A LARGE SCALE MULTI-CANCER, MULTI-OMICS BIOMARKER STUDY OF >1,800 SUBJECTS INCORPORATING DEEP UNBIASED PLASMA PROTEOMICS

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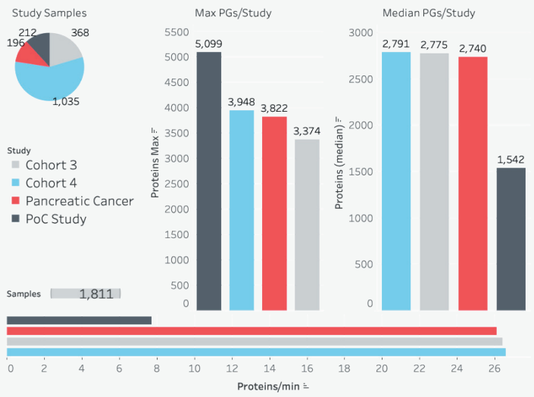
INTRODUCTION

- Cancer is a leading cause of death worldwide[1] and many cancers are diagnosed in the late stage of disease progression with poor prognosis, highlighting the need for early detection.[2]
- Blood-based tests that include multiple analytes may enable identification of biomarkers that provide high sensitivity and specificity for earlier detection and more selective treatment.
- PrognomiQ has developed a multi-omics assay and analysis platform to comprehensively profile blood samples and detect proteins, metabolites, lipids, messenger RNA (mRNA), micro RNA (miRNA), cell-free DNA (cfDNA) fragments, and methylation at CpG sites.
- Deep unbiased proteomics analysis in our platform is facilitated by recent advances in sample preparation (i.e. Seer's Proteograph™ Product Suite) coupled with improved mass spectrometry instrument sensitivity and speed. Together, these technologies provide the ability to quantify thousands of proteins from human plasma at the necessary throughput and reproducibility for large scale biomarker studies.
- To date, we have utilized our multi-omics and analysis platform to deeply profile the genome, epigenome, transcriptome, proteome, lipidome and metabolome of over 1,800 subject samples to identify biomarkers for the early detection of multiple cancers.

RESULTS

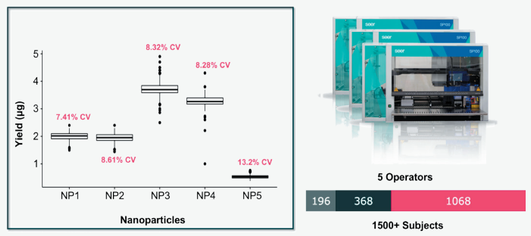
FIGURE 1. >1,800 subject samples were evaluated in four independent plasma cancer biomarker studies



DDA, Data-Dependent acquisition
DIA, Data-Independent acquisition

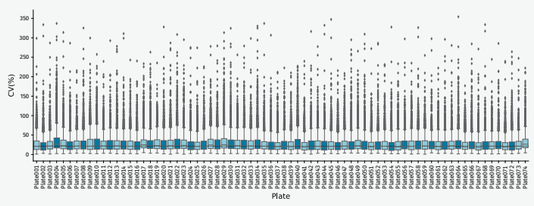
- *Proof of Concept study: 60 min DDA on Dionex Ultimate 3000 - Bruker timsTOF Pro 2. Additional studies: 21 min DIA on Evosep – Bruker timsTOF Pro 2 platform.*

FIGURE 2. Seer Proteograph reproducibility across multiple studies demonstrates capabilities for large scale plasma proteomics



- *191 replicates of a pooled plasma process control sample processed on 3 Proteographs, with 5 operators over 8 months were used to assess reproducibility.*

FIGURE 3. Platform robustness and reproducibility demonstrated by low CV's over 74 batches and multiple instruments in a 1,035 subject biomarker study



CV, Coefficient of Variation

- *Median peak areas of common protein groups for all batches of Process QC samples are below 26.2%.*

FIGURE 4. 3,948 Protein groups were detected across all 5 nanoparticles for 922 of 1,035 subjects with complete data

