Investigation of the anti-biofilm properties of purified biosurfactants in Enterococcal biofilms

Investigation of the anti-biofilm properties of purified biosurfactants in Enterococcal biofilms

Patricia Puyol McKenna[1], James S.G. Dooley[1], Nigel G. Ternan[1], Ibrahim M. Banat[1], Patrick Lemoine[2], Patrick J. Naughton[1].
[1]*Nutrition Innovation Centre for Food & Health (NICHE), Ulster University, Coleraine, BT52 1SA*
[2]*School of Engineering, Nanotechnology and Integrated Bioengineering Centre (NIBEC), Ulster University, Belfast, United Kingdom*

Ulster University
Faculty of Life and Health Sciences
ulster.ac.uk/niche

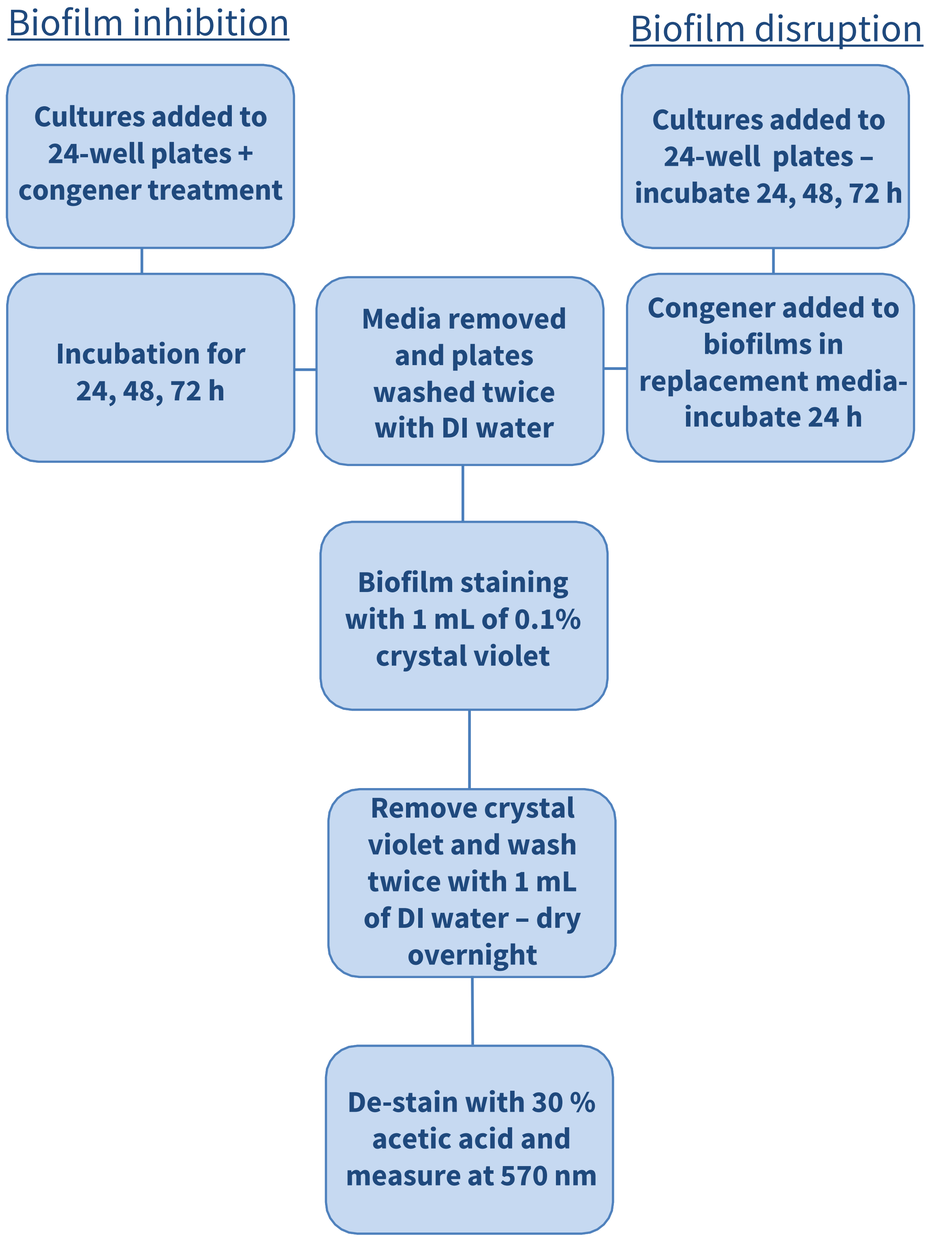
Introduction

- *Enterococcus* spp. have become a leading causative of healthcare-associated infections (HAIs) ranging from bloodstream infections to UTIs[1]. Some 30% of healthcare-associated enterococcal infections are caused by vancomycin-resistant enterococci (VRE), requiring prolonged antibiotic therapy or becoming resistant to antibiotic monotherapy[2].
- Biofilms, structured biological communities that grow enveloped in an extracellular matrix of protective polysaccharides that subsequently prevents the action of antimicrobial agents[3], can develop on an array of surfaces, from indwelling medical devices such as catheters to infected heart valves, facilitating chronic infections[4].
