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Table of Contents

PLENARY-1 HYDRODYNAMICS AND ENERGY EFFICIENCY
Eliasson, J 1

REGULATORY AND ENVIRONMENTAL (PERERIA, M):

RE-1 ANTIFOULING AND IMO'S GOALS - THE IMPACT OF FOULING PROTECTION ON SHIPPING'S GREENHOUSE GAS EMISSIONS AND TRANSFER OF NON-INDIGENOUS SPECIES
Berg, E 2

RE-2 REASSESSMENT OF ANTIFOULING BIOCIDES IN NEW ZEALAND
Koutsaftis, A 3

RE-3 THE CHANGING SCENE OF REGIONAL REGULATIONS AFFECTING THE ANTI-FOULING INDUSTRY AND THEIR IMPLICATIONS FOR DEVELOMENTS OF NEW ACTIVE SUBSTANCES
Poppleton, J 4

RE-4 A REVIEW OF THE GLOBAL STATUS OF ANTIFOULING PAINT REGULATORY MANAGEMENT, WITH A PARTICULAR FOCUS ON THE USE OF COPPER
Long, K 4

RE-5 REGULATORY IMPLICATIONS OF OLFACTORY AND BEHAVIORAL EFFECTS OF COPPER ON FISH
Gensemer, R 5

RE-6 PROPOSED U.S. AMBIENT WATER QUALITY CRITERIA FOR COPPER IN SALTWATER: OVERVIEW AND EXAMPLE APPLICATION
Santore, R 6

RE-7 REGULATORY PRESSURES ON RECREATIONAL BOATING: DO ALTERNATIVE PAINTS OFFER SOLUTIONS?
Holman, K 7

RE-8 TECNOLOGIC AND SCIENTIFIC FEASIBILITY OF ADAPTIVE MANAGEMENT ON ECOLOGICAL RISK CAUSED BY ANTIFOULING AGENTS
Shibata, K 7

BIOFOULING CONTROL TECHNOLOGIES-NEW COATINGS FOR AF AND FOULING RELEASE (WEBSTER, D):

AF-1 FOULING IN THE FACE OF A “LITTLE” SURFACE ROUGHNESS
Lamb, R 8

AF-2 HYDROGELS AS NON-FOULING SURFACES FOR MARINE APPLICATIONS
Olsen, S 9

AF-3 MACROMOLECULAR DESIGN OF SEBS AND PDMS BASED AMPHIPHILIC COATINGS FOR FOULING RELEASE APPLICATIONS
Yasani, B 10

AF-4 DEVELOPMENT OF ENVIRONMENTALLY BENIGN, DURABLE AND EFFECTIVE ZWITTERIONIC-BASED MARINE COATINGS
Jiang, S 10

AF-5 XEROGEL COATINGS WITH ANTI-FOULING AND FOUL-RELEASE PROPERTIES FROM LONG-CHAIN ALKYL AND FLUOROALKYL COMPONENTS
Detty, M 11

AF-6 NONFOULING RESPONSE OF DIFFERENT HYDROPHILIC UNCHARGED POLYMERS
Serrano, Â 12

AF-7 NOVEL ZWITTERIONIC COATINGS BASED ON RING OPENING METATHESIS POLYMERIZATION
Tew, G 13

AF-8 BOTTLE BRUSH-NANOGLASS: A NEW HYBRID APPROACH FOR ENGINEERED, FOULING RELEASE, FLUOROUS POLYOXETANE COATINGS
Chakrabarty, S 14

SHIP HYDRODYNAMICS AND ENERGY EFFICIENCY (SCHULTZ, M):

SH-1 A REPORT ON SOME RECENT DEVELOPMENTS IN SHIP HULL AND PROPELLER COATINGS WITH A VIEW TO FOUL RELEASE TYPE ANTIFOULINGS
Atlar, M 15

SH-2 EVALUATING HULL FOULING ON SHIP PERFORMANCE: UNDERWATER PHOTOS IN CONNECTION WITH SPEED/FUEL CONSUMPTION ANALYSIS
Kane, D 16

SH-3 HULL AND PROPELLER FOULING PENALTIES ON AUSTRALIAN ARMIDALE CLASS PATROL BOATS
Grandison, C........ 16

SH-4 PERFORMANCE OF FOULING RELEASE COATINGS ON NAVY PROPELLERS
Steinberg, M........ 17

SH-5 METHODOLOGY FOR SHIP'S HULL PERFORMANCE ANALYSIS AND ANTIFOULING MONITORING
Krapp, A........ 17

SH-6 FLOW-GENERATED FORCES ON HULL FOULING ORGANISMS AND HYDRODYNAMIC SELF-CLEANING OF FOULING-RELEASE COATINGS
Gowing, S 18

SH-7 FOULING-RELEASE OF BARNACLES AND ALGAE FROM A SHIP HULL
Granhag, L 18

SH-8 STUDY FOR FRICTION RESISTANCE AND GEOMETRY OF SURFACE ROUGHNESS OF ANTIFOULING PAINT
Mieno, H 19

BIOFOULING CONTROL TECHNOLOGIES-BIO-INSPIRED COATINGS (JIANG, S):

BI-1 BIO-INSPIRED ANTIFOULING MATERIALS – TRANSLATING INSPIRATION INTO APPLICATION
de Nys, R 20

BI-2 STRUCTURE-ACTIVITY RELATIONSHIPS OF AMIDES IN ANTI-SETTLEMENT OF DIFFERENT FOULERS
Chai, C 20

BI-3 A FIRST-PRINCIPLES THERMODYNAMIC MODEL FOR PREDICTING RELATIVE ATTACHMENT OF FOULING ORGANISMS TO ANTIFOULING TOPOGRAPHIES
Brennan, A 21

BI-4 TERNARY CROSSLINKED NETWORKS BASED ON HYPERBRANCHED FLUOROPOLYMER-POLYDIMETH-YLSILOXANE-POLY(ETHYLENE GLYCOL) BLENDS TOWARDS NON-TOXIC, ANTI-FOULING COATINGS
Pollack, K........ 21

BI-5 POSSIBLE MOLECULAR TARGETS OF AN ANTIFOULING COMPOUND (BUTENOLIDE) IN THE BARNACLE *BALANUS AMPHITRITE* AND THE BRYOZOANS *BUGULA NERITINA*
Qian,P 22

BI-6 LABORATORY AND FIELD TESTS OF AN ANALOGUE OF ANTIFOULING FROM MARINE SPONGE
Coutinho, R 22

BI-7 POTENTIAL OF MICROALGAE EXTRACTS FOR ANTIFOULING APPLICATION: LABORATORY SCREENING AND FIELD-TESTING
Id Daoud, G 23

BI-8 TRANSPORT OF SOFT PARTICLES VIA ACTUATED ARTIFICIAL CILIA
Balazs, A 23

POSTER SESSION 1

MP-01 ASSESSMENT OF COPPER CONCENTRATION, TOXICITY AND BIOAVAILABILITY IN SHELTER ISLAND YACHT BASIN, SAN DIEGO, CALIFORNIA
Capolupo, C........ 24

MP- 02 BIOSYNTHESIS, ASSEMBLY AND CHARACTERIZATION OF SILVER NANOPARTICLES TO CONTROL MARINE BIOFILM FORMATION
Inbakandan, D 25

MP-03 ABNORMAL DUCKWEED FRONDS OCCURRED BY METAL PYRITHIONE
Okamura, H 26

MP-04 BIOACCUMULATION AND EFFECT OF TRIBUTYLTIN ON THE SPERMATOGENESIS IN A MUD CRAB SCYLLA SERRATA
Iyapparaj, P 26

MP-05 REVISION AND UPDATE OF THE US AMBIENT WATER QUALITY CRITERIA FOR COPPER IN SALTWATER
Santore, R 27
MP-06 MATHEMATICAL PREDICTION AND SIMULATION OF BIOCIDE DIFFUSION IN ANTIFOULING SYSTEMS
Goodes, L 28
MP-07 GROWTH AND ADHESION STRENGTH OF DIATOM BIOFILMS CULTURED UNDER DYNAMIC CONDITIONS
Finlay, J 28
MP-08 A FLUOROPOLYMER FOULING RELEASE COATING SIGNIFICANTLY RESISTS THE INITIAL DEVELOPMENT OF BIOFILMS
Dobretsov, S 29
MP-09 BIOFILM DEVELOPMENT ON HIGH-PERFORMANCE MOLDED MATERIALS IN A TEMPERATE NORTHERN ESTUARY
Maranda, L 29
MP-10 MICROBIAL COMMUNITIES ON ANTIFOULING MARINE PAINTS
Muthukrishnan, T 30
MP-11 APPLICATION OF A NEW BIOFILM SENSOR FOR ANTIFOULING TREATMENTS OPTIMIZATION
Pavanello, G 31
MP-12 IN SITU ASSESSMENT OF BIOFILM GROWTH AND STRUCTURE ON NATURAL PRODUCT CONTAINING ANTIFOULING COATINGS
Salta, M 32
MP-13 ADHESION OF MARINE BACTERIUM BACILLUS SUBTILIS TO HARD SURFACE: ROLE OF NUTRIENTS AND ENVIRONMENTAL FACTORS
Satheesh, S 32
MP-14 ELECTROCHEMICAL SENSING AND CHARACTERIZATION OF AEROBIC MARINE BACTERIAL BIOFILMS ON GOLD ELECTRODE SURFACES
Wharton, J 33
MP-15 SURFACE WETTABILITY AND TEXTURE DO NOT AFFECT THE SETTLEMENT OF THE HYDROID ECTOPLEURA LARYNX
Bloecher, N 34
MP-16 ENHANCING THE SETTLEMENT AND ATTACHMENT OF PEDIVELIGERS OF MYTILUS GALLOPROVINCIALIS USING SURFACE WETTABILITY AND MICROTOPOGRAPHY
Carl, C 34
MP-17 WHEN NAILS ARE MORE COMFORTABLE: THE SETTLEMENT-INHIBITING CHARACTERISTIC OF AIR POCKETS ON SUPERHYDROPHOBIC SURFACES
Nakanishi, K 35
MP-18 BIOFOULING IN NATURAL SEAWATER OF THE ROUGHENED SURFACE OF PTFE
Lawrence, S 35
MP-19 WHALE BALEEN NANOSTRUCTURES AS A POSSIBLE MODEL FOR ANTIFOULING CONTROL
Pustam, A 36
MP-20 DESIGN OF OPTIMAL SURFACE TOPOGRAPHY FOR ANTI-FOULING SURFACES BY COMPUTER SIMULATION
Schoch, P 36
MP-21 TUNING SENSITIVITY TO THIN AIR: THE EFFECT NANOBUBBLES ON ATTACHMENT BEHAVIOUR OF MARINE ORGANISMS
Wu, A. 37
MP-22 ENGINEERING SURFACES FOR MARINE SERVICE BY COLD SPRAY
King, P 37
MP-23 COLD SPRAY ANTIFOULING PROTECTION FOR POLYURETHANE MARINE SEISMIC STREAMERS
Poole, A 38
MP-24 POLY (TRIALKYLSILYL METHACRYLATE)S: A FAMILY OF HYDROLYSABLE POLYMERS FOR MARINE ANTIFOULING COATINGS
Bressy, C 39
MP-25 BINDER RESEARCH FOR ANTIFOULING MARINE COATINGS
Brisset, H 40
MP-26 LABORATORY AND FIELD PERFORMANCE OF PDMS-PEO BLOCK COPOLYMER COATINGS OF VARYING WETTABILITY
Evariste, E 41

MP-27 SOLVENT-FREE ALKOXYSILANE-BASED HYDROLYZABLE POLYMER SYNTHESIS AND DEVELOPMENT
Lawrence, S 41

MP-28 CONTROLLED POLYMERIZATION BY THE RAFT PROCESS OF TRI-ALKYLSILYL METHACRYLATES AND STUDY OF THEIR HYDROLYSIS KINETICS IN ALKALINE MEDIA
Lejars, M..... 42

MP-29 DEVELOPMENT OF FRC/SPC HYBRID ANTIFOULING COATINGS
Lejars, M..... 42

MP-30 SYNTHESIS AND PROPERTIES OF WATERBORNE POLYURETHANE-IMIDAZOLE ANTIFOULING COATINGS
Rahman, M 43

MP-31 EFFECT OF SHEAR FORCES AND SURFACE HETEROGENIETY ON MARINE BACTERIAL RELEASE FROM PNIPAAM GRAFTED BRUSHES
Shivapooja, P 43

MP-32 SURFACE PROPERTIES OF BOTTLEBRUSH POLYMER FILMS
Verduzco, R 44

MP-33 FABRICATION AND INVESTIGATION OF SURFACE-PATTERNED AMPHIPHILIC PERFLUOROPOLYETHER/ POLY (ETHYLENE GLYCOL) NETWORKS: TOWARDS HIGH-PERFORMANCE ANTI-FOULING COATINGS
Wong, D 45

MP-34 POLY(SULFOBETAINE METHACRYLATE)-MODIFIED DIMETHYLPOLYSILOXANE WITH IMPROVED DIATOM ADSORPTION RESISTANCE PROPERTY
Xu, F 46

MP-35 LOW-FOULING AND HIGH-FOULING-RELEASE MARINE COATINGS BASED ON ZWITTER/PDMS COMPOSITES
Xu, X 46

MP-36 FUNCTIONAL POLYMER BRUSHES VIA SURFACE-INITIATED ATOM TRANSFER RADICAL GRAFT POLYMERIZATION FOR COMBATING MARINE BIOFOULING
Yang, W 47

MP-37 STUDY OF *PSEUDOMONAS* NCIMB 2021 ADHESION ON STAINLESS STEEL IN PRESENCE OF A PEO-PPO-PEO TRIBLOCK COPOLYMER – A POSSIBLE METHOD TO REDUCE BIOFOULING
Yang, Y..... 48

MP-38 SALT OIL ACIDS AS EFFECTIVE INHIBITORS FOR STEEL CORROSION IN SEAWATER
Abbasov, V 49

MP-39 FIELD TESTS OF MICROBIOLOGICALLY INFLUENCED CORROSION OF TYPE 304 STAINLESS STEELS: EFFECTS OF SEASONAL CHANGE IN BACTERIAL CONSORTIA AND CHLORIDE CONCENTRATION
Kawakami, H 49

MP-40 ELECTROCHEMICAL BEHAVIOR OF UNS S31600 STAINLESS STEEL IN THE PRESENCE OF CULTURED JUVENILES OF THE BARNACLE AMPHIBALANUS AMPHITRITE DARWIN, 1854
Messano, L 50

MP-41 A COMPARISON OF DNA EXTRACTION METHODS ON NOVEL TEST SAMPLES INCLUDING THOSE CONTAINING CRUDE-OIL
Oldham, A 51

MP-42 INFLUENCE OF MICROFOULING ON DRAG FOR FOUL RELEASE COATINGS, MEASURED USING A DISC ROTOR APPARATUS
Dennington, S..... 51

MP-43 BARNACLE GROWTH AT YACHT PROPELLERS – A SERIOUS REASON OF INCREASED FUEL CONSUMPTION
Elwing, H 52

MP-44 HYDRODYNAMIC DRAG FORCES ASSOCIATED WITH LOW-FORM FOULING
Hausmann, D..... 52

MP-45 SHIP HULL COATING INSPECTION
Koka, A 53

MP-46 THE PROTOTYPE OF A LASER HULL ROUGHNESS PROFILOMETER FOR USE IN DRYDOCKS
Politis, G 53

MP-47 BIOCIDE ANTIFOULING PAINTS-BENEFITS VS. CONTINUING ENVIRONMENTAL COSTS
Bray, S 54

MP-48 RIGID FIBER COATINGS AS EFFECTIVE ANTIFOULING SURFACES FOR BOTH HARD AND SOFT FOULING – 5 YEAR RESULTS AND MECHANISM INTERPRETATION
Breur, R 55

MP-49 HULLBUG, AN AUTONOMOUS HULL CRAWLING ROBOT FOR PROACTIVE HULL GROOMING AND INSPECTION
Darling, D 56

MP-50 BIOFOULING FIELD SURVEY ON THE HULLS OF EX-USS FORRESTAL AND EX-USS SARATOGA
Pinckard, N 56

MP-51 PROGRESS IN THE DEVELOPMENT AND FULL-SCALE TESTING OF ELASTOMERS PROTECTED FROM MARINE BIOFOULING BY AN ISOTHIAZOLINONE-BASED BIOCIDE
Ramotowski, T 57

MP-52 DURABILITY ASSESSMENT OF FOUL-RELEASE COATINGS
Tordonato, D 57

PLENARY-2 TOWARDS TOUGH FOULING-RELEASE COATINGS WITH TAILORABLE SURFACE COMPOSITION
Webster, D **58**

MICROBIOLOGICALLY INFLUENCED CORROSION (BEECH, I):

MIC-1 THE ROLE OF MOLECULAR TECHNIQUES IN ELUCIDATING MICROBIOLOGICALLY INFLUENCED CORROSION
Duncan, K 59

MIC-2 THE STUDY OF BIOFILM-INFLUENCED MARINE CORROSION OF CU-NI ALLOY
Beech, I 60

MIC-3 MILD STEEL CORROSION IN NEARSHORE MARINE ENVIRONMENTS - ASSESSING THE PRESENCE OF IRON-OXIDIZING BACTERIA AND CHARACTERIZING THE OVERALL BACTERIAL COMMUNITY
Emerson, D 61

MIC-4 MARINE MIC OF MILD STEEL - ELECTROCHEMICAL ANALYSIS OF MODEL CORROSION COMMUNITIES
Little, B 61

MIC-5 MARINE CORROSION IN FUEL SYSTEMS
Little, B 62

MIC-6 SULFIDE PRODUCTION AND CORROSION IN SEAWATER DURING EXPOSURE TO FAME ALTERNATIVE FUEL
Lee, J 62

MIC-7 ARE BACTERIA INVOLVED IN THE EARLY STAGES OF CARBON STEEL CORROSION PROCESS IN MARINE ENVIRONMENT?
Lanneluc, I 63

MIC-8 EXPERIMENTAL INVESTIGATION INTO THE EFFECTS OF ICCP AND SCALE FORMATION ON MIC ON HSLA STEEL
Bruin, A 64

GENERAL ASPECTS OF FOULING-MACRO FOULING (HADFIELD, M):

MACRO-1 POPULATION DYNAMICS OF THE BARNACLE *BALANUS AMPHITTRITE* IN THE TROPICAL MONSOON-INFLUENCED ENVIRONMENT OF INDIA
Anil, A 65

MACRO-2 A COMMON THEME IN BACTERIAL INDUCTION OF MACROFOULING
Hadfield, M 65

MACRO-3 DO BACTERIA EAT BARNACLE GLUE?
Essock-Burns, T 66

MACRO-4 OBSERVATIONS ON THE LOCATIONS OF ADRENERGIC-LIKE RECEPTORS IN THE BARNACLE (*BALANUS AMPHITRITE*) CYPRID LARVAE AND OYSTER (*CRASSOSTREA)*
Gohad, N 66

MACRO-5 BIOFOULING – UNSOLVED PROBLEMS, INSUFFICIENT APPROACHES AND SOME LIGHT AT THE HORIZON
Flemming, H 67

MACRO-6 LARVAL BIOLOGY OF THE TROPICAL OYSTER *DENDOSTREA FOLIUM* IN SINGAPORE WATERS
Juhel, G 67

MACRO-7 REARING TEMPERATURE AFFECTS THE ADHESIVE TENACITY OF BARNACLES GROWN ON NON-TOXIC MARINE COATINGS
Daugherty, M 68

MACRO-8 ADHESIVES OF CYPRIDS OF THE BARNACLE BALANUS AMPHRITE AND THE TUBES AND ADHESIVES OF HYDROIDES ELEGANS AND HYDROIDES DIANTHUS
Shi, W 68

MARINE CORROSION MATERIALS AND COATINGS (LEE, J):

MC-1 5XXX ALUMINUM SENSITIZATION AND THE NAVY
Golumbfskie, W 69

MC-2 ALUMINUM ALLOY SUSCEPTIBILITY TO SULFIDE DERIVITIZATION
Lee, J 69

MC-3 SEAWATER FIELD TRIALS OF MATERIAL LOSS IN COPPER ALLOYS IN THE NORTH ATLANTIC
Drach, A. 70

MC-4 BIOFOULING AND CORROSION OF SPRAY COATED CARBON STEEL
Kanematsu, H 71

MC-5 SOME NEW EVALUATION METHODS FOR BIOFOULING ON METALLIC MATERIAS ON LABORATORY SCALE AND THEIR RESULTS
Kanematsu, H 71

MC-6 ANTI-CORROSIVE METAL COATING USING SOL-GEL DERIVED LAYERED ORGANIC-INORGANIC HYBRID
Urata, C 72

MC-7 BIOFOULING ON SPRAY COATED CONCRETES IN MARINE ENVIRONMENT
Kanematsu, H 72

MC-8 THE ROLE OF IRON IN SRB INFLUENCED CORROSION OF STEEL IN SEAWATER
Duan, J 73

NEW APPROACHES TO ANTIFOULING TESTING (CALLOW, M):

NA-1 3D TRACKING OF FOULING EVENTS
Rosenhahn, A 74

NA-2 A NOVEL DESIGN FOR THE *IN SITU* EXAMINATION OF SETTLEMENT BEHAVIOR
Ralston, E 74

NA-3 A NOVEL MICROFLUIDIC APPROACH FOR THE ASSESSMENT OF ANTIFOULING TECHNOLOGIES
Salta, M 75

NA-4 A NOVEL GEOMETRY FOR LABORATORY-BASED LARVAL SETTLEMENT ASSAYS
Petrone, L 76

NA-5 BEHAVIOURAL ANALYSIS OF BARNACLE LARVAE
Aldred, N 76

NA-6 DEVELOPMENT OF ACCELERATED TEST METHODS FOR CHEMICALLY-ACTIVE ANTIFOULING COATINGS
Bressy, C 77

NA-7 NEW SELECTION CRITERIA FOR BIOCIDES IN ANTI-FUOLING COATINGS
Elwing, H 78

NA-8 AN ASSESSMENT OF *ELMINIUS MODESTUS* (DARWIN) A MEMBRANOUS-BASED BARNACLE AS A MODEL SPECIES FOR EVALUATING FOULING-RELEASE COATINGS
Herdman, R 79

POSTER SESSION 2

TP-01 BIOFOULING ON SALMON CAGE NETS IN NORWAY – A ONE YEAR STUDY
Bloecher, N. 80

TP-02 MARINE FOULING ON AQUACULTURE FACILITIES IN THE NORTHERN SOUTH CHINA SEA
Li, Z 80

TP-03 EFFECT OF CAGE DEFORMATION AND BIOFOULING ON THE WELFARE OF FARMED ATLANTIC SALMON *SALMO SALAR*
Perry, B. 81

TP-04 MICROSTUCTURE OF BASE-PLATE OF BARNACLE ON DIFFERENT SUBSTRATE
Chen, X 82

TP-05 DO BARNACLES HAVE COLOUR PREFERENCES IN THEIR CHOICE OF SETTLEMENT SUBSTRATUM?
Dürr, S 83

TP-06 BRICKS AND MORTAR MODEL OF BARNACLE ADHESION
Rittschof, D. 83

TP-07 BETWEEN A ROCK AND A HARD PLACE: STRUCTURAL COMPLEXITY OF THE BARNACLE ADHESIVE INTERFACE
Wahl, K 84

TP-08 INFLUENCE OF SURFACE CHARGE AND SURFACE ENERGY ON THE SETTLEMENT BEHAVIOUR AND 'FOOTPRINT' MORPHOLOGY OF BALANUS AMPHITRITE AND B. IMPROVISUS CYPRIDS
Di Fino, A 84

TP-09 IDENTIFICATION OF SMALL MOLECULE COMPONENTS OF BARNACLE GLUE
Wu, F 85

TP-10 COMPARATIVE TOXICITY EVALUATION OF ANTIFOULING AGENTS ON THE OVARIAN DEVELOPMENT OF BROWN MUSSEL PERNA INDICA
Iyapparaj, P 86

TP- 11 VISCOSLAINE ANALOGS: SYNTHESIS AND ANTIFOULING PROPERTIES
Legrave, N 86

TP-12 PRELIMINARY RESULTS OF THREE NATURAL EXTRACTS FOR ANTIFOULING PAINTS
García, M 87

TP-13 ANTIFOULING POTENTIAL OF MARINE SPONGES FROM BAY OF LA PAZ, MEXICO
Ortiz-Aguirre, I 87

TP-14 SCREENING OF NOVEL ANTIFOULING METABOLITES FROM MANGROVE ASSOCIATED STREPTOMYCES FRADIEA
Prakash, S 88

TP-15 ANTIBACTERIAL AND ANTIFOULING ACTIVITY OF SYMBIOTIC BACTERIA FROM APLYSINA GERARDOGREENI
Aguila Ramirez, R 89

TP-16 ANTIFOULING EVALUATION OF SYNTHETIC MERIDIANINS DERIVATIVES: ANTISETTLEMNT ACTIVITY AND TOXICITY AGAINST BARNACLE LARVAE
Trepos, R 89

TP-17 ANTIFOULING MOLECULE FROM THE BACTERIUM HALOMONAS MERIDIANA ASSOCIATED WITH A MARINE INVERTEBRATE
Viju, N 90

TP-18 INFLUENCE OF EXTRACTANTS FROM THE CALLYSPONGIA SPONGE ON THE LARVAL SETTLEMENT OF ACORN BARNACLE BALANUS (=AMPHIBALANUS) RETICULATUS
Yan, T 90

TP-19 MARINE NATURAL PRODUCT AS NOVEL ANTIFOULDING REAGENTS: STRUCTURE OPTIMIZATION AND BIOLOGICAL EVALUATION
Li, Y 91

TP-20 MOLECULAR LEVEL STUDIES ON ANTI-BIOFOULING AND FOULING-RELEASE POLYMER SURFACES
Liu, Y 91

TP-21 STEREOSCOPIC METHODS FOR THREE-DIMENSIONAL TRACKING OF EXPLORATORY BEHAVIOR OF BARNACLE CYPRIDS
Maleschlijski, S 92

TP-22 IN-SITU STUDY OF BARNACLE CYPRID AND JUVENILE BARNACLE CEMENT USING XRF MICROSCOPE AND MICRO-RAMAN SPECTROSCOPY
Mohamed, T 93

TP-23 ZWITTERIONIC BETAINES AS VERSATILE ULTRA LOW FOULING SURFACE COATINGS OR HYDROGELS FOR REAL-WORLD APPLICATIONS
Brault, N 93

TP-24 THE EFFECTS OF CHARGED AND UNCHARGED XEROGEL SURFACES ON THE ADHESION STRENGTH OF THE BROWN ALGA ECTOCARPUS CROUANIORUM
Gatley, C 94

TP-25 SELF-POLISHING ZWITTERIONIC NONFOULING COATINGS FOR MARINE APPLICATIONS
Jiang, S 94

TP-26 BIO-RESPONSIVE ANTIFOULING COATINGS
Koene, B 95

TP-27 ANTIFOULING ACTIVITY OF COATING CONTAINING MACROCYCLIC LACTONES (IVERMECTIN 0.1%) IN SUB-TROPICAL FIELD TESTS IN BRAZIL
Countinho, R 95

TP-28 A COATING PENETRATION DEPENDENT ANTIFOULING CONCEPT
Pinori, E 96

TP-29 AMPHIPHILIC POLYSACCHARIDES AS INERT SURFACE COATINGS
Finlay, J 97

TP-30 POLYMER BRUSHES AS MODEL INTERFACES FOR MARINE ANTIFOULING
Zhou, F 98

TP-31 SWITCHABLE SURFACES TO REGULATE ADHESION OF MARINE BACTERIA
Pranzetti, A 99

TP-32 NATURAL PEPTIDE NONFOULING MATERIALS AND COATINGS BY COMBINATORIAL AND RATIONAL DESIGN
Keefe, A 100

TP-33 CHEMISTRY DEPENDING SURFACE CONDITIONING AND ITS IMPLICATION FOR COLONIZATION BY MICROORGANISMS
Thome, I 101

TP-34 MIMICKING NATURE THROUGH MODELLING FOR NONFOULING MATERIALS
White, A 101

TP-35 AMBIGUOUS SURFACES FOR ANTIFOULING AND FOULING RELEASE APPLICATIONS
Taylor, W 102

TP-36 FOULING CONTROL USING AMPHIPHILIC COPOLYMERS
Martin-Tanchereau, P 103

TP-37 MULTI-LENGTH-SCALE POLYMER GRADIENTS FOR THE INVESTIGATION OF SURFACE-EXPLORATION BEHAVIOUR OF ZOOSPORES OF THE MACRO-FOULING ALGAE ULVA
Sterne, O 104

TP-38 INFLUENCE OF SURFACE TOPOGRAPHIC GRADIENTS ON ULVA SETTLEMENT
Xiao, L 105

TP-39 EFFECTS OF SURFACE WETTABILITY ON SECONDARY SETTLEMENT OF THE MUSSEL MYTILUS CORUSCUS PLANTIGRADES
Yang, J 105

TP-40 SETTLEMENT OF THE MUSSEL MYTILUS CORUSCUS PLANTIGRADES IN RESPONSE TO NATURAL BIOFILMS
Yang, J 106

TP-41 QUANTIFICATION OF THE ADHESION STRENGTH OF THE DIATOM NAVICULA PERMINUTA IN A MICROFLUIDICAL ASSAY
Alles, M 106

TP-42 FIELD TESTS OF THE EARLY COLONIZATION STAGE OF MODEL SURFACES
Arpa Sancet, M 107

TP-43 DEFOULING LOOP IN YIELDING SETTLEMENT SUBSTRATES
Baum, C 107

TP-44 A NOVEL METHOD FOR BIOFOULING PREVENTION
Heinonen, K 108

TP-45 INVESTIGATION OF TITANIUM'S FOULING IN SEA WATER
Kania, C 108

TP-46 THE EFFECT OF GROOMING AND CLEANING ON THE CONDITIONING OF TRANSPLANTED PANELS WITH SILICONE COATINGS
Ralston, E 109

TP-47 COLONIZATION PATTERN OF BIOFOULING COMMUNITY IN THE COASTAL WATERS OF KALPAKKAM, BAY OF BENGAL, INDIA
Satpathy, K 109

TP-48 OPTIMIZING BRUSH DESIGN FOR THE GROOMING OF SHIP HULL COATINGS
Tribou, M 110

TP-49 FUNCTIONAL ROLE OF FOULING COMMUNITY ON AN ARTIFICIAL REEF AT THE NORTHERN COAST OF RIO DE JANEIRO STATE, BRAZIL
Zalmon, I 110

TP-50 A FIFTEEN-YEAR ASSESSMENT OF FOULING COMMUNITY ASSOCIATED WITH ARTIFICIAL REEF ON THE NORTHERN COAST OF RIO DE JANEIRO, SOUTHEAST
Zalmon, I 111

PLENARY-3 ANTIFOULING REGULATIONS – 25 YEARS ON – IS IT TIME FOR A RETHINK?
Hunter, J **112**

BIOFOULING ISSUES FOR INDUSTRY (TEO, S):

BII-1 EVALUATION OF COATINGS FOR LONG-TERM BIOFOULING PREVENTION ON DEEP SEA STRUCTURES
Lewis, J 113

BII-2 MANAGING NET BIOFOULING IN TASMANIAN CAGE FINFISH AQUACULTURE
Yaxley, B 113

BII-3 INCREASE IN DRAG FORCE ON A TWINE WITH DIFFERENT LEVELS OF HYDROID FOULING
Lader, P 114

BII-4 ENZYMATIC ACTIVITY IN MARINE MICROBIAL BIOFILMS LINKED TO RISK OF BIODEGRADABILITY IN MARINE POLYURETHANE COATINGS?
Heusinger von Waldegge, T 115

BII-5 ANTIFOULING COATINGS: A DRY DOCK PERSPECTIVE
Swain, G 115

BII-6 FIELD TESTING OF FOUL RELEASE COATINGS IN STATIC AND DYNAMIC CONDITIONS FOR WATER RESOURCES INFRASTRUCTURE
Skaja, A........ 115

BII-7 ELIMINATING HULL-BORNE AQUATIC INVASIVE SPECIES – AN ALTERNATIVE, PRACTICAL APPROACH
Phillips, D........ 116

BII-8 THE EFFECTS OF LOW INTENSITY ULTRASOUND ON BARNACLE CYPRIDS
Guo, S 116

MARINE BIOADHESION-BIOADHESIVES (WILKER, J):

MBA-1 RESTRICTION ENZYME ASSOCIATED DNA SEQUENCING OF BARNACLES WITH HARD AND GUMMY GLUE PHENOTYPES ON T2 SILICONE FILMS
Rittschof, D........ 117

MBA-2 A DAY IN THE LIFE OF BALANUS AMPHITRITE: SPATIAL AND TEMPORAL VARIATIONS IN NEWLY FORMED BARNACLE ADHESIVE
Burden, D 117

MBA-3 EFFECT OF SURFACE CHEMISTRY ON SPREADING OF BARNACLE (BALANUS AMPHITRITE) CYPRID PERMANENT CEMENT
Conlan, S 118

MBA-4 THE MULTI-PART ADHESIVE OF THE MARINE SANDCASTLE WORM, *PHRAGMATOPOMA CALIFORNICA*
Stewart, R........ 119

MBA-5 THE INFLUENCE OF POLYMER THICKNESS AND CHARGE ON THE SETTLEMENT AND ADHESION BEHAVIOUR OF MARINE ALGA *ULVA*
Yandi, W 120

MBA-6 COACERVATION IN MUSSEL ADHESION
Joon Cha, H 121

MBA-7 ADHESION MECHANISM IN A DOPA-DEFICIENT FOOT PROTEIN FROM GREEN MUSSELS
Soo Hwang, D 121

MBA-8 CHARACTERIZING OYSTER CEMENT AND MUSSEL GLUE: WHAT IS DIFFERENT AND WHAT IS THE SAME?
Wilker, J........ 122

BIOFOULING CONTROL TECHNOLOGIES-COPPER AND CO-BIOCIDES (BLOSSOM, N AND LINBLAD, L):

CU-1 HIGH PERFORMANCE ANTIFOULINGS BASED ON ORGANOSILYL POLYMERS: RESULTS FROM LONG TERM EXPOSURE UNDER DYNAMIC CONDITIONS
Jackson, S 123

CU-2 EXPERIMENTAL OBSERVATION AND MODELLING OF BIOCIDE RELEASE FROM MODEL ANTIFOULING COATINGS
Goodes, L 123

CU-3 COLD SPRAY ANTIFOULING PROTECTION FOR POLYURETHANE MARINE SEISMIC STREAMERS
Vucko, M 124

CU-4 ANTI-BARNACLE PERFORMANCE OF MEDETOMIDINE IN SOFT (EROSIVE) AND SOLID (NON-EROSIVE) COATINGS
Dahlström, M 124

CU-5 THE ROLE OF ORGANISM ADAPTATION AND ACCLIMATION IN MANAGING COPPER EFFICACY AND TOXICITY
Long, K. 125
CU-6 A UNIFIED SALTWATER-FRESHWATER BIOTIC LIGAND MODEL TO PREDICT THE OLFACTORY EFFECTS OF COPPER TO SALMONID FISHES
Meyer, J. 125
CU-7 EFFECTS OF COPPER ON THE OLFACTORY AND BEHAVIORAL RESPONSES OF SALTWATER FISH, AND THE PROTECTIVENESS OF REGULATORY AQUATIC LIFE CRITERIA USING THE BIOTIC LIGAND MODEL
DeForest, D 126
CU-8 ECO-FRIENDLY SURFACE CHEMISTRIES FOR THE INHIBITION OF BIOFOULER GLUE POLYMERIZATION
Schetz, J. 126

BIOSECURITY/BIOINVASIONS (HOLM, E):

BIO-1 MAKING POLICY SAUSAGE: A BEHIND-THE-SCENES LOOK AT THE DEVELOPMENT OF BIOFOULING MANAGEMENT REGULATIONS FOR THE STATE OF CALIFORNIA
Scianni, C 127
BIO-2 BALANCING BIOFOULING, BIOCIDES AND BUREAUCRACY
Georgiades, E 128
BIO-3 BALANCING FOULING CONTROL STRATEGIES FOR MANAGING INVASIVE SPECIES AND WATER QUALITY: THE ROLE OF HULL COATINGS IN RECRUITMENT
Culver, C 129
BIO-4 BALANCING FOULING CONTROL STRATEGIES FOR MANAGING INVASIVE SPECIES AND WATER QUALITY: THE ROLE OF HULL CLEANING IN RECRUITMENT
Johnson, L 130
BIO-5 MILITARY SEALIFT COMMAND VESSELS: AN IMPORTANT CONTRIBUTION TO THE BIOFOULING VECTOR
Ashton, G 130
BIO-6 DETERMINING BIOFOULING RATES ON A MARINE CONSTRUCTION FLEET WHICH INCREASE OUR PREDICTABILITY OF BIOFOULING AND THE POSSIBLE PRESENCE OF MARINE PESTS
Booth, G 131
BIO-7 SETTING A NEW BENCHMARK FOR RISK ASSESSING MARINE VESSELS WHICH OPERATE IN ECOLOGICALLY SENSITIVE ENVIRONMENTS
Johan van der Merwe, C 131
BIO-8 NOVEL FIELD EXPERIMENTS INVESTIGATING THE EFFECT OF ORGANISM SIZE AND WATER TEMPERATURE ON TREATMENT OF MUSSEL BIOFOULING IN INTERNAL SEA WATER SYSTEMS USING QUATERNARY AMMONIUM COMPOUNDS
Grandison, C. 132

PLENARY-4 MARINE ANTIFOULING: PROGRESS AT THE INTERFACE OF DISCIPLINES
Clare, A **133**

GENERAL ASPECTS OF FOULING-MICRO FOULING (DOBRESTOV, S):

MICRO-1 DOES A BACTERIAL BIOFILM INFLUENCE THE ADHESION STRENGTH OF MARINE ALGAE?
Mieszkin, S 134
MICRO-2 GREEN INTELLIGENT MATERIALS: DEVELOPING NEW SURFACES WITH ANTI-BIOFILMS PROPERTIES
Hellio, C 134
MICRO-3 SUBSTRATE BACKGROUND COLOUR AFFECTS FORMATION OF MICRO- AND MACRO-FOULING COMMUNITIES
Abed, R. 135
MICRO-4 THE EFFECT OF STATIC AND DYNAMIC CONDITIONS ON THE SETTLEMENT AND ADHESION OF DIATOMS TO SHIP HULL COATINGS
Zargiel, K 135
MICRO-5 BIOFILM DISPERSAL, NITRIC OXIDE AND CONTROL TECHNOLOGIES FOR BACTERIAL FOULING
Steinberg, P 136
MICRO-6 FOULING DIATOM COMMUNITY SPATIAL STRUCTURING ON IN-SERVICE INTERSLEEK 900
Longyear, J 136

MICRO-7 MOLECULAR SIGNALS AND METABOLITES FROM ALGAE HAVE A POTENTIAL ROLE IN MARINE MICROFOULING CONTROL
Case, R 137

MICRO-8 ANTIFOULING COATINGS: *IN VITRO* BIOASSAY *VS* MARINE BIOFILM COMMUNITIES
Brian, J 138

MICRO-9 DIVERS, BOATS AND FLYING MACHINES REMOTELY OPERATED VEHICLES AS AN ALTERNATIVE TO DIVERS FOR MARINE VESSEL INSPECTIONS
Burrows, R 139

GENERAL ASPECTS OF FOULING-MISC (HASLBECK, E):

MISC-1 A HARD AND GUMMY PROBLEM: UNDERSTANDING VARIATIONS IN BARNACLE CEMENT PROPERTIES
Barlow, D 140

MISC-2 PHOSPHOPROTEINS IN BARNACLE GLUE
Dickinson, G 140

MISC-3 BIO-INSPIRED ANTIFOULING VIA ACTIVE SURFACE DEFORMATION
Zhao, X 141

MISC-4 IDENTIFYING VIABLE ALTERNATIVES TO COPPER HULL PAINTS FOR THE RECREATIONAL BOATER
Bauer, S 142

MISC-5 AQUEOUS BASED BLOCK COPOLYMER NANOSTRUCTURED COATINGS FOR ANTIFOULING APPLICATIONS
Gunari, N 142

MISC-6 QUANTIFICATION OF FRICTION DRAG INCREASE DUE TO PROPELLER FOULING
Klijnstra, J 143

MISC-7 SHORT- AND LONG-TERM FUEL SAVINGS FROM HYDROGEL-BASED FOULING RELEASE COATINGS
Meseguer-Yebra, D 143

MISC-8 DO BARNACLES HAVE COLOUR PREFERENCES IN THEIR CHOICE OF SETTLEMENT SUBSTRATUM?
Dürr, S 144

MISC-9 MONITORING THE IMPACT OF BIOFOULING ON SHIP POWERING CONDITION – CASE STUDIES FROM US NAVY EXPERIENCE
Haslbeck, E 144

PARTICIPANT LIST 145

INDEX 152

Plenary 1

PLENARY-1

HYDRODYNAMICS AND ENERGY EFFICIENCY

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The International Maritime Organization (IMO) Maritime Environmental Protection Committee (MEPC) issued regulations for the emissions of carbon dioxide from ships in July 2011.

Two initiatives are incorporated into the regulatory regime: (1) the Energy Efficiency Design Index (EEDI), a design target for new ships and (2) the Ship Energy Efficiency Management Plan (SEEMP), a documentation of energy efficiency practices for ships in service.

The MEPC also adopted voluntary guidelines on preventing/minimizing the introduction and/or spread of invasive aquatic species (IAS) via ship hull fouling.

Although the energy efficiency regulations and the guidelines for eliminating negative effects of hull fouling were developed to improve the environmental performance of shipping there are also potential commercial benefits from these regulatory developments. Since the cost of fuel represents a substantial amount of the operating cost for marine transportation, there is also a strong commercial incentive for reducing fuel usage. Fuel consumption is influenced by many factors and managing the hull resistance has a role in helping to minimize fuel usage. Who benefits from the savings in the fuel cost depends on the contractual arrangements in the transportation chain. Therefore, although IMO's goal of energy efficiency and industry's objective to save fuel are complimentary, the economics for the owner or the operator may not always support the investments needed to improve operational efficiency.

As vessels propel forward they have to overcome the hull frictional resistance, wave making resistance, wind resistance, and the added resistance from waves. In general terms, the hull frictional resistance is the major factor for slower ships in calm waters, while the wave making resistance becomes more important as the speed of the vessel is higher. Since hull fouling has a major impact on the hull frictional resistance its management plays an important role in the operational efficiency of ships. While regulations typically impose a cost on the industry, this is an occasions when regulations actually could reduce operating and capital expenses.

The interrelationship between all of the above factors will be discussed within the framework of the SEEMP.

REGULATORY AND ENVIRONMENTAL SESSION
Session Chair: Marianne Pereira

RE-1

ANTIFOULING AND IMO'S GOALS - THE IMPACT OF FOULING PROTECTION ON SHIPPING'S GREENHOUSE GAS EMISSIONS AND TRANSFER OF NON-INDIGENOUS SPECIES

Eivind A. Berg

Jotun AS

The reduction of international shipping's share of CO_2 and other greenhouse gas emissions is the single issue that has generated most debate and work at all the latest meetings in the United Nations' International Maritime Organization's (IMO) important Marine Environment Protection Committee (MEPC). Also, since the adoption of the Ballast Water Convention, IMO has worked efficiently with guidelines to reduce the risk of new marine invasions by species carried on the outside of ships' hulls. These guidelines were adopted last summer. Fouling protection plays an important role with respect to finding efficient measures to meet both to the greenhouse gas (GHG) and the non-indigenous species (NIS) challenge.

The paint industry's International Paint and Printing Ink Council (IPPIC) is a non-governmental organization accredited by the IMO. IPPIC has for years actively been using its status with the IMO to bring information and expertise on both these issues to the relevant IMO committees. IPPIC submitted in 2010 a document to MEPC on the importance of effective antifouling systems for greenhouse gas emissions. Since then the IMO has further developed indexes to quantify how the design and operation of ships affect fuel consumption and a lot of new data have been brought to IMO's attention both from the paint industry and other parties. The conclusion that adequate antifouling protection is a cost-effective and immediate means for significantly reducing the air emissions from shipping has been supported by data from ships. Similarly, the IMO Biofouling Guidelines point at proper selection and maintenance of the antifouling system as a highly important means for managing biofouling risk. The paper will present the latest developments from this year's IMO meetings with respect to consequences for antifouling.

RE-2

REASSESSMENT OF ANTIFOULING BIOCIDES IN NEW ZEALAND

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The New Zealand Environmental Protection Authority (EPA) has established the use of specific scenarios within the MAMPEC model based on the research of the National Institute of Waster and Atmospheric Research[1]. These scenarios better predict the environmental concentrations of antifouling biocides within the New Zealand context. Initial application of inputs into these scenarios indicated that predicted environmental concentrations were markedly higher in New Zealand ports than the OECD default scenarios.

As a result of the development of New Zealand specific scenarios within the MAMPEC model, the EPA is conducting a reassessment of all (14) active ingredients (sea nine, chlorothalonil, copper pyrithione, copper, dichlofluanid, diuron, irgarol, mancozeb, octhilinone, thiram, tolyfluanid, ziram, zinc pyrithione, zineb) approved for use in antifouling paints in New Zealand. Initially, the active ingredients were divided into three categories in terms of expected risk based on recent overseas regulatory decisions:

- “Green”– low expected risk;
- “Orange”– moderate/uncertain level of risk; and,
- “Red”– high expected risk.

This characterization was employed as a communication tool for stakeholders regarding the likelihood of biocides to be subject to stricter controls as a result of the reassessment process. A discussion paper on proposed new controls was also drafted for stakeholder consultation. Stakeholders were invited to comment on the above characterization and the proposed controls and also to provide feedback on required technical information necessary for the risk assessment and on benefits of the use of specific substances.

The methodology used for the environmental risk assessment is based on the TGD[2] and for the human health risk assessment on the TNsG on Human Exposure[3].

At this point (January 2012) the results of the risk assessments and consequent regulatory actions have yet to be finalised. However, these will be concluded by end of May, 2012 according to the project plan timelines and will be presented at the congress.

Acknowledgement - The author thanks Georgiades E, Depree C, Gadd J, Hickey C

[1] Relevance to New Zealand of the OECD Emission Scenario Document for Antifouling Products (2011) Phase 2 Report Prepared for the Environmental Protection Authority (EPA)

[2] Technical Guidance Document on Risk Assessment (2003) in support of Directive Directive 98/8/EC of the European Parliament and of the Council concerning the placing of biocidal products on the market

[3] Human Exposure to Biocidal Products Technical Notes for Guidance (2007) Commission Directive 98/8/EC

RE-3

THE CHANGING SCENE OF REGIONAL REGULATIONS AFFECTING THE ANTI-FOULING INDUSTRY AND THEIR IMPLICATIONS FOR DEVELOMENTS OF NEW ACTIVE SUBSTANCES

Jack Poppleton, Lonza Microbial Control

Nicholas P Skoulis, Lonza Microbial Control

The anti-fouling industry continues to be challenged by changing regulations. The major overhaul of the existing Biocidal Products Directive which is to become the Biocidal Products Regulations and which will enter the European Union statute books in 2012 is a prime example. Other regions also see changing and ever more onerous regulations including Japan, Australia and the US. Issues raised in such regulations include scope, data requirements, risk versus hazard methodologies, data protection and data sharing, and practicability. This presentation reviews the impending changes and looks to the impact of these both upon existing active substances and products but also looks to the significant impact that previous legislation has had for the investment by the industry in new active substances and products. Politicians react to the wishes from the public and other interested parties for ever decreasing impacts of these products on both the environment and human health and purportedly legislate to this end. However, for regulations to be effective they must be both capable of and be seen to achieve the goals set. This presentation further looks to the experiences in the development of potential additive chemistries and the impact that legislation has had on such projects.

RE-4

A REVIEW OF THE GLOBAL STATUS OF ANTIFOULING PAINT REGULATORY MANAGEMENT, WITH A PARTICULAR FOCUS ON THE USE OF COPPER

Kevin W J Long

Regulatory Compliance Limited

Pursuant to the unintended environmental consequences of the use of tributyltin (TBT), over the last two decades national and international regulatory authorities have begun to develop and implement mechanisms to manage TBT alternatives to ensure the risk of similar issues is minimised in the future. Internationally, an ISO draft standard has been developed to describe a risk assessment process on anti-fouling systems on ships and on a regional level, the EU has introduced the Biocidal Products Directive which obliges an assessment of risk for all current and future antifouling substances, describing procedures to allow this. In addition, on a national level, several countries have developed, or are developing, separate schemes to address the hazard and risks associated with antifouling substances, to ensure long-term acceptability and safe use. Various national and international schemes will be presented, focusing on the regulatory status of copper within each of these schemes.

