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Evaluation of transparent exopolymer particles and microbial communities found post-UV light, multimedia and cartridge filtration pre-treatment in a SWRO plant

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ABSTRACT

Seawater reverse osmosis desalination is affected greatly by membrane biofouling which reduces membrane lifetimes and increases cost of permeate production. This work reports on the analysis of pre-treated seawater from a small-scale desalination plant operating with a three-stage pre-treatment system namely, (1) medium-pressure ultraviolet (MP-UV) disinfection, (2) multimedia filtration (MMF), and then finally, (3) cartridge filtration. Transparent exopolymeric particles (TEPs), chlorophyll *a*, phytoplankton, bacteria and viruses were evaluated in the pre-treated seawater after each pre-treatment stage over a one-year period (July 2012–July 2013). The concentration of TEPs was found to occasionally increase after MP-UV disinfection. MP-UV disinfection had no effect on the phytoplankton, bacterial or viral cell counts. In contrast, MMF was shown to be the most efficient step in removing TEPs and micro-organisms from seawater, while this removal was less significant for viruses. Cartridge filters had limited efficiency. Phytoplankton was observed to be more efficiently removed compared to bacteria. Although phytoplankton removal rates varied over time and were dependent upon cell size and shape, most of the micro-organisms were removed from seawater throughout the period of study. Investigating the seawater pre-treatment system during different season provided, thus, useful insights on its efficiency.

Keywords: Desalination; TEP; Pre-treatment; Biofilm; Microalgae; Micro-organisms; Fouling

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1. Introduction

Seawater reverse osmosis (SWRO) plants, as well as other water treatment systems based on reverse osmosis (RO), face periodic clogging by chemical precipitates (i.e. scaling) or live biofilms (i.e. biofouling) which decrease the permeate flux. While scaling mainly affects brackish water and can be efficiently minimized by pH adjustments and other chemical treatments, efficient strategies to control biofouling are still under debate [1,2]. The performance of RO membranes and their life duration in SWRO plants are highly affected by the conditions of the RO feed water. Specifically, plankton cells and organic compounds which have passed through the pre-treatment systems of the raw seawater prior to the RO feed tank can bind to the RO membrane causing biofouling [3,4].

The succession of microbial communities on RO membranes is dependent on a wide range of physical, chemical and biological conditions occurring in the RO feed water, including temperature, pH, nutrients, particulate and dissolved organic matter [5,6]. Furthermore, biofilm growth is influenced by the dynamics of the microbial communities present in the RO feedwater. These conditions can either promote or inhibit further biofilm growth on the RO membrane, thus, affecting membrane biofouling. It is clear that biofouling consists of the accumulation of active and senescent micro-organisms along with organic compounds including transparent exopolymeric particles (TEPs). TEPs are formed by abiotic coagulation of phytoplankton, and to a lesser extent by bacterial exudates [7,8]. Once formed, TEPs are known to form a sticky matrix which promotes cell adhesion [9] as well as biofilm formation [3] on the RO membranes. Since TEP concentration, as well as the composition of microbial communities, varies both locally and seasonally it is, therefore, of utmost importance to quantify these parameters within a SWRO pre-treatment system prior to the RO feed tank in order to better understand the biofouling potential of seawater.

Typical pre-treatment strategies include coagulation and/or flocculation, biocide spiking, ultraviolet (UV) irradiation and filtration [2]. The combination of biocides and/or medium-pressure ultraviolet (MP-UV) disinfection (400–190 nm) followed by filtration has been shown to remove cells, viruses, debris and sediment from seawater [10,11]. However, biocide suitability is under question because of its efficiency and safety. Often biocides can exacerbate biofouling by causing some bacteria to produce exudates in order to protect themselves from biocidal attack [12]. Furthermore, biocides have been shown to damage RO membranes [13,14] and degrade into toxic by-products [15]. A main disadvantage of UV light and biocide treatments is that large amounts of particles and dead cells, which still remain in the seawater that can potentially contribute to biofouling by either making up the biofilm matrix or providing living micro-organisms with fresh substrates for growth. Thus, one or more filtration steps are necessary to decrease the particle content in the treated seawater. Such steps typically employ multimedia filtration (MMF) and cartridge filtration (CF), the former typically consisting of a combination of sand and gravel layers. This leads to a minimal pore size of approximately 1 μm, whereas cartridge filters have a pore size of 5–20 μm [16]. Conventional pre-treatment systems are designed to remove suspended particles >0.5 μm although a low efficiency has been reported for particles <15 μm [17]. However, improved pre-treatment ultrafiltration (pore size <0.02 μm), for retaining both colloids and suspended particles [17], and nanofiltration (molecular weights >200 daltons), for retaining solutes [2], can also be used.

In the present study, we monitored the concentration of TEPs, phytoplankton, bacteria and viruses after each pre-treatment stage, namely MP-UV disinfection, MMF and CF, in a small-scale SWRO plant. The study took place over a one year period, from July 2012 to July 2013, to monitor seasonal variations that may arise from changes in raw seawater input (temporal dynamics) into the pre-treatment process. For the study, the fully operational Penneshaw SWRO plant on Kangaroo Island, South Australia, was selected due to its small size and simple configuration along with the lack of biocide and coagulant applications.

