

Table 1. Target sequences in the DEN-2 DNA used to PCR amplify viral genes

Protein	Target 5' sequence	Target 3' sequence
C	ATGAATAACCAACGGAAAAA	GATTCCAACAGTGATGGCG
PrM	TTGAATAGGAGACGCAGATC	CTGTCACTCCTTCAATGACA
E	ACACATTTCCAAAGAGCCCTG	TGGGAGTCATGGTGCAGGCC
NS1	TCACGCAGCACCTCACTGTC	TCACTCCTTGGTCACAGCT
NS2A	GGACATGGGCAGGTCGACAAC	CAAGAACCAGCAAGAAAAGG
NS2B	AGCTGGCCATTAAATGAGGC	GGGAAGTGAAGAAAACAACGG
NS3	CCGGAGTATTGTGGGATGTTCC	GAATTTGCAGCCGGAAGAAAG
NS4A	CTCTGACCCTGAACCTAAT	TCCAGAACCTGAAAAACAGAGA
NS4B	ACACCCCAAGACAACCAACTGA	ACACAACCAACACAAGAAGG
NS5	GGAACTGGCAACATAGGAGAG	GAAGAAGCAGGAGTTCTGTGG

Sets of two primers were designed for the shown target 5' and 3' DNA sequences on the infectious clone PD2/IC-30P-A (DEN-2 16681, Thailand, 1996). PCR fragments were cloned with appropriate restriction sites in pCAGGS [3' hemagglutinin (HA)]. Start codons were inserted at the 5' ends of each gene, and 3' ends were fused in frame to the HA tag sequence encoded in the plasmid and ending on stop codons. Each cloned DEN-2 gene was sequenced to ensure accurate cloning.