"This is an Accepted Manuscript of a book chapter published by Routledge in Polymetallic Coatings to Control Biofouling in Pipelines on 2021, available online: https://doi.org/10.1201/9781003193449

## 2.1 Introduction

Biofouling is an inevitable phenomenon demonstrated by the attachment and buildup of microorganisms and the development of a biofilm (She et al. 2016) on the inner surfaces of pipelines and on membranes used in water treatment and desalination (Wingender, Neu, and Flemming 1999). The biofouling phenomena and remedies are the same for pipelines and reverse osmosis membranes used in seawater desalination and wastewater treatment for reuse. Two other chapters in this book discuss on biofouling in pipelines and their characterization methods. Thus, this chapter discusses mainly on biofouling in reverse osmosis membranes. This chapter aims to elucidate the biofouling mechanisms and their adverse effects, biofouling detection, and remediation methods. It also highlights the key issues related to the use of pretreatment schemes for biofouling mitigation.

The researchers reported that within a few years of the service, potable water distribution pipelines were found to be accumulated with a fine film of microbes that also pose major public health issues (van der Kooij, Visser, and Hijnen 1982). The microorganisms can regrow on the water-carrying pipe network in the presence of certain most significant constituents in water such as biodegradable organic matter (BOM), ammonia, iron, manganese, nitrite, soluble hydrogen, and reduced sulfur compounds. Moreover, it makes water biologically unstable due to the biofilm development caused by biodegradable organic matter (BOM), ammonia, iron, manganese, nitrite, dissolved hydrogen, and sulfur in reduced form. Water quality often adversely is affected by chlorine dosing that produces undesirable disinfection by-products (DBPs), which are unsafe and carcinogenic in nature.

In the seawater desalination process, RO membrane fouling is a major hurdle that reduces permeate flux and increases operating cost OMBRs (Sun et al. 2018). RO membrane commonly encounters colloidal fouling, organic fouling, inorganic scaling, and biofouling (Matin et al. 2011). The deposition of colloidal particles on membranes is called colloidal fouling and deposition and adsorption of macromolecular organic compounds on membranes is termed as organic fouling. The precipitation of dissolved inorganic compounds on the membrane surface is called inorganic scaling, while biofouling is the adhesion and accumulation of microorganisms on the membrane surface (She et al. 2016; Vrouwenvelder et al. 2009). The complex biofouling phenomena occur at the inner surface of pipes and on the RO membrane surface, which is accompanied by the agglomeration of soluble microbial products (SMP) and extracellular polymeric substance (EPS), thereby forming biofilm on the respective surface (Wingender, Neu, and Flemming 1999; Chun et al. 2017).

In response, the feed water pretreatment (such as microfiltration, ultrafiltration, chlorine dosing, and biocides addition) is highly effective in the elimination of 99.99% of microbes. Though only a few colonies of bacteria enter the system, they stick to the surfaces and then reproduce and grow on the surfaces in contact with even an oligotrophic environment (Chun et al. 2017). Therefore, biofouling is significant and inevitable in the RO membrane process even after periodic cleaning cycles and adapting other antifouling pretreatment strategies such as ozone and chlorine dosing or biocides application (Flemming et al. 1997). Furthermore, the polyamide RO membrane is susceptible to oxidation by free chlorine species (HOCl and $OCl^-$). However, the growth of resilient strains of microbes can be adversely affected by the constant use of disinfectants (Kang et al. 2007; Shannon et al. 2008). Biofouling leads to a drop in permeate velocity, selectivity, and membrane service life. Furthermore, it increases cleaning frequency and operational cost of chemicals and electricity (Luo et al. 2018; Vrouwenvelder, van Loosdrecht, and Kruithof 2011; Flemming et al. 1997).

In RO desalination, plant fouling is a major concern that requires frequent cleaning, shortens the membrane life, reduces permeate flow, and increases up to 50% total operating cost (Ridgway 2003; Bell, Holloway, and Cath 2016). Different cleaning tactics include the use of antiscalants and acids and importantly all such chemicals act very differently, and even certain commercially available chemicals cause biofouling (Vrouwenvelder et al. 2000).

## 2.2 Biofouling Mechanisms

Biofouling is a thin and compact gel-like biofilm layer formed on the inner surface of pipes or on the RO membrane surface. This biofilm formation involves three subsequent phases as shown in Figure 2.1.

i. Movement of microbes to the membrane surface,
ii. Adhesion to the surface, and
iii. Formation of nuclei and layer-by-layer addition of microbes on the surface (Al-Juboori and Yusaf 2012).

Biofilm formation in the membrane process is separate from the simple deposition of particles on the membrane, which are readily removed by physical washing (Miura, Watanabe, and Okabe 2007).