2. Materials and methods

2.1. Seawater pre-treatment system prior to SWRO

The Penneshaw SWRO desalination plant has a capacity of 3×10^5 L d^{-1} and has been described in detail in previous studies [18,19]. Seawater from a depth of 6 m is pumped from the coastal waters north of Kangaroo Island at a site located 190 m from the Penneshaw desalination plant. Seawater enters the system (Fig. 1) through two pre-filtration screens (10 cm and 0.5 mm pore sizes, respectively). This is then followed by the pre-treatment system which includes an MP-UV disinfection unit, four parallel multimedia filters (gravel, garnet, sand and coal with grain size ranging from 0.3 to 10 mm), and two consecutive sets of three cartridge filters each with a pore size of 15 and 5 μm, respectively. The flow rate through the system is typically 8.4 L s^{-1} after which the seawater enters the RO feed tank (Fig. 1). Seawater

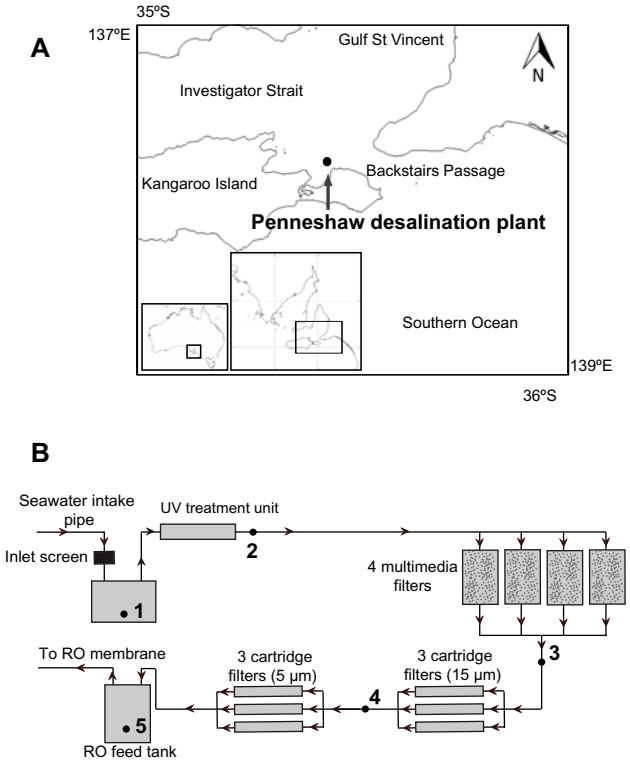

Fig. 1. (A) Location of Penneshaw desalination plant and (B) schematic of the seawater pretreatment prior to RO. Numbers indicate the five different sampling sites (Sites 1–5).

was collected from five sampling sites within the seawater pre-treatment system, every two weeks between July 2012 and July 2013. Site 1 was located prior to MP-UV treatment (raw seawater), Site 2 post-MP-UV treatment, Site 3 post-MMF, Site 4 post-15 µm CF and Site 5 within the RO water feed tank located post-5 µm CFs (Fig. 1). For the measurement of selected parameters seawater was collected at each sampling site (Fig. 1) using polyethylene (PET) bottles. The PET bottles were then stored on ice and transported to the laboratory for immediate processing.

2.2. Transparent exopolymeric particle evaluation

TEP were measured every four weeks from November 2012 to March 2013 and then every 2 weeks thereafter (Table 1). Seawater (1 L) was filtered in triplicate through 47 mm isopore membrane filters (0.4 µm pore size, Millipore, Kilsyth, Australia) using a vacuum pump and a filtration ramp. The filters were then placed in 0.4 µm filtered seawater (5 mL) and stored at −20˚C, as described by Klein et al. [20]. Determination of the concentration of TEP was carried out following published methods [21,22]. Samples were thawed and the material on the filters was resuspended followed by centrifugation at 3,200 × *g* for 30 min, the filters were then discarded. Samples were recentrifuged as above, the supernatant removed and 0.02% w/v alcian blue (2 mL, Sigma–Aldrich, Castle Hill, Australia) in 0.06% v/v acetic acid (Merck Millipore, Kilsyth, Australia) added to the pellet which was resuspended as above. Samples were immediately centrifuged, the supernatant removed, and the pellet washed twice with distilled water (2 mL) to remove excess alcian blue. Subsequently, 80% v/v sulfuric acid (4 mL, Merck Millipore, Kilsyth, Australia) was added to the pellet and the pellet resuspended (using a vortex mixer). The solutions were incubated for 2 h at room temperature, resuspended and centrifuged at 3,200 × *g* for 20 min. The absorption of the supernatant (0.3 mL) was measured at 787 nm using a FLUOstar Omega fluorometer (BMG Labtech, Mornington, Australia). TEP values were calibrated using a standard weight of xanthan gum (Sigma–Aldrich, Castle Hill, Australia) ranging from 10 to 800 µg suspended in ethanol [22]. TEP values of relative fluorescence were thus converted to µg equivalent of xanthan gum L^{-1} (µg Xeq. L^{-1}).

2.3. Chlorophyll-a evaluation

Chlorophyll-*a* (Chl-*a*) evaluation is used as an estimator of phytoplankton biomass. The concentration of Chl-*a* was measured every four weeks (Table 1) using methanol extraction and subsequent fluorometric determination [23]. Seawater (0.5 L) was filtered in triplicate through 47 mm, glass microfiber filters (1 µm pore size, Filtech, Fairy Meadow, Australia), using a vacuum pump and a filtration ramp. The filters were then wrapped in aluminium foil and stored at −20˚C. For analysis the filters were placed in methanol (5 mL) for 24 h at 4˚C in the dark and the concentration of Chl-*a* dissolved in the methanol was determined using a Turner 450 fluorometer previously calibrated with Chl-*a* extracted from *Anacystis nidulans* (Sigma Chemicals, St Louis, MO, USA).

