-order decomposition kinetics, suggesting a half-life time of about 26 min (at 18 °C). The estimated half-life time ($t_{1/2}$) of tap water is in agreement with the degradation of dissolved ozone in distilled water, where the ozone $t_{1/2}$ is only 20 min (at 20 °C).[35,36] On the other hand, in water with the highest salinity (15 PSU), the residual ozone was lower than 20% of the initial ozone concentration within 10 min of reaction. At the highest ozone concentration (2.6 ppm), the $t_{1/2}$ of TRO levels at salinities 1.5, 4, 8, and 15 PSU were found to be 3.4, 0.98, 0.52,

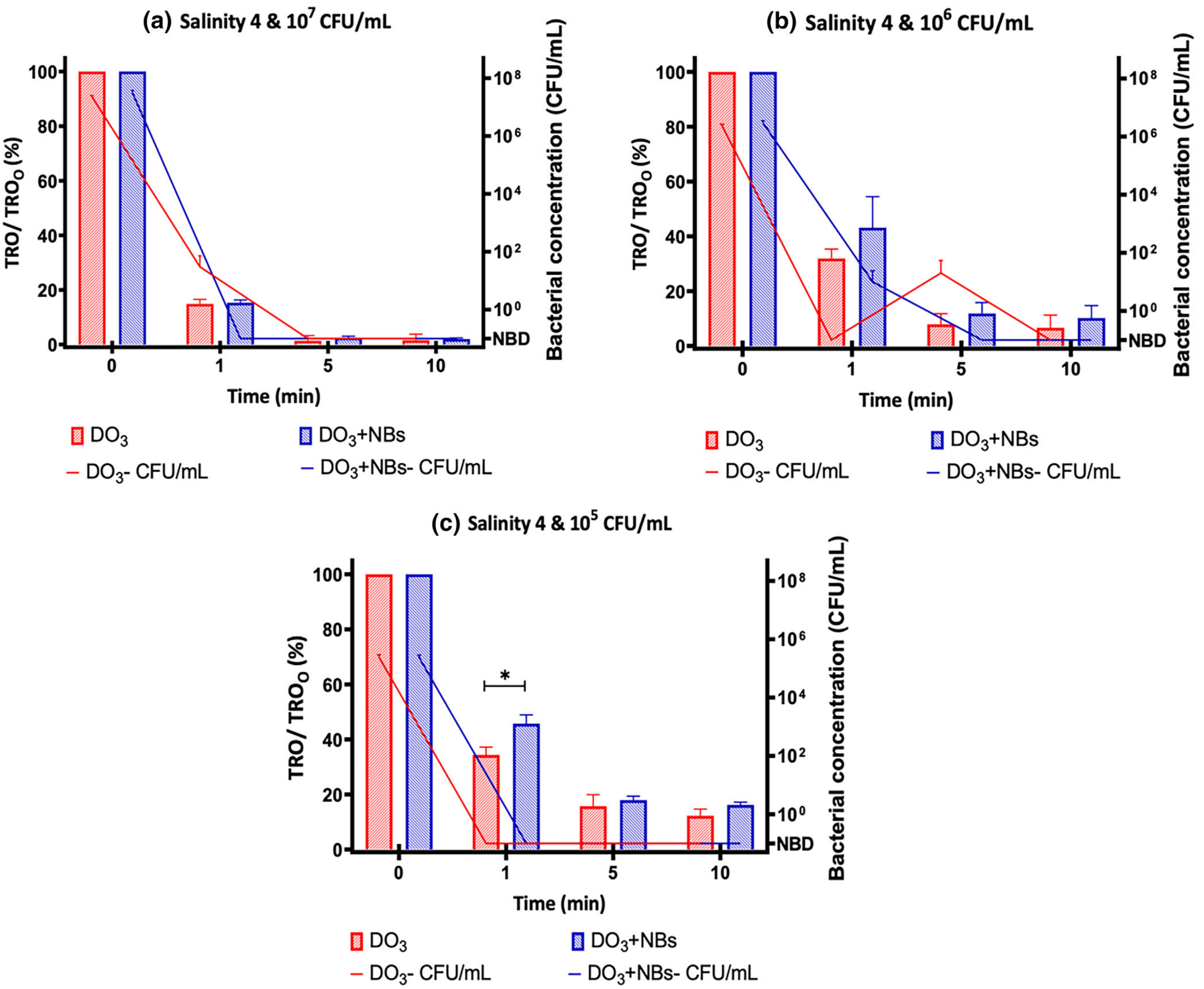

Figure 5. Total residual oxidants (TRO) remaining and bacterial concentrations after disinfection experiments with and without the presence of OzNBs in salinity 4 PSU and at bacterial concentrations: (a) 10^7, (b) 10^6, and (c) 10^5 CFU mL^{-1} (NBD, No bacteria detected).