The first stage which occurs in minutes to hours is a reversible process. The initial attachment of organic matter, colloids, nutrients, and bacterial cells onto a membrane surface occurs at the time of contact of feed water and RO membrane (Subramani and Hoek 2010). During the second stage, cell attachment and micro-colony formation begin on the membrane surface where attached bacteria consolidate the bonding by secreting soluble microbial products that form complex with organic constituents. Consequently, biofilm adhesion becomes irreversible, and the organism becomes attached to the surface in a stack. This

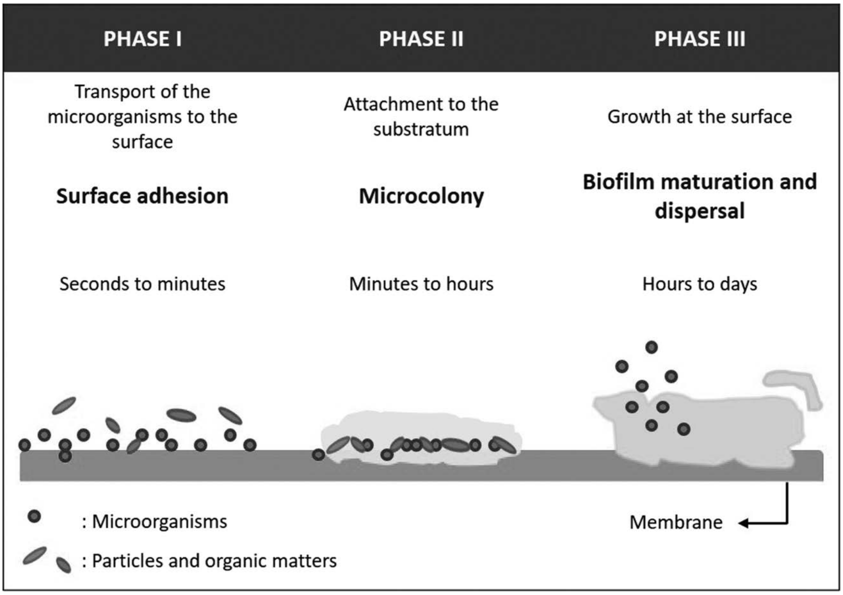


**FIGURE 2.1**
Schematic of biofilm formation.

phenomenon happens so rapidly when feed water meets the membrane skin layer. This biofilm is extremely hard to disengage from the membrane surface, which demands rigorous feed water pretreatment prior to its contact with the membrane (Adham et al. 1991; Habimana, Semião, and Casey 2014).

The adhesion of microbes onto the membrane surface is caused by complex physicochemical and biological processes. Feed characteristics, temperature, membrane properties, and module geometry play a role in this (Miura, Watanabe, and Okabe 2007; Subramani and Hoek 2010). The feed characteristics include pH, TDS, conductivity, and dissolved organic carbon. The properties of the membrane and pipes such as surface roughness, charge, and hydrophobicity are also significant parameters. The water flux is also an important factor in cell attachment to the membrane surface; however, the microbial community can attach via Brownian deposition in the absence of permeation (Subramani and Hoek 2010; Schneider et al. 2005).

## 2.3 Biofouling Control and Prevention Strategies

Biofouling poses major challenges, and its monitoring and control incur a huge financial burden on the RO water plants (Flemming 2002). Biofouling can be minimized by fouling control and prevention techniques. In the biofouling control strategy, chemical cleaning is performed to reestablish permeate flow. This can be achieved by increasing cleaning cycles. However, the frequency of chemical cleaning must be minimized as it increases the operating cost of chemicals and decreases the membrane life (Onoda 2016).

On the other hand, biofouling prevention is a more suitable option as compared to fouling control. Two common strategies for biofouling prevention are:

1. Membrane modification
2. Feed water pretreatment

Membrane modification technically controls the adhesion of microbes or inactivates the bacteria that get attached to the surface. This technique is still in the experimental stage and not explored yet for fouling prevention. On the other hand, the second alternative of feed pretreatment removes microbes, organics, and nutrients from the feed water. To achieve this objective, various pretreatment techniques are employed. They are MF/UF membrane filtration, ultraviolet light treatment, hypochlorite disinfection, or biocide use prior to RO filtration. The prevention techniques are beneficial as they incur less cost and energy consumption due to fewer amounts of chemicals used and lessen the adverse impact on the ecosystem. Furthermore, seawater desalination by the RO process becomes smooth and economical as feed pretreatment produces consistent water quality with a negligible amount of potential foulants to RO inlet. Raw water characteristics vary with geographical location and influent water quality analysis is essential to quantify biofouling potential. As a result, it is important to visualize and characterize the spatial distribution and transport of key membrane foulants. These fouling characteristics provide insights into the design of the pretreatment process and fouling mitigation strategies (Adham et al. 1991; Luo et al. 2018; Chun et al. 2017).

The design and operation of seawater reverse osmosis desalination (SWRO) processes relies heavily on the feed water characteristics. Thus, SWRO plant performance and selectivity are governed by the pretreated RO influent. Hence, the pretreatment process is a very significant section of the desalination plant. In this pretreatment stage, all particulate matters, colloidal particles, organics, nutrients, scalants, and bacterial impurities are removed from saline water in order to prevent them from reaching the sensitive RO membrane skin layer and prevent membrane fouling. The peculiarities and number of contaminants in source saline water (influent) directly relate to the pretreatment processes, produced water yield, and selectivity.