RE-5

REGULATORY IMPLICATIONS OF OLFACTORY AND BEHAVIORAL EFFECTS OF COPPER ON FISH

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Copper Development Association Inc, Webster, NY; and Joseph S. Meyer, ARCADIS U.S., Inc., Lakewood, CO

Aquatic life criteria are derived on the basis of laboratory toxicity studies that quantify the effects of chemical stressors with respect to survival, growth, or reproduction. According to EPA guidelines, criteria derived using these data provide adequate levels of protection for aquatic life as set forth under the Clean Water Act. It has been suggested that sub lethal effects can occur at chemical concentrations below more traditional chronic growth and reproduction endpoints, which has been interpreted as challenging the effectiveness of aquatic life criteria. For copper, it has been suggested that the olfactory systems of salmonids (e.g., Pacific salmon) and other fish are impaired at relatively low concentrations that ultimately lead to behavioral responses (e.g., defense against predation) that negatively impact fish populations. However, the ultimate protectiveness of these regulatory criteria are dependent on 1) the relevance of these other sub lethal endpoints with regard to ecological or recreationally important species, 2) the extent to which criteria derived for all aquatic life are also protective of any one group of aquatic organisms, and 3) the effects of water quality on the response of that endpoint or organism. This presentation will compare the olfactory and related behavioral effects of copper on fish compared to both freshwater and saltwater regulatory aquatic life criteria. Although some sub lethal studies suggest adverse impacts at relatively low concentrations, we argue that aquatic life criteria are protective of all aquatic life at levels of protection set forth under the Clean Water Act, particularly when the influence of water quality on copper bioavailability is properly considered.

Acknowledgements: This work was funded by the Copper Development Association, Inc. and the International Copper Association Ltd.

RE-6

PROPOSED U.S. AMBIENT WATER QUALITY CRITERIA FOR COPPER IN SALTWATER: OVERVIEW AND EXAMPLE APPLICATION

Robert Santore, Adam Ryan - HDR|HydroQual, Syracuse NY*

Casey E. Capolupo- University of San Diego, San Diego, CA

Gunther Rosen, Patrick Earley, Brandon Swope, Ignacio Rivera-Duarte, - SPAWAR System Center Pacific, San Diego, CA

Charles Delos - US EPA

A draft update to the US EPA ambient water quality criteria for copper in saltwater has recently been developed. This update incorporates a large amount of new marine and estuarine toxicity data that have been published since the last saltwater document for copper was released by US EPA[1]. This update also incorporates bioavailability relationships using a saltwater Biotic Ligand Model (BLM) for copper in a manner similar to the use of the BLM in the US EPA ambient criteria document for copper in freshwater[2]. The importance of bioavailability in determining metals criteria has long been recognized by the U.S. EPA and a provision for developing site-specific modifications to criteria that considers bioavailability has existed based on Water Effect Ratios (WERs) [3]. The practical utility of the direct incorporation of the BLM in the revised criteria is that for sites where bioavailability is important, the cost and effort associated with developing a WER is no longer needed. To evaluate the effectiveness of the updated approach using the saltwater BLM for copper, an example application of the model has been conducted for Shelter Island Yacht Basin (SIYB) in San Diego Bay, CA. For this evaluation, ambient toxicity in SIYB was assessed, and a site-specific modification to the saltwater criteria for copper was developed for SIYB using a WER approach. Both of these approaches are based on measured copper toxicity to the mussel *Mytilus galloprovincialis.* The saltwater BLM calculation requires just three parameters that describe the water chemistry at the site: pH, salinity, and dissolved organic carbon. This presentation will compare the results from each of the two approaches and will discuss the costs and relative merits of each.

[1] U.S. Environmental Protection Agency, Glen Thursby and David J. Hansen. 1995. Ambient Water Quality Criteria - Saltwater Copper Addendum (Draft). EPA 440-5-80-036.

[2] U.S. Environmental Protection Agency; Luis A. Cruz, Cindy Roberts, Mary Reiley, Robert Santore, Paul Paquin, Gary Chapman, Jennifer Mitchell, Charles Delos, Joseph Meyer, Rooni Mathew, and Tyler K. Linton. 2007. Aquatic Life Ambient Freshwater Quality Criteria - Copper. 2007 Revision. EPA-822-R-07-001.

[3] US EPA. 1994. Interim Guidance on Determination and Use of Water-Effect Ratios for Metals. EPA-823-B-04-001. National Technical Information Service, Springfield, VA.

RE-7

REGULATORY PRESSURES ON RECREATIONAL BOATING: DO ALTERNATIVE PAINTS OFFER SOLUTIONS?

Karen Holman

Port of San Diego

Boat hull paints have been used for years to prevent marine organisms from fouling the boat hull, which can result in a loss of speed, maneuverability and damage to the boat hull. Antifouling paints work by using a biocide, or pesticide, that leaches into the surrounding water and is toxic to the fouling organisms. Although antifouling hull paints are an effective tools for protecting boat hulls and preventing fouling attachment, the biocide from these paints has been linked to adverse impacts on the water, sediment, and the marine environment.

Due to the phase out of tributyltin (TBT), copper soon emerged as an effective and popular antifouling product in hull paints. Copper-based paints are now the preferred paint for recreational boaters. Currently, scientists are realizing that an over-reliance on copper paints can result in water quality problems.

The elevated copper level in marina basins has led to various regulatory measures at the local, state and federal level. In San Diego specifically, the Shelter Island Yacht Basin has been issued a Total Maximum Daily Load (TMDL), requiring a 76 percent reduction of copper loading by 2022, with other marina basins throughout California soon to follow.

Studies previously completed by the Port of San Diego show that alternative hull paints can be viable solutions for recreational boaters. Paint manufacturers are continuing to improve on their formulations, making the paints easier to apply and more affordable. Providing boaters with effective alternatives to copper hull paint is a necessary first step before regulations oblige boaters to make the change. This presentation will focus on how regulatory pressures may start to impact a boater's choice of hull paint, and focus on the various mechanisms that can be used to encourage a successful transition to copper alternatives.

RE-8

TECNOLOGIC AND SCIENTIFIC FEASIBILITY OF ADAPTIVE MANAGEMENT ON ECOLOGICAL RISK CAUSED BY ANTIFOULING AGENTS

Kiyoshi SHIBATA

Chiba Institute of Technology

Environmental regulation on antifouling agents has been well developed based on risk based principle. However, environmental fate and concentration of antifouling agents are not understood well. The author himself developed a simulation model to predict the concentration in aquatic environment and found that the calculated concentration is depending on various boundary conditions. The predicted concentration changes with time, location, weather, etc. That is so sensitive that it is difficult to predict. The uncertainty might lead overestimation of the risk.

To cope with uncertain in environmental risk, adaptive management has been developed, especially in fishery or forestry industries. The adaptive management is based on strategic combination of step-wise trial and environmental monitoring. If this strategy can be applied for the environmental risk management of antifouling agents, it may encourage developing new antifouling agent or wise use of existing new antifouling agent.

In this presentation, possibility and future task in environmental risk management using the concept of adaptive management will be discussed.

BIOFOULING CONTROL TECHNOLOGIES-NEW COATINGS FOR AF AND FOULING RELEASE
Session Chair: Dean Webster

AF-1

FOULING IN THE FACE OF A "LITTLE" SURFACE ROUGHNESS

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Readily processible coatings based on integration of hydrophobic materials with hierarchical micro/nanoscale roughness were produced using silica sol-gels[1]. These generate superhydrophobic coatings (SHC) that exhibit water contact angles in excess of 160º and low hysteresis (< 10º). Subtle variations in the micro/nanoscale coatings architecture have resulted in surfaces that exhibit marine antifouling properties[2]. The origin of this behavior is investigated with synchrotron small angle x-ray scattering (SAXS). This technique, sensitive to local changes in electron density, is adapted to the study of nanoscale wetting of immersed interfaces[3]. The experimentally observed variations in wetting behavior were examined with respect to settlement assays of major fouling species (*Amphora sp., Ulva rigida, Polysiphonia sphaerocarpa, Bugula neritina, Amphibalanus amphitrite*).

Macroscopic wetting behaviour does not appear to exert any major effect, as initial contact angles are shown to be independent of subtle changes in nanoroughness. The same changes however have a significant influence on the settlement of tested fouling organisms. Enhancing hierarchical nature of the surface structures resulted in even higher attachment inhibiting behaviour[4]. The concept of experimentally measured pseudo-fractal dimension to quantify these subtle roughness changes will be discussed and possible correlation with fouling behaviour examined.

1. Zhang, H, Lamb, R. and Jones, A. *US Patent* 6743467 (1999)
2. Zhang, H.; Lamb, R. and Lewis, J., *Sci & Tech Adv. Materials* (2005), 6, 236- 239
3. Zhang, H., Lamb, R. and Cookson, D. J. *Applied Physics Letters* 91, 254106, (2007).
4. Scardino, A., Zhang, H.,Cookson, D. J.,de Nys R, Lamb R, *Biofouling* (2009), 25, 757-67.

AF-2

HYDROGELS AS NON-FOULING SURFACES FOR MARINE APPLICATIONS

Stefan M Olsen, Peter Thorlaksen, Anders Blom, Dorthe Hillerup, Ulrik Bork, Micha Berner

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Hydrogels are well-known for their resistance to fouling[1]. They are exploited widely in the biomedical industry to hinder the accumulation of bacteria and biopolymers on artificial surfaces[2]. Since 2008, hydrogel technology has also been exploited commercially in marine fouling release coatings (e.g. Hempel's Hempasil X3[3]). The non-fouling working mechanism of a hydrogel is connected to its ability to retain water in a thin layer at the surface of the coating. The effect of this water layer (the hydrogel) on fouling molecules and organisms is not yet understood in full detail. However, it is generally recognised that a hydrogel, consisting primarily of water, is a poor substrate for water-suspended foulers for two reasons. Firstly, the molecules constituting the non-water part of the hydrogel are suspected to sterically hinder the access to the surface of the molecules involved in the fouling adhesion process. Secondly, the bound water of the hydrogel does not offer physico-chemical properties that differ significantly from the bulk water. Therefore, globular proteins such as bio-adhesives have very little or no entropic gain from adsorbing to the surface[1].

This paper describes the investigation of different experimental hydrogel-based marine fouling release coatings, by surface characterisation and gravimetrical means. The characterisation methods have been applied in order to correlate hydrogel parameters to antifouling performance. The results indicate that the ability of a hydrogel based fouling release coating to retain water only at the surface of the coating can be related to the long term antifouling potency.

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1: Ekblad T. Hydrogel coatings for biomedical and biofouling applications. Linköping studies in science and technology. Dissertation no 1302. 2010.

2: Henriques M, Sousa C, Lira M, Elisabete M, Oliveira R, Aliveir R, Aceredo J, Optometry and vision science. Vol 82 (6). 2005.

3: Thorlaksen P, Yebra D M, Catalá P. Hydrogel-based third generation fouling release coatings. EUROCOAT. 2009.

AF-3

MACROMOLECULAR DESIGN OF SEBS AND PDMS BASED AMPHIPHILIC COATINGS FOR FOULING RELEASE APPLICATIONS

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A recent concept in designing a coating for antifouling/fouling release purposes exploits the interactions intervening between the fouling organisms and the specific surface features at the nanoscale.[1]

We are developing fouling release coatings in which a surface heterogeneity at the nanoscale is created by means of the thermodynamically driven phase segregation of polymer assemblies with varied architectures.[2]

By following this strategy, novel copolymers and terpolymers based on styrene monomers carrying poly(ethylene glycol), poly(dimethyl siloxane) and perfluoroalkyl pendant chains were prepared. The amphiphilic character of the polymers was systematically tuned by the appropriate combination of the hydrophobic, hydrophilic and hydrophobic/lipophobic properties of the constituent counits. The polymers were then dispersed in two different elastomeric matrices (SEBS and PDMS) in order to obtain polymer films with both low elastic modulus and low surface tension. The surface segregation of the surface-active polymer in the blend was assessed by X-ray photoelectron spectroscopy (XPS), atomic force microscopy (AFM) and measurements of contact angles with different probing liquids. PDMS based coatings were proven to have better fouling release performances against Ulva sporelings than the corresponding SEBS based coatings. Notably, coatings incorporating a copolymer with poly(ethylene glycol)-poly(dimethyl siloxane) side chains had superior fouling release properties. The data show that the intrinsic amphiphilic nature of the coatings can add in a synergistic way to create a dynamic, chemical and topographical surface complexity that can deter settlement of fouling organisms and reduce the interfacial bonding with bioadhesives.

Acknowledgments. The work was funded by the EU-FP7 Marie Curie Initial Training Network 'SEACOAT' and the Italian MiUR (fondi PRIN 2008).

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AF-4

DEVELOPMENT OF ENVIRONMENTALLY BENIGN, DURABLE AND EFFECTIVE ZWITTERIONIC-BASED MARINE COATINGS

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In this work, I will overview our efforts to develop zwitterionic-based ultra low fouling surface coatings. Key parameters to control coating performance in complex media and interactions of these coatings with a variety of biomolecules and microorganisms will be discussed. Two types of zwitterionic-based marine coatings will be highlighted – (a) self-polishing/nonfouling and (b) low fouling/high fouling-release. The objective of this work is to develop non-toxic, durable, effective, and low-cost coatings with excellent mechanical strengths for marine applications.

For self-polishing/nonfouling coatings, marine microorganisms cannot attach. These coatings are as effective as anti-fouling coatings, but do not contain or leach any metal ions or biocides while they are much more effectively than conventional fouling-release coatings, particularly at low ship moving speeds. For low fouling/high fouling-release coatings, an integrated zwitterionic and PDMS technology is developed. These coatings have very low fouling. At the same time, microorganisms can be easily released if they are attached onto these coatings. These coatings can be easily applied using a spray gun to a surface covered with an epoxy primer. They are tested extensively in both laboratory and field tests.

Acknowledgment: This work has been supported by the Office of Naval Research.

AF-5

XEROGEL COATINGS WITH ANTI-FOULING AND FOUL-RELEASE PROPERTIES FROM LONG-CHAIN ALKYL AND FLUOROALKYL COMPONENTS

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Organically-modified silicon-derived xerogels (ormosils) are relatively ≨ 25 μm), robust, optically transparent coatings that function as anti-fouling (AF) and foul-release (FR) materials. Xerogels can be applied via dip coating, spin coating, spraying or brushing to a variety of surfaces including aluminum, glass, fiberglass, and epoxy-primed surfaces. By their physical nature, xerogels are porous and reagents found in seawater (halide salts, oxygen, hydrogen peroxide) can permeate the xerogel surface and reach additives that are physically entrapped or covalently attached in the xerogel. As an example, hydrogen peroxide and the bromide/iodide found in seawater can produce oxidized species that discourage the settlement of marine organisms.

Ormosils are prepared by the acid-catalyzed hydrolysis of organically-modified trialkoxysilanes in the presence of tetraalkoxy silanes to give an oligomeric "sol" precursor to the xerogel with a typical molecular weight on the order of 3000-4000 amu. Additives at this point become physically entrapped in the sol or covalently attached to the sol. After the sol is applied to the surface, further crosslinking to the xerogel film occurs with evaporative loss of alcohol or water from the sol pecursors.

Four-component ormosils prepared from sols containing a few mole-% of long-chain alkyl(trialkoxy)silane and tridecafluorooctyl(trialkoxy)silane components in primarily an octyl(trialkoxy)silane/tetraethoxysilane matrix give spontaneous segregation of fluorocarbon and hydrocarbon phases on the xerogel surface as detected by AFM and IR reflectance microscopy. These surfaces function as AF/FR materials toward juvenile barnacles and the macrofouling marine alga *Ulva* in laboratory assays. Field testing of 4" × 8" panels gives results consistent with those observed in the laboratory assays. Imidazolium-based ionic liquids that activate peroxide can be covalently attached in the four-component xerogels, which give improved AF characteristics without sacrificing FR characteristics in both laboratory assays and field testing of 4" × 8" panels.

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AF-6

NONFOULING RESPONSE OF DIFFERENT HYDROPHILIC UNCHARGED POLYMERS

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Marine biofouling has long been acknowledged as a persistent problem with severe consequences both in the ecologic and economic point of view[1]. For this reason several techniques are being studied in order to prevent or diminish accumulation of biomass in wetted surfaces[2] among which the use of polymeric ultra-thin films has been gaining a growing interest. Many polymers are being studied towards this purpose but no work has focused on developing a protocol that allows a reliable comparison between the efficiency of different well-known nonfouling polymers. This is one of the aims of this work and has been achieved by using a common, azide-terminated monolayer to which different non-fouling polymers, such as PEG, PEOXA, PVP, PVA and Dextran with various molecular weights, have been covalently bound. The nonfouling surfaces were then subjected to a comparative biological study by exposure to complex proteins solution, marine bacteria, *Ulva* zoospores and barnacle cyprids, in order to validate the developed protocol and evaluate performances against a wide range of organisms. Further characterization and structure-property relationship study with these latter results was performed by using techniques such as dynamic contact angle, XPS, ellipsometry, QCM-D and AFM.

Acknowledgements:

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AF-7

NOVEL ZWITTERIONIC COATINGS BASED ON RING OPENING METATHESIS POLYMERIZATION

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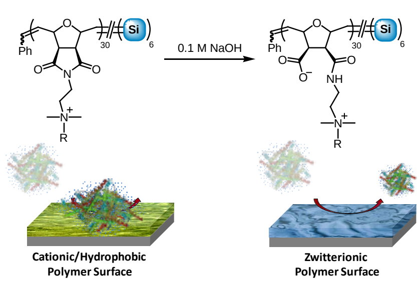
Foundational materials for non-fouling coatings were designed and synthesized from a series of novel dual-functional zwitterionic polymers, Poly[NRZI], which were easily obtained via ring-opening metathesis polymerization (ROMP) followed by a single step transformation of the cationic precursor, Poly[NR(+)], to the zwitterion, Poly[NRZI]. The resulting unique dual-functional structure contained the anion and the cation within the same repeat unit but on separate side chains, enabling the hydrophilicity of the system to be tuned at the repeat unit level. These dual-functional zwitterionic polymers were specifically designed to investigate the impact of structural changes, including the backbone, hydrophilicity, and charge, on the overall non-fouling properties. To evaluate the importance of backbone structure, and as a direct comparison to previously studied methacrylate-based betaines, norbornene-based carbo- and sulfobetaines (Poly[NCarboZI] and Poly[NSulfoZI]) as well as a methacrylate-based sulfobetaine (Poly[MASulfoZI]) were synthesized. These structures contain the anion-cation pairs on the same side chain. Non-fouling coatings were prepared from copolymers, composed of the zwitterionic/cationic precursor monomer and an ethoxysilane-containing monomer. The coatings were evaluated by using protein adsorption studies, which clearly indicated that the overall hydrophilicity has a major influence on the non-fouling character of the materials. The most hydrophilic coating, from the oligoethylene glycol (OEG)-containing dual-functional betaine, Poly[NOEGZI-*co*-NSi], showed the best resistance to non-specific protein adsorption (Γ_{FIB}=0.039 ng/mm^2). Both norbornene-based polymers systems, Poly[NSulfoZI] and Poly[NCarboZI], were more hydrophilic and thus more resistant to protein adsorption than the methacrylate-based Poly[MASulfoZI]. Comparing the protein resistance of the dual-functional zwitterionic coatings, Poly[NRZI-*co*-NSi], to that of their cationic counterparts, Poly[NR(+)-*co*-NSi], revealed the importance of screening electrostatic interactions. The adsorption of negatively charged proteins on zwitterionic coatings was significantly less, despite the fact that both coatings had similar wetting properties. These results demonstrate that the unique, tunable dual-functional zwitterionic polymers reported here can be used to make coatings that are highly efficient at resisting protein adsorption.

Acknowledgments: ONR is acknowledged for financial support.



AF-8

BOTTLE BRUSH-NANOGLASS: A NEW HYBRID APPROACH FOR ENGINEERED, FOULING RELEASE, FLUOROUS POLYOXETANE COATINGS

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Hard and soft fouling that forms on hulls of naval vessels in the marine environment constitute a problem resulting in increased energy to achieve a given speed. Fouling release coatings promise easy cleaning but basic understanding of complex requirements is lacking. Thus, interdisciplinary research holds promise for the evolution of a truly “green” FR coating with high performance. Current coatings having the greatest promise for advanced FR are those based on silicones and fluoropolymers, but those comprised of a combination of hydrophilic and hydrophobic or omniphobic groups represent a new direction for nontoxic fouling release coatings.

This research involves a new hybrid “bottle brush”-nanoglass (BB-NG) approach, which provides engineered control of near surface modulus and surface free energy. Feasibility studies for BB-NG hybrid coatings has focused on the fluorinated “P[3FOx]” bottle brush having a 1,3-polypropylene oxide main chain and semifluorinated side chains. Guiding this development is a new theoretical model that combines mechanical and surface free energy considerations. In order to quantitatively correlate the model with experiment, a new test for adhesion has been devised using a TA Instruments RSA III DMA. A combination of mechanical and spectroscopic characterization has provided important insight into the correlation of composition and near surface morphology with adhesion. Initial field measurements for the force required to remove hard fouling will be correlated with laboratory determined measurement of release. In turn, these results will be discussed in light of theory for release energy that contains terms for modulus, thickness, and work of adhesion.

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SHIP HYDRODYNAMICS AND ENERGY EFFICIENCY
Session Chair: Michael P. Schultz

SH-1

A REPORT ON SOME RECENT DEVELOPMENTS IN SHIP HULL AND PROPELLER COATINGS WITH A VIEW TO FOUL RELEASE TYPE ANTIFOULINGS

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Because of the increasing environmental pressures allied with energy efficiency enhancement of ships, the current antifouling systems are under close scrutiny. The complete removal of the TBT based antifouling systems from commercial shipping has placed the foul release type coating system as the real contender to the Self-Polishing Copolymer (SPC) type antifouling systems. Apart from their almost fully environmental friendly characteristics, there are other performance improvement myths, e.g. relatively smoother and hence better drag performance over SPCs, that have been attracting the attention of ship owners not only for hull coating but also for propeller coating.

According the data base of one of the world's major stake holding paint manufacturers, the current share of the foul release coating applications on ship hulls is approaching to a 10% of the total paint applications on commercial ships whilst the number of coated propeller has reached to over 200 with an increasing rate during the last decade.

While the above developments have been taking place there are current concerns about the foul release coatings including, e.g.: their relative drag performance over the SPC types; different types of foul-release coatings; deleterious effect of biofilm on their performance; their effect on ship performance; robust measurements of their surface characteristics in a dry dock; and worthiness of their applications on propellers.

This presentation is a relatively recent review of the developments in foul release coatings to discuss some of the concerns based on the presenter's own experience through some hydrodynamic research and development activities conducted in his institution over the last decade. These include relative drag performance comparisons of foul release coatings over the SPC types using different type hydrodynamic testing facilities; comparative hydrodynamic performance comparisons of newly developed different nano-structured foul release coatings; surface characterisation of foul release coated surfaces with recently enhanced BMT roughness analyser equipped with a laser sensor; development of a new flow cell facility in combination with a full-scale biofilm samples collector facility on board of a ship to study for the hydrodynamic effect of biofilms on foul release coatings.

The presentation will continue with an update on the propeller coating including a short historical review to follow with the recent upsurge in the foul release coating applications. Some performance myths associated with the open water efficiency of propellers, cavitation inception and full developed cavitation as well as the radiated underwater noise will be discussed based on the presenter's research and development work in his institutions. Interaction of a foul release coating on the propeller based on full-scale observations and other practical aspects of the propeller coatings including the potential saving in propeller maintenance will be highlighted.

SH-2

EVALUATING HULL FOULING ON SHIP PERFORMANCE: UNDERWATER PHOTOS IN CONNECTION WITH SPEED/FUEL CONSUMPTION ANALYSIS

Daniel Kane

Co-Founder

Propulsion Dynamics Inc.

With the increasing demand by the International Maritime Organization for improving ship fuel efficiency and mitigating the translocation of hull borne invasive species, the focus on hull condition from a fouling view point is higher than at any time in maritime history. While there is some theoretical formulas and statistical information that demonstrate the detrimental effect of slime and marine growth on ship fuel efficiency, there has not been a lot of independent publicized case studies involving underwater photos of hull coating condition and corresponding ship fuel efficiency.

Rather than focus on the biological aspects of hull fouling or the methods of analyzing ship performance data - the purpose of the presentation paper is to display actual underwater photos before and after in-water hull cleanings on a variety of commercial vessels and the direct real life effect on speed and fuel efficiency improvements. Photos will cover a wide range of anonymous merchant ships and hull coating types, including Foul Release and Self-Polishing Copolymers and different cleaning methods, along with precise ship speed and fuel consumption measurements before and after cleanings.

The evidence from these case studies covers the past half a decade and indicates the economic and environmental importance of maintaining a smooth hull and propeller and efficiency of newer hull cleaning methods.

KEY WORDS: Hull and propeller condition; ship technical performance; fuel efficiency; maritime emission; hull resistance; added resistance; hull coatings; hull husbandry.

SH-3

HULL AND PROPELLER FOULING PENALTIES ON AUSTRALIAN ARMIDALE CLASS PATROL BOATS

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The Royal Australian Navy (RAN) operates in varying geographical localities which each pose unique biofouling challenges. The Armidale Class Patrol Boats (ACPBs) face some of the most severe biofouling pressure being deployed predominately in tropical waters. The aluminium hulled ACPBs are covered with fouling release coatings (FRCs). This presentation outlines various ship trials conducted to assess the hull and propeller fouling penalty on the ACPBs. Torsionmeters has been fitted to one ACPB and torque and shaft power has been recorded over two years. Regular diver inspections have mapped the rate of hull and propeller fouling. Significant increases in shaft power at fixed speeds have been recorded which correlates to the emergence of calcareous fouling on the hull. The resultant increased fuel usage has been calculated. Measurable improvements in speed have been recorded after propeller polishing trials. The results of new FRC trials on the hull and propeller will be outlined including in-dock support options for stationary vessels.

SH-4

PERFORMANCE OF FOULING RELEASE COATINGS ON NAVY PROPELLERS

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Unprotected propellers on U.S. Navy vessels rapidly accumulate extensive surface roughness from biofouling and from calcium carbonate deposits related to impressed current cathodic protection systems. This increase in roughness degrades the efficiency of ship operations by increasing drag and decreasing the lift and thrust coefficients of the propellers. To mitigate this loss in performance, the propeller blades of a surface combatant were painted with a fouling release coating system in November 2008. Over 2.25 years, the coating has minimized accumulations of mineral deposits and biofouling, and while the other ships in the class required an average of six interim propeller cleanings, the ship with coated propellers required none. On top of these maintenance savings, there is the added benefit of fuel savings from operating with a smoother propeller surface. Calcium carbonate deposits are estimated to have a Rubert C/D roughness ranking that corresponds to a 0.7-1.9% fuel penalty, and heavily biofouled propellers on surface combatants can reduce powering efficiency by 6.5%. Here we present data from the ongoing demonstration and relate the observations to fuel savings for the Navy. Additionally, we will discuss service life, payback periods, and the potential to use other candidate coating systems and other ship classes.

SH-5

METHODOLOGY FOR SHIP'S HULL PERFORMANCE ANALYSIS AND ANTIFOULING MONITORING

Andreas Krapp

Jotun A/S

Fouling on a ship's hull leads to an increase in frictional drag and thereby to increased fuel consumption and greenhouse gas emissions. Being able to quantify the impact of fouling of in service vessels is prerequisite to being able to manage the ship hull condition in a rational way and to choose from different surface treatment possibilities. The potential economic and ecological impact of reliable hull fouling monitoring is considerable given the fact that the market average overconsumption due to hull fouling is estimated to be at least 15%. This quantification, however, is not a trivial endeavor for a vessel in service. The present paper will give an overview over these challenges and about different proposed strategies to quantify the impact of fouling. Under the perspective that the output of hull performance analysis is used for comparing and documenting the effect of different hull surface treatments for single vessels, but also between different vessels, an hull performance analysis standard method is desirable. Such a standard should be based on the principles of transparency, generality and simplicity. The present paper will present such an open and transparent analysis method based on the statistical treatment of automatically logged measurement data from ship's propulsion and navigation systems. Several case studies will be discussed in detail.

SH-6

FLOW-GENERATED FORCES ON HULL FOULING ORGANISMS AND HYDRODYNAMIC SELF-CLEANING OF FOULING-RELEASE COATINGS

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Fouling-release coatings represent a non-toxic approach to the control of ship hull fouling. These coatings allow organisms to attach but promote their release from the hull as a result of hydrodynamic forces experienced during ship transit (hydrodynamic self-cleaning). More complete knowledge of the forces experienced by fouling organisms improves predictions of the efficacy of fouling-release coatings. These data also provide performance targets for polymer chemists or materials developers. An experimental approach is used to measure the flow forces on organisms recognized as important ship hull foulers. Initial experiments focus on three types of sessile invertebrates; barnacles, serpulid tubeworms and bivalve mollusks. Organism models are made by scanning representative specimens with a laser, producing three-dimensional point clouds that are processed into CAD files. The models are then reproduced at an enlarged size scale by rapid prototype manufacturing. Hydrodynamic testing is performed in a water tunnel with the models mounted on a force balance embedded in a plate. The three orthogonal forces on the models are measured as they are rotated at all angles to the flow. Screens are used to replicate the boundary layer velocity profiles flowing over the models to simulate the effect of different locations of the fouling organisms along the ship hull. This approach and the results may also be applicable to sessile invertebrates occurring in wave-swept or other hydrodynamically-challenging environments. This research was funded by the NSWCCD ILIR program.

SH-7

FOULING-RELEASE OF BARNACLES AND ALGAE FROM A SHIP HULL

Lena Granhag

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Fouling-release of various life stages of the barnacle *Balanus improvisus* and the green alga *Ulva* sp. was measured from different surfaces on a boat hull. Barnacles settle as 1 mm cypris larvae and green algae as 10 µm spores that grow into adults of cm-size. The hydrodynamic forces acting on the different life stages of fouling organisms vary as the organisms change size and shape during development. It is of interest to find the flow speeds required for fouling-release of the different life stages. In measurements of fouling-release under defined hydrodynamic conditions and flow speeds of 20 knots significantly more newly settled cypris larvae were removed than metamorphosed barnacles. The detachment failure mode is believed to be different between the two barnacle stages. The material tested were two silicone based products and a reference Plexiglas (PMMA). For the alga *Ulva* sp. newly settled (12 h) spores were difficult to remove in 20 knots while a large proportion of the 3 week old sporelings were removed in 10 knots. The density of barnacles was not found to influence detachment, whereas for algae detachment increased with higher density. The importance of bacterial film composition for the early growth in *Ulva* sp. is also described. Based on field trials, laboratory experiments and calculations, the flow speeds critical for fouling-release of various life stages of *Balanus improvisus* and *Ulva* sp. is presented and discussed. The results are important from coating development and user perspective. Ships spending only short periods in port may be self-cleaned of barnacle-larvae while for metamorphosed barnacles a considerably higher speed is needed for detachment.

SH-8

STUDY FOR FRICTION RESISTANCE AND GEOMETRY OF SURFACE ROUGHNESS OF ANTIFOULING PAINT

Hirohisa Mieno

Chugoku Marine Paint

Depletion of fossil fuels and global warming resulting from emission of Green House Gas are global concerns. International shipping cause about 3 % of the worldwide GHG emission. In July 2011 IMO adopted a revision of MARPOL Annex VI to make establishing the Energy Effective Design Index (EEDI) for new build ships. EEDI for new build vessels has to be calculated in the design stage and confirmed by sea trial. The target EEDI will be tightened step by step until 2030. The EEDI is largely determined by a ships resistance. The ships' hull roughness is one of the most important factors for the resistance.

In this study friction measurements of cylinders painted with antifouling paint was conducted using Double Rotating Cylinder equipment. Surface roughness was measured minutely with a Laser Displacement Meter. The roughness parameters such as Rt50, Rzjis, Ra, Rq (amplitude parameter) Sm(wavelength) Sk, Ku were calculated. A strong correlation between Rt50, Rzjis, Ra, Rq was confirmed, and Sk and Ku approximate to 0 and 3 respectively. The friction resistance caused by surface roughness can be estimated using these amplitude and wavelength parameters. Foul Release Coatings and the newest generation self polishing antifouling paint showed lower Rt50 and longer Sm values. These two aspects result in a lower Friction Increasing Ratio (FIR) than that of currently available antifouling paint. Both lower roughness and longer wavelength are important for reducing the friction resistance. Actual ship hull surface roughness is currently measured using a BMT Analyzer that can only measure Rt50. However as has been shown above, Rt50 values only is not enough information to predict the friction. Therefore Replicate Method was developed for analyzing the hull roughness geometry. This roughness analysis with both roughness amplitude and wave length parameters is very useful to predict the ships' friction resistance more accurately.

BIOFOULING CONTROL TECHNOLOGIES-BIO-INSPIRED COATINGS
Session Chair: Shaoyi Jiang

BI-1

BIO-INSPIRED ANTIFOULING MATERIALS – TRANSLATING INSPIRATION INTO APPLICATION

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Bio-inspired antifouling technologies build on an understanding of the mechanism of action and efficacy of natural antifouling defences. Marine organisms utilise multiple strategies combining physical, mechanical and chemical mechanisms to deliver the most effective broad spectrum antifouling defence. As we identify natural models and begin to understand the complexity of natural defences, particular in terms of their mechanism(s) of action, we are also beginning to understand the complexity of mimicking these systems. This translation from inspiration to application, with the manufacture of a surface that can control micro- and macro-fouling, is a significant and difficult challenge. In this presentation we review our research into bio-inspired materials that combine surface material properties, surface micro-topography, activated surface chemistry and biomolecules. We highlight our successes, and failures, in translating bio-inspired mechanisms into innovative materials to identify principles that can contribute to improved antifouling materials.

BI-2

STRUCTURE-ACTIVITY RELATIONSHIPS OF AMIDES IN ANTI-SETTLEMENT OF DIFFERENT FOULERS

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In this paper, we present a QSAR study for some amides[1] which prevent the settlement of larval of four phyla of common fouling organisms, namely, barnacles *Amphibalanus(=Balanus) amphitrite*; tubeworms *Pomatoleios krausii*, bryozoans *Bugula neritina*, and colonial ascidians *Polyclinum* sp. Our ultimate goal is to develop a series of rapidly biodegradable paint additives that disrupt the metamorphic cascade in macrofouling organisms. A series of amides with varying structural features have been synthesized and assayed for their ability to disrupt larval settlement. The ED50 for the two hard foulers, barnacles and tubeworm, were relatively similar, whereas ED50 against the other two non-planktonic larvae was much higher. The results were also examined in relation to physical and structural properties of the compounds. From the calculated logP values of the compounds, it appears that the hydrophobicity of the compounds may have an impact on how they are taken up by the larvae.

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BI-3

A FIRST-PRINCIPLES THERMODYNAMIC MODEL FOR PREDICTING RELATIVE ATTACHMENT OF FOULING ORGANISMS TO ANTIFOULING TOPOGRAPHIES

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The preparation and implementation of materials to deter biofouling in the marine and medical environments has gained considerable interest over recent years. Our group has focused primarily on the use of microtopographies to deter attachment. These topographies have been shown to be effective against a variety of organisms, but their antifouling mechanism remains unknown. We have developed a model based in first principle thermodynamics to explain the antifouling effect of microtopographies and help aid in the development of future technologies. The model uses an appropriately defined energy function to describe the relative probability of organism settlement between a smooth surface and a topographically modified surface of the same chemistry. We have demonstrated the model's effectiveness for the green algae *Ulva linza*, gram negative bacteria *Cobetia marina*, gram positive bacteria *Staphylococcus aureus*, diatoms *Navicula permintua* and *Seminavis navicula*, and cyprids of *Balanus amphitrite*. All organisms fit the linear model with an R2 value of .93 or higher.

BI-4

TERNARY CROSSLINKED NETWORKS BASED ON HYPERBRANCHED FLUOROPOLYMER-POLYDIMETHYLSILOXANE-POLY(ETHYLENE GLYCOL) BLENDS TOWARDS NON-TOXIC, ANTI-FOULING COATINGS

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The development of environmentally-benign, anti-biofouling surfaces is desirable for the prevention of the accumulation of biofilm and marine organisms over current paint options that show environmental toxicity. One approach that has shown to be promising is the use of novel polymeric networks as anti-fouling coatings. Three classes of widely investigated polymers have been shown to inhibit fouling of specific marine organisms; that of fluoropolymers, siloxanes, and poly(ethylene glycol) (PEG) based materials. In this study, a two-dimensional array of amphiphilic polymeric networks was generated by the crosslinking of hyperbranched fluoropolymers (HBFPs), with varying concentrations of polydimethylsiloxane (PDMS) and PEG. The coatings in this matrix were obtained through a crosslinking reaction between the terminal amines of the PDMS and PEG with the alkyl bromides of the HBFP and subsequently deposited as coatings. This synthetic method allows kinetically driven crosslinking of all three materials simultaneously. Crosslinking is immediately followed by a degree of thermodynamically driven phase separation within the bound network and results in a complex surface that displays heterogeneity in its topography, topology and chemical composition on both the nano- and the micro-scale. The coatings were characterized extensively using atomic force microscopy (AFM), contact angle measurements, wide field fluorescence microscopy, and thermal measurements to probe the surface chemistry of the systems as well as their bulk properties. These findings suggest that our ternary films will contribute to a novel class of anti-fouling coatings that are environmentally benign and offer improved performance due to their blend of chemical composition, topography and topology on both the nano- and micro-scale.

BI-5

POSSIBLE MOLECULAR TARGETS OF AN ANTIFOULING COMPOUND (BUTENOLIDE) IN THE BARNACLE *BALANUS AMPHITRITE* AND THE BRYOZOANS *BUGULA NERITINA*

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Butenolide [5-octylfuran-2(5H)-one] is discovered and patented by my laboratory as an efficient and environmental friendly antifouling compound. According to OECD's guideline, the predicted no effect concentration (PNEC) of butenolide against both non-target and target species is 0.168 μg l^{-1}. To understand molecular mechanisms, we used butenolide to pull down its binding partners in cell lysate of the barnacle *B. Amphitrite* and the bryozoan *Bugula neritina*. We identified several potential molecular targets and important genes, including 1) the *B. Amphitrite* ACAT1 (also known as T2 thiolase) – a mitochondrial tetrameric enzyme involved in ketone body synthesis and degradation, 2) the *B. Neritina* ACADVL – the first enzyme of the very long chain fatty acid beta-oxidation pathway, 3) the *B. Neritina* actin, and 4) two *B. Neritina* GSTs (glutathione S-transferase) – a group of enzymes involved in phase II detoxification or the isolation, transport and synthesis of endogenous hydrophobic compounds. Since the ACAT1 and ACADVL are acyl-CoA binding enzymes involved in fatty acid metabolism while the actin and SCSβ are NTP-binding proteins and the two GSTs are glutathione binding proteins. No known proteins involved in cell cycles, cell proliferations, cell differentiations, cell death and neuronal transmissions were found to directly bind butenolide in these species, we conclude that butenolide inhibits larval settlement of these two organisms through alteration of larval energetic metabolisms. This is the first report on the protein targets of active antifouling compounds.

BI-6

LABORATORY AND FIELD TESTS OF AN ANALOGUE OF ANTIFOULING FROM MARINE SPONGE

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Several compounds of ***Glycerophospholipids with a*** chemistry structure analogue that produced by marine sponges from Arraial do Cabo, Brazil, were synthesized and tested in laboratory against bacteria and microalgae. The most active compounds were incorporated it in a paint matrix and tested in the field, using 5 a 10% and different concentration of main and auxiliary biocide from a commercial paint. The panels were placed at racks tests of International Paints at Guanabara Bay, a highly eutrophic Bay, and in oligotrophic field test area of Instituto de Estudos do Mar Almirante Paulo Moreira, in Arraial do Cabo. The results showed a significant reduction in the fouling growth compared with a panels control without antifoulouling paint. The performed of the natural biocide was better that obtained by the auxiliary biocide used in the commercial paint. Therefore, the results shows that our biocide synthesized base on marine sponge structure could be used as an auxiliary biocide in existing commercial paints.

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BI-7

POTENTIAL OF MICROALGAE EXTRACTS FOR ANTIFOULING APPLICATION: LABORATORY SCREENING AND FIELD-TESTING

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Biofouling has major economic and environmental impacts that justify the need to develop novel preventive treatments. Biocides and heavy metal-based antifouling paints are widely used, but their toxicity against non-target organisms is driving the search for more specific and environmentally friendly antifouling compounds.

There is a growing interest in screening marine organisms for the production of biogenic compounds that have potential for use in antifouling applications. Microalgae have attracted attention in this area as they have high growth rate and can be cultivated sustainably on a large scale under controlled conditions. Several strains of marine microalgae have been cultivated for this purpose and biogenic compounds were obtained by solvent extraction. Their antifouling activity (growth inhibition, adhesion/settlement inhibition) were tested through laboratory-based bioassays targeting several strains of bacteria, diatoms, macroalgae spores, and nvertebrates such as barnacles and mussels. Toxicity testing was also performed to confirm the eco-friendly properties of the natural products. Promising compounds were incorporated into paint formulation and several panels were tested by static immersion in 3 different locations: Portsmouth (UK), Toulon (France) and Martinique (French West Indies). The results of these studies will be presented and discussed.

BI-8

TRANSPORT OF SOFT PARTICLES VIA ACTUATED ARTIFICIAL CILIA

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Biological organisms often manipulate particles in their surroundings using hair-like appendages called cilia. In this work, using simulation and analysis, we show how an array of actuated, fluid-immersed, artificial cilia interacts with soft particles via adhesive forces. For each cilium in the array, one end is tethered to the wall, while the other end is actuated by an external periodic force. The simulations are performed using the Lattice Boltzmann Method for the flow, with a chain of point-forces, connected by springs, used to represent each cilium. Our results show that, for a given cilia stiffness, the trajectory of the particle strongly depends on both the particle-cilia adhesion and the particle stiffness. Specifically, the stiffer the particle, the higher the critical adhesion required for the cilia to start 'propelling' the particle forward. Analysis is presented to explain the scaling of the critical adhesion with respect to the particle stiffness. The results presented here indicate that adhesive artificial cilia can be used to reliably manipulate and sort particles according to their compliance. Furthermore, the findings reveal how to harness active, ciliated surfaces to prevent the adsorption of soft particles on substrates and thus, can be affective in antifouling applications.

Poster Session 1

MP-01

ASSESSMENT OF COPPER CONCENTRATION, TOXICITY AND BIOAVAILABILITY IN SHELTER ISLAND YACHT BASIN, SAN DIEGO, CALIFORNIA

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Metal bioavailability can be assessed by metal addition to water samples and observing the response by appropriately sensitive organisms relative to the observed response in a reference water sample. Recent studies have shown that the presence of natural organic matter (NOM) is one of the most important environmental factors that affect copper bioavailability at coastal marine sites. Since bioavailable metal is a better predictor of effects to aquatic organisms than dissolved or total metal, it is important to assess bioavailability before concluding that effects are expected, even at sites with a strong gradient in metal concentration. A copper gradient has been reported for Shelter Island Yacht Basin (SIYB), which is probably due to hydrological conditions and the relatively large number of vessels coated with copper antifouling paint. Evaluation of spatial and temporal changes in concentration, toxicity and bioavailability of copper in SIYB was accomplished by sampling surface and bottom waters in the wet and dry seasons, quantifying total and dissolved copper and copper toxicity to the mussel *Mytilus galloprovincialis.* Bioavailability was also assessed by predictions with the marine Biotic Ligand Model (BLM) for copper, which is currently under USEPA review. Dissolved copper concentration gradients can be described as three distinctive areas, one with relatively low concentrations (1-2 $\mu g\ L^{-1}$) by the mouth, an area with high concentrations (2-5 $\mu g\ L^{-1}$), and an area with a steep gradient to the highest copper concentrations (5-9 $\mu g\ L^{-1}$) in the back. In spite of the high copper concentrations, toxicity was only observed once in surface waters at one station. This finding is supported by measured EC50s in spiked copper samples using the WER methodology, which ranged from 8 to 11 $\mu g\ L^{-1}$, and were also consistent with BLM predictions. These results support including copper bioavailability for evaluation of environmental effects.

MP- 02

BIOSYNTHESIS, ASSEMBLY AND CHARACTERIZATION OF SILVER NANOPARTICLES TO CONTROL MARINE BIOFILM FORMATION

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Marine biofilms are the preliminary process due to bacteria that attach to surfaces interact or immersed in seawater and aggregate in a hydrated polymeric matrix. Adhesion of these sessile communities and their inherent resistance to chemical antifoulants are the sources of many relentless and never-ending maritime hitches. Such biofilms or microfouling play a major role in the succession of marine biofouling which attracts the settlement of the larvae of barnacles, mussels and other sessile invertebrates. Different approaches have been used for preventing micro and macrofouling on marine industrial settings. These methods have their own demerits that include chemical based complications such as dumping of toxic substances in the marine environment, emergent antibiotic resistant strains, etc. Silver nanoparticles are renowned for their influential anti-microbial activity.

On this back drop the present work focused on the silver nanoparticles, biosynthesized using the extract of marine sponge, and characterized by UV visible spectroscopy, High Resolution Transmission electron microscopy, X-ray diffraction pattern and by Fourier transforms infrared spectroscopy. The isolates of marine biofilm forming bacteria scraped from ship hull's were identified by 16S rDNA sequences and tested against silver nanoparticles resulted in more than 90% inhibition of biofilm formation. The antibacterial activity and the rate of inhibition vary to species to species of the microbes tested. Further the antifouling property of the biosynthesized silver nanoparticles were confirmed by preparation of silver nanoparticles assemblies on titanium and stainless steel surfaces that were exposed in seawater for microfouling formation and characterized by Scanning electron microscopy and compared with the images of LIVE/DEAD staining. Optimistic results of this study demonstrate the futuristic application of silver nanoparticles in addressing marine biofouling problem based on their potential anti-microfouling activity.

MP-03

ABNORMAL DUCKWEED FRONDS OCCURRED BY METAL PYRITHIONE

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Copper pyrithione ($CuPT_2$) and zinc pyrithione ($ZnPT_2$) are two popular antifouling agents that prevent biofouling. Research into the environmental effects of metal pyrithiones has mainly focused on aquatic animal species such as fish and crustaceans, and little attention has been paid to primary producers. There have been few reports on residues in environmental matrices because of the high photolabile characteristics of the agents. Residue analyses and ecological effects of the metabolites and metal pyrithiones are not yet fully understood. This study was undertaken to assess the effects of $CuPT_2$, $ZnPT_2$, and six metabolites (PT_2: 2,2′-dithio-bispyridine *N*-oxide, PS_2: 2,2′-dithio-bispyridine, PSA: pyridine-2-sulfonic acid, HPT: 2-mercaptopyridine *N*-oxide, HPS: 2-mercaptopyridine, and PO: pyridine *N*-oxide) on a freshwater macrophyte. A 7-day static bioassay using axenic duckweed *Lemna gibba* G3 was performed under laboratory conditions. Toxic effects of test compounds were assessed by biomass reduction and morphological changes were determined in image analysis. Concentrations of $ZnPT_2$ and $CuPT_2$ and those of PT_2 and HPT in the medium were determined by derivatizing 2,2′-dithio-bispyridine mono-*N*-oxide with pyridine disulfide/ethylene diamine tetra-acetic acid reagent that was equimolar with pyrithione. The toxic intensity of the compounds was calculated from the measured concentrations after 7-day exposure. $ZnPT_2$, $CuPT_2$, PT_2, and HPT inhibited the growth of *L. gibba* with EC_{50} ranging from 77 to 140 μg/l as calculated from the total frond number as the conventional index, whereas the other four metabolites had less effect even at 10 mg/L. The presence of the former four toxic derivatives resulted in abnormally shaped and unhealthily colored fronds, whose size was about 20% of the control fronds. EC_{50}, calculated from the healthy frond area determined in image analysis, ranged from 10 to 53 μg/l. Thus, image analysis as part of a duckweed bioassay can detect the toxic effects of pyrithione derivatives with 3–10 times higher sensitivity than the traditional index.

MP-04

BIOACCUMULATION AND EFFECT OF TRIBUTYLTIN ON THE SPERMATOGENESIS IN A MUD CRAB SCYLLA SERRATA

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Effect of tributyltin (TBT) on the bioaccumulation and spermatogenesis of a mud crab *Scylla serrata* was studied, at varying concentrations of TBT (10, 100 and 1000nl/l) for 50 days exposure period. All the concentrations of TBT resulted disrupted the reproductive activity as evidenced by Gonado Somatic Index (GSI). The control crab showed normal testis; whereas the treated crabs showed variation in the weight of the testis with respect to TBT concentrations. Bioaccumulation of TBT level varied in different tissues. Histological results clearly indicated that the control crab showed spermatophores and granular substances were seen filled up in the lumen of the seminiferous tubules. Besides, the treated crab testis illustrated that disruption in the seminiferous tubule membrane architecture, arrangement of spermatophores, granular substance, vacuole formation and reduction in the number of epithelial cells of the membrane were apparent in the seminiferous tubules. Biochemical constituents such as protein and glycogen contents also decreased in the treated crabs; whereas glucose level elevated in the treated groups, compared to the control. Thus, the study amply demonstrates that the TBT had significantly impaired the spermatogenesis in a mud crab *Scylla serrata*.