- Biosurfactants are unique amphiphilic molecules of microbial origin that are capable of interacting with the lipidic components of microorganisms[5]. Biosurfactant interactions with different surfaces can affect their hydrophobic properties; their ability to alter microorganisms' adhesion abilities and consequent biofilm formation[6] could make biosurfactants suitable for targeted use in medical and pharmaceutical applications[7].

Aim/Objectives

- To determine the inhibitory effects of 24-hour biosurfactant congener treatment (Mono Rhamnolipid) on 24, 48 and 72 hour *E. faecalis* biofilms.
- To determine the biofilm disruption effects of the congeners on established 24, 48 and 72 hour *E. faecalis* biofilms.
- To determine minimum inhibitory and biocidal concentrations of the congener by broth microdilution.

Methods



Conclusions

- **Mono-RL** shows significant (P<0.001) efficacy in biofilm inhibition and disruption. Results show a **dose-dependent response**, with higher concentrations of biosurfactants leading to greater inhibition/disruption of biofilm growth.
- Preliminary results suggest that biosurfactants may offer a promising alternative or adjunctive treatment option against enterococcal biofilms.

Mono-Rhamnolipid structure

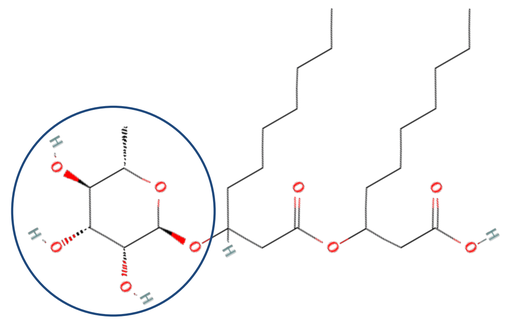


Fig 1. Rha-C_{10}-C_{10}. Molecular weight = 504.7 g/mol. Structure of RL consists of 2 β-hydroxyl fatty acids attached by a glycosidic linkage to 1 rhamnose sugar (PubChem, 2021).

