MS06-1-8 Structural and functional insight into glycocin-glycosyltransferases
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Abstract
Antimicrobial resistance is on the rise and thus, there is an urgent need to discover novel antimicrobial compounds and determine their mechanism of action.A promising class of such antimicrobials are bacteriocins: Short ribosomally synthesized and post-translationally modified peptides (RiPPs).One subclass of these peptides requires a glycosylation to show antimicrobial activity and thus have been termed glycoactive bacteriocins, Glycocins.[1]Despite their high potency, the challenging synthesis renders the elucidation of the mechanism of action difficult. The correct glycosylation is installed by a dedicated glycosyltransferase that is able to either glycosylate a serine or threonine, resulting in canonical *O*-glycosylation or a cysteine resulting in a rare type of *S*-glycosylation.[2-4]Insights into the function of the cognate Glycocin-Glycosyltransferases (GGTs) will help to understand the unusual specificity of theses transferases of cysteine and/or serine and threonine. Furthermore, these transferases might be useful tools to synthesise glycocins, glycopeptides and neo-glycopeptides in general.We were able to discover several putative Glycocins and their cognate GGTs based on sequence similarity and cluster characterisation.The recombinant production and subsequent purification of five GGTs from *Bacillus subtilis*, *Gottfriedia acidiceleris*, *Laceyella sacchari*, *Enterococcus faecalis* and *Streptomyces platensis*, was established.The recombinant GGTs were characterised in regard of the metal ion dependency and the carbonucleotide specificity by direct measurements of the binding constants with isothermal titration calorimetry.To investigate the molecular determinants for the observed differences in specificity in terms of preferred sugars and their *S*/*O*-selectivity, we use X-ray crystallography as method of choice. For four GGTs crystals could be obtained and the crystal structure for the *B. subtilis* GGT could be solved to 2.6 Å resolution.

References
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Crystal structure of the B. subtilis GTase