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Review on biofouling prevention using nanotechnology

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Abstract

Biofouling refers to undesirable colonization by microorganisms, macroalgae and invertebrates, leading man-made surfaces to subsequent bio deterioration. In the marine environment, this bioprocess affects surfaces such as pipes, water intake systems, desalination devices, probes and sensors, ship's hulls, building materials and filters. It also damages mariculture facilities such as pipelines, cages etc. Antifouling is the process of controlling fouling of a surface. Commercial antifouling techniques include mechanical cleaning, biocides, toxic coatings etc. Copper based mixture works well for short term and serves as an ideal antifouling agent at least for three years after application. Copper based antifouling formulations affect the organisms other than the fouling organisms too. However their non-target effect is not as much as organo-metallic compounds. When copper is used in nano level, the impact on environment is much lesser. Recently nanotechnology has been evolved as a tool for the formation of antifouling coating. The result of scientific studies has revealed that the nanocoating prevents biofilm formation, bacterial adhesion besides the attachment of macro foulers. It has promising future in maritime industries including shipping in controlling the biofouling. Nanocoating of the metals with antifouling properties have shown positive results for the effective control of fouling in shipping industry in different parts of the world.

Keywords: Biofouling, antifouling, nanocoating, biofilm and nanotechnology

Introduction

Biofouling can be defined as "the undesirable phenomenon of adherence and accumulation of biotic deposits on a submerged artificial surface or in contact with seawater" (Eguia, 1996) [33]. This accumulation or incrustation consists of a film composed of micro-organisms affixed to a polymeric matrix created by biofilm, where inorganic particles (salts and / or corrosive products) may arrive and be retained, as a consequence of other types of fouling developed in the course of the process. Biofouling accelerates the process of corrosion of the materials and causes loss in the performance of the structures. These damages takes place on movable and stationary structures such as boats, gas platforms, oceanographic investigation implements, thermal energy conversion plants and subaqueous sounding equipment. It also damages mariculture facilities such as pipelines, cages etc (Yebra *et al.*, 2004) [94]. Antifouling is the process of controlling fouling of a surface. Commercial antifouling techniques include mechanical cleaning, biocides, toxic coatings etc. Marine biofouling, broad spectrum metal biocides, such as tributyltin (TBT) and all the organic compounds of tin are extremely toxic to non-target organisms. In 1970's, there was continuous use of antifouling paints based on the biocide performance of the organic derivatives of tin, especially tributyltin (TBT). They were found to be the most potential antifouling chemicals used in ships as they covered the ship's hulls and turned out to be an effective and economically viable antifouling agents. Despite the restrictions imposed on use of organo-metallic compounds, copper compounds have re-emerged as a main active ingredient of antifouling coatings in recent years. Copper based mixture works well for short term and serves as an ideal antifouling agent at least for three years after application (Clare, 1995) [21]. Copper based antifouling formulations affect the organisms other than the fouling organisms too. However their non-target effect is not as much as organo-metallic compounds. When copper is used in nano level, the impact on environment is much lesser. The result of scientific studies has revealed that the nanocoating prevents biofilm formation, bacterial adhesion besides the attachment of macro foulers. In recent years, the application of nanotechnology has revolutionized many areas such as material science, agriculture, fisheries, engineering and medicine.

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It has promising future in maritime industries including shipping in controlling the biofouling. Nanocoating of the metals with antifouling properties have shown positive results for the effective control of fouling in shipping industry in different parts of the world.

