Cell-specific bioorthogonal tagging of glycoproteins

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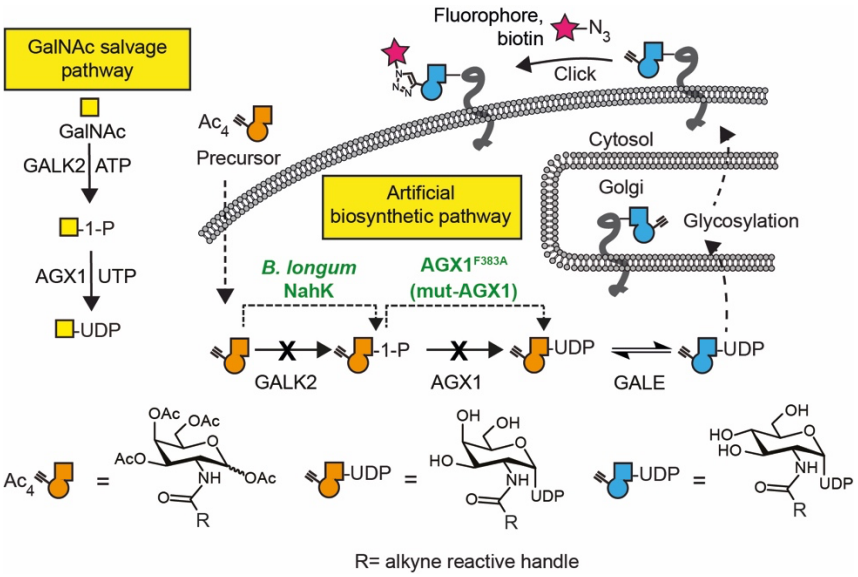
Altered glycoprotein expression is an undisputed corollary of cancer development. Some highly glycosylated proteins such as mucins are clinically recognised cancer biomarkers.[1,2] However, understanding the cancer-related alterations of glycoprotein expression is hampered by limitations in both glycan detection and cellular model systems. For instance, the intricate interactions between tumour and host cannot be adequately recapitulated in monoculture of tumour-derived cell lines. More complex co-culture models usually rely on sorting procedures for proteome analyses and rarely capture the details of protein glycosylation. Bioorthogonal ("clickable") monosaccharides have greatly contributed to our ability to reveal such details, but classically lack specificity for individual cell types.

Here, we develop a tactic to specifically study the cancer-derived glycoproteome in the presence of non-cancerous cells *in vitro* and *in vivo*.[3]Bio-Orthogonal Cell line-specific TAgging of Glycoproteins (BOCTAG) features an artificial biosynthetic pathway that transforms clickable sugars into the corresponding nucleotide-sugars. Only transfected cells incorporate clickable tags into glycoproteins in the presence of non-transfected cells. Modification with suitable reporter probes such as fluorophores or biotin allows for analysis of labelled glycoproteins.[4,5,6] We show that BOCTAG can be tuned to preferentially label either N- or O-GalNAc glycans through engineering of glycosyltransferases. We employ BOCTAG as an imaging technique and to annotate cell-specific glycosylation sites in mass spectrometry-glycoproteomics. Application in co-culture and mouse models allows for profiling of the glycoproteome as an important modulator of cellular function in cancer.



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