QCPRECISE!™ A Novel Spatially Targeted Tissue Microdissection Platform Driven by AI Guided Digital Pathology Imaging Improves Extracted Solid Tumor DNA Purity Relative to Conventional Macrodissection.

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Background

Current methods for molecular analysis of solid tumors rely on pathologist review of a hematoxylin and eosin (H&E)-stained slide to select the region of interest (ROIs) followed by manual macrodissection (MMD) of subsequent serial unstained sections. QuantumCyte has developed a novel platform to extract material from complex ROIs using semi-conductor print masking technology integrated with artificial intelligence (AI)-based digital pathology (QCPRECISE!™). In this study, we compared the QCPRECISE!™ Platform (Figures 1 and 2) to standard MMD to test if the QuantumCyte solution can improve next generation sequencing (NGS) results.

Figure 1: The QCPRECISE!™ Platform workflow.

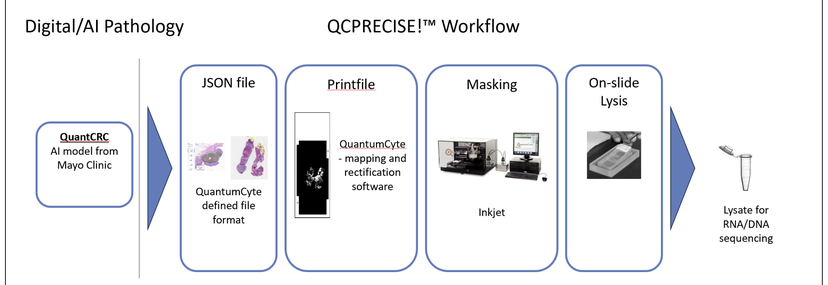
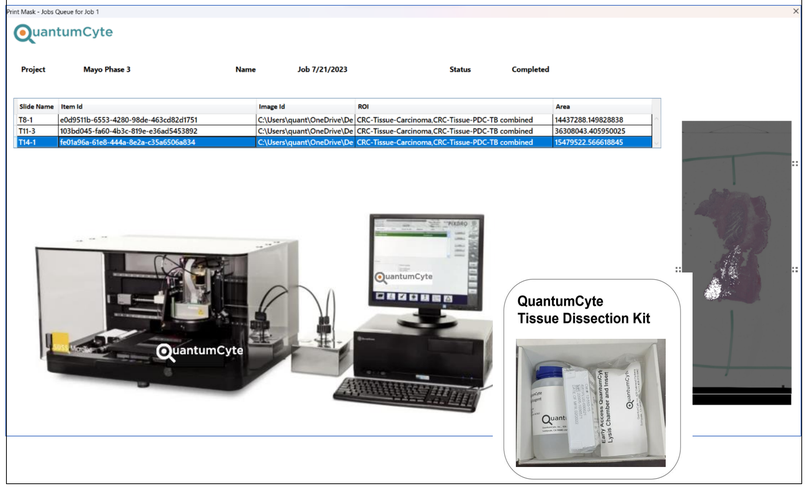


Figure 2: The QCPRECISE!™ platform.



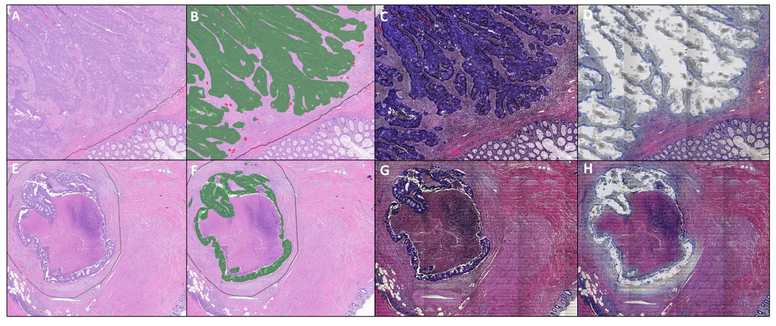
Disclosures

This study was funded by Mayo Clinic. CK, SS, JB, JV, SM, SG, BK and RP are full time employees of Mayo Clinic. BC, KK, DD, AA, JB & JB are full-time employees of QuantumCyte. QCPRECISE!™ Is trademark of QuantumCyte.

