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Colloque VIIIB : Biologie structurale en conditions extrêmes

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The Agarolytic system of the marine Flavobacterium *Zobellia galactanivorans* : Structural and biochemical insights into a model system for marine polysaccharide degradation

Zobellia galactanivorans (formerly *Cytophaga drobachiensis*) is a marine Flavobacterium capable of degrading complex polysaccharides such as agar [1]. Agar is widely used in biotechnology and food chemistry and due to the fact that it is a major algal cellwall compound of red algae also of ecological interest [2]. In the marine ecosystem photosynthetic organisms like algae and cyanobacteria are the main producers of organic carbon. This carbon source is subsequently used by marine bacteria, which secrete glycoside hydrolases to attack algae and or hydrolyse cellwalls of dead algae. To understand the mechanisms of carbon release from these nutritional hotspots we decided to analyse the agarolytic system of *Zobellia galactanivorans*. The complete bacterial genome showed the presence of nine family GH 16 agarases, indicating that *Z.galactanivorans* indeed secretes a complex agarolytic system to degrade the natural agar source [3]. Seven original sequences were implemented in a medium throughput cloning strategy. So far three new β-agarases (AgaD, AgaE, and AgaH) could be purified to homogenity, crystallised and diffraction data has been collected to 1.1, 1.5 and 1.8 Angstroem resolution and functional characterization is currently under way. Our work will present the functional and structural comparison of this enzyme family in the attempt to answer the question that intuitively arises of why a single bacterium needs such a relatively high number of agarases. Preliminary analysis of some of the new β-Agarases posess no activity on gelified agarose, which apparently indicates a new substrate specifity, possibly for substituted agarose. The high number of different agarases may furthermore reflect a biological adaption to cope with the highly variable grade of agarose substitution.

[1] Barbeyron, T., L'Haridon, S., Corre, E., Kloareg, B. and Potin, P. Int. J. Syst. Evol. Microbiol. 51 (2001), 985-997

[2] G. Michel, P. Nyvall-Collen, T. Barbeyron, M. Czjzek and W. Helbert. Appl. Microbiol. Biotechnol. 71, (2006), 23-33.

[3] Allouch J., Jam M., Helbert W., Barbeyron T., Kloareg B., Henrissat B., Czjzek M. J Biol Chem. 278, (2003) 47171-47180.