Impacts of Biofouling

Biofouling refers to undesirable colonization by microorganisms, macroalgae and invertebrates, leading man-made surfaces to subsequent biodeterioration. In the marine environment, this bioprocess affects surfaces such as pipes, water intake systems, desalination devices, probes and sensors, ship's hulls, building materials and filters (Hellio and Yebra, 2009) [38]. Marine biofoulers are divided into three groups such as: primary, secondary and tertiary colonizers. Primary colonizers are microorganisms (mainly bacteria and microalgae), which settle first on the surface. They can be considered as pioneering organisms and are found on unprotected surfaces less than few hours' of immersion. These organisms have been linked to biocorrosion, which is a result of synergistic interactions, between the metal surface, abiotic corrosion products, and microbial cells and their metabolites (Beech and Sunner, 2004) [12]. Secondary macrofoulers comprise of protozoa and spores of macroalgae, and, account for a frictional drag increase of ship up to 10% (Schultz, 2007) [84]. Significant technical and environmental damages of man-made structures are linked to algal fouling. Algal development on structures such as aquaculture nets, buoys and marine blazes can result in such a weight increase that they can consequently sink (Lebert *et al.*, 2009) [50]. Algal fouling on ship's hull is very abundant because ships move between different areas with different biological, physical and chemical properties and are always in the photic zone (Chambers *et al.*, 2006) [14]. Tertiary colonizers are hard macrofoulers which settle on unprotected man-made surfaces after 2-3 weeks of immersion. A great variety of organisms have been observed on surfaces, the main ones being, mussels, tubeworms, and bryozoans. Their presence lead to a dramatic increase of frictional drag up to 40% increase and in some cases to the damage of ships hulls (Gollasch, 2006) [36]. Biofouling and marine corrosion are the two challenging problems that have hindered the exploitation of the seas. Fouling costs the shipping industry millions of Euros every year worldwide due to vessels being out of service in order to have fouling removed, costly repairs and man hours lost. The first obvious effect is the increased frictional drag, thus slowing down the vessel in the water and leading to increased fuel consumption to maintain the same speed. Biofouling on the hull below waterline may lead to an increase of frictional resistance of the boat and thereby causes increased fuel consumption and reduction in speed (Venkatesan *et al.*, 2006) [90]. Additionally, engine will be overstressed and will lead to increase in wear, stress and fatigue. These adverse effects are significant when ships travel via tropical/subtropical zones lead to notable increase in the cost of maritime transportation, which account for 90% of the global exchange of goods (Rodrigue, 2006) [72]. Another detrimental effect of biofouling on ship's hull is the emission of greenhouse gases such as CO₂, CO and SO₂ into the atmosphere.

Studies on Biofilm Formation and Development

The first step in marine biofouling is the formation of a conditioning film with organic materials primarily of protein and carbohydrate. The second step after the formation of

conditioning film in marine environment is the adhesion and development of biofilm by bacteria and microalgae primarily diatoms (Abarzua and Jakubowski, 1995) [1]. Marine natural biofilm consists of bacteria, diatoms, protozoa and fungi. These organisms settle on submerged surface and can colonize fast (Cooksey and Wigglesworth, 1995) [16]. Biofilms are mainly constituted by bacterial populations, which are enclosed in a matrix. The bacteria are adhered to each other and to a surface/interface. Microbial communities in porous spaces, floccules and aggregates are also constitute biofilm (Costerton 1995) [17]. Since the marine environment is nutrient scarce, microbial growth is found at interfaces mainly between solid surfaces and water. The solid surfaces accumulate nutrients as organic macromolecules and inorganic molecules, which are required for microbial growth. Natural occurring surfaces are rocks, plants, the seabed, animals, plankton and other biological materials where manmade surfaces found in the sea are ship hulls, bridge pillars, harbours and piers (Marshall, 1980) [62].

The bacterial biofilm is formed as a result of planktonic cells encountering a surface. The cells use extracellular "sticky" polymers to adhere reversibly. These polymers being primarily glucose and fructose-based polysaccharide fibrils. (Abarzua and Jakubowski, 1995) [1]. Sessile bacterial colonies change their phenotype from planktonic behaviour to an adapted biofilm metabolic state, including increased production of extracellular polymeric substances (EPS) (Marshall, 2006) [63]. The phenotypical change is closely linked to a cell density dependent system called quorum sensing, which has been demonstrated with many distinct bacterial species. As the name indicates, the bacterial cell can "sense" that it is the part of concentration of cells of a certain size (the "quorum"), because of low-molecular-weight signal. Quorum sensing is considered to enable improved access to nutrients, more resilient colonization and higher levels of resistance to hostile environments and antibiotics (Reading and Sperandio, 2005; Waters and Bassler, 2005) [73, 91]. Bacterial biofilms are organized communities, which form intricate architectures with micro colonies of homogenous and mixed species, and water channels inside the matrix that can transport nutrients or metabolites through convective flow (Costerton, 1995) [17]. The community is analogous to eukaryotic tissues, where in cells achieve physiological efficiency and a high level of protection from outside threats.