MP-05

REVISION AND UPDATE OF THE US AMBIENT WATER QUALITY CRITERIA FOR COPPER IN SALTWATER

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A large amount of new information related to the toxicity of copper to aquatic organisms in marine and estuarine waters has been published since the most recent update to the US EPA ambient water quality criteria[1]. Over 700 toxicity endpoints were obtained from a recent literature review conducted to update the marine document. From these data, toxicity information for 80 species in 67 genera were suitable for the update to the saltwater ambient water quality criteria for copper, compared to 33 species in 26 genera used in the previous document. In the updated database, the most sensitive organisms to copper toxicity were embryo-larval forms of marine invertebrates. There was also considerable evidence in recent literature showing the importance of bioavailability in determining copper toxicity. In particular, natural organic matter (NOM) was one of the most important factors that affect copper toxicity. Chemical speciation measurements have shown that NOM can bind dissolved copper. A consistent reduction in copper toxicity was demonstrated for a number of organisms in waters from several sites that was proportional to the amount of NOM present. Measurements of copper accumulation in sensitive embryo-larval forms of marine invertebrates are also reduced in the presence of NOM, suggesting that NOM-bound copper is not bioavailable. A saltwater Biotic Ligand Model (BLM) for copper was calibrated to predict the speciation of copper in marine and estuarine waters, and accumulation of copper in sensitive invertebrates. The saltwater BLM for copper was used in this update to incorporate bioavailability effects in the marine criteria, using a methodology that was similar to the most recent update to the freshwater ambient criteria for copper[2]. This update recommends that the final acute value is lowered below the 5th percentile of the BLM normalized species sensitivity distribution to be protective for *Mytilus edulis* (blue mussel).

[1] U.S. Environmental Protection Agency, Glen Thursby and David J. Hansen. 1995. Ambient Water Quality Criteria – Saltwater Copper Addendum (Draft). EPA 440-5-80-036.

[2] U.S. Environmental Protection Agency; Luis A. Cruz, Cindy Roberts, Mary Reiley, Robert Santore, Paul Paquin, Gary Chapman, Jennifer Mitchell, Charles Delos, Joseph Meyer, Rooni Mathew, and Tyler K. Linton. 2007. Aquatic Life Ambient Freshwater Quality Criteria - Copper. 2007 Revision. EPA-822-R-07-001.

MP-06

MATHEMATICAL PREDICTION AND SIMULATION OF BIOCIDE DIFFUSION IN ANTIFOULING SYSTEMS

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Release modelling of biocides has focussed principally on the widely used pigment cuprous oxide and its reaction products. The antifouling activity of cuprous oxide is governed principally by chemical reaction of large pigment particles within the binder, at the interface of the pigment front with the seawater. However, the processes influencing movement of dispersed organic or inorganic biocides within the binder are likely to be predominantly diffusion-driven rather than controlled by chemical speciation. Diffusion in polymers is a highly complex phenomenon, particularly for polar molecules or those whose molecular weight is comparable to, or larger than, the repeating monomer unit, as is the case for compounds typically employed as antifouling agents. Furthermore, the effects of water swelling on the kinetics of diffusion within a polymer binder are hitherto rarely considered in the literature.

A study has been carried out to determine the applicability of physicochemical models from existing literature to the case of biocide diffusion in a series of model acrylic binders. A transition-state theory model for small penetrants in polymer systems below glass transition temperature was modified to allow for geometric variation of interstitial cavities as a function of temperature and water uptake. The model allows estimation of diffusion coefficients for input into mathematical models of idealised paint film structures, which in turn allows a prediction of biocide residence time. Validation of results was achieved by long term natural seawater immersion of acrylic coatings containing the biocide, and optical/fluorescence microscopy determination of biocide concentration.

MP-07

GROWTH AND ADHESION STRENGTH OF DIATOM BIOFILMS CULTURED UNDER DYNAMIC CONDITIONS

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The fouling of ships' hulls by diatoms is a problem on many traditional antifouling paints as well as on silicone-based elastomeric coatings. Diatoms generally attach strongly to low energy surfaces, and silicones, along with many other candidate fouling-release coatings, tend to have hydrophobic characteristics.

Current laboratory methods to evaluate the likelihood of fouling by diatoms on non-biocidal surfaces have concentrated on studies using single cells with short contact times (several hours). This provides useful information on the likely colonisation of coatings by diatom slimes. However, the adhesion strength data, pertinent to fouling-release studies, may be misleading since biofilms are likely to adhere more strongly than single cells as the confluent growth of the biofilm embeds the cells in a continuous matrix of polysaccharide and glycoprotein exopolymers. A more complete picture of diatom fouling is therefore gained by examining the adhesion of biofilms as well as single cells.

The growth of diatom biofilms is likely to be influenced by the surrounding environmental parameters. A major factor is the movement of water around ship hulls, which is never stationary and is responsible for nutrient delivery, the recruitment and erosion of cells (and polymers) and hydrodynamic adaptations in terms of biofilm structure and polymer generation. To facilitate the culture of diatoms under flowing seawater in the laboratory, a Perspex channel has been designed. Diatom biofilms have been grown under different shear stresses to provide an insight into the susceptibility of surfaces to biofilm development. Adhesion strength measurements have indicated that culture under flowing seawater increases attachment strength. The data are contrasted with information from single cell assays.

MP-08

A FLUOROPOLYMER FOULING RELEASE COATING SIGNIFICANTLY RESISTS THE INITIAL DEVELOPMENT OF BIOFILMS

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Anti-microfouling performance of two fouling release coating systems, namely a silicon based technology (Intersleek 700 = IS700) and a fluoropolymer technology (Intersleek 900 =IS900) was investigated in a 10 day field experiment in tropical waters in Oman. A tie coat (TC) was used as a control surface. Microfouling accrual on 256 coated glass slides was analyzed using epifluorescence microscopy. Both IS700 and IS900, as well as the TC, developed biofilms made up of heterotrophic bacteria, cyanobacteria, seven species of diatoms and algal spores. However, IS900 had significantly thinner biofilms, lower diatom species richness, no algal spores and the least number of bacteria. The differences in surface roughness, modulus, wettability and energy between the fluoropolymer and silicone technologies most likely account for this result. The greater efficacy of IS900 against both microorganisms and algal spores is likely to lead to lower fuel consumption for those vessels with an operational profile capable of utilizing fouling release coatings.

MP-09

BIOFILM DEVELOPMENT ON HIGH-PERFORMANCE MOLDED MATERIALS IN A TEMPERATE NORTHERN ESTUARY

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The composition of biofilms and the environmental factors controlling them are key elements in understanding and predicting surface responses to exposure in seawater, and resulting recruitment of larvae and spores. We are characterizing the development of marine biofilms during summer and winter on thermoplastic materials used in underwater connectors and cable assemblies. One objective is to compare patterns of microbial community composition between winter and summer biofilms. Metal slides coated with polyetheretherketone (PEEK), or polyetherketoneketone (PEKK) are immersed in flow-through running seawater directly pumped from Narragansett Bay, RI; slides of the metal alloy Monel are used as Controls. Replicate samples are retrieved after seven, 15 and 30 days for chlorophyll-a measurement, carbon and nitrogen analysis, and DNA extraction. Remaining slides are fixed for observations by confocal scanning, scanning electron, and atomic force microscopy to help identify the community and investigate structural characteristics of the biofilm and of the surfaces. Not surprisingly, for the summer immersion (June-July 2011), organic biomass, as measured by carbon and chlorophyll concentrations per unit area, increased with immersion time. A great deal of variability was observed between replicate slides of a given type. By day 30, the chlorophyll biomass on PEEK was not significantly different from the biomass on PEKK, but both were significantly higher than the biomass measured on the Control slides, 2.3 vs. 0.9 µg chlorophyll-a cm-2, respectively. However on a carbon basis, no significant differences were noted: an average biomass of 100 µg cm-2 was measured by day 30. A carpet of bacteria, pennate diatoms, seaweed germlings, and stalked ciliates comprised this early microbial community, and will further be characterized by a DNA fingerprinting technique, the automated ribosomal intergenic spacer analysis (ARISA). Temporal patterns of microbial community composition in biofilms will be contrasted with patterns obtained from an on-going winter immersion (January-February 2012).

MP-10

MICROBIAL COMMUNITIES ON ANTIFOULING MARINE PAINTS

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Although antifouling marine paints have been used to prevent biofouling, not much is known about their effectiveness in preventing attachment of microorganisms. This research project aims at qualitative and quantitative comparison of microbial communities developed on four different types of antifouling coatings in Marina Bandar Rowdha and Marina Shangri La, Oman. Coatings tested included zinc-based (Pettit #1863 and #1792, West Marine #11046620) copper-based (West Marine #5566252 and #10175206, Hempel's Hard Racing #76484 and Olympic #86950), copper and zinc-based (International Micron Extra) and silicone-based fouling release coating (Hempel's Hempasil X3). All coatings were applied on clean plastic slides. Slides without any coating were used as a control. Microbial biofilms were harvested after 2, 7 and 14 days of immersion. Bacterial densities were analyzed using epifluorescence microscopy and biomass of photosynthetic organisms was estimated using chlorophyll *a* concentrations. Antifouling performance of coatings in both marinas was different. At Shangri La zinc-based coatings showed the lowest bacterial density after 2 days in contrast to the control which had ten fold greater bacterial densities. Fouling by phototrophic microbes was prevented until 7 days. Copper-based coatings inhibited bacterial adhesion only after 7 days, while the silicone-based fouling release coating prevented microfouling after 14 days. However at Bandar Rowdha, copper-based coatings effectively reduced bacterial fouling throughout the experiment when compared with the control which showed a gradual increase in bacterial count. Silicone-based fouling release coating inhibited attachment of photosynthetic microorganisms until 14 days. The differential performance of tested antifouling coatings may be attributed to several factors including varying environmental conditions, difference in biofouling communities, time of exposure and physical and chemical properties of antifouling coatings. The temporal and spatial changes of microbial communities on antifouling paints are currently being assessed using molecular techniques such as 454 pyrosequencing and automated ribosomal intergenic spacer analysis.

MP-11

APPLICATION OF A NEW BIOFILM SENSOR FOR ANTIFOULING TREATMENTS OPTIMIZATION

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Even since its first phase, biofilm development on artificial substrates (fluid flow systems, water distribution devices, marine sensors, medical equipments, etc.) causes several technological problems (corrosion, equipment failure, energy loss, reduced performance, resistance to antimicrobial treatments), increasing management and maintenance costs. To avoid these problems, in water flow systems, different kinds of treatments exist, but the choice of active substances to be used, application time and frequency is usually based on an empirical approach. The possibility of monitoring bacterial growth, from its first phases, allows optimizing the treatment process achieving the best results in terms of effectiveness and costs reduction. In this contest we have developed a new sensor, which exploits the natural marine and freshwater biofilms electrochemical activity, proportional to surface covering. Applying this technique, it is possible to accurately keep tabs on bacteria settlement since surface first colonization, giving an early warning very useful for those applications which could be affected even by low biofilm covering (e.g. membrane ultrafiltration, ultrapure water systems, etc.), and allowing to treat the affected plant pipelines/sections right when the biological layer is easier to be eliminated. The existing biofilm sensors, on the contrary, can point out only thicker biofilms (microns), providing a late indication. Furthermore, the device presented here includes advanced technological features, such as distributed approach, real-time monitoring, data accessibility from remote, high sensitivity, precision and flexibility. During laboratory characterization and field testing in industrial environment, the sensor response to biofilm growth has proven to be quick and accurate, therefore this instrument has been employed to optimize cleaning treatments, and check their effectiveness, inside some industrial plants. The sensor adoption allowed to drastically reduce the amount of chemical substances used, without any loss in the plant efficiency, giving at the same time a real-time feedback on cleaning treatments effectiveness.

MP-12

IN SITU ASSESSMENT OF BIOFILM GROWTH AND STRUCTURE ON NATURAL PRODUCT CONTAINING ANTIFOULING COATINGS

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Biofouling is a surface related phenomenon and as such it is essential to investigate the relationship between fouling organisms and the water-substrate interface. Specifically, it has been shown that biofilms are not homogenous planar layers, but complex aggregations of cell clusters with channels where convective flow can facilitate the supply of oxygen and nutrients (Stoodley et al. 1994). This has significant importance for biofilms growing on antifouling (AF) coatings, since these channels will transport nutrients from the surrounding water into the biofilm, but also assist in the transport of biocides from the AF coated surface through the biofilm to the surrounding water.

This study has incorporated natural products (NPs) into a model coating system, using two formulations in order to assess the effect of NP-containing coatings on biofilm growth. Laboratory screening of NP-containing coatings is often largely unexplored mainly due to difficulties in assessing their activity over short experimental time scales (typically only a maximum of a few days). To date there are only a limited number of reports on laboratory assessment for antifouling paints and their effect on biofilm growth and/or attachment. In this study, NP-containing paints were applied on to glass coupons, placed in 24-well plates and then inoculated with the marine biofilm forming bacterium *Cobetia marina* for 48 h. This has been achieved by the development of a novel bioassay protocol that has allowed the *in situ* observation of biofilm formation and growth, by corroborating different techniques such as a multidetection microplate reader and confocal laser scanning microscopy (through nucleic acid staining). There was good correlation between the two techniques which showed that the NP containing coating significantly inhibited biofilm growth and also revealed marked differences in biofilm structure (e.g. bio-volume, morphology and thickness). The goal of this study was to develop a new protocol to allow assessment of biofilm formation on coatings in a high throughput non-invasive manner.

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MP-13

ADHESION OF MARINE BACTERIUM BACILLUS SUBTILIS TO HARD SURFACE: ROLE OF NUTRIENTS AND ENVIRONMENTAL FACTORS

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Adhesion of bacteria on hard surfaces is a common process in aquatic environments and has great ecological and economical significance. The bacterial adhesion on surfaces is influenced by a variety of physical, chemical and biological properties. In the present study, the influence of salinity, pH, calcium, magnesium, carbohydrate, protein, extracellular polymeric substance (EPS) and conditioning film on the adhesion of marine bacteria *Bacillus subtilis* was studied in laboratory condition. Result showed that glucose, bovine serum albumin and calcium reduces the bacterial adhesion while magnesium enhances the settlement. The bacterial adhesion was significantly high on coupons submerged in alkaline pH and 30 psu salinity mediums. Conditioning film induces the biofilm formation and the EPS secreted by the bacteria did not induce the adhesion. Statistically significant difference on the adhesion was observed on coupons submerged in different pH medium. In general, the present study showed that bacterial adhesion on surfaces is controlled by combination of factors in the surrounding environment and also on the surface.

MP-14

ELECTROCHEMICAL SENSING AND CHARACTERIZATION OF AEROBIC MARINE BACTERIAL BIOFILMS ON GOLD ELECTRODE SURFACES

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Marine bacteria are ubiquitous and they play a pivotal role in the initial biofilm development and the maintenance of a biofouling community[1]. The engineering issues of this troublesome phenomenon in naval applications and in merchant shipping are numerous and can cause excessive capital costs, *e.g.* excess of £100k ($155k) each to replace header castings[2]. To combine reasonable expenses and higher operational availability, ship owners are concerned by increasing maintenance schedule of marine platforms. Fouling can affect the hydrodynamic properties and reduce heat transfer performance of operating marine heat exchangers leading to frequent failures and blockages of fluid-handling components. In addition, marine organisms can interact with metallic surfaces and be involved in corrosion pathways through biocorrosion mechanisms[2,3]. Although the problems of biofouling in marine heat exchangers are abundant, suitable techniques capable of sensing for the presence and extent of biofilms on metallic surfaces are still required. Reliable and accurate *in situ* sensors capable of detecting and quantifying biofilmed metallic surfaces are becoming increasingly necessary. An electrochemical sensor using a 0.2 mm diameter gold electrode has been characterized in abiotic and biotic artificial sea water media within a modified continuous culture flow cell. The significance of extenal polarization on a biofilmed gold surface has been explored using cyclic voltammetry. Overall, the cyclic voltammograms for abiotic and biotic artifical sea water demonstrated that bacterial biofilms can change the electrochemical properties at the gold electrode interface. The objectives of this study were:

to establish a framework for electrochemical sensing within abiotic and biotic artificial sea water test media,

to electrochemically detect marine bacterial biofilms and address the relevance of using electrical imposed polarization to monitor naturally occurring bacterial biofilms.

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MP-15

SURFACE WETTABILITY AND TEXTURE DO NOT AFFECT THE SETTLEMENT OF THE HYDROID ECTOPLEURA LARYNX

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The hydroid *Ectopleura larynx* (syn. *Tubularia larynx*) is one of the main fouling organisms on salmon aquaculture cages in South to Mid Norway, and its excessive growth can negatively affect the farming process. The aim of this study was to find novel surface materials and textures that deter the settlement of hydroid larvae. The settlement preferences of *E. larynx* larvae for 11 materials of different wettabilities and a control surface were tested in a no-choice bioassay. The experiment was performed three times. Settlement was highly variable, ranging between 95 % and 33 %, and preferences were inconsistent across trials. For example, polytetraflouroethylene (PTFE) had the highest settlement in trial 1 (95 %) and was among the least preferred materials in trial 3 (37 %). Overall, *E. larynx* had no preference for any of the tested materials. Furthermore, there was no effect of texture on the settlement of *E. larynx* in both laboratory and field experiments. In the laboratory assay, 9 high-density polyethylene (HDPE) and 7 polydimethylsiloxane (PDMS) surfaces with textures ranging from 100 to 600 µm and 40 to 400 µm spacing, respectively, were tested with a smooth control. Larval settlement did not differ significantly between any surface. These results were confirmed in the 12-day field test with PDMS surfaces, where there was no preference for any of the examined textures. In conclusion, neither the tested materials nor textures are effective at deterring the settlement of the hydroid *E. larynx*.

MP-16

ENHANCING THE SETTLEMENT AND ATTACHMENT OF PEDIVELIGERS OF MYTILUS GALLOPROVINCIALIS USING SURFACE WETTABILITY AND MICROTOPOGRAPHY

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The surface properties of wettability and micro-topography can either enhance or deter larval settlement of many sessile marine organisms. This study quantifies the effect of these surface properties on the settlement of pediveligers of *Mytilus galloprovincialis*, using polymers spanning a wettability range and micro-textured polydimethylsiloxane (PDMS). Furthermore, the adhesion strength of settled pediveligers on micro-textured PDMS surfaces was quantified using a flow chamber. Settlement was enhanced at the hydrophilic end of the wettability spectrum, where mean settlement on nylon reached 35.2 ± 4.3%. In contrast, mean settlement on the most hydrophobic polymer (PDMS) was 4.4 ± 1.4%. Micro-topography had a much stronger effect compared to wettability, where 400 µm textured PDMS enhanced settlement above 90%. Settlement preferences were also positively correlated to adhesion strength at flow rates of 4 knots, with all initially settled pediveligers on smooth PDMS detaching, while 79.9 ± 5.7 % of pediveligers remained on the 400 µm texture.

MP-17

WHEN NAILS ARE MORE COMFORTABLE: THE SETTLEMENT-INHIBITING CHARACTERISTIC OF AIR POCKETS ON SUPERHYDROPHOBIC SURFACES

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The attachment-inhibition and foul-release behavior of super-hydrophobic surfaces were examined with the Amphora sp. Diatom. Various controllable parameters of the super-hydrophobic surface were isolated and varied in order to test the effect of nanoscale roughness, microscale roughness and surface chemistry on the settlement behavior of diatoms.

Super-hydrophobic surfaces were engineered with dual scale roughness features using two building blocks; silica nanoparticles ranging from 7-40 nm and PMMA templating latex ranging from 400-800 nm. The combination of these building blocks affords various roughness changes at the nanoscale.

Results suggest that there exists no clear relationship between the scale of nanoroughness and microroughness with attachment-inhibition and foul-release behavior of super-hydrophobic coatings. However, upon removal of the air layer present in immersed super-hydrophobic surfaces using ethanol-water exchange, a significant increase in attachements of diatoms was observed. This suggests that the presence of an entrapped air layer acts as a physical barrier against diatom attachment of super-hydrophobic surfaces.

MP-18

BIOFOULING IN NATURAL SEAWATER OF THE ROUGHENED SURFACE OF PTFE

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The goal of this project was to further evaluate, in a natural biofouling seawater environment, the observation reported from the laboratory of Professor Jonathan Rothstein (U. Massachusetts – Amherst), that simply roughening the surface of solid polytetrafluoroethylene (PTFE, e.g., Teflon®) results in a super-hydrophobic surface. [1,2]

A panel of PTFE was roughened on one side using an abrasive blasting gun, while the opposite surface was left in its original state. Contact angle measurements using purified water were carried out on both sides of the PTFE, and the results confirmed greater hydrophobicity of the grit-blasted surface.

The panel was immersed in Biscayne Bay, and it was found that the roughened surface initially resisted biofouling to a greater degree than the untreated (smooth) side. However, eventually the roughened surface became fouled, and the organisms adhered to the roughened side were not dislodged as readily in a flow channel as the untreated side.

The behavior of such super-hydrophobic surfaces will be discussed, including a comparison of their performance under conditions of intermittent wetting (e.g., terrestrial plants) vs. continuous immersion.

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MP-19

WHALE BALEEN NANOSTRUCTURES AS A POSSIBLE MODEL FOR ANTIFOULING CONTROL

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Nature has provided us with examples of non-toxic, environmentally friendly antifouling surfaces as well as ideas towards achieving this goal.[1] Recent research has indicated that topography of the surface is an important consideration for the creation of such a surface.[2-5] Our Centre for Biofouling Research has been investigating the fouling characteristics of mussel, *Mytilis edulis* and scallop, *Placopectin magellanicus* shells, but are currently focusing on baleen from the fin whale, *Balaenoptera physalus*. Baleen is composed mainly of keratin and serves to filter krill, plankton and small fish from sea water. As such, it is likely to have antifouling or anti-adhesive properties to ensure a consistent passage of flow without clogging. We tested whether the physical structure at the surface of *B. physalus* was responsible for the amount of fouling observed compared to fouling on glass slide controls. Biofilm development studies were carried out by exposing baleen pieces to a mixed community of foulers in seawater tanks in the StFXU Animal Care Facility, and to cultures of marine bacteria, *Pseuodomonas sp*. NCIMB 2021, in the laboratory. In addition, the cut edges of the baleen pieces fouled more relative to its natural surface. The percentage fouling of baleen from gray whale, *Eschrichtius robustus* and blue whale, *Balaenoptera musculus* were also determined. Similar fouling studies using replicas (both positive and negative impression molds) of the surfaces were carried out as described for the baleen pieces. Images from an atomic force microscope of the surface of *B. physalus* revealed a nanotextured surface topography with some variation in the depths (60-100 nm) and widths (0.4 and 0.7 µm) of the ridges. Differences among whale species were noted.

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MP-20

DESIGN OF OPTIMAL SURFACE TOPOGRAPHY FOR ANTI-FOULING SURFACES BY COMPUTER SIMULATION

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Biofouling is a major problem for many industries ranging from shipping to medical implants. Recent work in several research groups had pointed towards the importance of surface topography in limiting the adhesion of biofouling agents. Here we aim to use Monte Carlo computer simulations to show the importance of periodic topographical features as a means to mitigate the adhesion of foulers.

We have developed a basic Monte Carlo model to simulate the adhesion of spherical particles with sticky polymeric hairs on a variety of surfaces. Our approach allows for the input of any surface configuration or particle shape. In this work we explore adhesion of such particles with varying diameter and the number and length of hairs on a range of sinusoidal periodic surface structures. This approach allows us to establish the optimal surface parameters minimizing the adhesion of particles with varying properties. Specifically, we will report that surfaces with very large or very low periodicity are nearly indistinguishable from flat supports (in some cases they even perform worse). Optimal surfaces are those whose periodicities are comparable to the "hydrodynamic size" of the particle (i.e., particle and hair). Additionally the role of amplitude has shown to be less significant than wavelength as long as the amplitude is larger than the size of the particle. We have also developed an order parameter that characterizes how well the particles organize over the periodic structures on the surface. Finally a new algorithm has been generated that allows for a computer driven "search" for optimal surfaces instead of inputting a predefined set of surfaces for testing.

MP-21

TUNING SENSITIVITY TO THIN AIR: THE EFFECT NANOBUBBLES ON ATTACHMENT BEHAVIOUR OF MARINE ORGANISMS

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The effect of subtle changes in surface nano- and micro- roughness of nanoengineered superhydrophobic surfaces on the attachment behavior of several fouling species (*Amphora sp., Ulva rigida, Bugula neritina*) was investigated. Two types of superhydrophobic surfaces were fabricated from latex-templated silica sol-gels. Type A superhydrophobic surfaces possess engineered single-scale roughness features whereas Type B superhydrophobic surfaces possess engineered dual-scale roughness features. In attachment assays, motile spores (Ulva rigida, Bugula neritina were found to be sensitive to the variations in surface roughness, whereas diatoms (Amphora sp.) exhibited equivalent settlement rates across all tested superhydrophobic surfaces. A subtle change in surface roughness of less than 200 nm resulted in significant decrease of Ulva spore settlement from approximately 350 spores/mm^2 to 30 spores/mm^2. Whereas the settlement of bryozoan (Bugola neritina) was dettered on all type B superhydrophobic surfaces, but favoured type A superhydrophobic surfaces. Correlating this data with the measured size and distribution of air pockets on immersed type A and B surfaces, results suggest that motile spores of Ulva rigida and Bugula neritina are able to sense the minute changes in air-pockets induced by variations in surface nanoroughness.

MP-22

ENGINEERING SURFACES FOR MARINE SERVICE BY COLD SPRAY

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Cold Spray is an emerging materials processing tool which involves the supersonic deposition of micro-particles onto surfaces. It has found broad application in industry, including as a coating method for protection against corrosion. This presentation highlights two alternative uses of cold spray which have been developed specifically for fouling prevention. Both have the advantage of ready adaptation to large components and infrastructure.

Rigid, sub-millimeter "pillars" have been built up by cold spray on metal surfaces, including titanium, copper, aluminium alloys and stainless steel. Pillar shape, height and spacing may be controlled by masking. Textured titanium coatings with 0.25 mm peak spacings were shown in lab assays to prevent settlement by the barnacle cyprid, *Amphibalanus reticulatus*. The results were in accordance with attachment point theory[1], which states that topographical features less than the length scale of the fouling organism have the effect of reducing adhesion strength.

Cold spray has also been used to embed particles of an active material into soft polymeric substrates. Examples of active particulate materials include copper and its oxides, which are capable of sustained release of ions for chemical fouling control. Due to deformation of the polymer around the impacting particles, they remain physically trapped in the surface. However, generally a portion of the particles remains exposed to the environment, allowing reaction with seawater to occur. The embedment approach avoids the limitations imposed by coating, namely mismatch in coating/substrate elastic properties and inadequate adhesion, which is a common problem with conventional antifouling paints. Depth of embedment and total particle loading depend on polymer type, relative exposure to the cold spray jet, and cold spray gas parameters. Polymer mechanical properties have been shown to be not appreciably altered by cold spray.

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MP-23

COLD SPRAY ANTIFOULING PROTECTION FOR POLYURETHANE MARINE SEISMIC STREAMERS

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Marine seismic surveys are the primary tool used by the oil and gas industry to explore, develop and manage undersea oil and gas deposits. Seismic streamers contain arrays of seismic sensors that are themselves towed in arrays measuring up to 90 km combined length allowing 3D imaging of the substrata. Fouling by pelagic goose barnacles is a severe and costly disruption, reducing data quality and increasing fuel consumption, drag and the likelihood of streamer breakage. The streamer jacket is made of flexible and low surface energy polyurethane, preventing effective coating with antifoulant.

Cold spray is a metal coating process that forms a supersonic spray of metal particles. Using cold spray equipment, we deposited a discontinuous layer of copper particles in polyurethane streamer jackets. The polymer retained its flexibility and did not become electrically conductive, demonstrating the discontinuous sub-surface nature of the treatment. Samples with 22 and 101 g/m^2 copper loading were mounted on frames and suspended 200 mm below the water's surface and 200 mm from the harbour wall at Townsville Yacht Club in tropical northern Australia and monitored weekly.

After 250 days monitoring, no macrofouler attachment had occurred on the high copper loading polyurethane, compared to 57% macrofouler coverage for low copper loading samples and 96% coverage for controls as determined by image analysis. The fouling organisms were barnacles, bryozoans, mussels and polychaetes. Soft fouling is not problematic on seismic streamers, however it is noted that fouling by *Ulva sp.*, a copper-tolerant algae, occurred on all surfaces, with total (*Ulva sp*. plus macrofoulers) coverage ranging from 40% for high copper loading back facing samples to 96% for back facing control samples. These results predict cold spray antifouling will be an effective technique for preventing biofouling of marine seismic streamers.

MP-24

POLY (TRIALKYLSILYL METHACRYLATE)S: A FAMILY OF HYDROLYSABLE POLYMERS FOR MARINE ANTIFOULING COATINGS

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Any object immersed in seawater will rapidly attract the attention of marine fouling, causing severe problems on dynamic or static structures. Coatings containing self-polishing or controlled erodible copolymers are used on surfaces likely to come into contact with marine fouling organisms. Such coating compositions comprise generally biocides or active compounds which could inhibit the settlement of marine organisms. The antifouling efficiency of these chemically-active coatings depends on the diffusivity of water inside the polymer matrix, the dissolution of the hydrolysed polymer chains and the diffusion of the dissolved active compounds towards the coating surface.

This study is part of the ECOPAINT PACA project and is focused on the use of trialkylsilyl-based (meth)acrylate polymers as potential tin-free substitutes and controlled erodible binders for marine antifouling coatings [1, 2]. Diblock (meth)acrylic copolymers bearing tert-butyldimethylsilyl-, tributylsilyl- and triisopropylsilyl-ester groups were synthesised by the Reversible Addition Fragmentation chain Transfer (RAFT) process. The chemical structure of comonomers aimed at adjusting desired dissolution rate profiles and also served to improve other coating properties, such as mechanical properties and degree of film formation. Antifouling paints derived from the silylated diblock copolymers were prepared and their antifouling efficiency was assessed through *in-situ* immersion tests. Samples were withdrawn every six months and the thickness of the biocides depleted layer was evaluated using scanning electron microscopy (SEM) and energy-dispersive X-ray analysis (EDX). The molar proportion of the silylated monomer units, the chemical nature of comonomers, and the molecular weight of the polymer chains were varied to control the thickness of the depleted layer with immersion time. Relationships between the chemical structure of the diblock copolymers and the *in situ* antifouling efficiency of the corresponding coating were demonstrated. Dynamic immersion tests were also performed. Data were compared with a commercially available self-polishing marine coating to classify the antifouling coatings developed in this study as self-polishing coatings.

MP-25

BINDER RESEARCH FOR ANTIFOULING MARINE COATINGS

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The Laboratory MAPIEM has worked in the research field of antifouling paints since the early eighties. The multidisciplinary knowledge of the research team (organic synthesis, polymer chemistry, biology, and biochemistry), and the natural sea water facilities (Mediterranean Sea) significantly strengthen the outcomes of antifouling projects. Our main antifouling activities are divided into five parts:

- the development of new acrylic binders for self-polishing or erodible marine paints,
- the extraction of natural biocides from marine organisms,
- the chemical and physicochemical characterisations of binders and natural biocides,
- the formulation of new tin-free antifouling marine paints,
- and the assessment of the efficiency of coatings combining erosion test, raft immersion exposure and bioassays.

Since triorganotin self-polishing paints were declared as toxicants towards non-target marine organisms, different tin-free self-polishing coatings have been developed in our lab. Our approach consists in synthesizing acrylic polymers containing a hydrolysable bond as the well-known TBT-copolymers in order to obtain paints with long time efficiency. New acrylic titanium- and silylated- based polymers were synthesised and characterized. Binders containing hydrolysable ionic bonds (ammonium salts) were also studied. Aqueous based binders with core-shell morphologies were developed for waterborne antifouling paints. Actually, copolymers containing silylated and poly(dimethylsiloxane) blocks are investigated combining erosion and low surface energy properties. A new strategy based on electroactive copolymers is explored.

An erosion test on rotor allows us to control the thickness decrease versus time in artificial/natural sea water and therefore to define the type of binders. Physicochemical characterisations which are useful to understand the mechanisms occurring in hydrolysable binders are available including Fourier Transform Infrared Spectrometer equipped with a microscope, gas/liquid chromatography, contact angle meter, NMR.

MP-26

LABORATORY AND FIELD PERFORMANCE OF PDMS-PEO BLOCK COPOLYMER COATINGS OF VARYING WETTABILITY

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Algae are well-recognised for their impact on the fouling of immersed surfaces. However, studies on the influence of surface physico-chemical properties on algal adhesion has focused mainly on the green macroalga *Ulva* and some diatom species. In this presentation we will explore the relationship between surface wettability and the adhesion preferences of a new test species, *Ectocarpus crouaniorum*, which is a filamentous brown alga and a significant member of fouling communities. The characteristics of a novel laboratory-based adhesion bioassay for *Ectocarpus crouaniorum*, at an appropriate scale for the screening of sets of experimental samples in well-replicated and controlled experiments will first be described.

A range of siloxane-based elastomeric coatings with systematic variations in surface wettability and polarity (hydrophilic/hydrophobic balance), but similar elastic moduli, was prepared using coatings based on PDMS-PEO block copolymers. The wettability and surface energies of the coatings were characterised by contact angle analysis, in air and underwater after various periods of immersion. These fouling-release coatings were then used in laboratory adhesion bioassays with *E. crouaniorum*, and for comparative purposes, with *Ulva*. The results of these assays showed that adhesion of neither species was directly or simply correlated with surface wettability and their adhesion preferences were different.

The same range of coatings was subjected to a fully replicated static field immersion test for 157 days. Results showed that colonisation by ectocarpoid algae and other fouling organisms, was strongly influenced by the wettability properties of the coatings. One of the most effective coatings in the laboratory assay with *E. crouaniorum* was also the most effective in the field tests.

MP-27

SOLVENT-FREE ALKOXYSILANE-BASED HYDROLYZABLE POLYMER SYNTHESIS AND DEVELOPMENT

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Recent advances in the development of hydrolyzable diisocyanate-cured alkoxysilane polymers will be discussed, along with the parallel development of laboratory and field analytical methodologies. Efforts have been directed toward developing the best means to provide rapid turn-around in laboratory analyses to enable improved chemical formulations, as well as ascertain the candidate prototype polymers that offer the most promise for yielding promising results in longer term field (natural seawater) evaluations.

Silicon alkoxide-based polymer chemistry poses particular challenges in achieving steady hydrolysis rates that do not compromise surface smoothness as that surface undergoes the aqueous ablation process. Improvements in both hydrolysis rate and restriction of the hydrolysis to the surface layer have been achieved through variations in the selection of (poly)diisocyanate, selection of curing catalyst, and nature and quantity of hydrolysis modification additives for control of reaction kinetics, the latter being typically done to retard hydrolysis.

Laboratory analytical methods are also under development for determining hydrolysis rates, primarily through quantification of species that are released into the immersion aqueous media during hydrolysis testing. Variations in polymer hydrolysis behavior as a function of immersion water (inorganic) composition have also been encountered (quite unexpectedly), and discussion will include the possible mechanisms by which these variations take place.

MP-28

CONTROLLED POLYMERIZATION BY THE RAFT PROCESS OF TRI-ALKYLSILYL METHACRYLATES AND STUDY OF THEIR HYDROLYSIS KINETICS IN ALKALINE MEDIA.

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Any surface immersed in seawater is rapidly colonized by marine fouling organisms. This colonization has serious impacts on the ship navigation (i.e., increased hull roughness, lost of maneuverability, increased fuel consumption). The most commonly used antifouling paints are the Self-Polishing Copolymer coatings (SPC). These coatings contain a polymer matrix which can be degraded in the marine environment, resulting in the erosion of the coating surface and the controlled release of the biocides incorporated within these coatings.[1]

Polymers based on tri-alkylsilyl methacrylate can be used as polymer binders for the formulation of SPC coatings. Actually, the tri-alkylsilyl methacrylate monomers can be hydrolyzed in alkaline and acidic media, as well as in seawater. This hydrolysis influences the hydrophilic character of the synthesized polymer and its erosion upon time.[2,3]

Polymers based on tri-isopropylsilyl and tri-*n*-butylsilyl methacrylate were synthesized by radical polymerization controlled by the RAFT process (Reversible Addition Fragmentation chain Transfer). A kinetic study by ^{1}H-NMR and TD-SEC demonstrated the control of the polymerization (linear evolution of the molecular weight versus monomer conversion).

The hydrolysis or saponification reaction in alkaline media of the ester bond of several poly(tri-alkylsilyl methacrylate)s was followed by *in situ* ^{1}H-NMR investigations. The influence of the structure of the tri-alkylsilyl moiety on the hydrolysis kinetics was evidenced.

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MP-29

DEVELOPMENT OF FRC/SPC HYBRID ANTIFOULING COATINGS

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Two main types of antifouling coatings are available on the market in order to limit the colonization of ship hulls by fouling organisms:

- The Self Polishing Copolymer coatings (SPC)[1]: their high antifouling efficiency relies on the release of biocides into the marine environment resulting from the controlled erosion of their polymer matrix.
- The Fouling Release Coatings (FRC)[2]: these coatings are based on polysiloxanes or fluorinated polymers and they do not contain biocides. The adhesion strength of fouling organisms is limited on such hydrophobic and soft coatings so as even if they may be fouled during idle periods, fouling organisms are easily removed from the hull at ship speeds higher than 10 knots.

In this study, environmentally friendly FRC/SPC hybrid antifouling coatings have been developed combining the hydrophobic properties of polysiloxanes and a controlled erosion of the coating. The aim of this work is to increase the antifouling efficiency of FRCs during idle periods and at low speeds, and to limit the impact of biocides on the marine environment.

The polymer matrixes of such coatings have been synthesized by radical polymerization of a FRC/SPC hybrid monomer. The hydrophobic and erosion properties of both the hybrid polymer matrix and the hybrid coatings have been studied in artificial seawater. The hybrid coatings have also been immersed in the Mediterranean Sea to evaluate their antifouling performance.

MP-30

SYNTHESIS AND PROPERTIES OF WATERBORNE POLYURETHANE-IMIDAZOLE ANTIFOULING COATINGS

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Marine biological fouling which is undesirable accumulation of microorganisms, plants and animals on artificial surfaces that are exposed to the marine environment is clearly observed on every suitable substrate that is immersed in seawater, such as ship hulls, marine cages, pipelines, heat exchangers and other structures including offshore platforms and bridges. Initial combat to fouling was successfully done by using TBT based coatings which has been banned from 2008 all over the world. Thus, the paint industry has forced on developing tin-free products that are able to replace the TBT-based ones. Another environmental driver for changing the paint industry is the reduction of volatile organic compounds (VOC). Therefore, solvent-based paints have also been targeted, resulting in enormous effort to find water-based alternatives or less toxic solvents. Waterborne polyurethane (WBPU) has shown promising result to protect the surface from fouler when the resin contained some silane compound. In this report we used synthesized poly(quaternized vinyl imidazole-*r*-vinylamine) (pQVIm-*r*-VA) in WBPU. Synthesized pQVIm-*r*-VA act as both hydrophilic agent (quaternized vinyl imidazole) and chain extender (amine react with NCO terminated prepolymer). The coating showed significant antifouling properties when the pQVIm-*r*-VA was present in WBPU.

MP-31

EFFECT OF SHEAR FORCES AND SURFACE HETEROGENIETY ON MARINE BACTERIAL RELEASE FROM PNIPAAM GRAFTED BRUSHES

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Surface active polymers represent an efficient and non-toxic approach to achieve release of biofouling. Among these, poly (N-isopropylacrylamide) (PNIPAAm) is one of the most studied synthetic responsive polymers that exhibits change in its relative hydrophobicity at its lower critical solution temperature (~32 °C). Above and below this transition temperature, PNIPAAm grafted brushes undergo collapsed and extended chain confirmation that has been shown to reversibly modulate the attachment and release of various bacteria, for example, *Cobetia marina* and *Staphylococcus epidermidis*. In our recent studies, we used interferometric lithography (IL) to pattern PNIPAAm brushes synthesized by surface initiated ARGET-ATRP as a fast and convenient method to graft nano-patterned polymer brushes over large areas under ambient conditions. IL is also being used to produce fouling resistant nanopatterned self-assembled monolayers (SAMs) as model surfaces for the study of marine bacterial attachment and release. Using a custom-built spinning disk apparatus, we are presently investigating the effect of shear forces on the attachment and release of bacteria from PNIPAAm brushes and nano-patterned SAMs. Under controllable applied shear forces, quantitative results can be obtained from spinning disk assays (i) that have shown dramatic differences on bacterial release from PNIPAAm brushes above and below 32 °C, and (ii) that can be used to study the effects of surface chemical heterogeneity on bacterial attachment and release from model surfaces.

MP-32

SURFACE PROPERTIES OF BOTTLEBRUSH POLYMER FILMS

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Bottlebrush polymers are macromolecules with polymeric side-chains on each repeat unit. These materials are structurally analogous to polymer brushes, which are made by end-tethering polymers to a surface and are effective in deterring fouling of man-made surfaces. The use of polymer brushes in large-area applications may be impractical, while bottlebrush polymers can be made in large quantities and solution processed onto a surface. Furthermore, the grafting density and size of the chains in a bottlebrush polymer can be precisely controlled, enabling systematic studies of the effect of chain conformation and composition on fouling resistance. Here, we present a study of the surface properties of bottlebrush polymer and copolymer thin films. Bottlebrush polymers with mixed and copolymer side-chains are prepared via living ring-opening metathesis polymerization of norbornene-functionalized macromonomers. Contact angle measurements, x-ray photoelectron spectroscopy (XPS), and atomic force microscopy measurements on bottlebrush polymer thin films show that polymeric side-chains have significant conformational flexibility and that bottlebrush polymers with mixed and copolymer side-chains can be used to prepare stimuli-responsive surfaces. For bottlebrush polymer with mixed poly(styrene) (PS) and poly(ethylene glycol) (PEG) side-chains, the contact angle depends on processing conditions. When exposed to a hydrophilic solvent which favors PEG, the surface becomes more hydrophilic, while a hydrophobic solvent which prefers PS results in a hydrophobic surface. The reversibility of the surface properties is revealed by both contact angle measurements and XPS. This study represents the first measurement of the surface properties of bottlebrush polymer thin films and indicates these materials may be applicable to creating stimuli-responsive surf aces and non-toxic antifouling coatings.

MP-33

FABRICATION AND INVESTIGATION OF SURFACE-PATTERNED AMPHIPHILIC PERFLUOROPOLYETHER/ POLY (ETHYLENE GLYCOL) NETWORKS: TOWARDS HIGH-PERFORMANCE ANTI-FOULING COATINGS

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The facile solvent-free preparation of amphiphilic network coatings for biofouling marine applications has been demonstrated previously in our group1,2. These high-performance materials, consisting of a hydrophobic dimethacryloxy-functionalized perfluoropolyether (PFPEDMA; *Mw* = 1500g mol-1) crosslinked with hydrophilic monomethacryloxy functionalized poly(ethylene glycol) macromolecules (PEG-MA), are excellent candidates for non-toxic highperformance anti-fouling coatings due to their high chemical and thermal stability, low surface energy and tunable modulus.3 Thus far, only ultra-flat PFPE-PEG coatings have been fabricated with surface roughness of less than one nanometer. Given the reported significant relationship between the interfacial contour and biofouling performance,4 we ultimately became interested in the combinative effects of chemical composition and surface topography. Therefore, using soft lithographic techniques, PFPE-PEG coatings patterned with various pillar-shaped topographies were fabricated. Dimensions of these surface features include 2x2x2 µm, 5x5x2.5 µm, 5x5x5 µm, 5x5x10 µm, and 10x10x10 µm patterns. These films demonstrate lower advancing and receding contact angles relative to the flat PFPE analogues, showing promise to further enhance the anti-fouling properties of our coatings. Comparative studies on the mechanical properties, deterred settlement of algal cells, barnacles and spore adhesion between the prepared PFPE-PEG flat and patterned surfaces will also be reported.

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MP-34

POLY(SULFOBETAINE METHACRYLATE)-MODIFIED DIMETHYLPOLYSILOXANE WITH IMPROVED DIATOM ADSORPTION RESISTANCE PROPERTY

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This paper describes a facile and novel approach for the surface modification of Dimethylpolysiloxane (PDMS) membranes by introducing poly(sulfobetaine methacrylate) [poly(SBMA)] groups grafted via atom transfer radical polymerization (ATRP) for their resistance to bacterial adhesion and biofilm formation. Chemical changes of the membrane surface were characterized by attenuated total reflectance Fourier transform infrared spectroscopy (ATR/FT-IR). Results revealed that poly(SBMA) groups have been successfully introduced to the PDMS membrane surface. The surface properties of membranes were characterized by water-contact angle and the diatom bioassays were measured further to evaluate the antifouling property for the studied membranes. Results showed that poly(SBMA)-modified PDMS membranes had higher diatom-adsoption-resistance property. And the poly(SBMA)-modified PDMS should be potential marine antibiofouling materials and biomedical materials.

Figure 1. *Water-contact angles of PDMS(a) and poly(SBMA)-PDMS(b).*

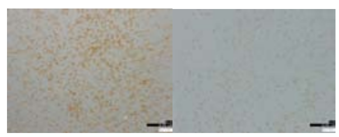

Figure 2. *Diatom adsorption on PDMS(a) and poly(SBMA)-PDMS(b) membrane.*

MP-35

LOW-FOULING AND HIGH-FOULING-RELEASE MARINE COATINGS BASED ON ZWITTER/PDMS COMPOSITES

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Biofouling on ship hulls and other marine surfaces has been a worldwide environmental problem. We have developed a novel polymer coating by combining the advantages of non-fouling zwitterionic and fouling-release PDMS polymers. The coating was prepared from the Zwitter/PDMS composite consisting of both zwitterionic and PDMS segments. The resulting polymer coating renders both hydrophobic and hydrophilic characters. The wettability of these coatings is changed after exposure to water due to surface reconstruction. The coating was evaluated using a number of lab assays and field tests. Low *Ulva* spore settlement indicates that the Zwitter/PDMS composite coating has low-fouling property. High percentage removal of sporelings under low shear stress using a water channel apparatus indicates that the Zwitter/PDMS composite coating has excellent fouling-release property. The field tests of this coating were performed in Florida, Hawaii, California and Singapore with promising results.

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MP-36

FUNCTIONAL POLYMER BRUSHES VIA SURFACE-INITIATED ATOM TRANSFER RADICAL GRAFT POLYMERIZATION FOR COMBATING MARINE BIOFOULING

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Dense and uniform polymer brush coatings were developed and tested to see if they can reduce protein adsorption, bacterial attachment, and barnacle cyprid settlement. Hydrophilic, 2-hydroxyethyl methacrylate, hydrophobic, 2,3,4,5,6-pentafluorostyrene, cationic, [2-(methacryloyloxy)ethyl] trimethylammonium chloride, anionic, 4-styrenesulfonic acid sodium salt and zwitterionic *N,N'*-dimethyl-(methylmethacryloyl ethyl) ammonium propanesulfonate monomers were polymerized via surface-initiated atom transfer radical polymerization (SI-ATRP) to prepare functional polymer brush coatings. The polymer brush-functionalized surfaces show different efficacies in preventing bovine serum albumin (BSA) adsorption, Gram-negative and positive bacteria (*Pseudomonas sp.* NCIMB 2021 and *Staphylococcus aureus (ATCC 25923),* respectively) adhesion and barnacle (*Amphibalanus (=Balanus) amphitrite*) cyprid settlement. Hydrophilic, anionic and zwitterionic polymer brush coatings resist BSA adsorption effectively. Hydrophilic, cationic and zwitterionic surfaces resist bacterial fouling and barnacle settlement. The hydrophobic surface reduces protein and bacterial adhesion, as well as barnacle settlement. The anionic polymer brush coating is the least effective for deterring the attachment of bacteria and barnacles. Based on the present results, the best approach to combat biofouling is to combine hydrophilic and zwitterionic polymer brush coatings on the material surfaces.

MP-37

STUDY OF *PSEUDOMONAS* NCIMB 2021 ADHESION ON STAINLESS STEEL IN PRESENCE OF A PEO-PPO-PEO TRIBLOCK COPOLYMER – A POSSIBLE METHOD TO REDUCE BIOFOULING

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Any materials in contact with a biological aqueous environment will be colonized by microorganisms after a given time. This phenomenon is called biofouling, and microbial adhesion on metal surfaces may cause biocorrosion, which is also named microbially induced corrosion (MIC).

The protection against biofouling and biocorrosion is still a challenge for researchers, with strong consequences in individual application, including cooling condensers treat by seawater. Chlorination is still the most common method to reduce biofouling and biocorrosion in spite of its harm to marine environment. Therefore, an eco-friendly coating to reduce biofouling is highly demanded in marine-related environment.

The initial step of marine biofouling starts by the adsorption of biomolecules (e.g. proteins, polysaccharides) present in seawater, which occurs within minutes after immersion of the material. The organic layer formed this way on metal surfaces, called conditioning film, may facilitate further colonization of the surface by bacteria in reason of modification of the surface properties (surface charge and wettability) as well as of specific interaction. Hence, an effective method to prevent biomacromolecule adsorption may in turn reduce bacteria adhesion.

