

Potential biofoulants in open-ocean SWRO desalination station in Jeddah, KSA

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ABSTRACT

Currently, water desalination is an essential solution for the high demand for water worldwide. A sea water reverse osmosis (SWRO) facility fulfills the need for pure water. Conventionally, these plants use open-ocean water that is rich with natural organic matter (NOM) and transparent exo-polymers (TEP). Marine flora increases the demand for fouling the membrane in the SWRO facility that raises the pressure and results in the halting of the station. Therefore, water samples were collected from SWRO stages during high-pressure problems to probe the factors that play a key role in membrane biofouling. NOM and TEP particles physically disappeared after the dual-media filter (DMF). However, turbidity significantly increased after the DMF stage, which is indicative of the fragmentation of NOM and TEP particles. Chlorophyll and phycoerythrin disappeared after the DMF stage but were observed in the reject stage (brine). Therefore, NOM and TEP are playing a role indirectly in membrane biofouling. Fifteen potential species of heterotrophic prokaryotes are identified and recorded in all stages of the SWRO. The characteristics of these species imply that they form a cooperative consortium that potentially creates the biofilm in the RO membrane. Therefore, SWRO facilities that use open-ocean water must develop highly sophisticated pretreatment technology to eliminate the seeds of the biofilm that fouls RO membranes.

Key words | biofoulants, natural organic matter, Red Sea, reverse osmosis pretreatment, sea water, transparent exopolymer particles

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INTRODUCTION

If the current global water consumption rate continues to increase, almost two-thirds of the world population will suffer from water troubled situations by the year 2025. Sea water represents 97.5% of the planet's water, therefore, desalination of sea water is a promising solution to meet the future call in the water supply (Dehwah *et al.* 2016). Water desalination is capital- and energy-intensive (Fane *et al.* 2015) making the cost of purified water very expensive (Pérez-González *et al.* 2012). Therefore, development of innovative and inexpensive desalination processes is imperative (Amezaga *et al.* 2014). There are two widely used desalination technologies, reverse osmosis (RO) and multistage flash (MSF)

(Zhou *et al.* 2012). Membrane-based RO uses high-grade electrical energy (Liu *et al.* 2016), and the membranes are prone to fouling and thus need frequent replacement (Boo *et al.* 2016).

Fouling happens when the foulant accumulates on the membrane exterior surface and diminishes water yield and cuts membrane lifetime, which aggravates the cost of water purification (Khan *et al.* 2014). Ineffective fouling characterization severely hinders fouling alleviation and control (Kumar *et al.* 2006). Therefore, accurate fouling characterization is the key to running water desalination plants optimally (Kim *et al.* 2016). Colloidal, organic, biological and scale are four major classes of foulant in RO

technology (Ji *et al.* 2010). Unfortunately, membrane fouling in RO practices is a difficult phenomenon that cannot be appropriately or effectively described by flux failure (Bucs *et al.* 2014). Although biofilm development in RO systems is currently documented, there is insignificant identification of the variety and complexity of the microbes that are accountable for the construction of biofilms (Manes *et al.* 2011). Accordingly, the structure and strength of the species forming biofilm populations that linger on RO membranes is principally unknown (Khan *et al.* 2015).

Saudi Arabia is fronting a real challenge to secure its fresh water supply (Dehwah & Missimer 2016). Currently, the country produces 3 million m³/day desalinated water, which is a quarter of world capacity and is expected to become 5.7 million m³/day. Desalinated water supply is only utilized for public and industrial consumption, however, agriculture depends on nonrenewable groundwater (Parkinson *et al.* 2016). Jeddah, the second largest city, is located on the Red Sea coast. The population is around 3.5 million and is projected to double in the next 20 years. As an arid area, the city is fully reliant on desalinated sea water. A total of 3 million m³ is produced to fulfill a clean, healthy water supply. Unfortunately, Jeddah's location and climatic characterization pose a challenge for desalination stations due to high salinity and growth conditions that favor frequent algal bloom and growth of diverse bacterial species (Khan *et al.* 2013). These complex biological components have critical impacts on the operation of sea water reverse osmosis (SWRO) plants (Dehwah & Missimer 2016). Indeed, this study aims to identify bacterial species that directly or indirectly contribute membrane fouling. In this project, we identified the dominant species that are usually present in the Red Sea explicitly that inhibit feed sea water for the selected desalination stations, and those that escape, and bloom within any stage of the RO process, colonize and form a biofilm.