Biofilm Composition

Bacterial cells constitute about 2-5% of the biofilm mass. The rest of biofilm is made up of the EPS matrix, consisting of exopolysaccharides, proteins, released nucleic acids, glycoprotein, and phospholipids and other surfactants. The matrix also includes trace amounts of ions and humic substances from the surroundings (Allison, 2003) [2]. Many of the proteins in the biofilm are polymer-degrading enzymes, and serve to release cells and provide nutrients for the immobilized cells (Allison, 2003) [2]. Pretentious substances and colonies of *Staphylococcus aureus* have been identified in various biofilms (Lasa, 2006; Latasa *et al.*, 2006) [51, 52]. Furthermore, high abundance of amyloid fibril proteins acting as adhesion has been documented in diverse aquatic habitats (Larsen *et al.*, 2007) [53]. The inherent hydrophobicity, structural stability and plasticity of amyloid fibrils could be important to biofilm structure. *Pseudomonas putida* biofilm, for instance, is reported to consist of up to 75% protein in the water-soluble extractable EPS (Jahn *et al.*, 1999) [47].

Pseudomonas aeruginosa, the best studied biofilm forming bacterial species, has been demonstrated to produce EPS consisting of about 40% (dry weight) neutral polysaccharides, while primarily proteins and lipids as well as some extracellular DNA composed the rest of the EPS (Chang and Gray, 2003) [18].

Biofilm on Different Substrates and Surface

In tropical waters, all surfaces are also heavily covered with biofilms and other biofouling (Lau *et al.*, 2002; Lee & Qian, 2003; Dobretsov *et al.*, 2006; Dobretsov & Qian 2006; Huang *et al.*, 1990; Huang, 1981a) [27, 28, 39, 40, 54, 55]. Biofilm bacteria such as, *Bacillus* sp., *Pseudomonas* sp., and *Staphylococcus* sp. (Sarala *et al.*, 2011) [83], *Vibrio* sp. (You *et al.*, 2007) [95], *Aeromonas* sp., *Micrococcus* sp. (Ramasamy and Murugan, 2002) [74] and *Alcaligenes* sp. (Mary *et al.*, 1993) [64] have been frequently isolated from marine biofouling samples and they clearly indicated that all the above bacteria have the ability to form biofilm. Bacterial strains have been isolated from biofilms formed on glass slides submerged in seawater in Dae-Ho Dike, Korea, by Kwon *et al.* (2002) [48]. The bacterial numbers on glass slides exposed to seawater were increased with exposure times and reached 3.74×10^5 cells/mm² after 72h and the increase rate was 4530/mm²h (Lee *et al.*, 1999) [56]. Dobretsov *et al.* (2006) [27] has mini-reviewed the research works on the inhibition of biofouling by marine microorganisms and their metabolites. Drake *et al.* (2007) [30] have carried out research work on the potential microbial bio invasions via ships' ballast water, sediment, and biofilm. From this study, they have explained how microorganisms are transported within ships in a variety of ways. Using temperature tolerance as a measure of survivability and the temperature difference between ballast-water samples and the water into which the ballast water was discharged, they estimated 56% of microorganisms could survive in the lower bay on the order of 10^{20} microorganisms (6.8×10^{19} viruses and 3.9×10^{18} bacteria cells) are discharged annually. Jin *et al.* (2008) [46] investigated the structure of pioneer communities of marine biofilms developed on three kinds of artificial surfaces (acryl, glass and steel coupons) kept submerged in seawater. The composition of bacterial communities was analysed by terminal restriction and nucleotide sequencing of 16S rRNA. From this study, it indicated some species of γ -*Proteobacteria* were more important as the pioneering population. Dhanasekaran *et al.* (2009) [31] studied the screening of biofouling activity in marine bacterial isolate from ship hull. In this study, 11 isolates were obtained from three ships from Royapuram Harbour, Chennai, Tamil Nadu. Among the 11 isolates only DR4 showed maximum biofouling activity in the micro titer plate assay with a significant optical density of 0.596 and the isolate was similar to *Bacillus* sp. Marine bacteria from the hull of a ship in the form of biofilms or microfouling were isolated, cultured, and identified by phylogenetic analysis using 16SrDNA sequences by Inbakandan *et al.* (2013) [44]. Among them, 16 strains of the Firmicutes were dominant (12.5%), CFB group bacteria (6.25%) and Enterobacteria (6.25%). Chen *et al.* (2013) [19] reported the results of early adherent bacterial diversity and dynamics on a toxic copper-based antifouling paint using a polyphasic approach, including ribosomal intergenic spacer analysis (RISA), conventional culture isolation and 16S rRNA gene clone library analysis. Information of the early adherent marine bacteria will serve as a basis for understanding of copper-related marine microbial diversity in the ocean.