Here we show that, the adsorption of a PEO-PPO-PEO triblock copolymer on hydrophobized stainless steel surface is useful to prevent protein (BSA, fibrinogen, cytochrome C) and polysaccharide (dextran) adsorption, which was examined by quartz crystal microbalance (QCM), X-ray photoelectron spectroscopy (XPS) and time-of-flight secondary ion mass spectrometry (ToF-SIMS). Bacterial (*Pseudomonas* NCIMB 2021) adhesion on these PEO-PPO-PEO-coated stainless steel surfaces is currently under investigation using parallel plate flow chambers. The amount of adhering bacteria will be numerated by fluorescent microscopy. Ultimately, the brush structure of PEO-PPO-PEO formed on hydrophobized stainless steel surfaces is expected to simultaneously reduce biomacromolecule adsorption and bacterial adhesion.

MP-38

SALT OIL ACIDS AS EFFECTIVE INHIBITORS FOR STEEL CORROSION IN SEAWATER

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For the extraction of oil from the depth of the sea which is injected into oil reservoirs and the sea water. In this case, the risk of corrosion field equipment is greatly increased. Therefore, in the protection of steel equipment operating in offshore oil production, from various forms of corrosion is necessary to use reagents of integrated action. For these purposes, salts of carboxylic and dicarboxylic acids with different structures were used. The inhibitory effect of which is based on their complexing properties. These compounds have good water-and oil-soluble and can form a solid metal surface chemisorption film. We have carried out such studies on the synthesis and testing of these inhibitors of the electrochemical and microbiological corrosion of steel in seawater based on salts of petroleum acids. Synthesis of the salts was based on petroleum acids isolated from the light fractions of Azerbaijani oil. The resulting aqueous solution was treated with saturated sodium soluble mineral salts (RCOONa), taken in equimolar ratios: $MgCl_2$, $AlCl_3$, $CaCl_2$, $CuSO_4$, $ZnSO_4$, $NiSO_4$, $MoCl_4$ and $Ba(NO_3)_2$. The reaction proceeded with vigorous stirring at room temperature. The precipitate was separated and the resulted salts were dissolved in gasoline. The yield of the salts was 97-98% after distillation of gasoline; these salts were well dissolved in aqueous-alcoholic medium. The bactericidal action of the salts was studied in enrichment cultures sulfate-reducing bacteria (SRB) isolated from the water of the Caspian Sea. Potentiodynamic method was used to study the kinetics of anodic and cathodic reactions at the salt concentrations of 100-200 mg/liter. The auxiliary electrode was made of platinum. The investigations revealed that naphthenates-alkaline and alkaline-earth metals effectively inhibit the growth of sulfate-reducing bacteria (SRB) in sea water and have a protective ability in neutral corrosive environments. Naphthenates Ni, Ca, Co inhibits the anodic and cathodic reactions occurring on the steel surface. This indicates that the corrosion inhibitors are mixed action in sea water.

MP-39

FIELD TESTS OF MICROBIOLOGICALLY INFLUENCED CORROSION OF TYPE 304 STAINLESS STEELS: EFFECTS OF SEASONAL CHANGE IN BACTERIAL CONSORTIA AND CHLORIDE CONCENTRATION

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Type 304 stainless steel and copper alloyed antibacterial stainless steel were subjected to field tests. Plain coupon specimens surface dry polished with #1500 emery paper were prepared from type 304 stainless steel, as well as copper alloyed stainless steel. For type 304 stainless steel, tungsten inert gas welded coupon specimens with/without scale removal polish treatment were also prepared. The specimens were subjected to field tests at two eras in Osaka, Japan; one was the Osaka bay era and the other was in the Dotonbori River. The latter era was about five kilometers above the Osaka bay era, where sea water from the estuary and flesh water from an upper side of the river were alternatively fed in by operating a lock gate twice in a day. Seasonal change in bacterial consortia and its effects on microbiologically influenced corrosion was studied.

MP-40

ELECTROCHEMICAL BEHAVIOR OF UNS S31600 STAINLESS STEEL IN THE PRESENCE OF CULTURED JUVENILES OF THE BARNACLE AMPHIBALANUS AMPHITRITE DARWIN, 1854

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Barnacles are the most conspicuous organisms in macrofouling community. Balanid barnacles' growth is one of the principal causes of biocorrosion in marine environments. In the present experiment, cultured *Amphibalanus amphitrite* juveniles were used as a biological model to study balanids' influence on electrochemical behavior of UNS S31600 stainless steel. Larvae were maintained in laboratory until Cypris stage. For settlement assay, one hundred cyprids were added to four individual containers, where each coupon (70x50x2mm) was immersed. After attachment, juveniles were reared for 50 days with *Skeletonema costatum* (Greville) Cleve. Open circuit potential (OCP) was registered every other day during the first week and each three days until the end. Breakdown potential was obtained each week at the first three weeks and each two weeks until the end. For control measures, four coupons were immersed in sterile seawater and four were immersed in seawater with *Skeletonema* solution. Average density was 0,330 $\pm$ 0,129 orgs/cm^2 and basal diameter of the juveniles varied between 1,88mm and 5,80mm. OCP did not show sharp differences between barnacles and control replicate groups during three weeks. However, the last OCP values showed a decreasing tendency in barnacles' replicates. Breakdown potential showed a difference of 300mV between barnacle and control groups. Lowest values were observed in barnacles' coupons, indicating that juveniles' attachment altered UNS S31600 electrochemical behavior when compared to control replicates. Crevice corrosion around barnacles' calcareous bases was observed, even though the small basal diameter. This experiment is part of the first attempts of use this kind of bioassays in corrosion studies. Methodology adaptations seem to be adequate and results were very promising.

MP-41

A COMPARISON OF DNA EXTRACTION METHODS ON NOVEL TEST SAMPLES INCLUDING THOSE CONTAINING CRUDE-OIL

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The direct extraction of nucleic acids from environmental samples has enabled the identification and enumeration of microbes associated with microbially-induced corrosion. Data gleaned in this fashion rely on the extraction of high-quality nucleic acids and amplification of the 16S rRNA or functional genes. Herein, we compared the extraction performance of three different methods: the MOBIO Powerbiofilm, widely used throughout the fields of environmental and industrial microbiology, and two more streamlined methods routinely used in the biomedical and forensics fields: the Fujifilm Quickgene, and Promega Maxwell. Three test samples were analyzed, two of which contained substantial amounts of crude oil, corrosion products such as iron sulfides and extrapolymeric substances (EPS) from "biofilms" and one, from a pipeline that transported seawater, contained lesser amounts of corrosion products and EPS. The two more automated methods (Quickgene and Maxwell) extracted 2- to 10-fold more DNA from the same starting sample volumes compared to the standard Powerbiofilm. Furthermore, extraction concentrations among subsamples were more consistent with these two methods. DNA extractions from all three methods were amplifiable in both quantitative real-time PCR experiments to assess 16S gene copy numbers and end-point PCR to generate libraries for 454 pyrosequencing analysis. The quantitative analysis of 16S rRNA gene copies reflected amounts of DNA extracted, with higher estimates among the more automated methods. Furthermore, the relative abundance of microbial membership in the samples were comparable among the three methods tested. We conclude that the two more automated DNA extraction methods displayed more consistent and higher DNA yields and that extracted DNA from all three methods performed similarly in a series of PCR-based analyses. Therefore, our demonstration that streamlined, automated methods produce high-quality products for molecular analysis means that such analytical tools now become more feasible for monitoring marine corrosion and biofouling.

MP-42

INFLUENCE OF MICROFOULING ON DRAG FOR FOUL RELEASE COATINGS, MEASURED USING A DISC ROTOR APPARATUS

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Foul release coatings facilitate the release of macrofouling organisms but are prone to the growth of microfouling (slime) on their hydrophobic surfaces. Rigid discs coated with commercial foul release coatings were exposed in estuarine seawater prior to the start of the main fouling growth season in order to favour the growth of slime. Visual identification of the main microfouling organisms (diatoms) was made using a variable focus optical microscope which also generated three-dimensional surface profiles. The frictional torque due to slime was measured using a disc rotor apparatus. The flow of water over the 28 cm rotating discs was visualised using neutrally buoyant polymer microspheres. Continuous monitoring of the rotor shaft torque at varying speeds of rotation enabled the drag force on the disc at different Reynolds numbers to be calculated. The drag caused by slime fouling was compared to the drag due to various controlled disc roughness profiles. Some slime was hydrodynamically removed at low speed while a significant proportion was retained even at the highest rotor speed (1000 rpm). The pattern of slime removal was recorded photographically and attempts were made to correlate the degree of removal with flow velocity. Improved knowledge of the flow conditions necessary to remove slime will aid in understanding the overall performance of foul release coatings.

MP-43

BARNACLE GROWTH AT YACHT PROPELLERS – A SERIOUS REASON OF INCREASED FUEL CONSUMPTION

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Barnacle growth on propellers is a well known phenomenon among owners of small yachts as well as larger ships. There is a rumor that growth of the barnacles at the propeller blades may partially destroy the intended propulsion dynamics of the propeller and make them inefficient. To learn more we made a detailed hydrodynamic investigation of the propulsion characteristic of a yacht type-propeller, densely covered with barnacle and compared with the polished propeller.

Method. The hydrodynamics of the freely rotation barnacle covered type-propeller (370 mm) was analyzed in a test bed at the SSPA (www.sspa.se) at different revolutions and speed of the water surrounding the propeller. The type-propeller was then rinsed from barnacles, polished, and then tested again. Beside this, we also made static pull force determinations with dynamometer of five yachts with propellers partly covered with barnacle.

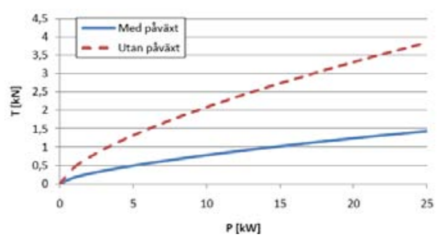

Figure. Left: Barnacle covered propeller Right: The push force (kN) of the tyrpe propeller has been plotted against the applied axis power (kW) of the propeller..

Results. When the power parameter was plotted against propulsion parameterfor the type-propeller we concluded that the barnacle covered type- propeller sometimes needed 4 times more power for one unit of propulsion compared to the polished propeller (right figure) Similar findings were made at the yachts experiments (left figure).

Conclusion. The barnacle growth at small propellers destroy much of the intended propulsion dynamics of the propeller, giving rise to a dramatic decrease in propulsion efficiency and, when used on yachts, a dramatic increase in fuel consumption per unit distance of travelling.

MP-44

HYDRODYNAMIC DRAG FORCES ASSOCIATED WITH LOW-FORM FOULING

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Low form fouling is present on most modern day ship hull coatings. These include biofilms, incipient fouling, algae, bryozoans and hydroids. This study measured the drag forces associated with different fouling communities that were grown on 250 x 300 mm epoxy coated panels. The panels were mounted to a custom made through-hull hydrodynamic drag meter which is incorporated into a wet-well of a 10 m power boat. This enabled the real-time measurement of drag forces at velocities up to 15 m/s. The panels were exposed under static conditions on the east coast of Florida and monitored for the development of biofouling. The fouling communities were characterized before and after each test run and related to the drag measurements. This data is being compiled to produce a library of drag coefficients with known fouling communities.

MP-45

SHIP HULL COATING INSPECTION

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Ship hull coating inspection was used to evaluate antifouling performance and estimate the powering penalty due to biofouling and hull coating condition on two ships during dry docking. The hulls were divided into eight zones and inspections were made immediately after drawdown, after pressure washing and after paint application. The methods included visual inspection, biofilm sampling, dry film thickness and hull roughness measurements. The data will be presented and discussed with respect to coating type, application, biofouling and performance.

MP-46

THE PROTOTYPE OF A LASER HULL ROUGHNESS PROFILOMETER FOR USE IN DRYDOCKS

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Hull roughness on ships is measured and most commonly presented as the maximum peak to lowest trough height (Rt50) parameter expressed in microns, in any given length of 50mm along the underwater hull. The BMT Hull Roughness Analyser has been the industry standard instrument for the measurement of hull roughness for over 6 decades. A hand propelled measuring head is traversed over the hull surface and the roughness determined by monitoring the displacement of a stylus relative to a curved skid. After the ban of TBT SPC coatings new technologies have been developed which require a new, ideally non contact device as the stylus and the skid are not effective over the new silicone based technologies. It has also been realised that the use of a single roughness parameter gives an incomplete representation of the concept of roughness, because two surfaces with the same value of height or amplitude parameter like Rt_{50} may have quite different textures which also affect frictional drag. At least one additional parameter is therefore necessary to describe the texture. The presentation will introduce the prototype laser hull roughness analyser developed as well as the results of the roughness parameters as compared to the results of a commercial desktop laser profilometer. This device could potentially give some indication on the effect of slime on the roughness characteristics of coated surfaces.

MP-47

BIOCIDE ANTIFOULING PAINTS-BENEFITS VS. CONTINUING ENVIRONMENTAL COSTS

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There are around 2000 biofouling marine species of which barnacles, a family which evolved impermeable, insoluble glue long before humans invented similar materials, are among the most significant. For shipping, the speed decrease and maintenance increase led to attempts to control biofouling, using concoctions including arsenic, sulphur and gunpowder and metal coatings (lead, copper). Latterly antifoul compounds became based on toxic biocide organometallic compounds which poison and self-polish thus impacting fouling and non-target species.

There is no doubt that hull-fouling control has significant environmental benefits. Efficient shipping burns less fuel thus reducing CO_2 output and the inhibition of fouling organisms reduces exotic species transport, one of Jared Diamonds quartet of evil, whilst facilitating global trade. However, the adverse effects of biocide paints have become infamous, most notably through the endocrine disruption and bioaccumulation effects seen from tributyltin (TBT). Furthermore, the long-term, though possibly decreasing, legacy of TBT in marine sediments also creates a “reservoir” which can be problematic to manage in dredge spoil and water quality terms.

TBT efficacy is claimed to have stalled alternative antifouling research, however with the ban on smaller craft and the 2008 IMO agreement on the banning of TBT and other harmful antifoulants, a return to heavy metal (primarily copper) based antifoul with booster biocides, was promoted. As with TBT non target organisms have proven to be susceptible with potential wider trophic-diversity implications through bioaccumulation.

Copper antifoul is more species specific than TBT hence the addition of booster biocides, but, the reliance on metallic paints to achieve fouling inhibition is receiving increased attention as it still has species and community implications and still creates a long term sediment reservoir.

It has been stated that “there is no simple and nontoxic solution for the biofouling problem [but that] copper containing coatings are considered as a transition between toxic and non-toxic coatings.” To support this, greater cost benefit research is needed into nontoxic coatings, perhaps through the wider ecosystem services concept. Accordingly greater consideration of benign alternatives, regular hull cleaning leading to fouling minimisation, reduction in exotics transport and contaminant build up in marine sediments appears warranted.

MP-48

RIGID FIBER COATINGS AS EFFECTIVE ANTIFOULING SURFACES FOR BOTH HARD AND SOFT FOULING – 5 YEAR RESULTS AND MECHANISM INTERPRETATION

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Today, antifouling paints are recognized as one of the substantial water polluting sources.[1]Therefore, the demand for environmentally friendly alternative is evident. Until recently, it has been unsuccessfully tried to develop effective nontoxic antifouling surfaces based on fiber technologies. Effectiveness was found for some organisms, for others - accelerated growth.[2] However, it turned out possible to prevent a complete range of macrofouling species relevant to industrial fouling problems by changing to a whole new spectrum of fiber lengths, thicknesses and densities.

A fundamental study on the working principle of the fibers was conducted. A broad range of fibers was trialed on flat panels in the North Sea. Based on these results, an optimal fiber was selected. In April 2007 full scale trials in aquaculture were launched. Since then, nets of 20 to 50 m diameter were deployed in warm Mediterranean water as well as in cold Norwegian/Scottish waters. Inspections were done periodically during the following 5 years. The results were evaluated with respect to copper. During these trials it was established that the curvature of an object plays an important role in the overall fiber effectiveness. Trials in shipping were also carried out in 2010 and 2011. Professional craft and pleasure craft were coated in the Netherlands and Florida, respectively. The results are compared to commercial antifouling coatings as reference.

In the paper a model will be presented regarding the working principle of the fibers. The results of the panel tests and the results of the aquaculture trials will be summarized. An outlook about the shipping industry and critical issues for this industry will be presented, i.e. application methods, durability, drag and antifouling performance. Finally, conclusions are drawn regarding the potential and the effectiveness of rigid fibers as antifouling technology.

1 US Environmental Protection Agency. Safer Alternative to Copper Antifouling Paints for Marine Vessels. Issued January 2011. Project NP00946501-4.

2 Aimee L. Phillippi. Surface flocking as a possible anti-biofoulant. Aquaculture 195 (2001), pp. 225–238.

MP-49

HULLBUG, AN AUTONOMOUS HULL CRAWLING ROBOT FOR PROACTIVE HULL GROOMING AND INSPECTION

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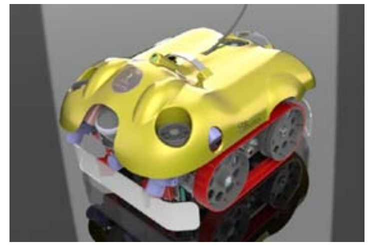

The HullBUG, or Hull Bio-inspired Underwater Grooming System, is an autonomous, hull crawling bio- film grooming robot. Recent studies performed by the US Navy suggest significant fuel savings will be achieved through the use of a proactive grooming process. Proactive grooming utilizes a frequent, gentle cleaning cycle intend to maintain ship hull fouling at a bio-film level. The increased energy efficiency and fuel savings associated with proactive grooming has been independently verified by ship operators through financial and fuel efficiency studies of their operations. Higher efficiency leads to reduced fuel cost, increased speeds, and a reduction in the ship's carbon footprint. Additionally, it is noted that the transport of non-indigenous species is reduced and hull coating longevity improves. Enabling technology is being developed to allow the unmanned grooming process to proceed as frequently as once per week, in a cost effective manner. The small lightweight system can be deployed frequently without coating damage due to minimal attachment forces and brush contact pressure. Autonomous operations are enabled through the utilization of low cost attitude sensors, rate sensors, and accelerometers. The addition of situation awareness sensors and bio-film detection allows the HullBUG to navigate on progressively complex areas of the hull. Test results on Navy ships and surrogate test platforms indicate a near term solution is at hand. Navigation techniques and grooming capabilities are explored. The HullBUG platform has also been adapted to ship hull inspection tasks, starting with coating and plate thickness potentially leading to more sophisticated crack and pitting corrosion detection.

The HullBUG system is adaptable to a broader range of bio-fouling removal solutions. Significant challenges in aquaculture, oil and gas asset production, and power generation can be addressed by the HullBUG system.

MP-50

BIOFOULING FIELD SURVEY ON THE HULLS OF EX-USS FORRESTAL AND EX-USS SARATOGA

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In-water biofouling surveys on the decommissioned aircraft carriers Ex-USS *Forrestal* and Ex-USS *Saratoga* were conducted in the summers of 2009 and 2010 to support planning of the eventual displacement of these inactive vessels from their current docking location in Newport, Rhode Island. Qualitative surveys aimed to document fouling organisms at niche areas (i.e., struts, rudders, and propellers), while quantitative surveys were based on randomly sampled 0.5m^2 quadrats of percent cover on submerged surfaces of the vessels (i.e., waterline, hull, bilge keel, and bottom of the ships). Surveys were monitored and recorded from the surface using real-time audio and video, which allowed for collaborative sampling and on-site taxonomic identification. As anticipated, long residence times in Newport, combined with no hull husbandry since last drydock, have allowed for the development of high-density biofouling assemblages. Heterogeneous 'niche' areas were extremely susceptible to fouling accumulation. The biofouling communities of the vessels were comprised of a number of taxa including soft-bodied fouling organisms (algae, anemones, sea stars, sponges, sea squirts, tunicates, and bushy bryozoans), as well as hard fouling organisms (corals, barnacles, mussels, calcareous polychaetes, and encrusting bryozoans). Differences in biofouling communities existed between the vessels. Sixteen species contributed to approximately 67% of the differences observed between the biofouling communities of the two vessels and may reflect variability from patchiness in biofouling cover and duration of time in port. In-water hull surveys have proven to be successful in examining hull fouling on Navy vessels and could result in savings in dry-docking, maintenance, and repair costs for active vessels.

MP-51

PROGRESS IN THE DEVELOPMENT AND FULL-SCALE TESTING OF ELASTOMERS PROTECTED FROM MARINE BIOFOULING BY AN ISOTHIAZOLINONE-BASED BIOCIDE

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The U.S. Navy has used biocide-impregnated elastomers for a number of specialty applications including surface ship SONAR domes and small acoustic windows/domes. Conventional antifouling paints and coatings perform poorly in these applications because of extensive flexing by the rubbery substrates and/or the need for smooth water-elastomer boundaries for good acoustics. Cavitation induced by underlying acoustic sources or high speed maneuvers can also damage/dislodge paints and coatings applied to elastomers. Traditionally, rubbers containing tributyltin oxide (TBTO), an effective, but toxic and environmentally persistent biocide, were used for these applications. International agreements banning the sale and use of TBTO have eliminated the use of TBTO-containing elastomers within the U.S. Navy with the exception of bow SONAR domes for certain destroyers and frigates. We have been assessing the efficacy of a "greener," more environmentally friendly, isothiazolinone-based biocide known as "DCOIT" or 4,5-dichloro-2-n-octyl-4-isothiazolin-3-one (Dow Chemical Company) as an alternative for TBTO in these applications. DCOIT breaks down very quickly in sea water and sediments, exhibits essentially no bioaccumulation factor, and has a maximum allowable environmental concentration about 300 times greater than TBTO. We have successfully incorporated DCOIT into EPDM and neoprene rubber, and several different polyurethanes. Long-term static panel immersion results from multiple test sites will be presented, along with diffusion and rotating cylinder release-rate data for several DCOIT-containing elastomers. State and Federal approvals have been obtained for a full scale test of DCOIT-containing rubber/polyurethane as a boot on an experimental composite-based surface ship bow SONAR dome, and the antifouling performance of these materials will be monitored from 2012-2015 as a step in the possible replacement of the TBTO-containing rubber currently used in this application. The development and testing of a small acoustic sensor dome for submarines made from DCOIT-containing polyurethane (in place of a copper (I) oxide-coated polyurethane dome) will also be discussed.

MP-52

DURABILITY ASSESSMENT OF FOUL-RELEASE COATINGS

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The Bureau of Reclamation (USBR) is currently investigating the feasibility of using foul-release coatings to mitigate impacts caused by macro-fouling organisms such as zebra and quagga mussels. Durability of these coatings must be considered as a factor in determining overall life cycle costs. To this end, the Bureau of Reclamation has developed testing protocols to evaluate the durability of foul-release coatings with respect to abrasion, erosion resistance and the ability to overcoat existing equipment. This paper details each testing methodology and presents results. The tests show a wide variation in the durability of commercially available foul-release coatings. In order to estimate the expected product life for specific service environments, the results are compared to several benchmark coatings which have an extensive service history within Reclamation. These results show that silicone coatings have the potential to work under certain conditions where floating debris is not present. Silicone epoxy and polyurethane systems are more durable but less resistant to erosion damage caused by entrained particulates such as sand. None of the coating systems are recommend as an overcoat material for coal tar enamel.

Plenary 2

PLENARY-2

TOWARDS TOUGH FOULING-RELEASE COATINGS WITH TAILORABLE SURFACE COMPOSITION

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In the search for non-toxic ship hull coatings, the fouling-release coating concept has gained considerable attention and most major marine coatings suppliers have commercialized fouling-release coatings. Typically based on silicone elastomers, these coating have several drawbacks including poor mechanical durability, challenges with adhesion to marine primers, and the build-up of slime fouling. An alternative approach to improving the properties of fouling-release coatings involves decoupling the bulk and surface properties through the phenomenon of self-stratification. Hybrid coatings containing a functional low surface energy polymer and a mechanically robust bulk composition will result in a system having a low surface energy due to surface segregation of the low surface energy polymer. For example, poly(dimethylsiloxane) (PDMS) having reactive functional groups can be incorporated into a polyurethane coating system to yield a mechanically robust coating having fouling-release performance. Careful tuning of the compositions enabled by combinatorial and high throughput experiments have been shown to be essential in identifying successful compositions. Siloxane-polyurethane coatings have been shown to be robust, have good adhesion and withstand repeated cleanings. To further adjust the surface composition yielding coatings having amphiphilic surfaces, the PDMS component can be modified via the incorporation of hydrophilic groups such as polyethylene glycol and carboxylic acids.

MICROBIOLOGICALLY INFLUENCED CORROSION
Session Chair: Iwona Beech

MIC-1

THE ROLE OF MOLECULAR TECHNIQUES IN ELUCIDATING MICROBIOLOGICALLY INFLUENCED CORROSION

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Molecular techniques are widely applied to determine the structure and species composition of microbial communities and to infer their major physiological processes. Molecular techniques such as polymerase chain reaction (PCR), quantitative PCR (qPCR), denaturing gradient gel electrophoresis (DGGE), cloning, and pyrosequencing have begun to be more commonly used in the study of microbiologically influenced corrosion. They hold great promise for detecting and monitoring the microbes responsible for fuel biodegradation and biocorrosion. However, every technique has strengths and weaknesses, which cannot be ignored if scientifically defensible conclusions are to be drawn. The types of downstream analyses proposed will dictate sample volumes and the types of samples collected, e.g. metal scrapings, biofilm material, filtered seawater, etc. Generation of good data begins with proper handling and preservation of samples and requires a series of quality control measures at each step. Documentation of pertinent metadata, such as temperature, detailed sample descriptors, corrosion status, volume or weight of the sample is also crucial for interpretation of results. Obtaining usable nucleic acids from biocorrosion samples containing metals is challenging, as metals such as iron are inhibitory to many downstream applications. Published techniques may need to be revised to provide high quality nucleic acids. There are a large number of oligonucleotide primers available for PCR-based analyses and the primer set chosen will determine the types of microbes and processes detected. Therefore, the specificity and sensitivity of primers must be evaluated and may need to be specially designed for particular projects. The application of molecular techniques in our laboratory will be illustrated by elucidating the microbial communities and processes associated with marine biocorrosion 70/30 Cu-Ni alloys and with the biocorrosion of carbon steel exposed to seawater and FAME diesel.

MIC-2

THE STUDY OF BIOFILM-INFLUENCED MARINE CORROSION OF CU-NI ALLOY

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Investigations were undertaken to determine the cause of unexpected accelerated marine corrosion of 70/30 Cu-Ni alloy at three different geographical locations. Sites coded T1 and T2 represented corroding systems at an average temperature of 24^oC, while site V, at an average temperature of 10^oC, was a non-corroding location. To determine whether reported field failures were due to biofilm-influenced corrosion, Cu-Ni specimens were exposed to complex prokaryotic communities, recovered from T1, T2 and V systems. For each system, aerobic and anoxic enrichments, selective for slime- and acid-producing and sulfide-generating microorganisms, respectively, were obtained from fouling layers. Combined enrichments, representing each system, served as inocula for continuous flow bioreactors operating for 6 months with filter-sterilized natural seawater at two different temperatures (10^oC and 24^oC for systems T1 and V and 24^oC for system T2). Controls consisted of specimens exposed in sterile bioreactors. Field emission scanning electron microscopy and digital light microscopy imaging of Cu-Ni specimens exposed in inoculated bioreactors revealed abundant biofilms. Varying levels of pitting attack were observed on specimen surfaces after biofilm removal. Each system had produced a distinct pitting morphology. Localized pitting, similar to that reported in field failures, was observed in the V-24 ^oC and T1-, T2 -24 ^oC bioreactors. Micro pits detected on Cu-Ni surfaces of control specimens did not exceed depths measured for Cu-Ni alloys in marine environments. Molecular characterization of inocula and resulting biofilms, carried out employing functional gene microarray (GeoChip, 4th generation) revealed similarities between enriched microbial communities obtained from different geographical locations. The GeoChip results confirmed a high abundance of Cu-resistant genes in biofilm populations, irrespective of their origins. The frequency of genes representing S-redox pathways and methanogenesis was considerable. The study has demonstrated unambiguously that microbiologically-influenced corrosion (MIC) caused the failure of Cu-Ni alloys and that microbial metabolism plays a key role in MIC.

MIC-3

MILD STEEL CORROSION IN NEARSHORE MARINE ENVIRONMENTS - ASSESSING THE PRESENCE OF IRON-OXIDIZING BACTERIA AND CHARACTERIZING THE OVERALL BACTERIAL COMMUNITY

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Little is known about the microbial ecology of corroding steel in marine environments[1,2] or of the natural abundance of iron-oxidizing bacteria (FeOB) in these systems. We hypothesized that coastal sediments are reservoirs for the marine FeOB 'Zetaproteobacteria' (Zetas), and that they can colonize and become numerically abundant on mild steel surfaces. A 40 day time series incubation was conducted in a salt marsh (summer 2010). Corrosion community DNA was extracted and analyzed for bacterial diversity with tagged pyrosequencing (V4 region, 16S rRNA gene). Several relevant communities were quantified using qPCR: bacteria and archaea[3] and Zetas[4] using 16S rRNA gene specific primers, and sulfate-reducing bacteria (SRB) using a dsrA gene specific primer[5]. The pyrosequencing data showed the presence of Zetas in sediments and throughout the incubations on the steel samples. Iron oxyhydroxide stalk biosignatures were observed on samples, further evidence that these sequences likely represent FeOB. Relatives of the H2-oxidizing genus Hydrogenophaga and members of the family Rhodobacterales were also identified as important members of the biocorrosion community and were present both on steel and in sediments. Gene copies assessed with qPCR remained fairly constant in sediments during the study, and Zetas were ca 10-fold lower than SRB. Zetas colonizing the steel increased rapidly over the first 10 days, exceeding copies quantified in the sediment by an order of magnitude. The SRB numbers on the steel were 10 fold lower than in sediments during the first days of incubation, but increased to near the sediment levels by 40 days. This work illustrates that coastal sediments are a reservoir for Zetas who, though numerically low in sediments, can quickly colonize environments where free Fe(II) is abundant.

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MIC-4

MARINE MIC OF MILD STEEL - ELECTROCHEMICAL ANALYSIS OF MODEL CORROSION COMMUNITIES

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Previous studies have shown that Fe(II)-oxidizing bacteria (FeOB) and Fe(III)-reducing bacteria (FeRB) are involved in steel corrosion, and enhance mild steel corrosion in laboratory studies[1,2]. The objective of this work was to determine the electrochemistry of mild steel challenged with single strains of FeOB and FeRB vs co-cultures of FeOB and FeRB. Batch experiments containing mild steel coupons in marine medium were conducted in green flasks. Pure and mixed cultures of marine FeOB (*Mariprofundus ferrooxydans* strain M34)[3] and FeOB (*Geothermobacter* sp. strain HR-1)[4] were used in each system, and controls containing no added FeOB and FeRB were also prepared. Pure FeOB were grown in an aerobic bulk medium and pure FeRB were grown under anaerobic conditions. Corrosion rates were monitored electrochemically, and following incubation, steel surfaces were evaluated with ESEM and profilometry. An FeOB and FeRB co-culture was successfully grown in an bulk aerobic environment, and the FeOB-generated iron oxide stalks in this treatment appeared denuded in comparison with those formed in the pure FeOB system. Profilometry demonstrated less uniform corrosion attack in the presence of FeOB and FeRB co-culture compared to all other exposures. Electrochemically monitored polarization resistance suggested that all aerobic corrosion rates were similar and orders-of-magnitude higher than anaerobic corrosion rates. Further work developing model systems for assessing the individual and collective influences of key microbes on corrosion include incorporation of sulfate-reducing bacteria.

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MIC-5

MARINE CORROSION IN FUEL SYSTEMS

Brenda J. Little[1], Jason S. Lee[1], Richard I. Ray[1], Deniz F. Aktas[2], Kathleen E. Duncan[2], and Joseph M. Suflita[2]

The relationship between corrosion and biodegradation of bio- and petroleum-based fuels exposed to seawater is being evaluated. To date the fuels have included petroleum diesel (F76) and jet propellent (JP) 5, hydroprocessed (HP) bio-based lipids from renewable stocks (e.g. camelina and algae) and blends. Experiments have been conducted with aerobic seawater and unprotected carbon steel coupons under stagnant conditions. i.e., there were no attempts to influence the distribution or concentration of oxygen in the sealed vessels. In all cases the dissolved oxygen (DO) in the seawater was below the detection limits of the DO probe (100 ppb) within a few days of incubation, independent of fuel composition. Corrosion was due to microbiologically produced sulfides reacting with the carbon steel. There were few differences in electrochemically measured corrosion rates in incubations amended with any of the fuels or their blends. In the experiments that have been examined in detail, transient oxygen influenced the microbial biodegradation of fuels and resulted in a suite of characteristic metabolites. Detection of catechols confirmed the exposure of incubations to oxygen. Clone library analysis indicated higher proportions of Firmicutes, Deltaproteobacteria (primarily sulfate-reducing bacteria), Chloroflexi, and Lentisphaerae in incubations exposed to fuels than the original seawater. Relative proportions of sequences affiliated with these bacterial groups varied with fuel. Methanogen sequences similar to those of *Methanolobus* were also found in multiple incubations. Despite the dominance of characteristically anaerobic taxa, sequences coding for an alkane monooxygenase from marine hydrocarbon-degrading genera was observed, suggesting that organisms with this metabolic potential survived the incubation. The current hypothesis is that initial aerobic oxidation of fuel components resulted in the formation of a series of intermediates that were used by anaerobic seawater microbial communities to support their metabolism, sulfide production, and carbon steel microbiologically influenced corrosion. The more precise relationship between oxygen, microbial activity and corrosion is underway with more precise DO probes (4 ppb resolution).

MIC-6

SULFIDE PRODUCTION AND CORROSION IN SEAWATER DURING EXPOSURE TO FAME ALTERNATIVE FUEL

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Experiments were designed to evaluate corrosion-related consequences of storing/transporting fatty acid methyl ester (FAME) alternative diesel fuel in contact with natural seawater under anaerobic conditions. Coastal Key West, FL, and Persian Gulf seawaters, representing an oligotrophic and a more organic- and inorganic mineral-rich microbial coastal seawater environment, respectively, were used in 60-day studies with unprotected carbon steel. Despite low numbers of sulfate reducing bacteria in the original waters and after FAME diesel exposure, sulfide levels and corrosion increased markedly due to microbial sulfide production. The original microflora of the two seawaters was similar with respect to major taxonomic groups but with markedly different species. After exposure to FAME diesel the microflora of both waters changed dramatically, with Clostridiales (Firmicutes) becoming dominant. Microbial sulfide production was stimulated in both seawaters by the presence of FAME.

MIC-7

ARE BACTERIA INVOLVED IN THE EARLY STAGES OF CARBON STEEL CORROSION PROCESS IN MARINE ENVIRONMENT?

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The aim of this study was to better understand the mechanisms involved in marine corrosion of steel structures. These mechanisms are known to be influenced by micro-organisms, in particular by the sulphide-producing bacteria [1;2]. It was then necessary to couple physico-chemical investigations with microbiology and molecular biology techniques. In order to evaluate the part played in biocorrosion by different microbial groups, we immersed carbon steel surfaces (coupons) for increasing times (from 1 week to 2 months) in a French Atlantic harbour and we characterized the corrosion products and the associated bacteria simultaneously. We wanted to obtain cultured representative bacteria, but we knew the limitations of cultivation-based profiling of microbial communities. Then, a combination of culture-dependent (traditional cultivation techniques) and culture-independent approaches (TTGE and cloning 16S rRNA genes) was applied.

The simultaneous monitoring of microbiological and physico-chemical data showed that the influence of bacteria on the rust layer formation was negligible at these early stages. However, bacteria whose growth is associated with iron and sulphur were detected. Moreover, the process influenced by sulphate-reducing bacteria was detected locally. Whatever the immersion time, an important biodiversity was found for the total bacterial flora as well as for the cultivated bacteria, despite a low rate of individually cultivable bacteria (less than 1.8%). Mixed cultures allowed the detection of sulphate-reducing bacteria which we were enable to cultivate individually, suggesting the existence of symbiotic relations between bacterial species. In conclusion, by comparison with previous results [3;4], the studied corrosion process seems to move from the abiotic process to a process influenced by sulphate-reducing bacteria between the 2nd and the 6th month of immersion.

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MIC-8

EXPERIMENTAL INVESTIGATION INTO THE EFFECTS OF ICCP AND SCALE FORMATION ON MIC ON HSLA STEEL

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Microbiologically Influenced Corrosion (MIC) is extremely aggressive and in its worst form it will lead to unpredictable damage to a structure within a short time period. It is virtually impossible to predict when and where bacteria will strike although to a certain extent the risk of corrosion due to microbes could be established. Hence, prevention of MIC is of utmost importance for the assurance of the integrity of materials and installations.

Cathodic protection (CP) is a corrosion protection system widely used both in the maritime field and in the energy sector and has been found to have influence on MIC processes. The objective of this study was to investigate in a laboratory setup the effects of continuous impressed current cathodic protection (ICCP) and pre-deposit calcareous layer applied through CP, on MIC mixed biofilm of *P. fluorescens* and *D. indonesiensis*, on HSLA type 355 steel. Microscope images of surfaces were taken from all panels before and after the experiments. Corrosion of panels was assessed through visual observation and light microscopy. Biofilms were assessed using epifluorescence microscopy after staining with 4',6-diamidino-2-phenylindole (DAPI).

Based on the results the following conclusions were drawn: CP did not inhibit biofilm formation. In presence of biofilm CP of -1.050 V (Ag/AgCl) showed less or no corrosion on smooth steel surfaces even after two months exposure period, while -0.950 V (Ag/AgCl) cathodically protected steel showed pitting attack within 10 days. Bare steel without a biofilm is protected against corrosion at -0.850 V (Ag/AgCl). Steel panels with a calcareous layer (developed during 5 days exposure to – 1.050 V (Ag/AgCl)) and subsequent exposure to MIC bacteria showed similar biofilm growth but corrosion attack could not be observed.

GENERAL ASPECTS OF FOULING: MACRO FOULING
Session Chair: Mike Hadfield

MACRO-1

POPULATION DYNAMICS OF THE BARNACLE *BALANUS AMPHITTRITE* IN THE TROPICAL MONSOON-INFLUENCED ENVIRONMENT OF INDIA

A C Anil

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Barnacles often dominate fouling communities. *Balanus amphitrite* (syn. *Amphibalanus amphitrite*), a shallow water acorn barnacle, cosmopolitan in distribution, is capable of inhabiting expanses from supra-littoral to sub-tidal levels, and as an epibiont. Integrating facets of developmental biology and larval ecology into research is important to unravel the complexities in the larval settlement of this organism. The availability of right food type is of fundamental importance in the development of planktotrophic nauplii. In nature, the larvae fulfill their energy demands from a soup of available food material. From larval fecal pellet analysis, it is now evident that successful larval development in barnacles is possible on varying forms of food, and it can vary temporally. Larval abundance can drive variations in settlement at a given locality, and the dispersion or retention processes regulate this factor. The need to settle close to conspecifics is yet another requirement so that reproduction is facilitated. In view of this, understanding the basis of gregarious behavior in barnacles is of utmost importance in elucidating the mechanisms involved in structuring their populations. Recent studies have shown that barnacle larvae may be able to detect parentally associated biofilms and use this information to settle close to members of their own species. The talk will provide an overview of settlement and recruitment of *B. amphitrite* with special reference to the tropical monsoon- influenced environment of India.

MACRO-2

A COMMON THEME IN BACTERIAL INDUCTION OF MACROFOULING

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Bacteria are the earliest foulers on submerged surfaces and often serve as inductive cues for macrofouling organisms. The serpulid polychaete *Hydroides elegans* is major contributor to fouling communities in subtropical waters, and larvae of *H. elegans* are induced to metamorphose by surface bound cues from microbial biofilms. We have isolated a strain of *Pseudoaltermonas luteoviolcea* (strain HI1) from Hawaiian biofilms that induces larvae to metamorphose. However the nature of the inductive cue has remained elusive. We used mutational analysis of the bacterial chromosome to identify four genes that are required for the induction of metamorphosis of *H. elegans* by *P. luteoviolecea* strain HI1. The mutated genes may be involved in cell-adhesion and bacterial secretion systems and not directly involved with the formation of a biofilm. Comparisons between biofilms generated by the four mutant strains and wild-type strain showed no differences in factors relating to the physical composition of the biofilms (bacterial density, biofilm thickness, biomass and EPS biomass), indicating that induction is most likely due to a specific molecular entity. Larvae of the common Indo-Pacific coral *Pocillopora damicornis* also settle in response to *P. luteoviolacea*, and preliminary data indicate that the same bacterial genes are involved in the induction process. Recent published data reveal that species of the bacterial genus *Pseudoaltermonas* are inductive to settlement of a spectrum of benthic invertebrate animals, raising the strong possibility that recruitment of many marine macrofouling animals may result from not only a small group of bacteria, but also common products from the bacteria.

MACRO-3

DO BACTERIA EAT BARNACLE GLUE?

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Biofilms often influence settlement of fouling organisms. We are examining the interaction between marine bacteria and barnacle glue. Bacterial strains that comprise biofilms and interact with glue may influence ecological processes. Our specific interests are to determine whether bacteria use glue as a food source, if barnacles manage bacteria, and if peptide barnacle pheromones attract bacteria. Using epifluorescent microscopy we observed that bacteria localize on cyprid permanent adhesive and are present under young barnacles (0-6 days old). Over 10-14 days, there is a striking decrease in the bacterial load present. In a direct contact test, bacteria grow in the presence of barnacle glue in aerobic conditions. Bacteria that secrete proteases and bacteria that do not secrete proteases grow faster in the presence of glue, demonstrating the potential of bacteria to grow on small and large components of the glue. Experiments with aged glue (the span of 0-14 days) show that fresh glue (initial stage of curing) stimulates bacterial growth more than older glue. We are currently investigating whether growth is stimulated under anaerobic and/or microaerophilic conditions as would be found under barnacle base plates and if peptide pheromone mimics attract bacteria. Our data support the hypothesis that bacteria utilize the barnacle glue as a food source. The relationship between barnacles and bacteria is complex and there is the potential for negative and positive interactions.

MACRO-4

OBSERVATIONS ON THE LOCATIONS OF ADRENERGIC-LIKE RECEPTORS IN THE BARNACLE (*BALANUS AMPHITRITE*) CYPRID LARVAE AND OYSTER (*CRASSOSTREA)*

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Exploration of substrata prior to cementation and metamorphosis is a critical step in the life cycle of many invertebrates. To this end, barnacle cyprid larvae and oyster pediveliger larvae have evolved sophisticated sensory mechanisms to investigate chemical as well as topographical composition of the substratum. Although both barnacles and oysters are major biofoulers, the sensory structures of barnacle cyprids alone have been described in detail using ultrastructural techniques like SEM and TEM, the same is not true for the oyster pediveliger sensory structures. Settlement assays have shown that pediveliger and cyprid settlement is

deterred by adrenergic receptor agonists covalently linked to polymer surfaces1, indicating ability of the larvae to sense surface-tethered molecules. Yet the exact location of these receptors on cyprid and pediveliger sensory structures is unknown. Here, using a fluorescent adrenergic receptor antagonist and neuron-specific antibodies with the aid of confocal microscopy we describe the putative locations of adrenergic-like receptors in *B. amphitrite* cyprids and *Crassostrea gigas* pediveligers. Our results indicate that postaxial setae 2 and 3 of the 2nd and 3rd segments respectively, the sub-terminal setae of the 4th antennular segment and the cement gland secretory cells seem to bear the adrenergic-like receptors. The foot of the oyster pediveligers also bears the adrenergic like receptors and we show the innervations of the pedal ganglion in the foot of the pediveliger larvae. With the use of laser scanning confocal microscopy, virtual optical sectioning allows imaging of live or fixed larvae in hydrated and native states, eliminating the need for conventional histology where samples are dehydrated, embedded in a matrix and physically sectioned.

Acknowledgements: Authors NVG and ASM would like to acknowledge support of US Office of Naval Research awards N00014-1-11-0183 and N00014-08-1-0157 at Clemson University.

References: 1Gohad, N.V., Shah, N.M., Metters, A.T., Mount, A.S., 2010. Noradrenaline deters marine invertebrate biofouling when covalently bound in polymeric coatings. Journal of experimental marine biology and ecology 394(1-2), 63-73.

MACRO-5

BIOFOULING – UNSOLVED PROBLEMS, INSUFFICIENT APPROACHES AND SOME LIGHT AT THE HORIZON

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Microbial biofouling is a very costly problem. Five general reasons can be identified which continuously contribute to costs: i) Biofouling is detected by its effect on process performance or product quality and quantity. Early warning systems are very rare although they could save costly countermeasures necessary for removing established fouling. ii) For diagnosis, the common practice is to take water samples which give no information about site and extent of biofouling deposits. iii) When finally the diagnosis is established, biocides are employed which, in many cases, for the best kill microorganisms but do not remove them. Killing is not cleaning, and in many cases, the presence of biomass and not its physiological activity is the problem. iv) Nutrients are not considered as potential biomass. Biofouling is a biofilm phenomenon and based on the fact that biofilms grow at the expense of nutrients; oxidizing biocides can make things even worse by breaking recalcitrant molecules down into biodegradable fragments. v) Efficacy control is performed again by process performance or product quality only and not optimized by biofilm monitoring, verifying successful removal. Thus, further biofouling is inevitable. In order to overcome this vicious circle, an integrated strategy is suggested. It includes the selection of low-adhesion, easy-to-clean surfaces, good housekeeping, early warning systems, limitation of nutrients, improvement of cleaners, strategic cleaning and direct monitoring of deposits. The goal of a holistic anti-fouling strategy is: to learn how to live with biofilms and keep their effects below the level of interference in the most efficient way by combining the approaches.

MACRO-6

LARVAL BIOLOGY OF THE TROPICAL OYSTER *DENDOSTREA FOLIUM* IN SINGAPORE WATERS

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The tropical Southeast Asian coastal waters of Singapore's exhibit high fouling rates as well as high biodiversity. Particularly, bivalves can be a serious problem for the shipping industry in these waters since they can attain considerable biomass in biofouling communities. Therefore a better understanding of the biology of some key species might prove to be useful for the antifouling industry as well as for the academic world. *Dendostrea folium*, also know as leaf oyster, is a significant component of biofouling communities in Singapore waters. This tropical oyster is known to be brooding its larvae inside its pallial cavity but there is little information about the biology and ecology of its larvae, particularly concerning their morphology and physiology as well as the utilization of energy reserves during development. Larval morphology was therefore characterized using Scanning Electron Microscopy and also provided information regarding the potential feeding habits of the larvae. Furthermore, various algal diets were tested to establish what influence food source may have on larval development and give some indications on weather the larvae are lecitotrophic or planktotrophic. This study also focused on the final stages of larval development, particularly on the factors (chemical cues, substrate type, etc.) that contribute to the settlement and metamorphosis of the larvae into spat. Ultimately this study addresses some of the knowledge gaps necessary for the development of a potential new settlement assay, similar to the already-existing barnacle cyprid assay, using oyster larvae as a new test organism to evaluate the efficiency of antifouling technologies.

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MACRO-7

REARING TEMPERATURE AFFECTS THE ADHESIVE TENACITY OF BARNACLES GROWN ON NON-TOXIC MARINE COATINGS

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Barnacles are often the dominant hard foulers in marine waters and they attach to substrates by secreting a proteinaceous adhesive. Understanding the chemical composition and molecular interactions between the constituents of this multi-protein underwater adhesive is central to developing non-toxic solutions to control biofouling. Environmental variables such as temperature may influence the adhesive properties of barnacle cement, which could be of practical important as ships move among different oceanic environments. Previous experiments in our lab have shown an inverse relationship between critical removal stress (CRS) and the temperature at which barnacles were reared. Further investigation showed that removal temperature had no effect on the CRS of barnacles reared at different temperatures and reinforced the results previously found that barnacles reared at lower temperatures showed higher CRS values. Additionally, a regression analysis to investigate CRS as a function of size for each temperature showed no significant relationship between size and CRS. Therefore the observed effect seems to be a result of the development of barnacles as they are reared at different temperatures and not a result of barnacle cement changing viscosity when moved to different temperatures over short periods of time. The mechanism accounting for the temperature effect is still unknown and could be mediated through changes to the composition and amount of adhesive. To investigate the underlying mechanism responsible for the temperature effect we are using a combination of 2D-E proteomics, direct observations of the density of growth ring and cementing systems, and by conducting FTIR microscopy to confirm chemical differences between cement secreted at different temperatures. Our working hypothesis is that the temperature at which animals grow may affect the density of the cementing system and thus the composition and abundance of cement proteins.

