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Biofouling- Bioadhesion of micro-organisms and its prevention: A Review

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Abstract: **The fouling organisms cause effective problems in several strategies by settling on ships' hulls, aquaculture systems, fishing nets, pipelines and other marine infrastructure. Ships suffer increased drag and surface corrosion, leading to lower speeds, thereby causing higher fuel consumption, additional CO_2 emissions and maintenance costs, hence this current review focused on prevention of biofouling by several physical methods like membrane ultrafiltration by surface modifying macromolecules and reverse osmosis. Ultrasonification is also used widely as a physical treatment to prevent biofouling bacteria's adhesion to the marine surfaces. Membrane ultra-filtration by silver nanoparticles (nAg) which are greatly known for antibacterial activity with polysulfone ultrafiltration membrane (nAg-psf) exhibits a wide antimicrobial resistance against several bacterias like *Escherichia coli* thereby reduced the potential of fouling. Tri-n-butyltin (TBT) is reasonably effective in controlling microbial fouling but it would affect marine life at both lower and higher concentrations. To replace this chemically modified antifouling agent, there is a wide need for the development of "eco-friendly" antifouling agent, so the most potential marine microbes like marine algae were identified, characterized by antimicrobial assay and several metabolites were purified by Thin layer chromatography and column chromatography and it's been used as an effective antifoulants. The chemical structures of the metabolites were further assayed by spectral methods. Thus, our present review also presented the benefits associated with the use of different marine organisms, including marine bacteria, algae, sponge, coral, bryozoa, ascidian, etc. as source means that they are the organisms of choice for obtaining natural products for antifouling coatings.**

Keywords: **Biofouling, antifouling agents, biofilm, prevention, reverse osmosis, membrane ultra-filtration, nAg, tri-n-butyltin (TBT), Chromatography, marine organisms.**

1. INTRODUCTION

Fouling is the accumulation/growth of unwanted material on solid surfaces which leads to the loss of function. The fouling materials also consist of either living organisms (bio fouling) or a non-living substance (inorganic or organic). Biofouling refers to the natural process in the marine ecosystem in which the micro and macro foulers like bacteria, algae etc. develops and forms colonization on the submerged or manmade marine surfaces, leading to a great economic and environmental losses. Biofouling is classified into micro fouling, a biofilms formation and bacterial adhesion and macro fouling, an attachment of larger organisms like barnacles, mussels, polychaete worms, bryozoans and sea weeds (Bott and Melo *et.al*, 1997). Damaging of metal surfaces by adherence of marine micro-organisms is directly related to biofouling. (Woods Hole Oceanographic Institution (WHOI, 1967) says among the marine foulers, macrofoulers (invertebrates and algae) are the major fouling organisms creating extensive problems in marine technology like roughness in the ship's hull, and reducing the speed. To avoid the macrofoulers (Secondary fouling organisms), preparing the antifouling biological substances against primary fouling microorganisms especially against to the bacteria are the emerging need in worldwide. Because bacteria are the first organisms responsible to foul the surfaces (Rao *et al*., 2005) and forms complex, three dimensional biofilms which serves as a focus for the attachment and plays a major role in the growth of other organisms, such as invertebrates, sessile plants, and animals (Stoodley *et al*., 2002).

The colonisation of a substratum in the aquatic environment occurs through a four-step process, viz. biochemically conditioning the surfaces, bacterial colonisation, diatom and protozoan colonisation and settlement of invertebrate larvae and algal spores (Wahl, 1997; Maki, 2002,Sergey Dobretsov *et al*, 2006, Guiamet and Gomez De Saravia, 2005).

The bacteria and diatoms are the primary colonizer of any freshwater surface. Marine organisms like *Vibrio proteolyticus* (Paul and Jeffrey, 1985), *Escherichia coli*, *Pseudomonas aeruginosa*, *Shewanellaoneidensis* (Lee and Newman, 2003) and *Bacillus subtilis* (Omoike and Chorover, 2004) have been found to be involved in the biofouling process. Generally, reversible attachment to the substratum followed by irreversible adhesion are the two-step process of bacterial colonization (Biancitto *et al*., 2001). The cells are held by physical forces and can be easily removed by gentle washing in reversible attachment, whereas in Non reversible attachment of cells is often mediated through specific mechanisms such as hydrogen bonding, cation bridging, specific receptor ligand interactions and the production of extracellular polysaccharides (EPS) (Jayaraman and Seetharaman, 2003). Development of this biofilm is simultaneously followed by a build-up of microorganisms (such as, bacteria, fungi, diatoms and other microbes) and the secretion at their cell surface of extra cellular polymeric substances (EPS) during the attachment, colonization, and population growth.

Bacterial cells that can enclose themselves into a hydrated matrix of polysaccharides or associates with extra-cellular polymeric substance (EPS) and proteins forming a slimy layer known as biofilms (Vrouwenvelder *et al.*, 2008) and is the general term for accumulation of bacteria on a surface. EPS is important for the development and maintenance of biofilm structure and accounts for roughly 50–90% of the total organic carbon of biofilms (Chathuri Piyadasa *et al*, 2017).The first step in biofilm formation involves pre-conditioning of the membrane surface by the adsorption of organic macromolecules. Kirchman *et al*. 1982; Lau & Qian, 1997; 2001; Qian *et al*. 2003 states that biofilms can enhance or retard larval settlement of marine invertebrates and attachment of algal spores (Rodriguez *et al*. 1993; Egan *et al*. 2000; 2001; Dobretsov & Qian, 2002; Huang & Hadfield, 2003). The disruption of biofilm formation or prevention of epibiosis can be made possible by chemical compounds produced by bacteria and diatoms, as well as biofilms of live microorganisms. Therefore, they may be useful for the biotechnological development of an ‘‘environmentally-friendly’’ protection against marine biofouling (Clare *et al*. 1992; Holmstrøm & Kjelleberg, 1999; Armstrong et al. 2000, Sergey Dobretsov *et al*, 2006).

