MIC-STAND 2024

BOOK OF ABSTRACTS

Mitigation of Microbiologically Influenced Corrosion:

Towards Scientific & Industrial Standardization

Faculdade de Ciências da Universidade de Lisboa

Lisboa, Portugal
July 24-26, 2024

MIC-STAND 2024

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Towards Scientific & Industrial Standardization

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TITLE

Book of abstracts of the *Mitigation of Microbiologically Influenced Corrosion: Towards Scientific & Industrial Standardization* - MIC-STAND hybrid meeting

24 to 26th July 2024, Lisboa – Portugal

EDITORIAL BOARD

Elisabete R. Silva (Chair) | Ana P. Carapeto and Noelia L. García

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WELCOME TO MIC-STAND

Dear Esteemed Participants,

We are delighted to welcome you to take part in the MIC-STAND hybrid meeting 2024, hosted by the Faculdade de Ciências at the University of Lisboa in Portugal.

Set against the picturesque backdrop of Lisboa, this hybrid meeting is prepared to be an encouraging exploration of challenges and breakthroughs in Microbially Influenced Corrosion (MIC) Mitigation. Our core mission is to foster dynamic discussions within the expansive MIC community, bringing together young researchers, high-level scientists, industry stakeholders, policymakers, and regulators.

Our inclusive approach emphasizes cross-disciplinary and interdisciplinary collaboration, fostering an environment where the exchange of knowledge flourishes. The focal point of our discussions will span the entire spectrum of MIC Mitigation, covering Diagnosis, Monitoring, and Mitigation.

Beyond the exchange of ideas, this meeting aspires to be a catalyst for innovative, sustainable, and environmentally friendly strategies in the battle against MIC. Our goal is to translate these strategies into practical, real-world applications that make a tangible impact.

Join us in this journey of exploration, collaboration, and transformation.

Let's collectively shape the future of MIC Mitigation!

Warm regards,

The local organizing committee

Elisabete R. Silva, Noelia L. García and Ana Carapeto

COMMITTEES

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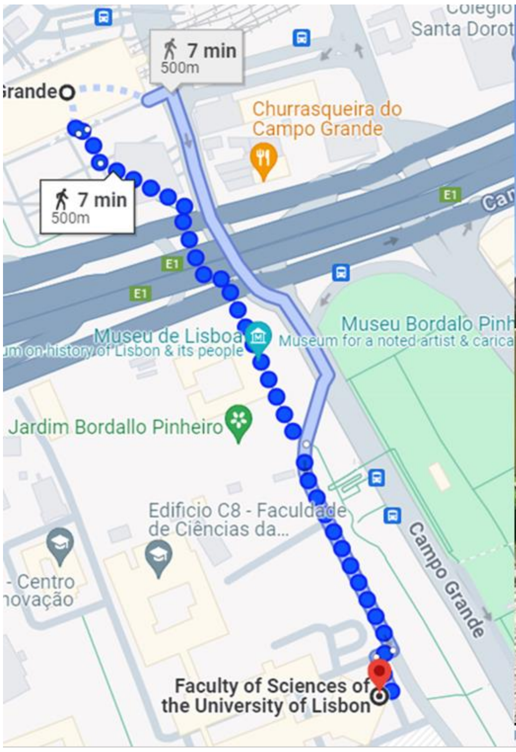
The National Biofilms Innovation Centre (NBIC) is an Innovation Knowledge Centre (IKC) funded by BBSRC and Innovate UK. Biofilms are central to our most important global challenges – from infections, antimicrobial resistance and food safety to water security, marine biofouling and corrosion – and exert significant economic, social and environmental impacts estimated at $5 trillion per annum. NBIC was launched in 2017 by its four lead universities (Edinburgh, Liverpool, Nottingham and Southampton) to address these challenges and bring together the best of UK research and businesses to drive the translation of biofilm research into innovative solutions.

ACADEMIC MENTOR

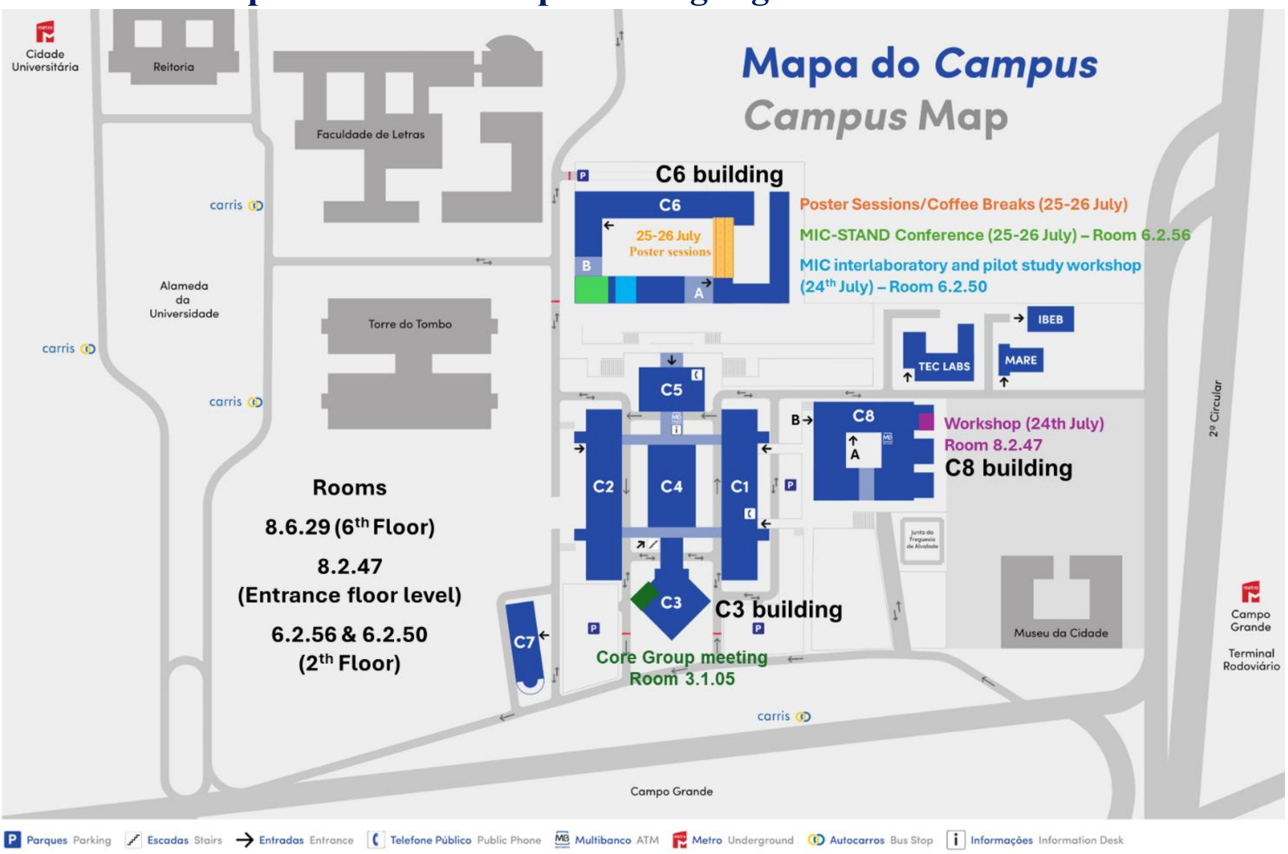
Coatings (ISSN 2079-6412) is an international, peer-reviewed and open access journal devoted to the science and engineering of coatings, thin and thick films, surfaces and interfaces. The journal covers Coatings (ISSN 2079-6412) is an international, peer-reviewed and open access journal devoted to the science and engineering of coatings, thin and thick films, surfaces and interfaces. The journal covers coatings, surface and interfaces in the broader sense.

SUPPORTERS

MEETING VENUE



Campus Ciências - Map with Highlighted Event Locations



HOW TO GET HERE?

Getting to FCUL/ULisbon is easy and quick - by bus, subway, car, or even bicycle (cycle lanes).

Available transportation:

Bus

Carris: 701, 717, 731, 735, 736, 738, 747, 750, 755, 764, 767, 768, 778, 783, 796, 798

Carris Metropolitana: bus routes available at Carris Metropolitana - Escolas

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Underground

Cidade Universitária and **Campo Grande** Stations

Train

Entrecampos Station

Car

2.ª circular - **Campo Grande / Cidade Universitária** exit

Taxi

https://lisboataxis.pt/en/

Others

Uber, Bolt

DETAILED PROGRAMME PER DAY

Wednesday, 24 July 2024

9:45-10:00 **Registration**

9:45 -13:30 **Fluorescence Microscopy and applications to microbiology workshop - Morning Session**

10:00 **Introduction to microscopy**
- Fundamental properties of light
- Main families of fluorophores
- Introduction to fluorescence microscopy

Instrumentation (I)
- Anatomy of the epifluorescence microscope

11:10 **Coffee Break**

11:30 **Instrumentation (II)**
- Confocal microscopy

Applications to microbiology
- Best practices for imaging microbiological samples

12:30 **Tour of the imaging facility**

13:00-14:15 **LUNCH**

14:15-17:30 **MIC interlaboratory and pilot study workshop**
C6 building, 2nd Floor, Room 6.2.50

14:15 • Overview of Pilot study test protocols

15:15 • Provisional test results Euro-MIC members

16:15 • Open discussion of protocols and engagement (new sites and inoculum samples)

17:15 • Wrap up

19:30 **Dinner**

PROGRAMME

14:30-17:30 **Fluorescence Microscopy and applications to microbiology workshop**
C8 building, Entrance Floor Level, Room 8.2.47

14:30 **Introduction to microscopy**
- Fundamental properties of light
- Main families of fluorophores
- Introduction to fluorescence microscopy

Instrumentation (I)
- Anatomy of the epifluorescence microscope

15:40-16:00 **Coffee Break**

16:00 **Instrumentation (II)**
- Confocal microscopy

Applications to microbiology
- Best practices for imaging microbiological samples

17:00 **Tour of the imaging facility**

19:30 **Dinner**

Thursday, 25 July 2024

Location: C6 building, Room 6.2.56

8:30-9:00	**Registration**
9:00	**Opening/Welcome**
SESSION 1	**Early detection of MIC for effective control: new advances in Diagnosis and Monitoring** Chairs: Geert Potter & David Culliton
9:15	KN1 **Use of Complimentary Microbial Monitoring Technologies for Effective Asset Integrity Threat Assessment** Matthew Snape (SGS) & Danika Nicoletti (LuminUltra)
10:10-10:30	**Coffee-Break**
10:30	O1 **Application of Machine Learning Techniques for Detection of Microbiologically Influenced Corrosion** • Miroslav Milovanović
10:45	O2 **Electrochemical Sensing of Aerobic Marine Biofilms and the Influence of Nitric Oxide Attachment Control** • Julian Wharton
11:00	O3 **Exploring Biomimetic Texturing for Antifouling Applications: Insights from a Computational Fluid Dynamics (CFD)** • Samira Nazari
11:15	O4 **Case Study on Progressive Corrosion of Carbon Steel Cooler Treated with Epoxy-ceramic Coating** • Veronika Hlavackova
11:30	**Q&A Session**
12:15-14:00	**LUNCH | POSTER SESSION**
SESSION 2	**MIC mitigation strategies towards eco-friendly and sustainable systems** Chairs: Torben L. Skovhus & Tugce Tuccar
14:00	KN2 **Demyth mechanism of microbial corrosion to design microbial corrosion-resistant materials** • Dake Xu (Northeastern University, China)
14:50	O5 **Marine Microbial Induced Mineralization: Exploring Mechanisms for Bioinspired Anticorrosion Solutions** • Maria J. Marques
15:05	O6 **Chitosan-based Coatings for Marine Biofilm Prevention** • Filipe Mergulhão
15:20	O7 **Polyoxometalate Ionic Liquids (POM-ILs) as Protective Coatings for Cultural Heritage against Acid Corrosion and Biodeterioration** • Archismita Misra
15:35	**Q&A Session**
16:00-16:30	**Coffee Break & Poster Session**
16:30	O8 **Investigating the use of freshwater in simulations of water compensated fuel tanks to impede corrosive activity by SRB** • Gareth Williams
16:45	**Q&A Session**
17:05	**Closing Remarks**
20:30	**Social Dinner**

Friday, 26 July 2024

Location: C6 building, Room 6.2.56

8:30-9:00	**Registration**
9:00	**Welcome /Opening Remarks**
SESSION 3	**Standardization: the lab-to-field transition** Chairs: Julian Wharton & Rick Eckert
09:15	KN3 **Building Consensus: Advancing Standards and Innovation in Microbiologically Influenced Corrosion and Biofilms** • Paulina D. Rakowska (National Biofilms Innovation Centre, UK)
10:10-10:30	**Coffee-Break**
10:30	O9 **Integrating Multiple Lines of Evidence (MLOE): Essential for Diagnosis of Microbiologically Influenced Corrosion (MIC)** • Torben L. Skovhus
10:45	O10 **Pilot Study of a Laboratory-Based Microbiologically Influenced Corrosion (MIC) Test Using Microbiological Consortia Sampled from the Field** • Sara Kalajahi
11:00	O11 **Investigating the Efficacy of THPS and Glutaraldehyde Treatment for Enhanced Performance on North Sea Offshore System** • Tanmay Chaturvedi
11:15	O12 **Dual anaerobic reactor model to study biofilm and microbiologically influenced corrosion interactions on UNS G10180 carbon steel** • Liam Jones
11:30	**Q&A Session**
12:15-14:00	**LUNCH | POSTER SESSION**
SESSION 4	**Industry insights: Technology and regulatory challenges** Chairs: Matt Snape & Maria Salta
14:00	KN4 **Biofouling assessment in the offshore renewable energy sector: challenges and expectations** • Pedro Vinagre (WAVEC Offshore Renewables)
14:50	O13 **Challenges and Opportunities in Corrosion Mitigation to Meet the Decarbonisation Journey** • Invited Talk (Lloyd's Register): Martyn Wright & Heather Hughes & James Kwame
15:15	O14 **Investigating Microbiologically Influenced Corrosion in Hot Water Boilers: Mechanisms and Mitigation Strategies** • Milena Rajic
15:30	O15 **New strategies for active corrosion protection of aluminium parts based on encapsulated inhibitors** • Andreu Ruiz
15:45	**Q&A Session**
16:10-16:40	**Coffee Break & Poster Session**
16:40	O16 **Quick Enrichment and Detection of Electrochemically Active Microorganisms** • Satoshi WAKAI
16:55	**Q&A Session**
17:00	Pitch **From Patents to Programmes - Supporting IPR Holders and Exploring Grants for all MIC-STAND members** • Theo Zacharis
17:15	**Closing Remarks**
20:00	**Social Dinner**

KEYNOTES

KEYNOTE SPEAKERS

Matt Snape (KN1)

Principal Consultant – *Facility Integrity & Oilfield Microbiology – SGS MIRAS, Global Bioesceinces Centre of Excellence, Lisbon, Portugal*

Experienced Global Subject Matter Expert for Production Chemistry, Microbiology and Facility Integrity. Industry recognised Technical Authority & Principal Consultant with a demonstrated history of working in the global oil and gas industry delivering value add, safety leadership and technical competency to customers projects and oil and gas field developments.

Responsible for leading multiple SGS global customer projects relating to integrated Production Chemistry services and Industrial Microbiology using advanced DNA meta-genomics, Reservoir Souring and Corrosion Forensics incorporating SGS MiRAS (Microbial Risk Assessment) technology, analytical programmes and consultancy services.

New service developments and continued focus on the role of microbes in green hydrogen cyclic storage in the subsurface, Carbon Capture and Underground Storage (CCUS), Geothermal, sustainable Biofuels, Biogas and a refreshed drive to support the wider remit of the renewable energy sectors.

Danika Nicoletti (KN1)

Applications Scientist - *LuminUltra Technologies Ltd.*

In her role as Applications Scientist, Danika leverages her industry and academic experience to work on product development, product applications, and thought leadership related to microbial monitoring and diagnostics in the energy industry. She has an MSc in Petroleum Microbiology with expertise in microbiologically - influenced corrosion, reservoir souring, and molecular microbiology. Danika is an active member of industry associations AMPP, IATA, IASH, EURO-MIC COST Action, and SPE, and is based in Calgary, Alberta.

Dake Xu (KN2)

Professor, Director of the Talent Affairs Office - *Northeastern University, China*

Dake Xu received his PhD degree from the Institute for Corrosion and Multiphase Technology, Ohio University in 2013. His research interests fall in the mechanism, detection, and mitigation of microbiologically influenced corrosion and antibacterial materials, which is an interdisciplinary research of material science, corrosion, and microbiology. He has published more than 110 peer-reviewed journal articles, and he served as editor or guest editor for the Journal of Materials Science & Technology, NPJ Material Degradation, Bioelectrochemistry, and other SCI-indexed journals.

Paulina D. Rakowska (KN3)

Biofilm Standards & Regulatory Programme Lead - *National Biofilms Innovation Centre, UK*

Paulina joint NBIC in 2020, where she has been leading a transdisciplinary biofilm standardisation programme by creating networks, linking academia with industry, contributing to influencing industry standards, research and metrology project development, securing funding and influencing policy. She built her scientific career at the UK's National Physical Laboratory (NPL), where she worked for 14 years leading metrology and standardisation projects in biometrology and surface chemical analysis, with the focus on biological systems. She has a vast experience in analytical R&D, interfacing physics, biology, chemistry and nanotechnology.

Pedro de Almeida Bica Lança Vinagre (KN4)

Researcher, Marine Communities specialist - *WavEC Offshore Renewables*, PT

Pedro, Biologist and Marine Ecologist, has large expertise in marine communities. He works actively in EIA and Environmental Monitoring, including biofouling (macrofouling), of coastal and offshore activities. He participates in expert groups (ICES WGORE, ICES WGMBRED, UNESCO-IOC GESAMP WG44) that aim to increase understanding and develop guidelines for more sustainable management of industries working in the maritime space.

INVITED SPEAKERS (O13)

Mr Martyn Wright

Principal Technology Specialist - *Lloyd's Register*

He is a technical author for Lloyd's Register Rules, in the subjects of materials, welding, and NDT. He manages LR's research portfolio in these subjects, actively engaging research in academia, Joint Industry Projects (JIP's), as well as their own internal research.
Additionally, he contributes to the IACS Expert Group for materials and welding, on behalf of LR. Furthermore, he serves as the Panel Chair for Annex T of BS7910 committee (BS7910: Guide to methods for assessing the acceptability of flaws in metallic structures).