and 0.40 min, respectively. The half-life time of TRO is clearly and substantially influenced when salt concentration is increased. As shown in Supporting Information, Fig. S3, the ozone dose did not have any strong effect on the ozone consumption at any salinity level, since the reduction is essentially the same among all the ozonation experiments. On the other hand, salinity displayed a significant impact on ozone depletion. The highest ozone consumption was observed at the highest salinity, as shown in Fig. 1.

Upon ozone addition, the ORP values rapidly increased from 221.8 to 883.7, 806.0, 795.0, and 779.8 mV at salinity 1.5, 4, 8, and 15 PSU, respectively (as shown in Table 4). During seawater treatment experiments, various oxidant residuals were formed, exhibiting a lower disinfection capacity than ozone. Thus, the highest ORP value was observed at salinity 1.5 PSU, where the concentration of oxidants formed in the ozonated saline water was lower due to reduced quantity of chlorides and bromides.

Size distribution of ozone nanobubbles (OzNBs) over time

As shown in Fig. 2, the effect of time after the preparation of the OzNBs water on the median diameter was examined. It is demonstrated that the OzNBs grow over time and become larger in size. This can be elucidated by the fact that the growth mechanism of OzNBs might follow the Ostwald ripening process, in which smaller bubbles tend to dissolve and redeposit to larger bubbles.[37] Before the ozonation experiment, samples were collected to evaluate the median diameter of the nanobubbles. On the first day, the median diameter was found to be 136 nm, as shown in Table 5. After 1 day, the ozone median diameter increased to 328 nm. The size of the ozone nanobubbles was further increased with time to 388 in 3 days and to 484 nm after 4 days.

Comparison of effects on ozone reaction with and without the presence of OzNBs

To investigate if the addition of OzNBs has any effect on TRO concentration, an experiment was conducted with the same transferred ozone dose with and without the presence of OzNBs, where the concentration of residual oxidants was recorded at time intervals (1, 5, and 10 min) in each case. Figure 3 displays the remaining percentage of TRO