MATERIALS AND METHODS

Physical parameter measurements

Physical parameter analysis requires on-site measurements to avoid unknown reactions of thermal waters and gather water properties in original conditions. Therefore, the EXO2

Multi-Parameter Water Quality Sonde (Yellow Springs, OH, USA) is used. It is equipped with titanium sensors, biofouling protection with copper-alloy components and anti-fouling wipers. The sonde is attached to an ROX® dissolved oxygen probe, pH probe, conductivity/temperature probe, turbidity probe, and a chlorophyll/BGA probe. Each water quality datum is associated with a date, time, and GPS latitude and longitude coordinates, and Bluetooth wireless technology connects the sonde and handheld to a personal computer by KOR software. The sonde is left in water samples for measurements, and three readings are captured every 3 minutes.

Analyses of chemical species

Subsurface water samples were collected at all sites in polyethylene bottles of 1 litre capacity. Three replicates were received from different locations surrounding each sampling site at various distances. All chemical analysis of the macronutrients and micronutrients was by YSI kits used with a YSI 9500 Photometer (Yellow Springs, OH, USA) according to the manufacturer's protocols. Seasonal field visits to the study area started in winter 2015 till autumn 2016.

Identification of bacterial isolates

BioMérieux's (VITEK®2) is a system to identify bacteria and yeast with test kits using a robust database. VITEK®2 customs single ID and antimicrobial susceptibility test (AST) cards that are ready-to-use, with a comprehensive list that provides identification in 5–22 hours. After organism isolation, a simple standardized inoculum is placed into the VITEK®2 cassette at the Smart Carrier Station™. Once the sample is loaded, sequential steps for each card are accomplished by the system without human interference. Isolated strains were sub-cultured to ensure purity in LB medium. To prepare the inoculum for analysis, each isolate was developed within 18–24 h on LB medium at 35 °C. Suspensions of the isolate cultures were prepared in 0.45% saline before loading turbidity adjusted to a 0.6 McFarland Stock. Samples were loaded on cards well suited to VITEK®2 according to the manufacturer's directions. VITEK®2 has been tested in several countries and has been used for over 15 years (Joyanes *et al.* 2001; Horstkotte

et al. 2002; Sader *et al.* 2006; Lupetti *et al.* 2010; Winstanley & Courvalin 2011).

Quantitative and qualitative analysis of TEP

Water samples were collected from five stages in the RO desalination station in Jeddah port. Feed water, first pretreatment, second pretreatment, cartilage filtration, and brine samples were collected and transported to the laboratory and stored in a 4 °C room. A concentrate of transparent exo-polysaccharides was prepared as follows: three litres filtered by 0.22 millipore filter, permeate was discarded, however, retentate was washed from the filter by 3 ml deionized water and was used for transparent exo-polymer (TEP) analysis.

RESULTS AND DISCUSSION

Sea water in Jeddah port is rich in a massive amount of organisms that are hard to enumerate with around 1,000 species of fish and 150 species of corals, hundreds of seaweeds, phytoplankton, zooplankton, bacteria, and others. Therefore, marine water is expected to produce a reasonable amount of TEP and natural organic matter (NOM) particles as well as biofoulants with the ability and tendency to

develop biofilm and engender membrane biofouling problems in the water purification station.

Quantitative and qualitative analysis of TEP

TEP dominated the feed water with a concentration of 20×10^6 – $100 \times 10^6/\text{m}^3$. The size of the TEP (Figure 1) is about 600 – $2,500 \mu\text{m}^2$. The nature and composition of the TEP as inferred from the microscopic examination is different from one particle to another. Seaweed pieces seen in some TEP, phytoplankton and picoplankton are present where all these mostly stick together by natural adhesives produced by organisms that inhibit TEP or exudate from external organisms like jellyfish. Definitely, bacteria are present in water and in TEP. The first pretreatment stage in the RO desalination station was enough to reduce TEP to zero. Interestingly, in all stages in the desalination station bacteria were the dominant particle in the water as seen by dark and phase contrast images (Figure 1). The presence of bacteria was doubled in the brine stage. The observations seen by light images and plate count supported this statement.

