Intercepting signalling mechanism to control environmental biofouling

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Abstract

Biofouling in environmental systems employs bacterial quorum sensing signals (autoinducers) and extracellular polymeric substances to onset the event. The present review has highlighted on the fundamental mechanisms behind biofilm formation over broad spectrum environmental niches especially membrane biofouling in water systems and consequent chances of pathogenic contamination leading to global economic loss. It has broadly discussed on bioelectrical signal (via, potassium gradient) and molecular signal (via, AHLs) mediated quorum sensing which help to propagate biofilm formation. The review has illustrated the potential of genomic intervention towards biofouled membrane microbial community and has uncovered possible features of biofilm microenvironment like quorum quenching bacteria, bioelectrical waves capture, siderophores arrest and surface modifications. Based on information, the concept of interception of quorum signals (AHLs) and bioelectrical signals (K^+) by employing electro-modified (negative charges) membrane surface have been hypothesized in the present review to favour anti-biofouling.

Keywords Membrane biofouling · Quorum sensing · Bioelectrical wave · Siderophore · Genomic

Introduction

Biofouling is an emerging and trouble-causing natural event which rapidly occurs in liquid–solid (biotic as well as abiotic surface) contact. In fact, the phenomenon itself manifests limelight not only in recent research interest but also in huge economic and ecological issues at the global scale (Jiang et al. 2017). Bacteria get readily attached to the moisture contacts, especially on submerged platform and by a complex successional process they architect a 3D dynamic heterogenic biolayer called biofilm. Later on, this

primary biofilm provides a hotspot for opportunistic micro- and macro-foulers to aggregate (Stover et al. 2000), in due courses of biofouling. Ranging from medical equipment (protein fouling) contamination events (Dancer 2014) to surface damage events, biosensor, milk, food, ship industries (Liu et al. 2014; Rosenhahn et al. 2010) and especially water treatment industry (Chiellini et al. 2012) have been drastically facing economic loss for biofouling (Kristensen et al. 2008; Chen et al. 2013; Das et al. 2013). Eventually, the impact of biofouling is considerably vast and diverse according to technology and processing. For example, in milk industry, biofouling causes reduction in quality and quantity of milk, whereas in water treatment system (Douterelo et al. 2017), it may infer membrane fouling (Chede et al. 2017) and subsequent higher energy consumption, pressure drop, flux decline and also maximises the chance of epidemic break through pathological contamination in pipeline (Liu et al. 2015; Yang et al. 2016). Even ship industries face biofouling issue, which comprises not only the physical damages, metal corrosion of ship hull, yard platform, reduced heat transfer efficiency, increased drag force and energy consumption, but it also introduces ‘alien’ species from prior destinations (Chien et al. 2012). Considering the global loss, studying biofouling has become a spin point

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of novel and multidisciplinary research since late nineties (Lindner 1992).

Research interest about biofouling problem is increasing day by day as shown in Fig. 1 using polynomial model (Jiang et al. 2017). However, if the collective number of last 10 years of publication regarding emerging problems of biofouling is taken, the biological insights about biofouling is very less with R^2 value of 0.339 in polynomial model (Fig. 1) and that unsaturated area can become a “thrust topic” for biological investment in the coming future.

The dynamic complexity and structural arrangements of biofilms on natural as well as artificial biotic or abiotic surfaces have been entertained in the last two decades and the recent excellency in sophisticated microscopic imaging (Powell et al. 2017), molecular approaches and high-throughput sequencing platform have added more research value to the subject. Eventually with the help of high-throughput genomic tools like metagenomics, metaproteomics, whole genome annotation, microarray and microchip, a large amount of data can be generated and dig down to come out with new ideas to combat biofouling issues (Davey and O’toole 2000). In this paper, the necessity and scope of using genomic tools to design novel ideas of antifouling strategies have been delivered.

Biofouling: signalling privilege of bacteria

Biofilm formation process has historical records in fossil and living fossil morphology, particularly in hydrothermal niches. Putative evidences came from 3.3 to 3.4 billion years

old Kornberg formation (Rasmussen 2000), and 3.2 billion years aged deep ocean hydrothermal rock (Kiyokawa et al. 2006) of Australia (filamentous structure). Interestingly, bio-matrix can be noted in recent sea rocks, hot spring or hydrothermal vents (Javaux et al. 2010). The extreme conditions of primitive earth triggered the rise of complex architecture of signal interaction and biochemical pathways which thrive under bulk stress and that the genome information have been taken throughout to understand different periods of evolutionary adaptations (Hall-Stoodley et al. 2004; Koo et al. 2017). Harsh temperature, pH, oceanic gyre and ultraviolet (UV) exposure stimulated the individual behaviour of microorganisms to change into synchronized population by cell–cell interaction and maintain the mass balance by turning over of the generated waste by biofilm microcolonies structured with open water channels. Thus, environmental catalytic factors offered by life always play a critical attribute in making a population sessile or planktonic or opportunistically sessile (Koo et al. 2017).

