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Microcosm approach for brine impact assessment from seawater desalination on benthic assemblages

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ABSTRACT

The brine discharge from seawater desalination has a strong impact on marine community, especially benthic community near brine outlet. The aim of this study is to examine the effect of brine discharge over soft bottom benthic community and changes of sediment quality by periodic brine exposure and water circulation confinement using small-scale benthic microcosms. Transparent acrylic cylinder (1 m in length and 0.5 m in diameter) was used to examine the changes of benthic community structure and sediment quality in microcosm. Salt bags were placed inside of microcosm and the plastic cover partially sealed the open area to maintain salinity at a certain level. Various chemical and biological parameters were analyzed using the sediment samples collected with every two-week interval for 12-week experiment. Major estimated parameters were sediment chemical oxygen demand and total sulfur concentration for sediment quality, and meio/macrobenthic community structure for biological parameters. There were significant differences in some parameters in terms of sediment quality and benthic assemblages between treatment effects. This microcosm approaches to investigate brine impacts of benthic assemblages can a useful tool for gapping between laboratory and field estimations and also to make decision for permitting the level of brine to marine ecosystem.

Keywords: Benthic microcosm; Brine discharge; Benthic assemblage; Meiofauna; Macrofauna; Community structure

1. Introduction

The current world population has exceed seven billion people, with over 40% suffering from water shortage. As a result, unconventional water resources such as seawater desalination are increasingly becoming evitable sources to alleviate water scarcity. The introduction of desalination plants, however, has been associated with potential environmental impacts, mostly like open discharge of concentrated brine into marine environment. Desalination plants have induced significant changes of water quality, mainly due to the large volume of brine discharged into the sea, which can reach salinities of between 38 and 90 psu. The importance of all impacts depends on the vulnerability of the species and ecosystems receiving the discharge, general hydrodynamic field, and the brine flux, among other factors [1,2]. Furthermore, the discharge water contains high saline water including

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various chemicals during the pre-treatment of the desalination process and there has been damage to marine fauna and flora in the area of the brine outlet [3]. The extent of vulnerability of the marine environment to salinity differed from place to place. And the range of the geographic distribution and resistance to changes by the nature of the marine and by the origin of the surrounding organisms can be applied. The rarity of the natural habitat and the importance of the environment are also significant [4].

Mostly, microcosm studies have been focused to explain the relationship between environmental stresses, such as reclamation, contamination, or physical disturbance under field and experimental conditions, and community responses for benthic organisms [5–8]. However, ecological information including recruitment and survivals of benthic organisms was not clearly specified with salinity limit which definite damage was caused to benthic organisms [9]. Several studies have been conducted to assess the effects of hyper-saline water of some species or communities. For example, Pagés et al. demonstrated sensitivity of the seagrass *Cymodocea nodosa* to hyper-saline condition using microcosm approaches and they indicated that shoot may suffer deterioration or mortality under hyper-saline conditions at preliminary experimental study [10], while no significant changes were detected in a field study carried out by meadow close to the brine discharge from a desalination plant [11].

It is not well known about experimental designs to assess ecological impacts of benthic organisms in microcosm experiment. The range of the effect of the brine on the marine environment depends on bathymetry and on the hydrological characteristics of the benthic environment such as the type of bottom currents and waves. Despite recent advances in the field study, knowledge of the effects on marine benthic community with brine exposure restricted to a few species [12,13]. Some species exposed to changes in salinity cannot suffer osmotic stress, with the consequent changes at the biochemical and physiological levels [14,15]. There has been few studies that tried to assess brine impact of benthic organisms caused by changes between the brine flux and hydrodynamic field through the microcosm experimental approach. Specially, ecological information such as recruitment and survivals of benthic organisms is little.

Therefore, the main objective of present study is to obtain the expanding knowledge of brine impact of benthic organisms on periodic short-term brine exposure and changes of hydrodynamic condition and to estimate the potential ecological effects associated benthic fauna inhabiting a coastal area from the performance of microcosm approach.

2. Study area and experimental design

The study area, Ganghwa tidal flat on the western coast of Korea, has a very strong tidal movement, with a maximum tidal range of 9 m and mixed semidiurnal M_2 tides [16]. The study area is shallow with a depth ranging from 0 to 2 m and the substrate is predominately covered by a thin layer of fine and medium sands. Microcosm study was conducted to assess the impacts of water circulation and brine exposure on sediment environment and benthic fauna during 12 weeks.

Microcosms were made of transparent cylindrical chamber (100 cm high $\times$ 50 cm diameter) and buried to an approximate depth of 50 cm on the mud tidal flat (Fig. 1). Treatment effects were water circulation confinement and brine exposure with four replicates for each treatment group. A total of 20 microcosms were installed over 400 m^2 area and filled with 10 cm sediment on the bottom, sieved with 0.5 mm to remove the macro-invertebrates and large-sized particles to make the each microcosm homogeneous at initial condition. Top open area of microcosm was partially sealed with plastic cover, 50 and 75% in terms of area, to maintain the brine effect by limiting the water circulation by flooding within the microcosm. Eight microcosms were placed with salt bags inside of pipe (90 cm high and 30 cm diameter) to increase the salinity inside of microcosms during study periods.