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MACRO-8

ADHESIVES OF CYPRIDS OF THE BARNACLE BALANUS AMPHRITE AND THE TUBES AND ADHESIVES OF HYDROIDES ELEGANS AND HYDROIDES DIANTHUS

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Tubeworms and barnacles are severe marine biofoulers in many practical contexts. We report recent studies of the mechanical and compositional properties of several glue types, which could assist in the development of antifouling and fouling release strategies. In the first case, we examine the conformations of the settlement inducing protein complex of the cyprid of Balanus amphrite, using force spectroscopy. We find evidence of adhesion structures that correlate with the regions of hydrophobicity within the protein. In the second case, we report on protein components of the calcareous tubes of Hydroides dianthus that induce biomineralization of specific calcium carbonate morphologies. In the third case, we make a comparison with the mechanical properties of the adhesives and matrix in the primary tubes of Hydroides elegans, as well as its larval adhesives.

We gratefully acknowledge ONR for financial support.

MARINE CORROSION MATERIALS AND COATINGS
Session Chair: Jason Lee

MC-1

5XXX ALUMINUM SENSITIZATION AND THE NAVY

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In recent years, structural aluminum alloys have become more prevalent in marine applications, with the Navy's need to build lighter, faster ships. Aluminum alloys have been considered because of their light weight, abundance and cost. Of the structural aluminum alloys, the high Mg 5xxx alloys are the best candidates, combining high specific strength, corrosion resistance, and high as-welded strength.

Of particular concern is an aluminum alloy's susceptibility to sensitization. The operating environment for U.S. Navy ships is particularly harsh, with wide variations in temperature, as well as exposure to, and immersion in, seawater. It is known that the aluminum alloys that are currently in service are subject to sensitization, which increases the likelihood of stress corrosion cracking (SCC), exfoliation, or intergranular corrosion (IGC), often necessitating the replacement of compromised material. Aluminum becomes sensitized when beta phase (Al_3Mg_2) is precipitated at grain boundaries, which may lead to stress corrosion cracking. It has been widely established that 5xxx alloys having greater than 3.5% magnesium are susceptible to sensitization when exposed to temperatures greater than 50°C. The exact mechanisms and specific microstrucural conditions that represent sensitization, however, are not clearly understood.

This presentation will provide a historical survey of aluminum alloys in the Navy (including 6xxx), chronicling their use leading up to the discovery of sensitization related issues and beyond. The primary focus will be on the current state of marine grade 5xxx aluminum in the Navy, with respect to sensitization. The current state of the art for sensitization prediction, detection, mitigation, and repair will be discussed, including the development of sensitization resistant materials. Additionally, knowledge gaps and future needs will be addressed in an attempt to promote critical thinking on this subject.

MC-2

ALUMINUM ALLOY SUSCEPTIBILITY TO SULFIDE DERIVITIZATION

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Experiments have been designed to examine the susceptibility of aluminum alloys to sulfide derivitization. Alloys (1xxx -7xxx) will be exposed to environments with either biotic sulfide (e.g., anaerobic natural seawater) or supplemental inorganic sulfide addition. The effect of alloying elements (e.g., Cu and Mg) on sulfide derivitization will be highlighted. Corrosion morphology, weight loss and surface chemistry will be documented.

MC-3

SEAWATER FIELD TRIALS OF MATERIAL LOSS IN COPPER ALLOYS IN THE NORTH ATLANTIC

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A set of experiments was conducted to evaluate material loss rates and fouling resistance of several copper alloys in marine environment. Nine alloys and two reference materials (pure copper and mild steel) in a plate form were deployed for one-year in the Portsmouth Harbor, USA (North Atlantic Ocean). The testing program included three studies: material loss in tensioned and unloaded specimens, and biofouling resistance study. Three replicates were used for each of the studies, and one sample of each alloy was reserved for future comparison between mechanical properties of the virgin material and after a one-year deployment. For the material loss studies two deployment setups were used: one setup with specimens attached to a PVC frame (*Figure 1a*), and the other setup with tensioned specimens suspended through the water column from the buoy at the water surface and anchored to the seafloor (*Figure 1b*).

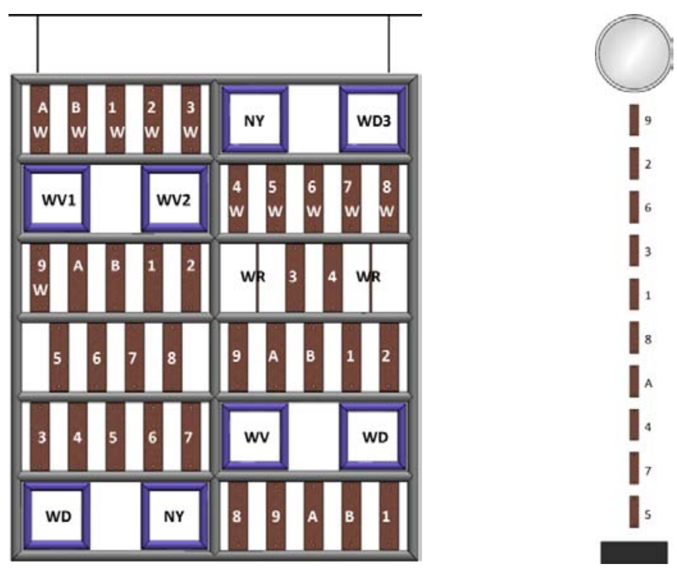
(a)  (b)

FIGURE 1. Deployment setup: (a) PVC frame with specimens for unloaded testing; (b) tensioned specimens suspended through the water column. Numbers and letters designate specimens of different materials

Material loss rates were quantified by measuring the difference in weight between the initial virgin specimens and the cleaned specimens after recovery. As part of the trials, material loss rates from continuous and interrupted deployments (three-month intervals) were also compared. It was found that the material loss from continuous deployment is approximately two times lower than that from the interrupted deployment. The effect of local corrosion damage was evaluated by analyzing micrographs and by quantifying the difference in stiffness and strength of alloys before and after exposure to seawater. The results of this study were compared to the accelerated lab testing in artificial seawater.

This work was supported by the International Copper Association, Ltd. The authors are grateful to Wieland-Werke (Germany) for supplying the test specimens and collaboration in research.

MC-4

BIOFOULING AND CORROSION OF SPRAY COATED CARBON STEEL

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The marine corrosion leads to serious economic loss on national scale and the counter measure was very hard to be established, since the number of factors is huge and complicated. One of the complicated factors in marine corrosion is biofouling. Biofouling should be mitigated as much as possible. In this study, we investigated the biofouling and corrosion deterioration for various spray coated carbon steels in a marine environment, being compared with the biofouling result in an usual clean water environment. The specimens used for this investigation are aluminium, zinc, aluminium-zinc, chromiumnickel, tin spray coating which were formed on carbon steel (JIS SS400). All of those specimens were immersed into sea water located at Marina Kawage (854-3, Chisato, Tsu, Mie Prefecture) in Ise Bay for several weeks. The depth of the specimen was two meter from sea water surface and the distance was always kept constant, since they were suspended from the floating pier. The biofouling behavior for each specimen was investigated by low vacuum SEM (Hitachi Miniscope TM1000), X-ray diffraction analysis (XRD), X-ray fluorescent analysis, a micro-video scope. Iron as substrate tended to attract bacterial the most due to the strongest affinity between trivalent iron ion and bacteria. Unlike the bulk material themselves, it is the most important for the coated specimens to inhibit the dissolution of iron for the viewpoint of corrosion protection and antibiofouling capability. Both have a close relation. And the inhibition capability against corrosion and biofouling depended on the coating materials due to our proposed model.

MC-5

SOME NEW EVALUATION METHODS FOR BIOFOULING ON METALLIC MATERIAS ON LABORATORY SCALE AND THEIR RESULTS

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It is well known that biofouling brings about various industrial problems. Even if it might be restricted to marine environments, the attachement of organism on boats and ships to reduce their driving power, microbiologically influenced corrosion, function deterioration of cooling system and so on can be mentioned immediately. From the background, the mitigation of biofouling has a significant industrial meaning. The countermeasure is required urgently. However, the correct and appropriate evaluation for biofouling in actual and simulated environment should be carried out properly. Since biofouling involves various complicated factors, it is usually very difficult for us to establish the appropriate evaluation process. In this study, we developed the two kinds of new processes to evaluate biofouling and the following deterioration of metals.

One of them is a circulation system. A special column was attached to the circuit and sea water was filled with the water circuit. The sea water was circulated in the circuit by a pump at a certain speed. Varois specimens were put in the inner part of the column within the circuit and after the series of experiment, circulation was stopped and the specimens were taken out. All of the specimens were served to various instrumental analysis later. The results were compared to those in clean water.

The other method was to use the filtration. Sea water in Ise Bay was filtrated by filter papers which had different pore sizes, respectively. Then the filers were immersed in artificial sea water (ASW), separately. Specimens (a stainless steel and carbon one) were put on the filters in ASW in a couple of days and weeks. Then the biofilm formation and corrosion behavior were observed and compared each other.

All of the results were examined from the viewpoint of reproducible and effective simulation for biofilm formation and the possibility for new labo-scale experiments about biofilm formation in sea water and cooling systems was discussed.

MC-6

ANTI-CORROSIVE METAL COATING USING SOL-GEL DERIVED LAYERED ORGANIC-INORGANIC HYBRID

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Many efforts have been made to protect metals from corrosion in a wide variety of marine applications. In this study, we report a novel strategy to prevent the corrosion of metals based on the use of a sol-gel derived layered organic-inorganic hybrid (LH) film. Our LH films, composed of alternating alkyl chain and siloxane layers with nanometer-scale interval distance between the layers, are derived from the co-hydrolysis and co-condensation of an alkylsilane and an alkoxysilane. Such hybrid materials offer several attractive advantages. First the resulting coatings are highly transparent and curable at room temperature without the need for any special treatments or equipment. Second the surface of the coatings shows excellent dynamic hydrophobicity due to the low surface energy and flexibility of the alkyl chains. Third the layered nanostructures are capable of carrying corrosion inhibitors (CIs) between the layers, and are expected to improve anti-corrosion performance and show self-healing properties when the samples are subjected to harsh environments. For example, our copper samples coated with CI-loaded (5-methyl-1*H*-benzotriazole) LH film showed not only excellent water repellency but also corrosion resistance against salt spray test more than 2000 h, while the regular LH film-coated sample was corroded within 100 h. Furthermore, after submerging the CI-loaded samples in a 5 wt% NaCl aqueous solution for 2 h, no corrosion was observed even in the deliberately cut region (self-healed), while regular LH film-coated samples allowed corrosion to progress around the cut regions. We believe that the 5-methyl-1*H*-benzotriazole between the layers is gradually released and forms stable CI-Cu complexes on the newly exposed metal surface. Therefore, thanks to the relative contributions of the hydrophobic and self-healing properties, long-term corrosion resistant coatings have been successfully prepared.

MC-7

BIOFOULING ON SPRAY COATED CONCRETES IN MARINE ENVIRONMENT

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The concrete is a sort of composite materials composed of cement pastes and aggregates. Usually, it is the portland cement (23.1%SiO2-5.0%Al2O3-3.0%Fe2O3-63.8%CaO-1.6%MgO-2.0%SO3-0.4%NaO-0.5%K2O). Being mixed with water, it is hardened and developed the gell structure including various pores and crevices to form a cement. From the viewpoint, the concrete can be considered as complex oxide. In marine environment, the concrete can be used for various purposes, not only as coast levee, but also as fish reef, seeweed bed etc. In marine environments, biofouling generally occurs on solid matters. Since it occurs also on concretes applied in marine environments, the control of biofouling is very important for the application purposes mentioned above. We have investigated the correlation between metallic materials and biofouling behaviors and found that the latter depends on the kinds of metallic materials. In this investigation, some metals, Fe, Cu, Ni, 50Ni-50Cr, 80Ni-20Cr, Zn, Ti, Sn, were spray coated on concrete surfaces and immersed in a marine environment under certain conditions. Then they were raised out from sea and the surface was observed by a low Scanning Electron Microscope, a video scope, a X-ray analysis, a fluorescent X-ray spectroscopy etc. And we discussed how metallic coating by spray coating on concretes affected the biofouling behavior. Iron coated concrete showed the remarkable biofouling and corrosion on the surface, while zinc and tin coated concrete materials showed few amount of biofouling. The evaluation and the mechanism of controlling by metallic elements for the biofouling of concrete were discussed.

MC-8

THE ROLE OF IRON IN SRB INFLUENCED CORROSION OF STEEL IN SEAWATER

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Sulfate-reducing bacteria (SRB) were found widely in marine environment, especially it was usually related the microbiologically influenced corrosion of steel and many other metal conductor materials in seawater. Although many studies related to SRB influenced corrosion in literatures, it is still an interesting and unclear subject why the SRB was often connected with the iron and steel materials? What's the role of iron in SRB corrosion process? It is important to understand the SRB corrosion mechanisms and develop SRB corrosion control technologies. In this study, from the microbial physiology and bioelectrochemical views, the interaction of a kind of SRB, *Desulfovibrio caledoniensis* cultured from the rust of steel, and conductor materials, especially steel, was further studied. It was found that there exists obvious microbiological difference when steel specimens contained in the SRB culture medium. Compared with the control tests, SRB showed about tenfold fast growth rate, higher maximum cell concentration and faster sulfate reduction rate. Hydrogen gas was also detected when steel immersed in SRB medium. We suggested iron may act as direct or indirect electron supplier to accelerate the growth of SRB, the corrosion rate also increase as a result. A serial of electrochemical tests were carried to study the cathodic electrochemical and corrosion behaviors influenced by SRB biofilms on steel and also on graphite electrodes. We founded that SRB biofilm could catalyze the hydrogen oxidation and reduction and decrease hydrogen evolution potential. Iron, with its negative redox potential, it is possible that SRB could acquire energy for growth through direct or indirect electric contact with steel substrate. SRB influenced cathodic corrosion process was further discussed.

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NEW APPROACHES TO ANTIFOULING TESTING
Session Chair: Maureen Callow

NA-1

3D TRACKING OF FOULING EVENTS

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Colonization of surfaces by biofouling organisms usually occurs through their dispersal stages [1,2]. This could be the planktonic stage of bacteria or, for larger organisms, spores or larvae. The selectivity during this dispersal stage is crucial for the long-term fate as it decides on secure anchoring and germination on the surface. Especially the three-dimensionality of the exploration process requires techniques, which are capable to record the full 3D scenes with video frequency. Different holographic [3] and stereoscopic approaches will be compared and discussed in terms of laboratory and field applicability. Tracking data will be compared to 2D data in literature [4-5]. In order to automatically extract position information we developed software algorithms capable to automatically extract the 3D positional information and to obtain the trajectories of multiple organisms in the field of view over time. From such 4D tracking data the sensitive response of spores and larvae as indication of their interaction with surfaces can quantitatively be studied. Statistical analysis of velocity and turning motions on surfaces with different chemical termination can be correlated with marked differences in the exploration patterns. By this we obtain a quantitative access to the interaction between single organisms and surfaces. The obtained results are discussed in the context of physicochemical surfaces properties (e.g. hydration, surface energy, and steric repulsion), time depend formation of conditioning layers, and the mechanism of surface selection.

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NA-2

A NOVEL DESIGN FOR THE *IN SITU* EXAMINATION OF SETTLEMENT BEHAVIOR

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Most settlement behavior studies are performed in the lab. This does not take into account variables associated with real world conditions. A flume has been developed to examine the settlement behavior of larvae *in situ*. It was designed to accommodate standard test panels (100x200 mm), observe behavior using HD video over a 75x50 mm test area and operate using native seawater and larvae at realistic flow rates, as well as being portable and robust. A novel S-shaped design was selected to optimize flow characteristics. Results from settlement experiments will be presented.

Acknowledgements:

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NA-3

A NOVEL MICROFLUIDIC APPROACH FOR THE ASSESSMENT OF ANTIFOULING TECHNOLOGIES

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Marine biofouling is the accumulation of organisms on underwater surfaces, causing increased hydrodynamic drag, resulting in higher fuel consumption and decreased speed and range. Biofilms constitute a major component of the overall biofouling, for example, fuel penalties from increased surface roughness due to biofilms (5 µm – 1 mm) are commonly reported (e.g. Schultz, 2007). Recent commercial antifouling technologies have managed to significantly reduce the effect of macrofoulers, however, marine biofilms are still an issue as they are known to remain attached even at high ship speeds (30-50 knots; Townsin and Anderson, 2009). The majority of reported biofilm studies involve the use of macro-scale reactors. However, more recently, there has been increased awareness that microfluidic systems provide several advantages, including inexpensive fabrication, highly parallel throughput, small size, and greater control over the microenvironment for cell culture (Meyer et al. 2011).

For this reason, we have developed and fabricated a novel lab-on-a-chip device for the investigation of the biofilm response to different hydrodynamic conditions. The microfluidic flow channel is designed using computational fluid dynamic simulations so as to have a pre-defined, homogeneous wall shear stress in the channels, ranging from 0.07 to 4.5 Pa, which are relevant to in-service conditions on a ship hull. The applicability of this approach has been demonstrated using a selected natural product (juglone - 5-hydroxy-1,4-naphthalenedione), which has previously been shown to have antifouling efficacy in static bioassays, where it allowed the investigation of the simultaneous effect of wall-shear stress and the natural product on biofilm structure. The results allowed for the first time the direct observation of the natural product influence on newly attached marine biofilms and the evolution of the antifouling effect with time. Biofilm attachment behaviour appeared to be markedly different in the presence of the natural product, illustrated by limited cluster and extracellular polymeric substance formation which suggests an interference of the bacterial attachment mechanisms.

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NA-4

A NOVEL GEOMETRY FOR LABORATORY-BASED LARVAL SETTLEMENT ASSAYS

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Laboratory bioassays are widely used in the research and development of novel antifouling coatings since they provide rapid means to screen surfaces. Supported by scientific knowledge of the biology of these model organisms, bioassays have added value in providing important clues on mechanism of action.[1] The cyprid settlement assay using larvae of the cosmopolitan barnacle *Amphibalanus amphitrite* is widely used to evaluate the affinity of barnacle settlement on materials with specific surface properties. However, existing assays have been conducted using widely differing assay systems with markedly different geometries, many of which are not optimal. In general, low or negligible settlement is observed when cyprids are placed in small water droplets on a flat substrate compared to the standard methods using petri dishes and 24-well plates. This effect is attributed to a larger seawater volume, which allows the cyprids' movement and exploration.[2] However, not all surface modifications are easily implemented on substrates in a 3D geometry, such as tubes and dishes, and this makes it difficult to study such material surface properties in larval settlement assays.

We will present a novel settlement assay system with geometry that can be applied to planar surfaces, including substrates with texture modulation and patterning. As this geometry does not require a confining material, it avoids the resulting choice assay artifact. This assay system also mitigates loss of (hydrophobic) cyprids trapped at the air/water interface. Cyprid confinement, exploration behavior and the development of settlement patterns were tracked using optical microscopy. Results were compared against experiments where cyprids were settled on flat planar surfaces in a droplet and standard assay in the wells of 24-well cell culture plates. This novel assay system expands our ability to screen a wider range of surface properties, with minimal change in methods currently used for surface fabrication in materials science laboratories.

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NA-5

BEHAVIOURAL ANALYSIS OF BARNACLE LARVAE

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Measuring behaviour is an invariably complex task, but one that has potential to provide limitless practical information for model organisms used in antifouling studies. In the marine environment planktonic organisms, such as the fouling larvae of sessile invertebrates, often have pelagic stages during which they inhabit the water column and benthic stages where they explore potential surfaces for settlement. Understanding the behavioural response of larvae to surface bound cues and/or inhibitory surface characteristics may therefore provide valuable information for the design of antifouling materials. This approach may be particularly useful in situations that require discrimination between numerous surfaces, all of which resist larval settlement in short-term laboratory assays. In this presentation, barnacle cypris larvae are used as a model organism and their behaviour in the laboratory is analysed in three ways. First, macroscopic behaviour is observed in two dimensions – what can be learnt from viewing the movement of larvae on test surfaces and what are the limitations of this approach? Second, what extra information may be yielded by expanding this method to three dimensions? A novel method will be presented for high-throughput 3D video analysis of the behaviour of barnacle larvae using Simi Motion 3D software and a custom-designed observation system. Third, how may observation of micro-scale behaviours during surface exploration aid our understanding of the adhesion mechanisms of these organisms and potential ways to impede their settlement? High-magnification light microscopy and high-speed photography are applied to visualising and elucidating the temporary adhesion mechanism of barnacle cyprids. It is proposed that, as antifouling technologies improve and discrimination between them becomes more challenging with our current suite of standard assays, it may be necessary to develop novel assays to answer important research questions. Multi-scale behavioural analysis could be one such method.

NA-6

DEVELOPMENT OF ACCELERATED TEST METHODS FOR CHEMICALLY-ACTIVE ANTIFOULING COATINGS

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One of the main objectives of this study is to develop a methodology able to predict long term antifouling (AF) paint performances. The challenge is to succeed in accelerating the ageing of AF coatings with a mechanism close to the natural one in less than one year. Cycles of static/dynamic exposure reported in ASTM D4939–89 (2007) were used. Before performing cycles, several artificial static phases were investigated. The most influent (sea)water parameters were determined through five subsequent protocols performed on two chemically-active AF coatings, i.e. a Self-Polishing Copolymer (SPC) and Controlled Depletion Polymer (CDP)-based coatings. Binder hydrolysis and diffusion phenomena are key mechanisms governing the AF performances of SPC and CDP coatings. Salinity of the water may influence the erosion of the coating and interfere with the dissolution of soluble compounds. Increasing the water temperature may increase the kinetics of water diffusion, the dissolution rate of bioactive compounds and the hydrolysis rate of the binder. The addition of an organic solvent in the electrolyte may swell the binder and thus improve the diffusion of water and dissolved compounds through the coating.

A dynamic step was performed using a rotating drum immersed in natural seawater; a new apparatus was purchased by the MAPIEM laboratory and is in service since April 2011. Regular thickness measurements were performed to assess the SPC and CDP erosion rate at 20 knots. AF performances were also assessed on aged and non aged coatings by performing monthly inspections to evaluate the degree of macrofouling and mechanical failures. In addition, coatings were mainly characterised by SEM-EDX analysis to assess compositional changes within their cross-section.

In this communication the relationships between artificial and natural ageing tests will be presented and discussed

NA-7

NEW SELECTION CRITERIA FOR BIOCIDES IN ANTI-FUOLING COATINGS

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We have investigated the binding affinity of some commonly used biocides in anti-fouling paint to “paint surfaces” in water environement, with the use of optical surface sensitive method like ellipsometry and surface plasmon resonance. Methods. Layers of abietic acid (“rosin”, a common binder in anti-fouling paints) were spin-coated on solid surfaces. Binding of the biocide, dissolved in water, to the rosin layers were then determined. Results. High binding affinity to the rosin layers was found for ivermectin, medium binding affinity for econea and medetomidine but no other biocides had any detectable affinity for the rosin layers.

Table. Binding of some biocides to rosin layers in water environment

Biocide	Binding to rosin/water (this study)	Leaking rate (ug/cm2/day)(Literature)	Concentration in paint (%)(Literature)
Ivermectin	High	0.003 – 0.00003	0.1
Medetomidine	Medium	0.05 – 0.005	0.1
Econea	Medium/Trace	> 1	> 1
Sea-Nine	Low	> 1	> 1
Tetrabromoheptanone	Low	> 1	> 1
Tolyfluanid	Low	> 1	> 1
Spinosad	Low	n.d.	> 1
Copper	Low	10-30	10-70

Conclusion. There is an excellent correlation between the high binding affinity of biocide at rosin/water interface and optimal low leaking rate (literature) and as well as the optimal concentration of biocide in the coatings (literature). The reason for this correlation is unclear but the use of the described surface sensitive method in selection of Biocides for anti-fouling purpose may lead to the discovery of new biocides/binder system with low leaking rates of biocides from the paint formulations

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NA-8

AN ASSESSMENT OF *ELMINIUS MODESTUS* (DARWIN) A MEMBRANOUS-BASED BARNACLE AS A MODEL SPECIES FOR EVALUATING FOULING-RELEASE COATINGS

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Fouling-release (FR) coatings based on polysiloxane and fluoropolymer technologies are environmentally benign alternatives to biocidal antifouling paints. A standardised protocol for determining the efficacy of the coating is to measure the force required to remove a fouling organism from the coating, normalised by contact area – the critical removal stress (CRS) or adhesion strength. *Balanus amphitrite* (*Amphibalanus amphitrite*), a calcareous-based barnacle, is a model species for fouling studies. This study aims to evaluate the potential of *Elminius modestus* (*Austrominius modestus*), a membranous-based barnacle, as a model species for use in standardised testing on FR coatings. Cyprid settlement and growth rate of laboratory-reared barnacles of both species was evaluated on Silastic-T2 and Sylgard 184 silicone test coatings. The CRS of *E. modestus* and *B. amphitrite*, to 7 coatings (5 polysiloxanes and 2 fluoropolymers) plus Silastic-T2 and Sylgard 184 were measured. The bulk properties of the polysiloxanes and fluoropolymers were modified by changing the polymer chain length and cross linker density, providing a modulus range from 0.2 to 19MPa, as determined by DMA. After 48h percentage settlement showed no significant difference between the two species, thus providing an adequate quantity of individuals to test. *E. modestus* took 28 weeks to reach 4.4mm and 4.7mm in diameter of the basis for Silastic-T2 and Sylgard 184, respectively. This is a slower growth rate than published reports for *B. amphitrite.* CRS increased with modulus for both species. There was no significant difference in removal force for lower modulus polysiloxanes. For the higher modulus fluoropolymers, shell failure occurred before adhesive failure for *B. amphitrite.* The CRS of *E. modestus* was less than that for *B. amphitrite* for Silastic-T2 and Sylgard 184. A hard basis (calcareous) – coating interface could behave differently than a soft (membrane) – coating interface. *E. modestus* provides a valuable comparative for studies in adult adhesion.

Poster Session 2

TP-02

BIOFOULING ON SALMON CAGE NETS IN NORWAY – A ONE YEAR STUDY

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The accumulation of biofouling on the cage nets is one of the main problems in Norwegian salmon aquaculture. By reducing the water flow through the net and subsequently the oxygen supply and waste removal, the settlement of organisms on the nylon nets may threaten the health of cultured fish. A 1-year field study at a commercial salmon farm in Mid Norway was conducted to determine the effects of immersion time (1, 3, 6 and 12 months), mesh size (13 and 25 mm half mesh) and cage (3 different cages) on the fouling biomass and community composition on net panels. For the net panels immersed for 1 month at a time, a total of 69 species and 7 fouling groups were identified, with species richness ranging between 1 (January) and 55 (September) species (or groups) per month. Interactions of sampling time with mesh size and sampling time with cage affected the biomass only in the first half of the year. In the second half only sampling time had an effect. The most frequently occurring macrofouling organisms were amphipods (*Corophium* sp. and *Jassa* sp.), blue mussels (*Mytilus edulis)* and hydroids (*Ectopleura larynx*). A similarity analysis of species composition demonstrated an influence of sampling time only, while mesh size and cage did not influence the community composition. The results of this study contribute to the optimisation of current antifouling treatments of Norwegian aquaculture nets.

TP-02

MARINE FOULING ON AQUACULTURE FACILITIES IN THE NORTHERN SOUTH CHINA SEA

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Marine fouling is a big problem around the world and can cause serious economic losses in aquiculture also. In the northern South China Sea, the settlement of fouling organisms occurs throughout the year, and the species composition and fouling biomass may change greatly with the season. Moreover, the environment factors, including salinity and location, also play an important role in the structure of the fouling community.

On aquaculture installations, fouling organisms are composed of acorn barnacles, bivalves, bryozoans, ascidians, hydroids, polychaetes, sponges and algae. Of them, the major fouling group is acorn barnacles, and the dominant species is *Balanus reticulatus*, following *B. cirratus* & *B. trigonus*. Regarding bivalves, the major fouling species are *Perna viridis* and *Brachidontes variabilis*. Other common species are *Dendostrea crenulifera*, *Saccostrea cucullata* and *Musculus nanus*.

The dense fouling normally occurs in summer and autumn, particularly acorn barnacles and bivalves. Bryozoans mainly occur from December to May, and the dominant species is *Bugula neritina*. The polychaetes such as *Hydroides elegans* occur in the fouling community at any time also, particularly from May to September. Ascidians mainly occur from April to October, and macro-algae in winter and spring. The settlement of sponges mainly occurs in summer and autumn. Although hydroids can be found in the fouling community at any time, most species occur in spring.

For fouling control effectively, further work should be focused on the revelation of dominant fouling species characteristics, elucidation of their adhesion mechanisms and development of novel and non-toxic antifouling technology.

TP-03

EFFECT OF CAGE DEFORMATION AND BIOFOULING ON THE WELFARE OF FARMED ATLANTIC SALMON *SALMO SALAR*

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In recent years there has been increasing interest from the public, government and industry, in the welfare of intensively farmed fish, and stocking density has been identified as an area of particular concern. Stocking density within marine cages is regulated by a set of stringent guidelines which rely upon the premise of a cage being a static structure of predetermined volume. However, cages are deployed in a dynamic environment and their volumes are defined not by the dimensions of their components but by the nature of the conditions within which they are exposed.

In this study, cage net deformation was measured on sea-cages of two full-scale commercial Atlantic salmon farms in Scotland for a total of 12 months; changes in cage volume were related to fluctuations in incoming current velocity, net aperture occlusion by biofouling and cage structure. Ambient current measurements were recorded using Doppler current meters, and cage volume reductions were deduced from pressure sensors placed at specific points on the cage net. To estimate the potential effect of these deformation events on fish welfare, water quality parameters critical to fish health (i.e. dissolved oxygen, salinity and temperature), were recorded and related to both observations of fish behaviour and production data.

Increase in cage net occlusion was found to adversely affect cage volume at similar ambient current flow by increasing the drag coefficient of the net. However, while in some instances the observed reductions in cage volume could be dramatic (e.g. over 40% during periods of current speeds exceeding $0.07 m.s^{-1}$ with high net occlusion), these events rarely lasted for more than 5% of the tidal cycle, and cage net deformation only represented a 16% loss in total volume for over 85% of the recorded time scale.

Increased net aperture occlusion by fouling communities also reduced water flow though the cages and was the main factor influencing within-cage water quality. Large reductions in current flow were accompanied by a reduction in levels of dissolved oxygen in 'down-current' cages. While wide variations were seen both within and between sites, our results highlight the relation between net deformation and cage volume, and have significant implications for on-site management of fish health and welfare.

TP-04

MICROSTUCTURE OF BASE-PLATE OF BARNACLE ON DIFFERENT SUBSTRATE

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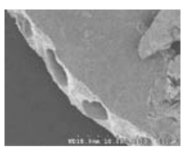
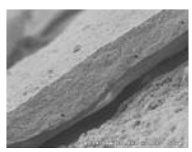
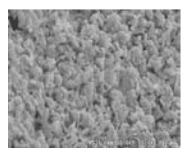
Barnacles firmly attach their base-plates on various substrates through barnacle cement, and which causes serious marine fouling problem and damage in many maritime affairs (1). The base-plate is one of the key parts in barnacle adhesive process, but the structure and functions of base-plate are less explored and far from our comprehension.

In this experiment we studied the microstructure of base-plate of *Balanus reticulates* on three different substrates by scanning electron microscope (SEM) to give an insight in the structural properties of barnacle base-plate. It has been found that the significant impact of substrate on the microstructure of barnacle base-plate including both inner and outer surface. The results also indicate that the microstructure shows different appearance in different regions of the of base-plate (F.1, F.2, F.3), and it is suggested that such characteristics may be related to some of the important functions of base-plate, such as the hollow structure of the basis periphery (F.1) can be the complementary part to fit to the parietal shell wall-plates (2), and the channels with different aperture sizes (F.2) in the base-plate can be acted as ways for transportation, distribution and steering of barnacle cement into the gap between base-plate and substrate (2). Obviously, the barnacle base-plate displays a complicated microstructure and high structure plasticity. Such characteristics of base-plate structure can help barnacles to effectively adapt to the harsh marine environment. This study can better our understanding of the mechanism of barnacle adhesion and provide useful knowledge for antifouling strategies.


F.1


F.2


F.3

F.1 The hollow structure in the basis periphery of the barnacle on a brick

F.2 The inner surface and cross section of the barnacle base-plate on the bamboo material, showing the apertures

F.3 The rough outer surface of the barnacle base-plate on a piece of fabric material

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TP-05

DO BARNACLES HAVE COLOUR PREFERENCES IN THEIR CHOICE OF SETTLEMENT SUBSTRATUM?

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Settlement into biofouling communities is influenced by factors such as larval supply, light, presence of conspecifics and substratum characteristics. Colour of the substratum as a settlement cue so far has been investigated from a human perspective, neglecting the visual capabilities of the organism. The aim of this study was firstly to determine if barnacle cyprids have a preference for different coloured surfaces in settlement. This would not necessarily mean that the larva uses colour vision; instead it may be relying on brightness differences. Therefore the second aim was to determine if the cyprid's settlement behaviour is influenced by the true colour of the substratum – as perceived by the cyprid visual system - or by brightness levels. Colour preferences of cyprids of the barnacle *Amphibalanus amphitrite* in settlement substratum were tested in the lab. Tested colours were specifically selected based on presumptive spectral sensitivity of the visual receptors of the barnacle larval nauplius eye. The lab experiment was conducted as a 2-choice experiment. Statistical analysis was done using Exact test and Regression Analysis. These results were compared to a field experiment with *Semibalanus balanoides* using the same colours. Cyprids preferred to settle on the tested colours black, violet, red and green over dark blue and white. The colour of the substratum has an effect on the choice of settlement substratum for the two particular barnacle species tested. Brightness of the substratum is an important determinant for settlement, though true colour perception cannot be dismissed as results of the field experiment indicate.

TP-06

BRICKS AND MORTAR MODEL OF BARNACLE ADHESION

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Multidisciplinary approaches and modern technology provide stimulating insights on glue curing. Barnacles adhere by creating a multifunctional interface composed of proteins, creating a "bricks and mortar" composite structure piece by piece. To understand the biophysical processes involved, we use multidisciplinary approaches to study barnacle adhesion. Approaches include physical measures, bacteriology, behavior, physiology, biochemistry, microscopy, spectroscopy, tomography, tandem mass spectrometry, molecular biology and proteomics. We combine experimental observations with theory grounded in evolution and earlier work. Use of analytical tools is facilitated by thorough knowledge of culture and manipulation of all life stages of *Amphibalanus (= Balanus) amphitrite.* Here we synthesize data from recent publications to support the hypothesis that barnacle glue curing is similar to blood clotting. Then we provide new data that will be published in detail by a subset of members of our group and add ideas that culminate in the Bricks and Mortar model of barnacle glue curing. Barnacle larval settlement, bacteriology and biochemical data show glue contains large amounts of small peptides. Many peptides are generated by trypsin-like serine proteases and end in lysine or arginine. The new peptide components to the story help explain how the glue might cure. Because they have little secondary structure, these peptides can associate with binding domains on the substrate and interface with the larger, well-described structural proteins known in barnacle glue. We hypothesize the process prepares the glue for calcification.

TP-07

BETWEEN A ROCK AND A HARD PLACE: STRUCTURAL COMPLEXITY OF THE BARNACLE ADHESIVE INTERFACE

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We are tackling a common marine fouler, the barnacle, with *in situ* approaches and tools that allow us to probe the bioadhesive interface as it develops. From these studies, we have learned that barnacles secrete and cure their adhesive through a multistage process. Further, the composition and structure clearly varies across the interface, in both lateral and perpendicular orientations relative to the barnacle/substrate interface. In this poster, we explore the compositional, chemical and mechanical differences found in the barnacle interface. We examine this interface "from the outside-in", and "from the inside-out" using a suite of high resolution analytical tools including optical and confocal microscopy, UV-visible microscopy, small spot x-ray photoelectron spectroscopy (XPS), scanning electron microscopy (SEM), energy dispersive x-ray spectroscopy (EDS), Fourier Transform Infrared Spectroscopy (FTIR), and nanoindentation. We combine these approaches to reveal local chemical, structural, and mechanical differences and correlate these with biophysical structures in the interfacial region that develop as the barnacle expands its base plate.

TP-08

INFLUENCE OF SURFACE CHARGE AND SURFACE ENERGY ON THE SETTLEMENT BEHAVIOUR AND 'FOOTPRINT' MORPHOLOGY OF BALANUS AMPHITRITE AND B. IMPROVISUS CYPRIDS

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Barnacle fouling is a major issue for maritime operations and there is considerable interest in methods to mitigate their impact. To this end, improved understanding of the factors that influence settlement and adhesion success are important. Among the many species used to examine the interfacial processes involved in colonisation of immersed surfaces, *Balanus amphitrite* has become a model species. Here, self-assembled monolayers (SAMs) of CH_3-, OH-, COOH-, $N(CH_3)3^+$- terminated thiols were deposited onto gold-coated polystyrene to investigate the cyprids' responses to surface charge and surface energy. Settlement of *B. amphitrite* (1) was compared to that of *B. improvisus*, which has been reported to differ in its settlement behaviour with respect to surface wettability. Contrary to expectations, neither species showed a preference for a particular surface energy. Surface charge, however, had a significant effect on settlement of *B. amphitrite*, with higher settlement on negatively-charged surfaces. *B. improvisus* showed a preference for hydrophobic CH_3 in agreement with previous work focused on surface wettability (polystyrene vs. glass (2)) and a variable response to surface charge. Further insight into these surface preferences was sought by investigating the deposition of cyprid temporary adhesive (footprints) on the SAMs, using imaging ellipsometry. Footprints were thicker on positively charged surfaces, which attracted the lowest settlement for both species.

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TP-09

IDENTIFICATION OF SMALL MOLECULE COMPONENTS OF BARNACLE GLUE

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Barnacles secrete glue which adheres to any solid surface under water. This inspires us to develop novel and practical bio glue for dental and surgical applications based on barnacle glue components. Though proteins in barnacle glue have been recognized as the major components and detailed studies have been done by several research groups, comprehensive studies related to the small molecule components in barnacle glue are lacking. Here, we present data from experiments targeted at identifying the small molecule components of barnacle glue. The small molecule components were isolated from freshly secreted barnacle glue by standard biochemical techniques. Mass spectrometry identified the presence of peptides, a heparin-like molecule, lipids and antibacterial compounds. These initial observations were followed by infrared spectroscopy and biochemical assays. Preliminary results showed that the small peptides (MW less than 3 kDa) could comprise more than 20% of freshly taken glue, indicating they may play an important role in curing. The percentage of small peptides also varied from experiment to experiment and this result was speculated to be related to molting or other biological cycles. Lipids with various molecular weights (e.g. various structures) were found in mass spectra and their structure will be further determined. Heparin and antibacterial compounds were also examined in detail. The authors gratefully acknowledge support from the ONR Coatings/Biofouling program and the McGowan Institute for Regenerative Medicine

TP-10

COMPARATIVE TOXICITY EVALUATION OF ANTIFOULING AGENTS ON THE OVARIAN DEVELOPMENT OF BROWN MUSSEL PERNA INDICA

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Most of the conventional/chemical antifoulants are reproductive toxicant to various marine organisms especially to molluscans and bivalves. To overcome this issue, natural products/compounds are being assessed for antifouling activity with no or ignorable toxicity. In this view, the present attempt has been made to compare the toxic effect of antifouling activity bearing methanolic extract of seagrass *Syringodium isoetifolium* (25µg/ml) over the conventional antifoulant, tributyltin (TBT; 100nl/l) on the ovarian development of brown mussel *Perna indica*. Appropriate controls were also maintained. The TBT exposed mussels exhibited decrease in Gonado Somatic Index (GSI) and Hepato Somatic Index (HSI) compared to *S. isoetifolium* extract treated group and control. Interestingly, the control and *S. isoetifolium* extract treated mussels showed normal cellular architecture and no damages in gills, digestive gland, muscle and ovary. However, histological results revealed that TBT caused increase in interfilamentar space and fusion of the filaments in gills, disruption in the digestive tubules and reduction in basement membrane thickness in digestive gland. Besides in adductor muscle, TBT caused muscular degeneration, necrotic muscle layer and in ovary; it inflicted fusion of the developing oocytes. Moreover, drastic decrease in total protein, lipid and glycogen content was noticed against the progression in free amino acid and glucose content in TBT treated mussels compare to other groups. TBT had significantly retarded the ovarian development and substantially reduced other biochemical constituents which led to the impairment of oogenesis as against the null effects noticed in *S. isoetifolium* extract treatment.

TP- 11

VISCOSLAINE ANALOGS: SYNTHESIS AND ANTIFOULING PROPERTIES

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3-Alkylpyridinium salts (3-APAs) are common secondary metabolites isolated from marine Haplosclerid sponges. Two major groups of 3-APAs metabolites with fully saturated alkyl chains have been identified to date: linear and macrocyclic structures. Each group can be further divided according to the number of pyridinium rings in the molecule. Most of the linear compounds are constituted by only one pyridine unit, excepted viscosaline (**1**), a potent antibacterial derivative, and the halitoxins (**2**). The group of cyclic 3-APAs is dominated by dimeric compounds with two pyridine moieties, e.g. cyclostelletamine A (**3**) and haliclamine A (**4**).

Despite their rather simple structure, 3-alkylpyridinium salts display a broad variety of biological properties, such as: cytotoxic, antibacterial, antitumoral and antifouling activities. However, the entire potential of these molecules is still understudied due to the difficulties to prepare them and the lack of SAR studies.

We have synthesized several viscosaline analogs and the derivatives were evaluated for their antifouling properties. Our results suggest that viscosaline analogs could be a promising class of substances with antifouling activity

TP-12

PRELIMINARY RESULTS OF THREE NATURAL EXTRACTS FOR ANTIFOULING PAINTS

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Natural extracts with potential activity were used in antifouling soluble matrix paints: an ethanolic extract of the marine sponge *Clathria microxa* and two polar subextracts of the plants: *Hyalis argentea* and *Nardophyllum bryoides*. The plant extracts were selected on the basis of their chemical diversity while the sponge extract was chosen for the empirical observation of the absence of fouling on the organism´s surface and the presence of complex secondary metabolites. For the preparation of the soluble matrix antifouling paint, colophony (resin) and oleic acid (plasticizer) were dissolved in a xylene/white spirit (1:1 % by weight) mixture using a high-speed disperser. Then, a laboratory scale ball mill was loaded with the vehicle followed by zinc oxide and calcium carbonate which were dispersed for 24 h. The extracts (0.8% v/v) were incorporated to the paint and then, dispersed during 1 h. Sandblasted acrylic tiles (4 cm×12 cm) were used for field trials. Paints were applied by brush on tiles previously degreased with toluene. Three coats of paint were applied and allowed to dry for 24 h between each application; resulting in a final dry thickness of 150±5_m. Panels were hung in a marina in Mar del Plata harbor to a depth of 50 cm below the water line. In addition, two controls were used; one of them was unpainted acrylic tiles and the second one, tiles treated with paint without extracts. After one month exposure in the sea, natural products-based paints were effective in inhibiting settlement. A great decrease in micro and macro-fouling density and diversity was observed in relation to controls ($p<0.05$ through ANOVA and contrast LSD test, using STATISTICS program). Preliminary results showed that *C. microxa, H. argentea* and *N. bryoides* extracts can be employed as bioactive agents for antifouling paints.

TP-13

ANTIFOULING POTENTIAL OF MARINE SPONGES FROM BAY OF LA PAZ, MEXICO

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All surface submerged in the sea is initially colonized by bacteria, forming biofilm, being the base for the colonization of macroalgae and invertebrates, causing the deterioration of the man-made structures, generating huge economic implications. The marine sponges have chemical defense mechanism for growth inhibition of epiphytic the micro and macro organisms. The objective of this study was evaluated the antifouling potential of six sponges (*Haliclona caerulea*, *H. turquiosa*, *Ircinia* sp., *Callyspongia* sp. *C. californica* and *Mycale* sp.) present in bay of La Paz, Mexico. The sponge specimens were collected in two localities (La Bruja and Agua de Yepiz) in cold and warm seasons, The specimens were repeatedly extracted with acetone/MeOH (1:1) and partitioned in ether and n-buthanol extracts. The antifouling activity of the extracts was tested against five bacteria and eight marine microalgae. The results indicated that the sponges collected in cold season in both localities have the best antifouling activity. The bioassay against bacteria strains showed that ether and n-buthanol extracts of *Haliclona caerulea* and *Mycale* sp. of La Bruja locality have a MIC of 0.1 $\mu g\ ml^{-1}$ against three of the five bacterial strains. In the case of Agua de Yepiz the ether extract of *H. caerulea* was very active against *Vibrio aestuarianus* (MIC=0.1 $\mu g\ ml^{-1}$). The results against marine microalgae showed that n-buthanol extracts of *Mycale* sp and *H. caerulea* of La Bruja locality had the highest activity with a MIC=0.1 $\mu g\ ml^{-1}$ against *Cylindrotheca clostridium* and *Navicula jeffreyi* respectively. These results show that sponges presents at both locations show interesting antifouling activity, specifically the species *Mycale* sp. and *Haliclona caerulea*, with a potential resource to provide novel antifouling agents

TP-14

SCREENING OF NOVEL ANTIFOULING METABOLITES FROM MANGROVE ASSOCIATED STREPTOMYCES FRADIEA

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Colonization by fouling (micro and macro) organisms is a problem for all submerged structures in marine environments. Economic loss due to damage caused by fouling organisms is so stupendous throughout the world. Application of antifouling paints containing tributylin (TBT) and copper compounds is the most widely used primitive coatings to overcome this problem. However TBT is considered to be potentially toxic and proved to have adverse effects on non-target marine organisms. Therefore intensive research is now being carried out to find out alternative non-toxic eco friendly antifoulant to prevent the use of TBT based antifouling paints. In this context, the present work was carried out to evaluvate the non/less toxic antifouling metabolites from mangrove associated actinomycetes. For this, actinomycetes were isolated from mangrove sediments of Mankudi estuary, Kanyakumari coast, Tamilnadu, India and all the strains were individually subjected to cross - streak and agar well diffusion assay against biofilm bacterial strains. The most prominent result yielding actinomycete was identified by polyphasic characters and then mass cultured. Then the culture supernatant was extracted with ethylacetate and reduced under vaccum pressure. 250 μg of ethylacetae extract was tested for antimicrofouling activity against seventeen biofilm bacterial strains through disc diffusion method. Results inferred 100% growth inhibitory activity against all the tested biofilm bacteria with considerable Minimal Inhibitory and Minimum Bactericidal Concentrations (MIC & MBC) against the biofilm bacteria. Further, the antimacrofouling potential of crude extract was tested through molluscidal assay against *Patella vulgata* and inhibition of byssal thread formation was tested with brown mussel *Perna indica.* Also, the crude extract was tested for cytotoxicity against *Artemia*.

TP-15

ANTIBACTERIAL AND ANTIFOULING ACTIVITY OF SYMBIOTIC BACTERIA FROM APLYSINA GERARDOGREENI

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The application of natural antimicrobial compounds in prevention of marine biofouling has tremendous interest in the industry. Fouling organisms cause severe damage to the marine structures raised for commercial purposes. The key area in marine anti-foulant research, is finding compounds with activity to prevent formation of biofilms, the succession of flora they attract and also prevent bio-corrosion of ship hulls and submerged objects as a result of acid produced by anaerobic bacteria in marine biofilms. In the present study we report the potential of marine sponge bacteria associated to produce compounds with potential application as natural anti-foulants. A collection of 63 bacteria were isolated previously from *Aplysina gerardogreeni* sponge. The bacterial biomasses were extracted with hexane and ethyl acetate (60:40). Extracts were assayed for their antifouling activity against 16 marine and four terrestrial bacteria and five microalgae. The bioassay showed that 57 strains was active against any of the strains tested. The most sensitive strain was *Bacillus subtilis*, being inhibited by 69% of the extracts most with low MIC (0.01 µmg ml^{-1}). On the other hand, 37 extracts showed activity against any of the five strains of microalgae tested. The most sensitive was *Cylindrotheca closterium,* 29 extracts were able to inhibit their growth at low concentrations. Sequencing of partial 16 S ribosomal DNA analysis showed that the bacterial isolates from sponge *A. gerardogreeni* with more activity were related to *Bacillus* species. The results revealed that bacteria marine sponge associated are a potential source of novel antifouling agents.

TP-16

ANTIFOULING EVALUATION OF SYNTHETIC MERIDIANINS DERIVATIVES: ANTISETTLEMNT ACTIVITY AND TOXICITY AGAINST BARNACLE LARVAE

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Biological fouling of engineered materials is a significant problem for any immersed surfaces and especially for ships. Materials deterioration, losses in heat-transfer efficiency, and mechanical blockages of fluid transport systems can result from biological fouling activities. Among the great variety of marine-derived compounds, indole alkaloids have emerged as an important structural class based upon their high degree of biological activities (including antimicrobial, antiviral and antitumor properties). Biological evaluation of two naturally occurring 3-(2-aminopyrimidine)-indoles, meridianins C and G, together with thirty-one of their synthetic derivatives, has been completed. Larvae of *Balanus amphitrite* (nauplii and cypris) were used to monitor anti-settlement activity and toxicity.