International Studies on Biofouling and Antifouling Measures

Various studies have been carried out globally on biofouling and antifouling measures.

International Studies on Biofouling

Studies on the settlement pattern of marine fouling organisms have been studied by many authors (Morals and Arias, 1979 and Ardizzone *et al.*, 1980) [03, 65]. Huang *et al.* (1981a) [40] observed that the biomass of fouling organisms on test panels was as high as 0.7g/m². Xruming *et al.* (1979) [93] have studied interrelation between the service conditions of ships and fouling organisms and the settlement differed between 31.5g/m² to 28 kg/m². The fouling organisms were found to settle first on weld joints, reverts and sheltered corners and then extended to other panels. Huang *et al.* (1990) [39] observed as much as 25 kg of fouling organisms/m² surface area of underwater structures in Chinese waters. Tseng and Huang (1980) [42] had discussed the biology of marine biofouling organisms and the relationship between marine biofouling and fisheries along the coastal regions of Hong Kong. They specially focused on fin-fish cage culture industries on Hong Kong, which were damaged by marine biofouling. There were about 700 species of foulers recorded in Chinese coastal regions (Huang and Cai, 1984) [41] and 250 species were recorded from Hong Kong (Tseng and Yuen, 1978; Huang and Mak, 1980; Mak, 1983) [42, 59, 87]. El-Komi. (1991) [34] observed the saturation point of fouling over the submerged panels within 3 to 6 months in Eastern harbour of Alexandria of Egypt. Ramadan *et al.* (2006) [79] compared the fouling communities between the years 1960 and 1990 in the Eastern harbour of Alexandria, Egypt and studied the controlling factors of these communities. They recorded minimum diversities with 20 species during the study carried out from 1960 to 1970, while the maximum diversity with 35 species was observed during 1991. They found that small shift among the four dominant groups (*Polychaeta*, *Cirripedia*, *Bryozoa* and *Amphipoda*) was noted during the four decades of the studies. Darbyson *et al.* (2007) [26] tested how clubbed tunicate settlement patterns are different among the most common boat hulls surfaces and colours as well as the ability of these tunicates to survive extended atmospheric exposure similar to that of boats being transported on trails during summer months. Farrapeira *et al.* (2007) [35] surveyed the identity, frequency of occurrence and distribution of dominant species of foulers associated with the various shipping trade routes in the area of Port of Recife, Pernambuco. This research showed that eleven taxa are involved in ship hull fouling in the Port of Recife comprising 28 sessile species, 8 sedentary, and 23 free-living organisms. Coutts *et al.* (2009) [20] tested the effect of vessel speed on biofouling assemblages up to 7 days following voyages of 20 minutes duration. They found that vessel speeds of 5 and 10 knots had little effect on biofouling species richness, however species richness decreased by 50% following voyages of 18 knots and percentage biofouling cover decreased with increasing speed of 10 and 18 knots species decreased by 24% and 85%, respectively. Davidson *et al.* (2009) [23] studied comparison of fouling on containerships as transfer mechanisms of marine biofouling species. The smaller vessel with the underwater surface area of 4465 m² had an estimated coverage of 90% fouling on the hull due to shorter voyage and slower speeds and the ships with the submerged surface area of less than 7,000m² had an estimated coverage of less than 17m² per