Antifouling is generally defined as preventing the accumulation of fouling micro/macro organisms (Briand, 2009).Currently, in order to prevent the marine biofouling, broad spectrum metal biocides, such as tributyl tin (TBT) and copper have been used as antifouling compounds (Albert *et al*., 1992; Thomas *et al*., 2001). These biocides were very effective, but highly toxic to non-target organisms (Alzieu, 2000; Konstantinou and Albanis, 2004).

Antifouling paints mostly with organotin, especially tri-n-butyltin (TBT), copper and organo nitrogen compounds are used as very effective active agents (Willemsen and Ferrari 1993, Isensee et al. 1994). The worldwide application of TBT-based paints has caused a growing pollution in the environment and in foods on a worldwide scale (Suzuki *et al*. 1992, Burridge *et al*. 1995, Horiguchi *et al*. 1995, Ruiz et al. 1995). Due to present and risky regulations on the use of TBT (Dalley 1987), and other polluting antifouling compounds, there is a wide need for other methods for the prevention of marine biofouling. Several physical, mechanical, chemical and biological methods for the prevention of marine biofouling have been developed earlier for the past 30 years (Kuhl and Neurmann 1969, Dhar *et al*. 1981, Branscomb and Rittschof 1984, Fletcher and Baier 1984, Humphries et al. 1986, De Nys *et al.* 1995). The non-poisonous coatings from the isolated produced biogenic agents and marine organisms seems to be the most promising and effective method for the prevention of marine biofouling (S. Abarzua et al, 1999)

In particular, seagrassess (Mayavu *et al*., 2009; Umamaheshvari *et al*., 2009), seaweeds (Bianco *et al*., 2009), mangrove plants (Manilal *et al*., 2009), bacteria (Ravikumar *et al*., 2010) and sea cucumber (Nagi *et al*., 2010) have the efficient eco-friendly antifouling activity which can inhibit the biofilms forming bacteria. (S. Abarzua *et.al*, 1999) isolated the biological active compounds from the benthic cyanobacteria *Scytonema hofmanni* Ag and *Calothrix brevissima*. Suganya *et.al* aimed at determining the antifouling activity of four commercially available seaweeds such as *Amphiroa anceps, Enteromorpha intestinalis, Spyridia hypnoides, Ulva fasciata* and they isolated 48 morphological different strains from two biofilm samples**.**

2. ISOLATION OF BIOFOULING ORGANISMS

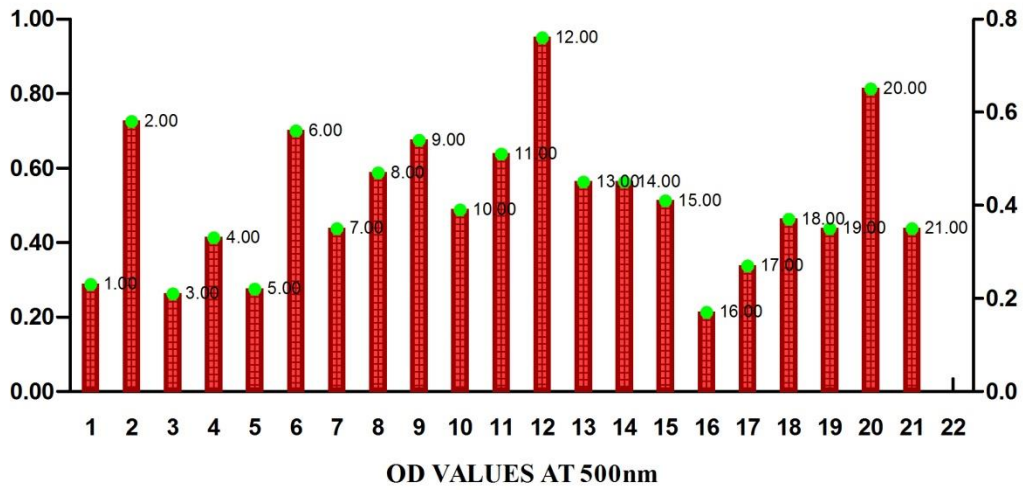
About one gram of fouling sample collected from different boat surfaces was serially diluted using sterile distilled water blanks. One hundred microlitre of aliquot from 10^{-3} to 10^{-5} was transferred to nutrient agar plates prepared in 50% filtered sea water and spreaded using sterile L-rod. Plating was done in duplicate and all the plates were incubated at 28^{0}C for 3-5

days. They were subjected to purification and sub-cultured on nutrient agar slants supplemented with 2% NaCl. Finally Gram's nature of the bacterial isolate were identified by Gram's staining (Bavya *et al.*, 2011, Suganya *et.al*)

3. SCREENING OF BIOFILM FORMING BACTERIA BY TUBE METHOD

The capability of biofilm formation by the bacterial isolates was noticed by the way of adherence to the walls of culture tubes. Inoculums were prepared using 2 ml of nutrient broth. After 24 hours of incubation at 28^0C, turbidity was adjusted to 0.5 McFarland standards. For biofilm experiment, 100µl of inoculums were transferred into 3ml of nutrient broth in 10 ml test tubes. All the test tubes were kept in shaker at 95 RPM speed for 24-48 hours. After incubation, culture broth which contains the free cells, if any, were discarded. The tubes were washed with 3ml of 1X phosphate buffer saline (PBS). About 3ml of 2 % crystal violet solution was added and allowed to act for 5 minutes. All the tubes were washed with sterile water after discarding the crystal violet solution and allowed to dry. All the tubes were visually observed for the presence of biofilms on the inner walls of the test tubes. All the tubes were added with 1.5ml of 33 % glacial acetic acid and mixed gently. Optical density (OD) value was measured in colorimeter at 570 nm. The OD values of the test samples were compared with the PBS present in the control tube (Mathur *et al.*, 2006).At last, while visually observing the presence of biofilms on the inner walls of the test tubes, among the 48 isolates 21 isolates were screened and they were named them as BFB01 to BFB21 (Suganya *et al.*) . The optical density (OD) in all the test tubes was measured in colorimeter at 570 nm (**graph 1**). Among the 21 strains, 6 strains namely, BFB02, BFB06, BFB09, BFB11, BFB12 and BFB20 were found to be higher than others. So, they screened the selected 6 strains for further study.