Dr. Heather Hughes

BSc Hons Applied Chemistry with Polymer Chemistry, FIMF, RMCI - *Lloyd's Register*

Heather is a coatings and polymer formulation specialist with 35 years of experience in the marine sector. Her career started by formulating alternatives to tin based SPC resin systems for Antifouling paints, and continued working for a world leading coatings manufacturer for many years before branching out into the wider world of OEM yacht and composite materials manufacture. Latterly taking on the role of Non-metallics and Coatings materials Team Lead for Lloyd's Register, Heather is heavily involved in the assurance of materials essential to the decarbonisation journey of shipping, and is passionate and proactively involved in the evolution of materials which substantially lower the environmental impact of shipping on our oceans and their life sustaining ecosystems.

Dr. James Kwame, CEng MIMMM

Senior Manufacturing Technologist - *Lloyd's Register*

James works to develop and maintain LR's internal and external technical publications including LR rules, procedures and guidance and also works closely with various technical discipline subject matter experts to provide input on assurance of materials selection, fabrication and inspection in client projects and R&D. Prior to joining LR, James worked as a Research Engineer at the Advanced Forming Research Centre (AFRC)- Scotland, where he undertook various research and development activities into sheet metal forming.
In addition to his work at LR, James is passionate about education and serves on the Industry Advisory Board as a Member for Mechanical Engineering and related programmes at the University of Aberdeen. He also works as a reviewer for several journals and a scrutineer for chartered engineer registration at the Institute of Materials, Minerals and Mining (IOM3). He is a Chartered Engineer with the Engineering Council UK and a professional member of IOM3.

ABSTRACTS OF KEYNOTE SPEECHES

KN1

USE OF COMPLIMENTARY MICROBIAL MONITORING TECHNOLOGIES FOR EFFECTIVE ASSET INTEGRITY THREAT ASSESSMENT

Matthew Snape[1], Danika Nicoletti[2]

[1] *SGS Global Biosciences Centre, Polo Tecnologico do Lumiar, Lisboa, Portugal*

[2] *LuminUltra Technologies Ltd., Fredericton, Canada*

As technology for microbial monitoring advances and new detection methods arise, it is important to understand the question they are answering to contextualize data for effective decision-making. Historically, culture-dependent methods have been solely relied upon for microbial detection and quantification, however, current microbial monitoring strategies now involve the use of advanced technologies to build a complete and more comprehensive picture of the microbial contamination present in a system. In recent years, molecular microbiological methods (MMM) like quantitative polymerase chain reaction (qPCR) and next generation sequencing (NGS) have been more widely adopted in the energy industry for characterization and quantification of the microbial taxa that populate its infrastructure.

Past studies have been published investigating field data sets for qPCR to understand the contamination thresholds that are comparative to those of other frequently used methods like adenosine triphosphate (ATP) testing and serial dilution bottles [1]. Also, a recent AMPP standard TM21465 provides guidance on the sample handling and processing related to MMM methods including qPCR [2]. Though there has been an uptick in publications and guidance materials related to the use of qPCR for microbial assessment in the energy industry, there is still a gap in understanding around the field application of the technology and the verified insights it can provide, particularly in combination with other monitoring methods.

In this presentation, a typical field data set will be presented where qPCR, ATP, and adenylate energy charge were used in combination with the use of biomonitoring standard key performance indictors [1] for an upstream oil and gas site audit. This permitted the user to pinpoint the source and downstream microbial contamination was occurring and to assess the threat of microbiologically influenced corrosion (MIC). qPCR, ATP, and adenylate energy charge data sets were used to build a picture broader than any single technology would offer on its own. This ultimately led to a more in-depth understanding of the microbial population at this oil and gas recovery site allowing the chemical treatment program to be optimized and its success monitored over time.

Keywords: MIC, Integrity, Assessment, Threat, Complimentary Methods

References:

1. Lomans, B.P., de Paula, R., Geissler, B., Kuijvenhoven, C.A.T., Tsesmetzis, N., Proposal of Improved Biomonitoring Standard for Purpose of Microbiologically Influenced Corrosion Risk Assessment. *Soc. of Pet. Eng.*, **2016**, SPE-179919-MS.
2. Association of Materials Protection and Performance (AMPP), (**2024**) *Molecular Microbiological Methods – Sample Handling and Laboratory Processing*. AMPP TM21465-2024, https://store.ampp.org/ampp-tm21465-2024-molecular-microbiological-methods-sample-handling-and-laboratory-processing.

KN2

DEMYTH MECHANISMS OF MICROBIAL CORROSION TO DESIGN MICROBIAL CORROSION-RESISTANT MATERIALS

Dake Xu[1*]

[1] *School of Materials Science and Engineering, Northeastern University, Shenyang 110819, China.*

**E-mail: xudake@mail.neu.edu.cn*

Microbiologically influenced corrosion (MIC) is a form of corrosion where the electrochemical reactions are mainly accelerated by microorganisms or their metabolic activities. The mitigation of MIC has gained serious attention from academia and industry due to the severity and widespread occurrence of this form of corrosion. A mechanistic understanding is required to develop mitigation strategies. We found that the attached biofilm was mainly responsible for the microbial corrosion. We demonstrated from the genetic level that the biofilm can uptake electrons from metal matrix via extracellular electron transfer, pointing out the targets for MIC mitigation.

To effectively mitigate corrosive biofilms, we designed novel high-entropy alloys (HEAs) provides a unique opportunity for the development of structure-function integrated materials with high mechanical and antimicrobial properties. We then fabricated a quasi-equiatomic CoCrFeCuNi high-entropy alloy by selective laser melting (SLM) and in-situ alloying of a blend of pre-alloyed CoCrFeNi powder and Cu elemental powder, performing with a broad-spectrum antibacterial ability and good mechanical properties. The addition of copper or/and silver into HEAs endowed the materials with excellent antiviral properties, and the antiviral rate exceeded 90% against COVID-19 after coculture for 24 hours. We recently demonstrated the excellent antibacterial performances of FeNiTiCrMnCux high-entropy-alloy nanoparticles (HEA-NPs). Benefitting from the synergetic effect of copper ions released and thermal damage induced by photothermal conversion, HEA-NPs achieved the best anti-biofilm efficiency of 97.4% in the nutrient medium which was a worst-case scenario for the antimicrobial material.

A better understanding of the MIC mechanisms also sheds light on the novel methods for detection and monitoring of MIC, which is a worldwide knotty concern for field applications. We can provide stable and accurate sensors for detection and monitoring of MIC caused by electroactive microorganisms via optical fiber-based technology.

Inspired by the amorphous/crystalline feature within nacreous platelets, a mechanically robust antifouling coating composed of biopolymer-based hydrogel and dense metal-organic frameworks (MOFs) is developed. The resultant coating as a suitable reservoir exhibits a unique active defensive behavior of intelligent MOF degradation or drug release, enabling a ground-breaking performance for broad-spectrum biofouling and corrosion control.

Keywords: Microbiologically influenced corrosion; Biofilm; Antibacterial material; Anti-biofouling coatings

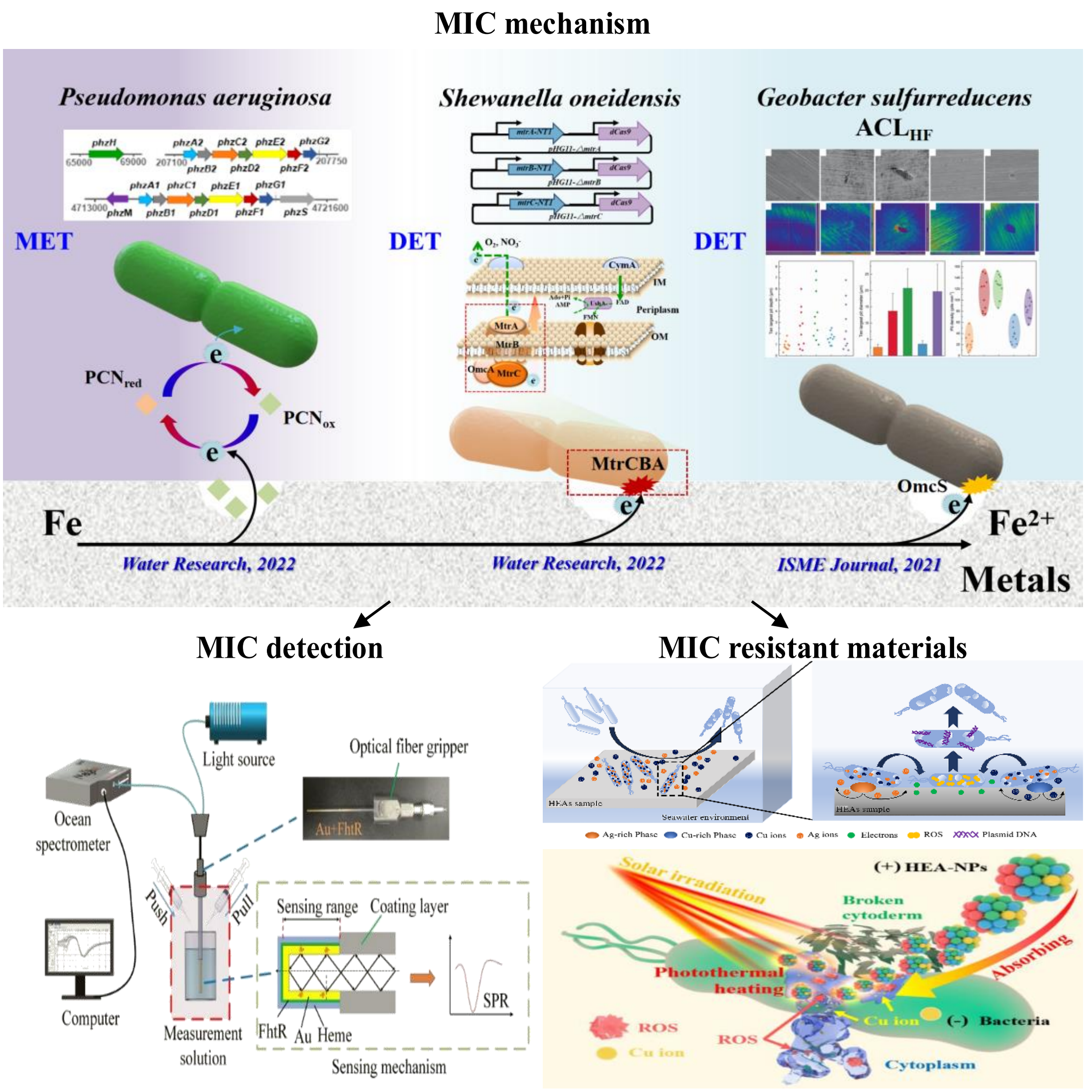

Figure 1. Demyth MIC mechanisms to design MIC-resistant materials and develop detection and monitoring technology.

KN3

BUILDING CONSENSUS: ADVANCING STANDARDS AND INNOVATION IN MICROBIOLOGICALLY INFLUENCED CORROSION AND BIOFILMS

Paulina D. Rakowska

National Biofilms Innovation Centre, University of Southampton, UK

Much of the world's industrial steel infrastructure is vulnerable to corrosion, with microbiologically influenced corrosion (MIC) being a major component. MIC affects multiple industries and has a significant economic impact, estimated at approximately $2.7 billion globally [1]. In the oil and gas industry, 20-40% of serious corrosion cases and 70-95% of pipeline internal leaks are attributed to MIC, with 20% of corrosion on metal surfaces caused by biofilms [2]. Despite the severity of this issue, MIC remains a largely intractable challenge, with fundamental scientific difficulties in its prediction, early detection, prevention, and control.

MIC is a complex process in which microbial biofilms play a crucial role. These biofilms can accelerate corrosion in various environments, including industrial settings such as pipelines, marine structures, and wastewater treatment facilities. One critical unmet need across all industry sectors dealing with MIC and biofilms is the infrastructure and support necessary to demonstrate alignment with relevant standards, guidelines, methods, and associated analytical competencies. The availability of agreed standards can facilitate translational innovation and stimulate business growth by allowing industries to make claims for their products and address global societal and environmental challenges.

Several challenges hinder the development of consensus in the biofilm and MIC communities [3]. The complexity of biofilm communities, their ubiquitous nature, and the broad range of affected sectors contribute to these difficulties. Consequently, there is a general belief that creating universal standards in this area may not be feasible. Various research groups and industrial stakeholders worldwide are addressing MIC, but discussions are often fragmented, information exchange is limited, and a true transdisciplinary and trans-sectoral approach is rarely seen. More initiatives like the Euro-MIC COST Action are needed.

This presentation will provide an overview of standards, standardisation processes, and some of the current initiatives within the community aimed at building consensus and accelerating the development of industry standards in the biofilms and MIC areas.

Keywords: Biofilms, MIC, Standards, Consensus

References:

[1] Cámara, M., et al. Economic significance of biofilms: a multidisciplinary and cross-sectoral challenge. *npj Biofilms Microbiomes,* **2022**, 8, 42. https://doi.org/10.1038/s41522-022-00306-y

[2] NACE International IMPACT Report, International Measures of Prevention, Application, and Economics of Corrosion Technologies **2016**, http://impact.nace.org/executive-summary.aspx

[3] NBIC Workshop Report. Microbe-Metal Interactions, **2019,** https://doi.org/10.5258/biofilms/007

KN4

BIOFOULING ASSESSMENT IN THE OFFSHORE RENEWABLE ENERGY SECTOR: CHALLENGES AND EXPECTATIONS

Pedro Vinagre[1]

[1] *WavEC Offshore Renewables, Environment & Licensing, Lisbon, Portugal*

WavEC Offshore Renewables has conducted several studies for the Offshore Renewable Energy (ORE) sector, antifouling companies, and environmental agencies aiming to support decision-making. The reason is that biofouling, namely macrofouling, (i) presents enormous costs to the ORE and other industries working in the marine environment, associated with equipment impairment and loss of functionality and performance, and (ii) has the potential to affect the ecosystems functioning and ecosystem services provision, for example by altering the type of habitat and enhancing the settlement and proliferation of non-native species.

This presentation intends to provide an overall understanding of the different aspects of macrofouling, with a focus on the ORE sector. The following topics will be tackled:

- WavEC at a glance
- Overview of biofouling-related legislation/regulation
- Macrofouling in the ORE
    - Definition and scope
    - Typical communities and dominant organisms
    - Factors influencing the communities’ structure and growth
    - Economic vs Ecological impacts
    - Control strategies
- WavEC’s case studies
    - Scope and design
    - Data acquisition, processing, and analysis
    - Reporting
- Future research demands

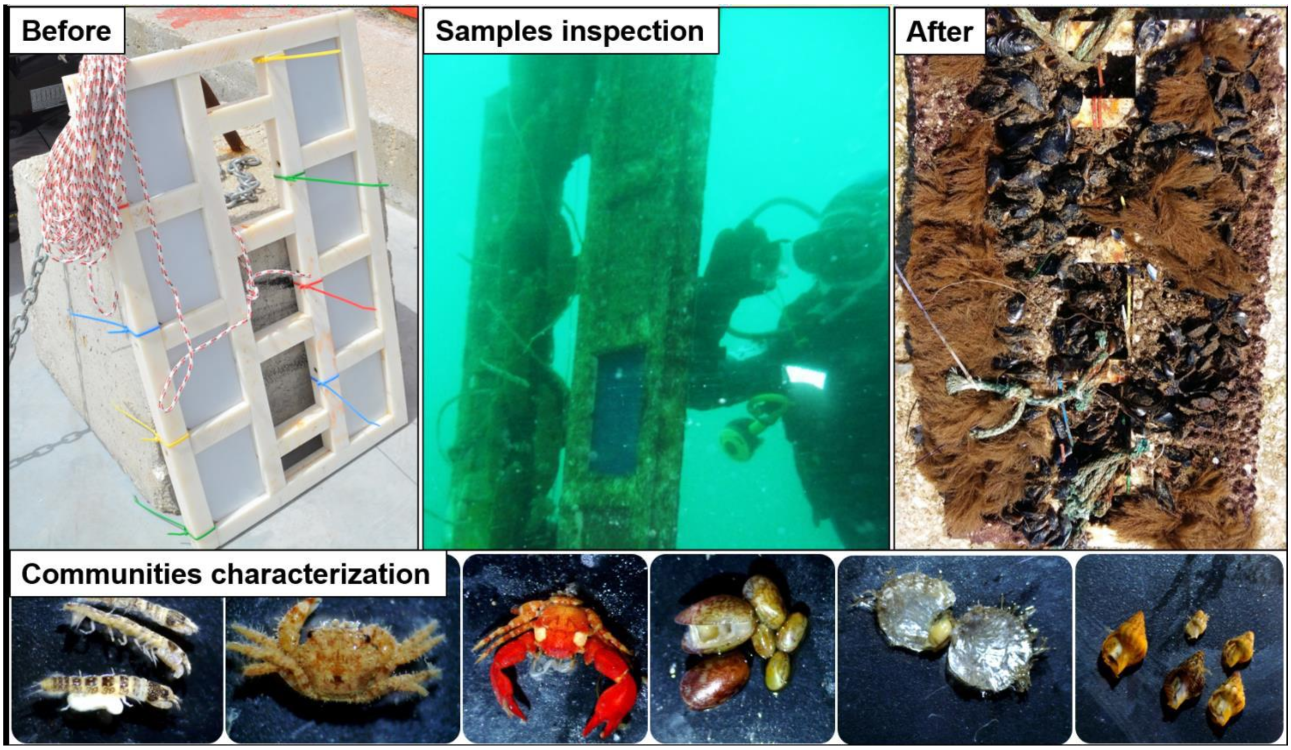

Figure 1. Overview of macrofouling assessment.

Keywords: Macrofouling; Offshore Renewable Energy; Assessment; Case studies

ORAL PRESENTATIONS

ORAL PRESENTATIONS

O1

APPLICATION OF MACHINE LEARNING TECHNIQUES FOR DETECTION OF MICROBIOLOGICALLY INFLUENCED CORROSION

Miroslav Milovanović[1], **Milena Rajić**[2], **Anđela Đorđević**[1], **Jelena Petrović**[1], **Saša S. Nikolić**[1]

[1] *University of Niš, Faculty of Electronic Engineering, Niš, Serbia.*

[2] *University of Niš, Faculty of Mechanical Engineering, Niš, Serbia.*

Microbiologically influenced corrosion (MIC) represents a challenge in various industries, particularly in sectors such as oil and gas, where infrastructure integrity is of the highest importance. In general, MIC occurs when microorganisms interact with metal surfaces, forming biofilms that accelerate corrosion processes. The importance of recognizing and mitigating MIC risks lies in its potential to cause significant damage and economic losses. Left unchecked, MIC can compromise the structural integrity of equipment and pipelines, leading to costly repairs, downtime, and safety hazards. Therefore, understanding the mechanisms and factors contributing to MIC is essential for developing effective mitigation strategies and safeguarding industrial assets against corrosion-related failures. The focus of this research is on application of Machine Learning (ML) techniques to recognize and quantify these factors contributing to MIC.