Transparent exopolysaccharide particles are abundantly present in the feed water (Dehwah & Missimer 2016). However, they almost disappeared after the first stage of pretreatments. It is unknown whether these particles were fully retained in the first filtration pretreatment stage or

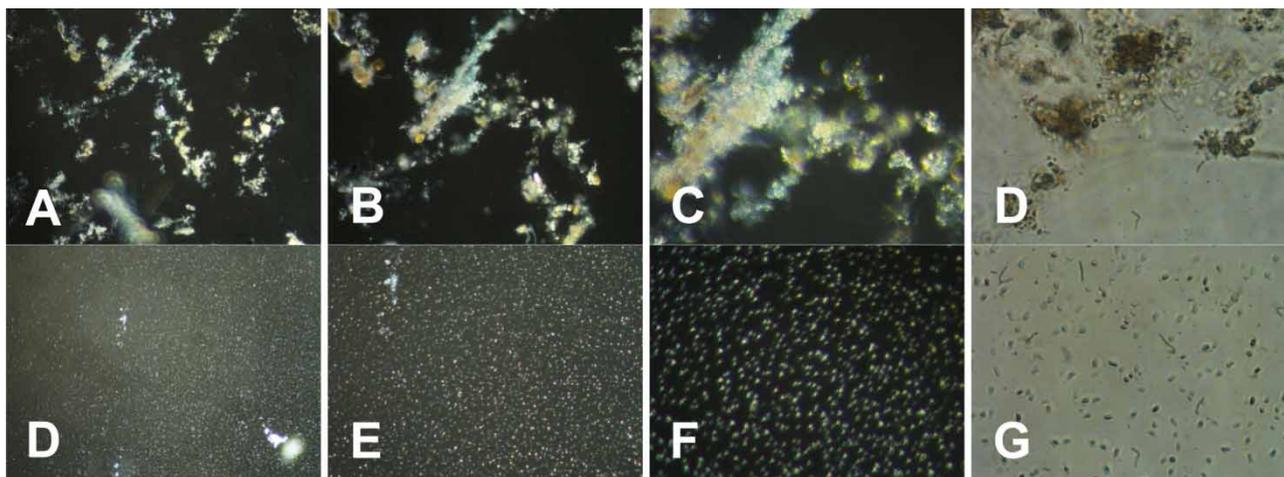


Figure 1 | Image of solid suspended particles and biofoulant candidates in the sea water inlet of the RO desalination plant. Dark field light microscope images (A)–(C) show aggregates of sea water organismal parts and plankton forming heterogeneous particles. Dark field light images (D)–(F) show sea water after pretreatment (multimedia filter: DMF stages) dominated by bacteria as a major biofoulant threat to the RO membrane. Phase contrast image (D) shows at $100 \times$ magnification the heterogeneous solid particles present in (A)–(C) images, (G) is a $100 \times$ magnification of the (D)–(F) images and provides evidence for the domination of bacteria in the water that is heading toward the RO membrane.

fragmented into nanoparticles that amplified their role in the biofouling problem. Concentrate of feed water after the first pretreatment stages was dominated by bacteria in all stages. The significant increase of bacteria in the brine is indicative that these species colonized and propagated in the previous stages of the RO membrane.

Chemical analysis of sea water

Physicochemical characteristics of sea water are shown in Tables 1 and 2. Sodium and chloride are the major ions followed by high concentrations of sulfate and magnesium. These analyses are indicative that sea water is a nutritive medium for many organisms that can cope with the high-level sodium. On the other hand, high ion concentration along with TEP and NOM particles collectively prepare the RO membranes' surface for biofilm-forming bacteria to colonize and foul the membrane.

Physical analyses of SWRO stages

Seven samples were analyzed to measure physical parameters that represent all stages of the SWRO station in Jeddah. These are sea water, DMF (dual-media filter), before cartilage filter, after cartilage filter, RO feed, brine

Table 1 | Inlet sea water chemical analysis of the desalination plant

Chemical species	Mean (mg/L)	SD
Ammonia (NH ₄ ⁺)	1	0.012
Nitrate (NO ₃ ⁻)	1	0.014
Phosphate (PO ₄ ⁺)	≤1	0.011
Chloride (Cl ⁻)	23,000	0.23
Sodium (Na ⁺)	12,748	210.25
Sulfate (SO ₄ ²⁻)	2,800	0.11
Magnesium (Mg ²⁺)	1,400	11.25
Calcium (Ca ²⁺)	480	4.45
Potassium (K ⁺)	476	22.11
Bicarbonate (HCO ₃ ⁻)	143	17.45
Bromide (Br ⁻)	69	2.03
Borate (BO ₃ ³⁻)	23	1.02
Fluoride (F ⁻)	2.97	0.11
Silicate (SiO ₃ ²⁻)	6.5	0.011