vessel. Lin and Siang (2012) ^[57] have surveyed the sub tidal biofouling organisms found on jetty pilings and seen differences across six sites within the Southern islands of Singapore. They found that even within small spatial scales, there were differences in the assemblages of biofouling organisms amongst some sites. At all pilings, algae dominated the top zone (along with the soft corals), followed by encrusting sponges in the middle zone and hydroids occurring primarily at the deepest zone of the pilings.

International Studies on Antifouling Measures

The primary way to prevent biofouling is to select the appropriate material to make the structure to be kept immersed under water. One of the earliest methods of solving the problem in shipping industry is to scrape the hulls of ships. When cleaning or scraping becomes time consuming or ineffective, industries turn to perhaps the most of controlling and preventing biofouling namely antifouling coatings. The best method to control the formation of biofouling on submerged surfaces has been found to be the use of anti-coating. From the dawn of maritime history, the growth of marine organisms on man-made surfaces, the first attempt to control biofouling goes back to the Greek and Roman civilizations, 700 BC, when copper or lead sheathing was used to protect wooden boats (Jones, 2009) ^[45]. Around 1860, ships were built of steel; however copper sheathing could not be used because electrolyte action accelerated the corrosion of the hull (Jones, 2009) ^[45]. This gave the need for alternative methods to protect ships and the dawn of modern paints systems. During the 1960s, the performance of Triphenyl Tin (TPT) paints further improved of self-polishing polymer paints (Yebera *et al.*, 2004) ^[94]. The deleterious effects of TBT released by anti fouling paints were first highlighted in Arcachon Bay of France during 1970s. Organotin, belong to the most toxic pollutants so far for aquatic life. These chemicals have been proven to contaminate the food chain and to be persistent in the environment and have been fully banned since September 2008. An efficient alternative on TBT is not available currently; therefore, nontoxic alternates are urgently needed to have eco-friendly compounds. Presently, 18 different compounds have been used for biocides-based coating. Since the ban of TBT- based paints from September 2008 as per AFS Treaty new formulations have been developed containing high levels of copper and herbicides such as Irgarol 1051, diuron, chlorothalonil, dichlorofuanid and zineb. These paints were first classified as environmentally friendly due to the facts that the active compounds were non-toxic towards non-target species and highly biodegradable when released in the water column. However, there are now significant evidences of a widespread use of these compounds in many countries such as Europe, North America and Japan with sizeable concentrations over marine structures and in harbours (Turner *et al.*, 2009) ^[88]. In order to be proactive, there is a real need for the continuous development of new non-toxic antifouling formulations. The industrial requests development of new coatings are as follow: minimal length of activity; a minimum of 5 years durable and resistant to damage, repairable, low maintenance, easy to apply, hydraulically smooth, compatible with existing anticorrosion, cost effective, non- toxic to non-target species, and effective port and sea (Ralston and Swain, 2009) ^[75]. So far, no new compound with such properties has been discovered despite a massive effort of research. After exploring a wide range of potentialities, many research teams