Graph 1: OD values of screened 21 Biofilm forming Bacteria



4. ANTIBACTERIAL ACTIVITY OF ANTIFOULANTS AGAINST BIOFOULING ORGANISMS

Seaweed extraction:

The dried samples were then placed on blotting paper to remove the excess moisture before preparation of the seaweed extracts, the samples were ground to fine powder prior to solvent extraction. Each 10 g of seaweed powder was mixed in 100 ml of solvents (v/v) like Methanol, Ethyl Acetate and Chloroform separately and they were extracted (Prabhakaran *et al*., 2012).

Antimicrobial assay:

Faulkner, 2000 stated that there is success and little attention has received in antibiotic drug discovery from non-marine microorganisms to marine microorganisms. Antibacterial activity was evaluated using the agar well diffusion assay method by using Nutrient Agar medium (NA) with 50% seawater. Biofilm bacterial isolates were spread on NA plates with sterile effusion and 100 µl of each solvent extract was loaded on agar wells, and the plates were incubated for 24hrs at 37°C. After incubation period, antibacterial activity of the seaweed extract was determined by measuring diameter of the inhibition zone around the wells. The solvent extract which showed the maximum inhibition zone against the tested biofilms forming bacteria will be chosen to characterize the antifouling compounds by using Thin Layer Chromatography and Column Chromatography. The separated fraction was again tested against biofilms forming bacteria by well diffusion

assay method and the compounds were separated by using TLC (S. Abarzua *et al*,1999) and the Rf value was calculated by using the following formula (Selvin and Lipton *et.al*, 2002).

$$\frac{\text{Distance Travelled by substance}}{\text{Distance travelled by solvent}} = \boldsymbol{RF}$$

Antibiotic or biogenic agent production:

The discovery of the first antibiotic compound produced by the marine bacterium *Alteromonas* sp. that was isolated from the Caribbean seagrass *Thalassia sp*. in 1966 (Fenical, 1993). Most metabolites with antibiotic effects were isolated from species that belong to the *Streptomyces*, *Alteromonas*, *Pseudoalteromonas* and *Roseobacter clades* (Fenical, 1993; Wagner-Dobler *et al*. 2002). The bacterial strains such as *Pseudoalteromonas luteoviolacea*, *P*. *tunicata* and *P*. *aurantia* associated with marine organisms that produce antibiotics are isolated from sponges and algae inhibited the growth of other bacteria from the marine environment (Holmstrøm *et al*. 2002). The bacteria *Bacillus pumilus, B. licheniformis, B. subtilis* and *Pseudomonas sp*. isolated from algae and from a nudibranch produced antibiotic compounds that inhibited bacterial attachment (Burgess *et al*. 2003). However, the true source of bioactive compounds from sponges, corals and other marine organisms remains unclear. It must be pointed out that in most previous antibacterial bioassays pathogenic strains were used. In order to investigate the antifouling activity of bacterial isolates, it is necessary to use bacterial isolates from the same environmental situation in order to target ecologically relevant bacterial taxa (Sergey Dobretsov *et.al*, 2006).

S. Abarzua *et.al*, 1999 selected *Scytonema hofmanni*, *Calothrix brevissima* and *Nitzschia pusilla*, a microfouling bacteria as a test diatom. It was isolated from sludge of the Jadebusen near to Dangast (North Sea). The diatom was grown in batch culture at 15◦C under continuous light (20 mEm22 s 21) and aerated with compressed air. Artificial seawater medium (Van Baalen 1962) was used as the growth medium. Bacterial contaminants were eliminated by the addition of a mixture of antibiotics to the growth medium (streptomycin, penicillin and amphotericin were used in a 40:20:0.25 ratio, at a concentration of 4 mg L21 streptomycin) (Round *et al*. 1990).

5. PREVENTION OF BIOFOULING

Prevention by TBT:

Coating marine structures with paints containing tri-butyl tin (TBT) is the most common method to deter fouling organisms from settling and attaching. Paints containing TBT are reasonably effective at controlling the fouling of ships remaining effective for approximately seven years. However, due to concerns raised regarding the safety of TBT in marine paint its use has been restricted. An alternative to paints containing antifouling compounds is foul-release coatings that do not allow fouling organisms to attach due to low surface energies. However, these coatings are as yet not able to withstand the rigours of practical applications such as shipping. In addition to foul-release coatings, a number of other approaches are being examined including electrochemical plating where the surface to be protected is electrically charged to prevent attachment of organisms (Evelyn Armstrong *et.al*, 2000, Sergey dobretsov *et.al*, 2005).

Ultrasonification:

The application of preventing biofouling on vessel hulls have used devices emitting mechanical waves in the ultrasonic (>20 kHz) and audible (20 Hz–20 kHz) frequency range. These devices are generally composed of a signal generator or self-oscillating circuit, power amplifier, and a transducer. Transducers used have included piezoelectric transducers (Branscomb and Rittschof et.al, 1984; Choi et.al, 2013, M. Legg et.al, 2015). Multiple transducers may be used together as an array to optimise the overall gain. Acoustic antifouling studies have been performed in the ultrasonic frequency range (Arnold and Clark, 1952; Berkowitz, 1957; Akselband, 1960; Mori *et al*., 1969; Kohler and Sahm, 1976; Fischer *et al*., 1984 ; Donskoy *et al.*,1996; Seth *et al*., 2010; Aquatic Science Inc, 1995; Mazue *et al.*, 2011 Guo *et al*., 2012, 2013, 2014;. A number of sea trials have been reported to have successfully used lower ultrasonic frequencies (tens of kHz) for preventing biofouling growth (M. Legg *et.al*, 2015).