Table 2 | Year-round average of the physical properties of the sea water inlet for the desalination plant

Physical parameters	Mean	SD
Temp °C	26.54	0.245
Cond μS/cm	60,313.4	21.04
TDS mg/L	38,085.33	125.3
Sal psu	39.1	0.275
ODO mg/L	5.73	0.014
pH	8.16	0.125
Hardness	7,200	115.23
Alkalinity	104	5.112

(RO reject), and final product. Conductivity and total dissolved salts (TDS) (Figure 2) are the same in all steps of desalination except for their increase in the brine by 40%. The pH and dissolved oxygen (OD) values (Figure 3) are the same in all samples except RO reject (brine). The pH increased by 6.5%, however, and OD increased 24%. The turbidity pattern across RO stage shown on Figure 4 is measured in Formazin Nephelometric Units (FNU) by infra-red light and dramatically increased after the DMF stage. However, turbidity was similar after the cartilage stage with that of the feed water, but was significantly increased after the RO feed and reject stages. This observation is critical for the whole process. It means that DMF is not working in the planned direction. Indeed, attentive assessment of the pretreatment stage is crucial to optimize feed water quality.

Chlorophyll and BGA-PE measurements

Chlorophyll measurement is indicative of green photosynthetic organisms that include picoplankton and seaweed. However, BGA-PE measures blue green algae only. This might describe the divergent performance drawn from these two factors in the RO stages (Figure 5). Chlorophyll and phycoerythrin are present in sea water. They were fully removed after the DMF stage. However, they were measured in the RO reject at a concentration higher than that of the original (sea water). The presence of chlorophyll in the RO feed may suggest the occurrence of algal contamination in the RO feed.

Pretreatment is a prerequisite step in the desalination process which removes living and nonliving particles to

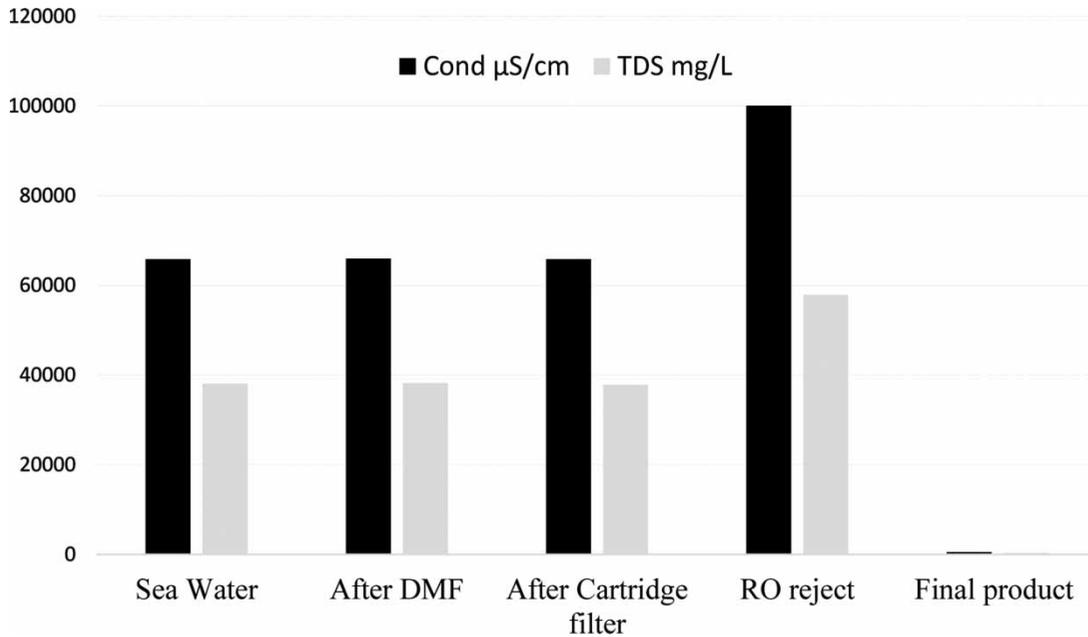


Figure 2 | Conductivity and TDS average values measured for different stages in the SWRO facility in Jeddah.