In the context of analysing factors contributing to MIC, several key considerations emerge. As a first point to highlight, understanding the microbial community involved is crucial. This community could include the types of microorganisms present, their metabolic activities, and their interaction with metal surfaces. Next, factors such as the composition and structure of biofilms, the presence of exopolymeric substances (EPS), and gradients of oxygen concentrations and pH values within biofilms play significant roles in MIC initiation and progression. In addition, environmental conditions (temperature, salinity, availability of nutrients) can significantly influence microbial activity and corrosion rates. Moreover, the material properties of the metal surface, including its composition, roughness, and surface energy, can impact susceptibility to MIC. Lastly, operational parameters such as flow rates, residence times, and fluid compositions must be considered as they affect the exposure of metal surfaces to corrosive environments and microbial colonization. Integrating these factors into ML models can facilitate the identification and quantification of MIC risks, enabling more effective corrosion management strategies.

To ensure optimal performance of ML models in recognizing and quantifying listed factors, careful preprocessing and preparation of MIC data are essential for the introductory phase. This includes data cleaning to remove noise and outliers, feature selection to identify the most relevant parameters influencing MIC, and normalization or scaling to standardize the data across different variables. Additionally, addressing missing values and imbalances in the dataset is crucial to prevent bias and improve model generalization. In addition, incorporating domain knowledge and expert insights during data preprocessing can enhance the interpretability and accuracy of ML models. Furthermore, validation techniques such as cross-validation and robust evaluation metrics should be employed to assess model performance and reliability. By ensuring high-quality input data, ML models can effectively harness the wealth of information contained within MIC datasets, facilitating more accurate predictions and informed decision-making for corrosion risk management strategies.

Focusing on types of algorithms that could be successfully utilized, the accent is on supervised and unsupervised models (Fig. 1). Supervised ML algorithms such as artificial neural networks, support vector machines, and decision trees can effectively analyze complex datasets comprising microbial, environmental, and operational parameters to identify patterns and correlations associated with MIC.

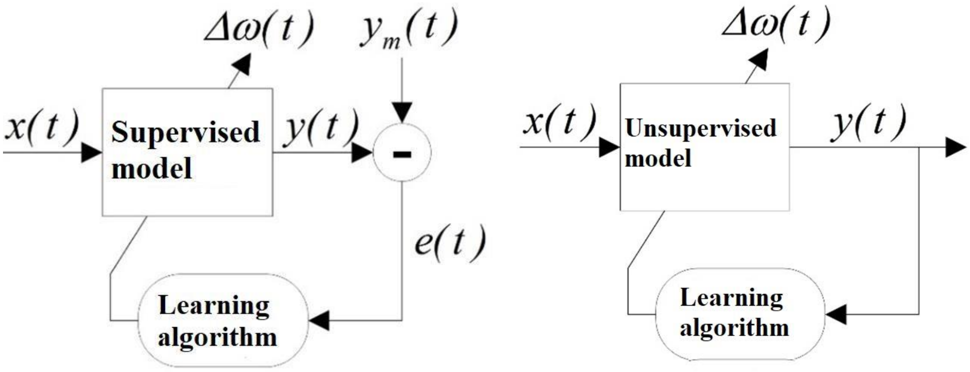

Figure 1. Supervised and unsupervised learning approaches of ML models

By training on historical data and incorporating appropriate features, supervised ML models can learn to predict MIC likelihood and corrosion rates. Focusing on unsupervised ML algorithms, clustering and dimensionality reduction techniques can reveal hidden structures within data, aiding in the discovery of novel insights and relationships. The adaptability of ML models allows for continuous learning and refinement, making them valuable tools for proactive MIC risk assessment and mitigation in various industrial settings.

Keywords: microbiologically induced corrosion; microbiologically induced corrosion factors; machine learning; supervised learning; unsupervised learning.

O2

ELECTROCHEMICAL PERFORMANCE OF AEROBIC MARINE BIOFILMS AND THE INFLUENCE OF NITRIC OXIDE ATTACHMENT CONTROL

Julian Wharton[1]

[1] *National Centre for Advanced Tribology at Southampton (nCATS), School of Engineering, University of Southampton, Southampton, Hants., SO17 1BJ, UK.*

The study presents a novel approach to managing biofouling in seawater piping systems, emphasizing the need for environmentally sustainable practices. By utilizing electrochemical impedance spectroscopy, the research offers a method to *in situ* monitor and evaluate biofilm formation, specifically focusing on the behaviour of *Pseudoalteromonas sp*. NCIMB 2021 within a controlled low laminar flow regime (Reynolds number, $Re \approx 1$). The biofilm attachment and growth to identify characteristic events sensed using a 200 μm diameter gold electrode surface. The findings suggest that the interfacial capacitive component could serve as a reliable indicator of biofilm presence and density, providing a quantitative measure that correlates well with confocal microscopy. This dual-analysis technique could be instrumental in developing early warning systems for biofouling, allowing for timely intervention with environmentally friendly biofilm disruptors utilising quorum sensing molecules to induce active biofilm dispersal. The study explored using sodium nitroprusside, a nitric oxide donor, as a potent coordinated disruptor and demonstrates its efficacy in dispersing biofilms, thus highlighting the role of nitric oxide in biofilm regulation. The electrochemical performances of a 72 h old biofilmed gold surface dosed with nanomolar concentration; 500 nM, of the nitric oxide donor, see Figure 1. This research demonstrates a viable strategy for in-service biofilm management that balances operational efficiency with ecological considerations.

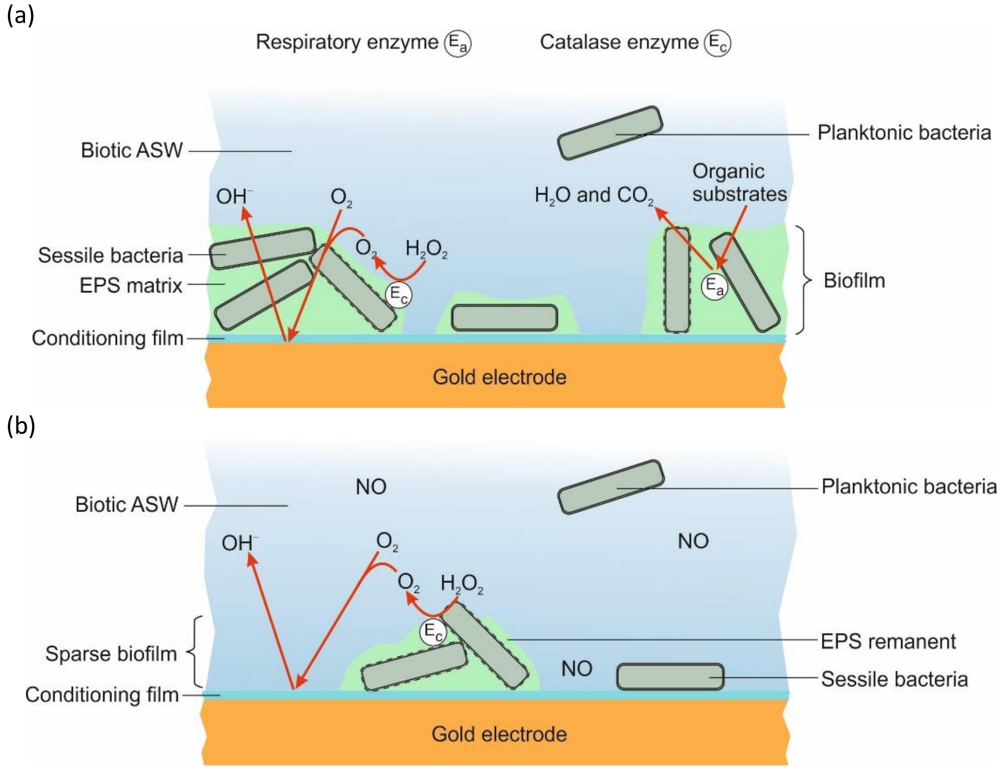

Figure 1. Electron transfer pathways within a *Pseudoalteromonas* biofilm: **(a)** biofilm growth and colonisation under aerobic biotic artificial sweater (ASW) conditions and **(b)** biofilm dispersion/disruption for aerobic biotic ASW with 500 mM NO donor.

Keywords: Marine biofilms; electrochemical; impedance; sensing; NO dosing.

Acknowledgements: Funding is gratefully acknowledged from the Defence Science and Technology Laboratory (Dstl).

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O3

EXPLORING BIOMIMETIC TEXTURING FOR ANTIFOULING APPLICATIONS: INSIGHTS FROM A COMPUTATIONAL FLUID DYNAMICS (CFD)

Samira Nazari, Lilibeth Zambrano, David Culliton

Aerospace and Mechanical Engineering Department, South East Technological University (SETU), Carlow Campus, Ireland

Biofouling and Microbially Influenced Corrosion (MIC) are two distinct processes that involve the growth and colonisation of microorganisms on surfaces. However, the initial stages of both MIC and biofouling are closely related to biofilm-formation due to the accumulation of microorganisms on exposed surfaces [1]. It can therefore be surmised that similar prevention strategies can be employed in both processes. In this study, we focussed on one of the most common passive anti-fouling/-MIC treatments: biomimetic antifouling coatings, which are morphologically inspired by natural structures and their physical and mechanical properties. Nature-derived biomimetic structures can exhibit either hydrophobic or hydrophilic behaviour. Such behaviour can then be enhanced using materials with similarly-programmed surface energies, to produce superhydrophobic or superhydrophilic surfaces. Superhydrophobic surfaces, such as lotus leaves, are water-resistant / repellent and have been promoted in research as one of the best options solutions for anti-fouling [2], anti-corrosion [3], and self-cleaning [4] surface treatments. There is a dearth of numerical studies investigating superhydrophobicity, with the majority of the studies limited to experimental investigations. In this study, we intend to model selected biomimetic surfaces, the results of which will better inform experimental analysis of these systems. This analysis will be based on computational fluid dynamic (CFD) simulations.

Preliminary simulations were conducted on three different micro-textured superhydrophobic surfaces with the same static contact angle of 150°, which investigated the impact of micro-textured shapes and density on the resultant spreading diameter. The morphology of the selected microtextures was either cubic, cylindrical, and hemispherical shapes (see Fig. 1). The Volume of Fluid (VOF) method was used to simulate a water droplet impacting on the micro-textured superhydrophobic surface, with a Weber number of zero. Each superhydrophobic micro-texture had three different densities, which are established by changing the distances between two adjacent micro-textures, which ranged from 10 to 30 microns. Preliminary results show that cubic structures with a density of 156 textures/mm^2 have more superhydrophobic features and a 54.4% smaller spreading diameter compared to a flat surface.

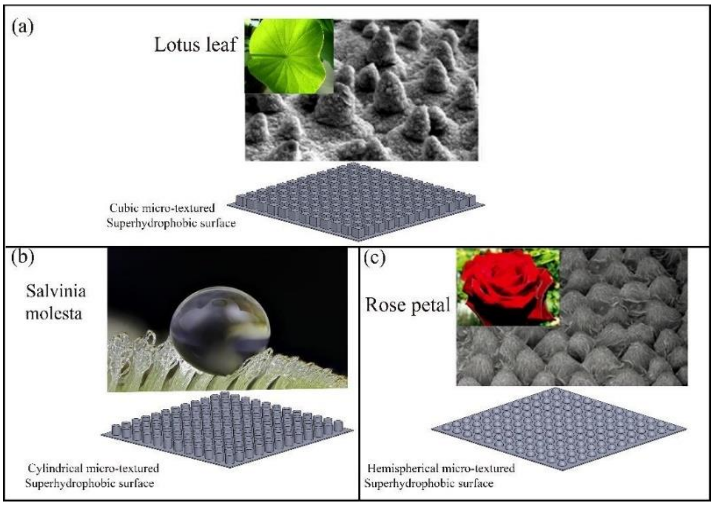

Figure 1. Micro-textured superhydrophobic surfaces (a) Cubic: inspired by Liu, et. al [5] (b) Cylindrical: inspired by © Science Photo Library (c) Hemispherical: inspired by Liu, et. al [5].

Keywords: Biomimetic coatings; Superhydrophobic surfaces; Numerical study; VOF method.

Acknowledgements: This Project has been funded by the Sustainable Energy Authority of Ireland (SEAI) through the SPOTBlade Project (Grant no. SEAI 21/RDD/671).

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O4

CASE STUDY ON PROGRESSIVE CORROSION OF CARBON STEEL COOLER TREATED WITH EPOXY-CERAMIC COATING

Veronika Hlavackova[1], **Petr Koutnik**[2], **Alena Sevcu**[1]

[1] *Technical University of Liberec, Institute for Nanomaterials, Advanced Technologies and Innovation, Dept. Applied Biology, Liberec, Czech Republic;*

[2] *ORLEN UniCRE a.s., Ústí nad Labem, Czech Republic*

Formerly, corrosion attack and/or products were found when cleaning the cooler. This motivated a case study to determine the mechanism of degradation and specify the reasons for progressive corrosion. The cooling system was fabricated in 1968 and made from carbon steel EN 1.0405 (dimensions: l = 6m, d = 0,4 m). Since the early observation of corrosion, the cooler had been repeatedly treated with commercially available repair composite to prevent further corrosion. Samples were taken from the cooler heads. Cooling water, tubercles and swabs were sampled aseptically and cooled down immediately. The microstructural, chemical and microbiological analyses were performed. Visual appearance of the surface of cooler tube after removing the cooler head revealed presence of orange to dark brown tubercles of corrosion products. After removal of tubercles and cleaning, the inspection of the surface has shown uneven corrosion in the form of pits with dimensions of up to tens of mm penetrating to a depth of mm units (Figure 1). XRD analysis has proved that the only identified mineral phase was goethite (α-FeO(OH)). Chemical analysis of the cooling water proved to be full of potential electron donors (organic matter, ammonium) and acceptors (sulfates, nitrates) for the microbial metabolism. The chemical composition of cooling water rich in organic compounds, sulfates, nitrates etc. hosted preferentially microorganisms typical for wastewaters, fermenters and nitrogen metabolizing microorganisms, but sulphur oxidizing bacteria, and iron reducing and oxidizing bacteria were also detected. In the carbon steel cooler we observed a more significant increase in ammonia and sulfur oxidizing representatives. Bacteria reducing or oxidizing nitrogen compounds were detected in the cooling water using qPCR. The coexistance of microorganisms oxidizing ammonia dissolved in water and members of family *Methylophilaceae*, here enriched in biofilms, ressembles the environment of wastewaters (1–3). Carbon steel corrosion has previously been associated with the occurrence of other methylotrophic bacteria - *Methyloversatilis* - under anaerobic conditions (4). The exact relationship of oxygen and other electron acceptors in relation to their putative involvement in induction of steel corrosion is not fully understood. The role of anaerobic microorganisms present in the biofilm, which are represented here by the poorly described and still uncultivated genera, cannot be sofar resolved. Advanced experimental program to better describe the role of indicated genera in corrosion induction must be performed in future.

Figure 1. The surface of the carbon steel cooler, after the removal of corrosion products, indicates a progressive degree of corrosion.

Keywords: cooler; carbon steel; ammonium; methylotrophic bacteria; wastewater

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O5

MARINE MICROBIAL INDUCED MINERALIZATION: EXPLORING MECHANISMS FOR BIOINSPIRED ANTICORROSION SOLUTIONS

M. J. F. Marques[1, 2], S. Zanna[3], P. Marcus[3], R. Basseguy[2]

[1]*LGC, Université de Toulouse, CNRS, INP, UPS, Toulouse, France*

[2] *LNEG, Laboratório de Materiais e Revestimentos, Lisboa, Portugal*

[3] *IRCP-PCS, Chimie Paris Tech-CNRS, PSL Research University, Paris, France*

In the field of corrosion, nature has proved to be a source of inspiration for developing new green protection concepts. Over the last decades, new emerging microbial-based technologies have been studied, based on the recognition that microorganisms can influence corrosion behaviour in an advantageous way, the so-called MICI (microbiologically influenced corrosion inhibition), opening different lines of research (1-5). In particular, the process of biomineralisation on the surface of materials was recently highlighted as a promising new environmentally-friendly approach to produce protective coatings.

In this context, the ANR MICOATEC project (2020/2024, https://micoatec.eu/) has proposed a new approach for developing anticorrosion solutions based on a biomimetic strategy. The guiding idea of MICOATEC was to draw inspiration from the interactions between an Al-Mg alloy and the marine environment, which lead to the formation of a protective layer. The present work proposed the used of combined surface characterization techniques to understand the natural biomineralization process that develops on the surface of Al-Mg alloy when immersed in a marine biotic environment.

A very interesting finding was the identification of different layer structures developed on the alloy surface, depending on whether it is exposed to direct sunlight or shade (light and dark side). On the surface exposed to sunlight, photosynthetic micro- and macro-organisms, such as green, red, and brown algae ("soft fouling"), induce the formation of a dual layer structure, in which a hydrated Mg-rich outer layer with extracellular polymeric substances (EPS) on the top has proven to play a role in the corrosion inhibition process (figure 1). No localised attack was observed over the course of 6 months of immersion of the Al alloy. In contrast, surfaces exposed on the dark side, were found to be covered mainly with a "hard-fouling" composed by barnacles and calcareous tube worms, which generate a single thick layer of Al/Mg oxide containing Ca, with a less protective behaviour. Localised corrosion of the Al-Mg surface was observed after two months of seawater immersion.

To sum-up, the formation of the protective layer on the Al-Mg surface exposed to the light side in biotic conditions is shown to be related to the soft fouling developed on the metallic surface. Indeed, marine algae are a source of sulphated polysaccharides (SPs), whose hydrophilic properties enhance absorption and retention of water, favouring the formation of the hydrated Mg oxide/hydroxide rich outer layer. This outer layer has proven to play a role in the corrosion inhibition process as a barrier to the penetration of Cl^-.

In conclusion, the MICOATEC results show that the use of biomineralisation to inhibit corrosion may become an important method in the field of corrosion prevention.