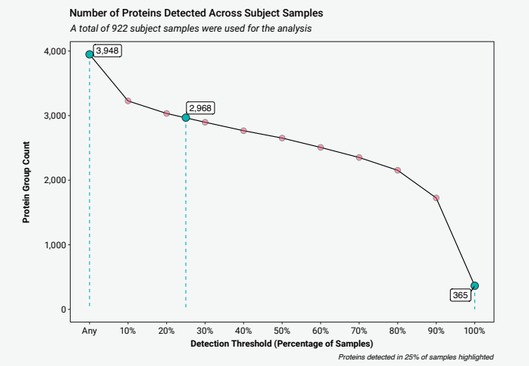
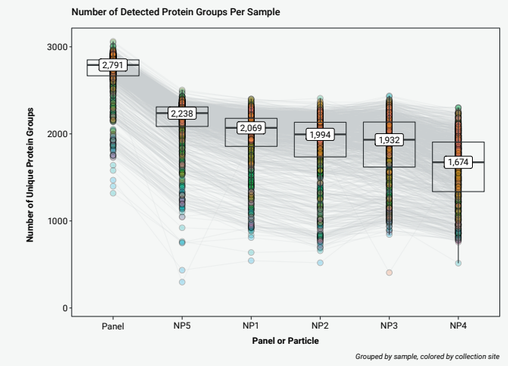
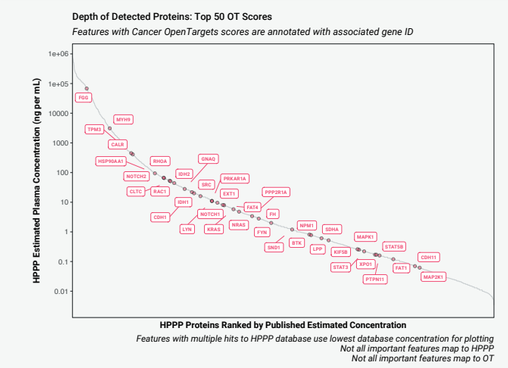


FIGURE 5. A median 2,791 unique protein groups were detected in 1.75 hrs of LCMS acquisition time across all subject samples and 5 nanoparticles



LCMS, Liquid chromatography–mass spectrometry

FIGURE 6. Canonical cancer associated proteins are detected,[3] along with thousands of proteins not currently associated with cancer



OT, Open target
HPPP, Human Plasma Proteome Project

CONCLUSIONS

- Identification of biomarkers from blood for the early detection of cancer requires the ability to conduct large-scale discovery studies with sufficient statistical power.
- We have demonstrated that liquid chromatography-mass spectrometry (LCMS) based plasma proteomics technologies are now capable of providing deep proteome coverage with sufficient reproducibility, robustness and throughput to conduct studies on thousands of subjects with simultaneous detection of thousands of proteins.
- Large-scale LCMS proteomics studies complement other high throughput phenotypic (i.e. metabolites & lipids) and genotypic (i.e. mRNA, miRNA, cfDNA fragments, methylation, etc.) measurements, thus creating an opportunity to develop tests with the necessary sensitivity and specificity for early cancer detection.

DISCLOSURES

Study funded by PrognomiQ. All authors are current or former employees of PrognomiQ.

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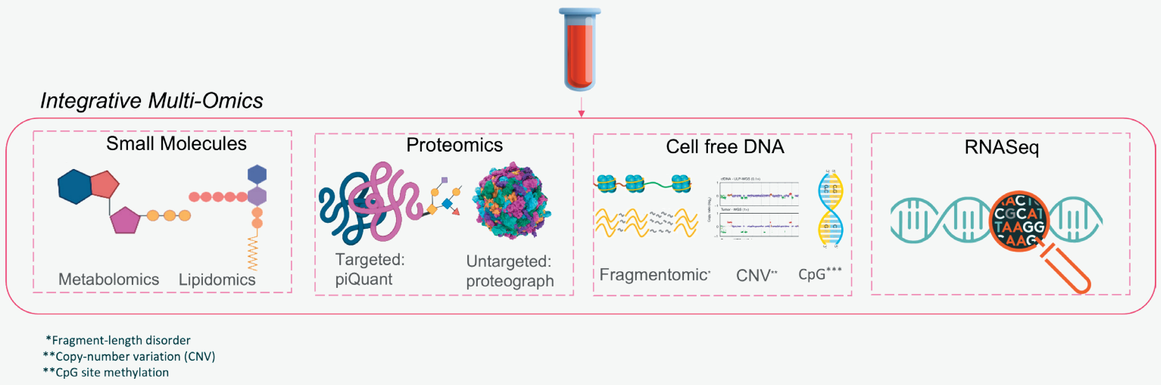
METHODS



- >1,800 human subjects enrolled at 37 collection sites were diagnosed with cancer or enrolled as either comorbid or healthy controls as part of an IRB approved study. Cancer cohort subjects were staged as 1-4.
- Individual assay samples underwent quality control and were prepared and processed using field-standard methods for their specific type. Metabolomics, lipidomics, transcriptomics and genomics data are not shown.
- Hemolyzed samples were excluded.
- Quantitation and normalization were done using field-standard methods specific to each omic.
- Plasma samples were processed with a Proteograph™ using the 5 nanoparticle panel. Peptides were analyzed on a timsTOF Pro 2 using Data Independent Acquisition mode with Parallel Accumulation-Serial Fragmentation (diaPASEF). The data were analyzed using DIA-NN v1.8 implemented on Proteograph™ Analysis Suite (PAS) v1.5.