The treatment labels of experimental microcosm units are given in Table 1. Four controls had no treatment effects with fully opened top and no salt bags inside, eight microcosms with 50% covered top with salt bags inside for four microcosms and without salt bags for the other four, and eight microcosms with 75% covered top with salt bags inside for four microcosms and without salt bags for the other four. The salt bags were replaced every two weeks and water and sediment samples were taken every two weeks to analyze water/sediment quality and benthic meiofauna.

3. Methods for assessment of sediment quality and benthic organism assemblages

About 200 g of surface sediment samples was taken from each microcosm to examine the chemical oxygen demand (COD) and total sulfur (TS) analyzed by marine environment standard test method [17]. Samples for meiofauna were taken by the modified disposable 50 mL plastic syringes (2.65 cm internal

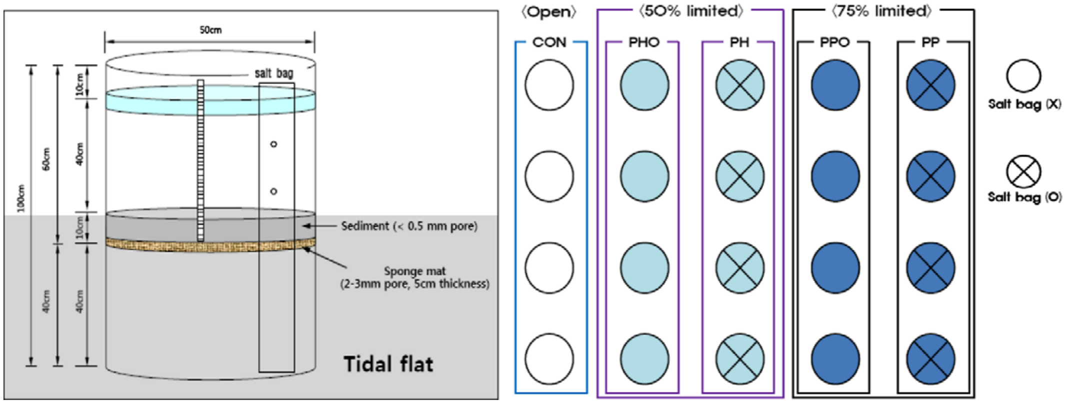

Fig. 1. Schematic description of experimental microcosms and treatment effects.

Table 1
Summary of the benthic microcosm experimental design and measurement parameters

Treatment labels	Test conditions	Measured parameters
BG	Natural mud flat without any treatment effects	TS, COD, benthic meiofauna, and macrofauna
Control	Microcosms without salt bags and with fully opened top	
PH-O	Microcosm with 50% closed top without salt bag	
PH	Microcosm with 50% closed top with salt bag	
PP-O	Microcosm with 75% closed top without salt bag	
PP	Microcosm with 75% closed top with salt bag	

diameter) inserted into the sediment. The sample was extruded from the syringe and each 2 cm horizon preserved 4% formaldehyde and Rose Bengal stain for separate sorting and identification. The formalin-preserved stained samples were washed gently in filtered fresh water then poured through a metal sieve with 0.5 mm mesh size in order to remove macrofauna and large particles. The passed sediment was then washed again on a sieve with mesh size 63 μm to remove fine silt and clay. Meiofauna in sediment retained on this last sieve was extracted using methods described in previous study [18]. The stained fauna were sorted and counted into major taxa, and the existing number of individuals was standardized into the number of individuals/10 cm^2. Macrofauna (0.12 m^2 to an approximate depth of 10 cm) was sieved in situ through a 1.0 mm mesh at final sampling. Faunal samples were fixed in 4% formalin, stained with drops of Rose Bengal, and subsequently preserved in 70% ethyl alcohol. Benthic samples collected in the field were transported back to the laboratory within 2-h of collection and sorted according to taxonomic groups, counted, and identified to the lowest possible taxonomic level using relevant identification guides [19,20]. For meiofauna and macrofauna, community indices such as Shannon-Weiner index (H′), Margalef species richness (d), and Pielou evenness (J′) were calculated from the species-abundance data from each microcosm using PRIMER v5 [21].

4. Variations of salinity concentrations in microcosms

The mean salinity variations with short-term brine exposure in each treatment (except background [BG]) are presented in Fig. 2. In this study, salinity concentration was not maintained as initial setup. In some cases, difference of salinity in each treatment was obviously observed during experiment. Especially, salinity concentration at first sampling for treatment group PP was measured to 51.0 psu and then gradually decreased to 16.2 psu. The salinity variations in other treatments were same as those observed at PP during experiment (Fig. 2A). It may be related to a great quantity of precipitation during the rainy season and periodic intrusion of normal seawater by strong tidal current in the study area. The mean concentration of salinity was high in treatment group PP (26.2 ± 13.1 psu) which had the limited seawater circulation