Molecules and mechanism of cell signalling in biofilms

Biofouling occurs when there is a natural or artificial surface in contact with liquid. The surface easily gets conditioned by drifting protein, organic and inorganic scaling particles coming from surrounding water stream, which leads to reversible adsorption and irreversible adhesion (Wahl et al. 2012). Reversible attachment is mainly due to physical parameters of surrounding liquid, like dragging force, Brownian motion, Reynolds number, velocity of fluid, Van der Waals force

Fig. 1 Graphical polynomial model presentation of last 10 years publication regarding membrane biofouling and biological insight into the phenomenon

and electrostatic charged particle interaction (Grobblor et al. 2015). The irreversible adhesion forms a permanent bonding of biolayers with the substratum through favourable microorganisms (mainly Proteobacteria) and their secreted sticky extracellular polymeric substances (EPS), which fill the gaps between cells and provide a basic matrix of biofilm network (Ghosh et al. 2017a, b; Nagaraja et al. 2017). Other populations including opportunistic bacteria, cyanobacteria, diatoms, protozoa and fungi then take part in the formation of the successful biofilm network. Although the cell percentage in a biofilm community is about 15–20% only, yet the composition of biofouled stratum has been highly diverse in nature (Table 1) and depends upon subjected surface material (biotic and abiotic), environmental conditions like temperature, pH, nutrient load, frictional factors and seasonal blooming conditions (Bengtsson et al. 2010; Qureshi et al. 2015). In marine biofouling, generally diatoms occupy the major percentage whereas in fresh water bacteria dominates (Kriwi and Uthicke 2011).

Literature reports have shown that signalling pattern plays the leading role in initiating and controlling the biofilm through quorum sensing (Nagaraja et al. 2017; Wang et al. 2018). Bacteria are known to use some specific auto-inducer molecules to generate signalling profile, which in turn synchronize social behaviour of the population like biofilm formation, antibiotic resistivity, drug production, xenobiotic degradation, flexible metabolism (Soumya et al. 2017), pigment generation, and bioluminescence (Ghosh et al. 2017a, b). Fungal quorum sensing study related to biofilm, in context of protein networking, has been majorly initiated in 2012 (Nobile et al. 2012; Wang et al. 2018), where authors identified and described signalling molecules. However, for bacterial system, these autoinducers were typically but not limited to acyl homoserine lactones with a varying carbon chain length of 4–12, which were produced from cysteine–methionine metabolism pathway (Anbazhagan et al. 2012). Apart from that, autoinducer 2 (AI2) box also can be found in many cases, where furanosyl borate diester controls the signalling mostly (Deep et al. 2011; Nobile et al. 2012). AI2 is produced by the phosphorylation of DPD ((S)-4,5-dihydroxy-2,3-pentanedione) at carbon 5 position by LsrK and readily gets accessed by response receiver proteins.

In eukaryotic biofilm, environmental signals and ‘cues’ play the major role. Researchers from University of Birmingham, UK (2002) have showed that Enteromorpha, which is globally known as sticky microfouling algae found typically in intertidal zones becomes a root problem of marine biofouling and it uses the mechanism of ‘first kissing’ to be attached with surfaces. Depending upon the environmental biotic and abiotic cues, they sense the matrix and swim (Zhao et al. 2018). Eventually upon reaching the suitable stratum, they make irreversible bonding with surface by

dropping down their motile part, become sessile and produce strong adhesive for the commitment. Hence, signal control and circuit disruptions can change the biofilm community, which in progress develops to biocorrosion.

Bioelectrical wave signalling favours biofouling process

Recently, Humphries et al. (2017) have reported signalling through bioelectrical wave and opened a new paradigm in biofilm signalling by uncovering the role of hidden electrical transduction in cell to cell communication, apart from quorum sensing. They showed extracellular potassium gradient released by the biofilm can mediate electrical wave and that wave can be channelled through population and can alter the behaviour response of distant individual or emboli that are not the part of community.

In bioelectrical signalling, a relay of spatially channelled electrical wave occurs from extracellular potassium threshold that leads to positive feedback circulation in communities, where the functional pathways and channelling proteins get induced to release intracellular potassium that depolarise the neighbouring individual or emboli. The depolarizing wave propagates and coordinates the metabolic flexibility of interior as well as peripheral cells. The electrically mediated signals propagate the wave through the population and induce the distant bacteria to synchronize with the biofilm by altering their motility, cell surface chemical response, EPS charge and thereby increase the cumulative fitness of the biofilm (Majumdar and Pal 2017).

Biofouling: big business

Supervising biofouling has become a big business (Table 1). Many reports showed that US navy alone has to pay a charge of \$1 billion per annum for biofouling attack in the ship industry. Biofouling on ship hull can increase drag force upto 60% and fuel consumption upto 40% which leads to decrease in speed by 10% (Callow 1986). A review from University of Glasgow, UK highlighted that biofilm costs every year billions of pounds due to energy loss, pathogenic aggregation, biocorrosion and product contamination (ISAAC 2006). Biofouling significantly charges high cost in water management and pumping in any industry especially petroleum and desalination (Perkins et al. 2010). Even cases have been reported where the total pipeline had to be replaced in the systems for biocorrosion. US government had to face indefinite shutdown of US largest oilfield which was connected by 16 miles of Alaska pipeline due to the pipe biofouling problem. As a result, production of 4 lakh barrels/day has to be suspended and oil price increased by a cost equal to 8% of US petroleum outcome. Pipeline biofilm is a very much common and serious issue at the industrial sector (Li 2013). The traditional pipeline cleaning techniques for

Table 1 Ubiquitous nature of biofouling in different ecological niches and damages offered by the problem

