Department of Food and Nutritional Sciences

Lipopeptide biosurfactants against foodborne pathogen and spoilage microorganisms

Constantina Kourmentza | Kimon-Andreas Karatzas | Vincent Lemoinge | Francois Coutte | Paula Jauregi

Introduction

Biosurfactants are surface-active-agents that are produced as secondary metabolites , secreted to the growth media or attached to the outer cellular membrane, by a variety of microorganisms.

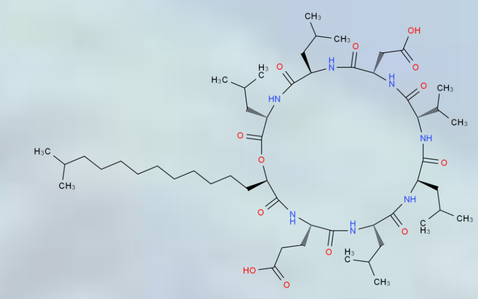
Lipopeptide biosurfactants are amphiphilic molecules that consist of cyclic peptide structures of 7-10 amino acids while their hydrophobic part comprises of a C_{13}-C_{19} fatty acid chain.

They are mainly produced by *Bacillus* and *Pseudomonas* species as well as *Serratia marcescens*[1,2]. Depending on their amino acid cyclic sequence they can be classified to different groups. For example, lipopeptides produced by strains of *Bacillus subtilis* fall into the groups known as surfactin, iturin and fengycin families.

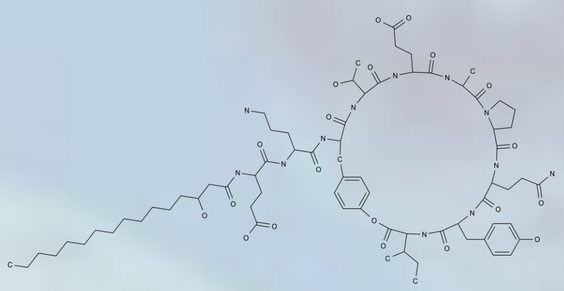
Scope of the study

Lipopeptides were produced by *Bacillus* sp. (Lipofabrik, SAS) and were assessed regarding their antibacterial activity against foodborne pathogen and spoilage bacteria. The antibacterial susceptibility tests were performed according to the broth microdilution methodology previously described[3].

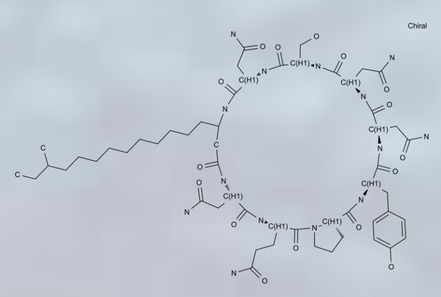
The lipopeptide structures tested are shown below.



Surfactin



Fengycin



Mycosubtilin

Iturin A

Mixtures of those lipopeptides were also examined.

Nisin, which is an approved food preservative of microbial origin, and EDTA, that has been used for the control of microorganisms and biofilms, were also tested for comparison.

Tests were performed against both gram-negative and gram-positive bacteria associated with food spoilage.

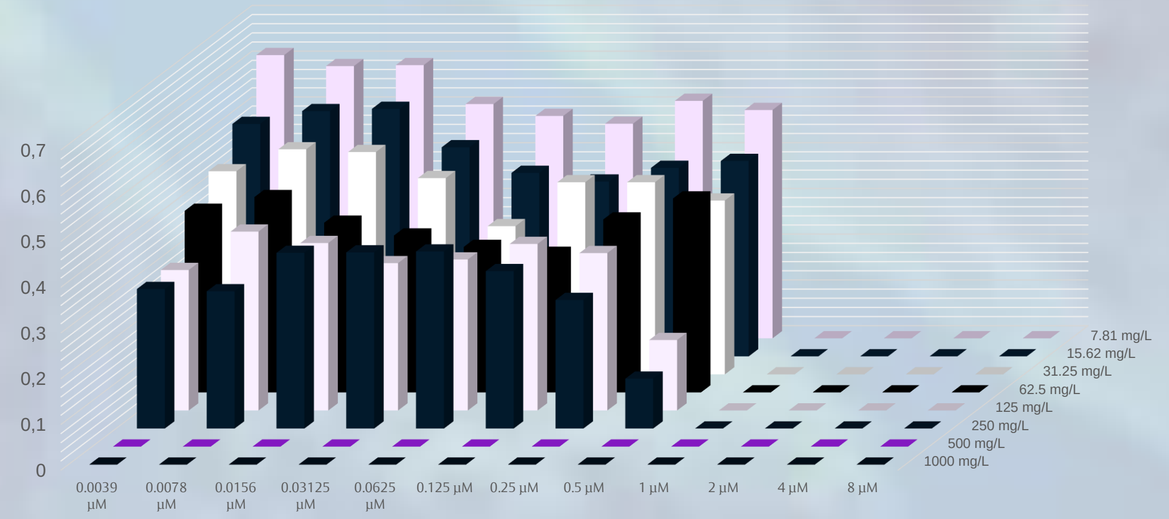
Results and discussion

- Since lipopeptides are water-insoluble compounds DMSO was used as diluent.
- Growth tests in the presence of different % of aqueous DMSO solutions were performed and results showed that DMSO percentage should not exceed 1% as it inhibits microbial growth.
- Results obtained on the minimal inhibitory concentrations (MIC) are summarized below:

Compound	Strain examined					
	P. aeruginosa	*S. enterica*	*E. coli*	*B. cereus*	*L. monocytogenes*	*C. divergens*
Nisin	> 32 µM	> 32 µM	> 32 µM	2 µM	2 µM	1 µM
EDTA	25 mM	50 mM	50 mM	0.78 mM	0.78 mM	0.19 mM
Surfactin (S)	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L
Fengycin (F)	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L
Mycosubtilin (M)	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	0.5 g/L
S/F	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L
S/I*/F *Iturin A	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L
M/S	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	> 1.0 g/L	0.5 g/L

- Combinations of nisin, EDTA and mycosubtilin against *C. divergens* did not show any synergistic effect.

Nisin (8 µM) - Mycosubtilin (1000 mg/L) against *C. divergens*
OD at time = 24h



Conclusions

- Nisin and EDTA are more active against gram positive bacteria,
- *C. divergens* was the only bacteria susceptible to mycosubtilin and the M/S mixture,
- Combinations of nisin, EDTA and mycosubtilin did not show any synergistic effect,
- Lipopeptides are active as antifungal agents but their antibacterial activity is quite limited, and reported only for mycosubtilin against *M. luteus*[4].

References

1. Matsuyama et al., 1992; **DOI:** 10.1128/jb.174.6.1769-1776.1992
2. Kourmentza et al., 2017; **DOI:** 10.1007/978-3-319-52666-9_2
3. Qaiyumi S. 2007, **ISBN:** 9780824741006
4. Besson et al., 1978; **PMID:** 658439

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Contact information

- Department of Food and Nutritional Sciences, University of Reading, Whiteknights, RG6 6AH
- Email: k.kourmentza@reading.ac.uk | www.reading.ac.uk/food/fns-meet-the-team.aspx
  p.jauregi@reading.ac.uk | https://www.reading.ac.uk/food/about/staff/p-jauregi.aspx