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E gene amplification V.1

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Protocol status: Working
We use this protocol and it's working

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1 The dengue virus strains obtained from cell culture were extracted by TGuide S32 magnetic bead method DNA/RNA extraction kit of Tiangen Biochemical Technology (Beijing) Co., Ltd. and amplified by one step RNA PCR Kit (Code No: DRR057A) reagent of Bao Bioengineering (Dalian) Co., Ltd.

2 The full sequence of E gene amplified by primer sequence was D1 (E) F: CAA GAA CCG AAA CA / GT GGA TGT C; D1 (E) R: GGC TGA TCG AAT TCC ACA CAC, and the length of amplified fragment was 1849 bp.

3 Reaction conditions: reverse transcription at 50 °C 30 min, reverse transcription at 94°C 2 min, reverse transcription at 94°C 30 s, reverse transcription at 52°C 30 s, reverse transcription at 72°C 2 min, 40 cycles and extension at 72°C. The amplified product was 5 micron L, and 1.5% agarose gel electrophoresis was used to confirm the reaction product according to Marker position.