Preston (2005) reported that dissolved organic carbon (DOC) in seawater varies from 1 to 3 mg/L. The seawater analysis data can be obtained by performing laboratory analysis for certain target compounds (Lu and Wang 2019). Also, instrument analysis with liquid chromatography-organic carbon detection (LC-OCD) is based on the size-exclusion chromatography. It is utilized to characterize the water-soluble organic carbon (OC) and it provides

MW fractions of organic matters in seawater in terms of biopolymers including polysaccharides and proteins (>20,000 Da), humic substances (highly UV-absorbable, hydrophobic, 800–1,000 Da), building blocks (Breakdown products of humic substances, 350–600 Da), low molecular weight (LMW) acids (aliphatic and low molecular weight organic acids, biogenic organic matter, 350 Da), and low molecular weight (LMW) neutrals (alcohols, aldehydes, ketones, amino acids, biogenic organic matter, 350 Da) (Huber et al. 2011).

## 2.4 Pretreatment and Membrane-Based Systems

Leparc et al. (2007) reported that for seawater pretreatment, conventional treatments such as dual media filtration (DMF) and cartridge filters and advanced pretreatments such as MF/UF filtration are employed (Leparc et al. 2007). The advanced pretreatment is getting more acceptance due to the complete removal of seawater contaminants as compared to DMF. Furthermore, few chemicals are consumed and MF/UF-based membrane filtration techniques are robust. MF/UF-based membrane filtration is gaining more popularity and acceptance as a pretreatment to RO. MF/UF provides consistent quality of safe and secure water to RO due to its absolute rejection for suspended solids irrespective of source seawater quality (Baig and Al Kutbi 1998; Pearce 2007). MF filters are available in pore sizes of 0.1–0.2 μm. UF membranes possess fine pores as compared to MF. UF is available in the range of 0.01–0.02 μm and sometimes even smaller to 0.005 μm. UF membranes are capable to reject all particulate matters and the majority of dissolved substances, including bacteria and viruses. The UF has better rejection than MF due to its low molecular weight cut-off (MWCO). The MF/UF rejection is subjected to the feed water characteristics and molar mass of the species (Baig and Al Kutbi 1998).

### 2.4.1 Biofilters

Biofilters are extensively used in air, water, and domestic sewage treatment characterized by biomass attached to its septum. A variety of biofilters are being used in water/wastewater treatment applications such as trickling filters, granulated activated carbon (GAC), sand filters, and horizontal rock filters to name a few. GAC-based biofilters were found useful typically in potable water purification due to the re-growth of bacterial colonies in a water distribution network pipe.

GAC-based biological treatments are more effective in oxidizing organic matters responsible for bacterial growth in a drinking water pipe. GAC biofilters are effective in water purification specifically after disinfection by ozone. Organic substances present in water impair water quality by imparting odor and taste that are esthetically undesirable. Also, organic micropollutants and organic precursors responsible for disinfection by-products (DBP) formation are undesirable.

## 2.5 Biofouling Reduction Strategies

As stated in the previous section, biofouling strategies are biofouling control and biofouling prevention. Chemical cleaning of the membrane is the major biofouling control technique. The biofouling prevention technique potentially includes feed water pretreatment, RO membrane surface modification, and water disinfection.

### 2.5.1 Direct Methods

In 'direct methods', biofouling is controlled from the time when membrane modules are manufactured. The immediate method is to control biofouling in situ by applying cleaning chemicals to the membrane directly. RO membrane life can be enhanced, and operational costs can be minimized by following appropriate cleaning protocols.

### 2.5.2 Modification

As a biofouling prevention strategy, antifouling agents are incorporated into the membrane during the preparation stage. Thus, membrane modification improves its physicochemical properties, thereby reducing biofouling. The membrane properties to be improved for fouling prevention includes functional group, smoothness, charge, and hydrophobicity (Louie et al. 2006; Chae et al. 2009; Malaisamy et al. 2010; Miller et al. 2012).

By enhancing membrane surface properties, biomass adhesion and attachment can be minimized and the bacterial species becomes inactive. Rough membrane surfaces are prone to increase biomass attachment than smooth surfaces (Louie et al. 2006). Microbes in the aqueous phase possess negative charge and so negatively charged membranes repel each other and thus negative charge on a membrane surface helps in preventing biofouling (Hori and Matsumoto 2010). However, negatively charged foulants are attracted to the membrane surface.

The hydrophilic and hydrophobic nature of the membrane is also linked to the membrane fouling. Kwon et al. (2005) reported that the more the hydrophilicity of the membrane, the lesser the degree of biofouling. However, a negatively charged organic foulant can easily attach to the membrane. Membrane modification with a chemical surfactant is also a promising option. The bio-surfactants are alternative to the chemical surfactants, which are derived from renewable materials. Bio-surfactants possess less interfacial and surface tensions in both water phase and organic solutions. The use of bio-surfactants is beneficial because they are biodegradable in nature, environmentally friendly, compatible under very high temperature, pH, and saline conditions, less toxic, and have high form formation and selectivity (Desai and Banat 1997; Wilbert, Pellegrino, and Zydney 1998).

One of the limitations of using bio-surfactants is the high cost. Furthermore, in the beginning, microbes may not attach to the membrane surface in the presence of bio-surfactants but in the longer run, microbes develop adaptability to grow under hostile environmental conditions (Hori and Matsumoto 2010). To achieve the best biofouling prevention using bio-surfactants, feed water must be free from any living biomass.