2.4. Evaluation of phytoplankton

For the enumeration of phytoplankton cells > µm, seawater (1 L) was collected in PET bottles and preserved with acidic lugol solution (5 mL, 0.6 M potassium iodide, 0.4 M iodine and 1.5 M acetic acid). Identification and enumeration of phytoplankton was carried out every 2 weeks by Microalgal Supply Service (Ormond, Victoria). The cells were identified to the genus or species level based on their key

Table 1
List of the parameters measured at each sampling date

Sampling date	TEP	Chl-*a*	Phytoplankton	FCM populations
4/07/12			yes	
18/07/12		yes	yes	yes
1/08/13			yes	yes
15/08/12		yes	yes	yes
29/08/13			yes	
12/09/12		yes	yes	yes
26/09/12			yes	yes
10/10/12		yes	yes	yes
24/10/12			yes	yes
7/11/12		yes	yes	yes
21/11/12	yes	yes	yes	yes
5/12/12			yes	yes
19/12/12		yes	yes	yes
16/01/13	yes	yes	yes	yes
30/01/13			yes	yes
13/02/13	yes	yes	yes	yes
27/02/13			yes	yes
13/03/13	yes	yes	yes	yes
27/03/13	yes		yes	yes
10/04/13	yes	yes	yes	yes
24/04/13	yes		yes	yes
8/05/13	yes	yes	yes	yes
22/05/13	yes		yes	yes
4/06/13	yes	yes	yes	yes
19/06/13	yes	yes	yes	yes
3/07/13	yes	yes	yes	yes
17/07/13	yes	yes	yes	yes
31/07/13	yes	yes	yes	yes

taxonomic features [24,25] and grouped according to their size and shape.

2.5. Evaluation of bacteria and viral abundances

Bacterial and viral counts were carried out using flow cytometry [26]. From each sampling site, triplicates of seawater (1 mL) were fixed with 0.5% gluteraldehyde (Proscitech, Thuringowa, Australia) and the samples flash frozen in liquid nitrogen and stored at −80˚C until further analysis. The samples were diluted 1:10 in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8; National Diagnostics, Atlanta, USA) and the DNA in the cells stained with 2.5% (w/v) SYBR I Green (Invitrogen, Carlsbad, USA). The samples were incubated at 80 C for 10 min and fluorescent marker beads (1 μL, Molecular probes, Eugene, USA) were added afterwards to all samples as an internal size and concentration standard prior to analyses which were performed using a FacsCanto instrument (Becton Dickinson, San José, USA). For each sample forward-angle light scatter, side-angle light scatter (SSC) and SYBR I Green (530 nm) fluorescence were recorded and total bacterial and viral count were then discriminated based on SSC and SYBR I green fluorescence, as described previously [26]. After each flow cytometry analysis the concentration of marker beads was estimated by epifluorescence microscopy and used to normalize the abundance of all microbial populations.

2.6. Statistical analyses

To investigate whether the changes in TEPs, Chl-*a*, phytoplankton, bacteria and viruses, observed after each seawater pre-treatment steps were significant we performed a one-way ANOVA using R. For each parameters comparisons were carried out between values measured before and after each pre-treatment step using a fitting linear model.

3. Results and discussion

3.1. Raw seawater properties

To evaluate the efficiency of Penneshaw seawater pre-treatment system we compared the concentration

of TEPs, viruses and microorganisms in raw seawater with those measured after different pre-treatment steps. The temperature of raw seawater followed typical seasonal trends of temperate latitudes, with lowest temperatures (~14˚C) occurring from mid-winter (July 2012) to early spring (October 2012), and highest values (~20˚C) from mid-summer (January 2013) to mid-autumn (May 2013). The salinity of the Gulf St Vincent, typically varies seasonally from 36 to 42 practical salinity units. These salinity values are similar to the salinity levels encountered in the Arabian Gulf [27].

The concentration of TEPs was measured from spring 2012 until winter 2013 and was found to vary over 2 orders of magnitude. This ranged from concentrations below the detection limit of 2–360 ± 120 μg Xeq. L^{-1}. The lowest concentrations were generally recorded in summer and the highest in autumn (Fig. 2). The highest concentrations found here for TEPs are lower than those typically found in other coastal environments [20,28–31].

The concentration of Chl-*a* was always <0.12 μg L^{-1}, except during early June 2013 where it reached 0.32 μg L^{-1} (Fig. 3). This peak was related to high phytoplankton abundances measured on that occasion (Fig. 4). We measured phytoplankton abundances higher than those found in early June in 4 occasions, when unfortunately we did not measure Chl-*a* concentration. Phytoplankton abundance varied between 1.7×10^4 and 3.5×10^5 cell L^{-1} (Fig. 4), their sizes ranged from 5 to 100 μm, and included round, conic as well as elongated cells (Table 2). Overall phytoplankton abundance peaked (>2×10^5 cell L^{-1}) once in spring 2012 and twice in autumn 2013 whereas the lowest abundance values were observed during summer 2013. Chl-*a* concentrations and phytoplankton abundances during the year were significantly lower compared to other environments where desalination plants operate, such as the Arabian Gulf [32], the Mediterranean Sea off the coasts of Spain [33,34] and Israel [35]. These low concentrations of TEPs and phytoplankton suggest that the seawater off Penneshaw might require a less rigorous series of pre-treatment processes, especially in terms of filter pore sizes compared to other desalination sites.

Similarly to phytoplankton, both bacterial and viral abundances peaked in autumn 2013, although the three populations did not always reach high abundances on the same dates. The bacterial abundance varied between 2×10^5 and 1.2×10^6 cell mL^{-1} (Fig. 5) during the period of study, whereas the abundance of viruses ranged from 6.5×10^5 to 1.6×10^7 mL^{-1} (Fig. 6). These abundances are similar to those occurring in the Mediterranean Sea [36,37], which is expected as the abundance values of both bacteria and viruses are known to be comparable in coastal seawater worldwide [38,39].

