# The structure of endo β-1,4-galactanase from *Bacillus licheniformis* in complex with two oligosaccharide products

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The β-1,4-galactanase from *Bacillus licheniformis* (BLGAL) is a plant cell wall degrading enzyme involved in the hydrolysis of β-1,4-galactan in the hairy regions of pectin. The crystal structure of BLGAL was determined by Molecular Replacement both alone and in complex with the products galactobiose and galactotriose (Figure 1), catching a first crystallographic picture of fragments of β-1,4-galactan. X-ray data for complexes were collected at 100K at EMBL beamlines X13 (galactobiose complex) to 2.2 Å resolution and X11 (galactotriose complex) to 2.5 Å resolution at the EMBL Outstation at DESY (Hamburg, Germany).

As expected for an enzyme belonging to GH-53, the BLGAL structure reveals a $(\beta/\alpha)_8$-barrel architecture. However, BLGAL βα-loops 2, 7 and 8 are long in contrast to the corresponding loops in structures of fungal galactanases determined previously [1, 2]. The structure of BLGAL additionally shows a calcium ion linking the long βα-loops 7 and 8 (Figure 1), which serves the same function as a disulphide bridge in the fungal galactanases. Compared to the substrate binding subsites predicted for *Aspergillus aculeatus* galactanase (AAGAL), two additional subsites for substrate binding are found in BLGAL, -3 and –4. A comparison of the pattern of galactan and galactooligosaccharides degradation by AAGAL and BLGAL shows that although both are most active on substrates with high degree of polymerization, AAGAL is active on short galactooligosaccharides whereas BLGAL shows only negligible activity, which can be explained by the presence of the extra subsites.

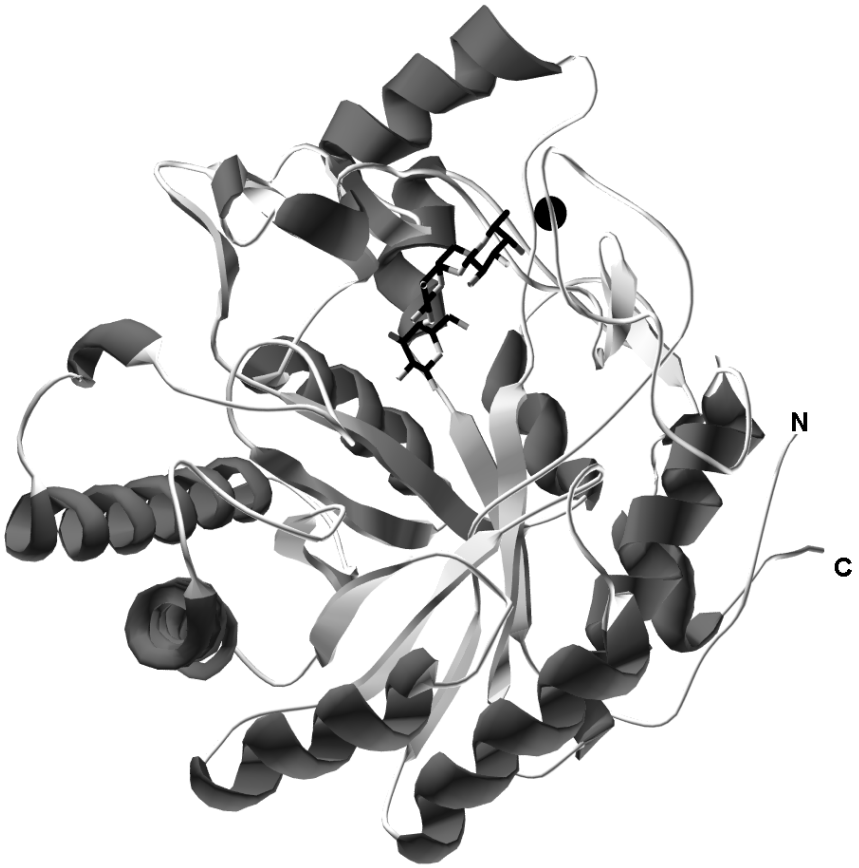


Figure 1: Overall structure of BLGAL in complex with galactotriose

[1] Ryttersgaard, C., Lo Leggio, L., Coutinho, P. M., Henrissat, B., and Larsen, S., Biochemistry 41, 15135-15143 (2002).

[2] Le Nours, J., Ryttersgaard, C., Lo Leggio, L., Østergaard, P. R., Borchert, T. V., Christensen, L. L. H., and Larsen, S., Protein Science 12, 1195-1204 (2003).