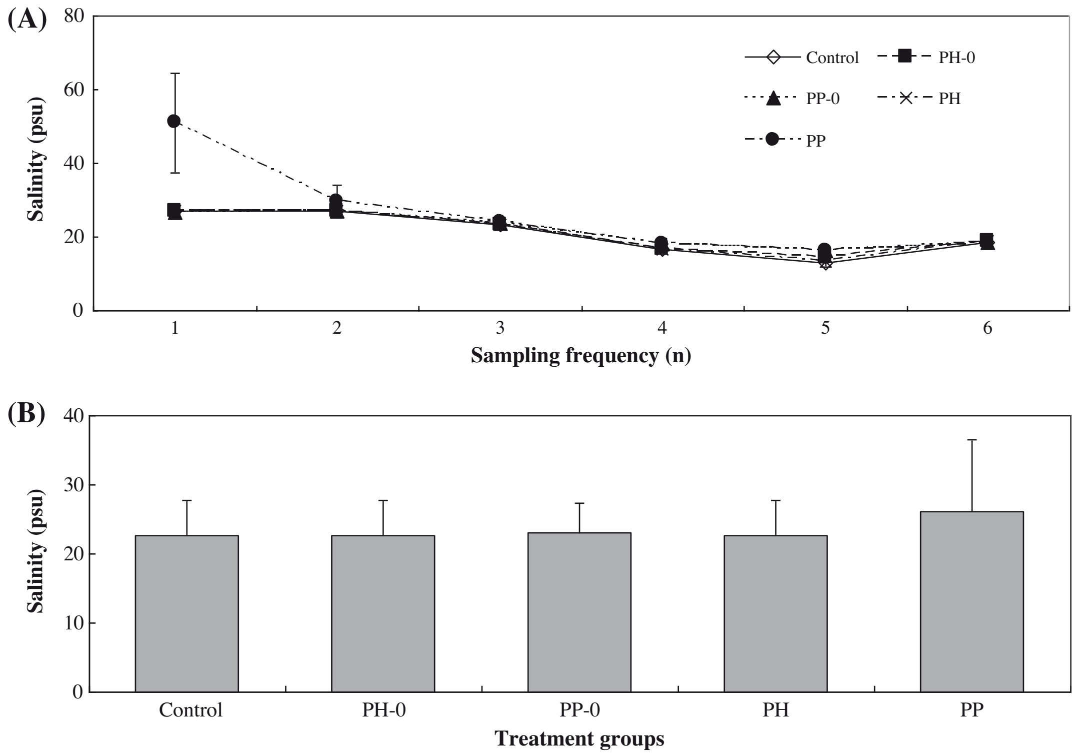

Fig. 2. Concentration of salinity measured at each treatment group. Values are means of four replicates at each treatment group.

and periodic brine exposure, but other treatments were maintained within 21.0 ± 6.0 psu (Fig. 2B).

5. Variations of sediment quality

COD concentrations were 6.17 ± 0.72 mg/L at BG and 8.18 ± 1.95 mg/L at control, respectively, and they gradually increased and distinguished between control groups (control and BG) and treatment groups (limited seawater circulation and periodic brine exposure) over the duration of the experiment (Fig. 3A). TS concentrations of sediment at each treatment group varied widely from 0.059 ± 0.009 to 0.101 ± 0.010% and their distributions were quite similar to COD. TS concentrations at PH-0 and PP-0 were higher than those of PH and PP treatments which were limited in water circulation and exposed to short-term salt stress. However, TS at control and BG were clearly lower than treatment groups (Fig. 3B). For TS, several studies suggested that areas with the highest sulfate leaching are associated with high precipitation, irrigated conditions, coarse-textured soils, shallow soils with low anion exchange capacity in soil environment [22]. Organic matter may be the major source of TS for the soils in the humid and semi-arid climate [23]. In this study, there were significant differences in sediment quality in terms of the COD and TS between non-treated and treated microcosms. This may be associated with the precipitation increment of organism matter, anaerobic condition, and short-term brine stress in sediment caused by lack of water circulation between inside and outside of microcosm. Several studies reported that redox boundary was caused by the release of metals into the bottom and anoxic environment where scavenging of dissolved metals and re-sedimentation take place in relatively short periods of time [24,25]. Some case studies suggested that redox reactions associated with natural organic matter oxidation may also play an important role in the salinity transition. Additionally, redox stratification of 4–5 cm light-grey coloured sediment over a dark layer was observed in some replicated treatment samples (PH, PP-0, and PP treatments) after about 2 months and this condition maintained until the end of the test period [26].

6. Changes of meiofauna assemblage

Sixteen meiofauna taxa were identified and densities ranged from 2,473 to 9,873 individuals/10 cm^2 during study (Table 2). Total abundance of meiofauna with sampling frequency at control, BG, and PP-0