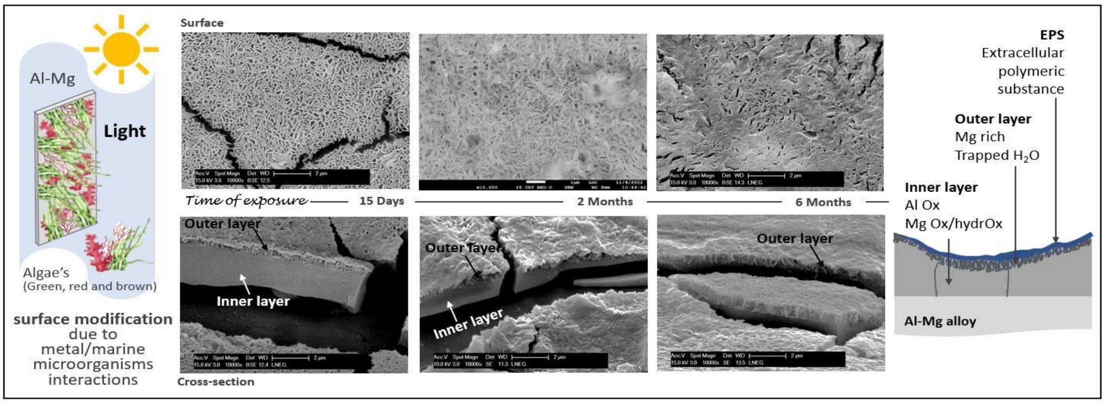

Figure 1. SEM observations of Al-Mg surfaces after seawater immersion on the light of exposure. Cross- section scheme based on the results of the different characterisation techniques involved in the work.

Keywords: Aluminium alloy; Corrosion inhibition; Biomineralisation; Marine algae/EPS; Bioinspired protection solution

Acknowledgements: This work was financially supported by the ANR, in the framework of the MICOATEC project (ANR-19-CE08-0018) coordinated by R. Basséguy-LGC.

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O6

CHITOSAN-BASED COATINGS FOR MARINE BIOFILM PREVENTION

Marta Lima[1,2], Luciana C. Gomes[1,2], Rita Teixeira-Santos[1,2], Maria João Romeu[1,2], Jesus Valcarcel[3], José A. Vázquez[3], Miguel A. Cerqueira[4], Lorenzo Pastrana[4], Ana I. Bourbon[4], Ed D. de Jong[5], Sjelmer Sjollema[5], Filipe J. Mergulhão[1,2]

[1] *LEPABE - Laboratory for Process Engineering, Environment, Biotechnology and Energy, Porto, Portugal;*

[2] *ALiCE - Associate Laboratory in Chemical Engineering, Porto, Portugal;*

[3] *Grupo de Reciclado y Valorización de Materiales Residuales (REVAL), Instituto de Investigaciones Marinas (IIM-CSIC), Vigo, Spain;*

[4] *International Iberian Nanotechnology Laboratory, Braga, Portugal;*

[5] *Department of Biomedical Engineering, Groningen, The Netherlands.*

Biofouling is a significant issue in marine industries, causing economic losses due to increased fuel consumption and maintenance costs, as well as ecological concerns related to the spread of invasive species and alteration of marine ecosystems. The search for non-biocide-release antifouling coatings has been on the rise, with chitosan (CS)-based surfaces proving to be a more sustainable and environmentally friendly solution.

CS is a natural polymer that has been widely used because of its outstanding biological properties, including antimicrobial activity, biocompatibility, and low toxicity. We have produced and characterised poly (lactic acid) (PLA)-CS surfaces with CS of different molecular weights (Mw) at different concentrations for application in marine paints. *Loligo opalescens* pens, a waste from the fishery industry, were used as a CS source. The antimicrobial activity of the CS and CSfunctionalised surfaces was assessed against *Cobetia marina* (a model proteobacterium for marine biofouling) over 7 weeks under controlled hydrodynamic conditions that mimic marine environments. The results demonstrated that CS targets the bacterial cell membrane, and PLA-CS surfaces were able to reduce the number of culturable cells by up to 68% compared to the control, with this activity being dependent on CS Mw. The antifouling performance was corroborated by Optical Coherence Tomography (OCT) since PLA-CS surfaces reduced the biofilm thickness by up to 36%, as well as the percentage and size of biofilm empty spaces [1].

Overall, CS coatings are a promising bio-friendly strategy for mitigating biofouling in marine settings, contributing to the valorisation of fishing waste.

Keywords: marine biofouling; antifouling coatings; *Cobetia marina*; chitosan; marine waste.

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O7

POLYOXOMETALATE IONIC LIQUIDS (POM-ILS) AS PROTECTIVE COATINGS FOR CULTURAL HERITAGE AGAINST ACID CORROSION AND BIODETERIORATION

Archismita Misra[1], Isabel Franco Castillo[2], Mariella Malefioudaki[2], Scott G. Mitchell[2]*, Carsten Streb[3]*, Andrea Koerdt[1]* and Oliver Hahn[1]*

[1] Bundesanstalt für Materialforschung und –prüfung (BAM), Unter den Eichen 87, 12205 Berlin, Germany

[2] Instituto de Nanociencia de Materiales de Aragon (INMA-CSIC) and CIBER-BBN, CISCUniversidad de Zaragoza, 50019 Zaragoza, Spain

[3] Johannes Gutenberg University Mainz, Duesbergweg 10-14, 55128 Mainz, Germany

Corrosion of stone by acid rain and deterioration from microbial biofilms are challenges pertinent worldwide for industrial or residential buildings as well as cultural heritage artefacts, like statues or historic buildings. One mitigation option might be the use of thin transparent films of polyoxometalate-based ionic liquids (POM-ILs). In this regard, different limestone samples were coated with hydrophobic, acid resistant POM-ILs which also have biocidal properties.[1] Exposure of the samples to simulated acid rain showed negligible corrosion compared to the significant deterioration of unprotected samples (Fig 1. Left). In addition, the biocidal properties of the POMILs suppress the formation of biofilms on coated stone slabs. The coating is mechanically stable and is not removed even by harsh mechanical and chemical treatment. Following studies successfully explored the effectiveness of the coating against lampenflora growing in the Pommery Champagne cellar [2] (Fig 1. Right); and the long-term performance of POM-ILs under outdoor environmental conditions [3].

So, POM-ILs are already proven to possess remarkable anticorrosion and antimicrobial properties against *aerobic microorganisms* and being water-insoluble, they don't get leached into aquatic ecosystem, which is extremely beneficial from an environmental sustainability and toxicological point of view.

The current project aims to continue the journey on protecting the cultural heritage, shifting focus from stones to metals and employ functional POM-IL nanocoatings to prevent MIC (Microbiologically Influenced Corrosion) of cultural heritage artefacts made of metal or metal alloy like carbon steel, brass, cast iron or bronze.

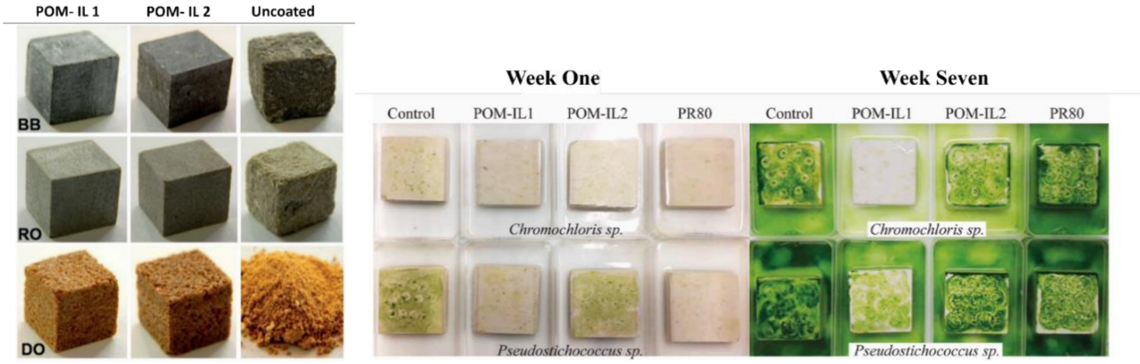

Figure 1. Left: Acid-vapour corrosion protection of two different POM-IL-coated stone samples compared to the non-coated controls. RO: Romery stone; BB: Belgian Blue stone; DO: Dom stone.[1]; **Right**: Inoculated controls and coated chalk slabs from Pommery Champagne cellar with *Chromochloris sp.* (upper slabs) and *Pseudostichococcus sp.* (lower slabs) at week one and week seven of incubation.[2]

Performance of both the coating materials and coating techniques via optimization of the adhesion of the nanocoating on the metallic surface on the corrosion rate and corrosion products in the MIC caused by *anaerobic microorganisms* like methanogenic archaea or Sulphate Reducing Bacteria (SRB) would be tested. The objective is to establish POM-ILs as efficient environmentally sustainable nanocoating materials against biocorrosion citing the already published success stories; and sketch the ongoing endeavours and prospects of these very efficient candidates in the context of MIC mitigation.

Keywords: Polyoxometalate; Ionic Liquid; Corrosion; Biodeterioration; Cultural Heritage

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O8

INVESTIGATING THE USE OF FRESHWATER IN SIMULATIONS OF WATER COMPENSATED FUEL TANKS TO IMPEDE CORROSIVE ACTIVITY BY SRB

Gareth J. Williams[1*] and Graham C. Hill

[1]ECHA Microbiology Ltd., Units 22&23, Willowbrook Technology Park, Llandogo Road, Cardiff, CF3 0EF, WALES, UK

**gwilliams@echamicrobiology.com*

Seawater is introduced into fuel tanks on some ships as the fuel is being utilised in order to maintain vessel stability. Sulphate is abundant in seawater and can be used as an electron acceptor by a range of sulphide generating microbes. Consequently, seawater compensated fuel tanks are susceptible to microbial sulphide generation and Microbiologically Influenced Corrosion (MIC).

A review was conducted to identify potential strategies to impede corrosive activity by Sulphate Reducing Bacteria (SRB) in seawater compensated fuel tanks on naval vessels. One of the strategies identified was the use of freshwater, instead of seawater, as compensate water. This could reduce the availability of essential nutrients for microbial growth and the amount of sulphate available for microbial sulphate reduction. Initial review established that the use of freshwater as compensate water was feasible, although the ingress of some seawater is inevitable in some operational circumstances. Therefore, a laboratory study was conducted to investigate the overall effectiveness of the strategy in simulations of seawater compensated fuel tanks. The amount of seawater ingress that might be tolerated without a corresponding significant increase in SRB activity was also explored.

Microcosms containing naval distillate fuel and water, comprising of varying proportions of seawater and freshwater, were set up and inoculated with field samples containing a consortium of microorganisms, including SRB. The extent of SRB growth, physico-chemical conditions, and levels of sulphide were monitored. Microcosm samples also underwent qPCR analysis to determine the abundance of SRB and other microorganisms implicated in corrosion. Under the conditions of the study, levels of microbial sulphide generation were significantly lower when seawater made up less than 25% of the compensate water.

O9

INTEGRATING MULTIPLE LINES OF EVIDENCE (MLOE): ESSENTIAL FOR DIAGNOSIS OF MICROBIOLOGICALLY INFLUENCED CORROSION (MIC)

Torben L. Skovhus[1], **Richard B. Eckert** [2]

[1] *VIA University College, Research Centre for Built Environment, Climate, Water Technology and Digitalisation, Banegaardsgade 2 DK-8700, Horsens, Denmark*

[2] *Microbial Corrosion Consulting, LLC, 2227 Palmetto, Commerce Township, MI, USA 48382*

It is well established that multiple lines of evidence (MLOE) are required to distinguish between abiotic corrosion and microbiologically influenced corrosion (MIC). The primary categories of evidence include 1) operation and design information, 2) the chemical environment, 3) the microbiological environment, and 4) materials and corrosion products. However, experience has shown that asset owners are basing their corrosion diagnosis on inadequate data and that clear guidance for integrating corrosion related data is lacking. Here, we discuss approaches that can be used to analyze and integrate MLOE for determining the causes of corrosion damage, as guided by the latest research, consensus standards, and by employing advanced analytical techniques and models. Accurate diagnosis of corrosion failures and operating assets is deemed essential, not only for identifying effective mitigation and prevention, but also as a vital step towards advancing materials sustainability.

Keywords: Microbiologically Influenced Corrosion, MIC, Multiple Lines of Evidence, Data Integration

O10

PILOT STUDY OF A LABORATORY-BASED MICROBIOLOGICALLY INFLUENCED CORROSION (MIC) TEST USING MICROBIOLOGICAL CONSORTIA SAMPLED FROM THE FIELD

Sara Taghavi Kalajahi[1], Maria Salta[2], Nanni Noël-Hermes [2], Torben Lund Skovhus[3], Andrea Koerdt[1]

Federal Institute for Materials Research and Testing (BAM), Berlin, Germany

[1] ENDURES B.V., Den Helder, The Netherlands

[3]VIA University College, Banegårdsgade 2, DK-8700 Horsens

Experts widely agree that the verification of microbiologically influenced corrosion (MIC) require the use of multiple lines of evidence (MLOE), including metallurgical, microbiological, operational/historical and environmental/chemical aspects. Existing standards related to MIC, such as NACE TM0106, TMO212 and TM0194, primarily aim to offer guidance on gathering such evidence. Working Group 5 of Euro-MIC COST action 20130 (Achieving standardization) proposed a laboratory-based approach that directly confirms the capability of the sampled consortium of microorganisms, obtained from the specific field location under investigation, to increase and/or alter the corrosion processes. It is necessary to verify this laboratory approach by conducting it in different laboratories, performed by different scientist. In the current study, the experiments based on the laboratory protocol of WG5 were conducted at Endures's laboratory using environmental samples (sediment and seawater) from the North Sea. Metal coupons (carbon steel C1010) were exposed to sediment and seawater samples with varying microbial inoculums and controls over 150 days. Microbial identification, corrosion product analysis, and surface morphology assessments were conducted using molecular and microscopy techniques. The results indicate distinct microbial influences, particularly in treatments containing yeast, which fostered heightened anaerobic activity, notably by sulfate-reducing bacteria (SRB). Weight loss measurements and surface analysis revealed elevated corrosion rates and susceptibility to pitting corrosion in microbial inoculum treatments, with yeast presence exacerbating corrosion processes. Elemental analysis confirmed the presence of iron sulfide in corrosion byproducts, affirming SRB activity. The findings emphasize the importance of employing a MLOE approach and investigating pitting corrosion to better understand the impact of microorganisms on metal corrosion in the marine environments. This study contributes to advancing knowledge in the field and developing standardization: the lab-to-field transition for MIC testing.

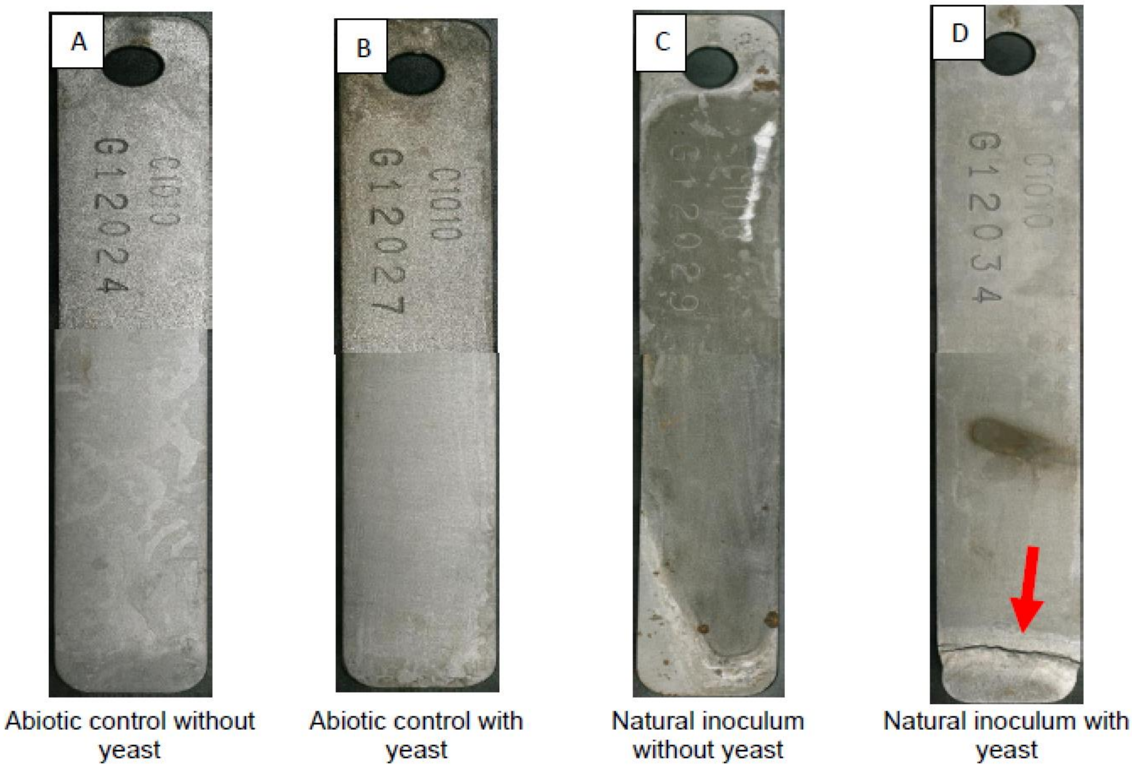

Figure 1. The appearance of coupons after cleaning. A B, and C) Controls and natural inoculum without yeast did not show any dramatic surface changes (pitting corrosion). D) Natural inoculum with yeast in the lower part of coupon (red arrow), which was closer to the sediment and anaerobic part of experimental system showed an obvious loss.

Keywords: Microbiologically influenced corrosion (MIC), Multiple lines of evidence (MLOE), Achieving standardization, Marine Corrosion.

Acknowledgements:

We would like to express our sincere gratitude to the Euro-MIC Cost Action CA20130 for providing the invaluable opportunity to conduct this Short-Term Scientific Mission (STSM). Their support has been instrumental in advancing our research in the field of MIC in marine environments. Additionally, we extend our heartfelt thanks to Endures Company for their generous provision of materials and necessary samples for this study, as well as the scientific advice and technical assistance provided throughout this work. Their expertise and guidance have been indispensable in carrying out the experiments and analysing the results effectively.

O11

INVESTIGATING THE EFFICACY OF THPS AND GLUTARALDEHYDE TREATMENT FOR ENHANCED PERFORMANCE ON NORTH SEA OFFSHORE SYSTEM

Pernille Ruby Lauritsen[1], Tanmay Chaturvedi[2]

[1,2] Aalborg University, Esbjerg, Denmark

The use of biocides plays an important role in maintaining the integrity of critical oilfield systems. At the North Sea offshore system, several locations have a history of elevated bacterial levels and significant corrosion, as observed by the intelligent pigging inspections. Common techniques for biocide qualification focus on the assessment of how effectively biocides minimize microbial activities, specifically sulphate-reducing bacteria (SRB). However, such testing cannot adequately answer the question of whether a particular biocide treatment regime can be expected to protect seawater injection systems against microbiologically influenced corrosion (MIC).

The optimization of biocide treatments is a challenge, as the efficacy of biocides can be influenced by various factors, including concentration, duration, and frequency of application. Therefore, this study conducts a thorough investigation into the efficacy of the treatment conditions for two frequently employed biocides, Tetrakis Hydroxymethyl Phosphonium Sulfate (THPS) and Glutaraldehyde. The primary objective is to enhance the understanding of biocide treatments, tailoring concentration and duration to match each biocide unique mechanism.