### 2.5.3 Cleaning

The strategies of membrane cleaning are plentiful and normally remain proprietary. The literature report suggests that permeate flux can be restored by adopting a proper cleaning protocol (Madaeni and Mansourpanah 2004). Membrane cleaning is recommended when the transmembrane pressure increases by 10% or the flux value drops by 10%. The major parameters that affect the chemical cleaning performance are the type of chemical selected for cleaning and its concentration, duration of cleaning, pH, and temperature (Al-Amoudi and Farooque 2005). Fane (1997) reported that about 5%–20% of the operating cost component is consumed toward membrane cleaning in an RO desalination plant.

Biofilms are attached to the membrane surface and to disengage the biofilm, two main techniques are used.

1. Application of air scouring or high crossflow velocity (high shearing velocity)
2. Detachment of the biofilm employing proper chemicals (Fleming 2002)

As a physical cleaning strategy, backwashing and/or relaxation are the commonly used techniques. These methods are performed on a regular basis, but their effectiveness will be reduced by filtration time. When irreversible fouling is observed on the surface ,different intensities of chemical cleanings can be injected on a regular basis (weekly to yearly). In chemical cleaning techniques, chemicals inhibit the microbial bonding with the membrane surface and loosen them. Sodium hydroxide (NaOH) (Kim et al. 2011) and sodium hypochlorite (NaOCl) (Subramani and Hoek 2010) are generally used cleaning chemicals. NaOH demonstrated excellent performance removing 95% biofouling from membrane when cleaning was performed for 20 minutes. NaOCl (0.3%) is usually utilized as a major chemical agent in the microfiltration membrane processes to remove the organic foulants, while citric acid is normally used for inorganic foulants (Le-Clech 2010). However, chemical cleaning could not absolutely remove the attached biofilm, and fast regrowth of biomass was observed (Kim et al. 2009; Vrouwenvelder et al. 2003; Bereschenko et al. 2011). Moreover, the thin polyamide skin layer gets damaged with cleaning agents. Also, the production of less amount of pure water and disposal of chemical waste are other issues that need to be tackled (Khan et al. 2010; Kang et al. 2007).

### 2.5.4 Indirect Methods

MF/UF-based membrane filtration and biofiltration are used as indirect pretreatment techniques to RO feed. Those pretreatment methods ensure that organics, nutrients, particles, and microbes are physically separated from raw water. Sometimes sodium bisulfite-based biocides and NaOCl as disinfectants are used to destroy microbes.

### 2.5.5 Pretreatment

Pretreatment is crucial in SWRO because it reduces the biofouling occurrence and thereby improves RO desalination plant efficiency. To achieve this, suspended solids, organics, nutrients, minerals, bacteria, and trace organics are removed from the source water (Kumar and Sivanesan 2006). Thus, RO feed water quality is improved, and it becomes free from biomass. This minimizes bacterial tendency to attach to the RO membrane. RO feed water treatment can be done either by employing conventional physicochemical methods or by recently used membrane-based separation. Leparc et al. (2007) reported that dual media filtration (DMF) and cartridge filters could not completely reject source seawater contaminants. The advanced pretreatment is getting more acceptance and MF/UF membranes could achieve better removal of harmful impurities as compared to DMF. Furthermore, few chemicals are consumed and MF/UF-based membrane filtration techniques are robust. MF membranes are fabricated from polyvinylidene fluoride (PVDF) as they provide mechanical strength and resist chemical attack (Ding et al. 2013). The MF membrane rejects suspended and dissolved particles, but absolute elimination of bacteria is not obtained. The MF process operates around 2 bar pressure, which is usually lower than that of nanofiltration (NF) and UF (Sachit and Veenstra 2014).

#### *2.5.5.1 Deep Bed Biofilter (DBF)*

Deep bed filters behave like a biofilter when they are operated at low filtration rates and allow the formation of a biofilm on their surface. When GAC is used as a filter medium, the process begins with sorption of organic molecules onto the filter media followed by enzymatic hydrolysis (enzymes secreted from bacteria are attached to the biofilm) of bigger molecules to the tiny fractions. Those small fractions then transport to the biofilm, which further metabolizes the biodegradable organics and consumes as a substrate from the feed water (Hu et al. 2005; Larsen and Harremoës 1994).

Naidu et al. (2013) examined the granular activated carbon (GAC) biofilter performance in biomass adhesion on its surface when treating seawater. Biomass activity was measured in terms of ATP and the bacterial population was expressed in CFU. The biomass accumulation and DOC removal were correlated. The authors reported that within 20 days of experimental operation, a high amount of bacterial mass (1.0 × 108 CFU/g media) was deposited on the top surface of the biofilter. They observed that with decreasing thickness of filter media, biomass accumulation was reduced. Moreover, in the early phase of the study, the microbial concentration was 0.9 ± 0.5 μg ATP/g media within 0–5 days period and after reaching the steady state within 15–20 days, it increased to 51.0 ± 11.8 μg ATP/g media and the filter produced good quality of treated water (the DOC concentrations were 0.51 ± 0.12 mg/L). It was reported that compared to sand filter and anthracite, the GAC performed better and this can be attributed to the high porosity of GAC capable of providing more surface area and accumulation of higher biomass (Wang, Summers, and Miltner 1995).