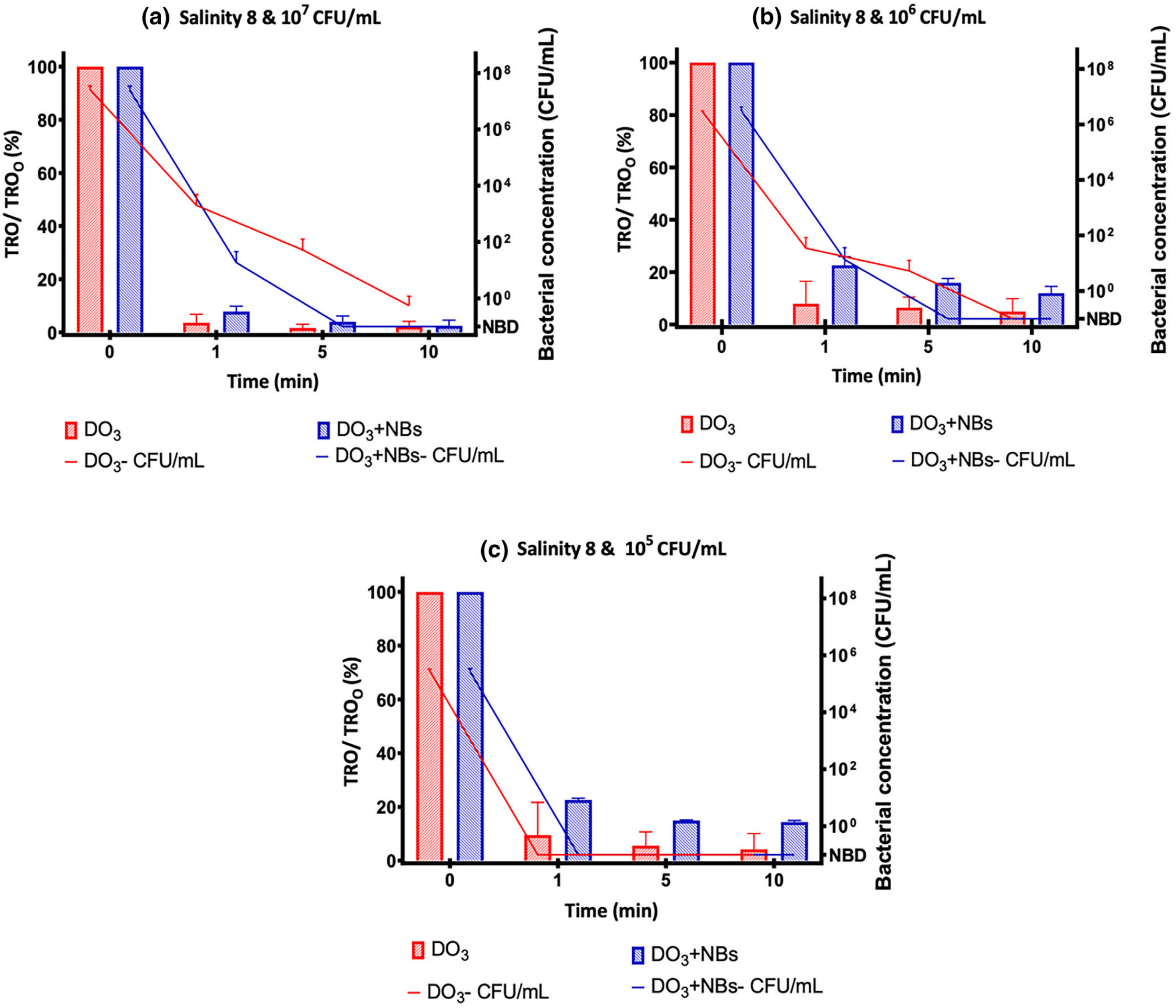

Figure 6. Total residual oxidants (TRO) remaining and bacterial concentrations after disinfection experiments with and without the presence of OzNBs in salinity 8 PSU and at bacterial concentrations: (a) 10^7, (b) 10^6, and (c) 10^5 CFU mL^{-1} (NBD, No bacteria detected).

concentration when the nanobubbles are present (or not) for every salinity level investigated in this study. As seen in Fig. 3, the difference of treatments is evident during the early minutes of reaction, where the presence of OzNBs enhanced the remaining concentration of TRO. In the fifth minute of reaction, only at salinity of 1.5 PSU, a difference can be observed, while at the higher salinities no significant variations are shown. In the last 10 min of the ozone reaction, no significant difference was observed in all tests.

Comparison of disinfection capacity of ozone with and without OzNBs at different salinities

Batch ozonation experiments with the addition of *E. coli* inoculum in three different concentrations in ozonated water containing OzNBs or DO_3 were conducted with a TOD of 0.89 ± 0.1 mg L^{-1}. Firstly, as shown in Fig. 4, at salinity 1.5 PSU, it is clear that the residual concentration of TRO is higher when the OzNBs are present for every bacterial concentration; therefore, it can be concluded that the residual activity is enhanced. On the other hand, it can be seen that the *E. coli* was completely disinfected in all initial bacterial concentrations. This is not the case for ozonated water containing DO_3 for the initial concentration of 10^7 CFU mL^{-1}, where the log reduction of bacterial concentration was approximately 4. It is worth mentioning that a t-test analysis was performed to investigate if the differences were statistically significant (Stars indicate significance levels: * for $P < 0.05$ and ** for $P < 0.01$). It is clear from the results that the concentration of residual oxidants was significant within the time of ozone reaction.

