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Automated permethylation for glycosylation analysis of biologics using MALDI-TOF-MS

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For most therapeutic glycoproteins the glycosylation patterns correlate strongly with the clinical safety and efficacy profiles. In biological tissues these patterns can also correlate with the state of health or disease of the individual. Given this, there is increasing interest in accurately characterizing changes in glycosylation, for example in Quality by Design studies throughout biopharmaceutical development as well as in glycan biomarker discovery for medical diagnostics. Changes in glycosylation patterns can be complex and subtle and the numbers of samples needed to be analysed can be large, ranging from hundreds to thousands. To perform these studies, reliable systems for high-throughput (HT) glycomics are needed. However, despite many advances in glycosylation analysis there are still problems with current technologies, including low sample throughput, long turnaround times, high cost per sample and labour intensiveness. This talk concerns "LongBow" — a system developed at Ludger for reliable HT glycomics. The "LongBow" system is made up of flexible, modular technologies for semi-automated processing of glycans from a variety of clinical and bio-therapeutic samples and analysis by mass spectrometry (MS) and/or ultrahigh performance liquid chromatography (UHPLC). The focus here will be on permethylated N- and O-glycans analysed by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF-MS). This automated, HT glycan preparation and permethylation method showed to be robust, convenient and fast and can be applied for biopharmaceutical glycan profiling and clinical glycan biomarker studies.

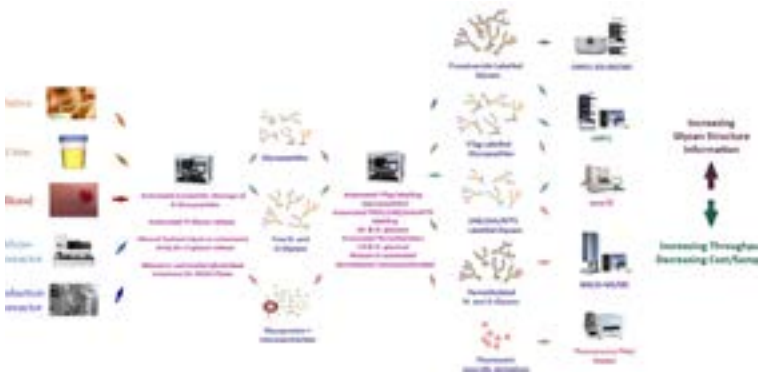

Figure: "LongBow" system developed at Ludger, is made up of flexible, modular technologies for semi-automated processing of glycans from a variety of clinical and biotherapeutic samples.

Recent Publications

1. Ventham N T et al. (2016) Integrative epigenome-wide analysis demonstrates that DNA methylation may mediate genetic risk in inflammatory bowel disease. Nature Communications. 7:13507.
2. Dotz V et al. (2015) Mass spectrometry for glycosylation analysis of biopharmaceuticals. TrAC Trends in Analytical Chemistry. 73:1-9.

3. Ventham N T et al. (2015) Changes to serum sample tube and processing methodology does not cause inter-individual variation in automated whole serum n-glycan profiling in health and disease. PloS One. 10(4):e0123028.
4. Shubhakar A et al. (2015) High-throughput analysis and automation for glycomics studies. Chromatographia. 78(5-6):321-333.

Biography

Archana Shubhakar obtained her Master's Degree in Biochemistry from Bangalore University in India. She worked in the public relations sector for a few years and gained experience in people skills. Currently, she is a Scientist at Ludger (UK) and has been an integral part of the development team, taking new glycobiology products from initial research to final product launch for several years. Her research involvement and accomplishments have been the implementation and development of high throughput analysis and robotization for N-glycan release, glycan derivatization and analysis using a number of orthogonal analytical platforms. As, Ludger is a consortium member of the European and UK funded projects she is also involved in market research, communication with other consortium members and project coordination/management of research projects and grants. She is pursuing her PhD in collaboration with the Department of BioAnalytical Chemistry at Vu University, Amsterdam, The Netherlands under the supervision of Dr. Daryl Fernandes and Prof. Dr. Manfred Wuhrer. Her PhD thesis is entitled "Method development for the discovery of glycosylation biomarkers of inflammatory diseases".

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