focus now on an interesting and promising line of research which is inspired by biomimetic solutions and marine biotechnology. Indeed, most marine organisms are prone to biofouling, and colonization of their surfaces leading to a dramatic stress. Organisms that settle on the body surface of other organisms are called the epibionts, the opposite of the basibionts, which are the hosts. Epibiosis refers to the assemblage of epibionts on a basibiont. Epibiosis is typically aquatic phenomenon. The threat of fouling is omnipresent and the list of fouled species is long. This complex association of species will affect the fitness of both the basibionts and the epibionts (Wahl, 2008) ^[96]. On the other hand, a great number of marine organisms do keep their body surface largely clean of epibionts though it is unlikely that there are many sessile species which are not occasionally (seasonally, locally, or on the level of weekend individuals) subject to epibiosis. Any potential basibiont, *i.e.* the majority of sessile, relatively long-lived organisms, must either defend itself against fouling or tolerable epibiosis. A better understanding of epibiosis avoidance would help to the design of new AF solutions. Marine organisms have developed natural AF strategies which can be classified in four groups: chemical, physical, mechanical and behavioural (Ralston and Swain, 2009) ^[75]. The first three are of great interest for new AF developments and have been the basis of biotechnological research on marine natural antifoulants and micro texturing of surfaces. However, even if researchers are focusing on single solution, the best solution would certainly be a mixture of these technologies (De Nys and Guenther, 2009) ^[32]. Marine natural products had been extensively studied for their potential antifouling bioactivities. Soft, fixed or slow moving, organisms showing no epibionts have been selected for bioassay-guided fractionation and purification procedures. To be selected as a new promising AF compound, the new products need to have an effective concentration $EC_{50} < LC_{50}$ (Dhams and Hellio, 2009) ^[25]. From the literature, it appears that the best sources of AF compounds are organisms such as sponges, corals, and macro algae and/or their associated microflora and/or symbionts (Clare, 1998; Fusetani, 2004) ^[21, 22]. Around 200 molecules with variable degrees of AF activities have been isolated and characterized (Hellio *et al.*, 2009) ^[43]. However, it has been regularly highlighted that the active compounds are quite often produced by the associated microflora on the surface of the organisms, which confers a great advantage in term of potential large-scale production. It is indeed less costly to produce a compound via microbial biotechnology than trying to elucidate a synthesis route. A limitation to the use of secondary metabolites within paint formulation is that they are usually rapidly breakdown when released in the environment, thus their incorporation in paint a formulation is very challenging. The best method developed so far is to use microencapsulation to ensure a control of the release rate (Price *et al.*, 1992) ^[69]. Regarding micro texturing of surfaces, studies have focused on marine organisms apparently deprived of physical and chemical defences, such as molluscan shells, crustose coralline algae, marine mammal and sharkskin (Scardinio, 2009) ^[80]. Methods have been developed to reproduce these micro-textured surfaces (laser abrasion, photolithography, moulds and casting and nanoparticles). Researchers are now developing multiple scales of topography with the goal of achieving broader deterrents effects.

Indian Studies on Biofouling and Antifouling Measures

In India also various studies have been carried on biofouling and antifouling measures

