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Development and evaluation a novel *in situ* target-capture approach for aptamer selection of human noroviruses ACCEPTED MANUSCRIPT

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Abstract

Human noroviruses (HuNoVs) is the primary non-bacterial pathogen causing acute gastroenteritis worldwide. Molecular approaches have been mainly used for detection of HuNoVs. Aptamer-based assay has been also applied for detection of HuNoVs through affinity binding of viral capsid. In a conventional systematic evolution of ligands by exponential enrichment process, the target protein-bound sequences in the library were recovered by complicated process including affinity chromatography, extraction, membrane-filtration or antibody-conjugated magnetic beads. In this study, a novel approach was applied to select aptamers for HuNoVs. The new approach incorporated an in situ capture assay and next generation sequencing (NSG) for selecting the aptamers. P particles of HuNoV (GII.4) were purified and coated on the module to capture sequences that were specifically bound with the protein. The unbound sequences were easily removed by washing. The sequences with high affinity were amplified just in the wells and selected by repeated in situ selection process. From the total of 30,622,226 tested sequences, two aptamers, APTL-1 and APTL-6, were finally selected to incorporate with in situ capture RT-qPCR assay for detection of HuNoVs from clinical samples. The sensitivity of these two aptamers was compared with porcine gastric mucin (PGM) that contains well-known viral receptors, and the reported aptamer APT-M6-2. Both GI and GII HuNoVs could be detected from 5 clinical samples tested. The selected aptamer APTL-1 was comparable to PGM and slightly superior to the reported APTM6-2 aptamer for detection of HuNoVs from clinical samples. The results demonstrated that this in situ target-capture approach for aptamer selection is practicable.

Graphical abstract

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