The study employs the use of anaerobic reactors for biofilm growth as a part of the setup to mimic offshore environment. Mixed microbial culture from Wadden Sea sediment is used as inoculum in Postgate medium to populate the media with sulphate reducing bacteria and archaea. The assessments of the biocide effectiveness are based on multiple lines of evidence (MLOE), to evaluate the efficacy of biocide treatments. Both indirect and direct assessments, including 16S/18S ITS rRNA sequencing, quantitative polymerase chain reaction (qPCR), adenosine triphosphate (ATP) activity, hydrogen sulphide (H_2S) concentrations levels, and surface corrosion and pitting rate analysis, are utilized to comprehensively assess the impact of biocide treatments on microbial communities and corrosion rates.

Through the integration of these diverse analytical techniques, a holistic understanding of the efficacy of the biocide treatments on microbial communities and corrosion rates are achieved. The

MLOE approach provides valuable insights into the complex interactions between biocides, microbial communities, and corrosion rate in offshore environments. By integrating the data, this study aims to elucidate the relationship between microbial activities and corrosion rates in order to ultimately inform a more targeted and effective biocide treatment strategies for offshore oilfield systems.

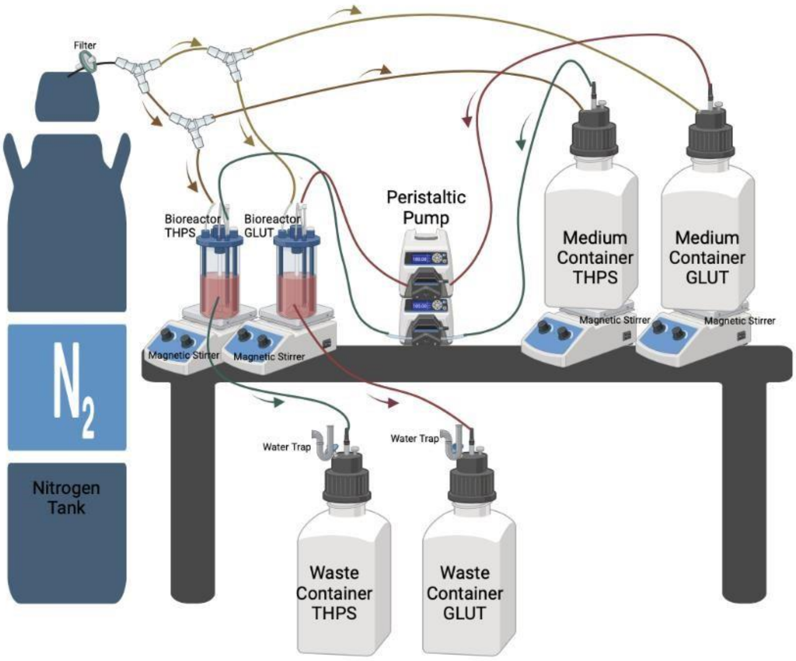

Figure 1. Setup of the anaerobic/sealed CDC biofilm reactors designed to mimic offshore environment, facilitating biofilm growth.

Keywords: Biocide; Tetrakis Hydroxymethyl Phosphonium Sulfate (THPS); Glutaraldehyde; Microbiologically Influenced Corrosion (MIC); Sulphate-Reducing Bacteria (SRB).

O12

DUAL ANAEROBIC REACTOR MODEL TO STUDY BIOFILM AND MICROBIOLOGICALLY INFLUENCED CORROSION INTERACTIONS ON UNS G10180 CARBON STEEL

Liam Jones[1*], Maria Salta[2], Torben Lund Skovhus[3], Kathryn Thomas[4], Timothy Illson[4], Julian Wharton[5], Jeremy Webb[1,6]

[1]*School of Biological Sciences, University of Southampton, Southampton, United Kingdom*

[2]*Endures, MIC and Biofilm Department, Den Helder, The Netherlands*

[3]*Research Centre for Built Environment, Climate and Water Technology, VIA University College, Horsens, Denmark*

[4]*DNV, Holywell Park, Ashby Road, Loughborough, United Kingdom*

[5]*School of Engineering, University of Southampton, Southampton, United Kingdom*

[6]*National Biofilms Innovation Centre, United Kingdom*

***** Corresponding Author: lmj1n20@soton.ac.uk

Continual challenges due to microbial corrosion are faced by the maritime, offshore renewable and energy sectors. Understanding the biofilm and microbiologically influenced corrosion interaction is hindered by the lack of robust and reproducible physical models that reflect operating environments. A novel dual anaerobic biofilm reactor, using a complex microbial consortium sampled from a marine littoral sediment, was used to investigate biofilms under conditions that mimic the MIC environment of interest. Overall, the aim is to develop a reproducible bioreactor-based model for MIC that will be able to: (i) gain new scientific insight and mechanistic MIC understanding that will improve predictive measures including next-generation sequencing of the MIC-associated microbiome and (ii) progress the first standard model and biofilm-relevant test method for the industry. Moreover, the protocol incorporates a multi-disciplinary approach, using multiple lines of evidence (MLOE) to gain a holistic understanding of biofilms and MIC. Electrochemical data indicated that the biofilm caused an increased incidence of localised pitting, resulting in active pit growth. Coupon analysis further confirmed that the biotic reactor had a significantly greater pit density, with a greater pit depth and size. Critically, DNA extraction and 16S rRNA amplicon sequencing demonstrated the principal biofilm activity was due to respiratory electrogens, specifically sulphate reducing and iron reducing bacteria. The dual anaerobic reactor model provided key insights into the different abiotic and biotic corrosion mechanisms under relevant environmental conditions.

From this, we can start to rationalise an idealised representation of the mixed-species biofilm and the microbial mechanisms that lead to corrosion of carbon steel under anoxic conditions at the electrode/electrolyte interface. Using multiple lines of evidence to gain a holistic understanding of biofilms and MIC, more sustainable prevention and mitigation strategies can be designed. Hopefully, the novel dual bioreactor protocol described here helps to progress the first standard model and biofilm-relevant test method for biocide efficacy testing within industry.

Graphical Abstract

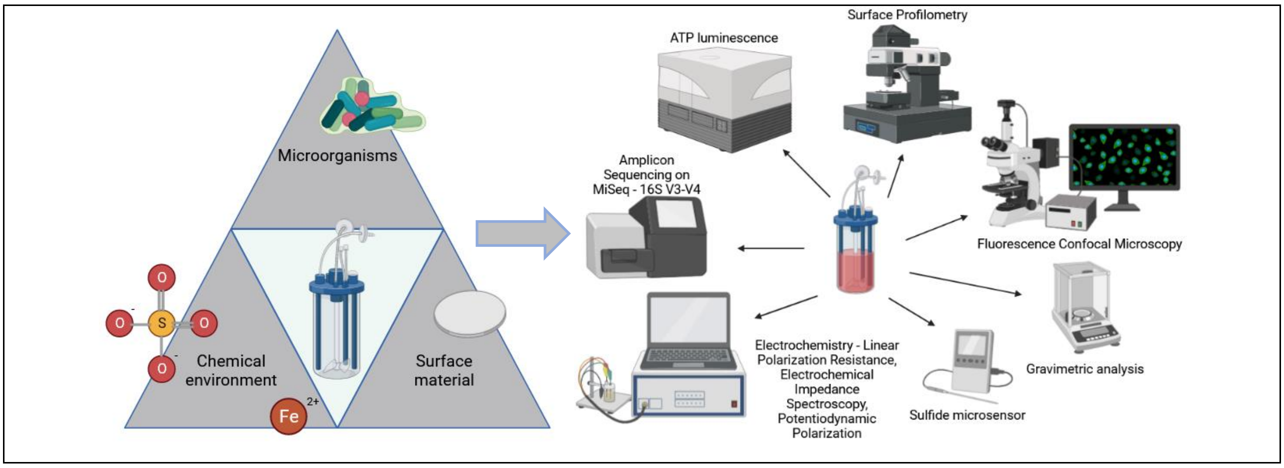

Figure 1. Graphical illustration of the research design, which incorporates multiple lines of evidence (MLOE) to research environmentally relevant biofilms and microbiologically influenced corrosion.

Keywords: biofilms1, microbiologically influenced corrosion2, electroactive microorganisms3, standardisation4, carbon steel5, multiple lines of evidence6.

Acknowledgements:

This work was supported by the South Coast Biosciences Doctoral Training Partnership (SoCoBio DTP), a Biotechnology and Biological Sciences Research Council (BBSRC) funded research training programme (reference number BB/T008768/1) in affiliation with DNV, and the National Biofilms Innovation Centre (NBIC).

O13 - INVITED TALK

CHALLENGES AND OPPORTUNITIES IN CORROSION MITIGATION TO MEET THE DECARBONISATION JOURNEY.

Martyn Wright[1], Heather Hughes[1], James Kwame[1]

[1] Lloyd's Register Group Limited, Global Technology Centre, Southampton/ United Kingdom

Calls from the 27th session of the Conference of the Parties of the United Nations Framework Convention on Climate Change (UNFCCC-COP 27) to pursue further efforts to limit the global temperature increase to 1.5 °C [1], has intensified the need to invest in technologies aimed at decarbonisation. With shipping contributing close to 3% of global greenhouse gas (GHG) emissions, there is the need to find alternative energy solutions to meet the 2050 Net-zero target set by the International Maritime Organisation [2]. LR in their role as maritime trusted advisors have carried out alternative fuels assessment and published rules for ships using alternative fuels such as ammonia, hydrogen, methanol, ethanol, liquified natural gas and liquified petroleum gas [3, 4], in support of the industry decarbonisation efforts. The need to decarbonise in these new environments also present significant challenges such as corrosion of deployed structural materials which poses significant risk to the safety of lives and assets. Providing the needed assurance for structures will require adequate understanding of the corrosion mechanisms and control methods in different natural environments, among others [5]. Current industry limitations in the availability of the requisite regulations and the general lack of standardisation governing product specifications such as the quality of CO_2 grades, present significant challenges in understanding their effect on corrosion and phase change during transportation and storage [6]. For ships operating across different marine environments, microbial adhesions such as biofouling results in significant ship GHG emissions [7]. Microbiological corrosion and biofouling have also been identified as the two main causes of marine corrosion posing major concern to the integrity of structures [8].

This brief session provides an insight into the main corrosion and regulatory challenges in energy transition in the efforts to decarbonise the maritime industry.

Keywords: Maritime industry, Corrosion, decarbonisation, energy transition, regulations

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O14

NEW STRATEGIES FOR ACTIVE CORROSION PROTECTION OF ALUMINIUM PARTS BASED ON ENCAPSULATED INHIBITORS

Andreu Ruiz[1], Antonio Montoliu[1], Frederico Maia[2], Marc Pallarès[3], Amaia Querejeta[4], Leticia Cabrera[5]

[1] Chemplate Materials SL, Santa Perpètua de Mogoda, Barcelona (Spain);

[2] Small Materials and Technologies Lda, Aveiro (Portugal);

[3] Grupo de Desarrollo Empresarial SL, Terrassa, Barcelona (Spain);

[4] CIDETEC Surface Engineering, San Sebastián, Guipuzkoa (Spain); [5] TITANIA Ensayos y Proyectos Industriales SL, Cádiz (Spain)

In the frame of European collaborative works, herein an Eurostars project is presented with primary results aiming to validate, for aeronautical and railway sectors, REACH compliant improved coatings to protect aluminium parts, based on the incorporation of encapsulated corrosion inhibitors (carriers: LDH and SiNC // inhibitors: VOx and MBT) [1] into Cr-free anodising and anaphoretic coatings. This project improves corrosion resistance (providing long term corrosion protection and in some cases filiform corrosion resistance [2], among others as microbial-promoted corrosion), going beyond what other Cr-free alternatives have achieved, this time using the advantages of metal-oxides based substances [3], delivering improved surface treatments (chemicals and methods) to protect Al alloys (namely, AA2024 and AA7075 for aeronautics and A5754 H111 and AA6063 T5 for railway) at an affordable price.

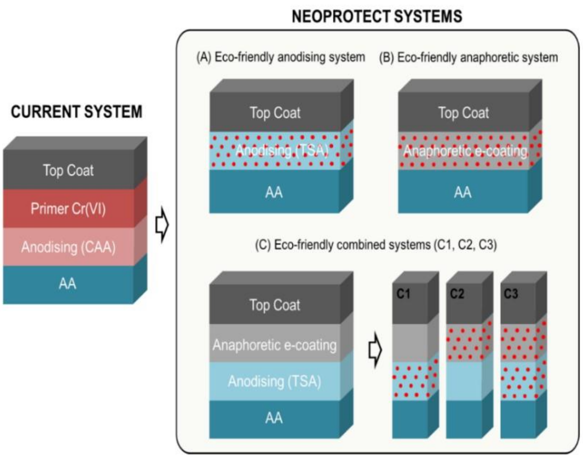

Figure 1. Strategy of C1, C2, C3 coatings with inhibitors against corrosion.

Keywords: Corrosion; Inhibitors; Aeronautics; REACH compliant; Anti-corrosion coatings

Acknowledgements:

This project (NEOPROTECT ID94) has received funding from Eureka Eurostars-3 Call-1 with support from national agencies (CDTI and ANI).

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O15

QUICK ENRICHMENT AND DETECTION OF ELECTROCHEMICALLY ACTIVE MICROORGANISMS

Satoshi Wakai[1]**, Chia-Lun Ho**[2]**, Akihiro Okamoto**[2]

[1] *Japan Agency for Marine-Earth Science and Technology (JAMSTEC), Yokosuka/Japan;*

[2] *National Institute for Materials Science (NIMS), Tsukuba/Japan*

Microbiologically-influenced corrosion (MIC) of metallic materials has been known for a long time. Recently, electrical-MIC (EMIC) caused by microorganisms with extracellular electron transfer ability has been attracting attention in the field of MIC [1]. Relative abundance of such electrochemically active microorganisms in a natural environment is relatively low, but as corrosion progresses, those of these microorganisms increased [2]. Although it is possible to infer that microorganisms were involved in EMIC by detecting the accumulation of electrochemically active microorganisms after the fact, it is currently difficult to detect the presence of microorganisms with extremely low relative abundance ratios with high sensitivity and to assess risk. Therefore, this study aimed to develop a culture-independent method to specifically accumulate microorganisms with extracellular electron transfer ability.

Many microorganisms with extracellular electron transfer ability have outer-membrane multiheme cytochromes (OMCs). We focused on the reactivity of heme in the OMC and 3,3'diaminobenzidine (DAB) [3]. DAB reacts with heme to form DAB polymer, which covalently binds with osmium oxide. Therefore, we performed targeted DAB polymerization using *Shewanella oneidensis* MR-1 with OMCs and *Escherichia coli* K12 without OMCs and demonstrated that only the cells of *S. oneidensis* MR-1 can polymerize DAB. Next, surface of magnetic nanobeads was treated with folic acid and osmium tetroxide to create Os-modified magnetic nanobeads (M@FA-Os NPs). The M@FA-Os NPs were mixed with microbial cells treated with DAB and observed the adhesion of M@FA-Os NPs to the cell surface of *S. oneidensis* MR-1 (Figure 1). The cells attacged with the M@FA-Os NPs can be collected using magnet.

Next, we demonstrated that the enrichment of microorganisms with OMCs from an environmental sample using DAB polymerization and M@FA-Os NPs. Microbiome analysis by amplicon sequencing for 16S rRNA gene fragment showed enrichment of two bacteria (order *Enterobacterales*) from the original sample.

Since it is unknown whether these bacteria possess OMCs, we performed metagenomic analysis of the enriched sample. As a result, an OmcA/MtrC family decaheme *c*-type cytochrome was detected.

Conclusively, we successfully developed the enrichment method for an electrochemically active microorganism using DAB polymerization and M@FA-Os NPs. This method would be useful for rapid and high-sensitive risk assessment of EMIC.

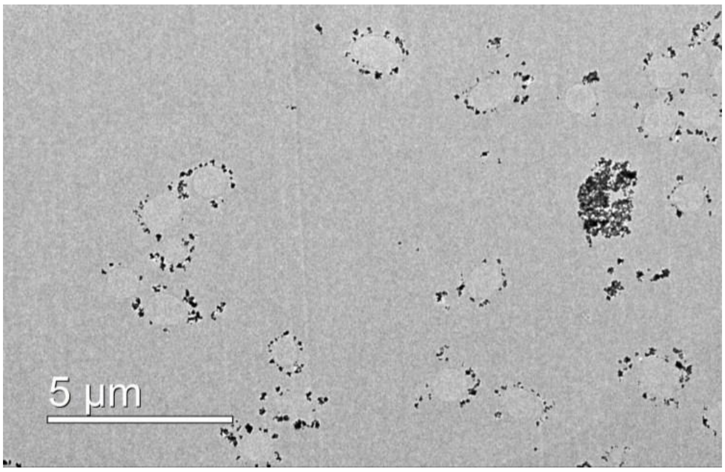

Figure 1. TEM image of the S. Oneidensis MR-1 cells attached with M@FA-Os NPs.

Keywords: risk assessment; extracellular electron transfer; electrochemically active microorganisms; outer-membrane multi-heme cytochrome.

Acknowledgements: This work was supported by a Grant-in-Aid for Research from the Japan Society for Promotion of Science KAKENHI [20H05590]; PRIME, the Japan Agency for Medical Research and Development [19gm6010002h0004]; JST, PRESTO [JPMJPR19H1]; JAEA Nuclear Energy S&T and Human Resource Development Project through concentrating wisdom [JPJA20P20333393]; and National Science and Technology Council, Taiwan NSTC 109-2113-M037-017-MY3 and NSTC 111-2320-B-037-035].

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PITCH

FROM PATENTS TO PROGRAMMES - SUPPORTING IPR HOLDERS AND EXPLORING GRANTS FOR ALL MIC-STAND MEMBERS

Theo Zacharis[1]

[1] *Greek Scientists Society*

Microbiologically Influenced Corrosion (MIC), a phenomenon causing significant material degradation due to the presence of microorganisms, is becoming an increasing concern across society. In Europe, various research groups and industrial stakeholders are tackling MIC, but their efforts remain fragmented with limited information exchange. This lack of a transdisciplinary approach hampers progress, despite the logical need for collaboration across material and biological sciences.

In contrast, countries like the USA, Canada, and Australia have established robust networks, developing methods, prevention measures, and standards that Europe often has to adopt. However, these solutions sometimes clash with European laws, such as restrictions on biocides. This dependency underscores the need for a strong European MIC network to foster collaboration and innovation tailored to European standards and regulations.