3.2. Post-medium pressure-ultra violet

No significant changes in the TEP concentration or microbial count were observed after post-MP-UV (Table 3). The concentration of TEPs after MP-UV ranged from values below the detection limit to 220 ± 85 μg Xeq. L^{-1} (Fig. 2). TEPs at this sampling

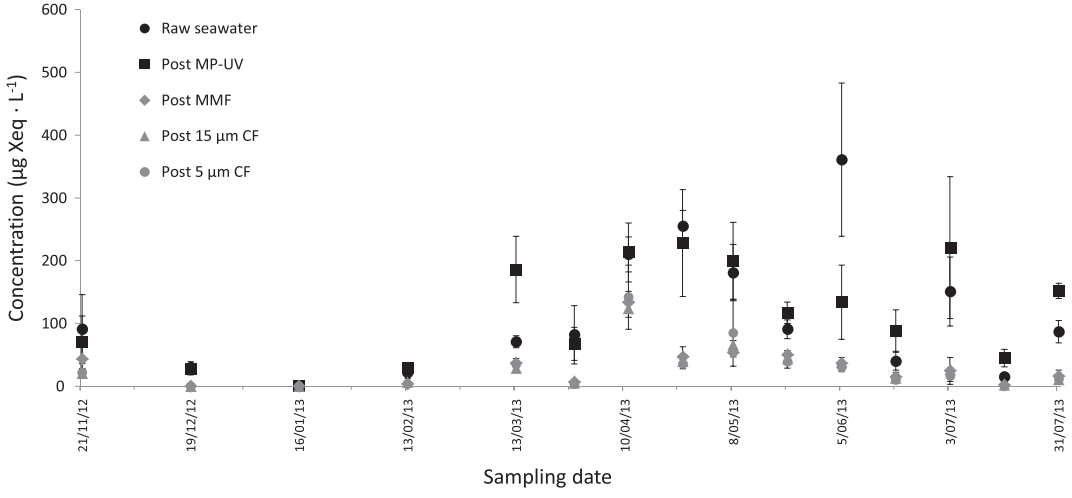

Fig. 2. Concentration of TEPs within Penneshaw pre-treatment plant from July 2013 to July 2013. TEPs were measured from November 2012 to July 2013 and their concentration was expressed as μg of xanthan gum equivalent per litre. Error bars correspond to the standard deviation. Abbreviations: medium-pressure ultra violet (MP-UV); multi-media filters; cartridge filters.

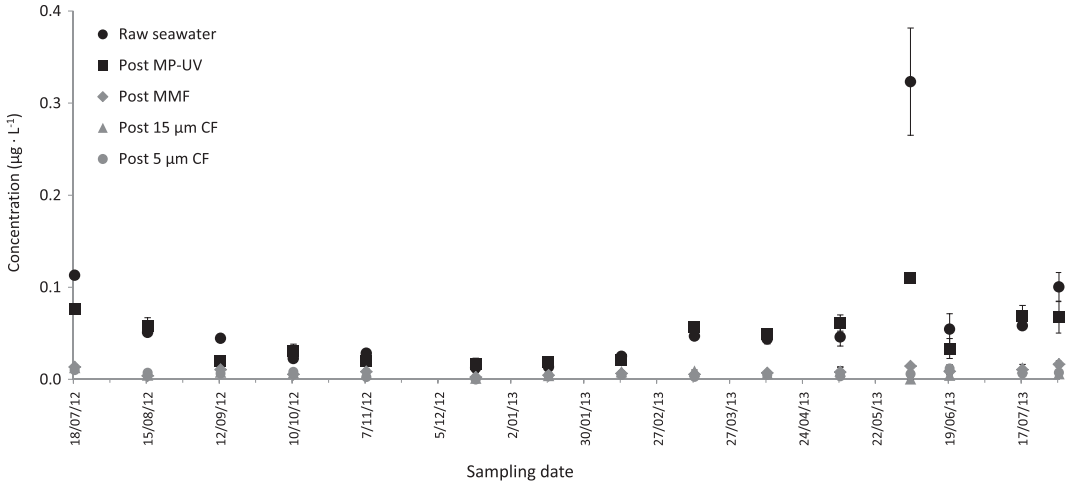

Fig. 3. Seasonal concentration of Chlorophyll-*a* within the Penneshaw pre-treatment plant. Abbreviations: medium-pressure ultra violet (MP-UV); multi-media filters; cartridge filters.

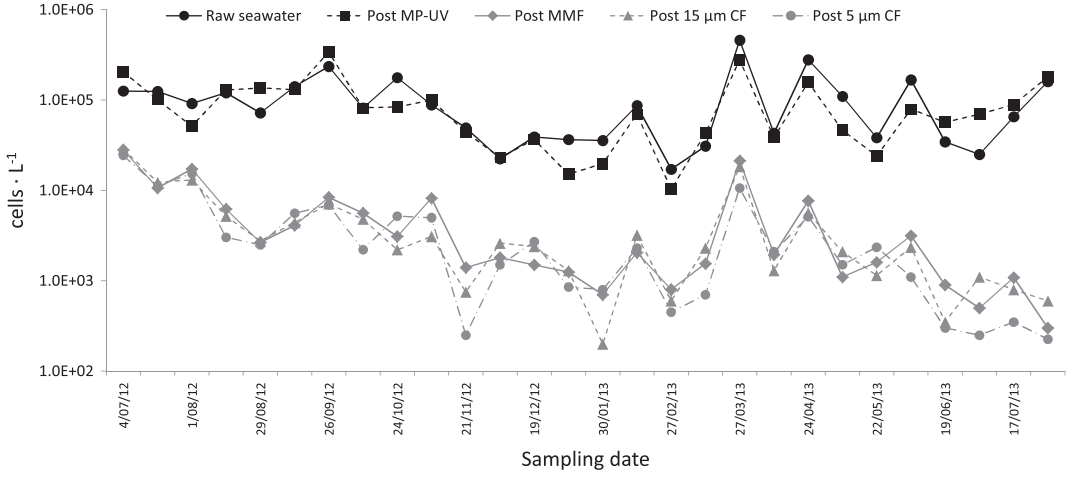

Fig. 4. Seasonal abundance of total phytoplankton within the Penneshaw pre-treatment plant.
Note: Values are on a logarithmic scale. Abbreviations: medium-pressure ultra violet (MP-UV); multi-media filters; cartridge filters.

site (Site 2, Fig. 2) did not always follow the seasonal trend observed in raw seawater. On four different occasions the TEP concentrations were found to be at least twice as high as those measured in raw seawater, whereas in early June 2013 their concentration post-MP-UV was lower than 50% compared to that measured in raw seawater. This irregular trend suggests an unpredictable effect of MP-UV irradiation on TEPs, and this is likely due to the heterogeneous nature of these substances [9,40]. In a previous study, an increase in particulate organic carbon (POC) was observed for a seawater mesocosm exposed to UV irradiation [41]. The authors suggested that the increase in POC observed was likely due to exudate release and that some of these exudates can promote aggregation into TEPs [41]. However the impact of MP-UV in promoting the release of cell exudates is not fully clear and the variations in TEPs observed here might be independent from the MP-UV treatment and simply associated with the water flow at high pressure, more observations are required to clarify this effect.