Prevention by reverse osmosis membrane and ultrafiltration:

Effective feed water pre-treatment is often considered as the most important factor for successful long-term operation of an RO membrane (H.K. Shon et.al, 2008, C. Piyadasa *et al*., 2017). Pre-treatment for RO biofouling includes ultrafiltration (UF) (C. Piyadasa *et al*., 2017, M. Ponti *et.al*, 2005, Y.S. Kim et.al, 2008) to remove colloidal material and

particles which can form a filter cake on the membrane surface and lead to cake enhanced osmotic pressure, and amendment of the feed-water chemistry with coagulants, dispersants and anti-scalants, for the removal of organic compounds to reduce subsequent organic fouling. Coagulation and activated carbon are considered conventional pre-treatment, whereas microfiltration (MF), UF and NF (Nano filtration) are considered advanced pre-treatments for RO (C. Piyadasa *et al*., 2017). Redesigning the RO membrane and/or module has also been proposed as a technique to reduce biofouling effects (AWWaRF- American Water Works Association Research Foundation, 2007). Rougher membrane surfaces result in higher rates of microbial adhesion (C. Whittaker *et.al*, 1984). Membrane/spacer modifications include a change of chemistry and coatings - and have been tested and used for biofouling management (P. Xu *et.al*, 2010, F. Macedonio *et.al*, 2012, D. Rana *et.al*, 2011).

Since excess of boron induce a problem due to an adverse effects on crop production as well as human health and aquatic life (E. Huertas *et.al*, 2008) examined the influence of biofouling by nanofiltration and reverse osmosis membrane on the performance of the membranes in removing boron from a synthetic water effluent. (E. Huertas *et.al*, 2008) used commercial thin biofilm composite RO and NF membranes, NF-70 (Dow-FilmTec, Minneapolis, MN) and LFC-1 (Hydranautics, Oceanside, CA), for the biofouling experiments. The average hydraulic resistances of the membranes were determined with deionized (DI) water to be 3.65(±0.07) $\times 10^{13} m^{-1}$ and 1.06(±0.018) $\times 10^{14} m^{-1}$ at 25.0±0.5ºc, respectively. Salt rejection was determined by synthetic wastewater at a cross flow velocity of 8.5 cm/s and at an applied pressure of 100 and 180 psi (6.9 and 12.4 bar) for the NF and RO membranes, respectively. Observed salt rejections (R_{obs}) of the NF and RO membranes were 85 and 98%, respectively, as determined by electric conductivity measurements. Both NF and RO membranes were received as flat sheet and stored in DI water at 4ºc.

Prevention by surface modifying macromolecules:

Membrane technology is now a days emerging as a vital technique in waste water treatment processes. Ultrafiltration membranes (UF) can disinfect the water by separating the colloids, macromolecules and smaller particles that are harmful. UF process is most importantly involve in separation of proteins, peptide drug from fermentation broths, protein isolated from blood plasma, decreasing the possibility of protein adsorption and cell adhesion in biomedical applications of tissue engineering. (P. Kanagaraj et.al, 2015)

Polyether imide is one of the familiar agent for molding of microfiltration membranes (MF), ultrafiltration membranes (UF), nanofiltration membranes (NF) for its well-developed film formation, mechanical properties, good thermal and chemical properties. This successfully induced the fouling resistance and permeation rate. P. Kanagaraj et.al, 2015 also described that various agents of surface modifying macromolecules such as polyethylene glycol, polyether sulfone, polyethylene oxide, polyvinyl alcohol, polyvinyl pyrrolidone increases the surface hydrophobicity on casting.

The molecular ratio of polyvinyl pyrrolidone (PVP) and polyether sulfone (PES) had most favorable effect on membrane modification process since it increases the size of the pores and effectively modify the ultrafiltration membranes (UF) (L. Y. Lafreniere et. al, 2006). J. Barzin et.al, 2004 evaluated that 12wt% PVP and 2.8wt% PES in N, N-dimethylacetamide had the greater effect in the removal of uremic toxins like urea, uric acid and creatinine from human blood serum compared to other compositions.

D. Rana et.al, 2006 synthesized a new type of hydrophilic surface modifying macromolecules (LSMMs) and investigated the performance of LSMMs by blending with PES ultrafiltration membranes. PES is also used for the ejection of endocrine disrupting chemicals (EDCs) and pharmaceutical and personal care products (PPCPs) (Dipak Rana et. al, 2014). These EDCs and PPCPs constitutes counter therapeutic drugs, fragrances, cosmetics, diagnostic agents, and other pharmaceuticals which greatly affect the purity level of the water.

Prevention by silver nanoparticles:

Biofouling reduces membrane permeability, increases energy costs, and decreases the lifetime of membranes. In order to effectively reduce the biofouling, nanofiltration and reverse osmosis must be used. Since, there is an urgent demand for low pressure membranes with anti-biofouling and antiviral properties the silver and the silver nanoparticles (nAg) which are known for the anti-bacterial properties are incorporated into a wide variety of consumer products as microbial control (Katherine zodrow et.al, 2009). Katherine zodrow et.al, 2009 incorporated nAg into polysulfone ultrafiltration membranes which exhibited antibacterial activity against several bacterias like *Escherichia coli* etc.

Millions of people have taken the advantage of the antibacterial activity of the silver nanoparticles and employed them in simple methods like storing drinking water and for the storage of milk (in silver containers) and as complex as silver-

coating medical implants and instruments. The antibacterial activity of silver is related to sulphur and phosphorous interaction. (Davies et.al, 1997, Katherine zodrow et.al, 2009). Incorporation of antimicrobial nanomaterials into membranes offer an innovative potential for the control of biofouling (Savage and Diallo, 2005; Li et.al, 2008; Katherine zodrow et.al, 2009). Researches is being carried out for the addressing of the long-term effectiveness of the incorporated nAg in polysulfone membranes but are not notably widespread in water purification as well as in biofouling control.