TP-17

ANTIFOULING MOLECULE FROM THE BACTERIUM HALOMONAS MERIDIANA ASSOCIATED WITH A MARINE INVERTEBRATE

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Marine biofouling continues to be a large problem in shipping and other marine associated activities. The application of heavy metal-based antifoulants has created environmental problem to non-target organisms. After the ban of TBT, there is an urgent need to find out effective and environmentally friendly antifoulants. Natural product antifoulants (NAPs) have been proposed as one of the best possible alternative antifoulants. For the isolation of NAP, marine organisms are used as the sources (invertebrates and seaweeds). In the present study, bacteria associated with the surface of the marine invertebrates were screened for antifouling activity. Eight bacterial strains isolated from the surface of sealilly were tested for antibiofilm activity against *Galionella* sp., *Alteromonas* sp. and *Pseudomonas* sp. Strain SL7 showed good activity against target bacterium and it was selected for further studies and characterized partially using TLC and HPLC analysis. The 16S rRNA of the strain SL7 was sequenced and identified as *Halomonas meridiana.* Further antifouling studies on this isolated compound could provide leads for the formulation of new natural product antifouling agent.

TP-18

INFLUENCE OF EXTRACTANTS FROM THE CALLYSPONGIA SPONGE ON THE LARVAL SETTLEMENT OF ACORN BARNACLE BALANUS (=AMPHIBALANUS) RETICULATUS

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While marine fouling causes serious problems around the world, some marine organisms, such as sponges and corals, are rarely epiphytized because they can produce secondary metabolites against fouling.

Acorn barnacle *Balanus* (=*Amphibalanus*) *reticulatus* is an important dominant fouling species in tropical and subtropical seas, particularly in the South China Sea. Its cyprid is therefore appropriate test representative for the antifouling bioassay.

The sponges of genus *Callyspongia* are widely distributed in the South China Sea. Eighteen compounds were isolated and purified by silica column and Sephadex LH-20 from the sponge. The structural identification was conducted by the spectroscopic analysis (NMR and MS).

The compounds were dissolved in 1 ml methanol (or chloroform) and transferred to Petri dishes, respectively. The control groups, one with filtered seawater alone and the other with 1 ml solvent before addition of filtered seawater, were prepared also. Petri dishes were dried on a shaker prior to the addition of filtered seawater and larvae. The mean values of larval settlement rate were compared with the LSD test.

Of them, 4 compounds, i.e. Cyclo-(Pro-Ala), Cyclo-(Pro-3hdroxy-2-aminobutanoic acid), Cyclo-(Pro-Leu) and Cyclo-(Pro-2-N-hydroxyl-Ile) could effectively impede the larval settlement of barnacle *B. reticulatus*. The results will contribute to reveal the chemical antifouling mechanisms of marine organism, and also would be beneficial to the research and development of new non-toxic antifouling agent.

TP-19

MARINE NATURAL PRODUCT AS NOVEL ANTIFOULDING REAGENTS: STRUCTURE OPTIMIZATION AND BIOLOGICAL EVALUATION

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Antifouling paints containing organotins, copper, lead, mercury, or arsenic that were widely used to control fouling in the past are very effective, but also highly toxic and persistent in the marine environment [1-3]. They must eventually be replaced by new, effective, and environmentally benign antifouling compounds. One of the most ecologically relevant antifouling strategies is to develop products that are based on the natural chemical defenses of sessile marine organisms. [4] To date, more than 400 natural AF compounds have been found in the world, but none of them have been developed into commercial antifouling agents for two reasons, the yield of compounds are normally poor and structurally too complexity to synthesize. Therefore, instead of investing effort in the search for additional bioactive compounds, it may be wiser to optimize structure of antifouling compounds base on SAR. Herein, we optimized the structure of antifouling compounds using chemical synthesis base on structure-activities relationship study of antifouling marine natural products. A series of new butenolide derivatives with various amine side chains were synthesized and their anti-larval settlement activities on *Balanus amphitrite* and *Bugula neritina* were further evaluated. Of those screened, we identified the analogue with the Boc group at the amine of the side chain as the most potent anti-larval settlement compounds. The strong antifouling activity, relatively low toxicity, and simple structures of these compounds make them promising candidates for new antifouling additives.

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TP-20

MOLECULAR LEVEL STUDIES ON ANTI-BIOFOULING AND FOULING-RELEASE POLYMER SURFACES

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Anti-biofouling and fouling-release polymers are designed to minimize biofouling to occur on the surfaces. The surface structures of these materials and surface ordering of functional groups are therefore of great interest to synthetic chemists and analytical chemists to make polymers with improved performance. Sum Frequency Generation vibrational spectroscopy (SFG) is a powerful surface specific technique to study these materials by analyzing the interfacial structures and their orientations. Our research is focused on the interfaces between polymer coatings and air/water/biomolecule solutions and we are aiming to unveil the relationship between the interfacial chemical structure and the performance of these antifouling and fouling-release materials, aiding in directing the synthesis of such materials. We have successfully demonstrated the structure-performance relations for various polymer coating samples from our collaborators including biocide-doped polydimethylsiloxanes, zwitterionic polymers, and amphiphilic block copolymers. Surface vibrational spectra can be obtained using SFG from these materials, from which detailed surface structures can be deduced. We are also developing new experimental data-collection and new data analysis methods to better interpret the complicated SFG spectra (e.g., polarization mapping and two-dimensional correlation spectroscopy). Polarization mapping is designed to de-convolute overlapping peaks in order to accurately fit the SFG spectra. And 2D correlation method is incorporated to identify origins of different peaks in the polymer structure. The applications of these methodologies lead to successful interpretations of complicated SFG spectra for different polymer materials, providing in-depth understanding on their molecular surface and interfacial structures. Such understanding is vital to synthesize new materials with better antifouling and fouling-release properties.

TP-21

STEREOSCOPIC METHODS FOR THREE-DIMENSIONAL TRACKING OF EXPLORATORY BEHAVIOR OF BARNACLE CYPRIDS

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An important step before colonization of surfaces by sessile marine biofouling organisms is surface exploration. During this step, the microorganisms evaluate different surface properties as well as different environmental factors in order to find an appropriate site for permanent settlement. This exploration is a fast process involving three-dimensional swimming motions and its quantitative analysis requires respectively the recording of temporally resolved three-dimensional swimming trajectories. Two dimensional tracking [1] has been proven to be suitable for identification of attractive and repelling surfaces, in terms of biofouling, and provided a new insight into selection strategies [2]. However, a clear separation between active (exploration, swimming) and passive (floating) phases of the swimming trajectories proves to be complicated. Another imaging method (Digital Inline Holographic Microscopy, DIHM) provides the needed features, but works optimally only with objects at size of around 20 μm [3], which is not sufficient in the case of cyprids (depending on the species, size varies in the range of 100 μm – 1000 μm). We present a transportable, submersible stereoscopic system which can be applied to record three dimensional video data and extract swimming trajectories of multiple, label-free objects. The hardware setup is presented and discussed. Introducing the mathematical basics of stereoscopy, we describe the resolution and perform empirical error analysis in order to obtain the error values of the system. First trajectories of barnacle cyprids on chemically different surfaces (PEG2000-OH, $C_{11}NMe_3^+Cl^-$, acid washed glass (AWG)) are extracted and evaluated in respect to known patterns (swimming velocity, swimming angle, etc.).

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TP-22

IN-SITU STUDY OF BARNACLE CYPRID AND JUVENILE BARNACLE CEMENT USING XRF MICROSCOPE AND MICRO-RAMAN SPECTROSCOPY

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Among the different biofouling species, barnacles are one specific thread as they are difficult to remove, able to damage foul release coatings and increase the drag force of ships.[1,2] Furthermore, barnacles are a good model system for research on permanent underwater adhesion strategies [2]. This study aims on the understanding and comparison of the spatial organization and the chemistry of the adhesive secreted by different cyprid larvae and juvenile barnacles for settlement. We apply both, synchrotron based X-ray microprobe fluorescence and Raman spectroscopy, for the in-situ investigation of the chemistry of barnacle cement. The results of these studies will provide information on chemical composition and morphological structure of both barnacle species at different life stages. The derived mechanistic understanding of the adhesive is supposed to lead to new, environmentally benign antifouling solutions aiming on the interference with curing of the adhesive and thus the attachment process.

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TP-23

ZWITTERIONIC BETAINES AS VERSATILE ULTRA LOW FOULING SURFACE COATINGS OR HYDROGELS FOR REAL-WORLD APPLICATIONS

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Undesirable attachment of both biomolecules (e.g., proteins) and microorganisms (e.g., cells, bacteria and algae) can severely deteriorate surface performance in both biomedical and marine environments. Due to the large variety of surfaces which are used in real-world applications, diverse approaches for forming coatings must be developed. The ability to achieve excellent resistance to biofouling as well as immobilize biological reactive agents has been enabled by dual-functional zwitterionic carboxybetaine polymers (pCB). First, highly non-fouling surface coatings to complex media were developed using a “graft-from” approach. This includes the use of surface polymerization methods (e.g., atom transfer radical polymerization and controlled photo polymerization) to generate highly dense polymers with controlled density and thickness on both flat surfaces (e.g., gold, glass and PDMS) as well as nanoparticles. This concept was further exploited to develop hierarchical films with structurally regulated functionalities for improving both the biological activity of the surface coating for immobilizing active agents all the while maintaining excellent non-fouling properties. Surface coatings were also generated using the “graft-to” approach. Using this technique, pCB polymers were conjugated to the biomimetic adhesive group, DOPA, which subsequently enabled their grafting to multiple surface platforms such as gold, glass, iron oxide, as well as polymer fibers. Additionally, the development of a tri-block co-polymer consisting of pCB-pPO-pCB offered an additional material for the convenient and simple attachment onto hydrophobic surfaces. These “graft-to” approaches achieved the polymer density necessary for excellent resistance to complex media. Furthermore, a zwitterionic hydrogel was fabricated by copolymerization of a CB monomer and a CB crosslinker. This hydrogel demonstrated intriguing combination of unique properties such as high mechanical loading, excellent non-fouling property and enhanced functionalization. Hydrogels with both mechanical and chemical gradients were built in one single gel. Taken together, these techniques enable the versatile use of zwitterionic pCB polymers for forming biofouling resistant surface coatings in real-world applications.

TP-24

THE EFFECTS OF CHARGED AND UNCHARGED XEROGEL SURFACES ON THE ADHESION STRENGTH OF THE BROWN ALGA ECTOCARPUS CROUANIORUM

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Biofouling is the adhesion of species of sea life (including bacteria, plants, barnacles, tubeworms, bivalves and the like) to a variety of submerged structures. When biofouling accumulates on the hulls of ships, an increase in drag, results. This drag requires more force and more fuel to propel the ship through the water. As a result, the Naval Industry pays billions of dollars each year in extra fuel. Antifoul and foul release coatings have been investigated to help alleviate the adhesion of sea life to submerged surfaces. Ectocarpus Crouaniorum is a genus of brown algae that has been found to adhere to surfaces coated with antifouling paints containing copper. A series of five coatings were prepared with similar surface energies and contact angles but differing abilities to develop discrete positive charges. The five coatings were exposed to ectocarpus filaments and the adhesion strength and removal rate were studied. It was found that coatings with discrete surface charges had greater adhesion strength than the coatings without the surface charge. The charged surfaces also had a lower percent removal when exposed to shear stress. From the initial testing, it is suggested that a charged surface allows ectocarpus to adhere more strongly to surfaces. Emmanuelle Evariste is partially supported by International Paints. Caitlyn Gatley, Maureen E. Callow, James A. Callow, and Michael R. Detty thank the Office of Naval Research for funding.

TP-25

SELF-POLISHING ZWITTERIONIC NONFOULING COATINGS FOR MARINE APPLICATIONS

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Super-hydrophilic surfaces are essential for preventing the unwanted settlement and attachment of bioorganisms on submerged underwater surfaces. Zwitterionic polymers are well known for their nonfouling applications due to the formation of a strong hydration layer around zwitterions. In order to create surface coatings with strong mechanical strengths, we have formulated zwitterionic coatings based on a hydrophobic hydrolysable ester. The hydrophobic nature of the polymer before hydrolysis gives the mechanical strength of the coatings while the top surface of the coatings undergoes hydrolysis upon contact with water. The coatings are self-polished layer by layer. The ester is chosen in such a way that its hydrolysis occurs in the marine environment at the controlled rate. The ester group, polishing rate and the corresponding non-fouling behavior are extensively studied. An optimal composition was identified, combining excellent stability, desired hydrolysis rate and good non-fouling performance. This type of coating is suitable for applications demanding long-term applications.

TP-26

BIO-RESPONSIVE ANTIFOULING COATINGS

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Marine biofouling, the buildup of microbial slimes, plants, and animals on any surface immersed in seawater, is one of the most significant problems affecting ships today. On ships' hulls, biofouling causes an increase in surface roughness, which decreases fuel efficiency, and if left unchecked can lead to further damage including structural disintegration, corrosion, and ultimate failure of immersed structures. Increased fuel consumption, hull cleaning, paint removal and repainting, and associated environmental compliance measures all contribute to the high costs of biofouling, with government and industries spending and estimated $5.7 billion dollars annually to prevent and control biofouling. The US Navy spends nearly $1 billion a year in associated biofouling costs, and could expect to realize a 5% savings in fuel costs for every ship with a non-fouled hull. To date, the most successful antifouling coatings have relied upon slow and controlled release of various active ingredients such as metals (copper or zinc) or organic biocides (tributyltin) to deter marine fouling. However increasing environmental regulations in response to demonstrated lasting toxicity of these compounds has resulted in bans or prohibition of many of these coatings in international waters. There is a clear need to develop a new class of marine coatings that will prevent fouling through some other more environmentally responsible mechanism, preferably an approach that will not release any contaminants in the environment. Luna Innovations has developed novel coatings that will remedy biofouling by specifically attacking the organism's bioadhesives to prevent attachment to marine vehicles. This antifouling coating technology will combine a naturally bioresistant polymeric surface with chemical moieties that have demonstrated the ability to degrade bioadhesives. This technology will result in a huge cost savings by producing a maintenance free hull coating that improves fuel efficiency over 10-12 years.

TP-27

ANTIFOULING ACTIVITY OF COATING CONTAINING MACROCYCLIC LACTONES (IVERMECTIN 0.1%) IN SUB-TROPICAL FIELD TESTS IN BRAZIL

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A new "low emission" principle for inhibition of barnacle colonization at marine coatings have recently presented [1]. The anti-barnacle activity is related to intoxication of the metamorphosed *Balanus improvisus* by macrocyclic lactones (ivermectin 0.1%) during penetration of the barnacle in the coating. Emission of ivermectin in the marine environment is not needed in this anti-fouling method can be shut off with the use of suitable, non-erosive coatings (Pinori et al, This conference). In this study we have repeated the experiment in the cited work, using the same formulation and same set-up, but in a demanding tropical and sub tropical biofouling condition - the Guanabara Bay (Rio de Janeiro) and Arraial do Cabo, in Brazil.

Conclusion The 0.1% (w/v) ivermectin addition in the marine coatings showed convincing anti-barnacle activity against *Megabalalanaus coccopoma, Balanus amphitrite and Balanus trigonus* indicating that the new "peneteration sensitive anti-barnacle method" method is not restricted to *B. improvisus*, but have effects on most (if not all) acorn barnacle. We also made a new observation that the macrocyclic lactons (ivermectin 0.1%) shows significant anti-colonization effect also on *Bugula neritina (Bryozoa)* and *Styella plicata* (*Urochordata*). Beside Barnacle, the Bugula is also considered to be a serious biofouling in tropical and sub-tropical waters. Details of the field study will be presented in the poster.

1. Pinori, E., et al., Multi-seasonal barnacle (Balanus improvisus) protection achieved by trace amounts of a macrocyclic lactone (ivermectin) included in rosin-based coatings. Biofouling, 2011. **27**(9): p. 941-953.

TP-28

A COATING PENETRATION DEPENDENT ANTIFOULING CONCEPT

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We have recently presented a new strategy in fighting barnacles on man-made surfaces [1]. The hypothesis that the observed antifouling activity achieved on barnacles by trace of macrocyclic lactones (ivermectin) was depending on the penetration of the barnacles into the coating, rather than on biocide released from the coating, has now been finally demonstrated by a series of laboratory experiments and in the field. Two experimental coatings were prepared. On the harder styrene based coating, the barnacles could settle and grow on the coating surface without penetrating the film significantly. In contrast, on the softer versatate based coating the barnacles could settle, grow as well as penetrate the film all the way down to the underlying substratum.

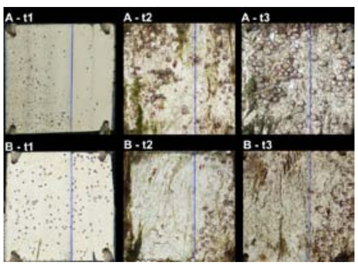

Figure 1. Panel (A) and (B) are painted with styrene based "harder" and versatate based "softer" paint respectively. The two panels have been immersed in Swedish west coast water for three summer months. t1, t2 and t3 are the three immersion intervals. The barnacles are highlighted in dark circles. The panels contain 0.1% ivermectin (w/v) on the left side of the blue line. The right side is control area. In the magnified pictures the penetration/non-penetration behaviour of barnacles on the two different control area is visible: penetration is occurring only in the soft (B) coating system.

When the two paints were added with 0.1% (w/v) of the ivermectin (Figure 1), the softer coating (B) showed 100% antibarnacle activity, while the harder coating (A) did not show any antibarnacle activity, despite same release rate for the biocide as for (B). The biocide release rates, as measured by the ISO1518 method, were for both formulations in the range of 0.05-0.1 ng/cm^2/day. Both (A) and (B) are waterborne polymer based paints, showing very low erosion rates and not containing any other biocide than 0.1% ivermectin. Further development of this new concept will include optimized binder materials and combining it with chemical or mechanical fouling protection strategies against other fouling organisms. Our findings can be the basis for development of novel antifouling coatings, with significantly lowered initial loading and release of biocides, and longer service life. This would improve both the economical and the environmental aspects, finally making an alternative to copper coatings feasible in the near future.

1. Pinori, E., et al., Multi-seasonal barnacle (Balanus improvisus) protection achieved by trace amounts of a macrocyclic lactone (ivermectin) included in rosin-based coatings. Biofouling, 2011. **27**(9): p. 941-953.

TP-29

AMPHIPHILIC POLYSACCHARIDES AS INERT SURFACE COATINGS

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For the development of the next generation of foul release coatings it is important to explore new classes of nontoxic and nonfouling materials. The potential of polysaccharides as fouling-resistant coatings lies in their chemical structure: due to the presence of hydroxyl-groups, they are highly hydrophilic and able to form water-storing hydrogels. Their resistance against bacteria and mammalian cells was e.g. demonstrated by Morra and Cassinelli.[1] Cao *et. al.*[2] applied these materials to the marine environment and showed that different acidic polysaccharides have a high anti-fouling potential in terms of protein resistance, but loose this promising property in the marine environment. This collapse is caused by a complexation of bivalent cations like Ca^{2+}.[2-3] In this study, the free carboxyl-groups of two polysaccharides, hyaluronic acid and chondroitin sulfate were postmodified with the hydrophobic trifluoroethylamine. With this strategy, different intentions could be realized: a blocking of free carboxyl groups to prevent complexation of ions and a preservation of the resistance in marine environment, a shifting of the contact angle towards the minimum in the Baier curve to maximize inert properties[4] and the introduction of amphiphilic properties due to the hydrophobic fluoro-groups.[5] Some of the coatings show very good protein resistance and a high resistance against a range of biofouling species.

TP-30

POLYMER BRUSHES AS MODEL INTERFACES FOR MARINE ANTIFOULING

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Biofouling and bioadhesion are complex process that involve living organisms and cells approaching and attaching to surfaces, and always caused serious problems, including reduced ship speed, increased fuel consumption due to drag, corrosion and malfunction. Surface chemistry, topography, and bulk properties of the substrate all affect the settlement and strength of biological adhesion[1]. So, the ability to tailor a surface property to control the biological adhesion would have the key application hints. Our current research are exploring new antibiofouling approaches by fabricating nano- and micro-structured surfaces and interfaces or tailoring polymer structure at the molecular-level, with the goal of preventing marine organisms from attaching. We anticipate that these approaches may be broadly applied for screening antibiofouling materials. With regard to marine antifouling coatings, restrictions on the use of biocide-releasing coatings have made developing less-toxic and nontoxic antifouling surfaces more important.

Polymer brushes are ordered polymeric films with polymer chains' one end chemically attached to substrate surface.[2] It provides an ideal model platform for studying bioadhesion. We recently designed novel antifouling polymer brushes based on universal macromolecular initiator which can be assuredly self-assemble onto variable inorganic and organic surfaces to improve the performance of protein and cell adhesion at laboratory scale. Ongoing studies include the investigation of composition and length of polymer brushes on cell and protein resistance, extension of this strategy for conferring fouling resistance to various substrate surfaces, and the development of patterning strategies for patterning adhesive and non-adhesive regions of a surface.

The study on the biological properties (e.g., antitumor, anti-cancer, toxicity, DNA cleavage) of ionic liquids is an emerging research field. The anti-microbial behavior is an important characteristic of ionic liquids. In this context, we have developed high-density poly(ionic liquid) brushes for use in biofouling prevention. Preliminary results confirmed that the poly(ionic liquid) coating have a significantly anti-chlorellas adhesion and excellent antibacterial properties.

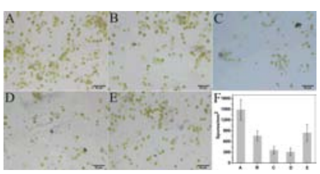

Fig. 1. Representative optical micrographs of settled organic Chlorella spores on unmodified Ti surfaces (A), PNM-MIm-Cl-Ti (B), PNM-MIm-BF4-Ti (C), PNM-MIm-PF6-Ti (D), PNM-MIm- N(SO2CF3)2-Ti (E), Organic Chlorella spores settlement data on bare and poly(ionic liquid) brushes (the different counter-ion) coating Ti surfaces (F).

Fig. 2. Optical microscope images of samples after hung in the laboratory seawater for 20 days: flat PDMS substrate (left); POEGMA brushes grafted from wrinkled PDMS substrate (right).

The success of the wrinkled PDMS surface design[2] with repelling spores has propelled our research to search for further improvement on antifouling properties by tethering PEG-based polymers or quaternary ammonium salts (QASs)-containing polymers onto wrinkled PDMS surfaces. Laboratory experiments indicate that it has exceptional nonadhesive/nonfouling properties due to the synergistic effect of chemical composition of the coating as well as surface topography. It is interesting to ponder the possible use of this strategy for preparing nonfouling surfaces resistant to attachment of mussels, barnacles, and other classic marine biofouling organisms.

Based on these fundamental research results, we are also developing environmental friendly antifouling coatings which have both low surface energy and self-polishing property by using novel self-prepared organic fluorine-silicon modified acrylic resin as film-forming substance. Seawater field marine fouling tests indicate these coatings can effective against some bacterial growth and attachments and exhibit excellent antifouling performance.

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TP-31

SWITCHABLE SURFACES TO REGULATE ADHESION OF MARINE BACTERIA

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Surface colonization by microorganisms, leading to the subsequent formation of a biofilm, is often described as one of the critical early events in marine fouling since biofilms may influence the subsequent settlement of spores of algae and larvae of invertebrates. Controlling the formation and detachment of unwanted biofilms requires an understanding of the mechanisms that organisms use to interact with submerged substrata. Advanced material engineering techniques, such as self-assembly can structure surfaces that allow dynamic tuning of their properties (*i.e.* wettability and superficial charge). Recently, switchable surfaces able to undergo conformational switch in response to an external stimulus applied have been shown to be a suitable platform for controlling cellular responses. Moreover, previous studies reported superficial charge to have a "key role" in the first steps of bacterial adhesion to a surface. Here we present a novel system to influence bacterial attachment based upon the conformational switching of negatively charged 11-mercaptoundecanoic-acid (MUA) tethered to a gold surface in response to an applied potential. MUA is homogeneously distributed on the substrate and spaced by shorter and neutral mercaptoethanol, (MET). This configuration allows enough space for conformational switches to happen as demonstrated by X-ray Photoelectron Spectroscopy (XPS) and Sum-Frequency Generation (SFG) spectroscopy. Electrochemical Surface Plasmon Resonance (E-SPR) has been used to monitor the adhesion of the marine bacteria *Marinobacter hydrocarbonoclasticus (Mh)* to the described switchable SAMs.

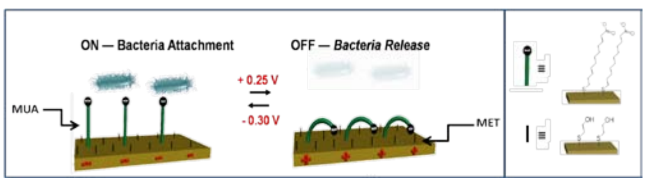

Figure 1. *Schematic representation of Mh adhesion to the switchable SAM.*

The results obtained show that when a positive potential is applied to the gold surface and the MUA charged end-groups are concealed, the surface becomes neutral and less hydrophilic reducing the adhesion of bacteria. On the contrary, when a negative potential is applied to the gold substrate the acid end-groups are exposed and bacterial adhesion to the surface increases dramatically. Such surfaces show great promise for the study of interactions controlling the formation and detachment of biofilms and the principles behind such tunable surfaces provide the basis for more practical coatings to control marine biofilms.

TP-32

NATURAL PEPTIDE NONFOULING MATERIALS AND COATINGS BY COMBINATORIAL AND RATIONAL DESIGN

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It has been established that surfaces modified with zwitterionic and mixed charge polymers such as poly(carboxybetaine) and poly(sulfobetaine) can effectively resist nonspecific protein adsorption and bacterial adhesion/biofilm formation in complex media. These zwitterionic materials have been successfully demonstrated to show resistance to spore/diatom and barnacle attachment, ideal for marine coatings. Mixed charge nonfouling polymers are also attractive for practical applications because of their simplicity, broad variations, and low-cost. Among mixed charge polymers, alternating charge peptides are of particular interest as natural materials. Peptide-based nonfouling polymers also offer potentially infinite variability in structure and properties. Marine environments are very complex, containing thousands of marine bacteria. Thus, it is necessary to provide a deeper understanding of nonfouling mechanisms and to search for a series of materials suitable for applications for marine coatings.

We have developed several experimental methods to introducing peptides onto surfaces, and demonstrated how different peptide sequences are able to alter fouling properties. We have the potential to produce incredibly large libraries of different surface sequences using combinatorial synthesis techniques. We have also been able to determine unknown peptide sequences in order to discover new non fouling sequences using mass spectroscopy. The entirety of the work described is unique because of its blend of experimentation and modeling. By being able to couple a peptides sequence to its fouling properties, a quantitative structure-property relation (QSPR) model can be used. Despite some progress in the fundamental understanding of molecular-level nonfouling mechanisms and the development of several new nonfouling materials, only a handful of nonfouling materials are available and their long-term performance in complex marine environments is still unknown. The implementation of peptides will enable us to determine molecularly what properties are useful and important for developing nonfouling marine coatings.

TP-33

CHEMISTRY DEPENDING SURFACE CONDITIONING AND ITS IMPLICATION FOR COLONIZATION BY MICROORGANISMS

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The adsorption of macromolecules is among the first steps in biofouling, which starts immediately after surfaces are immersed into the ocean. The influence of surface chemistry and physicochemical coating properties on the formation and composition of conditioning layers have been investigated. In our study, self-assembled monolayers (SAMs) on gold are used as highly controlled surface chemistries which allow to fine tune the physicochemical surface properties and to study their effect on the formation of conditioning layers. In agreement with previous work, chemical termination of the surface affects the settlement kinetics of spores of the macrofouler *Ulva linza* [1]. *Ulva* settlement on the pristine surface chemistries and the successive formation of conditioning layers depending on the surface chemistry was investigated in greater detail by surface analytical techniques. Spectral ellipsometry was used to measure the increasing conditioning layer thickness over a timescale of 48 hours. Contact angle measurements revealed that formation of this protein layer changes the wettability of the surfaces and with IRRAS it was possible to show, that the surface chemistry changes the composition of the conditioning film. It also could be shown, that organism settlement is significantly changed if pristine chemistries are compared to conditioned surfaces and especially for longer biological assays conditioning needs to be taken into account.

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TP-34

MIMICKING NATURE THROUGH MODELLING FOR NONFOULING MATERIALS

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It is still challenging to design materials which resist non-specific adsorption of bio-macromolecules, microorganisms, and bacteria. Nature, however, presents many examples of resisting non-specific adsorption. For example, the cytoplasm is a crowded environment, yet proteins are stable at high concentrations; proteins do not aggregate non-specifically. Another example is the interior surfaces of molecular chaperones, which assist hundreds of different proteins to fold. This is remarkable because the surface of chaperones must non-specifically bind hundreds of protein types and release them after they fold. We have used molecular modeling and bioinformatics to better understand these two examples of Nature's resistance to biological molecules. This understanding has allowed us to design peptides and peptide structure-function relations for non-fouling. The main conclusions are that Nature uses specific charged amino acids to create strongly hydrophilic surfaces on both the surfaces of proteins and on the interior surfaces of molecular chaperones. Additionally, the particular amino acids used on molecular chaperone surfaces and protein surfaces are especially poor at forming salt bridges or other specific interactions, thus preventing specific binding between proteins. Peptide materials based on these principles have synthesized and tested with excellent performance against a panel of proteins. The principles learned from Nature are being applied in high-throughput screening of new non-fouling materials, via a combination of quantitative structure-property relationship (QSPR) analysis and modeling. A software library with QSPR methods specific to peptide systems has been developed and validated on existing results in the literature of peptide screening. The lessons learned from this modeling work are the first steps towards a broader understanding of the principles of non-specific interactions in biology.

TP-35

AMBIGUOUS SURFACES FOR ANTIFOULING AND FOULING RELEASE APPLICATIONS

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The collaborative work presented here seeks to develop a fundamental understanding of new non-toxic, fouling resistant coatings for marine applications. Anti-fouling properties are typically achieved through the surface deposition of a hydrophilic polymer such as polyethylene glycol (PEG), which is well known for its excellent resistance to the adsorption of proteins and marine micro-organisms such *Navicula* diatoms. However, certain organisms such as *Ulva* zoopores have been found to adhere very strongly upon hydrophilic surfaces. *Ulva* zoopores attach in greater numbers to hydrophobic surfaces; however, their attachment is weak, making them easily released under shear stress. This has led to an effort to produce ambiguous surfaces, those containing both hydrophobic and hydrophilic moieties or regions. One general approach is to spray coat a surface layer of comb-like block copolymers with amphiphilic side chains (surface active block copolymer SABC) on top of a relatively thick thermoplastic elastomer coating whose morphology provides the low elastic modulus necessary for good fouling release. Bio-fouling assays reveal that these comb-like polymers can show high removal of both *Ulva* and *Navicula*, demonstrating that such amphiphilic surfaces are effective antifouling coatings. Another promising SABC system is PS-b-P(EO-co-AGE) block copolymers. In this approach the backbone incorporated AGE units can be chemically functionalized via UV-assisted thiol-ene chemistry. PS-P(EO-co-AGE) polymers functionalized with 1H, 1H, 2H, 2H-perfluorooctanethiol, show very low settlement of *Ulva* spores as well as exhibiting excellent foul release properties (~100%), far better than the PDMS standard. This P(EO-co-AGE) system also opens up the possibility of chemically patterning surfaces with UV photolithography. This research is supported by the U. S. Office of Naval Research

TP-36

FOULING CONTROL USING AMPHIPHILIC COPOLYMERS

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Amphiphilic networks based on the intermixing of hydrophobic and hydrophilic groups are of emerging interest to the Marine Industry as non fouling coatings that provide a topographical and chemical complexity to control the settling of fouling organisms. This study was conducted to evaluate the fouling control performance of amphiphilic surfaces prepared from acrylic copolymers and block copolymers.

A series of copolymers were prepared by radical polymerisation and living radical polymerisation to investigate the polymer architecture on the deterrence of a range of organisms. Block copolymers were prepared by sequential introduction of selected monomers (mPEG-MA and PDMS-MA) using RAFT (reversible addition fragmentation chain transfer) polymerisation. Then the surfaces made from these polymers were studied through AFM and surface energy measurements. Finally their fouling control performance was evaluated against different organisms. Three types of systems were studied which gave different surface architectures. The PEG-MA and PDMS-MA parts were statistically distributed along the polymer backbone to give a homogeneous surface, or mono-functional polymers were prepared and mixed to give microscale features on the surface, and finally block copolymers were prepared to control the surface architecture. Amphiphilic copolymers gave better performance than their hydrophilic or hydrophobic homologues and initial results indicate that the presence of chemical inhomogeneity on the surface has lead to a higher fouling removal.

TP-37

MULTI-LENGTH-SCALE POLYMER GRADIENTS FOR THE INVESTIGATION OF SURFACE-EXPLORATION BEHAVIOUR OF ZOOSPORES OF THE MACRO-FOULING ALGAE ULVA

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The prevention of marine biofouling on submerged man-made structures is undoubtedly complicated by the surface-exploration capability of the settling stages of higher organisms. For example, zoospores of the macro-fouling algae *Ulva* have been reported to select actively between hydrophobic and hydrophilic regions on a surface[1], and cyprids of *Balanus Amphitrite* are capable of selecting regions of surface morphology that are beneficial, in regard to their final adhesion strength[2], effectively increasing their success rate in finding a suitable habitat. Understanding this surface-exploration phase could potentially aid in the development of new non-fouling technologies. Surface gradients offer the possibility to gradually vary a surface property relevant to invoking or preventing settlement, and we hypothesise that by varying the slope of the gradient, information regarding exploration behaviour and area can be attained. A protocol for the fabrication of polymer gradients using perfluorophenyl azide chemistry and deep-UV lithography has been developed, which allows the length of the gradient to be readily varied. In an initial study, surface gradients in density of two non-fouling polymers, poly (vinyl pyrrolidone) and dextran, were tested against settlement of zoospores of *Ulva* and it was found that the number of settled spores decreased with increasing polymer density, showing the applicability of this system for future studies. The gradients have been characterised by means of ellipsometry, XPS and water contact angle. To investigate the chemical and morphological microscale characteristics of the gradients, TOF-SIMS and AFM will be used.

1 Finlay, J. A. *et al.* Settlement of UlvaZoospores on Patterned Fluorinated and PEGylated Monolayer Surfaces. *Langmuir* **24**, 503-510.

2 Aldred, N., Scardino, A., Cavaco, A., De Nys, R. & Clare, A. Attachment strength is a key factor in the selection of surfaces by barnacle cyprids (Balanus amphitrite) during settlement. *Biofouling* **26**, 287-299.

TP-38

INFLUENCE OF SURFACE TOPOGRAPHIC GRADIENTS ON ULVA SETTLEMENT

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For the development of environmentally friendly antifouling coatings, it is important to understand the interaction between marine organisms and surfaces in greater detail [1, 2]. Among the different surface cues, the settlement of cells and larvae has been found to be influenced by surface microtopography[3]. In this study, the influence of surface topographic gradients on the settlement of zoospores of the green alga *Ulva* was investigated. Honeycomb gradient structures were obtained by hot embossing [4], and the effect on the density of spores that attached in laboratory assays was quantified. The highest density of spores was found when the size of the microstructures was similar to or larger than the size of a spore. With decreasing size of the honeycombs, spore settlement decreased. Interestingly, spore settlement correlated with the Wenzel roughness of the surfaces. During settlement, "kink positions" on the surface played an important role and resembled preferred attachment positions. The gradients furthermore allowed determining the minimum pit size chosen by the spores to squeeze in and settle.

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[2] J. A. Callow, M. E. Callow, *Nature Communications*, 2011, *2,* 10.1038/ncomms1251.

[3] A. J. Scardino, R. de Nys, *Biofouling*, 2011, *27*, (1), 73-86.

[4] M. Worgull, W. Andrew, 2009, ISBN-10: 0815515790.

TP-39

EFFECTS OF SURFACE WETTABILITY ON SECONDARY SETTLEMENT OF THE MUSSEL MYTILUS CORUSCUS PLANTIGRADES

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In China, the mussel *Mytilus coruscus* is one of commercial marine bivalves and is also an important fouling organism. Although much research has focused on the mechanism of mussel larval settlement, the information on secondary settlement of plantigrades is little known. In the present study, the relationship between surface wettability and reattachment of plantigrade or juvenile of *M. coruscus* was invetigated. The interaction between surface wettability and plantigrade gregariousness and subsequent reattachment of plantigrades were also investigated. In the small size group, clusters in two surface wettabilities increased with the increasing density of plantigrades and significant differences in clusters were observed at the density of 50 plantigrades in two surface wettabilities. The percentages of plantigrade settlement increased with densities in different surface wettabilities, and significant differences in settlement were observed at the density of 5 plantigrades in these two surface wettabilities. In the big size group, clusters in two surface wettabilites also increased with density of plantigrades, significant differences in clusters were only observed at the density of 30 and 50 plantigrades between two surface wettabilities. Except for the density of one plantigrade, no differences were observed between two different surface wettabilities. When exposed to adult conspecifics, the percentages of settlement of big plantigrades were higher in glass Petri dishes than those in polystyrene Petri dishes. When exposed to adult shells, the percentages of settlement of big size plantigrades were not significantly different between those in glass Petri dishes and those in polystyrene Petri dishes. Overall, surface wettability plays an important role in secondary settlement of plantigrades in the presence of mussel density and/or size. The density is a key factor in secondary settlement of *M. coruscus*. Adult conspecifics also play an important role in secondary settlement in this species.

TP-40

SETTLEMENT OF THE MUSSEL MYTILUS CORUSCUS PLANTIGRADES IN RESPONSE TO NATURAL BIOFILMS

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The mussel *Mytilus coruscus* is one of commercial marine bivalves and is also an important fouling organism in China. Although many researches have focused on the mechanism of mussel larval settlement, the information on secondary settlement of plantigrades is little known. In the present study, settlement of plantigrades of *M. coruscus* in response to natural biofilms was investigated in the laboratory. Various aspects of biofilms were examined including age, dry weight, chlorophyll *a* concentration, bacterial and diatom densities, and bacterial community structure. Plantigrades of *M. coruscus* settled in response to biofilms. Settlement of plantigrades was independent of biofilm age. The dry weight gradually increased with the age of biofilms. Biofilm age significantly affected bacterial and diatoms density and chlorophyll *a* concentration. Dry weight, bacterial and diatoms density and chlorophyll *a* concentration significantly correlated with biofilm age, respectively. The biofilm activity was positively correlated with dry weight, bacterial and diatom densities, and chlorophyll *a* concentration. In addition, bacterial structure plays an important role in the settlement of *M. coruscus* plantigrades. Overall, this investigation indicates that biofilms can promote the settlement of *M. coruscus* plantigrades, and that bacterial and diatom densities, chlorophyll *a* concentration and bacterial structure are important factors influencing secondary settlement in this species.

TP-41

QUANTIFICATION OF THE ADHESION STRENGTH OF THE DIATOM NAVICULA PERMINUTA IN A MICROFLUIDICAL ASSAY

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Foul Release (FR) technologies have been proven as a suitable approach against marine biofouling and ongoing research aims on enhancement of coating formulations.[1] FR coatings make use of very weak adhesion strength of attached organisms enabling to shed them off easily by the shear forces present at the hull of a cruising ship. We developed a microfluidic shear force assay which allows to simulate this situation and to study cell adhesion strength on different substrates quantitatively, under static and dynamic conditions.[2,3] After an attachment phase, the adhesion strength of strongly bound cells can be measured by detaching them from substrates using a stepwise increased flow inside our microfluidic system. With this device we can determine both, the fraction of adherent cells and the critical shear stress which is necessary to remove 50% of the adherent cells. In the presented work we investigated the adhesion strength of the diatom *Navicula perminuta* on chemically different substrates testing the effect of different incubation conditions- statically and dynamically. Inert chemistries can readily be discriminated from surfaces with low foul release potential and be correlated with the adherent cell fraction. The sensitivity of the technique allows revealing even small differences in adhesion caused by a change in surface properties.

TP-42

FIELD TESTS OF THE EARLY COLONIZATION STAGE OF MODEL SURFACES

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While many studies correlate surface chemistry with results of laboratory assays, field tests are the real benchmark for antifouling performance. The majority of these studies concentrate on fouling composition on samples submerged over a couple of weeks or even months. The surfaces usually used are panels of promising coating formulations. There is no data about the composition of communities on model surfaces like SAMs or chemically coupled bio-macromolecules as frequently used in laboratory experiments. For this study three kinds of surfaces were used: SAMs with different termination and hydration properties, amphiphilic polysaccharides and poly[oligo(ethylene glycol) methacrylate] (POEGMA) brushes. The choice of the samples was based on the protein resistance of polysaccharides and POEGMA, and the possibility to study the influence of chemistry and hydration through different SAMs. The samples were immersed in November and December 2011 at the FIT test site on the east coast of Florida for 2, 6, 12 and 48 hours. The dominant organisms observed on every surface were the diatoms *Navicula, Mastogloia, Cocconeis* and *Amphora* and the protozoa *Peritrich*. The abundance of these organisms was found to be influenced by environmental conditions. The time depending colonization and the distribution of the community is correlated with the physicochemical properties of the coatings. Compared to polymeric coatings some self assembled monolayers show surprisingly good performance.

TP-43

DEFOULING LOOP IN YIELDING SETTLEMENT SUBSTRATES

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Macrofouling organisms exposed to water flow inevitably transmit lift, drag and torsion to their supporting substrates. A substrate unable to tolerate such pressures acts as a foulage release coating. Giving away under pressure, growth and release of settlers loop into a steady state of biomass accumulation over time without additional mechanical cleaning. Results from immersion tests indicate that the loss of settling substrate during defouling is a function of the rate of attachment and the size of organisms released from the substrate. In addition, it was evident from cleaning experiments under accelerated flow conditions that defouling was effective without disruption of organisms. Examples of the contact-release effect will be presented for each (1) the hydrodynamical termination of settlements, (2) the rheologically engineered limit for settlements and (3) the hyper-hydrodynamical termination of fouling in active cleaning. We will discuss options to apply the contact-release effect for net coatings in mariculture.

Our contribution is funded by the Deutsche Bundesstiftung Umwelt (az29456).

TP-44

A NOVEL METHOD FOR BIOFOULING PREVENTION

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Biofouling is a significant issue for Navy ships, fixed test gear, and sensors deployed underwater because it can compromise the operations of these systems, as well as increase fuel consumption. Although recent research efforts have focused on antifouling methods, these frequently are ineffective, costly, and pose a risk to the environment (e.g., chemically active coatings or heavy metals). This study evaluated a range of existing paints (i.e., SeaGuard Heavy Metal Free, Intersleek, and copper [28% cuprous oxide]) and novel antifouling techniques (i.e., aeration) on fiberglass test panels and a model boat hull in Narragansett Bay, Rhode Island; and aimed to determine which method maximizes efficacy against biofouling, while minimizing cost and environmental risk. Coated test panels and control panels were deployed in Narragansett Bay and sampled monthly from May through September 2011. Percent cover was estimated for biofouling species. Results indicated that panels treated with SeaGuard, copper paint, and Intersleek had less fouling than panels treated with aeration and control panels. Biofouling community composition was similar among treatments and included both soft-bodied fouling organisms (algae, bushy bryozoans, hydroids, sea squirts, sponges, and tunicates), as well as hard fouling organisms (barnacles, encrusting bryozoans, mussels, and calcareous polychaetes). SeaGuard was the most effective antifoulant, followed by copper paint, Intersleek, and aeration. Findings from the aeration treatment were contradictory to those obtained from other studies. A follow-on study in the summer of 2012 will assess those properties of an aeration system (i.e., minimum pressure and angle of air delivery) that make it most effective in removing biofouling on submerged surfaces. Results of both studies will be considered in the design of a full-scale aeration system that can be tested to provide a cost-effective preventative and remediation tool for both active and inactive Navy vessels.

TP-45

INVESTIGATION OF TITANIUM'S FOULING IN SEA WATER

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The paper presents the results of tests on fouling of titanium in sea water carried out by Ship Design and Research Centre in the scheme of own research project. Also the paper shows research on pitting and crevice corrosion of titanium in sea water and influence of chlorine dosing to fouling and corrosion of titanium.

Titanium and its alloys have high strength, low density and high resistance to corrosion at ambient temperature and at high temperatures up to 450°C during constant work and up to 700°C during short-time work. This metal shows also high corrosion resistance e.g. to crevice corrosion and to pitting corrosion in the marine atmosphere and sea water. All over the world titanium and its alloys are applied in production of yachts, screw propellers and heat exchangers. But chlorine dosing may damage passive layer on titanium what may be initiation of corrosion.

In literature was written that surfaces of constructions made of titanium that were immersed in sea water were not fouled by living organisms (crustacean and algae). Other authors inform that titanium is fouled by barnacles and seaweed. To clear up contradictory information about fouling of titanium by living organisms in sea water specimens made of titanium were exposed in Gdynia Shipyard sea water on our floating corrosion station. Moreover, fouling of titanium and PMMA was compared. Chlorine dosing in different concentrations to prevent fouling was carried out.

TP-46

THE EFFECT OF GROOMING AND CLEANING ON THE CONDITIONING OF TRANSPLANTED PANELS WITH SILICONE COATINGS

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Original location has been shown to affect subsequent fouling on transplanted panels on both silicone and copper coatings (Floerl et al. 2005; Ralston et al. *in prep*). This has implications for the antifouling performance of a coating and the attraction and transport of non-indigenous species. This experiment was designed to test whether coating husbandry would affect the conditioning of silicone coatings and their fouling post-transplant. Panels were coated with three different silicones and placed at two locations on Florida's east coast: Port Canaveral and Sebastian Inlet. The two locations are typified by different fouling communities. Panels were cleaned, groomed weekly, or allowed to foul freely. After conditioning, panels were cleaned and transplanted between the two locations and fouling was monitored to determine whether the original location and/or treatment affected subsequent fouling.

TP-47

COLONIZATION PATTERN OF BIOFOULING COMMUNITY IN THE COASTAL WATERS OF KALPAKKAM, BAY OF BENGAL, INDIA

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Biofouling studies in the coastal waters of Kalpakkam, south east coast of India, Bay of Bengal, were carried out during 2006-10 to evaluate the pattern of fouling, the dominant species, seasonal succession and the role of environmental driving forces acting upon them. Wide weekly as well as seasonal variations in the colonization of macrofoulants were observed. The succession pattern of the biofouling community was as follows; barnacle - hydroid - sea anemone - ascidian and finally green mussel (*Perna viridis* Linn. 1758). A noticeable shift in the peak settlement period of green mussel was observed as compared to earlier report and it was found to be the climax & most dominant fouling species. However, during entire period of 2010, green mussel appearance was negligible or almost absent. Appearance of oysters on panels was encountered in significant numbers (~7-10% of total density) during the present study unlike the previous reports from this locality. Throughout the year, barnacle settlement was observed and was at their peak during monsoon and post-monsoon period. Pre-monsoon period (July-September) showed relatively high species diversity index which coincided with relatively high temperature, salinity and chlorophyll a. The total number of fouling taxa observed was 103 and of it, 75 were identified. Bivalves showed a successful relationship between its larvae and adults. Macrofouling association indicated a positive relation of bivalves with barnacles. Correlation analysis indicated that salinity, temperature and chlorophyll-*a* were the principal factors that significantly influenced the intensity of larval recruitment as well as macrofouling community development at this location. The paper also delineates the biofouling control measures to be adopted for cooling water system of Fast Breeder Reactor which is under construction at this location. Moreover, seasonal variations in settlement pattern of biofouling organisms on nine types of metals are also discussed.

TP-48

OPTIMIZING BRUSH DESIGN FOR THE GROOMING OF SHIP HULL COATINGS

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Underwater grooming has been proposed as a method to maintain ship hulls in a smooth and fouling-free condition by dislodging potential fouling species before they become established on the surface. A five-head rotating brush system was used to investigate, understand, and optimize grooming tool function. The brush system was mounted to an instrumented gantry carriage capable of controlling the normal force, translation rate and rotation rate of brush operation while monitoring power requirements. A range of brush types were tested on several different hull coatings and applied forces monitored. Models including the power requirements of each brush type along with the forces applied to each coating will be presented.

TP-49

FUNCTIONAL ROLE OF FOULING COMMUNITY ON AN ARTIFICIAL REEF AT THE NORTHERN COAST OF RIO DE JANEIRO STATE, BRAZIL

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The northern coast of Rio de Janeiro State shows a scarcity of natural consolidate substrates, making it a proper environment to the development of artificial structures research. After an initial test of the type of substrate, concrete seems to be the most appropriate for fouling community studies. This research was carried out to investigate the functional role of biofouling on the ichthyic colonization at the northern coast of Rio de Janeiro State. Percentage data of the epibenthic organisms' coverage and samples of the fish community with gillnet and visual census showed that biofouling in artificial reefs might have more than one functional role, acting as a facilitator in the recruitment of fish species and as a link in the trophic marine chain. Through the increase of localized structural complexity provided by the reef itself and by the fouling organisms, which act as "engineering species", additional protection options are offered to the ichthyic community, especially recruits. Also, the epibiont biomass represents an important link in the food web, acting either as a direct source or in the transference of energy to higher trophic levels. Through the relationship between the ichthyic and fouling communities we concluded that the functional role of the latter in artificial reef habitats could be characterized mainly as shelter and feeding grounds for several fish species.