Events	Causes	Damage (economic + operational)	Pictorial information in web	References
US navy ship and ship-hull biofouling	Slime produced by marine bacteria, fungi, algae, barnacles and other larva	\$1 billion per annum Increased drag force Bio-corrosion High energy		Hull Fouling Control: The Era of Innovation, June 23, 2013 by FathomShipping Callow et al (2002)
UK navy-ship fuel burning	Clogging by marine bacteria, fungi, algae, barnacles and other larva	Billions of pounds per annum 18% higher fuel utilization 5% of UK CO ₂ emission		yachting-pages.com-2016 University of Glasgow 2006
Aquaculture industry (fishing net and fish pathogen)	Riverine and marine bacteria, fungi, algae, barnacles and larva	5–10% of production cost Weight of the netting by as much as two-hundred-fold, and the drag force fivefold Shut-down of aquaculture structure	 (12 weeks of immersion)	https://phys.org/news/2013-09 Dobretsov et al (2013)
Alaska Pipeline biocorrosion	Majorly bacteria and fungi	Suspension of 4 lakh barrels/day production Cost equal to 8% of US petroleum		process-worldwide.com, 10/04/2011 Gomez-Alvarez et al (2012)
Darkening dome of the Jefferson Memorial (Washington)	black patchy biofilm majorly dominated by bacteria, fungi and algae	Turned into mottled mess in only 10 years \$12.4 million repair project started up but found to be less effective		washingtonpost.com/ 2016/08/09/33be9040-5 nydailynews.comarticle-1.2746702
Membrane biofouling	Majorly bacteria and fungi	7% cost only for membrane maintenance in desalination plant If damaged it can include other consumable (26%), electricity (41%)		scribd.com/doc/10/Energy-Water-and-Land-Use-7/2009) Photo courtesy Chiellini et al (2012)
Hospital acquired pathogenic biofilm (UK)	Pathogenic bacteria and fungi	1 in 20 UK people (approx. number 300000) suffer from HAI (hospital acquired infection) Bacteremia (blood) 6.2%, urinary tract 23.2%, surgical wound 10.7%, lower respiratory tract 22.9%, skin 9.6%, others 27.4% 10% pathogenicity reduction can save £93.1million per annum		http://www.wales.nhs.uk , Nao 2014) Canadian union of public employee, 2014

CIP (cleaning in place) have been found to be usually ineffective, superficially partial and unable to remove microbes from the inner layers of the biofilm, as reported by Liu et al. (2014). However, they also reported that use of enzymatic cleaning (*N*-acetylmuramide glycanohydrolase) under optimal condition (pH 10 ± 1 , temperature 45 ± 2) in circulating water could arrest pipeline biofilm. However, pipeline biofilm has remained as a great challenge in field level, even when it was reported in the industries where antimicrobial and germicidal surgery grade povidone were being manufactured. A research from UK showed that 1 in 20 UK people (approx. number 300,000) suffer from HAI (hospital-acquired infection) that come from pathogenic biofilm (Dancer 2014, <http://www.bbc.com/news/health-27046990>). Medical biofilm majorly attacks mucosal surface inside the body or on implanted devices like catheters, pacemakers and artificial heart valves (Anbazhagan et al. 2012; Percival et al. 2015). It was found that 10% reduction in biofilm pathogenicity can save £93.1 million per annum.

Membrane biofouling: issues and challenges

Biofouling remains an unavoidable challenge for membrane systems (Li et al. 2016). Worldwide increase of water demand has led to two considerable and sustainable solutions, which are desalination and wastewater reclamation. In 2009, more than 13,000 desalination plants worldwide was producing more than 12 billion gallons of fresh water a day and the market is expected to be double within the next 10 years that is up to 2019. According to the hydropolitic academy report (2016), more than 16,000 desalination plants produce more than 66.5 million cubic/day fresh water and the current number of total desalination plants is more than 19,000 globally (Desaldata 2017, <https://www.desaldata.com/>), in which at present reverse osmosis technology is said to be predominant in the market. The reverse osmosis membrane (RO) technology is esteemed to earn a market prize of USD 5.00 billion by 2021 globally with uprise in the compound annual growth rate (CAGR) of 15.75% from 2016 to 2021. By 2021, at a CAGR of 15.75%, RO market is projected to reach USD 5.00 billion between 2016 and 2021 (<https://www.businesswire.com/news/home/20170926006593/en/Global-Reverse-Osmosis-RO-Systems-Market-2017-2022>). In terms of value addition due to increased concerns over safe drinking water and stringent regulations on potable water, high-growth application of sea water desalination systems are on count and drive the growth of the RO market. Market reports showed that Asia-Pacific zone is the fastest growing membrane market (2015–2025) and the reasons behind it are, (1) growing water crisis from the application sectors, like water and wastewater treatment, medical and pharmaceutical, food and beverage, paper and dyes

in major countries of the region, especially in China and India (2) increasing investment pull in the membrane market (<https://www.techsciresearch.com/report/india-water-purifiers-market-by-technology-uv-ro-gravity-by-sales-channel-direct-vs-retail-by-region-competition-forecast-andopportunities/1247.html>).