In another study, Jeong et al. (2013) evaluated the performance of GAC and anthracite biofilter in seawater desalination. Terminal restriction fragment length polymorphism (T-RFLP), principal component analysis (PCA), and 16S rRNA gene sequencing techniques were used for bacterial consortia analysis. The authors deduced that the GAC biofilter captured diverse heterotrophs and outperformed the anthracite filter during 75 days of operation. High AOC removal was linked to the abundance of the microbial community on GAC. When the process was in the early phase of the operation, effluent AOC concentration was high (18.0 ± 1.4 μg-C glucose/L). After attaining a steady state within a 15–20 days period, AOC in the effluent was low (0.6 ± 0.2 μg-C glucose/L). The high AOC in the early phase was linked to the higher molar mass of the organics, which assimilated to the low molar mass fractions. Once the steady state is attained, the specific microbial consortia proliferated over time and metabolized the low molecular weight organic reducing AOC in the permeate (Naidu et al. 2013). On the other hand, the anthracite biofilter was selective for sulfur-oxidizing and reducing bacteria. This can be attributed to the sulfur availability in the anthracite as a contaminant.

Though biofilter is an attractive pretreatment alternative, it has some limitations. When biofilter is put in service, it requires some time for acclimation. In this acclimation phase, certain microbes and nutrients pass through the biofilter and form a colony on the membrane surface. Similar phenomena were observed during the backwashing cycle (Sadr Ghayeni et al. 1998; Chua, Hawlader, and Malek 2003). Furthermore, for a biofilter to operate efficiently, some vital factors need to be considered such as filter media, feed flowrate, and cleaning cycles.

#### *2.5.5.2 Membrane Filtration*

Membrane filtration is another effective pretreatment since commercial membrane produces high water throughput and is cheaper. Furthermore, the small footprint of the membrane plant and few inventory requirements are the benefits of using membrane pretreatment.

The microbes can be rejected by employing membrane filtration and researchers suggested that this bacterial removal mechanism is the combination of two processes: (i) the effect of physio-chemical interactions between the membrane and microorganisms and (ii) the sieving effect (Košutić and Kunst 2002; Van der Bruggen et al. 1999). A membrane typically rejects the larger diameter of bacteria from passing through it. Also, the negatively charged membrane and microbes repel each other.

The membrane pretreatment potentially rejects nutrients from the raw water and thus prevents biofilm growth on the downstream RO system. As such, microbes do not receive enough nutrients from the feed. This malnourishment condition adversely affects reproduction and proper growth of microbes resulting in a thinner and unevenly distributed biofilm with less biofouling potential (Al-Juboori and Yusaf 2012; Flemming et al. 1997).

### 2.5.6 Biochemical Methods

Biochemical techniques such as bacteriophage, signaling molecules, and enzymes are employed to remove a rigidly attached biofilm on the surface (Flemming 2011). Bacteriophages are viruses that kill bacteria (Fu et al. 2010). The recently invented quorum sensing method uses extremely specific signaling biomolecules, which deliberately destroy cell–cell communication in microbes of the biofouling layer (Davies and Marques 2009). Although quorum sensing is a very promising biofouling removal method, it suffers from certain downsides such as high cost associated to process such biochemical molecules on a commercial basis and lack of consistency and efficacy in removing attached biomass using such methods (Richards and Cloete 2010; Flemming 2011).

### 2.5.7 Water Disinfection Method

Introducing disinfectants prior to the RO membrane is proved to be a very efficient pretreatment technique that simply prevents bacterial bonding onto the membrane surface (Hori and Matsumoto 2010). Disinfection methods include both chemical and thermal means as a conventional method and by application of such pretreatment, microbes are destroyed at the source and thus prevent them from reaching out to the membrane surface. Nonconventional treatments such as ultraviolet (UV) light treatment, mechanical treatment, and ultrasound treatment are also being employed.

Chemical pretreatment employs chemicals such as hypochlorite ($Cl_2$ based) and ozone molecules, which are generated in situ using an ozonizer. These disinfectants have a very wide potential to destroy microbes and are cheaper though the formation of disinfection by-products (DBPs) is a limitation with this process.

In this regard, solar energy (Davies et al. 2009) is considered another attractive alternative to the chemical pretreatment. It is one of the most promising economical pretreatments that suffers from limitations of low efficiency due to varying topographical and weather conditions. Specifically, during nighttime, solar energy is not available, so solar system efficiency falls to zero and storage is not much viable alternative techno-economically (Davies et al. 2009).

UV light source is also used as a disinfectant technique (Schwartz, Hoffmann, and Obst 2003). The high cost of a UV lamp, energy consumption, DBP formation are some of the issues associated with the application of UV light. Furthermore, water characteristics such as turbidity and color adversely affect UV light performance due to absorption and Tyndall effects in the aqueous phase (Harris et al. 1987; Parker and Darby 1995).

Ultrasound has emerged as an attractive ecofriendly pretreatment option. Ultrasound has the ability to destroy the microbes and detach the biomass from the membrane surface (Gogate and Kabadi 2009; Joyce et al. 2003).

