Implementation of structured metal packing for the design of biofilm reactor : analysis by high energy X-ray tomography and application to the production of lipopeptides by *Bacillus subtilis*

Quentin Zune[1], Delphine Soyeurt[1], Marc Ongena[2], Dominique Toye[4], Philippe Thonart[3], Frank Delvigne[2]

Context and objectives

Whereas multi-species biofilm reactors are commonly used for treatments of water and gas effluents, new strategies are arising for the development of mono-species biofilm reactors in order to produce high added value molecules. Thus, it is required to design new bioreactors able to promote biomass growth as biofilm and to identify the key physico-chemical parameters involved in order to optimize the bioprocess.

Methodology

- In this work, an experimental setting (Fig 1A) comprising a liquid continuously recirculated on a metal structured packing (Fig 1B) was used in order to promote *B. subtilis* GA1 growth as biofilm.
- X-ray tomography was performed in order to non-invasively visualize repartition of the biofilm mass inside the metal structured packing (Fig 1C).
- Mass balance of the process was calculated in order to know repartition of the biomass between liquid phase and packing and to measure bioconversion yield in cellular biomass and exo-polysaccharides (EPS).
- Quantification of lipopeptides by LC-MS was performed from samples of the liquid phase during the fermentation. These data were compared with those of a submerged culture carried out in a stirred tank reactor.

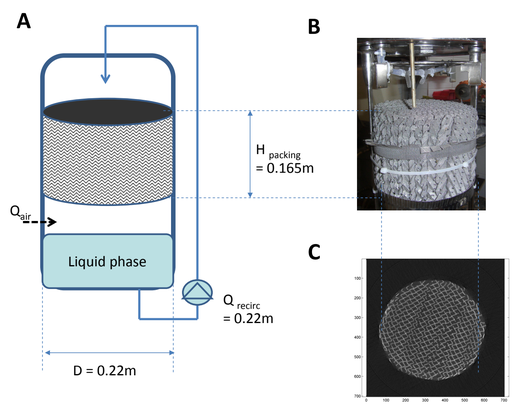

Figure 1

Results ➔ Discussion

- There is a conical repartition of the biofilm mass inside the packing as well as the presence of clogging (Fig 2).
- Cellular biomass yield is lower than Smf culture because of matrix production but surfactin abundance is higher than Smf culture to the detriment of iturin and fengycin (Fig 3a,3b and 4).
- The packing fills a part of useful volume and decreases productivity (Tab 1).
- Thanks to its design (no stirring but aeration above the liquid phase), there is no foam formation (Tab 1).

- X-ray tomography proves it is an efficient tool to study effects of physico-chemical parameters on growth and distribution of biofilm.
- Lower cellular growth rate in biofilm induces higher surfactin production rate (Ongena 2005) ➔ however, clogging causes bad mass transfer that impedes good surfactin recoveries in the liquid phase. Thus, there is a need to improve biofilm distribution in thin layers.
- No foam formation is a great technological progress for Bacillus subtilis fermentation.