Collaboration with industry, plant operators, and critical infrastructure owners will be key. By facilitating interaction, communication, and knowledge sharing, and providing training for personnel and researchers from different disciplines, we can position Europe as a leader in MIC mitigation. This initiative will align with European values, such as environmental protection, and offer enhanced protection for people, property, and the environment.

In my presentation, I will explore how we can support Intellectual Property Rights (IPR) holders and explore grant opportunities for all MIC-STAND members. I will provide insights on securing funding and leveraging patents, empowering our community to turn research and innovations into impactful, real-world programmes. Our collective efforts will not only advance MIC mitigation but also ensure that Europe can independently develop and implement sustainable, environmentally friendly strategies.

Keywords: MIC Mitigation, Intellectual Property Rights (IPR), Grant Opportunities.

POSTER ABSTRACTS

POSTERS ABSTRACTS

P1

CASE STUDY ON PROGRESSIVE CORROSION OF CARBON STEEL COOLER TREATED WITH EPOXY-CERAMIC COATING

Veronika Hlavackova[1], **Petr Koutnik**[2], **Alena Sevcu**[1]

[1] *Technical University of Liberec, Institute for Nanomaterials, Advanced Technologies and Innovation, Dept. Applied Biology, Liberec, Czech Republic*

[2] *ORLEN UniCRE a.s., Ústí nad Labem, Czech Republic*

Formerly, corrosion attack and/or products were found when cleaning the cooler. This motivated a case study to determine the mechanism of degradation and specify the reasons for progressive corrosion. The cooling system was fabricated in 1968 and made from carbon steel EN 1.0405 (dimensions: l = 6m, d = 0,4 m). Since the early observation of corrosion, the cooler had been repeatedly treated with commercially available repair composite to prevent further corrosion. Samples were taken from the cooler heads. Cooling water, tubercles and swabs were sampled aseptically and cooled down immediately. The microstructural, chemical and microbiological analyses were performed. Visual appearance of the surface of cooler tube after removing the cooler head revealed presence of orange to dark brown tubercles of corrosion products. After removal of tubercles and cleaning, the inspection of the surface has shown uneven corrosion in the form of pits with dimensions of up to tens of mm penetrating to a depth of mm units (Figure 1). XRD analysis has proved that the only identified mineral phase was goethite (α-FeO(OH)). Chemical analysis of the cooling water proved to be full of potential electron donors (organic matter, ammonium) and acceptors (sulfates, nitrates) for the microbial metabolism. The chemical composition of cooling water rich in organic compounds, sulfates, nitrates etc. hosted preferentially microorganisms typical for wastewaters, fermenters and nitrogen metabolizing microorganisms, but sulphur oxidizing bacteria, and iron reducing and oxidizing bacteria were also detected. In the carbon steel cooler we observed a more significant increase in ammonia and sulfur oxidizing representatives. Bacteria reducing or oxidizing nitrogen compounds were detected in the cooling water using qPCR. The coexistance of microorganisms oxidizing ammonia dissolved in water and members of family *Methylophilaceae*, here enriched in biofilms, ressembles the environment of wastewaters (1–3). Carbon steel corrosion has previously been associated with the occurrence of other methylotrophic bacteria - *Methyloversatilis* - under anaerobic conditions (4). The exact relationship of oxygen and other electron acceptors in relation to their putative involvement in induction of steel corrosion is not fully understood.

The role of anaerobic microorganisms present in the biofilm, which are represented here by the poorly described and still uncultivated genera, cannot be so far resolved. Advanced experimental program to better describe the role of indicated genera in corrosion induction must be performed in future.

Figure 1. The surface of the carbon steel cooler, after the removal of corrosion products, indicates a progressive degree of corrosion.

Keywords: cooler; carbon steel; ammonium; methylotrophic bacteria; wastewater

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P2

PREVENTING BIOFOULING WITH CYCLAM-DERIVED COATINGS

Luis G. Alves[1], Stephanie Almada[1,2], Inês Nunes[1,2], Sara I. Faria[3,4], Fábio M. Carvalho[3,4], Luciana Gomes[3,4], Filipe J. Mergulhão[3,4], Elisabete R. Silva[2]

[1] *Centro de Química Estrutural - Institute of Molecular Sciences, Instituto Superior Técnico, University of Lisboa, Lisboa, Portugal*
[2] *BioISI - Biosystems & Integrative Sciences Institute, Faculdade de Ciências da University of Lisboa, Lisboa, Portugal*
[3] *LEPABE - Laboratory for Process Engineering, Environment, Biotechnology and Energy, Faculty of Engineering, University of Porto, Porto, Portugal*
[4] *AliCE - Associate Laboratory in Chemical Engineering, Faculty of Engineering, University of Porto, Porto, Portugal*

Surface colonization by aquatic foulant organisms (biofouling) poses significant environmental and economic risks to industrial systems. For instance, in water supply and treatment circuits, it can lead to serious human infections, thus becoming a major public health concern[1]. On the other hand, the main underlying problem common to all these systems is their susceptibility to hosting biofilms, recognized as promoters of Microbially Influenced Corrosion (MIC), which contributes to the premature degradation of materials and adversely affects the sustainability of industrial systems[2].

The most effective bio-decontamination strategies comprise the controlled release of toxic and cumulative bioactive agents into the aquatic environment, implying a limited life cycle, considerable ecotoxicity, and the promotion of pathogen resistance. In this study, novel cyclam derivatives with proven antimicrobial activity[3] and relatively low toxicity were used as bioactive additives in silicone and polyurethane-based coating formulations[4]. The optimized coating formulations were applied to coat acrylic coupons and ceramic-based filters to assess their antimicrobial and anti-biofilm activity against various bacteria (both Gram-positive and Gram-negative). The results obtained show that the iron-based cyclam derivative $[\{H_2(^{4\text{-}CF_3}PhCH_2)_2Cyclam\}FeCl_2]Cl$ incorporated into silicone and polyurethane marine paints is highly active against *Pseudoalteromonas tunicata* and *Cobetia marina* biofilms under controlled hydrodynamic conditions[1,5] in comparison to its cyclam-free coating counterpart (see Figure 1).

These findings will be presented and discussed in the present communication.

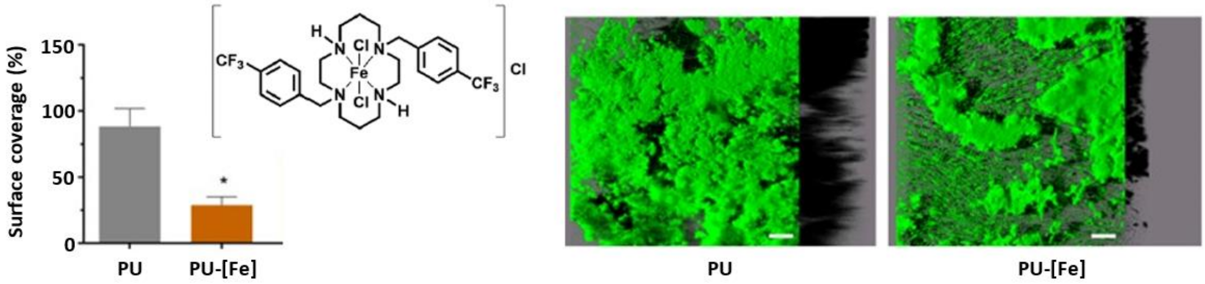

Figure 1. Representative biofilm structures of *Cobetia marina* on a reference polyurethane (PU) and polyurethane with [{H_2($^{4\text{-}CF3}$PhCH_2)$_2$Cyclam}FeCl_2]Cl (PU-[Fe]) surfaces after 42 days of biofilm formation. These images were obtained from confocal *z*-stacks using IMARIS software and present an aerial, 3D view of the biofilms (shadow projection on the right). The scale bar is 40 µm.

Keywords: bacteria; biofilm; bioactive cyclams; antifouling coatings.

Acknowledgments: This work was supported by national funds through FCT/MCTES (PIDDAC): UIDB/00511/2020 (DOI: 10.54499/UIDB/00511/2020), UIDP/00511/2020 (DOI: 10.54499/UIDP/00511/2020), LA/P/0045/2020 (DOI: 10.54499/LA/P/0045/2020), UIDB/04046/2020 (DOI: 10.54499/UIDB/04046/2020), UIDP/04046/2020 (DOI: 10.54499/UIDP/04046/2020), UIDB/00100/2020 (DOI: 10.54499/UIDB/00100/2020), UIDP/00100/2020 (DOI: 10.54499/UIDP/00100/2020) and LA/P/0056/2020 (DOI: 10.54499/LA/P/0056/2020). F.M.C. and E.R.S. thank Fundação para a Ciência e a Tecnologia, Portugal, for the Ph.D. scholarship (ref. 2022.12705.BD) and work contract through the Scientific Employment Stimulus-Individual Call (DOI: 10.54499/CEECIND/03530/2018/CP1553/CT0011, respectively.

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P3

MARINE BIOMINERALIZATION FOR ENHANCED CORROSION RESISTANCE: INSIGHTS FROM THE ANR MICOATEC PROJECT

M. J. F. Marques[1,2], C. Nkoua[3], J. Jaume[1], A. Diderot[1], D. Mercier[4], A. Seyeux[4], M-L. Delia[1], S. Da Silva[5], B. Fori[6], C. Blanc[3], S. Zanna[4], P. Marcus[4], R. Basseguy[1]

[1]*LGC, Université de Toulouse, CNRS, INP, UPS, Toulouse, France*

[2]*LNEG, Laboratório de Materiais e Revestimentos, Lisboa, Portugal*

[3]*CIRIMAT, Université de Toulouse, CNRS, INP, ENSIACET, Toulouse, France*

[4]*IRCP-PCS, Chimie ParisTech-CNRS, PSL Research University, Paris, France*

[5]*6T-MIC INGENIERIES, Castanet-Tolosan, France*

[6]*MECAPROTEC Industries, Muret, France*

Concerns about marine pollution and ecological threats caused by traditional corrosion protection technologies have driven the development of new environmentally friendly anti-corrosion solutions. In recent years, it became clear that microorganisms have the potential to positively impact corrosion behavior, a phenomenon known as MICI (microbiologically influenced corrosion inhibition) [1,2].

Although research on MICI mechanisms is still in the beginning, two main mechanisms have been outlined: direct and indirect inhibition. In the first one, the microorganisms are responsible for the segregation of slow-release inhibitors or surfactants or consume oxygen, which affects the cathodic reaction process. The second one, indirect inhibition mechanism, is associated to the formation of a protective layer on the surface of the material due to metabolic activity of microorganisms. In this context, biomineralization attracted the attention of researchers as a solution to inhibit metal corrosion.

Being aware of the potential of this microbial induced mineralization phenomenon, the French ANR MICOATEC project (www.micoatec.eu) established a new approach for the development of a bioinspired anticorrosion solution for metal protection based on the biomineralization process observed on an Al-Mg surface during exposure in marine field. The main goal is to translate the natural biotic process into an abiotic technological process for corrosion protection, without replicating the biofilm itself or incorporating active biocompounds into a coating matrix (figure 1). Three specific objectives were outlined:

(1) Understanding the interactions of aluminium alloys with marine biological activity which leads to the formation of a layer that can inhibit corrosion (protective layer).

(2) Mastering the growth process and the physico-chemical properties of the protective layer on the alloy surface under the influence of biological activity, and

(3) In the longer term, providing the industry with a bioinspired technology for anticorrosion coatings that is more environmentally friendly in order to increase the lifespan of metal structures.

The work of the multidisciplinary consortium has led to numerous results in research activities associated with the growth process (WP1) and anti-corrosion properties (WP2) of the protective layer, providing the bases for replication steps (WP3). Among the most important results, we can cite:

i The Al-Mg surface modifications showed to be strongly influenced by the type of fouling present, notably photosynthetic, such as algae.

ii The anticorrosion properties of the formed layers are intimately linked to their structure and chemical composition.

iii Replication in a biotic environment (seawater + salt marshes) is currently being mastered.

MICOATEC has allowed to confirm marine biomineralization as a potential new approach for the development of anti-corrosion solutions inspired from biological interactions with metals.

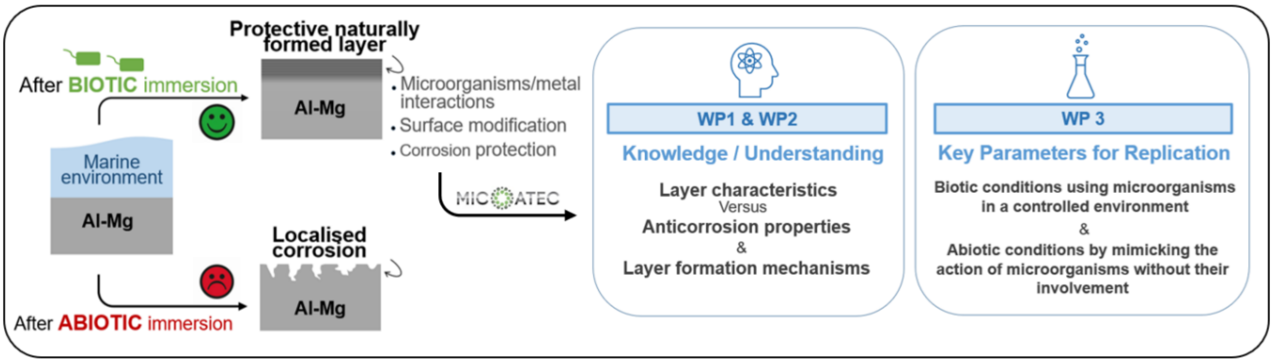

Figure 1 - MICOATEC project: New concept of Microbially Inspired anticorrosion coating technology. Work Packages (WP).

Keywords: Aluminium alloy; Marine Corrosion inhibition; Biomineralization; Bioinspired protection solution.

Acknowledgements: This work was financially supported by the ANR, in the framework of the MICOATEC project (ANR-19-CE08-0018) coordinated by R.Basséguy-LGC.

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P4

ANTIFOULING NANO-DESIGNED COMPOSITE AS A MITIGATION STRATEGY

Elisabete R. Silva[1,2*], Noelia Losada-Garcia[1], Ana P. Carapeto[1,3], M. Pereira[1,2], Nanni Noël-Hermes[4], Maria Salta[4]

[1]BioISI - Biosystems & Integrative Sciences Institute, Faculdade de Ciências, Lisboa, Portugal

[2]Departamento de Química e Bioquímica, Faculdade de Ciências da Universidade de Lisboa, Portugal

[3]Departamento de Física, Faculdade de Ciências, Universidade de Lisboa, Portugal

[4]Endures BV Den Helder, Netherlands

The pillars of a greener and healthier society's growth are the sustainable use of resources and materials, which form the foundation of all social infrastructures. However, the rising industrial activity in many sectors, including water and wastewater management systems, marine operations, renewable energy platforms, and climate change, raises one of the biggest global threats: surface corrosion and biofouling. The main underlying problem common to all these systems is the attachment and growth of microorganisms on surfaces in contact with aqueous streams, forming biofilms, promoters of Microbially Influenced Corrosion (MIC), which is estimated to affect up to 20% of corrosion in aqueous systems, costing billions of dollars only for rehabilitation costs [1]. Hitherto, the most effective anti-biofilm strategies have relied on toxic and persistent chemical agents (e.g., biocides) that act through release mechanisms. However, these agents inevitably end up in the environment, posing ecological issues and urging the search for long-lasting and environmentally friendly mitigation alternatives. This work aims to contribute to overcoming these mitigation challenges by providing a novel approach that involves prior know-how on bioactive agents grafting [2, 3] for the immobilization of biocidal agents on the surface of metal oxide nanoparticles (e.g., Ti and Si-based), allowing their bioactivity to be amplified or generated and improve their capacity against bacterial biofilms and MIC, without leaching agents into aquatic media, extending their potential application. XRD and DRIFT spectra (Fig. 1c) confirmed the biocidal agent grafting on the NPs′ surface, and the maintenance of their characteristic crystalline structure. The assessment of the morphological properties of the bioactive nanocomposites using scanning electron microscopy (SEM-EDS) revealed nanoparticles with a spherical geometry with higher agglomeration (Fig. 1b). The antimicrobial susceptibility of the nanocomposites against major waterborne microorganisms showed promising inhibition effects, with notable changes in bacterial cell morphology observed in AFM topographic images.

Fig. 1a shows a representative antimicrobial action against the Methicillin-resistant *Staphylococcus aureus* (MRSA) obtained with a TiO_2-based developed nanocomposite. Furthermore, to evaluate the potential applicability of this strategy, MIC-related enrichment cultures from industrial environments are being utilized in laboratory experiments to assess the efficacy of coatings incorporating developed bioactive nanocomposites against MIC.

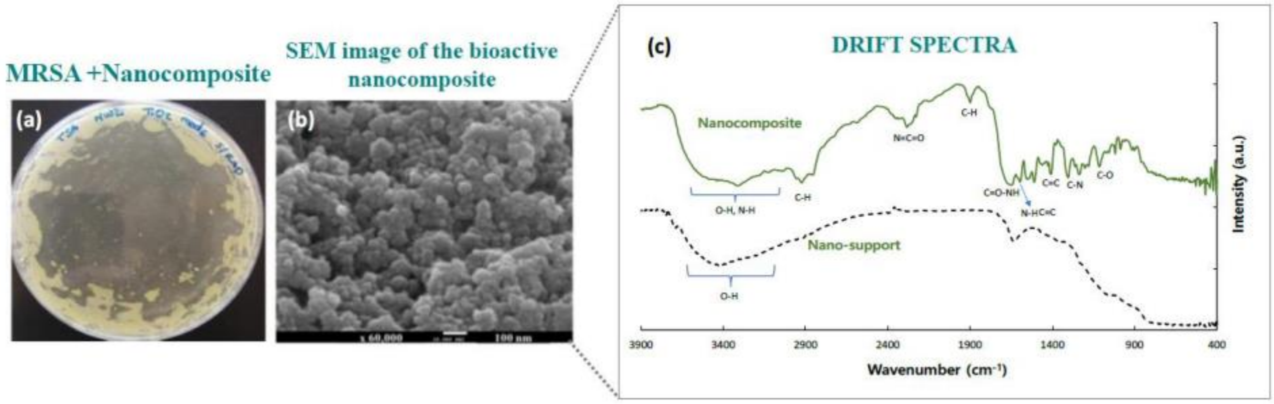

Figure 1. Antimicrobial action against the Methicillin-resistant *Staphylococcus aureus* (MRSA) of a bioactive TiO2-based nanocomposite (a), together with its morphological details through an SEM image (b); and Fourier transform infrared diffuse reflectance (DRIFT) spectrum in comparison with the pristine nano-support.

Keywords: Metal oxide nano-supports, grafting of bioactive agents, nanocomposite, antimicrobial.

