

## **Extended capability of Visium CytAssist to enable highly-sensitive transcriptomic profiling of PFA fixed, OCT embedded biological specimens**

**Anushka Gupta** (anushka.gupta@10xgenomics.com), 10x Genomics, **Anushka Gupta**, 10x Genomics, **Lauren Gutgesell**, 10x Genomics, **Janine Hensel**, 10x Genomics, **David Sukovich**, 10x Genomics, **Govinda Kamath**, 10x Genomics, **Stephen Williams**, 10x Genomics, **Hanyoung Kim**, 10x Genomics, **Augusto Tentori**, 10x Genomics

The Visium Cytassist platform from 10x Genomics allows spatially-aware, transcriptomic profiling of Fresh Frozen (FF) and Formalin-Fixed, Paraffin-Embedded (FFPE) samples, using probe-based RNA-templated ligation chemistry. Here, we demonstrate extended capabilities of CytAssist to make highly sensitive transcriptomic measurements from PFA fixed, OCT embedded (Fixed Frozen or FxF) samples. FxF tissue processing is suitable for users interested in superior morphological and protein-level measurements. Using CytAssist, users continue to have the built-in flexibility of executing tissue preparation steps independent of Visium slides. Users can select the FxF tissue slide and region that they want to align to the Visium slide based on the morphological, or protein-level landmarks of interest. Here, we characterized the performance of CytAssist on FxF samples by processing eye & brain FF, FFPE, and FxF sections derived from the same litter of mice. Notably, all three sample types had similar sensitivity with ~18,000 unique genes detected in all three sample types. All major morphologically relevant cell-types recovered in FF and FFPE samples were also recovered in FxF samples. Finally, differential gene expression analysis revealed similar transcriptional features in FxF samples as compared to FF and FFPE samples. Overall, Visium CytAssist now enables highly sensitive transcriptomic profiling of FF, FFPE, and FxF samples, while preserving the innate spatial architecture.