The investigation of saline water at 4 PSU with dissolved ozone supplemented with OzNBs showed that, in the highest bacterial concentration, no significant difference was observed compared to the treatment with dissolved ozone and without any OzNBs. The *E. coli* bacteria were successfully disinfected in both treatments. When the bacterial concentration was reduced to 10^6 CFU mL^{-1}, a higher residual concentration of oxidants was

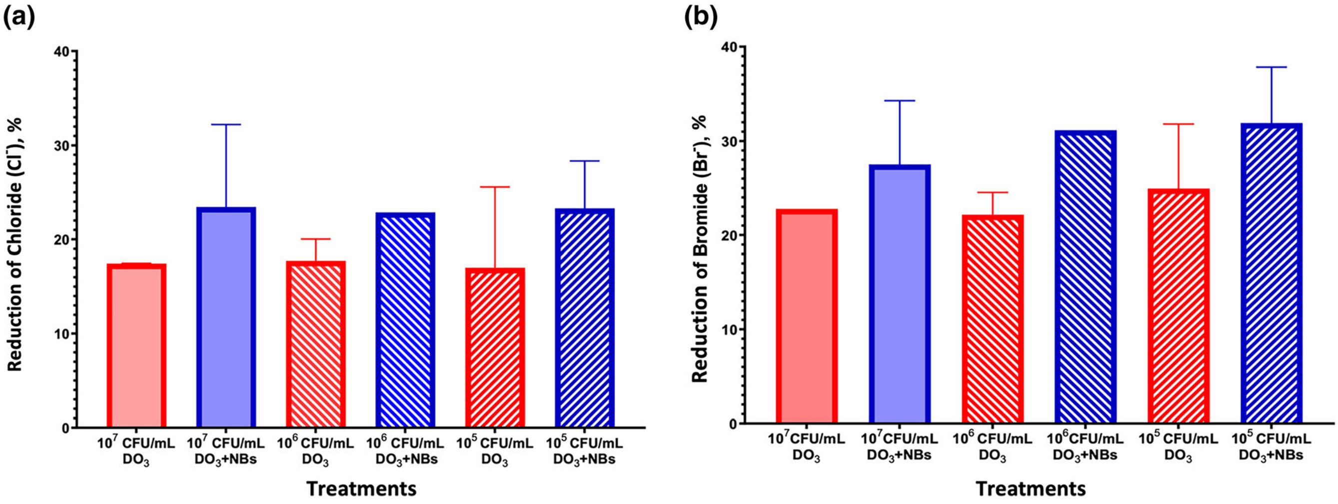

Figure 7. Reduction of chloride (Cl^-) and bromide (Br^-) at salinity 15 PSU after 10 min of exposure to dissolved ozone with and without OzNBs and for three different initial microbial concentrations.

observed; however, it was not statistically significant. After the first minute of ozone treatment, the concentration of TRO was stable, confirming the formation of by-products derived from the saline water ozonation that are more stable than ozone. The formation of stable oxidants can be confirmed in Fig. 5(c); it should be noted that, in this experiment, the remaining concentration of TRO was significantly higher when OzNBs were present at the first minute of reaction.

At salinity 8 PSU, the ozonation experiments with the highest bacterial concentration did not exhibit any difference in the residual concentration of oxidants, while a 10-min exposure led to total disinfection in both treatments, as shown in Fig. 6. For the other two bacterial concentrations, the TRO oxidant concentration was greater when OzNBs were present; however, no statistically significant difference was observed between the two treatments. Moreover, at the end of the experiment, a total inactivation of *E. coli* was achieved. When the salinity was increased to 15 PSU, there was no variation between the treatments, as seen in Supporting Information, Fig. S4. There was no significant effect on the residual concentration of TRO in all bacterial concentrations with and without the presence of OzNBs.

In all disinfection experiments, the microbial regrowth after 5 days was investigated and, in all cases, no regrowth was observed. More specifically, the bacterial concentration was maintained at the level that was previously recorded or it decreased. A control experiment without any ozone dose was performed to test any bacterial loss due to salinity or any other factor. During the experimental process, there was no decrease in bacterial concentration, since it was stable for 10 min. In 5 days, the bacterial concentration in some cases was reduced only by one order of magnitude; therefore, the reduction was due to the disinfection capacity of ozone (Supporting Information, Fig. S5). Treatment with ozone at all salinities formed bromate levels well below the maximum contaminant level of 10 μg L^{-1}.