The trends in Chl-*a* concentrations measured post-MP-UV followed those observed for the raw seawater, except in early June 2013 when the concentration of Chl-*a* post-MP-UV decreased by 66% (Fig. 3). This drop is likely due to UV damage on Chl-*a* when high Chl-*a* values are measured [42,43]. Previous studies on

Table 2
List of flow cytometry populations and phytoplankton species most abundant at Site 1 (raw seawater, average $>1 \times 10^3$ cell L^{-1}) and Site 5 (RO feed tank, average >10 cell L^{-1}) during the period of study

Species	Taxon	Shape[a]	Size or width[b]		Mean[c]		Removal rate		
			Min	Max	Raw seawater	RO feed tank	Min	Max	Total[d]
Pyramimonas spp.	Chlorophyta	conic	4	10	7.9E + 03	5.8E+ + 02	4	100	93
cf. *Gymnodinium*	Dinoflagellate	prolate spheroid	7	20	1.2E + 04	5.0E + 02	83	100	96
Hemiselmis sp.	Cryptophyta	prolate spheroid	3	7	1.3E + 04	4.2E + 02	76	100	97
Cylindrotheca closterium	Diatom	elongated	2	5	7.4E + 03	3.0E + 02	0	100	96
Plagioselmis prolonga	Cryptophyta	cone + half sphere	3	5	5.8E + 03	2.7E + 02	80	100	95
Pseudo-nitzschia delicatissima group	Diatom	elongated	10	15	2.0E + 03	2.4E + 02	25	100	88
cf. *Navicula*	Diatom	elongated	5	10	5.1E + 03	2.3E + 02	60	100	96
Chrysochromulina spp.	Haptophyta	prolate spheroid	2	5	2.8E + 03	2.2E + 02	0	100	92
Heterocapsa rotundata	Dinoflagellate	2 cones	5	15	5.8E + 03	1.9E + 02	73	100	97
Tetraselmis spp.	Chlorophyta	prolate spheroid	8	15	1.4E + 03	1.6E + 02	82	100	89
Nitzschia spp.	Diatom	elongated	2	10	3.2E + 03	1.3E + 02	88	100	96
Teleaulax acuta	Cryptophyta	cone + half sphere	5	15	3.8E + 03	1.1E + 02	70	100	97
Gyrodinium spp.	Dinoflagellate	prolate spheroid	10	50	1.9E + 03	1.0E + 02	82	100	95
Leucocryptos sp.	Cryptophyta	cone + half sphere	8	15	1.1E + 03	8.0E + 01	0	100	92
Eutreptiella spp.	Euglenophyta	cylinder + cone	5	10	6.3E + 02	6.2E + 01	60	100	90
Cyclotella spp.	Diatom	cylinder	1	3	1.5E + 02	5.3E + 01	0	100	65
Leptocylindrus danicus	Diatom	cylinder	5	10	1.3E + 03	4.8E + 01	93	100	96
Unidentified heterotrophic flagellates	unknown	prolate spheroid	2	5	6.6E + 02	4.6E + 01	75	100	93
Fragilariopsis sp.	Diatom	elliptic prism	4	10	2.6E + 02	4.2E + 01	0	100	84
Pleurosigma sp.	Diatom	elongated	5	10	4.7E + 02	3.8E + 01	64	100	92
Entomoneis sp.	Diatom	elliptic prism	7	15	3.4E + 02	2.8E + 01	78	100	92
Cocconeis spp.	Diatom	elliptic prism	10	20	2.4E + 03	2.7E + 01	97	100	99
Gyrosigma spp.	Diatom	elongated	5	15	1.3E + 02	2.4E + 01	75	100	81
Nephroselmis sp.	Chlorophyta	sphere	2	5	2.9E + 02	2.1E + 01	83	100	93
Chaetoceros spp.	Diatom	elliptic prism	5	20	2.8E + 03	2.1E + 01	73	100	99
Fragilaria sp.	Diatom	elliptic prism	4	10	7.0E + 02	2.1E + 01	86	100	97
Unidentified bodonids	Euglenophyta	ellipsoid	4	10	4.1E + 02	2.0E + 01	90	100	95
Unidentified flagellates	unknown	unknown	2	5	4.3E + 02	2.0E + 01	75	100	95
Ochromonas spp.	Chrysophyceae	cone + half sphere	5	10	1.1E + 03	2.0E + 01	70	100	98
Emiliania huxleyi	Haptophyta	cylinder	5	10	2.8E + 02	1.8E + 01	85	100	94
Unidentified amoeba	unknown	unknown	3	10	9.8E + 01	1.2E + 01	80	100	88
Thalassiosira cf. *mala*	Diatom	cylinder	5	10	1.0E + 03	3.6E + 00	100	100	100
Total large phytoplankton					8.7E + 04	4.0E + 03	81	100	95
Picoeukaryotes	Unknown				4.2E + 03	5.9E + 02	18	100	86

(*Continued*)

Table 2 (*Continued*)

Species	Taxon	Shape[a]	Size or width[b]		Mean[c]		Removal rate		
			Min	Max	Raw seawater	RO feed tank	Min	Max	Total[d]
Synechococcus	Cyanobacteria				1.3E + 04	5.3E + 03	0	85	60
Prochlorococcus	Cyanobacteria				4.2E + 03	5.9E + 02	19	100	86
Total cyanobacteria	Cyanobacteria				1.7E + 04	5.9E + 03			66
Bacteria	na				4.6E + 05	2.1E + 05	19	81	53
VLP	na				2.0E + 06	1.1E + 06	0	98	43
TEP	na				107	29	0	100	73

[a]Shape inferred according to Hillebrand et al. [52].
[b]For non spherical cells the size of their lowest dimension is indicated.
[c]Data are shown as cell L^{-1} for phytoplankton, cell mL^{-1} for bacteria, number mL^{-1} for viruses and μg Xeq. L^{-1} for TEPs.
[d]Removal of cells or particles from seawater integrated over the period of study.

