High-throughput quantification of N-glycans using 96-well plate based platform

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Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) has been used as a powerful analytical tool for qualitative analysis of glycans owing to mass accuracy, efficiency, and reproducibility, but it has been of limited success in quantitative analysis for both neutral and sialylated glycans. Here, we present an improved high-throughput N-glycan preparation method that enables N-glycan liberation to permethylation using a stack of three 96-well plates. N-glycans from chicken ovalbumin glycoprotein and porcine thyroglobulin glycoprotein have been identified by using this high-throughput system. Moreover, the absolute quantity of total N-glycans including neutral and sialylated oligosaccharides was determined using internal standards. Finally, we applied this technique to confirm the N-glycan from human serum prostate specific antigen (PSA). To the best of our knowledge, this study presents a first attempt to develop an in situ N-glycan preparation method with a high-throughput platform.