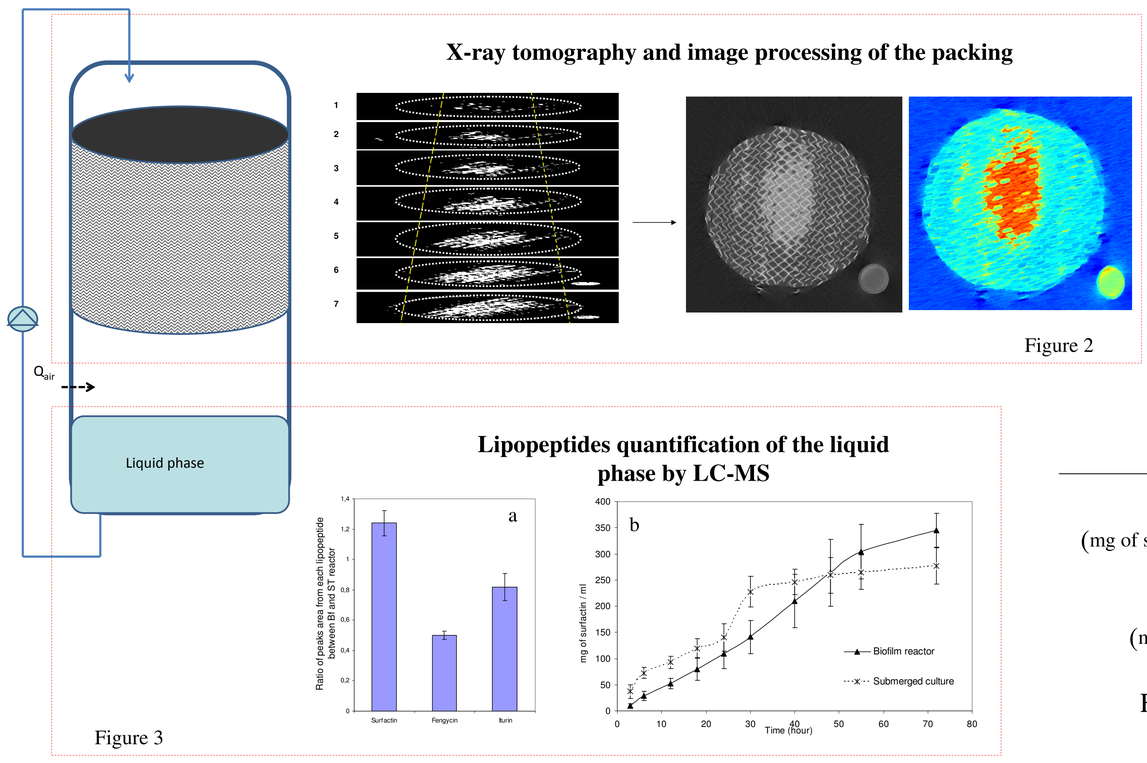

Figure 3

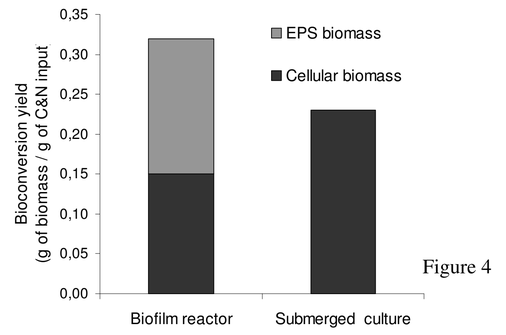

Figure 4

Table 1	Submerged culture		Biofilm reactor
$Y_{surf/X}$ (mg of surfactin / g of dry biomass)	22,8 ± 2,8	<<	37,9 ± 2,2
Productivity (mg of surfactin L^{-1} h^{-1})	3,3 ± 0,4	>>	2,1 ± 0,2
Foam formation	YES		NO

CONCLUSION

In conclusion, the experimental setting leads to a major technological progress avoiding foam formation and increasing surfactin production. Nevertheless, significant improvements are required at the level of the biofilm distribution in thin layers inside the packing in order to increase mass transfer and lipopeptides recoveries. Further investigations will be devoted to the optimization of the physico-chemical parameters involved in biofilm distribution.

[1]Univ. Liege- Gembloux Agro-Bio Tech. Bio-Industries Unit. Passage des Déportés, 2. B-5030 Gembloux (Belgium). Doctoral thesis funded by FRIA.
[2]Univ. Liege-Gembloux Agro-Bio Tech. F.R.S-FNRS. Bio-Industries Unit. Passage des Déportés, 2. B-5030 Gembloux (Belgium).
[3]Univ. Liege- Gembloux Agro-Bio Tech. Bio-Industries Unit. Passage des Déportés, 2. B-5030 Gembloux (Belgium).
[4]Univ. Liege. Chemical Engineering Laboratory. Allée de la Chimie, 3/6c. B-4000 Liege (Belgium).

Contact : qzune@doct.ulg.ac.be