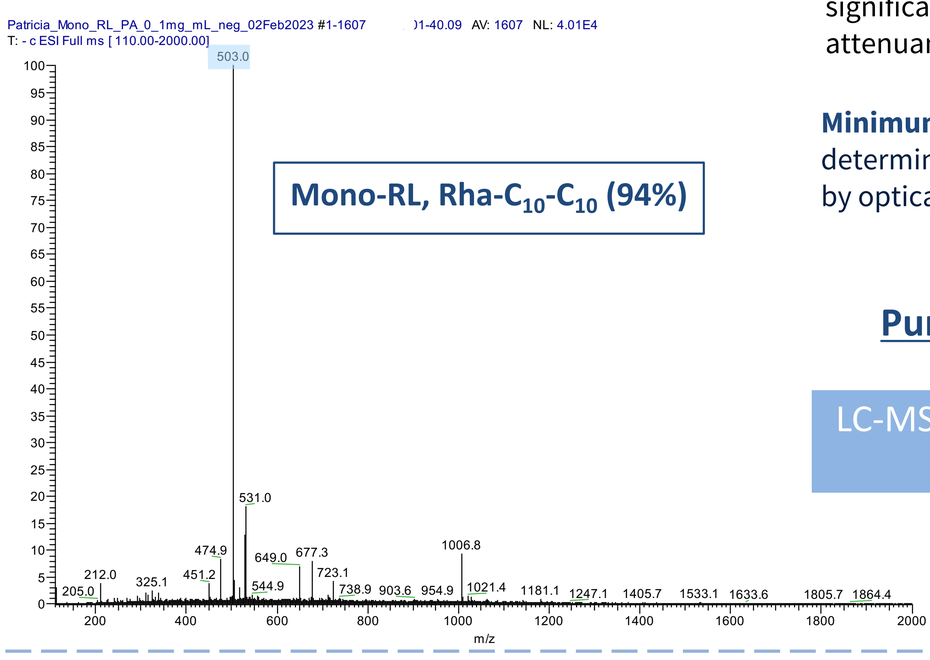


Table 1. MIC and MBC of Mono-RL for *E. faecalis* strains.

Strain (*E. faecalis*)	Mono Rhamnolipid (µg/mL)	
	MIC	MBC
MW01105	250	500
MF06036	250	500
MW02048	250	500

MIC –the minimum concentration at which there is a significant decrease in growth as measured by attenuance (D) at 600 nm.

Minimum inhibitory and biocidal concentrations– determined by broth microdilution, growth measured by optical density at 600 nm.

Purity of Mono-Rhamnolipid

LC-MS/ESI mass spectrum in negative ion mode – **purity 94%**

Results

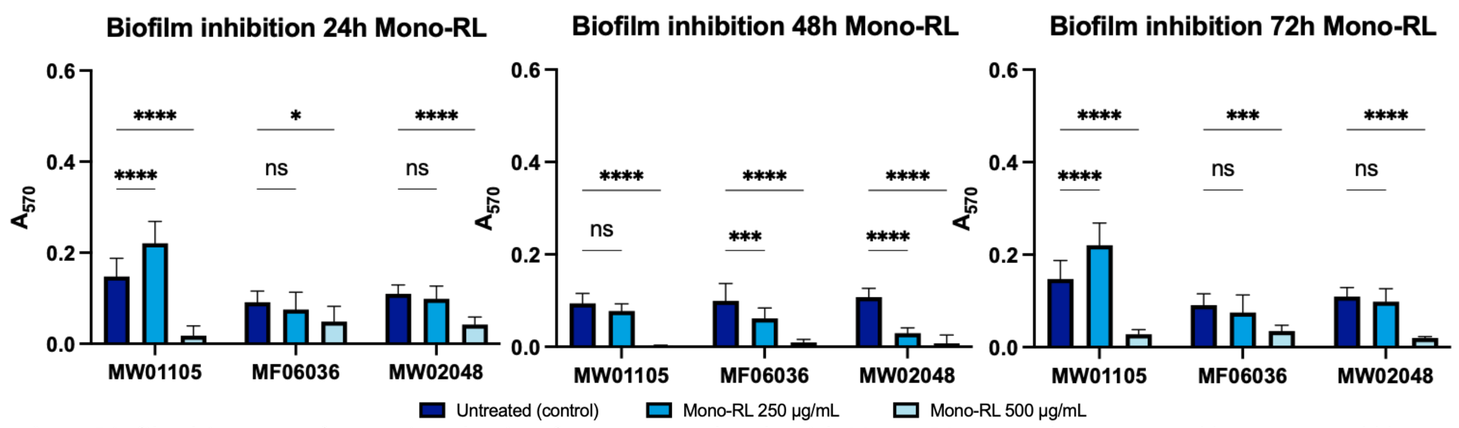


Fig 3. Biofilm biomass after co-incubation for 24, 48 and 72 h with Mono-RL. n = 9, * = $p < 0.05$, ** = $p < 0.01$, *** = $p < 0.001$, **** = $p < 0.0001$.