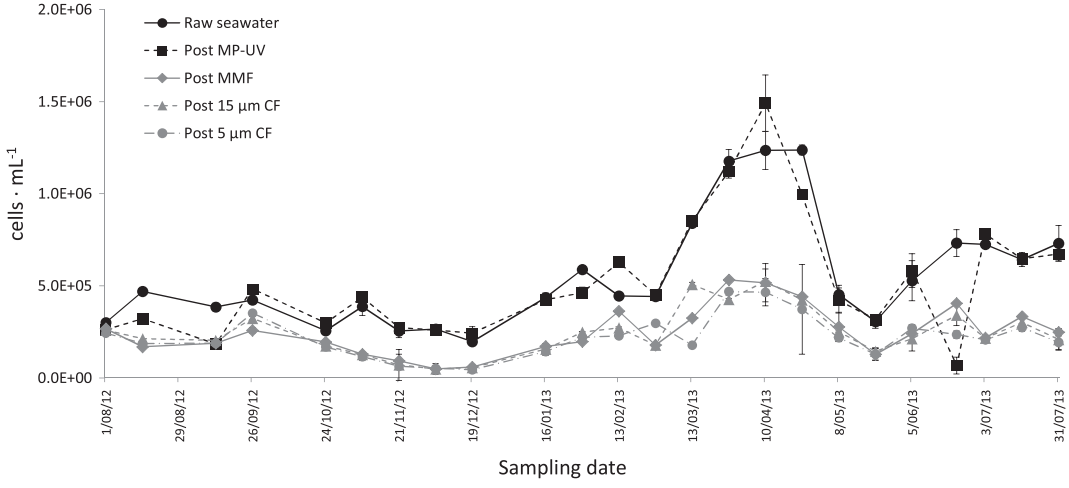

Fig. 5. Seasonal abundance of total bacteria within the Penneshaw pre-treatment plant. Abbreviations: medium-pressure ultra violet (MP-UV); multi-media filters; cartridge filters.

phytoplankton cultures do not highlight a clear decrease of Chl-*a* concentration after UV exposure [42,43]. The seasonal variations in the concentrations of phytoplankton, bacteria and viruses post-MP-UV were also similar to those observed in raw seawater (Figs. 4–6). The abundance values recorded were slightly different than raw seawater values although they were neither systematically lower nor higher. Even when microorganism abundances peaked (during blooming) the effect of MP-UV was still not clear. For example, the phytoplankton abundance measured post-MP-UV was higher than that of raw seawater during the spring 2013 maximum and lower during the autumn 2013 maxima (Fig. 4). Moreover cells were not completely inactivated after MP-UV since we successfully brought 12 phytoplankton strains to culture from the seawater collected after this pre-treatment step (data not shown). Beyond the ability of some phytoplankton cells collected after MP-UV to grow, we did not compare biological activities (i.e. proportion of active cells) before and after MP-UV treatment and therefore did not assess microorganism viability. Previous studies show a variable, species-specific, sensitivity to UV irradiation for both phytoplankton [42,43] and bacteria [44]. They report that after several hours of exposure to UV light a significant proportion of phytoplankton and bacterial cells were still able to grow. Since seawater flows within the Penneshaw pre-treatment system at a speed of 8.4 L s^{-1}, the exposure time of phytoplankton and bacteria to UV light might be insufficient to inactivate the majority of microorganisms.

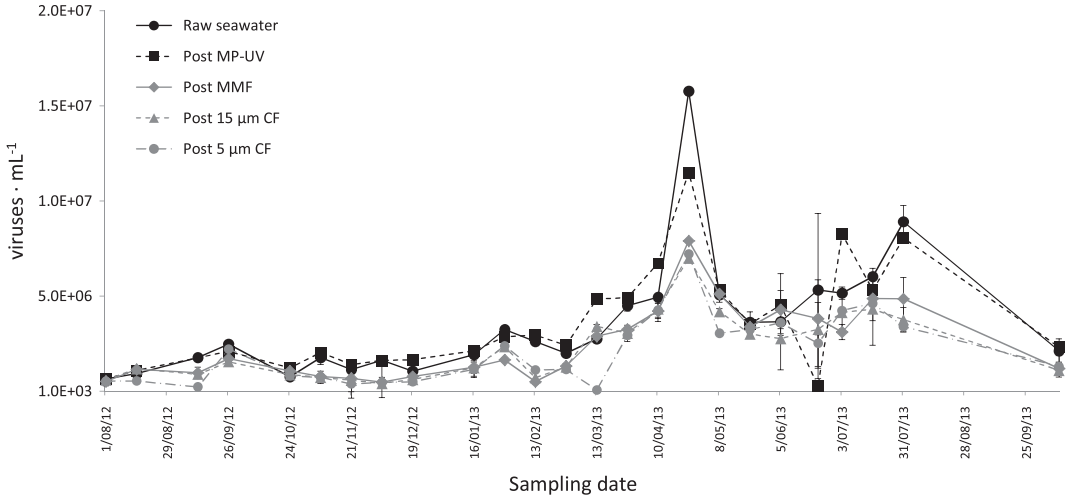

Fig. 6. Seasonal abundance of viruses (No mL^{-1}) within the Penneshaw pre-treatment plant. Abbreviations: medium-pressure ultra violet (MP-UV); multi-media filters; cartridge filters.