Materials and Methods

FFPE colorectal carcinoma (CRC) tissue blocks were sectioned at 5 microns thick on to QuantumCyte's premarked microscope slides. Alternate slides from each CRC sample were designated for MMD or for the QCPRECISE!™ workflow. For the QCPRECISE!™ workflow, all slides were H&E stained, scanned using a Leica Aperio scanner, and ROIs were identified using the QuantCRC AI-algorithm[1] as shown in Figure 3. For MMD sections, a board-certified pathologist reviewed each slide and manually marked tumor ROIs using a Sharpie. Then tumor tissue was harvested by a technician using a scalpel and placed into an Eppendorf tube. gDNA was purified using the Qiagen DSP DNA purification kit protocol. For the QCPRECISE™ process, printfiles were generated using the QCPRECISE!™ software and the masks were printed onto the tissue mounted slides (Figure 1). The mask inhibits access of Proteinase K (PK) digestion. Following the PK formulation and protocol as described in the DSP DNA purification kit, PK buffer was added directly to the slide using a GraceBiolabs Hybriwell placed directly over the ROI. Lysis was then performed but placing the slide into a humidity chamber and incubating it at 56C for 60 min. The crude lysate was recovered from the slide and gDNA was purified following the DSP protocol. NGS was done on the purified gDNA from the 55 CRCs using the MayoComplete Colorectal Cancer Panel. Data was analyzed at Mayo Clinic and results reported as discussed and illustrated in this poster.

Figure 3: Overview of the QCPRECISE!™ process. A, E. H&E image of two colorectal carcinomas. B, F. Tumor region identified in green after application of QuantCRC AI-algorithm[1]. C, G. Post-print image after QCPRECISE!TM chemical mask. D, H. Post-lysis image after on slide digestion demonstrating selective digestion of tumor regions.



Results

Successful sequencing results were obtained in both the manual and QCPRECISE!™ process for all 55 CRCs. Between the two methods, 111 concordant pathogenic variants were identified. In the QCPRECISE!™ samples, the variant allele frequency (VAF) was increased in 108/111 variants by a median of 73.5% (Figure 5a). Eighteen pathogenic variants were identified only in the QCPRECISE!TM samples, whereas only 1 pathogenic variant seen in the manual macrodissection sample that was not seen in the QCPRECISE!TM sample. There was a negative correlation between %tumor as determined by QuantCRC and increase in VAF (r = -0.537, P<0.001) (Figure 5b). This indicates that the QCPRECISE!™ process increases VAF compared to the manual process as %tumor within the FFPE section decreases. This further validates the QCPRECISE!™ process of selective enrichment of tumor DNA

Results (continued)

Figure 4: Pre- and post-lysis images of spatially targeted microdissection of clusters of Pseudo-Dentritic Cells (tumor buds) using the QCPRECISE!™ platform and the Mayo Clinic's QuantCRC AI model

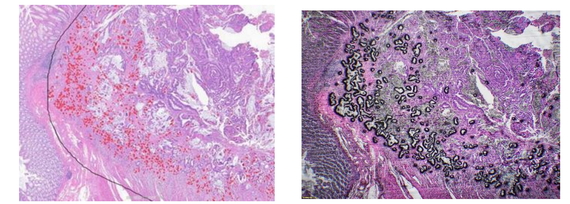
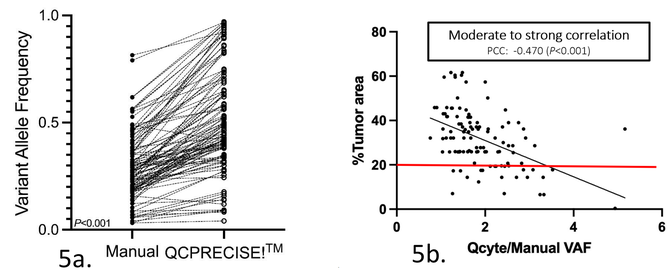


Figure 5: QCPRECISE VAFs compared to MMD in this study. See results for a discussion of the data.



Conclusions

QCPRECISE!™ has been shown to effectively interface with a front-end AI-enabled computational pathology solution and enabled a seamlessly integrated spatially targeted microdissection – thus providing highly specific data from sequencing at high throughput. While this study focused on the microdissection on H&E section, the platform is capable of microdissection from unstained sequential sections, where the image processing software from QuantumCyte transfers the annotations from the reference H&E taking into account tissue distortions.

References

1. Pai RK, Banerjee I, Shivji S, et al. Quantitative Pathologic Analysis of Digitized Images of Colorectal Carcinoma Improves Prediction of Recurrence-Free Survival. Gastroenterology. 2022;163(6):1531-1546 (QR Code).