## 2.6 Detection of Fouling

The most significant factor that contributes to the decline in RO performance in the desalination process is the fouling of the membranes caused by adsorption and accumulation of particulate and organic foulants into the pores and onto the membrane surface (Inaba et al. 2017). Fouling impedes the effectiveness and throughput of RO by declining water flux, membrane selectivity, and permeate quality (Luo et al. 2018; Ng and Elimelech 2004). The root cause and major fouling potential to the membrane process are feed water characteristics (Vrouwenvelder et al. 2003) though operating parameters such as operating flux and recovery are also contributing factors (Chen et al. 2004).

### 2.6.1 Organic Matter (OM) in Seawater

Organic contaminants of the feed water lead to the organic fouling, which combines with other foulings such as colloidal and biofouling to contribute to the overall fouling in the system. Biofouling is seen as a living form of organic fouling and organic matters are believed to be a non-living form of biofouling resulting from bacterial metabolisms and its cellular fractions (Amy 2008) (Table 2.1).

**TABLE 2.1**

Various OM Measurements and Characterization of Feed Water Samples

| Measurement Category | Protocol |
|---|---|
| Molecular weight (MW) distribution by size exclusion chromatography with online DOC detection (SEC–DOC) | OM in terms of chromatographic peaks corresponding to high molecular weight (MW) polysaccharides (PS), medium MW humic substances (HS) consisting of humic and fulvic acids, and low MW acids (LMA); this technique is conceptually equivalent to LC–OCD, liquid chromatography with organic carbon detection |
| Hydrophobic/transphilic/hydrophilic (HPO/TPI/HPI) DOC distribution | XAD-8/XAD-4 resin adsorption chromatography, revealing a polarity distribution of OM |
| 3-Dimensional spectra fluorescence excitation–emission matrix (3D-FEEM) | Distinguishing between humic-like and protein-like OM as well as providing a fluorescence index (FI) that is related to the OM source |

*Source:* Modified from Amy (2008).

### 2.6.2 Parameters Characterizing Biomass

Total direct cell count (TDC), adenosine 5′-triphosphate (ATP), and heterotrophic plate count (HPC) are significant parameters to measure biomass (Vrouwenvelder and van der Kooij 2001). The epifluorescence microscopy is used for TDC measurements with different dyes such as SYTO, acridine orange, and 4-6-diamidino-2-phenylindole (DAPI). The downside is that those dyes unfortunately stain the whole microbial communities available in the specimen. In other words, it stains both living and non-living cells. On the contrary, ATP is a more reliable, rapid, and easy method of biomass measurement that only considers and senses living cells. The active cells can be determined by light production through the enzymatic process by means of luciferin and firefly luciferase. The ATP amount and light produced had a linear relationship that determines ATP concentration. In the HPC analysis, samples are kept at 20°C or 28°C for an incubation period of 5–7 days on R2A plates to acquire heterotrophic bacterial cell counts in the mixed liquor. However, all the above-mentioned methods have limitations in assessing biomass when cells are in the cluster form. In this regard, still ATP is a more reliable, precise, and distinct method. Certainly, biomass detection in feed water and on the membrane employs a combination of ATP and TDC (Vrouwenvelder et al. 2008).

The choice of the pretreatment process largely depends on the quantity and the diversity of bacterial consortia (Schneider et al. 2005). A wide and diverse bacterial community is reported in saline water. It has been reported that certain groups of bacteria or dominant species are responsible for high molecular weight organics concentration or SMP (polysaccharides/protein) secretion (Frias-Lopez et al. 2002; Cottrell and Kirchman 2000).

### 2.6.3 Fouling Potential of Water

#### *2.6.3.1 Particulate Fouling Potential*

To measure and detect particulate fouling in feed water and on the membrane surface, a suitable fouling detection technique is essential (Boerlage et al. 2003). Particulate fouling details are useful during the design of the entire plant specifically for pretreatment. Also, this is significant to monitor plant performance and efficiency.

The particulate matters fouling can be measured and indicated by the silt density index (SDI) and modified fouling index (MFI). In the beginning, MFI with 0.45 μm filter media was employed for particulate matters measurement in feed water. Later, Moueddeb, Jaouen, and Schlumpf (1996) pointed out shortcomings of this method and then Boerlage et al. (2003) established a novel UF-MFI. UF-MFI is a promising technique in fouling characterization for a certain source of feed water and records any variation in RO feed water characteristics (Boerlage et al. 2003).

#### *2.6.3.2 Extracellular Polymeric Substances*

The amount of SMP and EPS significantly affects biofouling. Both are heterogeneous in nature and consist of a variety of organics mainly polysaccharides, proteins, humic acid, glycolipids, and deoxyribonucleic acid (DNA) (Wang et al. 2014). Thus, membrane biofouling is a dynamic and slow process revealed by an addition of self-originated microbial cells to the membrane surface by glue-like, autogenic soluble microbial products (Inaba et al. 2017). EPS originated from biomass are the main component that contributes to biofouling and causes membrane permeability decline with time (Wang et al. 2016).

Berman (2010) deduced that transparent exopolymer particles (TEP) are a major contributor to the biofouling on a RO membrane. The TEP role is very vital in bacterial growth and this gluelike TEP layer helps in biofilm formation over the RO membrane. In another report, it has been reported that about 68% of the total microbial community were attached to the TEP component and this TEP measurement and monitoring is very useful to understand RO fouling phenomena (Villacorte et al. 2009).