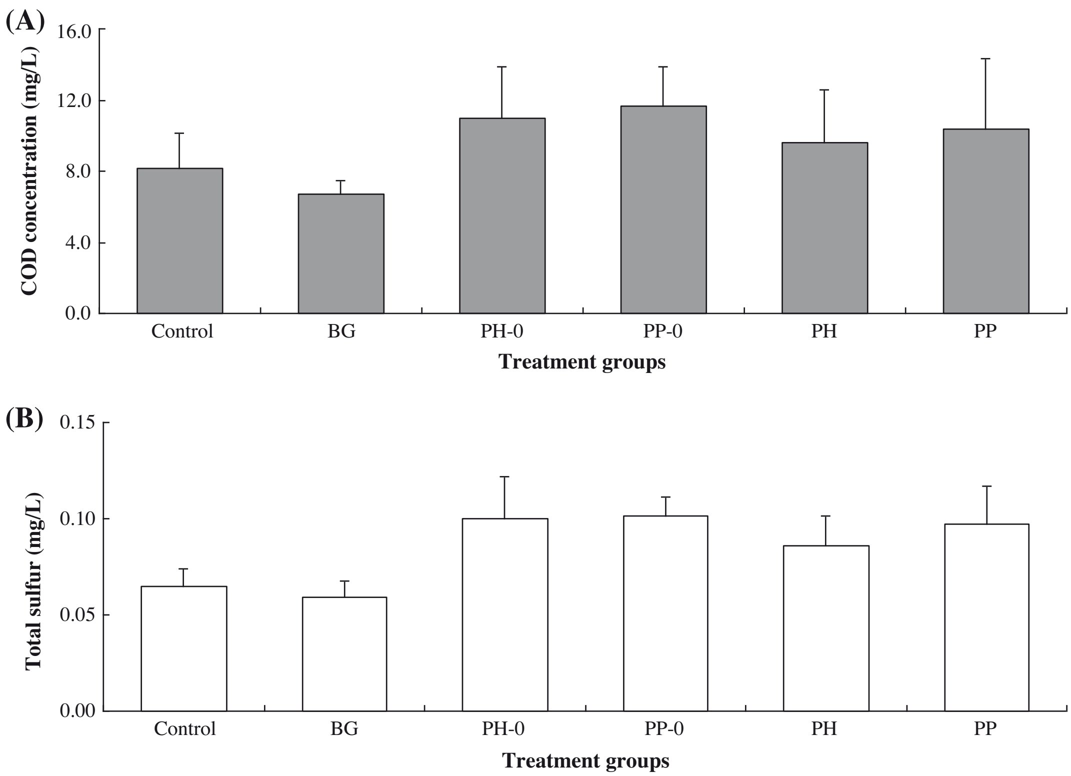

Fig. 3. Concentration of COD (A) and TS (B) at control, BG, and each treatment group. Values are means of four replicates.

treatment increased from the initial to third and fourth sampling, thereafter decreasing until the end of the experiment. Abundances in PH and PP treatment were relatively lower than those of the other treatments and variation of total individuals was partly the same as that observed by others over the duration of the experiment (Fig. 4A). At the end of experiment, the final abundance of meiofauna estimated at each treatment averaged from 412 ± 337 to 1,646 ± 1,038 individuals/10 cm^2. High abundance was observed at control and low abundance at treatment group PH and PP (Fig. 4B).

The species number at all the treatments increased from the initial to second sampling period and then gradually decreased until the end of the experiment. The species number of meiofauna obviously differed among sampling frequencies (Fig. 5A). However, it was not clear that mean species number was clearly influenced by treatment effects. Species number was relatively high at BG and PP-0 treatment but there was no difference between the other treatment groups (Fig. 5B). Over the entire study periods, dominant taxa collected in each treatment were foraminifera, turbellaria, nematode, harpacticoid copepod, and copepod nauplii. Nematoda was numerically dominant among the major meiofauna taxonomic groups representing 70.1 ± 10.7% of total population followed by copepod nauplii 13.3 ± 6.4%, turbellaria 7.9 ± 2.6%, harpacticoid copepod 4.7 ± 2.3%, and the remaining of the groups with 1.5%. Salt-treated groups (PP and PH) showed relatively higher composition copepod nauplii and lower contribution of nematoda, and the other groups with no significant differences in species composition (Fig. 6).

In this study, decrement of abundance and species number of meiofauna with sampling frequency were similar among treatment groups. Mean abundance at control groups was relatively higher than that of salt-treated groups. However, in terms of composition of dominant taxa, relative contribution of nematoda was higher than that of harpacticoid copepod and copepod nauplii. Several studies revealed that sediment quality can be differed with the degree of water circulation (or mixing strength), which reflects different hydrodynamic regimes, and is expected to have a significant influence on the abundance, diversity, and species composition of meiofauna assemblages (especially nematode) [27]. Additionally, some researchers concluded that salinity might be the principal factor correlated with assemblage structure and species diversity was highest at the higher salinity sites [9]. Therefore, differences in meiofauna abundance and species composition between treatment groups in this

Table 2
Total abundances and species numbers of meiofauna by experimental groups

Taxa	Meiofauna at each treatment group (individuals/10 cm^2)					
	Control	BG	PH-0	PP-0	PH	PP
Foraminifera	196	186	104	481	69	102
Tintinopsis	–	15	–	–	5	–
Turbellaria	444	417	507	894	322	159
Nemertina	3	2			1	17
Nematoda	8,195	4,828	5,002	5,457	1,683	1,462
Gastrotricha	–	–	15	11	–	5
Harpacticoida	171	367	131	382	214	158
Copepod nauplii	843	572	429	1,171	549	562
Unid. Crustacea	3	9	4	31	7	8
Ostracoda	3	53	2	8	–	–
Gammaridea	2	1	–	1	1	–
Cumacea	–	4	–	–	–	–
Bivalvia	6	6	4	3	1	–
Polychaeta	7	7	1	22	2	–
Gastropoda	–	1	–	–	–	–
Bryozoa	–	–	–	3	–	–
Total	9,873	6,468	6,199	8,464	2,854	2,473
Species number	11	14	10	12	11	8