The major limitation associated with membrane technology is fouling (Nguyen et al. 2012; Yoon et al. 2013), which includes subsequent higher energy consumption, pressure drop, pore blockage, flux decline and also maximises the chance of epidemic break through pathological contamination (Kim et al. 2013). One of the main drawbacks of the RO membrane is the biofouling problem, which in present scenario stands as the top challenge because, although many research teams have tried to overcome the problem and worked on fouling in general, too much research orientation in the past decades has been “wasted on studying it, describing it and publishing papers” (Logan 2017) but ultimately could not improvise towards fouling rates. As biofouling is the most practical problem faced by RO membrane operators, massive shutdown of desalination plant has been observed in the recent decade (especially Gulf of UAE) and the number of cases have been still increasing (Hess et al. 2017; Huang et al. 2013; Villacorte 2014).

Water flow under high pressurized condition in membrane thus allows a unique pattern of biofilm community in the membrane (Saeki et al. 2016). The rigid elasticity of biofilm mechanism of sessile community in the flowing channels elucidates the challenges to plan membrane antifouling strategies. The population may not behave like complete sessile, instead they somehow behave like partial floaters while attaching a head part of biofilm to the membrane filter substrate through adhesins, while the remaining tail part is left in drifting condition in open water channels in membrane fluid current. This typical adapting nature protects them from sudden increase of pressure drop or membrane flux.

Biofouling propagation issue: membrane to pathogens

Practically, the main pathological issue towards the membrane biofouling is the membrane leakage associated with polymer degradation by acidic byproducts secreted by residing bacteria and subsequent contamination of waterline. This drawback can lead to epidemic breakthrough of biofilm bacteria which are simultaneously multidrug resistant by default nature. Interestingly, environmental isolates also could act as opportunistic pathogens by the alteration of signalling cues of niches.

The multicellular topography of biofilm explains the perseverance of being attached to a solid surface with the head part, while at the same time being associated with the flow, by partially protruding the tail part. This flexibility

always remains a wonder in biofilm environment because very little information could be gained till now in context of mechanism or partial floating. Biochemical properties of EPS-based biomatrix uncovered showed that the slime associated with biofilm is practically hydrogel in nature, in spite of the connected nurturing environment, in vitro host or natural springs. This classical viscoelastic property of biofilm bed-morphology helps to repeal the stress factor or bears with the transient state when sudden increase in shearing occurs during context of seasonal current or tidal variation in marine, riverine environments and pumping/filtering activity in pipeline or membrane operations. However, during the transiently changing weather conditions or local flow current (elevated pressure of water in RO membrane), the hydroelasticity nature protects the biofilm community from shearing, but over long run of higher current or other elevated stress factors in the environment, hydrogel nature (supplementary Fig. 1) facilitates the dissipation of film layer through viscous elastic flow rather than detachment of individual cells of the community (Hibbs et al. 2016). Through this viscoelastic response and EPS modification, the film layer becomes streamlined with the flow of fluid and reduces the drag force or readily finds a new suitable hub point upon detachment from stationary membrane phase due to excessive fluid current (Fig. 2). This type of cue-response is not mutually exclusive depending on the surrounding environment, rather they are quite similar although the magnitude of response is highly variable between environmental biofilm and host in vitro pathogenic biofilm (Kusić et al. 2016).

The shear module factor, i.e., rigidity parameter G is about in the range of 10^1 – 10^3 Pa in in vitro biofilm and η (viscosity) is in the range of 10^5 – 10^8 , while the biofilms attached with natural hot spring biolayer has G value in the range of 10^3 – 10^5 Pa and viscosity is about 10^7 – 10^8 . Hence, viscoelasticity plays as a major option or adaptation component depending on earth to host conditions offered by the substratum (Koo et al. 2017). The biofilm pathogen invasion provides stability in the harsh environment by forming films and catalyzes the biofilm community growth through local synchronization of individual emboli, shields from challenges like UV exposure, acid leakage, metal toxicity, hydro scarcity, salt stress, avenging antibiotics or antimicrobial agents. Indeed, biofilms were likely antibiotic resistant in nature in both environment and in vitro host, but the feature has been more salient in pathogenic terms. Conceptually, the first barrier to the drug intrusion in localized cell cluster is the slime factor, which dilutes the effective concentration of antibiotic and renders it into a sublethal dose or neutralises the metal-charged particle (Swarupa et al. 2018), reactive oxygen or immunoglobulin. In addition, biofilm offers the production of extracellular beta-lactamase which cleaves the beta-lactam ring antibiotic targeting cell wall damage. During host attacks, other cell clustering pathogenesis differ from biofilm pathogenicity by the adherence capability of localized cell clumps with a substratum in the form of microcolonies aggregates (Flemming et al. 2016). Direct microscopy revealed that the biofilm-infected cells are encased in matrix and thus matrix-enclosed aggregates are resistant to antibiotics. Moreover, they can form biofilm ‘pockets’ in 3D

Fig. 2 Schematic representation showing role of EPS in two niches, viz, bio-slimes on biofouled membrane (environmental biofilm) and in host tissues (pathogenic biofilm)

clustering, where they arrest the drugs in the pockets and destroy them by secreting enzymes or diluting the effective concentration. Describing biofilm aetiology has been quite difficult (Davey and O'toole 2000), rather a massive localized inflammation and antibiotic resistant infection could indicate the presence of biofilm pathogen; however, the same drug might be effective in planktonic condition for the same case history.