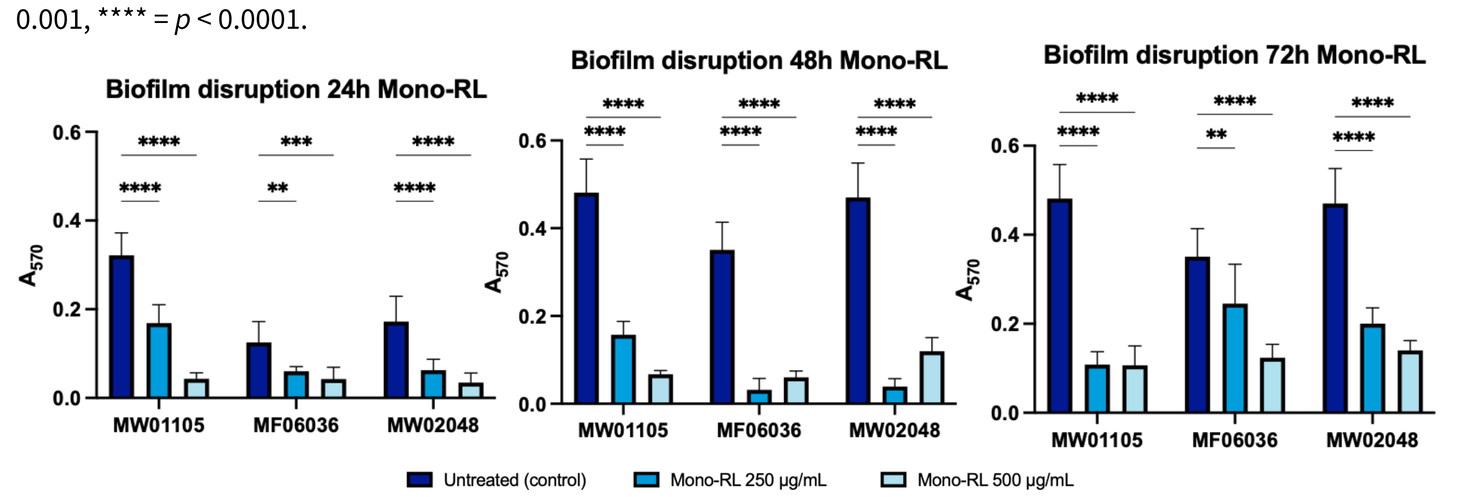


Fig 4. Biofilm biomass after 24, 48 and 72 h with a futher 24 h of Mono-RL treatment. n = 9, * = $p < 0.05$, ** = $p < 0.01$, *** = $p < 0.001$, **** = $p < 0.0001$.

Funding and acknowledgements. PPM was recipient of a PhD studentship awarded by Department for the Economy (DfE) NI. PPM acknowledges travel funding from the Medical Research Foundation.

1. Codelia-Anjum et al.,(2023). Enterococcal Urinary Tract Infections: A Review of the Pathogenicity, Epidemiology, and Treatment. *Antibiotics*.
2. CDC (2019). Biggest Threats and Data. Centers for Disease Control and Prevention.
3. Luo et al.,(2021). Mechanisms and Control Strategies of Antibiotic Resistance in Pathological Biofilms. Journal of Microbiology and Biotechnology
4. Conwell et al.,(2022). Enterococcal biofilm – a nidus for antibiotic resistance transfer? Journal of Applied Microbiology.
5. Ceresa et al.,(2020). The effect of sophorolipids against microbial biofilms on medical-grade silicone. Journal of Biotechnology.
6. Kaczorek et al., (2018). The Impact of Biosurfactants on Microbial Cell Properties Leading to Hydrocarbon Bioavailability Increase.
7. Ferreira et al., (2017). Novel cosmetic formulations containing a biosurfactant from Lactobacillus paracasei. Colloids and Surfaces B: Biointerfaces

@NICHE_ULSTER niche_ulster puyol_mckenna-p@ulster.com NICHE Ulster / UlsterUniversityNICHE