

# N°1647 / PC TOPIC(s) : Enzyme production, immobilization / Enzyme discovery and engineering

# Novel activities from an allogenic microbial glycosyltransferase

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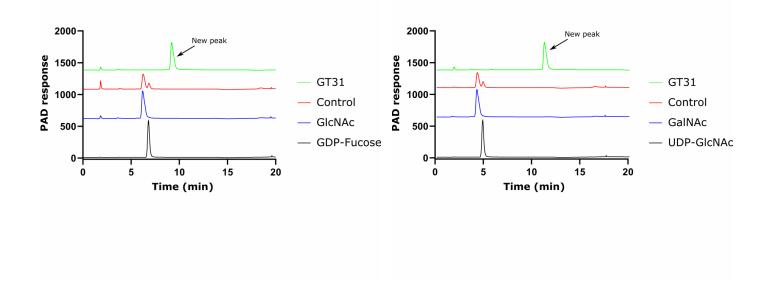
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## PURPOSE OF THE ABSTRACT

Glycosylation is a function prevalent in eukaryotes, prokaryotes, and archaea, and is required for the formation of important glycoconjugates including cell surface glycans, glycosphingolipids, and glycoproteins.[1] The synthesis of such complexes is possible through the use of glycosyltransferases (GTs), enzymes that are ubiquitous in nature, which catalyse such glycosylations through the transfer of activated sugar donors, frequently nucleotide diphosphates (NDPs), to specific acceptors.[2]

GT family classification derives from sequence homology, with each preserving the structural fold and mechanism, and although their activities are substrate specific, families can be utilised with several donor-acceptor substrates.[3] Here, we report studies on a GT from the family 31 (GT31), sourced from the gram-negative gut bacterium Akkermansia muciniphilia. After its expression in Escherichia coli, the enzyme displayed new activities not previously reported for this family.

The successful glycosylation of N-acetylglucosamine (GlcNAc) with fucose, and N-acetylgalactosamine (GalNAc) with GlcNAc indicate these new activities and open the question as to the main activity of this expressed GT. Future work will focus on uncovering the predominant activity of this GT, as well as to determine its structure before its classification can be deduced. Verifying this currently unestablished activity for the GT31 suggests many more activities could still be undiscovered in homologues of other families too. The potential to unlock a library of new activities for attainable, soluble GTs presents an alluring opportunity for the facile assembly of desirable enantiomerically pure glycoconjugates.



# FIGURE 1

#### GDP-fucose and GlcNAc enzymatic reactions

HPLC chromatograms of enzymatic reactions with a donor control (GDP-fucose) (black), an acceptor control (GlcNAc) (blue), an E. coli control reaction (red) and a GT31 reaction (green).

### FIGURE 2

#### UDP-GlcNAc and GalNAc enzymatic reactions

HPLC chromatograms of enzymatic reactions with a donor control (UDP-GlcNAc) (black), an acceptor control (GalNAc) (blue), an E. coli control reaction (red) and a GT31 reaction (green).

## **KEYWORDS**

glycosylation | enzymology | glycosyltransferase | glycoconjugates

### BIBLIOGRAPHY

- [1] Ito T, et al. Biochemistry. 2010; 49 (11): 2604-2614
- [2] Moremen KW, Haltiwanger RS. Nat Chem Biol. 2019; 15 (9): 853-864
- [3] Coutinho PM, et al. J Mol Biol. 2003; 328 (2): 307-317