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Cell Surface Capturing (Csc) allows for the MS-BASED Identification and Quantification of Cell Surface Antigens

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The molecular composition and dynamic organization of the plasma membrane (PM) determines how a cell can interact with its environment at any given moment. Cell surface proteins are crucial for cell-cell communication, interaction with pathogens, binding of chemical messengers, and responding to environmental perturbations. Alterations in the PM protein composition, as a result of a disease may well define phenotypic and functional differences between normal, and for example tumour cells. Cell surface proteins can be markers of disease and are potential targets for pharmaceutical intervention.

We developed a method for the targeted isolation of glycopeptides from the extracellular domains of PM proteins. This highly specific cell-surface capturing methodology (CSC) is equally applicable to cultured cells, cells isolated from relevant animal models of cancer, or cells recovered from human cancer subjects. CSC is compatible with in vivo isotopic labeling (SILAC) and with after-the-fact chemical isotopic labeling strategies (e.g. ICAT, iTRAQ, etc.) and has the potential to reveal quantitative information, as shown by a quantitative comparison of cell surface glycoproteins from lymphocytes. To date, more than 300 glycoproteins and glycosylation sites were identified from cultured human B and T cells.

The CSC module is integrated into an analysis pipeline, which allows for the mass spectrometric (MS) based identification of cell surface proteins via a modified Trans-Proteomic Pipeline (TPP), including the relational database SISYPHUS. SISYPHUS is a flexible, open source database for storage, annotation and curation of statistically validated TPP proteomic datasets. Furthermore, SISYHUS GlycoBase is a publicly available repository for MS identified cell surface peptides and proteins.

Together, we present a pipeline for the quantitative MS-based identification and analysis of cell surface glycoproteins.

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