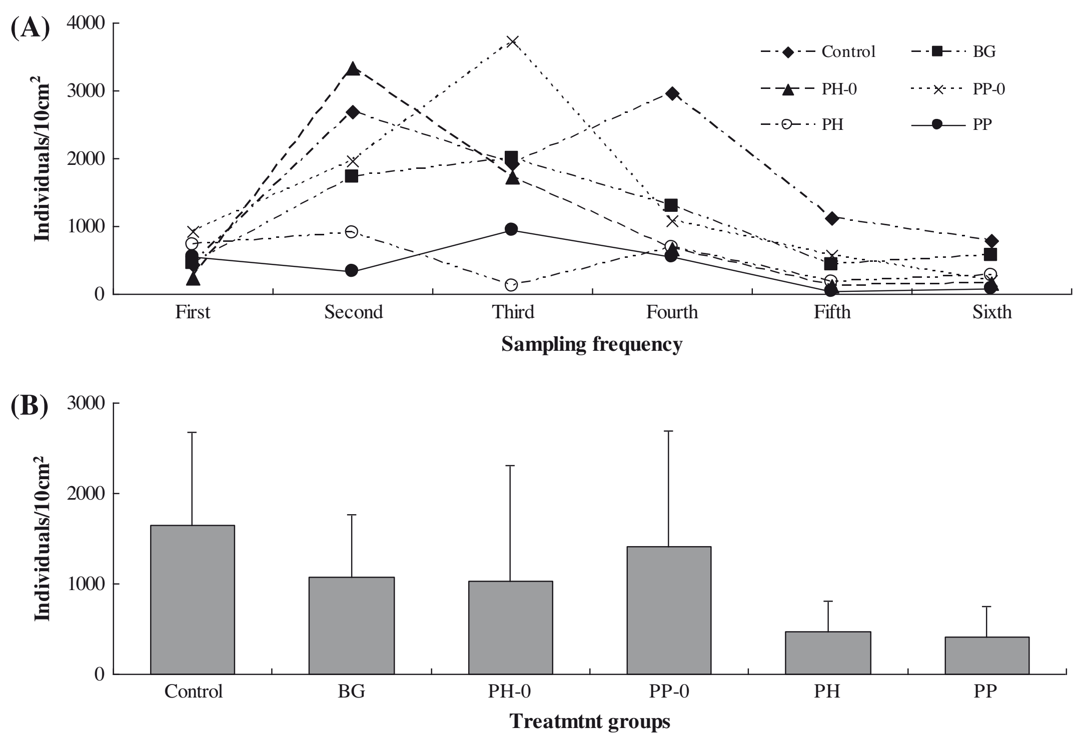

Fig. 4. Variations of total (A) and mean abundances (B) of meiofauna collected in each group of microcosm during the experiment.

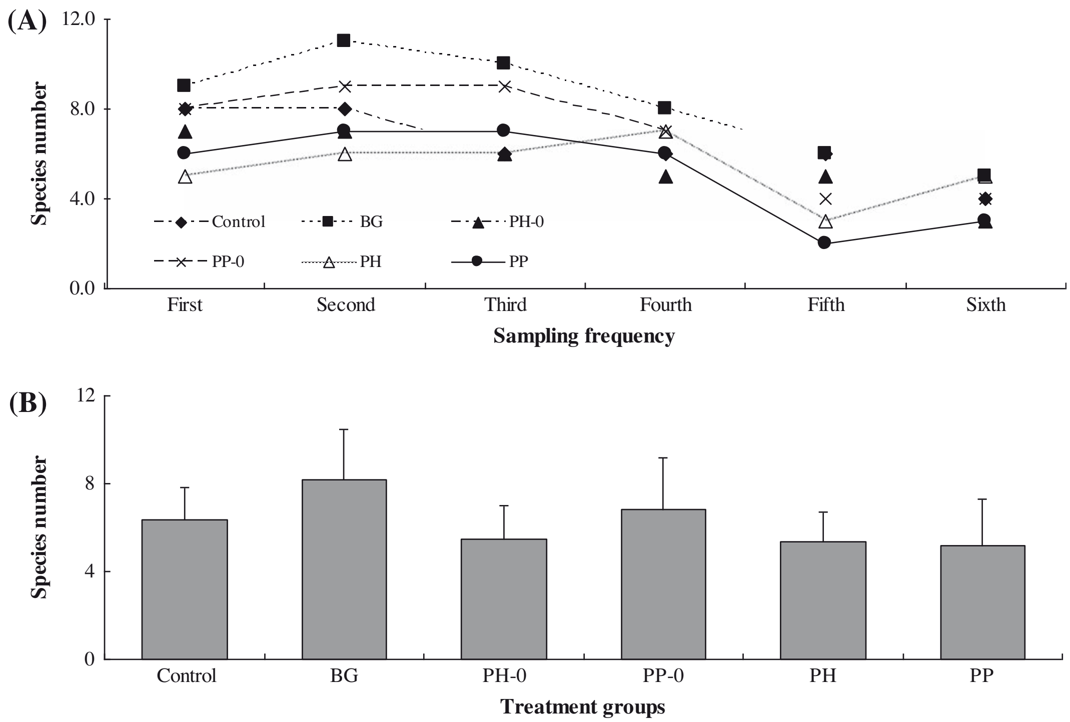

Fig. 5. Number of meiofauna species by sampling frequencies and at each treatment/control group.

study may be related to water circulation, periodical brine stress as well as habitat preference.

