BIOGNOSYS
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Biognosys AG
Wagistrasse 21
8952 Schlieren (Zurich)
Switzerland

t: +41 (0) 44 738 20 40
f: +41 (0) 44 738 20 49
info@biognosys.com
www.biognosyscom

Deep Blood Plasma Profiling Using Single Shot Data Independent Acquisition Mass Spectrometry

Marco Tognetti, Christopher Below, Sebastian Müller, Roland Bruderer, Lukas Reiter

Introduction

Blood flows through all organs and has the potential to be informative of the state of the whole human body. Further, it is readily available. Therefore, it is a highly relevant sample type. Even though one can speculate that blood carries all human proteins to a certain extent, deep plasma profiling was rather limited in the past due to the immense dynamic range of plasma of more than ten orders of magnitude. Improvements in sample preparation and total peak capacity of mass spectrometry based workflows have lead to substantial increases of proteome coverage in recent years. We show evidence that this is related to the protein abundance distribution of blood. Finally, we show some data how state of the art mass spectrometry workflows can quantify 4000 plasma proteins using single shot acquisition.

Methods

Plasma samples were measured using Biognosys' TrueDiscovery workflow. Samples were depleted of high abundant proteins in a first step, and analyzed using liquid chromatography ion mobility separation (FAIMS) mass spectrometry (Orbitrap Exploris 480). DIA data was either directly searched using directDIA or analyzed with a library using Spectronaut (Biognosys).

Results

Using a workflow with a high total peak capacity we could quantify more than 4000 proteins using single shot acquisition. This corresponds to a 800% improvement compared to the roughly 500 proteins which were achieved few years ago.

The data suggests that these substantial improvements are due to the particular protein abundance distribution of human plasma.

Conclusion

Due to the particular protein abundance distribution in human plasma substantial improvements in proteome depth could be achieved allowing to profile more than 4000 proteins using a single shot mass spectrometry workflow.