Acknowledgments: The authors acknowledge the FCT (Foundation for Science and Technology) support within the scope of the BioISI unit grant (https://doi.org/10.54499/UIDB/04046/2020) and under the project NanoBioMitig (https://doi.org/10.54499/2022.06149.PTDC). E. R. Silva thanks FCT for her work contract (https://doi.org/10.54499/CEECIND/03530/2018/CP1553/CT0011). Support from the EURO-MIC COST Action (CA20130) is also acknowledged.

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P5

ANTIMIOCBIAL ACTIVITY MECHANISM AND ECO-CYTO-TOXICITY OF CeO_2/CuO

Svetlana Vihodceva[1*] Andris Šutka[1], Mairis Iesalnieks[1], Liga Orlova[1], Arturs Pludonis[1], Maarja Otsus[2], Mariliis Sihtmäe[2], Heiki Vija[2], Alexandra Nefedova[3,4], Angela Ivask[3], Anne Kahru[2], Kaja Kasemets[2]

[1] *The Institute of Materials and Surface Engineering, Faculty of Natural Sciences and Technologies, Riga Technical University, Riga, Latvia*

[2] *National Institute of Chemical Physics and Biophysics, Laboratory of Environmental Toxicology, Tallinn, Estonia*

[3] *Institute of Molecular and Cell Biology, University of Tartu, Tartu, Estonia*

[4] *Institute of Physics, University of Tartu, Tartu, Estonia*

So far, limited research exists on the antimicrobial activity and mechanisms of CeO_2/CuO, and assessments of antimicrobial efficacy have relied on the agar diffusion test that indicates the dissolution of compounds from the M/NPs, including ions and residual materials from the synthesis process and does not provide information about the actual antimicrobial activity and its mechanisms. [Fathima Khyrun et al. 2023]

This research presents a new synthesis method for the CeO_2/CuO nanoparticles composite. It explores its antimicrobial properties, mechanisms of action, and potential ecotoxicity via bioluminescence test with environmental bacterium Vibrio fisheri and cytotoxicity to human keratinocytes in vitro. The antimicrobial properties of the CeO_2/CuO composite were assessed against various microorganisms, including Gram-negative bacteria *Escherichia coli*, *Pseudomonas aeruginosa*, Gram-positive bacteria *Staphylococcus aureus* as well as a fungus *Candida albicans*. Bacteria and fungi (10^7 CFU/ml) were exposed to CeO_2/CuO powder suspensions in deionized water for 2, 4, and 24 h (concentrations from 1 to 1000 mg/L). The CeO_2/CuO at concentrations from 100 mg/L upwards showed biocidal activity towards *E. coli* and *S. aureus* after 2 h of contact and towards *P. aeruginosa* after 4 h. In the case of *C. albicans*, no inhibition by CeO_2/CuO was evidenced at 2 and 4 h exposure times and only after 24 h 100 mg/L and higher concentrations of CeO_2/CuO inhibited the fungus. Notably, the CeO_2/CuO composite exhibited cytotoxic effects in vitro only at 1000 mg/L concentration after 24 h with cell viability of 33%. Also, according to the bioluminescence test with the environmental bacterium *Vibrio fisheri* shows low ecotoxicity. Study findings highlight the multimodal antimicrobial mechanisms of composite, and low eco- and cytotoxicity, suggesting its potential as a safe and effective antimicrobial agent.

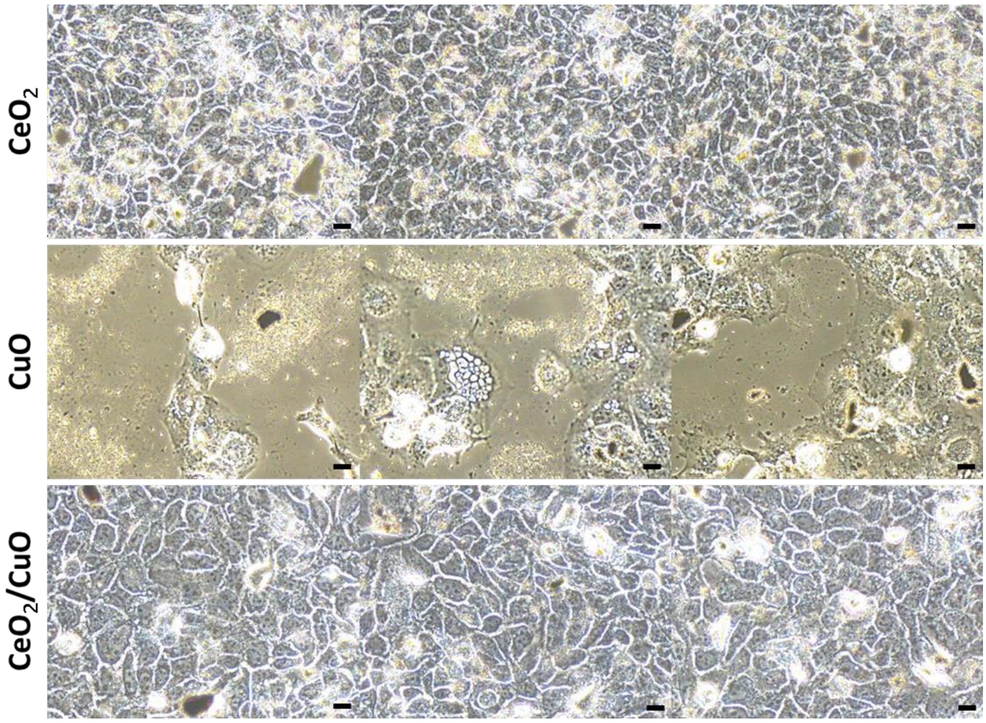

Figure 1. Light microscopy 20x magnification of the HaCat keratinocytes cells after 24 h incubation with CeO2 (upper row), CuO (middle row), and CeO2/CuO (lower row) at 500 mg/L concentration.

Keywords: antimicrobial; nanomaterials; ROS production; composite.

Acknowledgements:

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P6

FABRICATION OF NEW ANTIFOULING COATING BASED ON COMMERCIAL EPOXY RESIN AND HYDROPHOBIC IONIC LIQUID

Sergiy Rogalsky[1], Olena Moshynets[2], Oleg Dzhuzha[1], Oksana Tarasyuk[1], Geert Potters[3,4]

[1] *V.P. Kukhar Institute of Bioorganic Chemistry and Petrochemistry of the NAS of Ukraine, Kyiv, Ukraine*

[2] *Institute of Molecular Biology and Genetics of the NAS of Ukraine, Kyiv, Ukraine*

[3] *Antwerp Maritime Academy, Noordkasteel Oost 6 2030 Antwerp, Belgium*

[4] *University of Antwerp, Groenenborgerlaan 171, 2020 Antwerp, Belgium*

Epoxy resins have the largest share of materials used in marine coatings today, since they have excellent corrosion resistance and cost-effectiveness, as well as long overcoat time and good recoat ability. More than 20 t of epoxy resin is used in the construction of each ship as a primer, tiecoat and topcoat for protection underwater hulls and ballast tanks against corrosion, biofouling and abrasion [1]. The brittle nature and hydrophilicity of cured epoxies are serious drawbacks limiting their practical applications. The incorporation of a liquid phase as modifier into epoxy resin based coatings can be an efficient approach for the enhancement of fracture and impact characteristics [2]. However, modifying additives must be chemically inert toward the components of the coatings in order not to interfere with curing. From this point of view, the hydrophobic long-chain ionic liquids (ILs) seem promising candidates, since they are known to have plasticizing effect on engineering polymers, as well as broad range of biological activity [3].

In this study, new hydrophobic IL – 1-dodecyl-3-methylimidazolium dodecylbenzenesulfonate ($C_{12}C_1$IM-DBS) has been synthesized and studied as potential multifunctional modifying additive for commercial epoxy resin.

Two-component epoxy resin DER 331 (Dow Chemical) based on diglycidyl ether of bisphenol A was used for preparation of modified coatings. Ionic liquid was dissolved in the liquid resin (10 and 20 wt.%) followed by addition of curing agent D.E.H.™ (diethylenetriamine). The liquid epoxy was used to coat steel bars (8 x 2.5 x 0.05 cm) using a paint brush. The coatings were cured for 72 hours at 25 °C and 4 hours at 60 °C. Control coatings were prepared by using neat DER 331 resin.

Differential scanning calorimetry (DSC) analyses were performed from -90 to 200 °C on a TA Q2000 calorimeter with a heating rate of 20 °C/min.

Static contact angle measurements were taken using a Drop Shape Analyzer DSA25E (Krüss) by the sessile drop method. Antibiofilm efficiency of $C_{12}C_1$IM-DBS was evaluated by assessing the capability of two biofilm-forming model strains, *Staphylococcus aureus* ATCC 25923 and *Pseudomonas aeruginosa* PA01, to form attached biofilms on the surface coated with DER 331 and DER 331/$C_{12}C_1$IM-DBS following 48 hours of stationary incubation at 37 °C. To measure the level of total biofilm metabolic activity, MTT assay was performed [4].

The results of static contact angle (θ_ω) measurements indicate that the surface of neat DER 331 epoxy resin is completely wetted by the water droplet that makes it difficult to measure the θ_ω value. However, the modified DER 331/$C_{12}C_1$IM-DBS coatings are much less hydrophilic with θ_ω value of 23.5°. According to DSC analysis data, the neat DER 331 has glass transition temperature (T_g) of 122.8 °C. The introduction of $C_{12}C_1$IM-DBS (20%) to epoxy resin significantly reduced T_g value to 91.5 °C that reveal plasticizing effect of the ionic liquid on the polymer matrix.

The total biofilm metabolic activity of *P. aeruginosa* and *S. aureus* formed on the surface of modified DER 331/$C_{12}C_1$IM-DBS coatings was found to be significantly reduced (by 50% and 92%, respectively) compared to control epoxy resin. Overall, the obtained results indicate that modification of epoxy resin by hydrophobic ionic liquid can be efficient approach to prepare new promising protective coatings with improved elasticity, as well as potential resistance to biofouling.

Keywords: epoxy resin; ionic liquid; protective coating; plasticizing effect; antibiofilm activity.

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P7

AFM IMAGING OF BACTERIAL MEMBRANE ALTERATIONS INDUCED BY BIOCIDAL CONTACT

Elisabete R. Silva[1,2], T. Robalo[2,3], A.P. Carapeto[2,3]

[1]*Departamento de Química e Bioquímica, Faculty of Sciences, University of Lisboa, Campo Grande, 1749-016 Lisboa, Portugal.*

[2] *BioISI- Biosystems & Integrative Sciences Institute, Faculdade de Ciências, Lisboa, Portugal*

[3]*Departamento de Física, Faculdade de Ciências, Faculty of Sciences, University of Lisboa, Campo Grande, 1749-016 Lisboa, Portugal.*

Atomic Force Microscopy (AFM) has become an indispensable tool in studying bacterial membranes, offering unparalleled insights at the nanoscale. Unlike traditional microscopy techniques, AFM provides high-resolution, three-dimensional images of bacterial cells and biofilms under near-physiological conditions. This capability is particularly valuable for observing the initial stages of bacterial adhesion and biofilm development on various surfaces, which leads to biofilm formation - the undesirable colonization of microorganisms on surfaces. Biofilm formation poses significant challenges in many industrial and environmental contexts, including water treatment systems, oil and gas infrastructures, marine vessels, and medical devices. One of the most critical consequences of biofilm formation is microbiologically influenced corrosion (MIC), which can severely damage these infrastructures. Understanding the mechanisms of biofilm formation, adhesion, and resilience is crucial for developing effective strategies to mitigate biofilm formation and its associated issues, such as MIC.

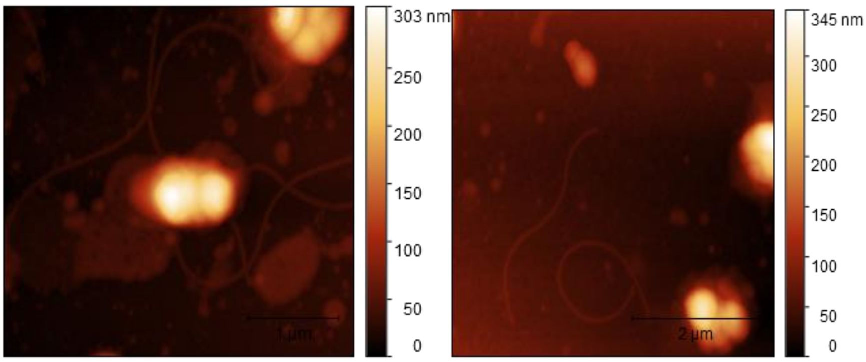

Figure 1. Topographic AFM images of a) untreated *V. cholerae* and b) *V. cholerae* treated with biocide.

In this work, AFM was employed to evaluate the effects of antimicrobial nanomaterials developed within the NanoBiomitig Project (https://nanobiomitig.pt/) on the morphological and topographical properties of bacteria, such as *S. aureus* (MRSA) and V. cholerae. The diameters and membrane roughness of the cells were measured before and after contact with the biocide agent.

For *V. cholerae* (Figure 1), exposure to the biocide resulted in noticeable changes in bacterial size and a significant increase in surface roughness, along with the loss of the flagellum, which is crucial for motility, adhesion, and biofilm formation. For MRSA, the cells became wider and rougher, with measurable increases in biophysical properties, including cell diameter and surface roughness (RMS), compared to the untreated bacteria.

In conclusion, Atomic Force Microscopy is a highly effective tool in the fight against bacterial biofouling due to its ability to provide precise nanoscale information about bacterial modifications upon contact with biocide agents. This capability allows for a detailed assessment of biofilm properties and the development of more effective antimicrobial strategies. Future investigations will focus on biofilm adhesion properties and their modulation by these nanomaterials.

Keywords: AFM, biocidal nanomaterial, biofilm, MIC prevention.

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P8

INVESTIGATING THE EFFICACY OF THPS AND GLUTARALDEHYDE TREATMENT FOR ENHANCED PERFORMANCE ON NORTH SEA OFFSHORE SYSTEM

Pernille Ruby Lauritsen[1], Tanmay Chaturvedi[2]

[1,2] Aalborg University, Esbjerg, Denmark

The use of biocides plays an important role in maintaining the integrity of critical oilfield systems. At the North Sea offshore system, several locations have a history of elevated bacterial levels and significant corrosion, as observed by the intelligent pigging inspections. Common techniques for biocide qualification focus on the assessment of how effectively biocides minimize microbial activities, specifically sulphate-reducing bacteria (SRB). However, such testing cannot adequately answer the question of whether a particular biocide treatment regime can be expected to protect seawater injection systems against microbiologically influenced corrosion (MIC).

The optimization of biocide treatments is a challenge, as the efficacy of biocides can be influenced by various factors, including concentration, duration, and frequency of application. Therefore, this study conducts a thorough investigation into the efficacy of the treatment conditions for two frequently employed biocides, Tetrakis Hydroxymethyl Phosphonium Sulfate (THPS) and Glutaraldehyde. The primary objective is to enhance the understanding of biocide treatments, tailoring concentration and duration to match each biocide unique mechanism.

The study employs the use of anaerobic reactors for biofilm growth as a part of the setup to mimic offshore environment. Mixed microbial culture from Wadden Sea sediment is used as inoculum in Postgate medium to populate the media with sulphate reducing bacteria and archaea. The assessments of the biocide effectiveness are based on multiple lines of evidence (MLOE), to evaluate the efficacy of biocide treatments. Both indirect and direct assessments, including 16S/18S ITS rRNA sequencing, quantitative polymerase chain reaction (qPCR), adenosine triphosphate (ATP) activity, hydrogen sulphide (H_2S) concentrations levels, and surface corrosion and pitting rate analysis, are utilized to comprehensively assess the impact of biocide treatments on microbial communities and corrosion rates.

Through the integration of these diverse analytical techniques, a holistic understanding of the efficacy of the biocide treatments on microbial communities and corrosion rates are achieved. The MLOE approach provides valuable insights into the complex interactions between biocides, microbial communities, and corrosion rate in offshore environments. By integrating the data, this study aims to elucidate the relationship between microbial activities and corrosion rates in order to ultimately inform a more targeted and effective biocide treatment strategies for offshore oilfield systems.

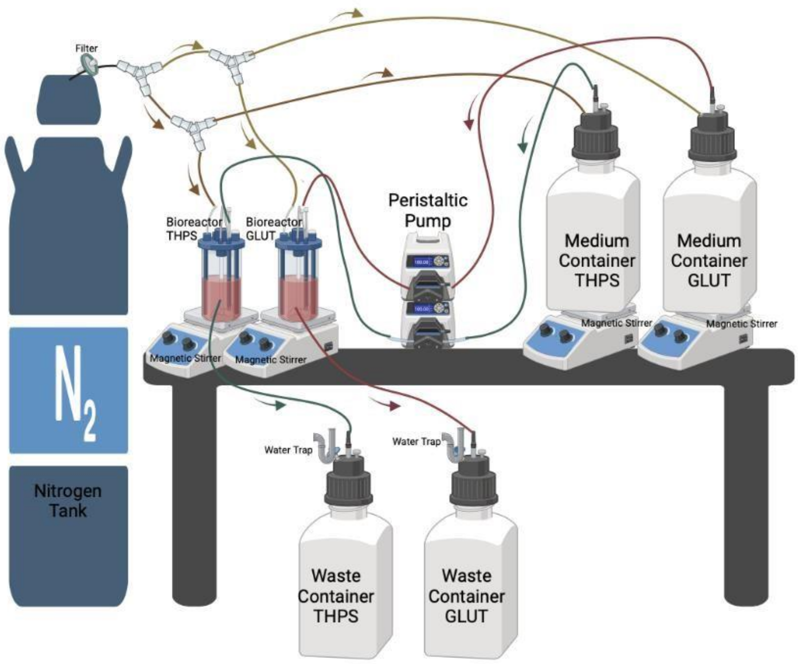

Figure 1. Setup of the anaerobic/sealed CDC biofilm reactors designed to mimic offshore environment, facilitating biofilm growth.

Keywords: Biocide; Tetrakis Hydroxymethyl Phosphonium Sulfate (THPS); Glutaraldehyde; Microbiologically Influenced Corrosion (MIC); Sulphate-Reducing Bacteria (SRB)

P9

QUICK ENRICHMENT AND DETECTION OF ELECTROCHEMICALLY ACTIVE MICROORGANISMS

Satoshi Wakai[1], **Chia-Lun Ho**[2], **Akihiro Okamoto**[2]

[1] *Japan Agency for Marine-Earth Science and Technology (JAMSTEC), Yokosuka/Japan;*

[2] *National Institute for Materials Science (NIMS), Tsukuba/Japan*

Microbiologically-influenced corrosion (MIC) of metallic materials has been known for a long time. Recently, electrical-MIC (EMIC) caused by microorganisms with extracellular electron transfer ability has been attracting attention in the field of MIC [1]. Relative abundance of such electrochemically active microorganisms in a natural environment is relatively low, but as corrosion progresses, those of these microorganisms increased [2]. Although it is possible to infer that microorganisms were involved in EMIC by detecting the accumulation of electrochemically active microorganisms after the fact, it is currently difficult to detect the presence of microorganisms with extremely low relative abundance ratios with high sensitivity and to assess risk. Therefore, this study aimed to develop a culture-independent method to specifically accumulate microorganisms with extracellular electron transfer ability.