#### *2.6.3.3 Biofouling Potential*

The omnipresence of bacteria and the amount of nutrients actually decide the growth of biomass and thus biofilm formation on the membrane surface. The biofilm formation growth depends on various operating parameters such as shear velocity, nutrient type (N, P, K), concentration, and robustness of the biofilm attached to the surface (Flemming 1997). The cleaning chemicals usage such as biocides and disinfectant quality directly depends on the biofouling detection (Vrouwenvelder et al. 2000; Vrouwenvelder, van Loosdrecht, and Kruithof 2011; Vrouwenvelder et al. 2007).

The biodegradable organic matter (BOM) is measured as assimilable Organic Carbon (AOC) and biodegradable dissolved organic carbon (BDOC) and the BOM is a limiting constituent for biomass growth (LeChevallier, Schulz, and Lee 1991). The AOC component of BOM is linked to the microbial population in feed water (Hambsch and Werner 1996; Weinrich, Schneider, and LeChevallier 2011) and it represents low molecular weight compounds such as acetic acids and amino acids.

#### *2.6.3.4 Assimilable Organic Carbon (AOC)*

The assimilable organic carbon (AOC) is typically 0.1%–9.0% of the TOC, which can easily be taken up by microbes for their metabolism and growth. This biomass growth is measured by the colony count in AOC analysis. For this, a standard concentration plot is prepared to show microbial growth yield and assimilable organic concentration. The growth monitored during the incubation is converted to the AOC from the standard curve. van der Kooij in 1992 reported that when AOC was <10 μg/L, the heterotrophic bacterial growth was limited. Based on van der Kooij's (1992) concept, many analytical techniques were developed to estimate AOC. Table 2.2 presents the representative AOC analysis techniques. Furthermore, some of these methods deviate from adapting natural bacterial consortia (Hammes and Egli 2005; Kaplan, Bott, and Reasoner 1993), rather than using pure cultures. Moreover, a majority of AOC available uses various growth measuring methods such as plating, ATP, turbidity, flow cytometry, and luminescence. The current research efforts are aiming at user-friendly and rapid AOC detection methods development.

Jeong et al. (2013) introduced a novel AOC measurement technique called the *Vibrio fischeri* method. In this method, frozen *V. fischeri* stock is allowed for incubation for a short time in seawater. Glucose is used as a carbon supply. The luminescence meter measures natural bioluminescence after the incubation period at 25°C wherein marine agar plate is employed for strain preparation. The distinctive feature of this method is the very less (30 minutes) incubation time due to the directly used marine agar plate for strain preparation. Moreover, *V. fischeri* strain outperformed the previously used *V. harvey* strain for AOC detection due to its good correlation with cell number and luminescence. Furthermore, as *V. fischeri* strain was derived from seawater, it can therefore be well adopted to high TDS concentration of source saline water. The luminescence detection approach is also favorable to flow cytometry as luminescence detection is much easier. Flow cytometry suffers from the limitation of detecting too small cell counts (<$10^2$ cells/mL).

#### *2.6.3.5 Biodegradable Organic Carbon (BDOC)*

The portion of available dissolved organic carbon (DOC) of the feed, which is easily metabolized by heterotrophic bacteria, is biodegradable dissolved organic carbon (BDOC) (Servais, Billen, and Hascoët 1987). The BDOC is

**TABLE 2.2**

Representative AOC Methods Available in the Literature

| Methods | Target (volume, mL) | Culture | Incubation Time (days/h) | Cell Counts | Substrate | Detection Limits | Ref. |
|---|---|---|---|---|---|---|---|
| Van der | Drinking | *Pseudomonas fluorescens* | 7–9 | Nutrient agar | Acetate | 10 μg/L | van der Kooij et al. (1982) |
| Eawag | Tap water | Precultured natural | 2–3 | Flow cytometer | Acetate | 10 μg/L | Hammes and Egli (2005) |
| *V. harvey* | Saline water | *V. harvey* | <1 h | Luminescence | Acetate | <10 μg/L | Weinrich et al. (2011) |
| *V. fischeri* | Seawater | *V. fischeri* | <1 h | Luminescence | Glucose | 0.1 μg-C | Jeong et al. (2013a) |

*Source:* Modified from Jeong et al. (2019).

obtained by subtracting the initial DOC from the final DOC detected after 28 days of incubation. For incubation, the inoculum chosen is simply environmental microbes. The incubation happens under suspended and attached growth conditions. For the attached growth mechanism, sand or porous bed supports are provided. During suspended incubation, 28 or 5–7 days are recommended time for bacteria attached to the sand support. The BDOC detection is a biodegradability indication parameter generally used in water treatment. Nevertheless, van der Kooij (1992) deduced that due to lack of proper correlation between bacteria and BDOC concentration, BDOC is not reliable in accurately predicting bacterial regrowth, and due to its extremely low detection limit (0.1 mg/L), it also measures AOC concentration (van der Kooij 1992).