Hence, Katherine zodrow et.al checked biofouling resistance of nAg-Psf The attachment of bacteria to the membrane surface was determined with *E.coli* in MD medium. Then the bacteria which attained stationary phase (~10^9 CFU/ml) were incubated with 1cm^2 PSf and nAg-PSf membrane coupons at 35ºc while shaking for 4hrs. It further resulted in the separation of visible layer in which bacterias are attached with low density which enables to count easily. Then they removed the membrane from the media and rinsed thrice with sterile water. Further, the bacteria on the membrane, specifically the nucleic acids were stained with 1µg/ml of DAPI (4', 6-diamidnia-2-phenylidole) for 5min and it was rinsed and they mounted the membrane on the glass slide and viewed under fluorescence microscope. Finally the antibacterial activity of the membrane was determined by filtering the stationary phased *E.coli* onto the nAg-PSf membrane using vacuum filtration cell and were placed on Luria-Bertani broth and they incubated at 37ºc for 24hrs. Finally, when the filtered *E.coli* suspension onto the nAg-PSf membrane was viewed under fluorescent microscopy they observed that incorporated nAg reduced the attachment of an *E.coli* suspension by 94% (Katherine zodrow et.al, 2009)

6. CONCLUSION

Biofouling increases the surface metal corrosion, cause effective problems in aquaculture systems and marine infrastructures. According to the exiting proof, the marine seaweeds and other marine species were found to have antifouling activity. So, the future work have to be focussed on isolating those antifouling agent in larger scale. Hence, the biofouling organisms are characterized and its prevention by all the physical, chemical and biological methods like reverse osmosis, ultrasonication (physical), TBT, nAg-psf membrane ultrafiltration (chemical) and the several biogenic agents separated from marine species like seaweeds etc has been demonstrated. Since antifouling agents from the marine species showed inhibitory activity against biofouling organisms it can be used an eco-friendly antifoulants rather than using the chemically modified antifouling agent.

REFERENCES

[1] Akselband A. 1960. Ultrasonic protection of ships from fouling. Trans. Oceanogr. Comission, Acad. Sci. USSR 13, 7.

[2] Albert R.S, Snyder S, Zahuranec B., (1992). Biofouling research needs for the United States Navy: program history and goals, *Biofouling*, Vol 6: pp.91-95.

[3] Aquatic Science Inc., October 1995. The Potential Use of Underwater Acoustics for Control of Zebra Mussels: Field Studies. Final Report RAC Project No. 690C. Ministry of Environment and Energy, Ontario, Canada.

[4] Armstrong E, Boyd KG, Burgess JG. 2000a. Prevention of marine biofouling using natural compounds from marine organisms. Biotech Ann Rev 6:221–241.

[5] Arnold, M.H.M., Clark, H.J., 1952. Project B: Ultrasonic Antifouling Shipboard Testing, 1951–52. Technical Report Technical Minute No. 93. Postans Limited.

[6] AWWaRF. Advancing the science of water: AWWa RF and membrane processes, AWWA Research foundation, 2007.

[7] Berkowitz, H., 1957. Acoustic Fouling Project: Final Report. NML/GSO Ref. Narragansett Marine Laboratory, University of Rhode Island.

[8] Biancitto V, Andreotti S, Balestrini R, Bonfante P, Perotto S., (2001). Extracellular polysaccharides are involved in the attachment of *Azospirullum brasilense* and *Rhizobium leguminosarum* to arbuscular mucorrhizal structures. *Eur.* J. Histochem.Vol 45 (1): pp 39-49.

[9] Bianco EM, Rogers R, Teixeira VL, Pereira RC. (2009). Antifoulant diterpenes produced by the brown seaweed *Canistrocarpus cervicornis*. J Appl Phycol. Vol 21: pp 341–346.

[10] Bott, T. R., and Melo, L. F M. Fletcher, and B. Capdeville, Kluwer, Dordrecht. (1992).'Particle-Bacteria Interactions in Biofilms'. In Biofilms*:* Science and Technology, Eds., pp. 199-206.

[11] Branscomb E, Rittschof D. 1984. An investigation of low frequency sound waves as a means of inhibiting barnacle settlement. *J. Exp. Mar. Biol. Ecol*. 79 (2). 149-154.

[12] Branscomb, E. S. and D. Rittschof. 1984. An investigation of low frequency sound waves as a means of inhibiting barnacle settlement. J. exp. mar. Biol. Ecol. 79: 149-154.

[13] Burgess JG, Boyd KG, Armstrong E, Jiang Z, Yan L, Berggren M, May U, Pisacane T, Granmo A, Adams DR. 2003. The development of a marine natural product-based antifouling paint. Biofouling 19:197–205

[14] Burridge, T. R., T. Lavery and P. K. S. Lam. 1995. Effects of tributyltin and formaldehyde on the germination and growth of Phyllospora comosa (Labillardiere) C. Agardh (Phaeophyta: Fucales). Bull. Environ. Contam. Toxicol. 55: 525-532.

[15] C. Whittaker, H Ridgway, B.H. Olson. Evaluation of cleaning stratergies for removal of biofilms from reverse-osmosis membranes. Appl. Environ. Microbiol. 1984. 395-403.

[16] Chathuri Piyadasa, Harry F. Ridgway, Thomas R. Yeager, Matthew B. Stewart, Con Pelekani, Stephen R. Gray, John D. Orbell. 2017. The application of electromagnetic fields to the control of the scaling and biofouling of reverse osmosis membranes - A review. Desalination. 418: 19-34.

[17] Choi C, Scardino A, Dylejko P, Fletcher L, Juniper R. 2013. Effect of vibration frequency and amplitude on biofouling deterrence. Biofouling 29 (2). 195-202.