Differences in community indices between control and treatment groups were not clear and comparable to BG level during this study (Fig. 7). Even with the clear changes of species composition between treatment groups overall community structures were similar between groups, even comparable to BG level. For the relatively closed environments such as lagoons and bays, nematode assemblages may be similar to those of estuarine habitats, structuring along salinity gradients, with diversity and abundance tending to reduce with decreasing salinity and increasing salinity ranges [28,29]. However, absolute tolerance of individual populations is likely to relate to the degree and duration of deviation from the average salinity to which the population is accustomed and the rate of change in salinity, factors which differ between closed lagoons and open estuaries [30]. These contemporary conditions may influence species fecundity, settlement, behavioural irritation, predation, and therefore relative abundances, while historic condition may influence species occurrence through the effects of recruitment and survivals [9,31].

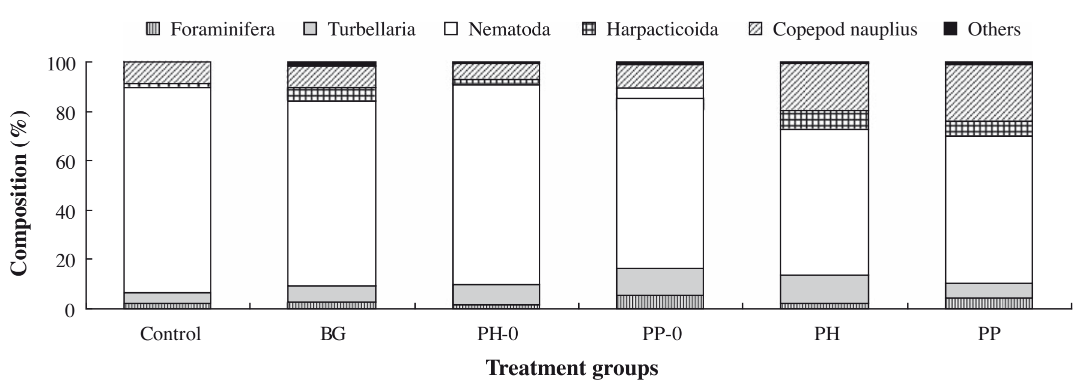

Fig. 6. Composition of meiofauna taxa at each treatment and control group. Values are means of four replicates at each group.

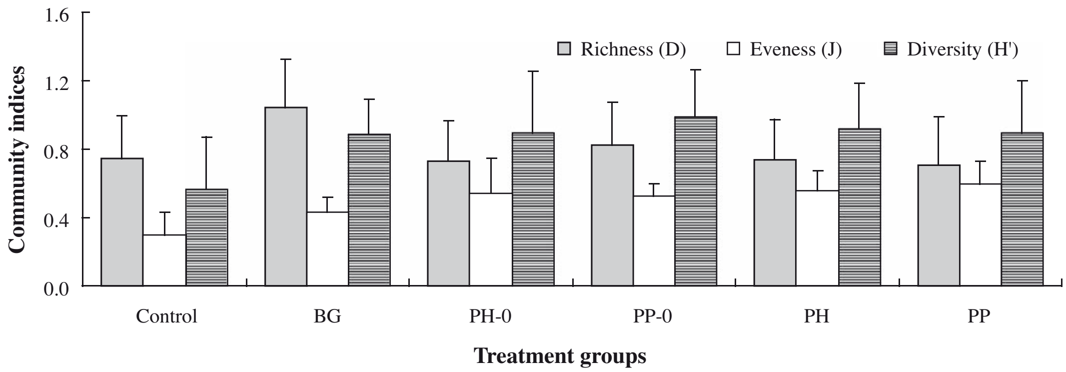

Fig. 7. Comparison of meiofauna community indices between control/BG and treatment groups. Values are means of four replicates for each treatment group.

Table 3
Abundances and species numbers of macrofauna at each treatment. Numbers in parentheses are species composition (%)

	Macrofauna abundance at each treatment group (individuals/0.12 m^2)					
	Control	BG	PH-0	PP-0	PH	PP
Polychaeta						
Neanthes japonica	1 (10.0)	1 (5.3)	2 (16.7)	1 (1.8)	–	–
Nephthys caeca	–	–	1 (8.3)	–	–	–
Brachyura						
M. japonicus	3 (30.0)	4 (21.1)	5 (41.7)	–	–	–
Macrura						
E. carinicauda	–	–	1 (8.3)	10 (17.9)	1 (6.3)	4 (33.3)
Bivalvia						
G. primeana	5 (50.0)	10 (52.6)	1 (8.3)	43 (76.8)	2 (12.5)	–
Cyclina sinensis		3 (15.8)				
Gastropoda						
Lunatia gilva	–	1 (5.3)	–	–	–	–
Fish						
A. flavimanus	1 (10.0)	–	2 (16.7)	2 (3.6)	13 (81.3)	8 (66.7)
Total	10	19	12	56	16	12
Species number	4	5	6	3	3	2

