

## REMOVAL KINETICS STUDY OF PSEUDOMONAS FLUORESCENS BIOFILM FROM CLOSED SYSTEMS

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### ABSTRACT

A major issue for food industries is the cleanliness of production lines, in order to avoid any cross-contamination of the processed food. Indeed, bacteria present in foodstuff are able to adhere to a wide range of surfaces and may lead to the subsequent development of biofilms, complex structures composed of a polymeric matrix in which bacteria are entrapped. Such structures have often been found in food industries, even in closed equipment (Brooks and Flint, 2008), and these biofilms can reduce heat transfer, increase pressure drop or bio-corrosion and can adversely affect the quality and safety of food in contact. Moreover, micro-organisms entrapped in biofilms are known to be highly resistant to high temperature, biocides, cleaning agents and cleaning procedures and may be 100–1000 times less susceptible than their free-living counterparts (Gram *et al.*, 2007).

Our objectives were to study *Pseudomonas fluorescens* biofilm detachment kinetics from cylindrical stainless steel pipes (2B glazed surface finish; 0.023 m diameter, 0.2 m long) during cleaning in place (CIP) procedures. The focus was first made on the effect of the mechanical action of the flow.

Biofilm growth was carried out under semi-static (low agitation conditions) conditions after filling the pipes by diluted milk (1/10) initially contaminated at  $10^5$  cfu ml<sup>-1</sup> as a nutrient medium for 48 h at 20°C, the milk being renewed after the first 24 h. The surface contamination (total viable count, TVC, per cm<sup>2</sup>) has been quantified after detachment by scraping the internal pipe surface with a piston ring. The biofilm removal was achieved using a pilot plant scale test rig. The flow rates varied from 300 to 5100 l h<sup>-1</sup>, giving mean wall shear stress conditions from 0.015 Pa to 0.8 Pa (Reynolds from 7000 to 120 000). According to the kinetics' shape observed, a two phase model (equation 1) proposed by GInaFiT, a freeware tool to assess non-log-linear microbial survivor curves (Geeraerd *et al.*, 2005), was initially proposed to describe and interpret the bacteria removal kinetics according to the flow parameters.

$$\text{Equation 1: } N/N_0 = f \exp(-k_{max1} t) + (1-f) \exp(-k_{max2} t)$$

with  $N_0$  being the initial bacteria count,  $f$  is the fraction of the initial population in a major subpopulation,  $(1-f)$  is the fraction of the initial population in a minor subpopulation (which is more adherent than the previous one), and  $k_{max1}$  and  $k_{max2}$  [1/time unit] were the specific inactivation rates of the two populations, respectively.

The relevance of this simple model was discussed and potential modeling improvements was proposed based on additional observations using coupons inserted in square pipes with a similar hydraulic diameter.

Brooks JD, Flint SH. 2008. Biofilms in the food industry: problems and potential solutions. *International Journal of Food Science & Technology*. 43:2163-2176

Gram L, Bagge-Ravn D, Ng YY, Gyomoese P, Vogel BF. 2007. Influence of food soiling matrix on cleaning and disinfection efficiency on surface attached *Listeria monocytogenes*. *Food Control*. 18:1165-1171

Geeraerd AH, Valdramidis VP, Van Impe JF. 2005. GInaFiT, a freeware tool to assess non-log-linear microbial survivor curves. *International Journal of Food Microbiology*, 102: 95-105.