High-Throughput Norovirus Gene Detection
Detection of Norovirus RT-PCR Amplification Products with "Norovirus Amplification Kit"

Pretreatment and detection operations are simplified by using the Norovirus amplification kit in conjunction with the MCE-202 MultiNA to achieve accurate, labor-saving and high-throughput Norovirus gene detection.

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Introduction

Norovirus is a genus of viruses considered to be the principal cause of acute gastroenteritis, and detection of these viruses is generally conducted using gene amplification techniques such as RT-PCR assays. However, since biological samples contain many enzymes (RNases), which powerfully catalyze RNA hydrolysis and interfere with the RT and PCR reactions, it is necessary to first separate the virus and extract and purify its RNA prior to conducting Norovirus RNA gene amplification. Additionally, further operations including agarose electrophoresis are required for the detection, adversely affecting the time required for gene amplification. Here we introduce an example of detection of RT-PCR amplification products obtained using the Norovirus amplification kit (Direct RT-PCR Kit for Norovirus G1/G2 RNA Detection), which does not require RNA purification, in conjunction with the MCE-202 MultiNA DNA/RNA analyzer.

Results

Figure 1 shows the analysis results of the Norovirus G1 and G2 amplification kits using the MCE-202 MultiNA. The amplification products originating from the 86 bp (G1) and 142 bp (internal control, IC), and 98 bp (G2) and 205 bp (IC) were detected in the Norovirus G1 and G2 positive samples, respectively.

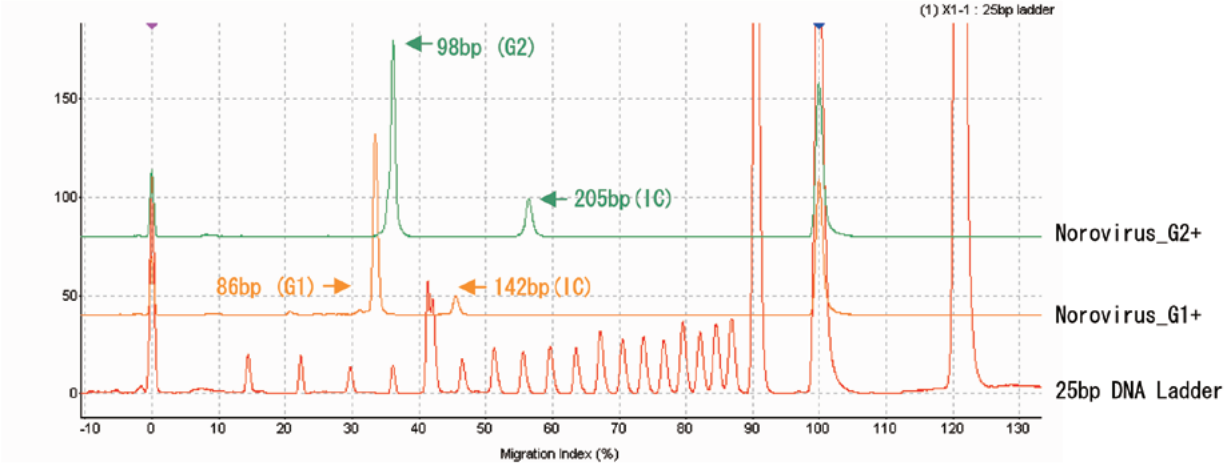

Fig. 1 Electropherogram of Samples Amplified with the Norovirus G1/G2 Amplification Kit

Analytical Conditions and Procedure

Instrument: MCE-202 MultiNA
Analysis mode: DNA-500 on-chip mixing
Samples:

G1+: Norovirus G1 positive sample processed with G1 amplification kit
G2+: Norovirus G2 positive sample processed with G2 amplification kit

Reagents:

- Direct RT-PCR Kit for Norovirus G1 RNA Detection (Shimadzu) P/N 241-08905-91
- Direct RT-PCR Kit for Norovirus G2 RNA Detection (Shimadzu) P/N 241-08905-92
- DNA-500 Reagent Kit for MultiNA (Shimadzu) P/N 292-27910-91
- SYBR® Gold nucleic acid gel stain (Invitrogen) S-11494
- 25 bp DNA Ladder (Invitrogen) 10597-011

(Note) For detailed information related to the Norovirus G1 and G2 amplification kits, refer to the instruction manuals provided in the kits.