minimize membrane exposure to foulant. However, there is a tendency to generate special foulant during the pretreatment like biological macromolecules. Therefore, careful examination of pretreatment impact on foulant status in the final feed water to the RO membrane is a prerequisite

to understand foulant dynamics. Foulant dynamics is expected to be site-specific for each water treatment station. Indeed, field comparison of different desalination stations is expected to resolve this issue and provide knowledge that is site-specific to each location.

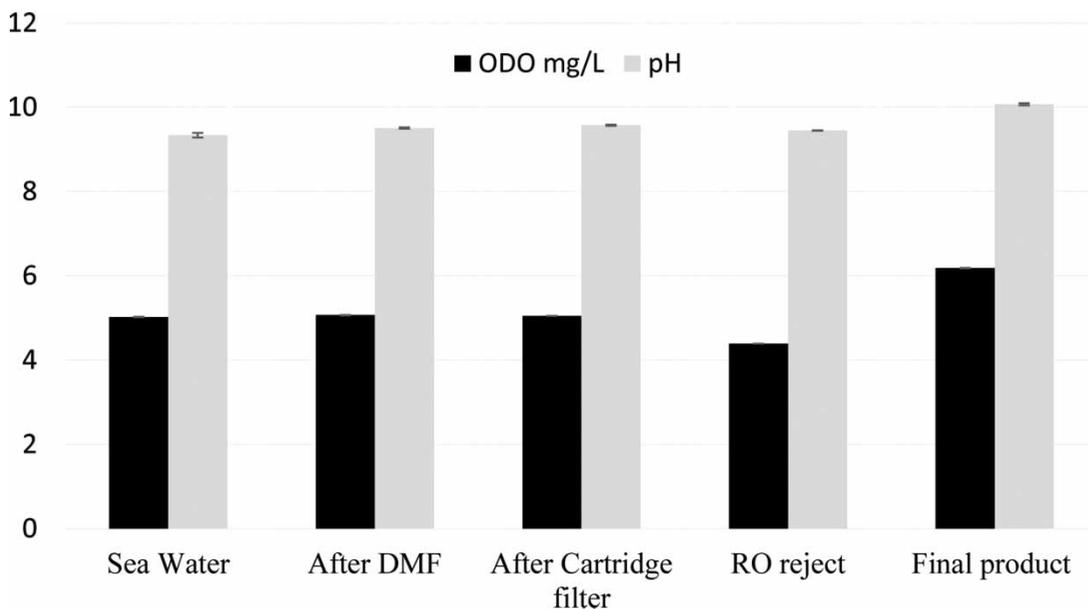


Figure 3 | Dissolved oxygen and pH average values measured for different stages in the SWRO facility in Jeddah.