Present scenario of membrane antifouling research

Considering the global loss, biofouling study has become a thrust area for research. Starting from physical attempts of bioinspired surface modifications to genetic-engineered circuits, a wide range of strategies were developed to understand and combat the problem. There were also a lot of trials and limitations in what to apply as antifouling strategies. Extensive care in feed pre-treatment, reactor designing, modification and development in membrane materials and different strategies in chemical cleaning were majorly established as membrane maintenance methods. Whittaker et al. (1984) evaluated the efficiency of five classes of chemical treatment for spirally wound cellulose acetate reverse osmosis membrane biofouling. They came to the conclusion that, in developing biofilm bacterial aggregation was the most contributing factor. Although the most common practices for membrane cleaning were periodic flushing of membrane with detergent and chelating agent, utilization of pH changer and antiprecipitant, however, in some cases strong bacteriocidal agents were not necessarily effective. It was shown that chlorine-inactivated bacteria could produce biofouling layers on RO. However, in such conditions enzyme-based treatment (papain-Triton-EDTA/protease-Triton-EDTA, pancreatin-Triton-EDTA etc) could give a more effective result, as they do not exhibit bacteriocidal activity, indeed they could actually remove the microbiota.

Biological treatments are gaining importance compared to other treatments (Xia et al. 2010). The use of chemical treatments are not promoted due to safety issues and bio-remedies for biofouling are being used as an eco-friendly, sustainable option for long-term membrane shelf life. Physical strategies actually detach the untied micro biolayer after cleaning while providing a good niche to form another layer very soon. Frequent chemical cleaning of membrane basically shortens the shelf life of membrane and includes extra cost for maintenance and extra man power (Matin et al. 2014).

Chemical means of control employ different types of biocides, which even in very low concentrations serve as broad spectrum toxins to target as well as non-target organisms and trigger higher probability for them to become resistant species. Applying biocides as antifouling agent basically gives stress factor to bacterial community and employ

them to produce more exopolysaccharides, which has been the principal element of biofilm formation. Hence, having alternative, safe, sustainable, eco-friendly control, and use of biological tools have been gaining attention rather than synthetic chemicals (Babu et al. 2016).

Biological-based strategies were reported to offer the advantages of lower toxic, highly efficient, more sustainable and less bacterial-resistant natural tools for controlling biofouling problems. Many reviews established that cell to cell signalling, which coordinates cell talking, conduct different features including expression of stress factors, biofilm formation, motility, antibiotic resistance and thus leads to a successful biofouling of membranes (Kalia and Purohit 2011; Malaeb et al. 2013; Weerasekara et al. 2016). Population driven by the quorum sensing (QS), which mostly involved acyl homoserine lactone (AHL) molecules for signalling in population, could switch the biofilm condition in aquatic environment in terms of controlling and replacing the biota (Hong et al. 2012; Wood et al. 2016). Hence, the use of quorum quenching (QQ) natural agents has become a promising solution to combat biofouling (Kalia 2013).

Due to the unavoidable nature of biofouling problem irrespective of field, the persistency of sticky nature of the phenomena has to be subjected to a wide range of integrated branches of research to produce solutions to this problem (Babu et al. 2016). Nguyen et al. (2012) have thoroughly described about the orientation of biofouling research, hence in this study, a time period of late 2012–2017 has been considered to discuss current biofouling status.

A wide range of model bacteria/fungi/algae/larva were subjected to understand the dynamic biofouling chemistry and to develop strategies for antifouling. Research over reactor-based mixed community and pilot scale applications have been reported, which highlighted the hidden behaviour of biofouling communities. Technology implementation process includes pre-treatment of resources, modification in reactor designing, use of nanoparticles, modification of surface chemistry, bioinspired mimicking, quorum control circuit, dispersal biomolecules and enzymes, etc.

Nanoparticle implementation on membranes

To address the challenges in context to membrane maintenance for quality-grade performance, various nanotechnology researches aligned consensus with each other to evolve nanotechniques to reduce membrane scaling and biofouling without any disturbances caused from operational point of view. Physical, chemical and biological events were manipulated by the introduction of nanoparticle in membrane coating (Chen et al. 2013). Ong et al. (2016) clarified the basic pros and cons of utilization of membrane surface modification through nanomaterials and nanoparticle implement.

They have summarized current challenges and highlighted the advanced technology in context of reliable pre-treatment and low biofouling scaling composites.

Das et al. (2013) developed and demonstrated protein-coated nanoparticle-mediated antifouling properties using synthetic nano-silica AgNP (NSAgNP) nanoscale material. Through application of the bioinspired technology, green chemistry have been involved in designing high surface area, less pore volume, antimicrobial protein coating and high surface active sites of nanostructured material, which enhanced functional activity, integrity and antifouling. Qu et al. (2013) proposed nanotechnology (nano-Ag, carbon nanotube) for advanced and improved fouling controlled water and wastewater treatment. By adding hydrophilic metal oxide nanoparticles (alumina, silica, zeolite, TiO_2), fouling can be reduced by enhancing the hydrophilicity of the polymeric ultrafiltration membrane surface. Partial success over biofouling can be noted by introducing aquaporin-incorporated lipid bilayers on artificial nanofiltration unit, because of aquaporin's selective rejection and specific permeability. Chen et al. (2013) and Lakshmi et al. (2017) reported that chemical modification of halloysite nanotube by addition of chitosan-Ag nanoparticles (HNTs-CS@Ag) and polyacrylonitrile, respectively. By SEM, AFM and measuring the contact angle, they showed that increased hydrophilicity resulted in higher pure water flux (23.3%) followed by 94.0 and 92.6% reduced biofilm of *Escherichia coli* and *Staphylococcus aureus*, respectively. Liu et al. (2015) employed biogenic silver nanoparticle grafted thin composite membrane, which in a long-term use maintained higher salt rejection and effective antibacterial inhibition.

