



Enabling Unprecedented Analysis of Precious Samples in Metabolic and Cardiovascular Diseases

Precious samples, such as aqueous humor, carotid plaque homogenate and plasma-isolated extracellular vesicles offer valuable insights and hold great potential as sources of informative biomarkers. However, their availability is limited by the invasiveness of collection procedures, the potentially challenging workflows, and/or the small volumes that can be extracted. Consequently, the potential for discovering novel and clinically relevant biomarkers is significantly constrained. These limitations are even more pronounced in pediatric populations. The Proximity Extension Assay (PEA) technology is the ideal platform for analyzing biomarkers in these precious samples. Distinguished by its ability to perform highly multiplexed protein measurements from just 1 μ L of sample, PEA technology delivers unparalleled specificity and outstanding sensitivity. Herein, we highlight publications showcasing the use of PEA with a qPCR readout across various sample types obtained through invasive procedures, challenging workflows and/or in limited quantities.

Extracellular vesicles

- Celik S, Sadrian J, Grossi M, et al. Extracellular Vesicle-Associated TWEAK Contributes to Vascular Inflammation and Remodeling During Acute Cellular Rejection. (2023) *JACC: Basic to Translational Science*, DOI: 10.1016/j.jacbts.2022.09.014
- Wu S, Noren Hooten N, Freeman, et al. Extracellular vesicles in diabetes mellitus induce alterations in endothelial cell morphology and migration. (2020) *Journal of Translational Medicine*, DOI: 10.1186/s12967-020-02398-6

Vitreous samples

- Lamy R, Farber-Katz S, Veves F, et al. Comparative Analysis of Multiplex Platforms for Detecting Vitreous Biomarkers in Diabetic Retinopathy. (2020) *Translational Vision Science & Technology*, DOI: 10.1167/tvst.9.10.3

Aqueous humor

- Haq Z, Yang D, Psaras C, Stewart J. Sex-Based Analysis of Potential Inflammation-Related Protein Biomarkers in the Aqueous Humor of Patients With Diabetes Mellitus. (2021) *Translational Vision Science & Technology*, DOI: 10.1167/tvst.10.3.12

Pediatric samples

- Stinson SE, Jonsson AE, Andersen MK, et al. High Plasma Levels of Soluble Lectin-like Oxidized Low-Density Lipoprotein Receptor-1 Are Associated With Inflammation and Cardiometabolic Risk Profiles in Pediatric Overweight and Obesity. (2023) *Journal of the American Heart Association*, DOI: 10.1161/JAHA.122.027042
- Manell H, Shen Q, Chowdhury A, et al. Biomarker screening in children and adolescents reveals that CUB domain-containing protein 1 is associated with obesity and that hepatocyte growth factor is associated with weight gain. (2023) *Obesity Medicine*, DOI: 10.1016/j.obmed.2023.100481

- Heydarian M, Oak P, Zhang X, et al. Relationship between impaired BMP signalling and clinical risk factors at early-stage vascular injury in the preterm infant. (2022) *Thorax*, DOI: 10.1136/thoraxjnl-2021-218083

Artery and coronary sinus plasma

- Kaye D, Nanayakkara S, Wang B, et al. Characterization of Cardiac Sympathetic Nervous System and Inflammatory Activation in HFpEF Patients. (2022) *JACC: Basic to Translational Science*, DOI: 10.1016/j.jacbts.2021.11.007

Carotid plaque homogenate

- Edsfieldt A, Singh P, Matthes F, et al. Transforming Growth Factor- β 2 Is Associated With Atherosclerotic Plaque Stability and Lower Risk for Cardiovascular Events. (2023) *Cardiovascular Research*, DOI: 10.1093/cvr/cvad079

Mouse plasma

→ Kumawat AK, Zegeye M, Paramel GV, et al. Inhibition of IL17A Using an Affibody Molecule Attenuates Inflammation in ApoE-Deficient Mice. (2022) *Frontiers in Cardiovascular Medicine*, DOI: 10.3389/fcvm.2022.831039

→ Li Z, Rasmussen T, Rasmussen M, Li J, Henríquez Olguín C, Kot W, Nielsen D, Jensen T. The Gut Microbiome on a Periodized Low-Protein Diet Is Associated With Improved Metabolic Health. (2019) *Frontiers in Microbiology*, DOI: 10.3389/fmicb.

Conclusion

The unprecedented ability to utilize 1 μ L of sample for extensive protein biomarker profiling ensures that virtually no sample is too limited or too challenging to interrogate, as proven by the highlighted publications. Samples obtained from difficult to access procedure or in limited volumes are a promising source of actionable biomarkers to assess risk, improve diagnoses, predict responses to treatments, and monitor disease progression. Additionally, the low sample volume requirement benefits pediatric cases, where available volumes are limited and minimizing invasive collection is crucial. In conclusion, PEA technology not only allows maximizing data yield from each precious sample, but also enables studies that were previously inconceivable.

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