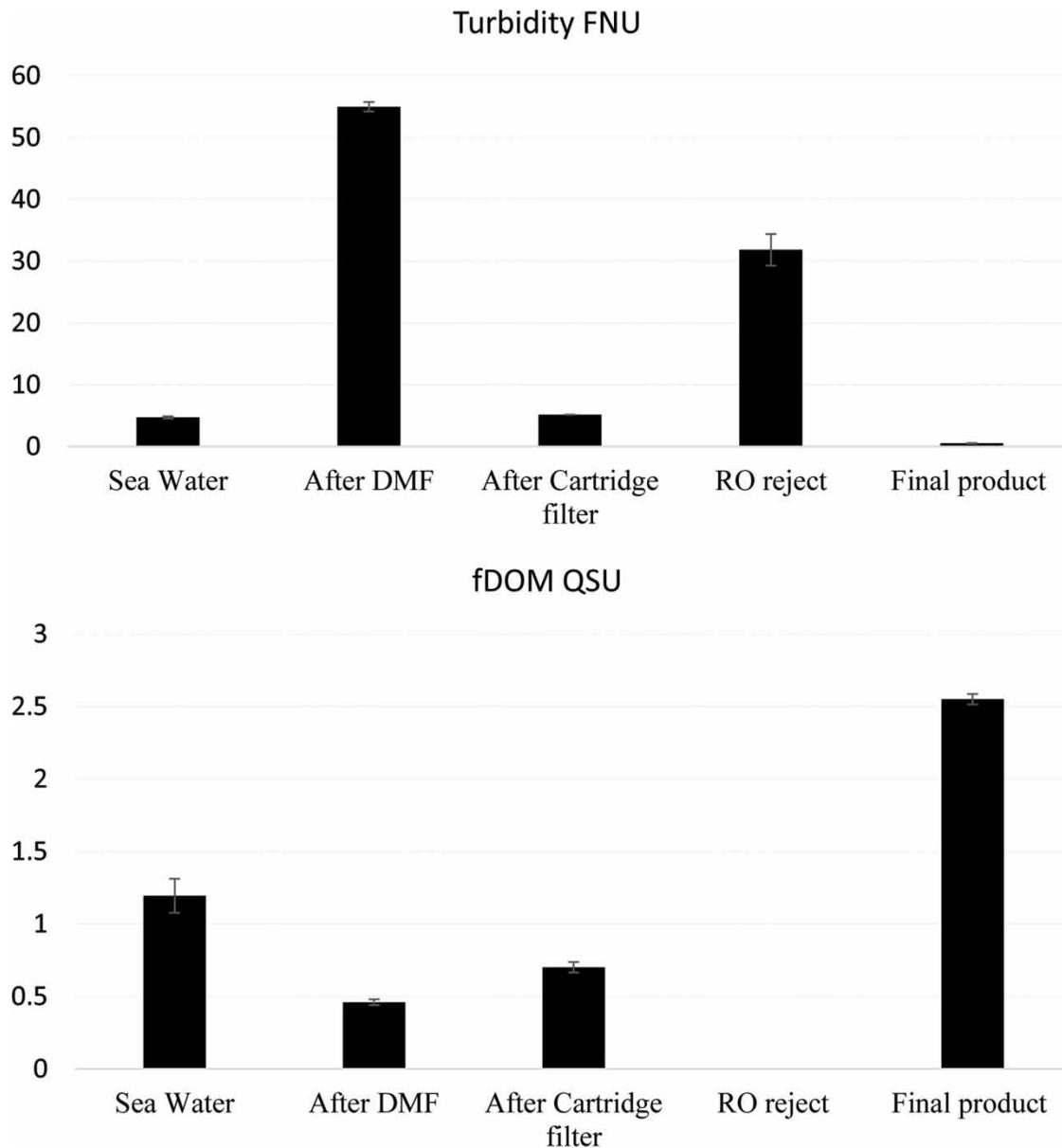


Figure 4 | Turbidity and fluorescence dissolved organic matter measured for different stages in the SWRO facility in Jeddah.

Diversity of microorganisms after pretreatment

A total of 15 bacterial species were recorded before and after the RO membrane (Table 3), and this result basically indicates that these species passed over the RO membrane surface, which makes them potential biofoulants. Only those species that have the ability to colonize and form biofilm are expected to be present on the RO surface. Among these species, *Sphingomonas paucimobilis*, *Brevundimonas*

diminuta, *Brevundimonas vesicularis*, *Acinetobacter lwoffii*, and *Micrococcus luteus* have a potential to secrete an extracellular matrix, pioneer biofilm colonizers, and utilize and live on an extracellular matrix, and some of them interact synergistically during biofilm formation. The coexistence of this consortium in RO desalination conditions only leads to a high risk of fouling.

The identified bacterial species and their nature provide conclusive evidence that these species are inhabiting the RO