Experimental Method:

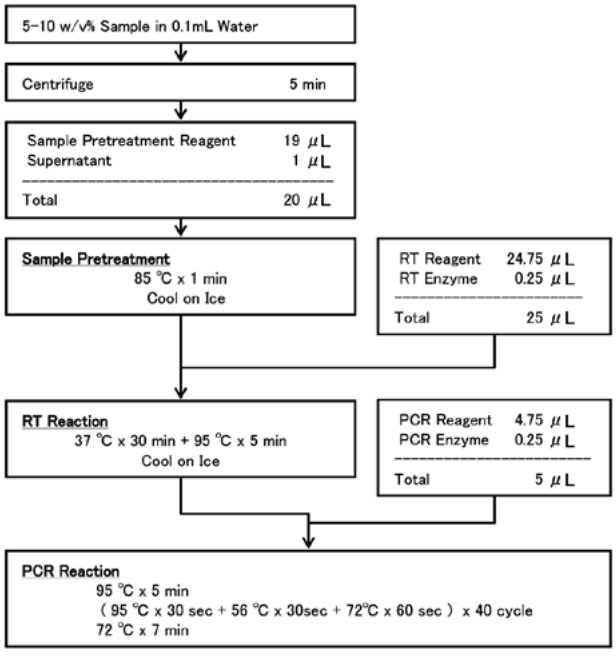

Fig. 2 Sample Preparation

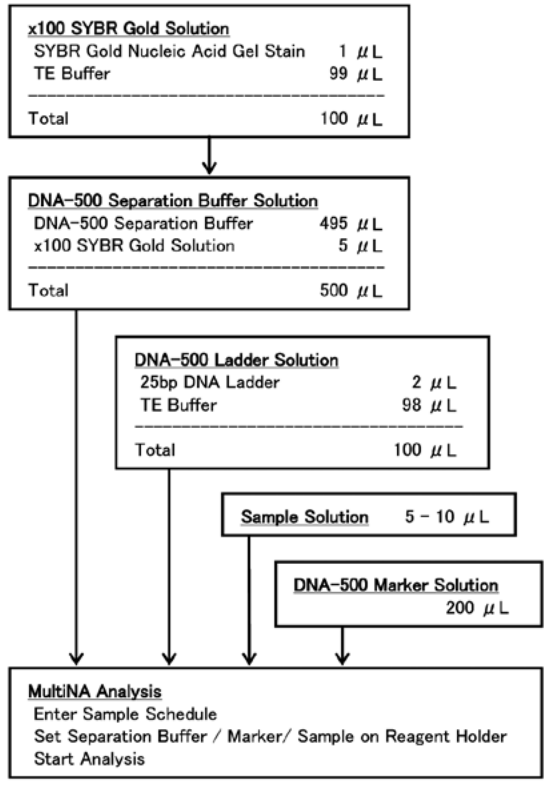

Fig. 3 Experimental Procedure (for 8 Samples)

(Note) For detailed operational information related to analysis using the MCE-202 MultiNA, please refer to the MCE-202 MultiNA Instruction Manual.

Reference Data

Norovirus Detection by Agarose Gel Electrophoresis

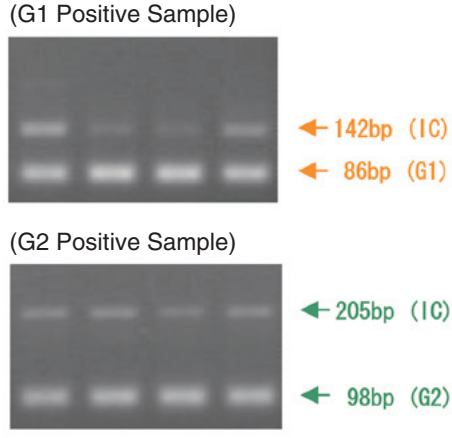

Fig. 4 Norovirus Detection by Norovirus G1/G2 Amplification Kit-Agarose Gel Electrophoresis

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