7. Changes of macrofauna assemblage

A total of 125 macrofauna individuals were collected and bivalvia and polychaeta were the most abundant taxa followed by brachyura and macrura. Other taxa were gastropoda and a fry of benthic fish (Table 3). Bivalvia were the most abundant representing 51.2% of the total population followed by a fry of benthic fish 20.8%, macrura 12.8%, brachyuran 9.6%, and polychaeta 4.8%, respectively. Bivalvia were also the most abundant among the major taxonomic groups representing 39.0 ± 35.3% of the total population followed by a fry of benthic fish 30.8 ± 34.6%, brachyura 18.7 ± 23.0%, and macrura 11.5 ± 12.8% (Fig. 8A). The most dominant species was *Glauconome primeana* (Bivalvia) and composed 11.1–78.2%. This species was mostly collected at control, BG, PP-0, PH-0, and PH groups. PP treatment, limited water circulation and short-term brine exposure, was estimated by lower compositions than those of other treatments. Also, *Macrophthalmus japonicus* of macrura represented high composition (ranged from 28.6 to 55.6%) at control, BG, and PH-0 treatments and this species were not found at PH-0, PH, and PP treatments (Fig. 8B). Individual composition for this species was quite similar to a phase of *G. primeana* in each treatment. However, reversely, the composition of *Acanthogobius*

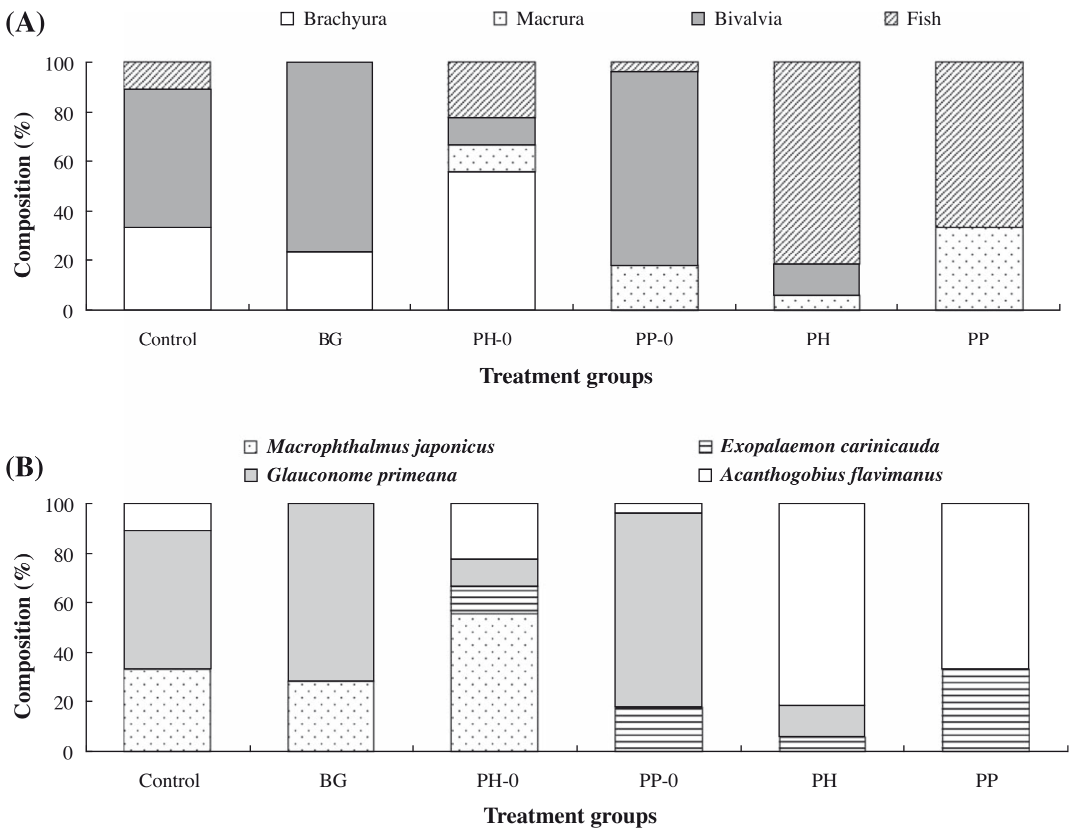

Fig. 8. Composition of dominant macrofauna (A) and dominant species composition (B) at each treatment group. Values are means of four replicates at each treatment group and control/BG.

flavimanus (a fry of benthic fish) was estimated by high values at PH and PP treatments (at stressed treatment groups from limited water circulation and short-term brine exposure). Species contribution in those treatments was estimated by 83.3 and 66.7%, respectively, against control groups (control and BG). The composition of *Exopalaemon carinicauda* (macrura) was similar to the distribution pattern of fish in treatment groups. In this study, distribution of macrofauna was obviously distinguished with ecological characteristic. Brachyura *M. japonicus* and bivalvia *G. primeana*, both burrowing species, were mainly found at control and BG which maintained full seawater circulation and nonbrine exposure during the experiment. However, macrura *E. carinicauda* and fry of benthic fish *A. flavimanus* having high swimming ability were dominated at treated groups which were partially limited in water circulation and periodic short-term brine exposure.

