Lipopeptides from *Bacillus subtilis*: potential biomolecules to functionalize bio-based smart packaging for the enhancement of food preservation (BIOSMART)

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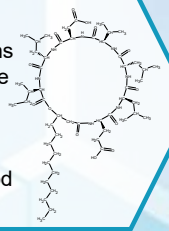
The BIOSMART project

- The BIOSMART project has the ambition to develop active smart bio-based and compostable packages addressing the needs of fresh and processed food applications.
- In this work, the lipopeptides from *B. subtilis* are evaluated for the first time as to cytotoxicity activities and their ability to functionalize bio-based packaging material.

BIOSMART
BIOMATERIALS FOR SMART FOOD PACKAGING

What are lipopeptides ?

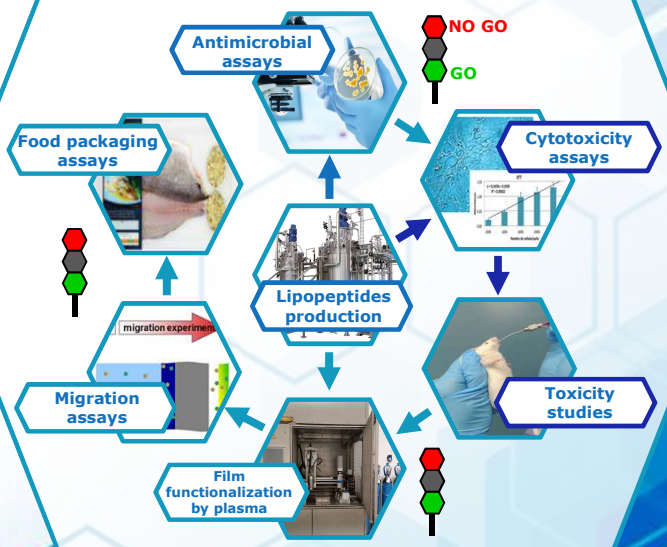
- Lipopeptides are secondary metabolites produced by *B. subtilis*. *B. subtilis* produces three different families of lipopeptides : surfactins, fengycins and iturins (mycosubtilin and iturin A). These molecules consist in cyclic peptide linked to fatty acid chain.
- Lipopeptides display bio-surfactant, anti-biofilm (surfactins), anti-fungal (fengycins and iturins), and/or anti-yeast activities (iturins). Thanks to their activities, they represent a good source of active molecules to functionalize food packaging.



Food packaging specification

- Two types of food packaging prototype (under vaccum and thermoformed) will be made with different food targets.
- The use of biomolecules in food packaging complies with specific regulations required by FDA (USA) and EFSA (EU).
- A key advantage will be the low cytotoxicity of the molecules and their good immobilization on various plastic package surfaces, avoiding migration into the food content of the package.

1. Procedure to select lipopeptides for use in food packaging



2. Cytotoxicity study on Vero-SF cell line

- First screening step on cytotoxicity was conducted on the 3 families of each lipopeptides and as mixtures on Vero-SF

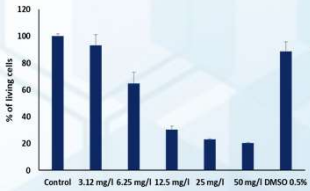


Figure 1: Example of cytotoxicity results obtained with a mixture of two lipopeptides (surfactin and mycosubtilin). Assay was realized by exposing Kidney epithelial cells extracted from an African green monkey (Vero-SF) with the molecule during 24h (n= 6)

Figure 2: EC_{50} of the different lipopeptides on Vero-SF. Values from literature for Nisin (bacteriocin authorized as food additive) and Ricin (strong poisoning agent) are added for comparison (n=6).

3. Film functionalization by plasma

- Study of package surface functionalized with these molecules is undertaking using Contact angle, ATR-FTIR, AFM and Raman spectroscopy.

- Contact angle
- FTIR / RAMAN
- AFM

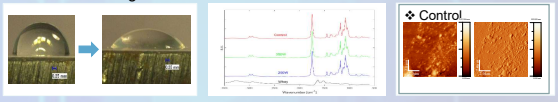


Figure 3: Methods used for surface characterization after plasma coating of the lipopeptides. Contact angle for the surface hydrophobicity, FTIR for the chemical surface composition and AFM for surface topography.

- These experiments are ongoing and will allow to select the best lipopeptide candidate to bring antimicrobial properties to a new packaging prototype.

Acknowledgements

This project has received funding from European Union's Horizon 2020 research and innovation programme under grant agreement n° 745782

Charles Viollette research Institute
Université de Lille
Lipofabrik
BIO-BASED INDUSTRIES Public-Private Partnership
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