Reduction of chloride (Cl^-) and bromide (Br^-) at salinity 15 PSU

Since the results from the ozonation experiments did not exhibit any differences between the treatments with and without the OzNBs in the highest salinity (15 PSU) (Supporting Information, Fig. S4), samples were collected to determine the chloride and bromide contents at the end of the disinfection experiments. Figure 7 demonstrates that utilization of ozonated water with OzNBs leads to an elevated reduction of chloride and bromide. This reduction cannot be captured by the indigo method since only ozone-based by-products were evaluated; this is malonic acid was used to avoid any interference from brominated and chlorinated disinfection by-products. Thus, it can be concluded that, when saline water with dissolved ozone contains OzNBs, it exhibits a higher residual activity.

This result is not surprising; although the total amount of ozone within the NBs is rather small, it remains for a longer period of time since the half-life of ozone at 20 °C in the gas phase is about 3 days (compared to only 20 min if it is dissolved in water). Hence, the ozone within the nanobubbles decomposes at a much slower rate than the dissolved ozone. Notably, among the different bacterial concentrations with the presence of OzNBs and with only DO_3, no substantial increase was detected. This can be elucidated by the fact that the transferred ozone reacts immediately with the chloride and bromide ions, all of which are in high concentrations and not with the bacteria, leading to total ozone depletion. Therefore, the reduction is the same in all treatments and the formed oxidants exhibit the observed disinfection capacity.

CONCLUSIONS

This study set out to compare the disinfection capacity and concentration of total residual oxidants (TRO) in saline water containing OzNBs compared to those of conventional ozone disinfection. As an emerging technology, nanobubbles have been widely utilized in water treatment; however, research on their use in ballast water treatment is limited. The results of the present study showed the efficacy of ozone (as TRO concentration) at different salinities. The implementation of OzNBs in the lowest salinity exhibited a significantly higher TRO than that of the dissolved ozone produced by bubbling ozone gas using a porous diffuser for all bacterial concentrations. At the highest bacterial content (10^7 CFU mL^{-1}) and at a salinity 1.5 PSU, the utilization of NBs led to a 6 times higher residual concentration of oxidants within the first minute of reaction and 1.6 times at the fifth minute; meanwhile, at a lower bacterial concentration (10^6 CFU mL^{-1}), the enhancement of TRO concentration was 7-fold and 5-fold at the 1st and 5th minutes, respectively. Dissolved ozone supplemented with OzNBs had a significant effect on the whole period of reaction at the lowest bacterial concentration. The utilization of nanobubbles in ozonation led to 5- fold, 4-fold, and 2-fold increases in the residual TRO concentration at the 1st, 5th, and 10th minutes of reaction, respectively. On the other hand, at the highest salinity, no difference was observed in TRO concentration; however, the results obtained from ion chromatography indicated that a greater reduction of bromide and chloride was achieved when OzNBs were used. Among the treatments at different bacterial concentrations, there was no statistically significant variation to indicate that the reaction with the halide anions was substantially rapid, leading to formed secondary oxidants that also exhibit a disinfection capacity. Our findings have practical implications for ozone nanobubbles to be used for ballast water treatment to inactivate microorganisms present in seawater because it was shown that OzNBs application leads to a more efficient ozonation since ozone utilization efficiency is higher. The most important limitation of this study lies in the fact that a high salinity (~30 PSU) could not be tested due to the experimental process being conducted under batch mode. However, this study has revealed that the application of OzNBs has the potential to enhance the disinfection capacity and the residual activity of ozone compared to conventional ozonation. Further work should be carried out under continuous mode to establish whether the delivery of ozone via nanosized gas bodies can ameliorate the inactivation efficiency in highly saline water.

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CONFLICT OF INTERESTS

The authors declare no conflict of interests.

AUTHOR CONTRIBUTIONS

Conceptualization, N.K. and P.S.; methodology, P.S.; formal analysis, P.S., N.K.; investigation, P.S., E.K., K.K.; chemical analysis, P.S., E.K., K.K.; data curation, P.S.; writing—original draft preparation, P.S.; writing—review and editing, N.K., P.S.; supervision, N.K.; funding acquisition, N.K. All authors have read and agreed to the published version of the manuscript.

DATA AVAILABILITY STATEMENT

Data available upon request.

SUPPORTING INFORMATION

Supporting information may be found in the online version of this article.

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