Indian Studies on Biofouling

Central Institute of Fisheries Technology (CIFT) has recommended sheathing of the wooden hulls of fishing boats with aluminium-magnesium alloys for protection against borers and foulers, in place of expensive copper sheathing Nair (1987) ^[11, 68] studied the marine biofouling in and around Cochin harbor. The quantum of biofouling accumulation was found to be high during the post monsoon period, followed by the pre-monsoon with the minimum fouling activities during the monsoon period. Balasubramanian and Srinivasan (1987) ^[11] analyzed the occurrences of fouling organisms on oysters in Paravanan estuary of Arcot district of Tamil Nadu. They recorded 445 organisms during April month and 75 organisms in July. Rao and Balaji (1988) ^[76] attributed the drastic reduction in species settlement at Kakinada port to the sewage and oil pollution. Of the 37 species recorded, only 11 could settle at the pollution site at Kakinada, which involved maximum number of polychaete species. They recorded maximum biomass on timber panels ranged, from 0.5 to 3.15 kg/m² (wet weight) and 0.28 to 1.87 kg/m² (dry weight). In the case of glass panels, the biomass ranged from 0.535 to 3.2 kg/m² (wet weight) and 0.31 to 1.9 kg/m². Alam *et al.* (1988) ^[4] studied the biofouling at Ratnagiri coast and they observed that the important mollusks fouling community were bivalves. Meenakumari and Nair (1988) ^[66] analyzed the growth rate of the barnacle *Balanus amphirite communis* in Cochin backwaters. The growth rate was found to be 0.47mm/day during December followed by 0.45mm/day in May. Eashwar *et al.*, (1990) ^[33] studied marine fouling and corrosion studies in the coastal waters of a Mandapam, India. They observed 70 species of fouling organisms in the coastal waters of Mandapam. They recorded that lowest rate of corrosion values were associated with the heaviest settlement rates (approximately 3x 10⁴m²) of *Balanus reticulates* and *Balanus amphirite*. Rajagopal *et al.* (1990) ^[77] also recorded heavy settlement of bryozoans and barnacles and the studies were related to the ecology of fouling organisms in Edaiyur backwaters of Kalpakkam. During this study, the maximum biomass settlement was observed 103g/cm² after 328 days of immersion and the short-term panels was 37g/cm² in 17 days immersion. Rao *et al.* (1991) ^[76] confirmed the successful spread of the exotic fouling bivalve, *Miytilopsis salei* in Visakhapatnam harbor. The fouling rate by this organism was found to be high as 100kg/m²/yr. Rajagopal *et al.* (1997) ^[77] studied the seasonal settlement and succession of fouling communities in Kalpakkam. They found maximum biofouling biomass accumulation of 64 kg/m² within 30 days. Swami and Udayakumar (2010) ^[81] aimed at seasonal influence on settlement, distribution and diversity of fouling organisms at Mumbai harbor. Sixty species were recorded during the investigation period (2000-2001). Among sixty recorded species, 16 were new records from the region. Species settled in pre-monsoon were significantly higher than species settled in monsoon and post-monsoon. Lakshmi *et al.* (2012) ^[58] has studied biofouling on six different polymers substrata with varying surface energy (18-40m/Nm) and surface roughness (R (a) 45- 175µm) in the Eastern coastal waters of India. The results showed that the substrata surface energy (SE) followed by the surface roughness (R (a) had profound effect on attachment of fouling organisms. Anand Babu *et al.* (2012)^[5]

investigated the distribution of macro fouling fauna in the coastal area of Puducherry, India. A total of 8 species belong to different groups such as barnacles, mussels, oysters and tubicolous polychaetes were the prevalent macro foulers encountered in this coastal area. Ananthan (2012) ^[6] studied the seasonal variations in the proximate composition of ascidians from the Palk Bay. Tamilselvi *et al.* (2012) ^[85] had analyzed the diversity and seasonal variations of Class, Ascidiacea in Thoothukudi coast, India. Palanichamy and Subramanian (2014) ^[70] had worked on hard foulers induced crevice corrosion of High S low A steel in the coastal waters of the Gulf of Mannar (Bay of Bengal), India.