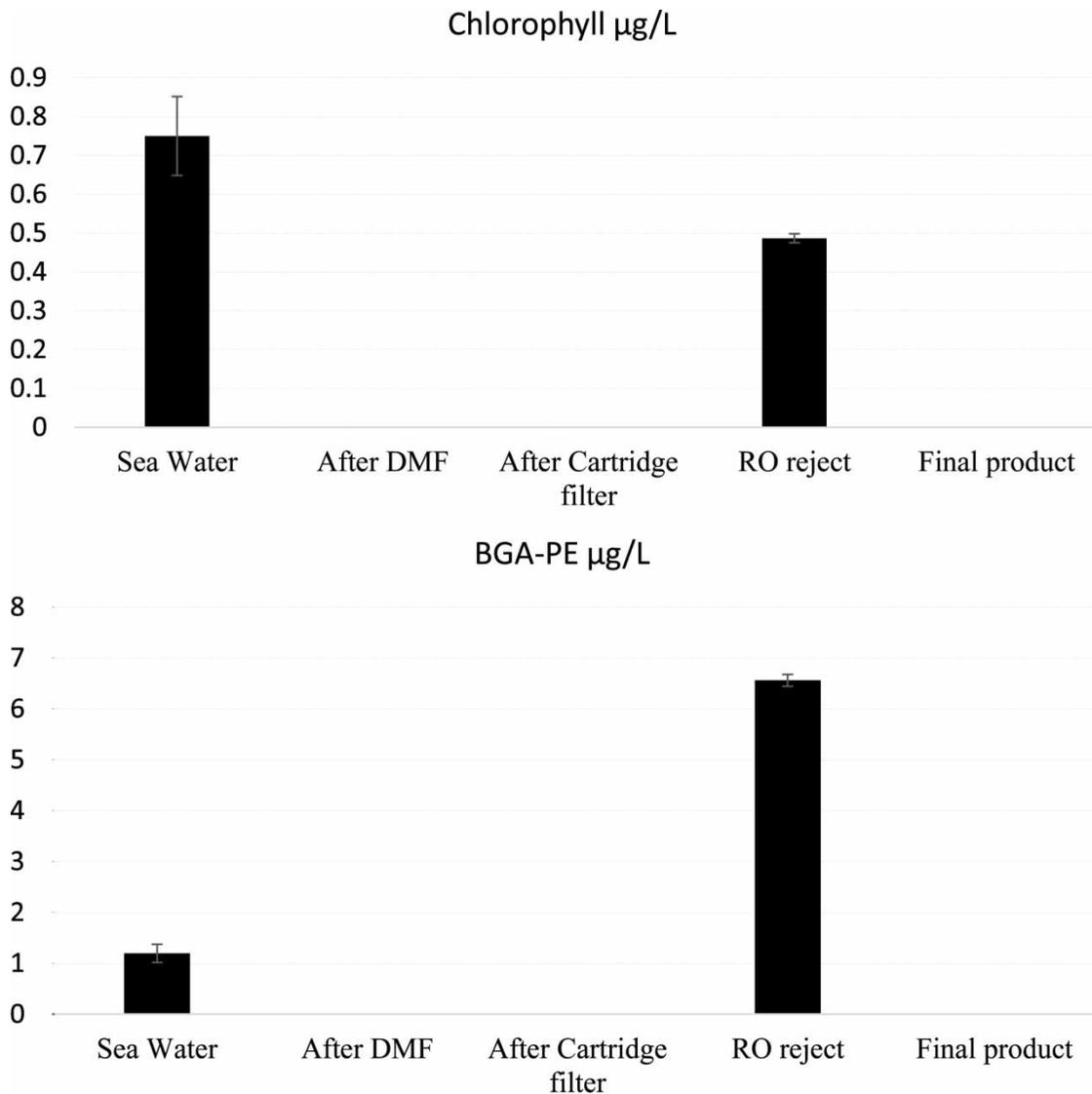


Figure 5 | Chlorophyll pigment and BGA-PE (blue green algae concentration measured as phycoerythrin) values in all stages of the SWRO facility in Jeddah.

membrane. *Brucella melitensis* is a small Gram-negative coccobacillus that is abundantly present throughout the different RO stages and increased significantly in the brine. The source of this bacterial species is mainly from dead animal discharge in the Red Sea (Ducrotoy *et al.* 2016). This species is known to form biofilm and produce exopolysaccharides (Godefroid *et al.* 2010). *Sphingomonas paucimobilis* is a Gram-negative aerobic bacillus that lives in soil. It possess a single polar flagellum that helps cells with gentle motility (Kim *et al.* 2008). The cell size is around $0.7 \times 1.4 \mu\text{m}$. This bacterium is able to produce up to 1.2 g/l exopolysaccharide (Manivasagan & Kim 2014)

and is a powerful foundation for biofilm formation because it can consume its own exopolysaccharide and other exopolysaccharides available by other bacteria (Herzberg *et al.* 2009). *S. paucimobilis* is capable of hydrolyzing lignin compounds and also fucoidan (Li *et al.* 2013). Lignin and fucoidan are two organic polymers that are produced by many marine plants and algae (Popper *et al.* 2011). Indeed, it is expected that these polymers are abundantly present in the port areas that are characterized by high mixing activities from cargo-ships (Tran *et al.* 2013). *Sphingomonas paucimobilis* is found to dominate biofilm (Ceyhan & Ozdemir 2008) after DUWL disinfection by 88.79% (Szymanska

