PEA – a high-multiplex immunoassay technology with qPCR or NGS readout

High Throughput Screening

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This poster will describe a high multiplex proteomics technology using DNA-linked antibody probes with NGS or qPCR readout. The Proximity Extension Assay (PEA) technology combines the best of antibody- and DNA-based methodologies to provide unique, enabling tools for protein biomarker discovery and development. The technique was commercialized by Olink Proteomics AB to develop its range of Olink® biomarker panels. PEA successfully merges an antibody-based immunoassay with the powerful properties of polymerase chain reaction (PCR), and readout using either quantitative real-time PCR (qPCR) or Next Generation Sequencing (NGS). This results in a scalable, multiplex and highly specific method where the concentration of hundreds of protein biomarkers can be quantified simultaneously. The basis of PEA is a dual-recognition immunoassay, where two matched antibodies labelled with unique DNA oligonucleotides simultaneously bind to a target protein in solution. This brings the two antibodies into proximity, allowing their DNA oligonucleotides to hybridize, serving as template for a DNA polymerase-dependent extension step. This creates a double-stranded DNA "barcode" which is unique for the specific antigen and quantitatively proportional to the initial concentration of target protein. The hybridization and extension are immediately followed by PCR amplification. The resulting DNA amplicon can then be quantified either by microfluidic qPCR on a Fluidigm® Biomark instrument, or on the Illumina® NovaSeq platform, depending on the specific Olink product used.