Some studies reported that distribution of echinoderm had disappeared from the meadow in front of the desalination plant and southern locality currently affected from brine, however, important increment was observed at the northern locality, which compared with dispersion of the brine since the northern locality is the less affected by the brine. Because live species of echinoderm are limited on the surface of bottom environment, their results may be focused on long-term brine exposure and flow direction of current using several target species without consideration of sediment condition [32]. However, in our results eco-physiological mechanism of burrow species may be influenced by strength of water flow and periodic brine exposure because benthic organisms may have strong tolerance of sudden change of high salinity concentration during a short-term period. Reversely, active species collected in stressed conditions can keep on normal feeding activity and eco-physiological metabolisms because they have avoidance capacity from serious environmental change using swimming organs. The current microcosm experiment revealed that the selected species in each treatment were differently affected by the disposed mud sediment, which could be attributed to the behavior and physiology of the buried organism [33]. In some cases, large and highly motile polychaetes showed the highest tolerance to sediment disposal

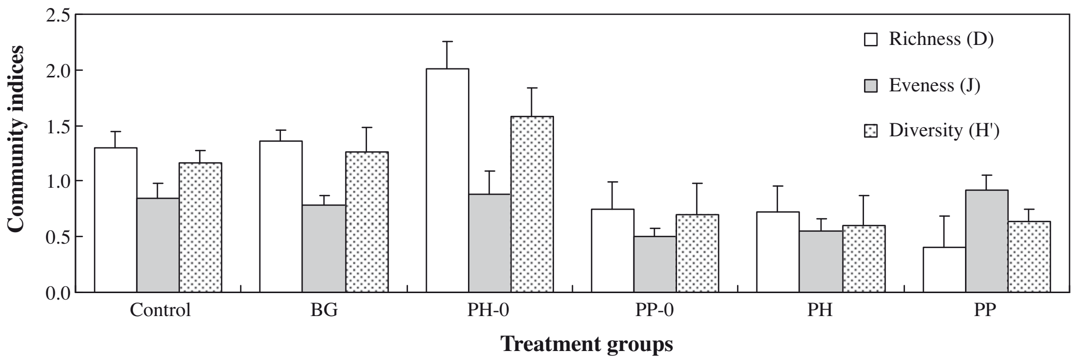

Fig. 9. Comparison of macrofauna community indices between control and treatment groups.

[34,35] and it suggested that motile polychaetes might be more tolerant to sediment dumping than mollusks. However, in this study, recruitment of macrofauna may be influenced by the strength of water circulation and periodic brine exposure rather than sediment disposal, although sediment deposition was observed within in microcosms. Differences of species recruitment at treated groups were related to sediment conditions. For macrofauna, burrow species was mostly collected at control, BG and PH-0 treatments which were not stressed by periodic brine exposure and measured low value of COD and TS concentration in sediment than others. However, recruitment of macrura and benthic fish, which may utilize surface sediment for feeding duration of full seawater condition, dominated in treated microcosms such as PP-0, PH, and PP microcosms. These relationships were clearly observed at differences in community indices (Fig. 9). The three community indices showed low values at treated groups which were limited in water circulation and periodic short-term brine exposure. Additionally, COD and TS concentrations were measured lower than control condition. These organisms may prefer increment of bottom sediment that occurred continuously in a deposited environment from water body to surface sediment because water circulations in these treatments were relatively stable condition than control groups (control and BG).

Microcosm experiments can contribute to providing rapid quantified evidences of expected ecological impacts associated with operation of desalination plant. Several researchers suggested that microcosm experiments could be utilized to achieve a balance between impacts prediction of related various environmental events and maintaining legal, time, and cost obligations [8]. Following this study, demonstration of the quantitative critical value of output parameters such as species number, individuals per certain area, and biodiversity caused by periodic brine exposure and change of water circulation is not enough. However, it may be able to refer to the levels of critical point that severely damage the marine environment benthic organism assemblages through the additional microcosm studies could be considered to be priority research objective including brine exposure and physical alternation of study area.

8. Summary and conclusions

One approach to investigate the expected impacts of water circulation and periodic short-term brine exposure on benthic organism assemblages is by utilizing microcosm experiments. The alternations of COD and TS concentration were distinguished between control groups (control and BG) and treated groups (limited seawater circulation and periodic brine exposure) over the experiment and the cause of differences may be from the increment of organism matter, anaerobic condition, and short-term brine stress in bottom sediment caused by lack of water circulation within microcosms. High abundance and species number were observed at control groups (control and BG) and low values at treated groups for meiofaunal community. However, evenness and species diversity indices at treated groups were slightly higher than those of the control groups.

For macrofauna, burrowing species were mostly found at control groups but most recruitments of macrura and benthic fish were observed at treated groups such as PP-0, PH and PP treatments. And these relationships were clearly observed at difference in community indices. The three community indices showed lower values at treated groups. As results of this study, some burrowing species may prefer increment of bottom sediment that occurred continuously in a deposited environment from water body to surface sediment because water circulations in these treatments were relatively stable condition than control groups of microcosms. For both of benthic meio-and macrofaunal community, there was no significant relationship between brine exposure and community indices but slightly decreasing trend in terms of the number of species and abundance at treatment groups.

In conclusion, microcosm experiments can contribute to providing rapid quantified evidences of expected ecological impacts associated with operation of desalination plants. It can provide us with what possible influential factors will produce a positive or negative reaction of our subject areas of interest within short time. Manipulating something can give us an idea as to what to expect if something were to occur in that ecosystem or environment. However, if microcosm (or mesocosm) is not adequately imitating the environment, the organisms can avoid giving off a certain reaction vs. its natural behavior in its original environment.

Acknowledgements

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