[18] Ciesluk, A.J., Dufilo, H.L., 1977. Ultrasonic Biofouling Prevention for Ships Intake Systems: FY 78 Report. Technical Report. Naval Coastal Systems Center, Panama City, FA, USA.

[19] Clare AS, Rittschof D, Gerhard DJ, Maki JS. 1992. Molecular approaches to non-toxic antifouling. Invert Reprod Dev 22:67–76.

[20] D. Rana, Y. Kim, T. Matsuura, H.A. Arafat. Development of antifouling thin-film-composite membranes for seawater desalination. J. Membr. Sci. 367 (2011) 110-118.

[21] Dalley, R. 1987. Legislation affecting tributyltin antifoulings. J. Coatings Technol. 59: 113-119.

[22] Davies. R.L, Etris. S.F. 1997. The development and functions of silver in water purification and disease control. Catalysis today 36,107-114.

[23] De Nys, R., P. D. Steinberg, P. Willemsen, S. A. Dworjanin, C. L. Gabelish and R. J. King. 1995. Broad spectrum effects of secondary metabolites from the red alga *Delisea pulchra* in antifouling assays. Biofouling 8: 259-271.

[24] Dhar, H. P., J. O. M. Bockris and D. H. Lewis. 1981. Electrochemical inactivation of marine bacteria. J. Electrochem. Soc. 128: 229-231.

[25] Dipak Rana, Roberto M. Narbaitz, Anne-Marie Garand-Sheridan, Amy Westgate, Takeshi Matsuura, Shahram Tabe and Saad Y. Jasim. Development of novel charged surface modifying macromolecule blended PES membranes to remove EDCs and PPCPs from drinking water sources. J. Mater. Chem. A, 2014, 2, 10059.

[26] Dobretsov S, Qian P-Y. 2002. Effect of bacteria associated with the green alga *Ulva reticulata* on marine micro- and macrofouling. Biofouling 18:217–228.

[27] Donskoy, D.M., Ludyanskiy, M.L., 1995. Low frequency sound as a control measure for zebra mussel fouling. In: The Fifth International Zebra Mussel and Other Aquatic Nuisance Organisms Conference. Pembroke (Canada), Toronto, Canada, pp. 103–112.

[28] Egan S, James S, Holmstrøm C, Kjelleberg S. 2001. Inhibition of algal spore germination by the marine bacterium *Pseudoalteromonas tunicata*. FEMS Microbiol Ecol 35:67–73.

[29] Egan S, Thomas T, Holmstrøm C, Kjelleberg S. 2000. Phylogenic relationship and antifouling activity of bacterial epiphytes from marine algae *Ulva lactuca*. Environ Microbiol 2:343–347.

[30] Esther Huertas, Moshe Herzberg, Gideon Oron, Menachem Elimelech. Influence of biofouling on boron removal by nanofiltration and reverse osmosis membranes. 2018. (318) 264-270.

[31] F. Macedonio, E. Drioli, A.A. Gusev, A. Bardow, R. Semiat, M. Kurihara. Efficient technologies for worldwide clean water supply. Chem. Eng. Process. 51 (2012) 2-17.

[32] Fenical W. 1993. Chemical studies of marine bacteria: developing a new resource. Chem Rev 93: 1673–1683.

[33] Fischer E.C. Castelli V, Rodgers S, Bleile H. 1984. Technology for prevention of marine biofouling a review. Mar. Biodeterior: An interdiscip study. 261-300.

[34] Fletcher, R. L. and R. E. Baier. 1984. Influence of surface energy on the development of the green alga Enteromorpha. Mar. Biol. Lett. 5: 251-254.

[35] Gitlitz, M. H. 1981. Recent developments in marine antifouling coatings. J. Coatings Technol. 53: 46-52.

[36] Guiamet, PS, Gómez de Saravia, SG. (2005). Laboratory studies of biocorrosion control using traditional and environmentally friendly biocides. An overview. LAAR Vol 35: pp 295-300.

[37] H.K. Shon, Vigneswaran J. Cho. Comparison of physicho-chemical pretreatment to seawater reverse osmosis: detailed analyses of molecular weight distribution of organic matter in initial stage. J. Membr. Sci. 320 (2008) 151-158.

[38] Hadfield MG, Paul VJ. 2001. Natural chemical cues for settlement and metamorphosis of marine-invertebrate larvae: In: McClintock JB, Baker BJ, editors. Marine chemical ecology. Boca Raton, FL: CRC Press. pp 431–461.

[39] Haglund A-L, To¨rnblom E, Bostrom B, Tranvik L. 2002. Large differences in the fraction of active bacteria in plankton, sediments, and biofilm. Microb Ecol 43:232–241.

[40] Hattori, T. and Y. Shizuri 1996. A screening method for antifouling substances using spores of the fouling macroalga *Ulva conglobata Kjellmann*. Fish. Sci. 62: 955-958.

[41] Holmstrøm C, Egan S, Franks A, McCloy S, Kjelleberg S. 2002. Antifouling activities expressed by marine surface associated Pseudoalteromonas species. FEMS Microbiol Ecol 41:47–58.

[42] Holmstrøm C, Kjelleberg S. 1999. Marine Pseudoalteromonas species are associated with higher organisms and produce active extracellular compounds. FEMS Microbiol Ecol 30:285–293.

[43] Horiguchi, T., H. Shiraishi, M. Shimizu, S. Yamazaki and M. Morita. 1995. Imposex in Japanese gastropods (Neogastropoda and Mesogastropoda): Effects of tributyltin and triphenyltin from antifouling paints. Mar. Poll. Bull. 31: 402-405

[44] Humphries, M., J. F. Jaworzyn and J. B. Cantwell. 1986. The effect of a range of biological polymers and synthetic surfactants on the adhesion of a marine Pseudomonas spec. strain NCMB 2021 to hydrophilic and hydrophobic surfaces. FEMS Microbiol. Ecol. 38: 299- 308.