Table 3
Percentage variation of microorganisms and TEPs after each pre-treatment step for the period of study[a]

	MP-UV	MMF	15 µm CF	5 µm CF	ANOVA ($p < 0.01$)
TEP	20 ± 75	−82 ± 20	−10 ± 31	−11 ± 33	MMF ≠ 15 umCF
Chl-*a*	13 ± 35	−82 ± 21	−3 ± 40	−25 ± 62	MMF ≠ 15 umCF
Phytoplankton	−9 ± 53	−96 ± 6	−14 ± 49	−20 ± 68	MMF ≠ 15 mCF
Bacteria	2 ± 26	−56 ± 53	−4 ± 24	−9 ± 21	MMF ≠ 15 umCF
Viruses	9 ± 40	−37 ± 53	−10 ± 23	−4 ± 33	ns[b]

[a]July 2012–July 2013.
[b]Non significant.

Current data along with previous studies on the effect of UV light on marine microorganisms and organic matter suggest that MP-UV disinfection does not seem to be an effective strategy to reduce the biofouling potential of seawater. Moreover, UV irradiation has been shown to break humic acids into lower molecular weight compounds likely increasing the availability of labile substrates to biofouling microbes [45].

3.3. Post-multi-media filters

A significant drop in TEPs, phytoplankton and bacteria was observed after MMF (Table 3). The concentration of TEPs ranged from values below the detection limit to 130 ± 20 µg Xeq. L^{-1} and were always lower than those recorded post-MP-UV (Fig. 2). The highest concentration was measured in early April 2013 and was associated with high, although not the highest, values recorded for the previous sampling sites. The removal rate of TEPs from seawater by MMF varied over time. For example, low TEP decreases across the MMF (37–38%) were observed in late November 2012 and early April 2013, whereas there was a decrease of >60% for the other sampling events, and in particular it exceeded 95% in mid July 2013 (Fig. 2). A high temporal variability in TEP removal from seawater has also been reported for rapid sand filtration [31,46,47]. Over the entire period of this study the concentration of TEP dropped ($\rho = -0.54$, $p < 0.01$), with average values decreasing from 120 to 32 µg Xeq. L^{-1} post-MMF. Although TEP concentration decreased by less than one order of magnitude the low background values measured in seawater off Penneshaw throughout the year suggest that MMF is still suitable for TEP removal in low-TEP environments.

The concentration of Chl-*a* decreased by 47–97% across MMF during the period of study. Very low values (≤0.02 µg L^{-1}) of Chl-*a* were measured post-MMF (Site 3, Fig. 3). In a previous study the concentration of Chl-*a* was found to decrease by 30–68% due to rapid sand filtration [47]. The abundance of phyto-

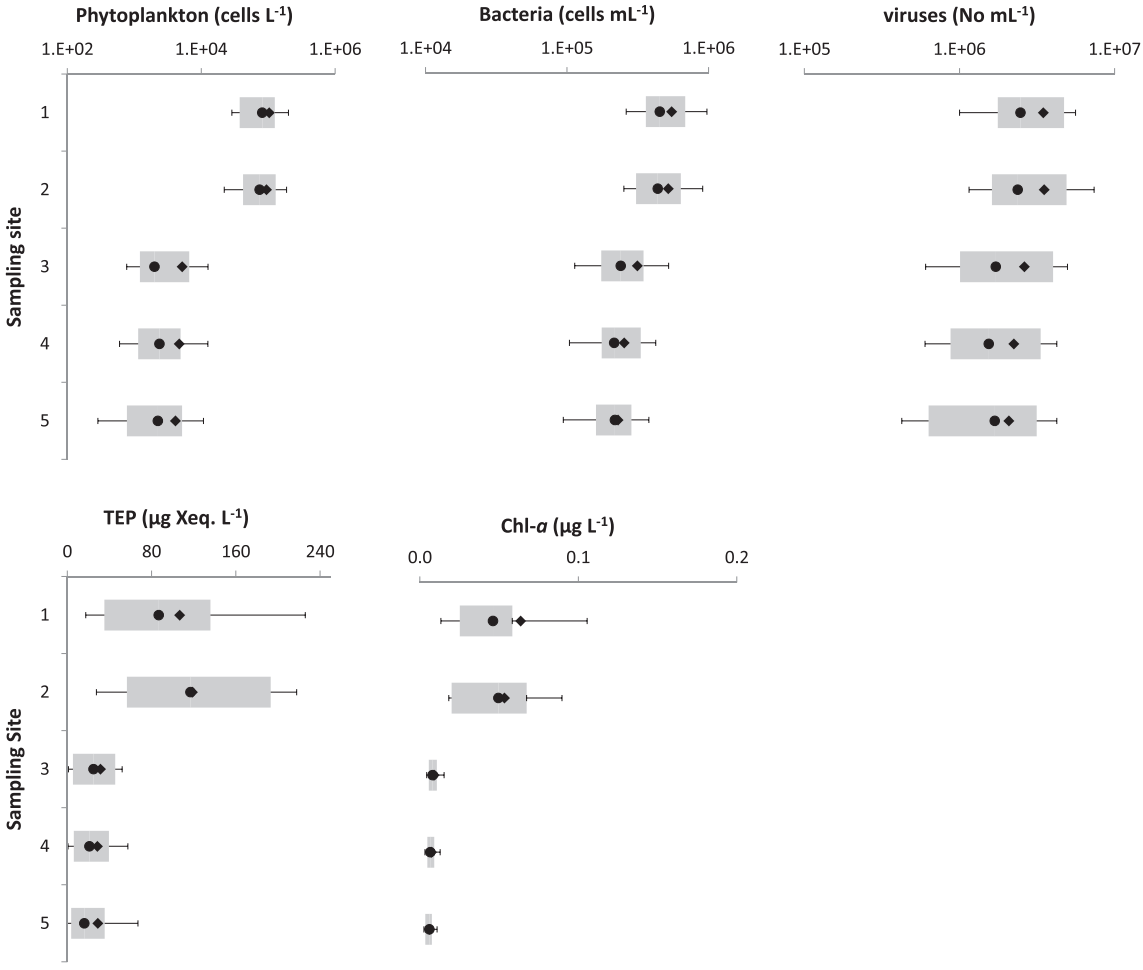

Fig. 7. Box and whisker plots highlighting the changes in the abundances of phytoplankton, bacteria, viruses as well as the concentrations of TEPs and Chl-*a*. Mean and median values are represented by diamonds and filled circles, respectively. Each box includes 25th and 75th percentiles from each parameter whereas the upper and lower narrow solid lines represent the 10th and the 90th percentiles. Values below the 10th percentile and above the 90th percentile are considered outliers and not shown in these plots. Please note that phytoplankton, bacterial and viral abundances are expressed in logarithmic scale.