Indian Studies on Antifouling Measures

In India, most of the antifouling studies have been carried out on green antifouling technology against biofouling. The antifouling property of water soluble and organic extract from two Gorgonian coral species. Mayavu *et al.* (2009) ^[58] made an investigation to explore the bioactive potential of sea grasses viz., *Cymodocea serrulata* and *Syringodium soetifolium* occurring commonly along the Thoothukudi coastal area and tested their ability of antifouling properties against marine biofilm forming bacteria. Bragdeeswaran *et al.* (2011) ^[9] have worked on antifouling activity by sea anemones (*Heteractis magnifica* and *H.aurora*) extracts against marine biofilm bacteria. Crude extracts of the sea anemone were assayed against seven bacterial biofilms isolated from three different panels. The extracts from seaweeds namely, *Ulva lactuca*, *Caulerpa scalpelliformis*, *Padina boergesenii*, *Caulerpa* sp. and *Chaetomorpha* sp. were tested against biofilm forming bacteria namely, *Micrococci* sp., *Aeromonas* sp., *Pseudomonas* sp., *Flavobacterium* sp., *Cytophaga* sp. and *Enterobacter* sp. Bavya *et al.* (2011) ^[10] concluded that the marine actinomycetes, *Streptomyces filamentous* (R1) would be a potential source for the development of eco-friendly antifouling compounds. Prabhakaran *et al.* (2012) ^[71] worked on antifouling potentials of extract from seaweeds (*Ulva reticulata*, *Sargassum wightii*, *Halimeda macroloba*), sea grasses (*Halodule pinifolia*, *Cymodocea serrulata*), and mangroves (*Rhizophora apiculata*) against primary biofilm forming bacteria. Ganapriya *et al.* (2012) ^[37] studied the antifouling activity of bioactive compounds from marine sponge, *Acanthella elongate* against the biofilm forming bacteria. Larvae of *Balanus amphirite* were used to monitor the settlement and the extent to inhibition due to its toxicity. Venugopal *et al.* (2013) ^[89] isolated and reported bioprospecting of culturable actinobacteria from less explored marine ecosystems against biofouling bacteria for antifouling compounds. The study was successful in identifying the marine actinomycetes from southeast coast of India, which remain unexploited for antifouling compounds. This study narrated the characterization and biofilm activity of different biofouling bacteria isolated from different coastal areas of Tamil Nadu, India. Therefore, the potential strains, PE7 and PM33 would be a promising antifouling agents and both are combined together with the knowledge of coating technology can be utilized for developing eco-friendly antifouling alternatives in future.

Nanotechnology as an Antifouling Method

Recently nanotechnology has been evolved as a tool for the formation of antifouling coating. Khanna (2008) ^[49] has reviewed work on the nanotechnology in high performance

paint coatings. Szewczyk (2010) ^[82] studied the role of nanotechnology in improving marine antifouling coatings. In this study, results of a preliminary literature review on the potential role of nanotechnology in solving ecological problems concerning antifouling coatings were presented. Theresa *et al.* (2010) ^[86] studied the effect of copper nanofilms on bacteria at Indira Gandhi Centre for Atomic Research, Kalpakkam. Preliminary studies have been initiated by Silesian University of Technology and AMBIO of European Union (Ravisubramanian *et al.*, 2013) ^[78] The aim of AMBIO was to study and develop different nano-structured surfaces to avoid the adhesion of marine fouling organisms. Nano-structuring of a coating controls many bulk properties that are relevant to antifouling, “non-stick” surface, such as surface energy, charge, conductivity, porosity, roughness, wettability, friction, physical and chemical reactivity. The research on nano-scale interfacial properties of different surfaces and how organisms adhere will allow understanding on how antibiofouling systems can work, starting at the nano-scale to scale-up to future industrial applications. Mathiazhagan and Rani Joseph (2011) ^[60] have reviewed different types of coatings different types of pigments used in paint formulation with the particular reference to the use of nanomaterials in coating application. Axel Rosenhahn *et al.* (2008) ^[7] reviewed the details of systematic strategy adopted by an FP6 EU Integrated Project “AMBIO” to develop fundamental understanding of key surface properties that influence settlement and adhesion of fouling organisms. Akesso *et al.* (2009) ^[8] have carried out studies on the potential nano-structured oxides deposited by plasma assisted chemical vapor deposition method for the control of aquatic biofouling. The coatings showed good performance against freshwater bacterium, *Pseudomonas fluorescens*, significantly reducing initial attachment and biofilm formation, and reducing the adhesion strength of attached bacterial cells under shear. Costello *et al.* (2012) ^[13] have carried out research for controlling bacterial adhesion on material surfaces using nano-technological applications.

Conclusion

Nanotechnology in biofouling can facelift the antifouling coating industry. Properties like corrosion resistance, chemical and mechanical properties are improved significantly using nanoparticles. The nanocoatings showed good performance against freshwater bacterium, *Pseudomonas fluorescens*, significantly reducing initial attachment and biofilm formation, and reducing the adhesion strength of attached bacterial cells.

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