Many microorganisms with extracellular electron transfer ability have outer-membrane multiheme cytochromes (OMCs). We focused on the reactivity of heme in the OMC and 3,3'diaminobenzidine (DAB) [3]. DAB reacts with heme to form DAB polymer, which covalently binds with osmium oxide. Therefore, we performed targeted DAB polymerization using *Shewanella oneidensis* MR-1 with OMCs and *Escherichia coli* K12 without OMCs and demonstrated that only the cells of *S. oneidensis* MR-1 can polymerize DAB. Next, surface of magnetic nanobeads was treated with folic acid and osmium tetroxide to create Os-modified magnetic nanobeads (M@FA-Os NPs). The M@FA-Os NPs were mixed with microbial cells treated with DAB and observed the adhesion of M@FA-Os NPs to the cell surface of *S. oneidensis* MR-1 (Figure 1). The cells attacged with the M@FA-Os NPs can be collected using magnet.

Next, we demonstrated that the enrichment of microorganisms with OMCs from an environmental sample using DAB polymerization and M@FA-Os NPs. Microbiome analysis by amplicon sequencing for 16S rRNA gene fragment showed enrichment of two bacteria (order *Enterobacterales*) from the original sample. Since it is unknown whether these bacteria possess OMCs, we performed metagenomic analysis of the enriched sample. As a result, an OmcA/MtrC family decaheme *c*-type cytochrome was detected.

Conclusively, we successfully developed the enrichment method for an electrochemically active microorganism using DAB polymerization and M@FA-Os NPs. This method would be useful for rapid and high-sensitive risk assessment of EMIC.

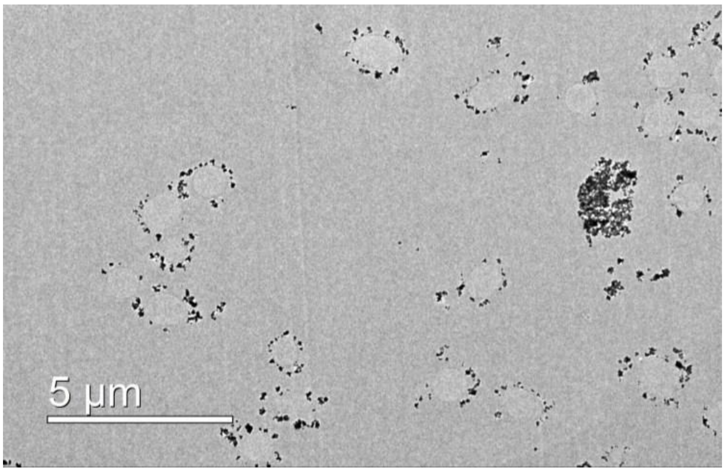

Figure 1. TEM image of the S. Oneidensis MR-1 cells attached with M@FA-Os NPs.

Keywords: risk assessment; extracellular electron transfer; electrochemically active microorganisms; outer-membrane multi-heme cytochrome.

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P10

INVESTIGATING MICROBIOLOGICALLY INFLUENCED CORROSION IN HOT WATER BOILERS: MECHANISMS AND MITIGATION STRATEGIES

Milena Rajić[1], Dragoljub Živković[1], Milan Banić[1], Miroslav Milovanović[2]

[1] *University of Niš, Faculty of Mechanical Engineering, Niš, Serbia;*

[2] *University of Niš, Faculty of Electronic Engineering, Niš, Serbia;*

Microbiologically induced corrosion (MIC) represents a process where microorganisms, such as bacteria, fungi, and algae, cause damage to materials, leading to significant deterioration and failure. This phenomenon is of particular concern in industrial settings, where it can affect the integrity and performance of hot water boilers, especially fire-tube boilers. Fire-tube boilers are the most commonly used units across various sectors due to their efficiency, durability, and ability to provide large volumes of hot water or steam. These boilers are commonly installed in industrial facilities, in manufacturing plants, processing plants, and other industrial settings for process heating, production operations, and providing central heating for large buildings or industrial complexes. The focus of this research is on a detailed numerical-experimental analysis of the MIC influenced corrosion on structural state of fire tube boilers.

Given their construction and operation, fire-tube boilers provide conditions that can favour the growth and proliferation of microorganisms, initiating the process of MIC. The water's low velocity within these boilers allows impurities, algae, and other microorganisms to settle on the water side of the boiler, particularly on the walls of the flue gas pipes. These pipes, being in close proximity to each other, create a micro-environment that not only supports but also enhances the activities of these microorganisms, leading to the acceleration of corrosion processes. This situation is exacerbated in environments where maintenance practices are inadequate, or where water treatment is not sufficiently aggressive to control microbial growth, especially in Serbia, where failures occur often.

The appearance of biofilm on the water side inside the boiler leads to overheating of the heating walls, contributing to an increase in the stress state of the boiler's structure. The increase in stress can lead to exceeding the allowable stresses in certain elements of the boiler, which are particularly sensitive. Numerous operational failures and accidents that have occurred are precisely the result of deposit accumulations and inadequate maintenance of the facility. This case needs special analysis, as one of the most prevalent in the real installed objects of the heating system. In some installed boiler plants in heating systems, there is no primary boiler feed water circuit, i.e., water from the heating network is directly introduced into the boiler. Some facilities do not have adequate preparation of feed water (due to significant water losses in the network.

Because of such experiences from operation, an analysis of the variable operating mode of the hot water boiler due to the occurrence of MIC biofilm accumulations was initiated. In the analysis, data from the boiler manufacturer were used. The cases were analysed using numerical analysis (FEM) to determine the impact of the deposits on the stress-strain state of the boiler structure. The results are then compared to measurements in the plant.

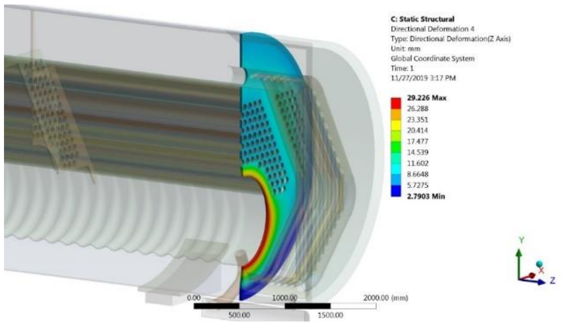

Figure 1. Directional deformation of the tube plate of the first turning chamber in the z-axis with 1mm MIC product deposits.

The analysis in this research analyses the specifics of how these conditions contribute to the onset and progression of MIC in fire-tube boilers. By understanding the interaction between the structural components of these boilers and the microorganisms, we can develop targeted strategies to mitigate the effects of MIC, thereby extending the operational life of these essential industrial units and ensuring their continued efficiency and safety in operation. The paper outlines the common reasons behind failures, which are often due to structural issues like vibration and leaks, performance challenges such as fouling and thermal stress, operational failures including improper handling during startup and shutdown, and structural damage from corrosion and erosion. The formation of biofilms on metal surfaces accelerates corrosion through electrochemical reactions. By analysing historical data, ML models can forecast potential issues, facilitating predictive maintenance. This approach can significantly improve efficiency, reduce costs, and improve safety in the process industry by allowing for maintenance to be scheduled in advance based on the anticipated needs.

Keywords: microbiologically induced corrosion; hot water boilers; finite element analysis; failure mechanisms; predictive maintenance.

P11

MODELLING ANAEROBIC MICROBIAL IRON CORROSION WITH DIRECT ELECTRON TRANSFER

Luis F. M. Rosa[1], Raúl Dávalos[1], Sarah Al-Aqeel[2], Cristian Picioreanu[1]

[1] *Biological and Environmental Science and Engineering (BESE), King Abdullah University of Science and Technology (KAUST), Thuwal / Saudi Arabia*

[2] *Saudi Aramco, Oil and Gas Network Integrity, Research & Development Center, King Abdullah University of Science and Technology (KAUST), Thuwal/ Saudi Arabia*

The study provides theoretical evidence on how the reversible electrical connection to the bacterial metabolism via cytochromes may enable the microbial use of electrons from Fe0 dissolution to supply the bacterial catabolism. The bidirectional exchange of electrons can interface with a natural corrosion cell.
The model combining electrochemical and biological reactions with the transport of ions explains the mechanism of anaerobic iron corrosion by coupling the bacterial metabolism of *Geobacter* sp. The model bacteria can both deliver electrons from the carbon-based metabolism to an external metal surface coupling with hydrogen evolution, the classic extracelular electron transfer metabolism[1], or take electrons from the iron dissolution and couple with inorganic carbon reduction, which can provide the bacteria with energy while interacting with the corroding metal.
Based on recently reported experimental data, we analyzed different scenarios function of local environmental conditions and we calculate corrosion potentials and currents explaining those determined experimentally. We also provide insight on practical conditions to inhibit direct electron transfer (DET) MIC.

Keywords: Microbial influenced corrosion; Geobacter; Extracellular electron transfer; Model.

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P12

HIGH-ENTROPY ALLOY-BASED MATERIALS FOR ANTIMICROBIAL AND CORROSION PREVENTION APPLICATIONS

Bogdan Postolnyi[1,2], Alexander Pogrebnjak[2,3], Volodymyr Buranych[3], Razet Basnukaeva[4]

[1] *Institute of Physics for Advanced Materials, Nanotechnology and Photonics, Faculty of Sciences of the University of Porto, Porto, Portugal;*

[2] *National R&D Institute for Non−Ferrous and Rare Metals, Pantelimon, Romania*

[3] *Institute of Materials, Faculty of Materials Science and Technology in Trnava, Slovak University of Technology in Bratislava, Trnava, Slovakia*

[4] *B. Verkin Institute for Low Temperature Physics and Engineering of the National Academy of Sciences of Ukraine, Kharkiv, Ukraine*

High-Entropy Alloys (HEAs) represent a revolutionary class of materials defined by the inclusion of five or more principal elements in near-equiatomic proportions. This unique composition disrupts traditional interatomic ordering, leading to exceptional properties, including outstanding mechanical strength, superior wear resistance, and remarkable thermal stability. These characteristics have positioned HEAs as strong candidates for a wide range of applications across various disciplines, including aerospace (e.g., high-performance turbine blades), nuclear energy (e.g., structural components in reactors), chemical processing (e.g., equipment handling corrosive fluids), and biomedical engineering (e.g., implants with enhanced wear resistance and biocompatibility).

Microbiologically Influenced Corrosion (MIC) is a significant concern in numerous industries. It arises from the synergistic interaction of microorganisms with the material surface, accelerating degradation. The exceptional properties of HEAs, particularly their inherent resistance to passive film breakdown, potential for tailored surface modifications, and the integration of antimicrobial agents into the HEA matrix, offer promising avenues for mitigating MIC.

The project delves into the interplay between HEA composition, microstructure, and surface chemistry to explore how these factors influence MIC resistance. It will examine various mechanisms by which HEAs can combat MIC, followed by a showcase of recent research findings.

Keywords: high-entropy alloys; microbiologically influenced corrosion; antibacterial coatings.

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P13

PREVENTING MICROBIOLOGICALLY - INFLUENCED CORROSION USING POLYOXOMETALATE - IONIC LIQUIDS

Mariella Malefioudaki,[a,b] Archismita Misra,[c] Andrea Koerdt[c], Rafael Martín-Rapún,[a,b] Scott G. Mitchell[a,b]

[a] *Instituto de Nanociencia y Materiales de Aragón (INMA-CSIC/UNIZAR), CSIC-Universidad de Zaragoza, C/Pedro Cerbuna 12, 50009 Zaragoza, Spain.*

[b] *CIBER de Bioingeniería, Biomateriales y Nanomedicina, Instituto de Salud Carlos III, 28029 Madrid.*

[c] *BAM, Department of Materials and Environment, Unter den Eichen 87, 12205 Berlin, Germany*

e-mail: malefioudaki@unizar.es

Corrosion of metals and other materials in the marine environment has an estimated global cost of $50-80 billion annually, representing severe problems for the oil & gas and maritime industries. Microbiologically influenced corrosion (MIC) accounts for a substantial portion of this corrosion, with sulfate-reducing bacteria (SRB) and methanogenic archaea (MA) being key contributors [1]. Conventional methods such as cathodic polarization have proven insufficient in mitigating the colonization of corrosive microbial communities in real marine environments, requiring the development of alternative, broad-spectrum antimicrobial strategies to prevent such biofilm formation. Recently, molybdate has emerged as a potential alternative to traditional biocides and nitrate [2]. Our hypothesis is that polyoxometalate-based ionic liquids (POM-ILs) coatings based on anionic polyoxomolybdates with tetraalkylammonium cations exhibiting antimicrobial and anticorrosion properties [3] could be suitable for protecting metal surfaces. Here we show how two prototype POM-IL variants, [(CH3(CH2)6)4N]2[Mo6O19] and [(CH3(CH2)6]4N]4[Mo8O26], have demonstrated significant antimicrobial activity at microgram per millilitre (µg/mL) concentrations and metal surface-active biocidal activity (on brass, iron, aluminium, and steel). Furthermore, electron microscopy and impedance measurements demonstrated how POM-IL-coated brass and iron coupons withstood extremely corrosive environments. These proof-of-concept results demonstrate that multi-functional POM-IL coatings offer promising prospects for mitigating MIC and addressing the challenges of marine biofouling by providing an acid-resistant hydrophobic protective layer and preventing biofilm formation. On-going research includes evaluating the effectiveness of POM-ILs against MIC-inducing microorganisms (e.g., SRB and MA), exploring the material-metal interactions, and evaluating the ability to prevent fouling in marine environments over longer periods of time.

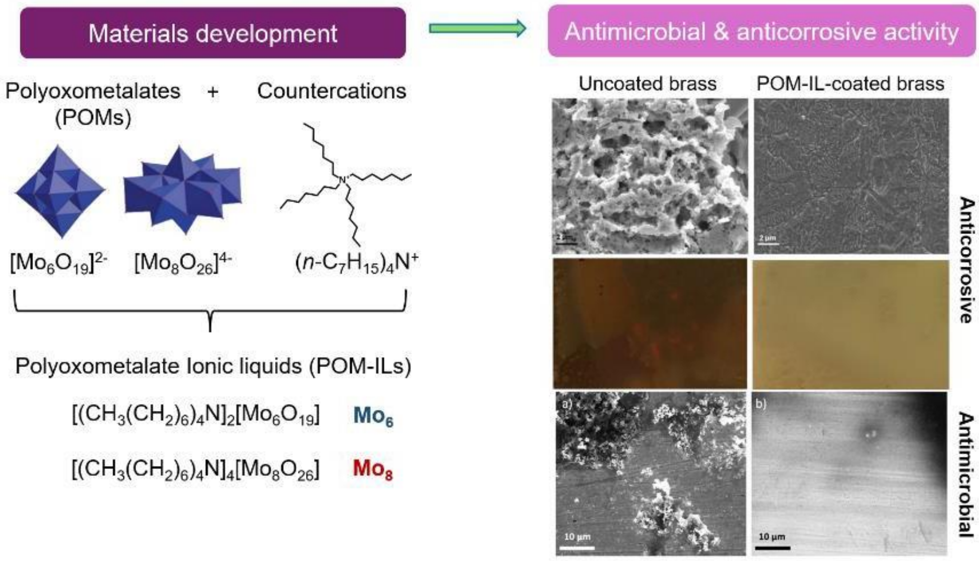

Figure 1. Illustration of the material development along with ESEM and photographic images of uncoated brass and POM-IL-coated brass exposed to aqueous acetic acid (10%) and microbial colonisation.

Keywords: polyoxometalate, ionic liquid, antimicrobial, Microbiologically - influenced corrosion, metal corrosion.

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P14

IN-VITRO DEVELOPMENT OF O&G MICROBIAL BIOFILMS AND ASSESSMENT OF BIOCIDAL EFFECT THROUGH CONFOCAL LASER FLUORESCENCE MICROSCOPY IMAGE ANALYSES

Francisco L. Massello[1], Joaquín Dos Santos[1], Agustina Piatti[1], Facundo Porcel de Peralta[1], Maria Clara Pagliaricci[1], Graciela Rojas[1], Walter Vargas[2], Isabel Vega[1]

[1] *YPF Tecnología S.A. (Y-TEC), Berisso, Argentina*
[2] *YPF S.A., Buenos Aires, Argentina*

Microbial biofilms in the Oil and Gas (O&G) industry are well known for their potential catalysis of surface corrosion (MIC); however, they can also compromise the production quality due to souring, plugging, and clogging. Furthermore, if failures occur, such as leakages, they may have severe impact on both the environment and personnel safety. Consequently, microorganisms, and especially microbial biofilms, are a concern for the integrity, quality, safety, and the production of the O&G industry, involving billions of dollars per year. Studying microbial biofilms in the O&G proposes serious challenges related to the heterogeneous nature of these biological structures, but also, due to the complexity of the environment in which they grow (i.e., the operating conditions of the facilities). Therefore, the improvement of methods and strategies for the study of biofilms is essential for the development of more effective products to limit their risk and damage. In this study, the efficiency of antimicrobial products on O&G biofilms grown *in-vitro* was assessed through *ad-hoc* analyses of Confocal Laser Fluorescence Microscopy (CLFM) images.

Produced waters from an unconventional O&G production facility in Argentina were cultured in CDC reactors (BioSurface Technologies, USA) and biofilms were collected on steel coupons. Cultures were designed to enrich the target microorganism of the shale, *Halanaerobium*. Coupons were treated with different antimicrobial formulations of interest that were previously evaluated on planktonic cells. Microbial activity was measured through ATP determination (LuminUltra, Canada), while the structure of treated and untreated biofilms was analysed by CLFM using three specific stains: Syto 9, Propidium Iodide and Calcofluor White. Resulting images were analysed with Fiji software (Schindelin *et al.*) using an *ad-hoc* workflow designed to obtain information related to the biofilm depth and volume, and to analyse the spatial distribution of living cells, dead cells, and EPS. These results provided information about how the evaluated biocides affect the biofilm structure, including the eradication potential, the effect on EPS matrix, the killing rate, helping to select and design a better antimicrobial product to apply at the facility.

Keywords: Biofilms; Biocides; Image Analysis; Halanaerobium

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