A green technology of in situ immobilization of silver nanoparticles (AgNPs) onto RO membrane followed by static insertion techniques have been demonstrated by Dong et al. (2017). The membrane surface was modified using two step process including intrusion of tannic acid–ferric ion–polyethylenimine complex layer followed by Ag revision. Strong microbicidal effect of Ag, coupled with hydrophilicity effect of tannic-coated layer revealed 100% mortality of model bacteria *E. coli* and *Bacillus subtilis*.

Quorum quenching bacteria: membrane biofilms

Earlier reviews suggested membrane cleaning technologies in membrane reactor systems (MBR) were partially effective and replacement of membranes has been a common practice in industries which leads to huge economic budget. They envisaged that, misguiding the signal throughout microbiolayer finds promise for designing new strategies and for that detailed data of core community needs to be generated in terms of genomics and proteomics (Eppinger et al. 2007; Fang et al. 2010; Feng et al. 2013; Katebian et al. 2016).

Weera, sekara et al. (2016) showed that chlorine dosing coupled with quorum quenching bacteria *Rhodococcus* sp. *BH4* could delay hollow fibre membrane biofouling, and save energy loss upto 74%. They also demonstrated that only high chlorine injection (100 mg/l) could not resist cell signalling and synchronized community behaviour in biofilm architecture which could be mismatched and controlled using *Rhodococcus* enhanced chemical backwashing.

For applying the fouling controlling bio-strategies, a well-cited organism has been used, *Rhodococcus* sp. *BH4* (Oh et al. 2012). As this organism produces quorum quenching enzymes, by stuffing the cells inside the lumen of microporous hollow fibre membrane (microbial vessel), lab scale membrane biofouling was achieved by 10 days delaying to build up the transmembrane pressure and established the fact that membrane biofouling could be controlled by quorum quenching. The study was further carried out for determining its success in the long run and its sustainable efficacy by Kim et al. (2013), where *Rhodococcus* sp. *BH4* was encapsulated in alginate beads. Cell entrapping beads (CEB) of porous microstructure made the biofouling event to occur at delayed times. Physical friction of the beads which was coupled to quorum quenching attribute together led to loosely bound biofilm which could be easily sloughed off from the surface of the membrane. The work was again evaluated by Lee et al. (2016) where they showed that QQ bacteria got trapped in the core part of the beads which were inadequate to reveal QQ activity, hence they designed a hollow column of QQ bacteria and reported higher efficiency in removing biofouling due to high surface area coverage.

Membrane reactor engineering and surface modification for biofouling issues

By modifying the reactor engineering, fouling was not be controlled in many cases because the fouling microbiota was robust and dynamic (Miller et al. 2012). Araújo et al. (2012) reported that neither the variation of spacer thickness, feed spacer orientation, coating feed spacer with silver, copper, gold nor using the biostatic feed spacer could meet considerable success in fouling control. By incorporating powdered activated carbon (PAC) as a pre-treatment of submerged membrane adsorption bioreactor, fouling could be controlled as it removed organic matter upto 72%, with a little marginal uprising of transmembrane pressure, as claimed by Jeong et al. (2013). The study also reported that 2.14 g of PAC/m^3 of seawater was sufficient to control reverse membrane fouling without hampering biological degradation and adsorption of organic matter by submerged reactor. Matin et al. (2014) reported the effect of surface-modified amphiphilic coating of RO membrane, where the biofilm gets formed by almost two orders of magnitude. They copolymerized

the hydrophilic hydroxyethyl methacrylate (HEMA) and the hydrophobic perfluorodecylacrylate (PFDA) were deposited on quartz crystals followed by adsorption of sodium alginate. Subsequently, less foulant absorption was recorded in comparison with pure homopolymeric surface.

Model bacteria in membrane treatment to prevent biofouling

Studies on model bacterial biofilm treatment have been started long back in nineties. Johansen et al. (1997) employed enzymatic cleaning against *S. aureus*, *Staphylococcus epidermidis*, *Pseudomonas fluorescens*, and *Pseudomonas aeruginosa* biofilm, prepared on steel and polypropylene substrata. They came to the conclusion that bactericidal enzymes may not be efficient in dispersion of biofilm from the surface, viz, glucose oxidase combined with lactoperoxidase. Although a synthesized complex mixture of polysaccharide-hydrolysing enzymes was able to clean bacterial biofilm from steel and polypropylene surfaces, but it was poor in bactericidal activity. Hence, a well-maintained combination of oxidoreductases with polysaccharide-hydrolysing enzymes, resulted in better membrane cleaning in terms of bactericidal activity as well as removal of the biofilm.

However, to explore basic mechanism behind biofilm–antibiofilm study, model bacteria especially *P. aeruginosa* have been used (Yang et al. 2016). Mutation-based study highlighted and identified a range of genes as the ‘required’ factor for biofilm, exclusively taking part in EPS production, motility, controlling surface adhesins, etc. Indeed, knocking out of these genes could not terminate biofilm formation in many cases; subtle vigilance revealed that it resulted in retardation or loose film formation. For example, Tremblay and Déziel (2010) highlighted involvement of motility especially the swarming pattern in biofilm formation and its spreading. By surfing the Gene chip *Affymetrix* microarray data, they differentiated between transcriptome profiles of tendrils migrating population and centre swarm population of swarming colonies and revealed that tendril trip cells act in scouting and rapidly spreading the planktonic surfaces, while inner cells allow the settlement of biofilm.