Table 3 | List of microorganisms present in different treatment stages of RO desalination station

No.	Species	Sea water	DMF	Cartilage filter	Brine
1	<i>Brucella melitensis</i>	+++	++++	+++	+++++
2	<i>Sphingomonas paucimobilis</i>	++++	+++++	+++	+++++
3	<i>Acinetobacter lwoffii</i>	+	++++	+++	+++++
4	<i>Brevundimonas diminuta</i>	+	+++	++	+++
5	<i>Brevundimonas vesicularis</i>	+	+++	++	+++
6	<i>Kocuria rosea</i>	+	++	++	++
7	<i>Dermaococcus nishinomiyaensis</i>	+	++	+	++
8	<i>Kytococcus sedentarius</i>	+	++	+	++
9	<i>Myroides</i> spp	+	++	+	++
10	<i>Roseomonas gilardii</i>	+	++	+	++
11	<i>Vibrio vulnificus</i>	+	++	+	++
12	<i>Moraxella</i> group	+	++	+	++
13	<i>Micrococcus luteus</i>	+	++	+	++
14	<i>Micrococcus lylae</i>	+	++	+	++
15	<i>Pontoea</i> spp	+	++	+	++

2006). Sphingomonadales are reported as primary colonizers of RO membranes (Bereschenko *et al.* 2010) and form biofilms under high shear rate with different bacterial compositions (Bereschenko *et al.* 2011). *Brevundimonas diminuta* is a potential foulant that is recorded in this work. It secretes a slime-like EPS matrix, where its biofouling affects the hydrodynamic backwashing of RO membranes (Badireddy *et al.* 2008). Protein production by this species contributes to biofilm formation (Miyoshi *et al.* 2015). One additional species of this strong biofoulant genus is *Brevundimonas vesicularis*, which produces 1.20 g/L heteropolysaccharides that are composed of glucose galactose, rhamnose mannose, and uronic acids (Rättö *et al.* 2005). Interestingly, at the high shear rate, the bacterial community that forms biofilm on the RO membrane is known to produce huge amounts of extracellular polymeric substances that may provide more protection for bacterial cells in the biofilm (Verhoef *et al.* 2002). *Roseomonas gilardii* which is Gram-negative, and strictly aerobic, was isolated from biofilm (Furuhata *et al.* 2007). *Vibrio vulnificus* is present in seafood (Raszl *et al.* 2016), which is indicative that this species is abundant in marine organisms (Jones *et al.* 2016). *Acinetobacter lwoffii* is one of the dangerous biofilm initiators (Burmölle *et al.* 2006), because it interacts synergistically with any biofilm-forming species and forms a powerful biofilm

(Guerrieri *et al.* 2008). *Micrococcus luteus* is a cultivable marine bacterium that is identified to generate a biofilm on the sunken surface of cargo-ships (Inbakandan *et al.* 2010). The other determined bacterial taxa are well known as potential biofilm-forming species and are usually inhabiting deep sea water and live in the muddy bottom (Meier *et al.* 2013).

CONCLUSION

This study shed light on the presence of bacteria as a potential biofoulant for the SWRO plant. It is evident that bacteria are the leading cause of biofouling and the generation of high pressure that causes temporary shutdown of the RO facility. Therefore, searching for a solution to control bacterial species in the feed water must minimize or eliminate the biofouling problem. Indeed, a desalination station using RO membranes in the port area must consider high precautions and select special protocols to remove biofilm-forming microbes. The bacterial biofouling in the RO desalination plant is a common problem with all open-ocean desalination stations. However, the presence of Jeddah port on the east coast increases the probability of bacteria that produce polysaccharides and natural adhesives. Polysaccharide mixture in addition to adhesive presents a

natural seed for biofilm formation in both natural and artificial environments. Indeed, a pretreatment stage that removes 100% of bacteria is a key solution for membrane biofouling in an open-ocean SWRO desalination facility.

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