The Biomass Production Potential (BPP) test is performed when the biodegradable chemical of a water sample cannot be consumed by the AOC test. BPP can detect maximum ATP of the microbial community of the water under 25°C incubation. BPP is measured in terms of ATPmax/mg product or liter of water (Vrouwenvelder et al. 2000). Another such parameter, the biofilm formation rate (BFR), is expressed as pgATPcm$^2$/day, which is nothing but biotic bacteria (ATP) deposition on the glass ring surface measured by an online biofilm monitor (van der Kooij et al. 1995).

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# Polymetallic Coatings to Control Biofouling in Pipelines

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## Challenges and Potential

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CRC Press
Taylor & Francis Group
Boca Raton London New York

CRC Press is an imprint of the
Taylor & Francis Group, an **informa** business

First edition published 2022
by CRC Press
6000 Broken Sound Parkway NW, Suite 300, Boca Raton, FL 33487-2742

and by CRC Press
2 Park Square, Milton Park, Abingdon, Oxon, OX14 4RN



CRC Press is an imprint of Taylor & Francis Group, LLC



ISBN: 9781032044897 (hbk)
ISBN: 9781032044903 (pbk)
ISBN: 9781003193449 (ebk)

Typeset in Palatino
by codeMantra

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# *Preface*

Biofouling is the major concern in most of the industries. Fouling-released coating with a special emphasis on pipeline industries is the focus of this book.

In oil and gas/fuel pipelines and storage tanks, biofilms cause significant operational problems due to microbial invasion, which leads to reduction of flow, souring, and reservoir plugging, thereby enhancing the corrosion of the bacterial adhered surface. With the constant use of biofuels, the study of the corrosive processes associated with microorganisms has also gained significance. Carbon steel (CS) or stainless steel is principally used for the transport of these materials because it is efficient and cost-effective. The major problems faced by these materials are the aggressive environment of fouling attack. Therefore, the study of protecting these pipeline surfaces with a protective coat is therefore quite significant. The surfaces of the pipeline colonize with the biofilm and form the complex microbial structure such as extracellular polymeric substance (EPS). The most abundant bacterial species, which are involved in the internal corrosion of the pipelines, interact with molecular hydrogen present on the surfaces of pipes and produce hydrogen sulfide as a by-product of metabolism. This process breaks down the iron and steel of even heavy-walled pipes, resulting in leaks and catastrophic pipeline failure, and thus reduces the quality of oil and gas/biofuel pipelines and storage tanks, therefore causing great financial losses worldwide. Surface modification of metallic materials by thin-film technology has achieved a considerable breakthrough to enhance its properties. Coatings on the surface provide a barrier between the surface and the environment, and provide fouling resistance to the surface. Various techniques have been used for surface modification such as chemical vapor deposition (CVD), physical vapor deposition (PVD), electrochemical and electroless plating. CVD and PVD techniques are relatively expensive and sophisticated. The choice for metallic coatings in recent years has emerged due to their low cost, fast deposition, good filling capability, good uniformity, and low processing temperature. These metallic coatings have been used extensively in the aerospace, automotive, and chemical processing industries in the past decade. It is the easiest method to deposit metallic films on arbitrary shapes with uniform thickness. It is a good choice of metal coating for oil and gas/biofuel pipelines and storage tanks, because of its excellent corrosion resistance and wear resistance when used as a barrier layer. It also has the benefit of an adjustable thickness of coating, good cohesion, and performance. Metal nanoparticles are known to exhibit enhanced physical and chemical properties when compared to their bulk counterparts because of their high surface-to-volume ratios. Copper (Cu), zinc (Zn), silver (Ag), nickel (Ni), etc. are known to have excellent toxicity for fouling organisms and thereby provide good resistance to biofouling. The fouling resistance is mostly due to toxic metallic ions on the surface, making it inhospitable to most organisms by blocking the respiratory enzyme system of these microorganisms in addition to damaging microbial DNA and the cell wall. Various surface characterization studies are there to evaluate the nanocrystalline nature of the film such as X-ray surface analysis (XRD), Field Emission Scanning Electron Microscopy (FESEM), Atomic Force Microscopy (AFM), thickness and contact angle measurement, corrosion behavior to distinguish the crystalline nature, morphology, chemical composition, and surface topography of the coating surface. The mechanism of corrosion resistance/improvement in the deposited layer should be studied by electrochemical techniques. Raman spectroscopy is used to characterize the corrosion deposits on the metal surface to find out the iron oxide phases and their transformation. Leaching of deposited materials study should be performed by using Inductively Coupled Plasma Mass Spectrometry (ICPMS)/Atomic Absorption Spectroscopy (AAS). The microbial culture methods such as isolation and identification of microbes are used to identify the biofilm and corrosion-causing bacteria, which are significant in diversity study and find the variation in the bacterial community.

The book contains eight chapters. The first part of the book discusses on biofouling mechanisms, assessment, and reduction strategies followed by biofouling in oil and gas/biofuel pipelines. The second part consists of surface coating methods to prevent the fouling and deterioration of the surface of pipes. The third part includes the ethical issues and environmental safety of coating and prospects of coating technologies.

Biofouling is the major concern in most of the industries. Fouling-released coating with special emphasis on pipeline industries is the better prospective for future direction research.

This book will be useful to learn the interdisciplinary skill for Chemical Engineering, Mechanical Engineering, and Biotechnology undergraduate students.

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