Yang et al. (2016) reported that H_2O_2 could be implied as the replacement of chlorine to control biofouling in membrane-based water treatment system. They established the efficacy of H_2O_2 by applying it against the model biofouling bacteria *P. aeruginosa* PA01. Hydrogen peroxide H_2O_2 at 625 ppm could inhibit the growth of 104 cfu/ml cells > 99%. They also suggested that low temperature and low cell density of bacterial growth can remarkably be prevented by a low concentration of H_2O_2 , while the remaining biofilm could be cleaned by H_2O_2 –bicarbonate mixture, eventually membrane biofouling could be controlled by H_2O_2 coupling.

Pal et al. (2016) reported application of quorum mimicking biomolecule ‘vanillin’ to prevent growth of membrane biofouling isolates and validated the biofilm inhibition at gene expression level. Wood et al. (2017) showed that incorporation of artificial beneficial biofilm (used model bacteria: *E. coli*, *P. aeruginosa* PA01) could prevent membrane clogging by limiting its own population density by sensing quorum gradient and simultaneously it prevented the biofilm formation as well by secreting nitric oxide (universal biofilm dispersal agent).

Humphries et al. (2017) have revealed the electrical potential that gets generated from membrane potassium gradient which could alter distance population behaviour and synchronize them with biofilm formation, with the help of microfluidic device and model bacteria *B. subtilis* (quorum sensing, gram-positive bacteria) biofilm and a distance species *P. aeruginosa* (quorum sensing, gram-negative bacteria) cells. *Bacillus* biofilm was cultured in microfluidic chamber and after reaching electrical threshold produced by biofilm extracellular potassium gradient, they introduced motile *Pseudomonas* cells to the chamber (Starling 2017). Their findings uncovered that the electrical oscillation produced by potassium channelling was positively correlated with the attachment of *Pseudomonas* with the edge of biofilm. Moreover, they were periodically synchronized with the population through the periodically emerging volts. Furthermore, there was no motile cell propagation, where the population could not start the electrical oscillation or reach the electrical threshold.

Population behaviour and individual role have been magnificently dynamic and complex in biofilm architecture and could be differentiated with the variation in cell signalling (Leary et al. 2014). Microorganisms produce chemical compounds (including signalling molecules, enzymes) which could induce or inhibit settlement, growth and leads to the variations in species richness or abundance factor. Biofilm-forming microorganisms generally tend to secrete antimicrobial, antifungal as well as antilarval compounds, especially marine heterotrophic bacteria, cyanobacteria and marine fungi (Gittens et al. 2013). The biocompounds have multiple activities and it could disrupt signalling production, mismatch signalling transduction profile as well as can break polymer and interfere with enzyme activity. Generally, epibiotic microorganism associate and prefer biotic surfaces like shark skin, kelp forest, coral bench and have aq greater capacity to act as antifouling resources than terrestrial sources.

To understand their hidden potency, genomic tools serve as an useful option along with routine lab practices. By surfing annotated genome, microarray data’s hidden capability towards antifouling can be revealed. Depending on metagenomic data and geo-tag facility, a worldwide network of zonation related to biofouling prone area and antifouling

resources can be monitored in the coming future. Meanwhile, biological means were getting more importance to combat biofouling, as signal disrupting has become a key, economically feasible and affordable choice in antifouling strategies. Lade et al. (2014) have vividly reviewed over pros and cons of using quorum quenching approach to control biofouling and they have found great promises hidden behind a wide range of bacterial species and their enzymes (Kristensen et al. 2008).

Genomic intervention in biofilms: uncovering possible antifouling strategies

To have a solution for membrane biofouling, the key answers have been reported to be hidden in genome profile of the population (metagenome) and as well as in individual robust biofilm former (Gomez-Alvarez et al. 2012), because they could modify total community behaviour according to their signalling pathways (Herzberg and Elimelech 2008). Hence, application of genomic tools has become a necessity for membrane biofilm study.

In the present review, we have illustrated the importance of genomics in membrane biofouling, using pre-existed database (GenBank, National Centre for Biotechnology Information, US). Amplicon metagenome data (Glass et al. 2010; Yergeau et al. 2010; Pal et al. 2016) was retrieved from MG-RAST (ID 4585128.3), that was derived from drinking water biofouled membrane community. Figure 3 illustrated the possible contamination from membrane to waterline emphasizing about membrane care (McLean et al. 2013). Presence of strong quorum producing *Corynebacteriaceae*, *Dermabacteriaceae*, *Mycobacteriaceae*, *Streptomyetaceae*, *Staphylococcaceae*, *Helicobacteriaceae*, *Brucellaceae*, *Bdellovibrionaceae*, *Myxococcaceae*, *Klebsormidiaceae* disclosed the risk factors associated with membrane leakage and subsequent threat for epidemic breakdown especially in developing countries. Second, how pathogen biofilms interacted with environmental biofilms and what key subsystems trigger them, were evaluated by the existing record of RAST Seed viewer (Bouchez et al. 2008; Copel et al. 2007; Overbeek et al. 2013). Comparative distribution of key subsystems (Fig. 4, supplementary Fig. 2) has been shown in 16 biofilm formers and four biofilm negative control bacteria (deficient in biofilm formation, although having stress