[45] Isensee, J., B. Watermann, H.-D. Berger. 1994. Emissions of antifouling-biocides into the North Sea 2 an estimation. Dt. hydrogr. Z. 46: 355-365.

[46] J. Barzin, S.S. Madaeni, H. Mirzadeh, M. Mehrabzadeh, Effect of polyvinylpyrrolidone on morphology and performance of hemodialysis membranes prepared from polyethersulfone, J. Appl. Polym. Sci. 92 (2004) 3804.

[47] Jayaraman M, Seetharaman, J., (2003). Physiochemical analysis of the exopolysaccharides produced by a marine biofouling bacaterium *Vibrio alginolyticus. Proc. biochem.*Vol 38 (6): pp 841-847.

[48] Jenner H. 1983. Control of mussel fouling in Netherlands: experimental and existing methods. In tous I, Miller M, Mussalli Y (Eds). Condenser macrofouling control technologies. Electric power research institute, Hyanis. MA, pp. 12-24.

[49] Katherine Zodrow, Lena Brunet, Shaily Mahendra, Dong Li, Anna Zhang, Quilin Li, Pedro J.J, Alvarez. Polysulfone ultrafiltration membranes impregnated with silver nanoparticles show improved biofouling resistance and virus removal. 2009. 715-723.

[50] Kirchman D, Graham D, Reish D, Mitchell R. 1982. Lectins may mediate in the settlement and metamorphosis of Janua (Dexiospira) brasiliensis Grube (Polychaetea: Spirorbidae). Mar Biol Lett 3:201–222.

[51] Kitamura, H., Takahashi, K., Kanamaru, D., 1995. Inhibitory effect of ultrasonic waves on the larval settlement of the barnacle. *Balanus amphitrite* in the laboratory, Marine fouling 12 (1), 9–13.

[52] Kohler, W., Sahm, K., 1976. Investigations into the use of ultrasonics to prevent marine fouling. In: 3rd International Conference and Exhibition for Ocean Engineering and Marine Sciences, Dusseldorf, Germany, June, pp. 827–834.

[53] Konstantinou IK, Albanis TA., (2004). Worldwide occurrence and effects of antifouling paint booster biocides in the aquatic environment: a review. Environ Int Vol 30: pp 235–248.

[54] L.Y. Lafreniere, F.D.F. Talbot, T. Matsuura, S. Sourirajan, Effect of polyvinylpyrrolidone additive on the performance of polyethersulfone ultrafiltration membranes, Ind. Eng. Chem. Res. 26 (1987) 2385.

[55] Latour M, Murohy P. 1981. Application of PFV2 transducers as pieozoelectric vibrators for marine fouling prevention. Ferroelectrics 32 (1), 33-37.

[56] Lau SCK, Qian P-Y. 1997. Phlorotannins and related compounds as larval settlement inhibitors of a tube-building polychaete Hydroides elegans (Haswell). Mar Ecol Prog Ser 159:219– 227.

[57] Lee, A. K. Newman, D. K., (2003). Microbial iron respiration: Impacts on corrosion processes. Appl. Microbiol. Biotech. Vol 62: pp 134-139

[58] Li. Q, Mahendra. S, Lyon. D.Y, Brunet L, Liga. M.V, Li. D, Alvarez. P.J.J. 2008. Antimicrobial nanomaterials for water disinfection and microbial control: potential applications and implications. Water Research 42 (18), 4591-4602.

[59] M. Leggn, M.K. Yücel, I. Garcia de Carellan, V. Kappatos, C. Selcuk, T.H. Gan. Acoustic methods for biofouling control: A review. 2015. 237-247.

[60] M. ponti, S. Rapenne, A. Thakkedath, J. Duchesne, V. Jacquemet, J. Leparc, H. Suty. Tools for membrane autopsies and antifouling stratergies in sea water feeds: a review. Desalination 181 (2005) 75-90.

[61] Maki J. 2002. Biofouling in the marine environment. In: Bitton H, editor. Encyclopedia of environmental microbiology. New York: Wiley & Sons. pp 610–619.

[62] Manilal A, Sujith S, Seghal Kiran G, Selvin J, Shakir C.,(2009). Biopotentials of mangroves collected from the South of India. Global J Biotech & Biochem .Vol 4 (1): pp 59-65.

[63] Mayavu P, Sugesh S, Ravindran VJ. (2009). Antibacterial activity of sea grass species against biofilm forming bacteria. Res J Microbiol .Vol 4(8): pp 314-319.

[64] Mazue G, Viennet R, Hihn J, Carpenteir L, Devidal P, Albaina I. 2011. Large scale ultrasonic cleaning system: deign of a multi-transducer device for boat cleaning (20 KHz). Ultrason. Sonochemistry. 18 (4), 895-900.

[65] Mori, E., Yamaguchi, Y., Nishikawa, A., 1969. The anti-fouling effect of ultrasonic waves on hulls. Japan: Mitsubishi Heavy Industries. Techn. Rev. 6, 1–9.

[66] Nagi A AL-Haj, Mashan NI, Shamsudin MN, Mohamad H, Vairappan CS, Sekawi Z.,(2010). Antibacterial activity of marine source extracts against multidrug.Vol 42: pp 1327-1329.

[67] Omoike, A.; Chorover, J., (2004). Spectroscopic study of extracellular polymeric substances from *Bacillus subtilis*: Aqueous chemistry and adsorption effects. Biomacromol. Vol 5 (4): pp 1219-1230.

[68] P. Kanagaraj, S. Neelakandan, A. Nagendran, D. Rana, T. Matsuurab and A. Muthumeenala. Performance studies of PEI/SPEI blend ultrafiltration membranes via surface modification using cSMM additives. RSC Adv., 2015, 5, 27594.

