Design of a biofilm reactor comprising a metal structured packing for the production of lipopeptides by *B. subtilis*

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Context and objectives

- During exponential and stationary growth phase, *B. subtilis* excretes lipopeptides which are metabolites with excellent surface active properties for a lot of applications. However, processes based on submerged culture in stirred-tank reactor involve important amount of antifoam and make difficult downstream processes.
- In this context, design of a biofilm reactor is investigated to suppress foam formation and improve productivity. Biofilm formation is monitored and visualized by X-ray tomography.

Methodology

- The biofilm of *B. subtilis* S499 is promoted by the media recirculation on a metal structured packing located in the top of the reactor.
- Matlab image processing of X-ray tomography data allows to monitor biofilm distribution on the packing and permits to estimate biofilm volume.
- Samples of liquid phase supernatant are taken during the culture and analysed by LC-MS in order to quantify lipopeptides concentrations.

Results and discussion

- This experimental setting (Figure 1) allows a good liquid distribution on the metal packing and promotes biofilm growth.
- Decrease of optical density in liquid phase (Figure 2) after 12 hours of recirculation means that cells begin to colonize the metal structured packing.
- Foam is completely avoided during the culture and surfactine concentration reaches 200 mg/l after 50 hours of recirculation (Figure 2).
- Raw X-ray tomography data are processed in Matlab. Each image contains pixel values proportional to the X-ray attenuation coefficient (Figure 3).
- Image processing by grayscale erosion removes pixels from metal structured packing and permits biofilm quantification (Figure 4).
- Integration of biofilm surface on each cross-sectional area leads to an estimation of the biofilm volume. In our experiments, biofilm volume corresponds to 20\% of the total volume and could still be improved by optimisation of flowrate and liquid distribution on the top of the packing.

CONCLUSION

In this study, an original process based on biofilm formation is developed with an experimental setting leading to the suppression of foam formation and the accumulation of lipopeptides in the liquid phase. However, some parameters like flow rate and liquid distribution must still be optimized in order to improve packing colonization by biofilm.

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