Fig. 3 Metagenomic analysis of biofouled membrane (MG-RAST ID 4585128.3) showing abundance, phylogenetic relation and co-existence of pathogenic bacteria with environmental isolates. Emerging pathogenic contaminants of biofouled membrane highlighted in ten groups

Fig. 4 Comparative distribution of biofilm and quorum signal-related genomic components in environmental and pathogenic bacterial genomes. 20 biofilm bacterial genomes were selected from environmental and pathogenic niches. 16 biofilm formers and 4 bacteria deficient in biofilm formation (stress mediated response)

response machinery) (supplementary table 1) (Firmani and Riley 2002; Gulhane et al. 2016; Heidelberg et al. 2000; Janssen et al. 2010; Liao et al. 2011; Ochiai et al. 2005; Read et al. 2003; Sikorski et al. 2010; Strnad et al. 2011; Suzuki et al. 2006; Wei et al. 2003). Functional gene diversity suggested that biofilm features were “mutually shared and partially compromised in pathogenic (P) and environmental (E) cases”, by allowing major component deviation in the factors assigned to (1) resistance to antibiotic and toxic (2) invasion and intracellular resistance, (3) siderophore formation (4) cell signalling and (5) oxidative stress (Fig. 4). It envisages that by taking these four subsystems, biofouling may be analysed and regulated.

Protein network interaction [of *Enterobacter asburiae* EGD_AQ_BF12(LYYC01), a membrane biofouling bacterial genome] was constructed in context of co-expression, fused pattern, mutual regulation, experimental and hypothetical records available at String database (<https://string-db.org/>) (Szklarczyk et al. 2014). Elements were manually decoded as 1–6 for better understanding of clustering pattern (1: proteins associated with cell signalling, including signalling peptide type I and II, autoinducers, twist arginine motifs and chemical signalling, 2: biofilm proteins, 3: siderophore formation proteins, 4: motility and chemotactic proteins, 5: drug resistance proteins, 6: osmoregulatory proteins). It was observed that, siderophore and siderophore-induced biofilm

proteins were exclusively clustered in environmental bacteria *E. asburiae* (supplementary Fig. 3). Hence, arresting siderophore production could lead to promising antibiofilm strategy, as mentioned earlier also by Harrison and Buckling (2009).

It has been reported that, bioelectricity generated from potassium gradient plays an unavoidable role in biofilm signalling and fouling phenomenon (York 2017). When the genome data of biofilm-forming bacteria (isolated from brackish water biofouled membrane, Pal et al. 2016) were explored, it was found to have potassium transporter and efflux protein (kef A, ATPase A) actively taking part in voltage gated ion channelling. Apart from that, any metabolic pathway that releases free potassium ion in cytoplasm could contribute a higher potassium pool in biofilm population than the surrounding K^+ concentration; whenever amino acid routing, phosphate compound channelling, inorganic and organic (having resonance stability) compound transportation carrying charged ion occur in biofilm matrix through the peripheral protein barrier, their movement across the membrane and charge difference causes voltage gradient and millions of ions cross the membrane gate per second. These machineries have a well-defined sense of voltage fluctuation and ligand variation, hence their gate open and close in a millisecond. For particularly addressing this problem, charge nullification

Fig. 5 Schematic representation of proposed hypothesis for mitigating membrane biofouling: Graphics showing quorum sensing (biochemical + bioelectrical) coupled with the electro-modified negatively charged membrane surface delinks signals to mitigate biofouling. 'A' is the quorum producing biofilm bacteria which oscillates bio-electrical (K^+) wave to generate signal to distant communities, i.e. B, C, D

(Piyadasa et al. 2017) by involving embedded negatively charged particle (Fig. 5) finds promiscuity. For example, graphene oxide rooted membrane (Chae et al. 2015; Guo et al. 2017) could impair biofouling by charge repelling while involving numerous hydrophilic and negatively charged functional groups. Membrane surface zeta potential could electrostatically repel bioelectrical wave generated through biofilm potassium gradient and subsequently interrupt signal conveying to distant population (Majumdar and Pal 2017).

By reviewing the genome and metagenome data, it could be suggested that quorum-based strategies (quorum mimicking or quenching) coupled with electrically/chemically modified surface could enhance the possibility of combating the issue of membrane biofouling (Fig. 5) by neutralizing the micro-charged environment of the biofilm.

Conclusion

Comparative omics analysis of microbial community of biofouled membranes revealed the convergence and co-occurrence of biofilm bacteria in pathogenic (P) and environmental (E) niches. Genome analysis of pathogenic and environmental (membrane) biofouling bacterial isolates indicated their salient role in viscoelastic EPS response. The new emerging mechanism proposed the gradient of potassium ions, which send a friendly bioelectrical impulse to distant population, and a phenomenon that leads to heterogeneous

colonization during biofouling. To impair the communication of coaggregating population, the interception of quorum signals and destabilization of the potassium signals using negatively charged surface modification would find promiscuity towards designing anti-biofouling strategies. But, the organization structure and space of inhabitants of biofilm still remains as an area which requires more understanding.

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Compliance with ethical standards

Conflict of interest All the authors declare that they have no conflict of interest.

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