[69] P. Xu, C. Bellona, J. E. Drewes. Fouling of nanofiltration and reverse osmosis membranes during municipal wastewater reclamation: membrane autopsy results from pilot-scale invesigations. J. Membr. Sci. 353 (2010) 111-121.

[70] Panchal, C., Takahashi, P., Avery, W., 1995. Biofouling control using ultrasonic and ultraviolet treatments. In: Fouling Mitigation of Industrial Heat Exchangers, p. 11.

[71] Paul VJ. (1992). Ecological roles of marine natural products'. Cornell University Press, New York.

[72] Qian P-Y, Rittschof D, Sreedhar B. 2000. Macrofouling in unidirectional flow: miniature pipes as experimental models for studying the interaction of flow and surface characteristics on the attachment of barnacle, bryozoan and polychaete larvae. Mar Ecol Prog Ser 207:109–121.

[73] Qian P-Y, Thiyagarajan V, Lau SCK, Cheung SCK. 2003. Relationship between bacterial community profile in biofilm and attachment of the acorn barnacle Balanus Amphitrite. Aquat Microb Ecol 33:225–237.

[74] Rao D, Webb JS, Kjelleberg S., (2005). Competitive interactions in mixed-species biofilms containing the marine bacterium *Pseudo alteromonastunicate.Appl Environ microbial*.Vol 71(4): pp 1729-1736.

[75] Ravikumar S, Thajuddin N, Suganthi P, Jacob Inbaneson S, Vinodkumar T.,(2010). Bioactive potential of seagrass bacteria against human bacterial pathogens. J Environm Biol .Vol 31: pp 387-389.

[76] Rodriguez SR, Ojeda FP, Inestrosa NC. 1993. Settlement of benthic marine invertebrates. Mar Ecol Prog Ser 97:193–207.

[77] Round, F. E., R. M. Crawford and D. G. Mann. 1990. The Diatoms. Biology and Morphology of the Genera. Cambridge University Press, 748 pp.

[78] S. Abarzua, S. Jakubowski, S. Eckert and P. Fuchs. 1999. Biotechnological Investigation for the Prevention of Marine Biofouling II. Blue-Green Algae as Potential Producers of Biogenic Agents for the Growth Inhibition of Microfouling Organisms. Botanica Marina Vol. 42, 459-465.

[79] Savage N, Diallo M.S, 2005. Nanomaterials and water purification: opportunities and challenges. Journal of nanoparticle research 7, 331-342.

[80] Seth N, Chakravarty P, Khandeparker L, Anil A, Pandit A. 2010. Quantification of the energy required for the destruction of *Balanus amphitrite* larva by ultrasonic treatment. J. Mar. Biol. Assoc. U.K. 90 (7) 1475-1482.

[81] Sheherbakov, P.S., Grigoryan, F.Y., Pogrebnyak, N.V., 1974. Distribution of highfrequency vibration in hulls of Krasnograd-class ships equipped with ultrasonic antifouling protection systems. In: Transaction. Technical Operations of the Maritime Fleet. Thermochemical Studies. Control of Corrosion and Fouling. No. AD 778380, February.

[82] Stoodley P, Sauer K, Davies DG, Costerton JW, (2002). Biofilms as complex differentiated communities.Ann Rev microbial .Vol 56: pp 187-209.

[83] Suzuki H, Konno k. 1970. Basic studies on the antifouling ultrasonic waves for ship's bottom fouling organism. Influences of ultrasonic waves on the larvae of barnacles, *Balanus Amphitrite hawaiiensis*and mussels. *Mytulis edulis.* J.Tokyo Univ. Fish. 56, 31-48.

[84] Suzuki, T., R. Matsuda and Y. Saito. 1992. Molecular species of tri-n-butyltin compounds in marine products. J. Agric. Food. Chem. 40: 1437-1443.

[85] Thomas K.V, Fileman T.W, Readman J.W, Waldock M.J., (2001). Antifouling paint booster biocides in the UK coastal environment and potential risks of biological effects. Mar Pollut Bull, Vol 42: pp.677-688.

[86] Umamaheshwari R, Thirumaran G, Anantharaman P., (2009). Potential antibacterial activities of seagrasses from Vellar Estuary; Southeast Coast of India. Advan Biol Res Vol 3(3-4): pp 140- 143.

[87] Van Baalen, C. 1962. Studies on marine blue-green algae. Bot. Mar. 4: 129-139.

[88] Vrouwenvelder. J, Manolarakis. S, Van der Hoek S. A, Van Paassen J. P, Van der Meer J. A. M, Van Agtmaal W. G. J, Prummel J. M. C., Kruithof H. D. M, J. C. & Van Loosdrecht J. C. & M. C. M.,(2008a). Quantitative Biofouling Diagnosis in Full Scale Nanofiltration and Reverse Osmosis Installations.Water Res. Vol 42: pp 4856–4868.

[89] Wagner-Dobler I, Beil W, Lang S, Meiners M, Laatsch H. 2002. Integrated approach to explore the potential of marine microorganisms for the production of bioactive metabolites. Adv Biochem Eng Biotechnol 74:207–238.

[90] Wahl M. 1997. Living attached: aufwuchs, fouling, epibiosis. In: Nagabhushanam R, Thompson M, editors. Fouling organisms of the Indian Ocean: biology and control technology. New Delhi: Oxford & IBH Publishing Company. pp 31–84.

[91] Waldvogel, C., Pieczynski, J., 1959. A Research Program for Marine Growth Prevention by Ultrasonics. Technical Report AD0219982. Defense Technical Information Center.

[92] Willemsen, P. R. and G. M. Ferrari. 1993. The use of antifouling compounds from sponges in antifouling paints. Surf. Coat. int. 10: 423-427.

[93] Y S. Kim, Y M. Kim, J H. Kim, I S. Kim, S Lee. Overview of engineering systems for large scale desalination plant with a reverse osmosis network. Desalination 238 (2009) 312-332.