TP-50

A FIFTEEN-YEAR ASSESSMENT OF FOULING COMMUNITY ASSOCIATED WITH ARTIFICIAL REEF ON THE NORTHERN COAST OF RIO DE JANEIRO, SOUTHEAST

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Artificial reefs are important elements of integrated management plan on several countries aiming to increase and diversify the coastal resources and the associated communities. Fouling community succession was evaluated since time zero (May 1996) on an artificial reef, 9 feet deep and 5 nm from the north coast of Rio de Janeiro (21°29'S, 41°00'W), considering seven sampling periods on concrete panels attached to each of 36 reefballs: 1 month, 6 months, 1 year, 6, 7, 10 and 15 years after the modules disposal. Numerical indicators of species richness and diversity reached maximum values (N_{tot} = 28 spp, N_{med} = 16.5 spp, H = 2.2) at 6 months, decreased and stabilized after 6 years (N_{tot} = 21 spp, N_{med} = 12.7 spp, H = 1.8). Multivariate analysis including the 15 years of community development showed the three distinct colonization stages of the reef complex: 1. Initial phase (1 month) with the dominance of the cirripede *Balanus* spp. (~ 43 %); 2. Intermediate phase (6 and 12 months) with the co-dominance of *Balanus* spp. (~ 26 %), the hydroid *Obelia* sp. (~ 10 %) and the bivalve *Ostrea* sp. (~ 10 %); 3. Posterior phase (6, 7, 10 and 15 years) with the dominance of *Ostrea* sp. (~ 25 %), the octocoral *Carijoa* sp. (~ 19 %) and the hydroid *Bougainvilla* sp. (~ 15 %). Higher turnover rates (~ 40 %) between 1 and 6 months and between 1 and 6 years reflected noticeable changes in the composition and abundance/dominance of representative species, emphasizing the three sucessional stages. Artificial reefs represent a useful tool on he north coast of Rio de Janeiro due to the scarcity of vertical hard substrate, and they are suggested as representative sites for observations of ecological changes in fouling community structure over long-term time scales.

Plenary 3

PLENARY-3

ANTIFOULING REGULATIONS – 25 YEARS ON – IS IT TIME FOR A RETHINK?

Dr Julian E Hunter

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The need to prevent fouling on the immersed areas of ships is undisputed and universally recognised as critically important to minimise the environmental impact of marine transport and for efficient commerce. Since the first restriction of use of products containing TBT biocides on pleasure craft in 1987, antifouling paints have been the subject of environmental scrutiny by academics, environmentalists and regulators alike and are now regulated by a plethora of stringent environmental and safety rules at a local, regional and global level. The reality of such a strict regulatory regime is that no approach to prevention of fouling can be seriously tabled without due consideration of the regulatory hurdles that must be overcome if it is to enter the market.

In this key note presentation the author will consider how antifouling regulations have evolved over the past 25 years and the positive and negative impacts they have brought the antifouling industry , the environment and human safety. Whilst the environmental impact of antifouling paints on aquatic and terrestrial environments can now be predicted using modern risk assessment techniques and strict pass/fail criteria applied to determine whether a product can enter the market, the positive impact of the environmental benefits effective antifouling paints bring to the marine industry is largely ignored by legislation.

Recently the focus of environmental regulation of shipping has turned to reduction of green house gas emissions from ships and translocation of invasive aquatic species into sensitive ecosystems on immersed areas of ships. Efficient antifouling coatings play an important part in reduction of both of these. The challenge for regulators is to balance the severe pass/fail environmental criteria applied during evaluation of antifouling coatings with the benefits they bring.

An integrated approach is needed to assess the impact of antifouling products and a more holistic approach adopted in regulation. It is time to take stock, rethink and redraft!

BIOFOULING ISSUES FOR INDUSTRY
Session Chair: Serena Teo

BII-1

EVALUATION OF COATINGS FOR LONG-TERM BIOFOULING PREVENTION ON DEEP SEA STRUCTURES

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Subsea structures to be located on the seafloor at a depth of about 100 m off the southern coast of Australia required a coating system to provide both anticorrosion and antifouling protection. Further, the structures were projected to have a 15 year life, well beyond the effective antifouling life of most coatings used in the shipping and offshore industries. Metallic copper, or copper-nickel alloy, was identified as most likely to provide the required long-term protection and, as fabricating the structures from cupro-nickel was not an option, coatings which incorporated metal particles within the coating matrix were investigated as an alternative. Test coatings of four systems were applied to pipe sections and immersed in a tropical estuary for periods of up to 18 months. At 3, 6, 12 and 18 months after immersion, all test pieces were visually inspected to assess antifouling performance and one test piece of each coating removed for laboratory study. Surfaces and cross sections of sample coupons cut from the latter were examined by scanning electron microscopy (SEM) and energy dispersive x-ray analysis (EDXA) to determine the abundance, composition and distribution of copper and copper compounds on, and through, the coating surfaces with increasing immersion time, and the relationship between this and antifouling performance. The study successfully identified a coating system to meet the specified requirements, and also provided insight into the mechanism of antifouling action in this type of coating.

BII-2

MANAGING NET BIOFOULING IN TASMANIAN CAGE FINFISH AQUACULTURE

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Currently there are a number of methods for controlling bio-fouling on nets within the finfish aquaculture industry. These include land- based cleaning of nets, in-situ cleaning of nets and the use of antifouling coatings. The industry is working to reduce its reliance on copper based antifoulant coatings because of their potential for causing adverse effects on local marine ecosystems. The increase in the use and technology associated with in-situ net cleaning may hold the key to the sustainable (economic and environmental) management of biofouling within the industry. But first we need to understand marine fouling settlement on nets and develop best practice guidelines for its removal.

In the present study we have developed a methodology for identifying the types of fouling organisms on selected net types on a monthly and seasonal basis. This will assist in understanding what fouling organisms are present on the finfish cages throughout the year and therefore adapting in-situ net-cleaning methods on a seasonal basis to reduce the growth of fouling as antifoulant paint is phased out. This will also extend to assessing the effect of in-situ cleaning on the dominance hierarchies of the fouling communities on fish farm nets.

Guidelines developed from this research are aimed at making in-situ cleaning more effective and efficient by better understanding the settlement of marine fouling organisms and improving water quality by reducing nutrient and sediment release that may occur with the action of in-situ net cleaning.

CARING FOR OUR COUNTRY

- A collaborative project between Huon Aquaculture Company Pty Ltd and Tassal Operations Pty Ltd made possible by the Federal government Caring for Our Country grant.

BII-3

INCREASE IN DRAG FORCE ON A TWINE WITH DIFFERENT LEVELS OF HYDROID FOULING

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The hydroid *Ectopleura larynx* is one of the dominating fouling species on fish farms in Norway between July and November, and represents an important issue for the aquaculture industry (Figure 1). The growth of hydroids on fish farm nets influences the water flow through the net, the sea loads on the structure and the dynamic response of the entire system. These factors subsequently affect the fish welfare in the cages and the potential risk of system failure. Biological material in general and hydroids in particular have complex hydroelastic behavior, which makes it difficult to predict the hydrodynamic effects fouling have on twines and nets. It was necessary to use laboratory experiments with physical models to study these effects, since it is impossible to keep hydroid specimens alive in a fresh water based hydrodynamic laboratory. These artificial models of hydroids (Figure 2) were made from multifilament nylon twines that were melted on the end to simulate the "head" of the hydroid (the hydranth). The density, thickness and length of the artificial hydroids were based on live hydroids grown for different periods of time. In the experimental tests, several growth levels were investigated, and the models were towed in different configurations and with different velocities (Figure 3). The drag force on the models was measured, and the findings showed an increase of up to seven times on a fouled twine relative to a clean one.

This work was funded by the Research Council of Norway trough the grant 190463/S40 Hydroids on aquaculture constructions in Norway.

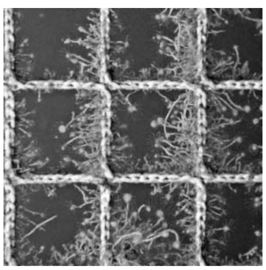

Figure 1. *The hydroid Ectopleura larynx growing on a typical fish farm net. Length scale of the picture is approx. 6 cm.*

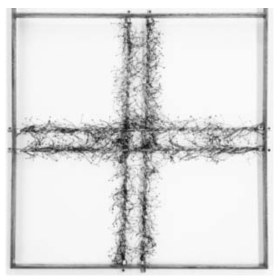

Figure 2. *Frame with the models of net twines fouled with hydroids. Length of the twines in the picture is 30 cm.*

Figure 3. *The frame with model attached to towing carriage in the hydrodynamics laboratory at the United States Naval Academy. Forces on the frame/model at different towing speeds were measured.*

BII-4

ENZYMATIC ACTIVITY IN MARINE MICROBIAL BIOFILMS LINKED TO RISK OF BIODEGRADABILITY IN MARINE POLYURETHANE COATINGS?

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Polyurethanes (PUR) represent a class of polymers that have found a widespread use in marine coatings. So far lipases, esterases, proteases, laccases and ureases have been described in literature as the key hydrolytic enzymes of the microbial induced polyurethane degradation at land. It is questioned, if marine biofilms provide similar degradative environments.

In order to highlight the risk potential a chromogenic micro plate approach has been applied to screen for exoenzymes. A common immersion test setup to let grow micro fouling and macro fouling organisms on the artificial surfaces but special microenvironments below suction cups were created in order to exclude macro fouling organisms from colonizing the whole area of panels immersed. After 75 days of dynamic tidal immersion scrap samples of the biofilms were subsampled for microbial community analyses by next-generation-sequencing of the 16S-DNA and for an enzyme assay.

Results showed that microorganism in biofilms synthesized enzymes belonging to the potentially degrading classes mentioned above. Additionally, change in color of the PUR-coating indicated an enzymatic pathway of biodegradation. Therefore, a more chemical detailed analysis of the PUR coated fragments by Time-of-Flight Secondary Ion Mass Spectrometry (*ToF-SIMS*) was conducted. All findings will be discussed at the conference to give an answer to the initial assumption.

Our contribution is funded by the Deutsche Bundesstiftung Umwelt (az29456) with respect to the research activity of IMARE in new non-biocidal solutions against biofouling in mariculture.

BII-5

ANTIFOULING COATINGS: A DRY DOCK PERSPECTIVE

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Dry docking is a time when decisions are made with respect to maintenance, refurbishment or replacement of the antifouling coating. It also provides an opportunity to conduct a thorough evaluation of the coating condition. This paper describes the inspection procedures and analyses that were developed to evaluate the outer hull condition of several cruise ships operated by Royal Caribbean Cruise Lines. Selected data are presented to illustrate the relationships between: coating type, coating roughness, biofouling, surface preparation, coating application, dry dock capabilities, ship operational schedule and fuel consumption.

Funding for this research was provided by Royal Caribbean Cruise Lines

BII-6

FIELD TESTING OF FOUL RELEASE COATINGS IN STATIC AND DYNAMIC CONDITIONS FOR WATER RESOURCES INFRASTRUCTURE

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Zebra mussels were first introduced in the United States in the 1980's into the Great Lakes. The migration of the mussels west prompted the US Bureau of Reclamation to examine technologies that might reduce their impact on water resources infrastructure. Many new materials and coating products have been developed to deter the attachment of fouling to infrastructure in recent years. In 2008, Reclamation's Materials Research Laboratory initiated a study to evaluate foul release coatings suitable for use on a variety of hydraulic equipment. Parker Dam, on the Colorado River, was selected for our test site due to the rapid reproduction rates of the quagga mussels and the capability to test samples in static and dynamic conditions. The current study includes long term test data from silicone based coatings as well as new foul release coatings technologies including silicone epoxy hybrid systems, nano-textured coatings, and proprietary polyurethane chemistries. Force measurements were also conducted to determine the force to remove mussels from various coated surfaces.

BII-7

ELIMINATING HULL-BORNE AQUATIC INVASIVE SPECIES – AN ALTERNATIVE, PRACTICAL APPROACH

David C. Phillips

Hydrex

Ship, boat and barge hull fouling has increasingly come to the fore as a vector for aquatic invasive species (AIS). The IMO recently issued guidelines for mitigating this threat, the introduction of exotic species being regarded as one of the greatest threats to global biodiversity. Australia and New Zealand are revising the ANZECC Code with a view to protecting their waters from bioinvasion. California is revising state regulations for the same reasons.

Conventional wisdom on the subject recommends the use of biocidal antifouling paint to prevent attachment of nuisance species. However, it is acknowledged that copper and other biocides are not effective in keeping the hull entirely free of macrofouling, especially the protected, niche areas, and that copper and biocide tolerant invasive species pose a worse threat of invasion than those which have not become tolerant to antifouling paint biocides.

It is acknowledged that in-water cleaning is needed to prevent the spread of hull-borne AIS, yet current biocidal paints are not suitable for in-water cleaning: the abrasive tools used damage and deplete the coatings and cause a pulse discharge of biocides hazardous to the local environment and non-target organisms and further afield when disposed of in dredge spoil. For these reasons in-water cleaning of biocidal antifouling coatings is prohibited in many areas.

Current stress is on preventing ships from arriving at their destination with excessive fouling, whereas global elimination of bioinvasion would require that ships sail with a clean hull. Fuel savings attributed to sailing with a clean hull more than cover the costs involved.

An alternative approach to eliminating the hull-borne AIS threat, is the use of a non-toxic surface treated coating system which can be cleaned in the water with no threat to coating or to the environment. This approach can eliminate the hull-borne AIS threat in an economical and environmentally benign way.

BII-8

THE EFFECTS OF LOW INTENSITY ULTRASOUND ON BARNACLE CYPRIDS

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The use of ultrasound to prevent barnacle settlement has been demonstrated in different studies. In this paper, we gave a short review of what is known, and present results in the application of low frequency, low intensity ultrasound on barnacle cyprid exploratory behaviour and settlement. In contrast to irradiation at the higher intensities used in many earlier studies, we found that exposure to low intensity ultrasound (5 kPa) at lower frequency of 23 kHz prevent cyprid settlement but there was no significant mortality or observable change in the cyprid's exploratory behavior. Higher frequencies of 63 and 102 kHz, however, generated no inhibitory effects with low intensity application. Considering energy saving purpose, the efficiency of intermittent ultrasound operation mode was explored and found effective. Results indicate that low frequency and low intensity ultrasound may present a promising and energy efficient strategy for antifouling applications.

Acknowledgement

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MARINE BIOADHESION-BIOADHESIVES
Session Chair: Jonathan Wilker

MBA-1

RESTRICTION ENZYME ASSOCIATED DNA SEQUENCING OF BARNACLES WITH HARD AND GUMMY GLUE PHENOTYPES ON T2 SILICONE FILMS

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Restriction enzyme Associated DNA (RAD) sequencing is an emerging technique for identifying genome-wide polymorphisms (SNPs) in organisms with unexplored genomes. We used RAD sequencing to compare barnacles that produce gummy glue (Gummy) when grown on T2 silicones to barnacles that produce hard glue (Hard) when grown on T2. We generated a RAD sequencing library from pooled genomic DNA for 50 Gummy individuals and for 54 Hard individuals. The DNA barcodes enable identification of each pool after DNA sequencing. The library was sequenced on a single lane of an Illumina HiSeq flow cell and generated 63 million reads (sequences) passing a standard quality filter. Eleven million reads were obtained from Gummy individuals and 21 million reads from hard individuals containing both the restriction site and the 6 bp barcode. The sequences from both populations were assembled into 91,426 contigs (RAD Tags) 38 base pairs in length. The sequences for each population (Gummy and Hard) were then mapped onto the 91,426 RAD Tags identifying a total of 15,426 Single Nucleotide Polymorphisms (SNPs). The frequency of each SNP in the populations was calculated and used to determine genetic differences between Gummy and Hard barnacles at a genome-wide scale. G-tests were used to identify allele frequency differences between the populations for 11,472 SNPs. Using a conservative cutoff of 50 for G-test values, 4,284 SNPs showed significant differences in allele frequencies between the populations. This result was unexpected and suggests that barnacles exhibiting distinct glue phenotypes on T2 silicone may be genetically differentiated at the population level.

MBA-2

A DAY IN THE LIFE OF BALANUS AMPHITRITE: SPATIAL AND TEMPORAL VARIATIONS IN NEWLY FORMED BARNACLE ADHESIVE

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Barnacles attach to nearly any submerged surface with a proteinaceous adhesive secreted and cured in a cyclical multi-step process. We are using epifluorescence microscopy, confocal fluorescence microscopy, and Fourier transform infrared (FTIR) microscopy to explore the chemical and optical properties of barnacle adhesive at the micron scale. Through *in situ* time-lapse optical microscopy of growing barnacles, we have shown that *Balanus amphitrite* releases an initial "barnacle cement secretion" (BCS1) continuously from the periphery of the barnacle during lateral growth and then injects an autofluorescent fluid (BCS2) into the interface periodically. Our experiments reveal that the strength of the nascent adhesive doubles upon addition of BCS2. We have found that this two-step process produces an adhesive with local compositional differences on the micrometer scale. *In situ* fluorescence emission spectra reveal variations in the adhesive that are mirrored by compositional differences observed by FTIR microscopy. The results demonstrate a significant change in protein composition and secondary structure over relatively short distances and time scales as the barnacle secretes and cures its adhesive.

MBA-3

EFFECT OF SURFACE CHEMISTRY ON SPREADING OF BARNACLE (BALANUS AMPHITRITE) CYPRID PERMANENT CEMENT

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Linnea Ista (The University of New Mexico, USA),

Luigi Petrone (Linköpings University, Sweden),

and Anthony S Clare (Newcastle University, UK)

The settlement stage of barnacles – the cyprid – commits to permanent attachment to the substratum by secreting 'permanent cement'. This could be considered to be the most important point in the life of the organism, as the cost of choosing an unsuitable site is likely compromised growth and/or reproduction and even death. There have been few studies aimed directly at the study of this adhesive. Here it is shown that the cyprid's permanent cement can be visualised using a common histological stain, Congo Red. Further the area the cyprid adhesive covers on a surface (around 6000 μm^2 on acid-washed glass) is shown to be strongly conserved between different batches of cyprids, and not to be significantly affected by the age of the cyprids used (up to 9 days post metamorphosis from 6th-stage nauplius) or the addition of 10^{-5}M of the settlement inducer 3-isobutyl-1-methylxanthine.

The effects of surface wettability, charge and surface free energy on the adhesive spreading were determined by utilising well characterised self assembled monolayers (SAMs). Cyprid adhesive secreted onto SAMs produced using R-OH, R-CH_3 and mixes of the two thiols is in accord with thermodynamic theory, with a strong positive linear correlation of adhesive cement area and wettability. The adhesive spreading is also affected by surface charge with a greater degree of wetting of the surface occurring on negatively charged surfaces. Finally a clear link is shown between settlement levels[1] and adhesive spreading on a number of SAMs covering varying surface free energies and charges.

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MBA-4

THE MULTI-PART ADHESIVE OF THE MARINE SANDCASTLE WORM, *PHRAGMATOPOMA CALIFORNICA*

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Sandcastle worms cobble together composite tubular dwellings with sand, seashell fragments, and dabs of a multi-part glue. Molecular components of the glue include at least four polybasic proteins (Pc1,2,4,5), a set of polyacidic phosphoproteins (Pc3A,B), polyacidic sulfated polysaccharides, divalent cations (Mg^{2+}/Ca^{2+}), and iron. Sets of oppositely charged components are electrostatically condensed into four distinct types of granules in at least four types of secretory cell types. The most prominent secretory cells, clustered around the cavity of the first three parathoracic segments, contain homogeneous and heterogeneous granules named for their distinct morphologies. The heterogeneous granules contain sub-granules of condensed polyphosphates (Pc3A,B) and divalent cations. The heterogeneous granules also contain the polybasic Pc1 and Pc4 proteins as revealed by immunolabeling.(*1*) The homogeneous granules contain polybasic Pc2 and Pc5 as well as sulfated polysaccharide counter polyanions. The multiple granule types are co-secreted. Shearing during or shortly after secretion ruptures the granules and their contents are partially mixed. Shear forces may be generated by a combination of three mechanisms: squeezing through narrow openings in the surface of the building organ (a strainer), the beating of bundles of paddle-shaped cilia that cover the building organ, or by twisting of the mineral particles as they are set in place. The glue hardens into a foamy load-bearing solid within 30 s after secretion. The sandcastle glue has served as a useful model for the development of synthetic underwater adhesives based on condensed polyelectrolytes (complex coacervates).(*2*) The sandcastle worm-inspired adhesives are being developed for repair of soft and hard living tissues.(*3*) General purpose versions of the adhesive designed for the high ionic strength of the ocean maybe useful for undersea applications, including coral seeding for reef restoration.

Acknowledgments:

This work was supported by grants from the Office of Naval Research, National Science Foundation, and National Institutes of Health.

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MBA-5

THE INFLUENCE OF POLYMER THICKNESS AND CHARGE ON THE SETTLEMENT AND ADHESION BEHAVIOUR OF MARINE ALGA *ULVA*

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Marine biofouling is a worldwide phenomenon which has become more and more important due to its great economic and environmental impact. The fouling behavior and adhesion mechanisms of marine organisms on different substrata is an important issue to address in order to open up new strategies for designing effective antifouling materials. Surface properties, for example topography, wettability, stiffness, thickness, charge and so on determine the surface selection by organisms prior to settlement on the substrate. In this research work we are focusing on the role of thickness and charge of polymer surfaces on the adhesion behavior of marine alga *Ulva* as there are few studies have been done on these two properties in correlation to marine biofouling field.

Different thicknesses (50, 100, 200, 400, 600 and 1000 Å) of poly(HEMA-co-PEGMA) and differently charged (positive, negative, neutral and zwitterionic) polymer brushes have been succesfully prepared via SI-ATRP. Positively charged polymer surfaces were prepared from 2-dimethylaminoethyl methacrylate (DMAEMA), negatively charged polymer surfaces were prepared from 3-Sulfopropyl methacrylate (SPMA), zwitterionic polymer surfaces were prepared from sulfobetaine methacrylate (SBMA) and neutral charged polymer surfaces were prepared from HEMA and PEGMA. These samples were prepared at an identical thickness (approximately 100 Å).

The spore settlement and adhesion strength assays of marine alga *Ulva* have been used in this work. Massive, rapid and anomalous settlement of spores occurred on the positively charged polymer surfaces. The adhesion strength assay also revealed that the interactions between spores and this surface were relatively strong. The characteristic of spore settlement and adhesion strength on negatively, neutral and zwitterionic charged polymer surfaces were expressed by lower settlement and weak adhesion strength. We also found that Polymer thickness influences the adhesion behavior of marine alga *Ulva.* The optimum polymer thicknesses, where *Ulva* spore settlement is lower, and spore removal is higher, was achieved in the range 200-400 Å. The settlement experiments are complemented with physical characterization of the prepared surfaces, using Fourier-transform infrared reflection-absorption spectroscopic (FT-IRAS), null ellipsometry , atomic force microscopy, and contact angle measurements.

Keywords: Marine biofouling, polymer brush, thickness, charged polymer, *Ulva*, zoospore

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MBA-6

COACERVATION IN MUSSEL ADHESION

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Marine mussels attach to substrates using adhesive proteins. It has been suggested that complex coacervation (liquid-liquid phase separation *via* concentration) might be involved in the highly condensed and non-water dispersed adhesion process of mussel adhesive proteins (MAPs). However, as purified natural MAPs are difficult to obtain, it has not been possible to experimentally validate the coacervation model. In the present work, we showed mussel adhesion mechanism can be explained by complex coacervation phenomenon using several recombinant MAPs. We also found that highly condensed complex coacervates significantly increased the bulk adhesive strength of MAPs in both dry and wet environments. In addition, oil droplets were successfully engulfed using a MAP-based interfacial coacervation process, to form microencapsulated particles. Collectively, we explained underwater adhesion mechanism of mussels using complex coacervation model and a complex coacervation system based on MAPs showed superior adhesive properties, combined with additional valuable features including liquid/liquid phase separation and appropriate viscoelasticity. Our microencapsulation system could also be useful in the development of new adhesive biomaterials, including self-adhesive microencapsulated drug carriers, for use in biotechnological and biomedical applications.

Acknowledgement: This work was supported by the National Research Laboratory program and the Brain Korea 21 program funded by the Ministry of Education, Science and Technology, Korea.

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MBA-7

ADHESION MECHANISM IN A DOPA-DEFICIENT FOOT PROTEIN FROM GREEN MUSSELS

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The holdfast or byssus of Asian green mussels *Perna viridis* contains a foot protein, pvfp-1, that differs in two respects from all other known adhesive mussel foot proteins (mfp): (1) instead of the hallmark L-3, 4-dihydroxyphenylalanine (DOPA) residues in mfp-1, for example, pvfp-1 contains **C**^{2}-mannosyl-7-hydroxytryptophan **(**Man7OHTrp). (2) In addition, pvfp-1 chains are not monomeric like mfp-1 but trimerized by collagen and coiled-coil domains near the carboxy terminus following a typical domain of tandemly repeated decapeptides[1]. Here, the contribution of these peculiarities to adhesion was examined using a Surface Forces Apparatus (SFA). Unlike previously studied mfp-1s, pvfp-1 showed significant adhesion to mica and, in symmetric pvfp-1 films, substantial cohesive interactions were present at pH 5.5. The role of Man7OHTrp in adhesion is not clear, and a Dopa-like role for Man7OHTrp in metal complexation (e.g., Cu^{2+}, Fe^{3+}) was not observed. Instead, cation-π interactions with low desolvation penalty between Man7OHTrp and lysyl side chains and conformational changes (raveling and unraveling of collagen helix and coiled-coil domains) are the best explanation for strong adhesion between pvfp-1 monomolecular films. Strong adhesion explained by cation-π interaction and conformational changes in pvfp-1 suggests new insights to design underwater adhesive.

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MBA-8

CHARACTERIZING OYSTER CEMENT AND MUSSEL GLUE: WHAT IS DIFFERENT AND WHAT IS THE SAME?

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If we want to make antifouling coatings by stopping marine bioadhesion we first need to understand how the organisms stick. Our laboratory is working to reveal the secrets of oyster and mussel adhesives. Experiments with small molecule models, extracted proteins, synthetic polymer mimics, and live animals provide insights on scales ranging from individual molecules up to animal behavior. Some chemical themes may be emerging in marine bioadhesion including oxidative cross-linking of macromolecules. The composition of these materials is also providing us with insights on how each class of animals adapts adhesion for their specific circumstances. For example, mussel glue appears to be soft when compared to harder oyster cement. We are also beginning to see how strong these materials are and why they have the strengths that they do. The latest characterization findings will be presented and placed within a context of general marine bioadhesion.

Acknowledgment: We are appreciative of support for this work provided by the Office of Naval Research.

BIOFOULING CONTROL TECHNOLOGIES-COPPER AND CO-BIOCIDES
Session Chairs: Neal Blossom and Lena Lindblad

CU-1

HIGH PERFORMANCE ANTIFOULINGS BASED ON ORGANOSILYL POLYMERS: RESULTS FROM LONG TERM EXPOSURE UNDER DYNAMIC CONDITIONS

Seamus M. Jackson and Terje Hansen
Jotun A/S

It is now approaching ten years since the widespread use of antifouling paints based on tributyltin (TBT) copolymers ceased. These paints provided excellent, predictable and long term performance at a low cost. Since then new antifouling coatings based on other technologies have been introduced. Most of the major suppliers of antifouling paints are now promoting coatings based on organosilyl polymers. These provide the same excellent, predictable and long term performance associated with TBT copolymer based coatings. The hydrolysis of organosilyl technology is outlined and the reasons for their success discussed. A major factor determining the success is the ability to give long term controlled release and limit the formation of a leached layer at the paint surface. Products using this technology have been on the market for more than twelve years. Results from dynamic testing and full scale commercial applications are presented. The dynamic testing includes products from several paint suppliers. The results from commercial applications for extended periods demonstrate that the leached layer thickness remains stable and this is a key factor determining their success. Test results of new coatings are also shown and potential future developments outlined.

CU-2

EXPERIMENTAL OBSERVATION AND MODELLING OF BIOCIDE RELEASE FROM MODEL ANTIFOULING COATINGS

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Prediction of biocide diffusion is crucial to estimation of efficacy. Their activity and leaching rate in coatings is the main factor governing their effective lifetime. However, diffusion in glassy polymers is a complex and oft-neglected topic. Furthermore, the chemically and physically changeable environment of the ocean and swelling of the polymer increase the complexity of models. As an attempt to model the diffusion of a furan derivative (FD) biocide of interest, existing literature models were adapted to take into account seawater conditions and obtain effective diffusion coefficients. These have been integrated into mathematical models to predict biocide lifetime. A test matrix of antifouling paint coatings including pMMA, an erodible (CDP) commercial binder and a novel trityl methacrylate copolymer, incorporating copper oxide and FD, was subjected to 10 months of natural immersion and 6 months of accelerated rotor immersion tests (17 knots, 25 °C). Fluorescence microscopy and optical microscopy techniques were applied to these coatings before and after immersion, allowing quantification of organic biocide and pigment distribution.

The biocide leached completely from the trityl methacrylate copolymer binder during rotor testing, compared to 35% from the pMMA binder. For pontoon immersions, 61% of the additive was lost from the pMMA coating, and 53% from the CDP. An accelerated loss of FD occurred in the surface of the CDP, due to rosin depletion. In all samples, release of the biocide was inhibited beyond the cuprous oxide front, which was congruent with the leached layer in samples where cuprous oxide release occurred. CDP was the only binder that demonstrated synchronous depletion of both additives, and it demonstrated a good resistance to fouling in immersion trials. Results of the mathematical modelling of the biocide diffusion were in good agreement with the observed data, highlighting in particular the importance of water uptake with respect to biocide diffusion.

CU-3

COLD SPRAY ANTIFOULING PROTECTION FOR POLYURETHANE MARINE SEISMIC STREAMERS

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Matthew J. Vucko, and Rocky de Nys

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Marine seismic surveys are the primary tool used by the oil and gas industry to explore, develop and manage undersea oil and gas deposits. Seismic streamers contain arrays of seismic sensors that are themselves towed in arrays measuring up to 90 km combined length allowing 3D imaging of the substrata. Fouling by pelagic goose barnacles is a severe and costly disruption, reducing data quality and increasing fuel consumption, drag and the likelihood of streamer breakage. The streamer jacket is made of flexible and low surface energy polyurethane, preventing effective coating with antifoulant.

Cold spray is a metal coating process that forms a supersonic spray of metal particles. Using cold spray equipment, we deposited a discontinuous layer of copper particles in polyurethane streamer jackets. The polymer retained its flexibility and did not become electrically conductive, demonstrating the discontinuous sub-surface nature of the treatment. Samples with 22 and 101 g/m^2 copper loading were mounted on frames and suspended 200 mm below the water's surface and 200 mm from the harbour wall at Townsville Yacht Club in tropical northern Australia and monitored weekly.

After 250 days monitoring, no macrofouler attachment had occurred on the high copper loading polyurethane, compared to 57% macrofouler coverage for low copper loading samples and 96% coverage for controls as determined by image analysis. The fouling organisms were barnacles, bryozoans, mussels and polychaetes. Soft fouling is not problematic on seismic streamers, however it is noted that fouling by *Ulva sp.*, a copper-tolerant algae, occurred on all surfaces, with total (*Ulva sp*. plus macrofoulers) coverage ranging from 40% for high copper loading back facing samples to 96% for back facing control samples. These results predict cold spray antifouling will be an effective technique for preventing biofouling of marine seismic streamers.

CU-4

ANTI-BARNACLE PERFORMANCE OF MEDETOMIDINE IN SOFT (EROSIVE) AND SOLID (NON-EROSIVE) COATINGS

Mia Dahlstrom (a,b) Mattias Berglin (a,b) and Hans Elwing (a)

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Medetomidine included in a rosin-based, erosive, coating prevents barnacle colonization in the marine environment (1). In this study we conducted field experiments with both a soft rosin-based erosive coating and a solid non-erosive coating (VC 17 without copper) Methods. Medetomidine was diluted in a commercial rosin-based coating and a solid coating to 0.1 % weight of the paint formulation. After different times of field exposure, the leaking rate of medetomidine from the coatings was determined using HPLC-UV with a reversed phase C18 column. The settlement rate at the painted surfaces was determined at the same time intervals.

Settlement frequency (right-hand y-axis) and release rate (left-hand y-axis) of B. improvisus *on soft, erosive coatings (left) and solid, non-erosive coatings (right).*

Results. *B. improvisus* recruits were present at day 8 on the "soft" and "solid", control coatings, without added medetomidine. The anti-barnacle effect was achieved only on the medetomidine containing soft coatings but barnacle colonization was not inhibited on the solid coatings (see fig) despite the higher leaking rate of medetomidine from the solid coatings. Conclusions: Medetomidine-mediated settlement inhibition at soft coatings is not a function of release rate of medetomidine since the release rate was much higher, from the solid coatings but without any effect on barnacle colonization. A plausible explanation to the difference may be that medetomidine, which have a high affinity for water/solid interface, is concentrated at the hydrated erosion layer of the soft coatings and that this layer is deterrent to the cyprid larvae. Such an erosion layer is not present non-erosive solid coating.

Dahlstrom M et al (2000) ¸ BIOFOULING 16, 2-4 , 191

CU-5

THE ROLE OF ORGANISM ADAPTATION AND ACCLIMATION IN MANAGING COPPER EFFICACY AND TOXICITY

Kevin W J Long

Regulatory Compliance Limited

Copper is a significant tool in the management of non-native aquatic invasive species, being the most commonly used component added to biocidal antifouling paints in order to prevent settlement of organisms on the hull surface, thus minimising the potential for species translocation. While recent developments in the understanding of the role of copper chemistry on its toxicity have answered many of the questions regarding impact of this use on the wider receiving environment, concerns have also been directed towards the development of copper tolerant species and their impact on local ecosystems. The issue of copper "tolerance" will be examined in respect of the existing understanding of organism adaptation and acclimation to varying levels of this naturally occurring essential element.

CU-6

A UNIFIED SALTWATER-FRESHWATER BIOTIC LIGAND MODEL TO PREDICT THE OLFACTORY EFFECTS OF COPPER TO SALMONID FISHES

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Recent studies have demonstrated short-term exposures to copper (Cu) concentrations as low as 1-2 µg/L in dilute laboratory waters can cause olfactory impairment in freshwater fish, which may limit their ability to migrate or detect predators. Olfactory impairment is of particular concern in the Pacific Northwest because many populations of migratory Pacific salmon and trout (*Oncorhynchus* spp.) are listed as threatened or endangered. However, water chemistry matters in determining lethal and sublethal effects of metals to aquatic organisms. For example, despite effects at low Cu concentrations in dilute laboratory waters, we previously demonstrated that the USEPA's BLM-based freshwater aquatic life criteria for Cu will protect against olfactory impairment across a wide range of fresh waters; and we developed a biotic ligand model (BLM) to predict olfactory effects of Cu to salmonid fishes in fresh waters. However, no olfactory-impairment studies have been conducted with fish exposed to Cu in salt water, leading to high uncertainty in risk assessments. Moreover, that lack of olfactory studies has impeded development of a saltwater olfactory BLM for Cu that would be analogous to the freshwater olfactory BLM. To address this situation, we propose a unified BLM to predict the olfactory effects of Cu to salmonid fishes across a wide range of both fresh and salt waters. Because of the relatively high concentrations of Na^{+} and Ca^{2+} in saltwater compared to freshwater, the competition of those ions with Cu for binding to olfactory tissue should generally cause the threshold-effects concentrations for dissolved Cu to be even higher in salt water than in fresh water. However, this unified olfactory BLM awaits empirical testing before it might be accepted for regulatory use.

Acknowledgements: This work was funded by the Copper Development Association, Inc. and the International Copper Association Ltd.

CU-7

EFFECTS OF COPPER ON THE OLFACTORY AND BEHAVIORAL RESPONSES OF SALTWATER FISH, AND THE PROTECTIVENESS OF REGULATORY AQUATIC LIFE CRITERIA USING THE BIOTIC LIGAND MODEL

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Several studies have evaluated the effects of copper on olfactory function and/or behavior in freshwater fish, including the freshwater life stages of several species of salmonids (e.g., salmon, trout, and whitefish). These studies have demonstrated that olfactory impairment is a more sensitive endpoint than the common acute lethality endpoint. However, the data available to-date indicate that ambient water quality criteria (AWQC) for copper in freshwater are protective against olfactory impairment in fish, particularly when the AWQC were derived using the freshwater biotic ligand model (BLM). With the pending release of draft BLM-based AWQC for copper in saltwater, toxicity data on the olfactory and behavioral effects of copper in saltwater fish species were reviewed. Overall, no olfactory and limited behavioral copper toxicity data were identified for saltwater fish species, and none of the water chemistry data from those studies included dissolved organic carbon (DOC) concentrations, which is one of the most important parameters influencing copper bioavailability. Accordingly, it was necessary to estimate DOC concentrations in the source water and use other approaches for evaluating whether draft saltwater BLM-based copper AWQC are protective against olfactory impairment and behavioral effects. The analyses to-date indicate that the draft AWQC are protective of these endpoints in fish. If proposed research on the effects of copper on olfactory impairment in saltwater life stages of Pacific salmon is conducted, those results will be used to validate a recently developed unified freshwater-saltwater olfactory-based BLM and further evaluate whether saltwater BLM-based AWQC are protective against olfactory impairment in saltwater fish.

Acknowledgements: This work was funded by the Copper Development Association, Inc. and the International Copper Association Ltd.

CU-8

ECO-FRIENDLY SURFACE CHEMISTRIES FOR THE INHIBITION OF BIOFOULER GLUE POLYMERIZATION

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An evolving mechanistic understanding of biofouler adhesion processes prompted us to develop novel polymer coatings that interfere with bioadhesive processing and/or curing. Our initial focus on marine hard foulers led us to create coatings decorated with chemical functionalities known to inhibit enzymes important for the polymerization of biofouler cement. Polymerization of novel cross-linkable monomers possessing enzyme-inhibiting functionalities resulted in some durable, non-toxic coatings which prevented the settlement of barnacle cyprids in the lab and in field studies. In order to validate a molecular mechanistic basis for the anti-settlement results, we sought to visualize the interactions of a mimetic model enzyme with our functionalized polymers coated onto a surface. This was achieved by measuring the binding of bioactive enzyme to functionalized and non-functionalized surface coatings. Functionalized coatings with cyprid anti-settlement properties also bound the model enzyme while the non-functionalized coatings did not. This suggests the anti-settlement effect correlates with our molecular target-based and mechanism-based hypothesis. Our findings provide a rational basis and opportunity for developing high throughput screening and optimization technologies for antifouling coatings analogous to those employed by the pharmaceutical industry (and in our lab) for drug discovery and development programs.

BIOSECURITY/BIOINVASIONS
Session Chair: Eric Holm

BIO-1

MAKING POLICY SAUSAGE: A BEHIND-THE-SCENES LOOK AT THE DEVELOPMENT OF BIOFOULING MANAGEMENT REGULATIONS FOR THE STATE OF CALIFORNIA

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The transformation of good science into effective marine policy is an underappreciated art form. This is especially true of state-level regulations that establish requirements for a global industry such as shipping, where a variety of stakeholders have a variety of priorities and voices, all of which are important. Weaving together all of these competing priorities, including good science, technological capabilities, pressures from the regulated industry, and political realities is an interesting and oftentimes frustrating endeavor. California's development of biofouling management regulations is presented here as an example of this process. In 2007 the California Legislature directed the State Lands Commission to develop regulations governing the management of biofouling in California. In the four years since, the Commission's Marine Invasive Species Program (MISP) has been working to satisfy this mandate and develop protective and achievable regulations. This endeavor began with efforts to fill key information gaps through the funding of targeted research and the annual collection of information on hull husbandry practices and voyage characteristics from the California fleet. Ultimately, this information was integrated into discussions of a technical advisory group (TAG) to inform the development of the regulations. The TAG process was essential, particularly in this instance where there were no existing models for holistic biofouling management regulations. Especially important was collaboration with international partners from Australia, New Zealand, and the International Maritime Organization, where biofouling management regulations and guidelines were being developed simultaneously. In essence, the development of California's biofouling management regulations became a group project and once complete, it will be a group success. This presentation will provide an honest look at the entire regulatory development process, from the initial discussion of the current science to the compromises necessary in today's political climate.

BIO-2

BALANCING BIOFOULING, BIOCIDES AND BUREAUCRACY

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In-water cleaning has been proposed as a tool for both routine hull maintenance and managing the risk associated with international vessel arrivals. However, in-water cleaning poses environmental risks that require consideration:

- the release and accumulation in the marine environment of chemical contaminants from antifouling coating(s);
- the release of non-indigenous species into new environments.
- Research was commissioned to investigate the following questions relating to in-water cleaning:
- What options are available to manage biofouling risk in New Zealand?
- Do the scenarios within the OECD Emission Scenario Document for antifouling products apply to New Zealand conditions using the MAMPEC model?
- When do the environmental costs of non-indigenous species and chemical contaminants, released by in-water cleaning, outweigh the risk of no action?

There are limited options available to manage biofouling on vessels entering New Zealand; these include no action, provision of educational materials and warnings, haul out and waterblasting (recreational and fishing vessels) and refusal of entry. Alternative technologies, such as in-water cleaning, although promising require further research.

The MAMPEC model predicted antifouling emissions from New Zealand ports to be lower than the OECD default values. However, predicted antifoulant concentrations are markedly higher in New Zealand ports than the OECD default. Higher antifoulant emissions and ambient concentrations were predicted in many of the New Zealand marinas compared to the OECD default.

The emission of biocides following the use of in-water cleaning technologies was able to be estimated and input to the New Zealand specific scenarios within the MAMPEC model. However, a range of variables have been identified that make any estimate of relative risk from different cleaning scenarios subject to uncertainty. These variables include type of fouling present, organism biology, the effects of cleaning on material released, and the nature of the receiving environment at the time of cleaning.

Acknowledgement - The authors thank Depree C, Gadd J, Floerl O, Hickey C, Inglis G,

Lewis J, Morrisey D, Page M, Woods C.

BIO-3

BALANCING FOULING CONTROL STRATEGIES FOR MANAGING INVASIVE SPECIES AND WATER QUALITY: THE ROLE OF HULL COATINGS IN RECRUITMENT

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Management of fouling on boat hulls continues to be an important issue worldwide, affecting boat operations and influencing the spread of non-native invasive species (NIS). Copper-based antifouling paints are often used to retard fouling growth. Water-quality impacts of these toxic paints have led to increased restrictions on their use in some areas. Such restrictions address water quality concerns but neglect important implications for NIS transport. To better understand potential implications of using non-toxic hull coatings, instead of toxic copper-based paints, we examined the response of fouling organisms to different hull coatings over short (1 month) and long (3, 6, 12 month) submersion-time periods. Our hypotheses were: 1) hull coating type affects recruitment of fouling organisms, and 2) NIS settle sooner and occupy more space over time than native species on surfaces coated with toxic, copper-based antifouling paint. Recruitment of fouling organisms on fiberglass panels was measured using percent cover and counts.

Copper antifouling paint inhibited settlement of most, but not all, fouling organisms over moderate (less than 6 months) time periods even when larvae were abundant. The few exceptions suggest copper tolerance and coating selectivity for certain NIS. On panels with toxic paint, NIS appeared sooner and occupied more space than native species. For panels with non-toxic coatings, recruitment was similar except that the non-native bryozoan, *Watersipora subtorquata*, had higher recruitment on epoxy and slick coatings, and the tube worms, *Hydroides* spp., had higher recruitment on epoxy coatings. Taken together, the risk of potential NIS spread is higher for non-toxic hull coatings, but risks also exist for copper-based paints especially over longer time periods. Thus, companion practices, such as hull cleaning, are needed for all boats regardless of hull coating. Our findings illustrate the importance of considering both water quality and NIS transport when developing fouling control strategies.

BIO-4

BALANCING FOULING CONTROL STRATEGIES FOR MANAGING INVASIVE SPECIES AND WATER QUALITY: THE ROLE OF HULL CLEANING IN RECRUITMENT

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Toxic antifouling paints have typically been used to deter recruitment of organisms on boat hulls. Concerns have been raised about their impacts on water quality. Further, recent data indicate that copper-based paints do not deter all fouling, particularly over time. They may select for copper-tolerant species, primarily non-native invasive species (NIS). In-water hull cleaning may help reduce water quality impacts and minimize NIS spread. However, efficacy of hull cleaning practices has been little studied, with some indications that certain methods may increase, not decrease, fouling.

We evaluated hull cleaning best management practices (BMPs) used in California and recruitment patterns at two study sites to test two hypotheses: 1) California BMPs do <u>not</u> stimulate recruitment of fouling organisms and 2) recruitment of fouling organisms differs among study sites and seasons For the first study, we assessed the biomass of fouling organisms accumulating on experimental panels with toxic and non-toxic coatings that underwent three treatments; not cleaned (new), cleaned once or continually cleaned using the California BMPs. For the second study, we evaluated monthly recruitment of fouling organisms among locations and over time on similar panels for one year. Recruitment to experimental panels was assessed using point contact methods and counts.

Fouling biomass accumulation was not significantly affected by cleaning treatment. However, significantly less fouling occurred on panels with toxic coatings than non-toxic coatings. Further, recruitment was affected by location and time of year, with significantly less fouling at the northern site and during the winter. Our findings support using California hull-cleaning BMPs, while considering location and season when determining cleaning frequency. Such BMPs represent a balanced approach for managing fouling on boats while minimizing impacts to water quality and reducing risks of spread of NIS.

BIO-5

MILITARY SEALIFT COMMAND VESSELS: AN IMPORTANT CONTRIBUTION TO THE BIOFOULING VECTOR

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Hull fouling is a key vector for the spread and introduction of marine species. Recent studies have used in-water surveys to demonstrate the real-time and real-life risk of NIS introductions via hull fouling. To date, military vessels have been a distinct exemption from these studies. During summer 2010, we surveyed six Military Sealift Command vessels and eight commercial container vessels whilst docked in Apra Harbor, Guam. Questionnaires to determine hull-husbandry characteristics and voyage histories were followed by in-water surveys to examine biofouling extent and composition. While hull husbandry practices of both in-service and commercial vessel types were similar, the voyage histories and biofouling communities were quite distinct. The long in-port durations and slow voyage speeds of MSC vessels enable the accumulation of extensive and diverse fouling communities, which are not limited to niche areas. Accompanied with the wide-spanning range of these vessels, their importance for the transport of diverse accumulations of fouling organisms is of concern.

BIO-6

DETERMINING BIOFOULING RATES ON A MARINE CONSTRUCTION FLEET WHICH INCREASE OUR PREDICTABILITY OF BIOFOULING AND THE POSSIBLE PRESENCE OF MARINE PESTS

Greg Booth

Chevron Australia

The Gorgon Project entails the development of a natural gas processing facility on Barrow Island, a globally significant nature reserve and marine protected area off the northwest coast of Australia. A key commitment of the Gorgon Project, Australia's largest resource project, is that no marine pests be introduced to the waters surrounding Barrow Island. The objective of this presentation is to address the question: "How long can vessels remain in water and still be considered free of secondary biofouling which could provide a niche for marine pests to attach, survive and reproduce in the protected waters surrounding Barrow Island?" This question is ecologically and economically significant for projects and fleet managers that require access to ecologically sensitive areas where their presence are closely monitored by the authorities and the community stakeholder groups and where the introduction of a marine pest may result in significant financial and reputation impact to project an fleet managers.

A comprehensive marine pest strategy supports the core commitment that all vessels mobilising to Barrow Island will be free of secondary biofouling (calcareous marine growth) at the time of mobilisation. Once free from secondary fouling, the rate of fouling development is of strategic importance in managing costs and risk. A range of vessels from a fleet of more than 180 vessels, including offshore support vessels, landing craft, dredges, barges, utility vessels and crew transport vessels, was considered to study biofouling rates on the project. The results of 20 vessel inspections were analysed. An unexpected pattern of randomness emerged which contradicts the classic succession paradigm. This result has significant implications for all vessel owners that work in marine sensitive areas. It also is an important indication that the current risk assessment methodologies under-estimate the risk the vessels present, more so when vessels operate in marine protected areas.

BIO-7

SETTING A NEW BENCHMARK FOR RISK ASSESSING MARINE VESSELS WHICH OPERATE IN ECOLOGICALLY SENSITIVE ENVIRONMENTS

C. Johann van der Merwe

Chevron Australia

Result from inspections on a large number of marine construction vessels participating on the Gorgon Project, one of the largest resource projects in the world, indicates that the prevaling risk assessment methodologies for marine vessels under-estimate the risk of marine pests establishing on marine vessels. It was also established that an infection event is more a stochastic event and not as deterministic as traditionally anticipated. This paper will present a new risk assessment methodology that goes beyond the classic approach to risk assessment that uses the proven likelihood and consequence approach as it falls short of anticipating this stochasticity.

