IPD Project Details

Project ID: IPD9355

Project Title: Salivary proteome signatures in the early and middle stages of human pregnancy with term birth outcome

Description: This prospective study is to find changes in salivary protein expressions from first to early third trimester of pregnancy in normal term birth using SWATH-MS strategy. Saliva at three periods of gestation, 6-13 (V1), 18-21(V2), and 26-29 (V3) weeks from 20 singleton pregnant women were used in a discovery phase. Selected proteins from the discovery study were verified by targeted schedule-MRM (multiple reaction monitoring) experiments in a separate subset of subjects (N=14). Using in-house generated saliva-specific protein library, 65 proteins (q-value<0.1) changed as a function of gestational age.

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Sample Preparation: The maternal saliva was collected from the enrolled participants. All samples were buffer exchanged with 100 mM ammonium bicarbonate (pH 8.0)through a 3 kDa MWCO membrane (Amicon Ultra, Millipore).

Peptide Separation: Trypsin were used for proteins digestion at 20:1 protein to enzyme ratio. The digested samples were desalted using C18 fast-flow tips (Pierce Thermo Fisher scientific) and spiked with iRT (Biognosys, Switzerland) at 1:10 ratio. Samples were vacumm dried and resuspended in 0.1% formic acid for LC-MS/MS analysis using an TripleTOF 5600 (SCIEX)in DDA or DIA (SWATH-MS) mode. MRM was performed on a QTRAP 6500+ mass spectrometer.

Protein Characterization: MaxQuant analysis software (version 1.5.4.1) with default settings were used to generate reference spectral library from DDA MS files. All DIA datasets (.wiff files) were converted to HTRMS format before analysis and Spectronaut (version 11) software was used for retention time normalisation and targeted data extraction from the DIA MS files. The protein quantitation and peptide quantities were calculated as a mean intensity within the XIC peak area of the respective fragment ions at MS2. The protein CVs were calculated based on the summed intensities of their

respective peptides.

Experiment Type: SWATH MS, SRM/MRM

Species: Homo sapiens

Tissue: Saliva (bto:0001202)

Cell Type: Saliva

Disease: Unknown

Instrument Details: TripleTOF 5600 (MS:1000932)

Protein Modifications: monohydroxylated residue, acetylated residue, iodoacetamide derivatized residue

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