plankton after MMF varied by two orders of magnitude (300–28,000 cells L^{-1}) and followed the seasonal trend observed in the upstream sampling points (Fig. 4). The removal rate of phytoplankton cells from seawater by MMF was relatively low in winter 2012 (66–89%) and increased afterwards (92–99.9%). In winter 2012 seawater was dominated by small sized (genera *Hemiselmis* and *Plagioselmis*) or elongated (*Nitzschia*) cells (Table 2) and a high proportion of these cells were able to pass through the MMF. Conventional filtration systems such as those studied here are poorly efficient in removing microorganisms smaller than 15 µm from seawater [17]. The size and shape of phytoplankton cells are highly variable and when seawater is dominated by microorganisms which have at least two dimensions <3 µm, a portion of them can easily cross the MMF.

The abundance of bacteria post-MMF ranged from $(4.9 \pm 1.1) \times 10^4$ to $(1.6 \pm 0.1) \times 10^6$ cell mL^{-1} (Fig. 5). During winter 2012 and in late June 2013 bacteria were not efficiently removed from the seawater by MMF since the values found here were similar to those measured post-MP-UV (Fig. 5).

However, beyond these sampling events bacterial abundances decreased sharply across the MMF up to one order of magnitude. A previous study carried out on three different seawater pre-treatment systems in the Mediterranean Sea, reported that bacterial abundances decreased by one order of magnitude after either microfiltration or slow sand filtration, whereas

such a decrease was less significant after dual media filtration [37]. In a Brazilian freshwater pre-treatment process bacteria decreased by 50% after sand filtration [48]. Overall, the removal rates of bacteria found in the present study are comparable to those previously reported in literature. The abundance of viruses post-MMF ranged from $(4.8 \pm 0.7) \times 10^5$ to $(7.9 \pm 0.8) \times 10^6$ mL^{-1} and, similar to bacteria, viruses were not efficiently removed from seawater during winter 2012 and late June 2013 (Fig. 6). In fact their abundance after MMF tripled in late July. Beyond these dates 4–83% of viruses were removed from seawater by MMF, therefore virus removal from seawater was not as efficient as bacteria and phytoplankton removal. Previous studies indicate poor virus removal when microfiltration is not preceded by iron coagulation [49–51].

Overall MMF was revealed to be an efficient pre-treatment in removing TEPs, phytoplankton and bacteria from seawater, whereas it showed a limited efficiency in decreasing viral abundances. The individual phytoplankton species were removed at different rates according to their size and shape and the decrease in phytoplankton across MMF was higher than that for bacteria. The less efficient removal of viruses compared to bacteria and phytoplankton is thus due to their smaller size which implied that a higher proportion of them crossed through the MMFs.

3.4. Post-cartridge filtration

No significant changes in seawater properties were observed after both 15 μm and 5 μm post-CF. The concentrations of TEPs and Chl-*a* post-CF were mostly similar to those measured in the previous sampling point (Sites 4–5, Figs. 2–3). Phytoplankton abundance was occasionally found to slightly decrease post-CF (Fig. 4) but this decrease was less significant than that observed post-MMF. Our data indicate that CF did not improve the seawater quality and was thus unlikely to decrease significantly the biofouling potential of seawater in this plant. CF in conventional seawater pre-treatment are used a final step protecting RO membrane should MMF fail to operate efficiently.

4. Conclusions

In the present study we assessed seasonal changes in the abundance of TEPs, Chl-*a*, phytoplankton, bacteria, and viruses over one year (July 2012–July 2013), within the pre-treatment system of the Penneshaw desalination plant in South Australia. All of these parameters underwent significant variability during different seasons, which is due to the seasonal variability of the feedwater. Such variability is likely to affect the biofouling potential of seawater and can potentially impact the performance of desalination plants.

The pre-treatment system of the Penneshaw desalination plant, which consists simply of MP-UV irradiation followed by three filtration steps, was shown to remove most of the microorganisms from seawater (Fig. 7). However lower removal efficiencies were found for some phytoplankton species (e.g. *Pyramimonas* sp., *Cylindrotheca closterium*, *Pseudo-nitzschia delicatissima*) which passed through all the pre-treatment steps because of their conic or elongated shapes. Viruses were also poorly removed from seawater and this is likely due to the lack of iron coagulation prior to MMF [49–51]. Furthermore, although this kind of pre-treatment system is thought to be inefficient during phytoplankton blooms, we did not observe a loss in performance (i.e. decrease in cell and particle removal rates) when microorganism abundance in seawater peaked over the course of the study. Within the seawater pre-treatment, the most efficient step was MMF, whereas both MP-UV irradiation and CF revealed poor efficiency. Our results questions the usefulness of two sets of cartridge filters located downstream to MMF in conventional seawater pre-treatment.

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Abbreviations

TEP	—	transparent exopolymeric particles
RO	—	reverse osmosis
SWRO	—	seawater reverse osmosis
UV	—	ultra-violet
FIA	—	flow injection analyser
SSC	—	side-scattered light
MMF	—	multimedia filtration
CF	—	cartridge filtration

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