In addition, the establishment of marine pests and other species of concern on the construction vessels of the Gorgon Project occur at a frequency which are significantly greater than previously understood but more importantly, at earlier stages than previously appreciated. In the marine pest strategy, the fleet remains under continuous risk tracking the risk status on a very fine scale, depending on the schedule parameters for each vessel. Qualitative risk values were weighted which assessed infection probability, survival, detectability and eradication on a 10 point scale ranging in values from an extremely remote chance of a marine pest being present to a marine pest being present every time. To date, with more than 180 vessels working in the limited confines of this marine protected area and with more than 2000 shipments mostly from ports where marine pests occur either naturally or as introduced species, no evidence exists of any marine pests being translocated to this sensitive environment.

As the frontier pushes in more remote and sensitive environments in our pursuit of resources a new benchmark in mobilizing marine vessels is urgently required to safeguard the sensitive environments. The Barrow Island experience can contribute to setting this new benchmark.

BIO-8

NOVEL FIELD EXPERIMENTS INVESTIGATING THE EFFECT OF ORGANISM SIZE AND WATER TEMPERATURE ON TREATMENT OF MUSSEL BIOFOULING IN INTERNAL SEA WATER SYSTEMS USING QUATERNARY AMMONIUM COMPOUNDS

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Vessel internal seawater systems (e.g. sea chests, sea water piping) remain one of the more challenging areas for the effective control of marine biofouling. Mussels are arguably one of the most problematic fouling taxa affecting ship internal sea water systems worldwide. In recent years, there have been several instances where the growth of mussels in the internal sea water system of Royal Australian Navy (RAN) vessels has resulted in significant disruptions to vessel operations and schedules, as a result of both operational and mechanical issues (e.g. restricted water flow to onboard cooling systems) and biosecurity concerns (e.g. discovery of nonindigenous mussel species following overseas deployment). Apart from physical removal, the primary method for treating mussel fouling of the internals of Navy vessels is to flush the affected system with a 1% (in sea water) detergent solution containing quaternary ammonium compounds (QACs; active ingredient benzalkonium chloride). Parameters for the application of this treatment are based on previous research on the effects of QACs on bivalve species. However, much of this research has been conducted at small-scales under controlled laboratory conditions, and has never properly addressed the influence of organism size and water temperature on the efficacy of QACs as a bivalve treatment method. This talk presents the findings of research examining the efficacy of QAC solutions for treating mussel biofouling, conducted under realistic field conditions using experimental sea water piping systems (including replica sea chests). We discuss the influence of treatment solution concentrations, mussel size and water temperature on the mussel mortality.

Plenary 4

PLENARY-4

MARINE ANTIFOULING: PROGRESS AT THE INTERFACE OF DISCIPLINES

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There are at least three historical truisms of marine antifouling research: 1) the field is multidisciplinary in scope; 2) colonization stages of fouling organisms are the target; and 3) a strategy that utilizes toxicity is best for broad-spectrum efficacy. Contemporary research on nontoxic solutions, including fouling release, to mitigate marine biofouling is more likely to be transdisciplinary, and the colonization stage of fouling is not necessarily the focus of study. Although it is debatable whether a practical, broad-spectrum, biocide-free antifouling solution is achievable, it is certainly a major challenge and one that requires a greater understanding of the biological and physico-chemical factors modulating the settlement of fouling organisms. This presentation will discuss examples of how our understanding of settlement has advanced in recent years, through the unprecedented application of new methodologies and analytical tools. An inter-/transdisciplinary approach will continue to be needed to address areas where our understanding of settlement remains poor, including the relative importance of behavioural and physical modes of surface rejection in flow, and the molecular mechanisms regulating settlement.

GENERAL ASPECTS OF FOULING: MICRO FOULING
Session Chair: Sergey Dobretsov

MICRO-1

DOES A BACTERIAL BIOFILM INFLUENCE THE ADHESION STRENGTH OF MARINE ALGAE?

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Previous studies in our laboratory have shown that a natural bacterial biofilm enhances the settlement of spores of the green algae *Ulva*[1,2,3]. However, the effect of such biofilms on the adhesion strength of algae, and how the interaction may be influenced by coating properties, is not known. For marine invertebrates, one study has shown that a natural bacterial biofilm on glass increased adhesion strength of the larvae of most species tested[4].

In the present study, the effect of a natural bacterial biofilm and 2 single-species biofilms (*C. marina* and *M. hydrocarbonoclasticus*) on the adhesion strength of *Ulva* was investigated. The settlement of spores and the adhesion strength of both spores and sporelings (young plants) were evaluated for 4 different surfaces: Intersleek® 700 (IS700), Intersleek® 900 (IS900), unpigmented poly(dimethylsiloxane) (PDMS) and glass. Bacterial biofilm density was quantified by staining and image analysis. Spore density and biomass of sporelings was quantified by auto-fluorescence measurements and their removal was evaluated using a water channel that generated a wall shear stress.

The results obtained showed that natural biofilm and the *M. hydrocarbonoclasticus* biofilm enhanced the settlement of *Ulva* spores on all the test surfaces, while for the *C. marina* biofilm, no consistent effect was obtained. The adhesion strength of sporelings was significantly enhanced by the presence of a natural biofilm on all test surfaces whatever the surface properties. On the other hand, biofilms of *M. hydrocarbonoclasticus* and *C. marina* reduced the adhesion strength of sporelings on some of the surfaces tested (PDMS and IS700).

The presentation will also explore the spatial relationship between marine algae and bacterial biofilm using confocal microscopy, the effect of biofilm density and age on algal adhesion strength, and the effect of a bacterial biofilm on the settlement and the adhesion strength of the diatom *Navicula* sp.

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[1]Joint *et al.* (2000), Biofouling, 16: 151-158 ; [2]Joint *et al.* (2002), Science, 298: 1207 ; [3]Patel *et al.* (2003), Environmental Microbiology, 5: 338-349 ; [4]Zardus *et al.* (2008), The Biological Bulletin, 214: 91-98.

MICRO-2

GREEN INTELLIGENT MATERIALS: DEVELOPING NEW SURFACES WITH ANTI-BIOFILMS PROPERTIES

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Green Intelligent Materials is a three years INTERREG project aiming to set up a trans-Channel Centre for the elaboration of green intelligent "smart" biomaterials.

The functionalization of surfaces by biocidal molecules, active against biofilms, is a challenge for the development of biomedical materials and new micro-organisms barrier materials for applications to packaging and marine antifouling surfaces.

We plan to develop strategies for the functionalization of polymeric materials by using biomolecules from marine origin, in order to modify the interactions between micro-organisms and these polymer surfaces. Our actions target both the support and the cell and we work on polymer by modifying their surface and studying the influence of the modifications on micro-organism adhesion. During the first year of the project we have focused on preventing the growth of micro-organisms by using natural biocides or anti-biofilm molecules . Algae have the ability to produce a large variety of chemical defences which prevent over-predation, defence against competition from foreign microorganisms or changes in environmental conditions .We have tested the anti-fouling and anti microbial activity of seventeen purified extracts of macroalgae, among these extracts three shows promising results and will be presented.

MICRO-3

SUBSTRATE BACKGROUND COLOUR AFFECTS FORMATION OF MICRO- AND MACRO-FOULING COMMUNITIES

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Previous investigations demonstrated the effect of substrate colour on recruitment of larvae of barnacles, abalones, pearl oysters, ascidians and spores of algae. Here, we investigated the effect of the substrate background colour on formation of microbial and macro-fouling communities. Acrylic panels were painted with black and white colours and were covered with transparent sheets in order to insure similar surface properties. All substrata were exposed to biofouling at 1 m depth for 40 days in Marina Bandar al Rowdha (Muscat, Sea of Oman). Development of macro-fouling communities was assessed using a dissecting microscope whereas the abundance of bacteria and phototrophs were assessed by direct microscopy and chlorophyll *a* extraction and quantification. Changes in composition of microbial communities were analyzed using 454/Roche pyrosequencing of 16S rRNA gene tags. Bacterial counts and chlorophyll *a* concentrations increased with time, but no effect of colour was found. However, there were drastic changes in the composition of bacterial communities as revealed by molecular tools. Macro-fouling communities changed with time resulting in different communities on the white and black panels. SIMPER analysis showed that *Folliculina* sp., *Ulva* sp. and *Balanus amphitrite* were the major species that contributed to the dissimilarities between the communities. The highest densities of macro-fouling species were found on black panels, while species richness was higher on white panels. Our results emphasized the importance of substrate colour in the formation of fouling communities that need to be taken into account in future antifouling studies.

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MICRO-4

THE EFFECT OF STATIC AND DYNAMIC CONDITIONS ON THE SETTLEMENT AND ADHESION OF DIATOMS TO SHIP HULL COATINGS

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Ship hull coatings experience both static and flowing seawater conditions; however, most *in situ* diatom studies are conducted under static conditions. This does not replicate the dynamic conditions that the ship hull and subsequent fouling organisms encounter. To compare diatom settlement and adhesion on antifouling coatings under static and dynamic conditions, four commercial antifouling coatings (one copper ablative and three fouling release systems) along with two controls (epoxy and Dow Corning 3140) were deployed on a rotating drum and under static conditions at the same test site. Biofilms were sampled after two weeks and analyzed for diatom composition and abundance. Biofilm and soft fouling adhesion was measured using the calibrated water jet method. Results show a difference in the adhesion strength and community structure of biofilms exposed to the different immersion treatments (static versus dynamic). Additionally a significant difference was seen in the performance of the commercially available coatings with regards to percent cover, diatom composition, and adhesion strength.

Acknowledgments: This research is funded by the Office of Naval Research Grant # N000141010919.

MICRO-5

BIOFILM DISPERSAL, NITRIC OXIDE AND CONTROL TECHNOLOGIES FOR BACTERIAL FOULING

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Bacteria form biofilms on almost all surfaces, ranging from ship hulls to water treatment membranes to indwelling biomedical devices. Therefore, there is a strong drive to understand the processes of biofilm formation, either to develop technologies that eliminate biofilm formation in industrial processes and human health, or, in some instances, to encourage their formation, such as in bioremediation. One recent advance in our understanding of biofilm biology is the realization that biofilms have a "life cycle" analogous to many marine invertebrates or algae, which includes a dispersal phase in which much of the biofilm biomass becomes detached from the surface[1]. This raises the potential of enhancing biofilm dispersal as a useful antifouling technology. We have investigated this possibility using the small signaling molecule nitric oxide (NO), which plays a key role in the endogenous regulation of biofilm dispersal in a number of bacterial species. Exposure to low, non-toxic concentrations of NO changes gene expression and induces the transition from a biofilm to a planktonic mode of growth in many bacterial species. Thus the use of NO, either *via* chemical compounds releasing NO in solution (NO donors) or *via* coatings that can be applied on surfaces and generate NO, may provide a low cost and environmentally safe solution to biofilm control. Our research has identified NO donors suitable for dispersing biofilms on reverse osmosis (RO) membranes, and that also when used as a combination treatment increased the efficacy of a standard industrial disinfectant.

[1] D. McDougald *et al.* (2012) Should we stay or should we go: mechanisms and ecological consequences for biofilm dispersal. *Nature Reviews Microbiology* 10: 39-50

MICRO-6

FOULING DIATOM COMMUNITY SPATIAL STRUCTURING ON IN-SERVICE INTERSLEEK 900

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Microfouling and the drag penalty it incurs to a vessel is a topic receiving focused interest and reducing "slime" fouling is becoming a recognized industry priority. A specific microfouling objective of the research labs at International Paint, Ltd is to devise a method to assess fouling-control coating in-service performance against biofilms in a meaningful way that generates data with a resolution fine enough to inform future development work. As part of this effort we are looking at how microfouling diatom communities are structured across vessel hulls. We present preliminary data on the fouling biofilms collected from the hull of a temperate/sub-tropical waters fast ferry coated in Intersleek 900. The vessel had last dry docked one year prior to the collection date. We sampled the biofilm at several points on the 190m horizontal transect along the vertical sides and down a vertical transect of the 6.5m draft. Samples were scraped from the side of the vessel and preserved in alcohol, then processed in the lab to isolate the silica diatom frustules. Cleaned frustules mounted in Naphrax were enumerated by light microscopy, and diatom species identified by morphology where taxonomic naming was not possible. The preliminary data suggest a vertical gradient in diatom community structure. This is not surprising as the marine euphotic zone is so often characterised by vertical gradients. Further results and implications for performance assessment will be discussed.

MICRO-7

MOLECULAR SIGNALS AND METABOLITES FROM ALGAE HAVE A POTENTIAL ROLE IN MARINE MICROFOULING CONTROL

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Roseobacters and marine algae commonly interact in the marine environment and engage in intermittent surface-associated relationships. Algal-derived compounds have been shown to be colonization signals for roseobacters1, and we have further explored the chemical cascade that mediates this symbiosis to identify compounds of interest in biofouling. Here we show that common breakdown products of lignin that are more abundant than previously believed in the ocean and have antimicrobial properties. Lignin itself has only recently been identified in a macroalgae2. We have employed a bioinformatics approach for to show that the genes required for its synthesis are present in a surprising diversity of marine microalgae. This suggests that breakdown products / precursor metabolites of lignin may be a common antifouling defense for marine phytoplankton.

In addition, these metabolites stimulate the roseobacter *Phaeobacter gallaeciensis* BS107 to produce a potent antialgal compound with an unusual structure3. This finding indicates that compounds produced by algae act as a signal and regulator of small molecule production in BS107. Nontoxic breakdown products of common metabolites from marine organisms (such as coumaric acid from lignin) could act as coatings in the marine environment if they are effective in deterring colonization through their antimicrobial properties and by stimulating production of inhibitory small molecules.

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2. Martone PT, Estevez JM, Lu F, Ruel K, Denny MW, Somerville C, Ralph J. (2009) Discovery of lignin in seaweed reveals convergent evolution of cell-wall architecture. *Curr Biol.* 19(2): 169-75.

3. Seyedsayamdost MR*, Case RJ*, Kolter R, Clardy J. (2011) The Jekyll-and-Hyde chemistry of *Phaeobacter gallaeciensis*, *Nature Chemistry*, 3: 331-335.

MICRO-8

ANTIFOULING COATINGS: *IN VITRO* BIOASSAY *VS* MARINE BIOFILM COMMUNITIES

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When immersed in sea water, any substrate would be rapidly colonized by micro and then macroorganisms. This complex and sequential natural process called biofouling induces economical and ecological prejudices, especially talking about ship hull or aquaculture nets (e.g. Schultz et al. 2011).

Following the development of a bioassay aimed to assess the anti-adhesive potential of molecules (Camps et al. 2011) and coatings, a first comparison of the results of these bioassays with the structure of *in situ* biofilms colonizing the same antifouling coatings in Toulon harbour (North-Western Mediterranean Sea) was performed. Immersion was performed in July during one month to get mature biofilms beyond pioneer stages (Briand et al. 2012). Complex biofilm communities were described, in term of both abundance and diversity, using flow cytometry, microscopy and PCR-DGGE on six different coatings including the poly(vinyl chloride) (PVC), four Self-Polishing Coatings and one Fouling Release Coating. Coatings without biocides (including the reference) showed higher densities than biocidal paints, whatever the groups of microorganisms (bacteria, cyanobacteria, pico- and nanoeukaryotes, diatoms). Significant variations for both abundance and diversity were observed between the coatings depending on the microorganism groups. Despite the low number of tested coatings and the lack of coatings with intermediate efficacy, when compared to *in vitro* data (% of adhesion for 3 marine bacterial strains in microplates), quantitative analyses suggested a coherence between the two approaches.

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MICRO-9

DIVERS, BOATS AND FLYING MACHINES REMOTELY OPERATED VEHICLES AS AN ALTERNATIVE TO DIVERS FOR MARINE VESSEL INSPECTIONS

Rae Burrows and Justin McDonald

WA Dept of Fisheries – Policy and research groups

The Department of Fisheries is the lead agency for managing the introduction and spread of harmful aquatic organisms into and around Western Australia. Western Australia is experiencing exponential growth in the exportation of natural resources and as a consequence unprecedented numbers and diversity of high risk vessel types and infrastructure are visiting Western Australia. Naturally, there is a greater risk of such vessels introducing harmful aquatic organisms, particularly Invasive Marine Species.

Vessel biofouling in Western Australia is largely being managed through State Ministerial Conditions via the Environment Protection Act 1986. Most Ministerial Conditions require vessels/infrastructure that is associated with a particular project to be "free of Invasive Marine Species". This usually requires the employment of Commercial Divers to inspect vessels to ensure they pose a low risk of introducing Invasive Marine Species into Western Australian Waters. However, there are Occupational Health and Safety issues associated with certain diving inspections and the cost of using Commercial Diving Companies is rather expensive.

As part of their research DoF evaluated cost-effective alternative inspection methods to diving that are capable of conducting biofouling inspections effectively and efficiently. One such approach was using Remotely Operated Vehicles (ROV)s as an alternative to commercial divers. In the trial mimic pests were deployed on vessels and two ROV types were trialled against divers under a range of conditions. The study examined:

- the effectiveness of ROVs and divers in inspecting all submerged areas of vessels, including niche areas;
- the effectiveness of the ROVs' and divers' capability of providing sufficient imagery to enable recognition/identification of IMS;
- whether ROVs are capable of collecting samples and if so, how effectively; and
- the benefits and costs (time and financial) associated with using ROVs relative to using experienced commercial divers when inspecting vessels.

GENERAL ASPECTS OF FOULING: MISCELLANEOUS
Session Chair: Elizabeth Haslbeck

MISC-1

A HARD AND GUMMY PROBLEM: UNDERSTANDING VARIATIONS IN BARNACLE CEMENT PROPERTIES

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**NRC/NRL Postdoctoral Associate*

Barnacle cement can vary in a number of different ways, and in order to fully understand barnacle adhesion, these variations must be taken into account. This talk will focus on two common, bulk-scale variations referred to as “hard” and “gummy” cements.

Visually, hard cement is a thin, clear adhesive, and gummy cement is thicker and white. Gummy cement production is typically observed with problematic adhesion on silicone substrates. The trait is heritable1 and can occur at repair and reattachment points at the adhesive interface.2 Results presented will provide a greatly expanded analysis of the comparative properties and origins of hard and gummy cements. This will include more definitive characterizations of hard and gummy cements in terms of biochemical structure, water content, and mechanical properties. New insights into gummy cement production are also provided by in situ FTIR spectroscopy of buried adhesive interfaces on silicone coatings, as well as X-ray tomography of barnacle shell structure. Discussion of the results will include how they relate to the bigger picture of barnacle cement production and curing.

1 Holm, et al., *Biofouling* 21, 121 (2005).

2 Saroyan, et al., *Biological Bulletin* 139, 333 (1970).

MISC-2

PHOSPHOPROTEINS IN BARNACLE GLUE

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The adhesive properties of highly phosphorylated proteins have long been known. For example, casein, a highly phosphorylated milk protein, has been used as glue for over a hundred years. Phosphate containing compounds are used as adhesion compounds in paints, dental adhesives, and within marine systems, phosphoproteins are just emerging as a common theme used by invertebrates for adhesion. Phosphorylated proteins have been identified in the adhesives of marine mussels, tube worms, Cuvierian tubules of sea cucumbers, and oysters. These observations led us to hypothesize that phosphoproteins may be present in barnacle glue and could contribute to its adhesive and cohesive properties. In addition, phosphoproteins have the potential to be involved in mineralization of the glue by inducing mineral nucleation. To test our hypothesis we stained uncured barnacle glue, separated by SDS-PAGE, using a phosphoproteinspecific stain (Invitrogen Pro-Q Diamond). We identified multiple phosphoproteins at approximately 125, 100, and 80 kDa. We found that the fluorescence intensity of the 80 kDa phosphoprotein varied among individual barnacles. These variations are likely due to molting or other biological cycles, consistent with previous observations of changes in glue production during the course of the molting cycle. Such variation in phosphoproteins may suggest significant differences in the adhesive and/or mineralization properties of glue secretions over time. Our current work focuses on understanding the biochemistry and function(s) of phosphoproteins in barnacle adhesion. The authors gratefully acknowledge support from the ONR Coatings/Biofouling program.

MISC-3

BIO-INSPIRED ANTIFOULING VIA ACTIVE SURFACE DEFORMATION

Xuanhe Zhao

Soft Active Materials Laboratory, Department of Mechanical Engineering and Materials Science, Duke University

Gabriel Lopez, Department of Biomedical Engineering, Duke University

Daniel Rittschof, Duke Marine Laboratory, Duke University

Biofouling of ship hulls and propellers increases drag and power usage and decreases fuelefficiency. Biofouling costs the US Navy alone approximately one billion dollars per year, and the decreases in fuel efficiency further increase green-house gas emissions. Traditional antifouling coatings, relying primarily on biocidal organics and metals, have negative environmental impacts, while newer polymer-based coatings are easily damaged and ineffective in long-term applications. On the other hand, nature has created an enormous number of biological surfaces that can effectively clean themselves via active deformation and motion. For example, tiny hairs called cilia on the surfaces of respiratory tracts constantly move back and forth, pushing inhaled foreign particles out of our lungs. The ciliary cleaning has also been used by molluscs, corals and many other marine organisms for active antifouling.

Inspired by active biological surfaces found in nature, we have developed a novel activeantifouling technology by harnessing on-demand deformation of polymer coatings in response to external stimuli. We discover that the surfaces of silicone-based coatings can be significantly deformed by applying a direct-current voltage across the coatings. The deformation is ondemand, dynamically switching the coating surfaces between deformed and flat states as the applied voltage is on and off (Fig 1A). The on-demand deformation can actively and effectively detach various biofouling organisms such as bacterial films and barnacles adhered on the polymer coatings (Fig 1B). The new technology can be readily integrated with existing or newlydeveloped polymer coatings, combining the advantages of various state-of-the-art antifouling technologies. This new active-antifouling system is environmentally friendly, autonomous, highly effective, and potentially durable over long-term applications.

Next, we will further discuss the fundamental effect of active surface deformation on marine organism-surface interactions. A new theory for biofouling detachment caused by substrate deformation, instead of external forces, will be presented.

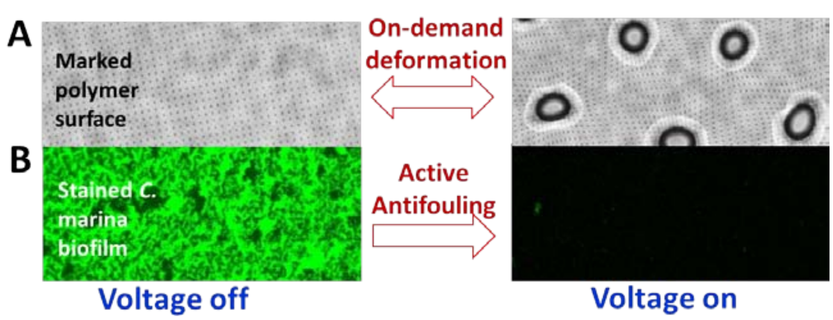

Figure 1. A silicone-based coating can deform significantly under applied voltages (**A**) and can actively detach over 99% of biofilms formed on the surface (**B**).

MISC-4

IDENTIFYING VIABLE ALTERNATIVES TO COPPER HULL PAINTS FOR THE RECREATIONAL BOATER

Stephanie Bauer

Port of San Diego

San Diego Bay is home to approximately 8,000 recreational boats with a large percentage using copper antifouling hull paints to prevent the growth of marine organisms. Copper antifouling paints were identified to be the primary source of dissolved copper into Shelter Island Yacht Basin (SIYB) and other San Diego Bay marina areas. Copper leached from these hull paints have been found to be toxic to marine life, resulting in a Total Maximum Daily Load (TMDL) issued for SIYB that requires a 76 percent reduction of copper loading by 2022. This is the first regulation of its kind in the nation to target a specific area of marinas as the primary source of a pollutant.

The San Diego Port District's EPA funded *"Safer Alternatives to Copper Antifouling Paints for Marine Vessels"* Project was a three year grant, designed to find viable alternatives to copper antifouling paints that could help reduce the reliance on copper paint. The project tested the effectiveness of several new and emerging alternative hull coatings. Viability was ascertained by assessing performance, longevity, and cost associated with the use of the alternative coatings and comparing them to conventional copper antifouling paints. Alternative hull paints were evaluated using a two phased testing approach; panel and boat testing with several successful viable alternative paints being identified, many of which are currently on the market.

The study also revealed additional findings that may facilitate the transition to alternative hull paints and assist boaters in selecting appropriate alternative hull paints for their boat hulls. This presentation will discuss the approach taken to assess alternative hull paints, the results, and key findings of the project.

MISC-5

AQUEOUS BASED BLOCK COPOLYMER NANOSTRUCTURED COATINGS FOR ANTIFOULING APPLICATIONS

Nikhil Gunari, Gilbert C. Walker

University of Toronto

Biofouling is a major problem for shipping and fishing industries leading to huge maintenance costs. Marine organisms such as algae, barnacles, tubeworms etc. settle and grow on structures that are immersed in seawater such as aquaculture cages, boat hulls or water pipes. The most effective antifouling paints to date are based on tin and copper biocides; however, these biocides are toxic to marine organisms. We have developed an aqueous based process to fabricate nanostructured surfaces of block copolymers. The method involves a phase transfer process wherein an amphiphilic block copolymer is transferred from water immiscible organic solvent to an aqueous solution. We transferred polystyrene-*block*-poly(2-vinyl pyridine)-*block*-polyethylene oxide (PS-b-P2VP-b-PEO) from chloroform to aqueous solution in a separatory funnel. A phase transfer agent is dissolved in the aqueous solution to assist and accelerate the phase transfer of the amphiphilic block copolymer from organic to aqueous phase to obtain a micellar solution. The process is versatile, does not require mechanical shaking and can be used for large-scale production. Thin films of polymer can be made by dip-coating, brush coating, spray-coating or simply pouring onto the substrate. Various substrates have been coated using this method such as, silicon, glass, stainless steel and nylon ropes that are used in the aquaculture industry. After one month of static immersion testing, glass slides coated with the aqueous block copolymer coatings showed no growth of macrofoulers such as barnacles and tubeworms. Further, these block copolymer micelles have been used to synthesize encapsulated inorganic nanoparticles and encapsulated organic molecules for the controlled release of antifouling biocides. Nanostructured thin films of encapsulated biocide can be used to control the leaching rate for better performance and environmental safety.

We gratefully acknowledge ONR for financial support.

MISC-6

QUANTIFICATION OF FRICTION DRAG INCREASE DUE TO PROPELLER FOULING

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The major consequence of fouling on ship propellers is the formation of a hydrodynamic rough surface that has negative influence on propeller function and propulsion efficiency. Different types of fouling are known to occur on propellers; most common are algae, barnacles, tubeworms and bryozoans next to slime fouling, a common term for various groups of marine micro-organisms.

The roughness of a propeller is determined either with advanced laboratory equipment to get quantitative data (Ra or Rt values) or with a so-called Rubert Gauge, a qualitative tool for comparative estimate of the surface roughness condition of the blades. Biological fouling on propellers cannot be described in concrete physical dimensions such as Ra or Rt values. This is due to the very different physical properties of biological organisms and secondly to the non-uniform distribution of fouling. Therefore, existing principles for roughness determination do not allow proper measurement of hydrodynamic roughness due to fouling.

The only way to investigate such roughness is to measure drag effects of bronze samples that have been exposed for some time to certain fouling conditions. With the Friction Disk Machine (FDM) available at TNO such measurements can be carried out. By comparing drag properties of bronze disks in fouled condition with those in clean condition, the added drag effects of fouling present were determined. Slime fouling may give added drag effects of 5 – 10 %; macrofouling such as barnacles and green algae was found to give increased friction drag up to 50 – 80 %.

The results in this study show that fouling organisms with very different morphology are able to give disturbance of hydrodynamic flow resulting in increased friction resistance. Using the FDM for direct measurement of drag effects of such “biological” surface conditions enables collection of relevant data of fouling induced drag resistance.

MISC-7

SHORT- AND LONG-TERM FUEL SAVINGS FROM HYDROGEL-BASED FOULING RELEASE COATINGS

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A wealth of information is available in the literature demonstrating that PDMS-based Fouling Release Coatings (FRCs) have a reduced friction coefficient with water compared to biocide-based antifoulings on smooth surfaces. For many years, commercial FRCs were exclusively applied after full abrasive blasting of the ship hull (equivalent to “smooth” conditions). In such cases, it is not easy for the ship owner to elucidate how much of the fuel savings experienced after vessel outdocking comes from the FR system and how much from surface preparation. Furthermore, early FRC generations had poor antifouling properties, with extensive slime fouling and possibly even macrofouling after long idle periods. The latter also raised doubts about the long term fuel saving properties of FRC coatings.

Using TNO’s a Friction Disk Machine we demonstrate that:

- Fouling Release systems have an important roughness damping effect compared to a model SPC system (silyl acrylate-based). Hence, Fouling Release coatings are also cost-efficient for non-blasted vessels.
- Fouling prevention properties are the key to long term fuel savings
- Some FRC coatings have fouling prevention capabilities in level with or better than the best SPC coatings.

To reach these conclusions, controlled roughness templates were transferred to epoxy coated aluminium disks which were subsequently applied with either a full FR system or a full SPC system. The drag resistance from these rough surfaces was then measured by means of the Friction Disk Machine. The results show that the higher the initial roughness of the substrate, the larger the difference in friction between the FR and the SPC systems. Finally, a set of smooth references was also immersed in natural seawater at Den Helder (The Netherlands) in order to test their fouling prevention capabilities. The results emphasize the importance of choosing the right FRC formulation so as to achieve the best long term fuel savings.

Acknowledgements: The professional support from Dr. Job Kljinstra and Mr. Mark Bakker (TNO) is greatly appreciated.

MISC-8

DO BARNACLES HAVE COLOUR PREFERENCES IN THEIR CHOICE OF SETTLEMENT SUBSTRATUM?

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Jeremy C Thomason (Ecoteknica SCP)

Anya C Hurlbert (Newcastle University, UK)

Settlement into biofouling communities is influenced by factors such as larval supply, light, presence of conspecifics and substratum characteristics. Colour of the substratum as a settlement cue so far has been investigated from a human perspective, neglecting the visual capabilities of the organism. The aim of this study was firstly to determine if barnacle cyprids have a preference for different coloured surfaces in settlement. This would not necessarily mean that the larva uses colour vision; instead it may be relying on brightness differences. Therefore the second aim was to determine if the cyprid's settlement behaviour is influenced by the true colour of the substratum – as perceived by the cyprid visual system - or by brightness levels. Colour preferences of cyprids of the barnacle *Amphibalanus amphitrite* in settlement substratum were tested in the lab. Tested colours were specifically selected based on presumptive spectral sensitivity of the visual receptors of the barnacle larval nauplius eye. The lab experiment was conducted as a 2-choice experiment. Statistical analysis was done using Exact test and Regression Analysis. These results were compared to a field experiment with *Semibalanus balanoides* using the same colours. Cyprids preferred to settle on the tested colours black, violet, red and green over dark blue and white. The colour of the substratum has an effect on the choice of settlement substratum for the two particular barnacle species tested. Brightness of the substratum is an important determinant for settlement, though true colour perception cannot be dismissed as results of the field experiment indicate.

MISC-9

MONITORING THE IMPACT OF BIOFOULING ON SHIP POWERING CONDITION – CASE STUDIES FROM US NAVY EXPERIENCE

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Biofouling increases roughness and therefore skin friction and drag and negatively impacts fuel efficiency of ships. The buildup of biofouling is most often managed through the application of marine coating systems a combination of antifouling coatings and, in some cases, in-water ship husbandry (cleaning). The concept of quantifying the relationship between speed and power as a function of biofouling has been explored for quite some time and has taken many forms. Numerous vendors offer commercial speed/power monitoring systems such as DEXTER and Casper®. The timely detection of the buildup of biofouling can be used to trigger maintenance (in-water hull and propeller cleanings) and could also potentially be used to detect improvements in performance attributable to a new materials or practices. Most monitoring systems analyze critical parameters such as speed over ground, speed through the water, shaft torque, shaft RPM, and wind speed and direction, but also include other important engineering and environmental parameters which can improve accuracy.

In the 1990's the US Navy began to develop in-house technique to quantify ship speed/power that involved manual data logging. The focus was also on relating the powering condition to hull and propeller biofouling condition (roughness). In-water inspections were executed just before data collection which initially was carried out during a dedicated power trial. In more recent efforts the focus was on automation of data collection and the application of an algorithm to identify and isolate ship operations conditions without interfering effects such as from high wind or current.

This paper will focus on a subset of the early US Navy efforts to relate speed/power to biofouling condition. We will also present our most recent data which were automatically collected continuously over time. We will relate the new data to earlier data sets and discuss lessons learned. Variables such as the number and type of sensors, data collection frequency, the impact of filter window width, and the length of running means will be discussed.

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Index

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Abbasov, V, 49
Abed, R, 30, 135
Achary, M, 109
Acosta, B, 89
Aguila-Ramirez, R, 87
Ajmal Khan, S, 25
Aktas, D, 62
Alberto, E, 11
Alberts, E, 122
Aldred, N, 66, 68, 76, 84, 92, 97, 118
Aliyeva, L, 49
Alles, M, 106, 107
Amade, P, 86
Anantharaman, P, 86
Anil, A, 65
Anton, C, 39
Araki, T, 49
Arpa Sancet, M, 97, 107
Arthi, A, 88
Asahina, A, 65
Ashton, G, 130
Atlar, M, 15, 53
Aufrecht, J, 70

B

Bai, T, 93
Bailey, J, 11
Bakker, M, 123, 143
Balazs, A, 23
Bao, W, 105
Barani, A, 138
Barlow, D, 68, 83, 84, 117, 140
Barteau, K, 102
Bauer, S, 46, 97, 107, 142
Baum, C, 107, 115
Beech, I, 59, 60
Bell, A, 128
Beniash, E, 85, 140
Berglin, M, 52, 78, 95, 96, 124
Berner, M, 9
Betts, D, 45
Bhattacharya, A, 23
Birch, W, 76
Bishop, C, 36
Biswas, S, 109
Blache, Y, 138
Blöcher, N, 114
Bloecher, N, 34, 80
Blom, A, 9
Blustein, G, 87
Booth, G, 131
Borges-Souza, J, 87
Bork, U, 9
Bouchez, A, 138
Brault, N, 93
Bray, S, 54
Brennan, A, 21
Bressy, C, 23, 39, 40, 42, 77, 138
Breur, R, 55
Brewer, L, 11, 68
Briand, J, 138
Bright, F, 11
Brisset, H, 40
Brive, L, 96
Brophy, S, 36
Brotto, D, 110
Bruin, A, 64
Burden, D, 68, 83, 84, 117, 140
Burkett, B, 20
Burrows, R, 139
Butler, B, 95

C

Caldwell, G, 79
Callow, J, 10, 11, 28, 41, 45, 94, 97, 99, 101, 102, 105, 120, 134
Callow, M, i, 10, 11, 28, 41, 45, 74, 94, 97, 99, 101, 102, 105, 106, 120, 134
Campos, L, 102
Camps, M, 138
Canilho, N, 39
Cao, W, 80, 90
Cao, Y, 82
Capolupo, C, 6, 24
Capretto, L, 75
Carl, C, 20, 34
Carr, L, 93
Case, R, 137
Cha, H, 121
Chai, C, 20
Chakrabarty, S, 14
Chen, X, 82
Chen, Z, 91
Cheng, D, 72
Chilkoti, A, 107
Cho, Y, 102
Choi, Y, 121
Christophis, C, 106
Cieslak, L, 60
Clare, A, 66, 68, 76, 79, 84, 92, 93, 97, 118, 133
Cole, A, 126
Colvin, M, 24
Cone, G, 28, 102
Conlan, S, 118
Corbel, B, 89
Couthon-Gourves, H, 89
Coutinho, R, 22, 50, 95
Culver, C, 129, 130
Curran, J, 17, 144

D

Dahlström, M, 78
Dario, C, 75
Darling, D, 56
Daugherty, M, 68
Davidova, I, 62
Davidson, I, 130
de Nys, R, 20, 34, 37, 38, 124
DeCew, J, 70, 114
Decker, J, 21
DeForest, D, 5, 125, 126
Delos, C, 6, 27
DeMont, E, 36
Dennington, S, 28, 32, 51, 123
Derksen, D, 36
DeSimone, J, 45
Detty, M, 11, 94
Di Fino, A, 84, 92, 93, 118
Dickinson, G, 83, 85, 140
Dima, H, 22
Dimitriou, M, 102
Dobretsov, S, 29, 30, 134, 135
Dong, Y, 90
Drach, A, 70
Drake, L, 41
D'Souza, F, 64
Duan, J, 73
Dugan, J, 129, 130
Dunbar, C, 143
Duncan, K, 51, 59, 60, 62
Dupont-Gillain, C, 48
Dürr, S, 83, 144

E

Earley, P, 6, 24, 27
Ederth, T, 84, 118, 120
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  Berg, E.A., **2**
El-Lateef, H, 49
Elwing, H, 52, 78, 95, 96, 124
Emerson, D, 61
Eppard, S, 35, 41
Essock-Burns, T, 66, 83, 117, 140
Evariste, E, 41, 94
Everett, R, 83, 140

F

Faimali, M, 31
Falck, L, 52
Fears, K, 84
Finlay, J, 11, 28, 97, 102
First, M, 41
Fischer, D, 102
Fladung, T, 115
Flemming, H, 67
Fletcher, L, 16
Fornalik, J, 11
Foster, R, 35, 41
Francisco GANEM, 22
Fredriksson, D, 114

G

Gabriel-Zadeh, C, 49
Galli, G, 10
Gansel, L, 114
Garaventa, F, 31
García, M, 87
Gatley, C, 94
Gawalt, E, 85
Gensemer, R, 5, 126
Genzer, J, 36
Georgiades, E, 128
Glenn, F, 34
Gohad, N, 66, 83
Golumbfskie, W, 69
Goodes, L, 28, 123
Gorsuch, J, 5, 126
Gowing, S, 18
Grandison, C, 132
Granhag, L, 18
Gravier, C, 39
Greco, G, 31
Gregori, G, 138
Griggs, D, 144
Grosicki, K, 36
Grunze, M, 37, 101, 105, 106, 107
Gu, Z, 105
Guenther, J, 34, 80, 114
Gulati, S, 41
Gunari, N, 142
Guo, S, 116

H

Hadfield, M, 65, 68
Hajiyev, S, 49
Han, X, 91
Hansen, T, 123
Haslbeck, E, 17, 57, 144
Hasneen, A, 43
Hausmann, D, 52
Hawker, C, 102
Hayes, P, 23
Heinonen, K, 56, 108
Heissler, S, 93, 101
Helena, M, 22
Hellio, C, i, 23, 86, 87, 89, 134
Henke, D, 14
Herdman, R, 79
Hernández Guerrero, C, 89
Hernández-Guerrero, C, 87
Heusinger von Waldegge, T, 115
Heyer, A, 64
Hillerup, D, 9
Hofmann, U, 70
Holappa, K, 56
Hollrah, R, 126
Holm, E, 17, 18, 144
Holman, K, 7, 46
Hou, B, 73
Hozumi, A, 72
Hu, Y, 80

Huang, R, 90
Huang, W, 101
Huang, Y, 65, 73
Hunter, J, 112
Hurlbert, A, 83, 144
Hwang, D, 121

I

Id Daoud, G, 23, 89
Ikegai, H, 71
Imbesi, P, 21
Immanuel, G, 86, 88
Inbakandan, D, v, 25
Israelachvili, J, 121
Ista, L, 43, 118
Itoh, H, 71
Iyapparaj, P, 26, 86, 88

J

Jackson, S, 123
Jahedi, M, 37, 38, 124
Jeannin, M, 63
Jiang, S, ii, 10, 20, 46, 92, 93, 94, 100, 101
Johnson, L, 129, 130
Juhel, G, 67

K

Kane, D, 16
Kanematsu, H, 71, 72
Kang, E, 47
Kania, C, 108
Kannapiran, E, 88
Kawakami, H, 49
Keefe, A, 93, 100, 101
Kennett, S, 16
Keramidas, P, 51
Khoo, B, 116
Kikuchi, Y, 49
Killea, L, 29
Killops, K, 102
King, P, 20, 37, 38, 124
Kirihara, S, 71
Klebert, P, 114
Klijnstra, J, 143
Klinstra, J, 123
Koelsch, P, 99
Koene, B, 95
Koka, A, 53, 115
Koutsaftis, A, 3, 128
Kowalski, M, 60
Koya, S, 71
Kramer, E, 102
Krapp, A, 17
Krishnan, M, 26
Kristiansen, D, 114
Krohlin, W, 110
Kruszewski, K, 85
Kuroda, D, 71, 72

L

Labeeuw, L, 137
Lader, P, 114
Lamb, R, 8, 35, 37
Langumier, M, 63
Lanneluc I., 63
Lawrence, S, 35, 41
Lay, S, 47, 76
Le Berre, B, 138
Leal-Taixe, L, 92
Lee, C, 76
Lee, H, 116
Lee, J, 59, 61, 62, 69
Lee, S, 20
Legrave, N, 86
Lejars, M, 42
Lewis, A, 84
Lewis, J, 113
Li, S, 105
Li, X, 44, 80
Li, Y, 91, 93, 105, 106
Li, Z, 80
Liang, X, 105, 106
Licina, G, 46
Liedberg, B, 84, 120
Lim, S, 121
Lin, C, 46
Little, B, 59, 61, 62, 69
Liu, Y, 90, 91
Long, K, 4, 125
Longyear, J, 41, 79, 136
Lopes, C, 22
Lopez, G, 141
López, G, 43
Lovelace, R, 56
Lu, Q, 121
Lund, G, 53, 115
Luo, W, 80
Lutkenhouse, C, 17, 144
Lynd, N, 102
Lynn, D, 17, 144

M

Makama, Z, 60
Maki, J, 66, 83
Malchev, P, 55
Maleschlijski, S, 92
Maranda, L, 29
Marceaux, S, 77
Marchisio, V, 77
Marcori, D, 10
Margaillan, A, 39, 40, 42, 77
Martin, C, 77
Martinelli, E, 10
Martin-Tanchereau, M, 103
Masheder, B, 72
McBeth, J, 61
McDonald, J, 139
Mehiri, M, 86
Mendes, P, 99

Meseguer-Yebra, D, 143
Messano, L, 50
Meyer, J, 5, 125, 126
Mieno, H, 19
Mieszkin, S, 10, 99, 120, 134
Mildon, W, 29
Miller, R, 29
Millichamp, I, 103
Mohamed, T, 93
Mohanty, A, 109
Mollica, A, 31
Mollica, Al, 31
Mollica, An, 31
Morales, F, 64
Mount, A, 66
Muller, L, 11
Munuswamy, N, 26
Murakami, K, 72
Murthy, S, 25, 33
Muthukrishnan, T, 30

N

Nakanishi, K, 35, 37
Nedved, B, 65
Neoh, K, 47
Neves. C, 22
Nie, M, 33
Nowinski, A, 100, 101

O

O'Brien, D, 113
Ober, C, 102
Ohara, C, 72
Okamura, H, 26
Oldham, A, 51, 59, 62
Olsen, S, 9
Orihuela, B, 66, 83, 117, 140
Ortiz-Aguirre, I, 87

P

Page, H, 129, 130
Paik, H, 43
Palavesam, A, 86
Palermo, J, 87
Patel, A, 23
Pavanello, G, 31
Pei, X, 98
Pérez, M, 87
Perez-Ibarra, M, 59, 60
Perry, B, 81
Perry, J, 103
Petrone, L, 76, 84, 118
Pettitt, M, 101
Pfeil, E, 115
Phillips, D, 116
Pinckard, N, 56, 108
Pink, D, 36
Pinori, E, 52, 78, 95, 96
Piola, R, 132
Politis, G, 53
Pollack, K, 21
Poole, A, 20, 34, 37, 38, 124
Pope, J, 89
Poppleton, J, 4
Prakash, S, 86, 88
Pranzetti, A, 99
Preece, J, 99
Presnall, D, 14
Preston, B, 144
Prukop, S, 44
Punitha, M, 90
Pustam, A, 36

Q

Qian, P, 22, 91

R

R. Johnny Eliasson, **1**
Rahman, M, 43
Ralston, E, 74, 109
Ramasubburayan, R, 86, 88
Ramirez, R, 89
Ramotowski, T, 57
Ray, R, 61, 62, 69
Raymond, J, 21
Refait, P, 63
Revathi, P, 26, 86
Reynolds, K, 41
Rice, M, 57
Rittschof, D, 20, 47, 66, 68, 83, 85, 117, 140, 141
Rivera-Duarte, I, 6, 24, 27
Röhrig, M, 105
Román, J, 122
Rosen, G, 6, 24, 27
Rosenhahn, A, 37, 74, 92, 93, 97, 101, 105, 106, 107
Rosenhahn, B, 92
Rouxhet, R, 48
Ruiz, G, 130
Ryan, A, 6, 27

S

Sabattini, R, 22
Sablé, S, 63
Sabot, R, 63
Sahu, G, 109
Salgado, D, 144
Salta, M, 32, 75
Sanchez de Lozada, M, 18
Sánchez, M, 87
Santore, R, 6, 24, 27, 126
Sarangi, S, 115
Satheesh, S, 32, 90
Sato, Y, 49
Satpathy, K, 109
Scardino, A, i, 8, 16, 37
Schetz, J, 126
Schoch, P, 36
Schultz, T, 117

Scianni, C, 127
Scott, V, 136
Scuppe, H, 123
Sendra, G, 92
Senkbeil, T, 93
Serrano, A, 104
Serrano, Â, 12
Severin, O, 86
Sexton, B, 34
Shankar, C, 90
Shen, P, 105
Shephard, B, 126
Shi, W, 68
Shibata, K, 7
Shivapooja, P, 43
Shohei, S, 71
Siew, S, 76, 116
Simon, G, 89
Skaja, A, 57, 115
Skalski, I, 108
Skoulis, N, 4
Smith-Palmer, T, 36
Sokolova, A, 11
Spencer, N, 12, 104
Spillman, C, 117
Spillmann, C, 84
Stamper, D, 17, 144
Steinberg, M, 17, 57, 144
Steinberg, P, 136
Stephens, A, 74
Sterner, O, 12, 104
Stewart, R, 119
Stokes, K, 28, 32, 33, 75, 123
Stupak, M, 87
Stuppy, S, 37, 107
Subramanian, H, 94, 150
Suflita, J, 62
Sukkaew, P, 84, 118
Sun, F, 100, 101
Sundaram, H, 102
Sunner, J, 60
Suppers, J, 143
Swain, G, 52, 53, 74, 101, 107, 109, 110, 115, 135
Swope, B, 6, 24

T

Tang, M, 82
Tanguy, B, 39
Tanur, A, 68
Taylor, S, 122
Taylor, W, 102
Telegdi, J, 48
Telfer, T, 81
Teo, M, 76
Teo, S, 20, 47, 116
Tew, G, 13
Thomason, J, 29, 83, 144
Thome, I, 101
Thomé, I, 107
Thompson, S, 105
Thorlaksen, P, 9
Togosmaa, L, 26
Tordonato, D, 57, 115
Tosatti, S, 12, 104
Tran, C, 65
Trepos, R, 86, 89, 134
Tribou, M, 110
Tsukrov, I, 70
Tucker, W, 57
Turnbull, J, 81
Tyson, L, 41, 103

U

Urata, C, 72

V

Vaksmaa, A, 135
van der Merwe, J, 131
Vega, I, 83
Velandia, F, 67
Venkatesan, R, 25
Verborgt, J, 35, 41
Verduzco, R, 44
Véron, B, 23
Viju, N, 90
Vincent, P, 16
Voermans, A, 143
Vuck, M, 34
Vucko, M, 20, 37, 38, 124

W

Wahl, K, 68, 83, 84, 117, 140
Waite, H, 121
Walker, G, 68, 142
Walsh, A, 126
Waltz, G, 11
Wang, C, 14, 119
Wang, J, 82
Wang, Y, 45
Webb, A, 41
Webster, D, 58
Wendt, D, 11, 45, 68
Werwinski, S, 33
Whalan, S, 34
Wharton, J, 28, 32, 33, 51, 75, 123
White, A, 100, 101
Wilker, J, 122
Williams, D, 103
Williams, M, 34
Willsey, E, 68
Wilson, A, 16
Wong, C, 69
Wong, D, 45
Wood, R, 28, 51, 123
Wooley, K, 21
Wu, A, 8, 35, 37
Wu, F, 83, 85, 140
Wyeth, R, 36
Wynne, K, 14

X

Xiao, L, 105
Xu, F, 46
Xu, X, 46

Y

Yan, T, 80, 90
Yan, W, 90
Yandi, W, 120
Yang, B, 90
Yang, J, 105, 106
Yang, W, 47, 93
Yang, Y, 48
Yasani, B, 10
Yaxley, B, 113
Ye, Q, 98
Yokoyama, S, 72
Yoo, H, 102
Yoshida, K, 71
Yuting, S, 94

Z

Zabin, C, 130
Zalmon, I, 110, 111
Zargiel, K, 52, 107, 135
Zeng, H, 121
Zhang, H, 8
Zhang, J, 46
Zhang, W, 14
Zhang, Y, 98
Zhao, X, 141
Zhou, F, 98
Zhou, X, 106
Zhou, Z, 102
Zodrow, J, 126
Zürcher, S, 12, 104