Efficient Recovery of Cell-Free DNA from Synthetic Cerebrospinal Fluid Enables Sensitive Molecular Profiling in Brain Cancer

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nRichDX POWERING LIQUID BIOPSY

INTRODUCTION

Cerebrospinal fluid (CSF) is an increasingly important biofluid for liquid biopsy for brain tumors, offering improved sensitivity over plasma due to proximity to the tumor and reduced peripheral DNA background. However, cfDNA concentrations in CSF are low, and extraction inefficiencies can limit the reliability of downstream molecular assays. This study evaluates cfDNA recovery performance from CSF-like matrices to support sensitive molecular profiling relevant to brain cancer applications.

MATERIALS & METHODS

Synthetic CSF samples spiked with fragmented cfDNA reference material were processed using the nRichDX Revolution Max 20 Kit workflow. Low-input volumes representative of clinical CSF collections were evaluated. cfDNA yield and recovery were quantified using fluorometric and quantitative PCR-based measurements, and fragment size distributions were assessed to confirm recovery of short cfDNA fragments typical of CSF-derived material.

RESULTS

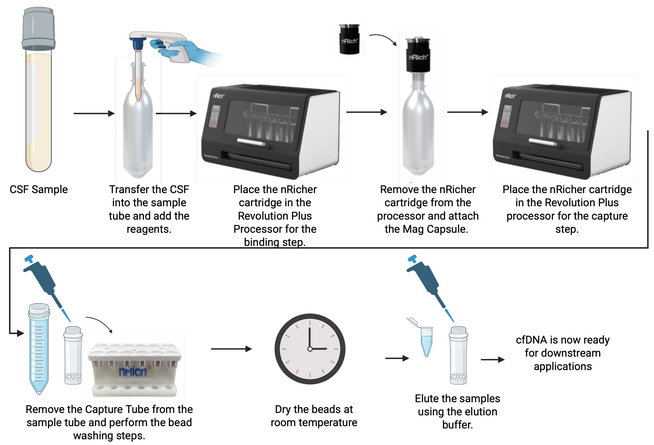

Figure 1. nRichDX Revolution cfDNA Max 20 Kit workflow.

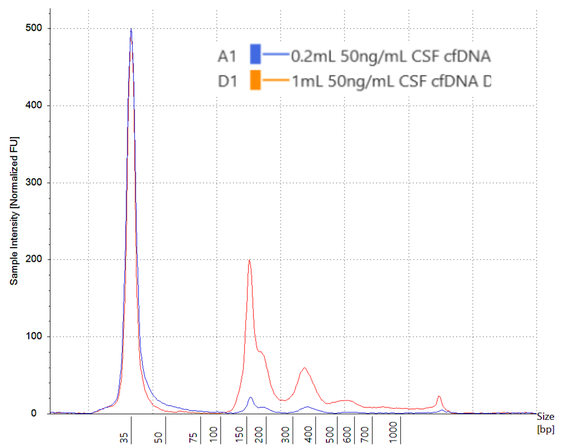

Figure 2. cfDNA recovery from synthetic CSF spiked with reference standard. Agilent cfDNA ScreenTape gel analysis confirmed successful recovery of cfDNA fragments within the 50–700 bp size range.

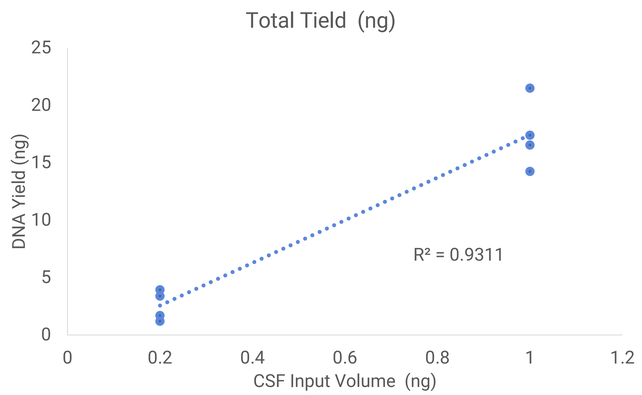

Figure 3. Total cfDNA yield (ng) as a function of CSF input volume. Qubit fluorometric quantification was performed on synthetic CSF samples processed at 0.2 mL and 1.0 mL input volumes following extraction with the nRichDX Revolution cfDNA Max20 Kit. The data showed strong linearity across the tested input volumes (**R^2 = 0.93**).

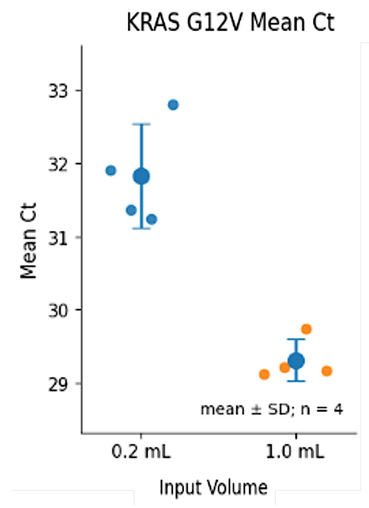

Figure 4. KRAS G12V mean Ct values for 0.2 mL and 1.0 mL synthetic CSF input volumes spiked with 50 ng/mL cfDNA reference standard. Large points represent mean Ct, error bars represent standard deviation, and smaller points represent individual replicates (**n = 4**). The 1.0 mL input produced a lower mean Ct than the 0.2 mL input, indicating improved target detection at the higher sample volume.

CONCLUSION

Efficient and reproducible cfDNA extraction from CSF-like matrices enables molecular profiling approaches that are critical for brain cancer research and clinical translation. The performance demonstrated in this study supports clinical applications including detection of tumor-derived genetic alterations, longitudinal disease monitoring, and assessment of treatment response in central nervous system malignancies. Reliable cfDNA recovery from limited CSF volumes is a key enabling step for integrating CSF-based liquid biopsy into precision oncology workflows for brain cancer.

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