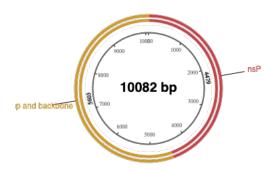
Component Fragments

Name	Length	Produced by 5' End 3' I		3' End
nsP	4479	PCR	Fwd Primer (custom)	Rev Primer (custom)
sp and backbone	5643	PCR	Fwd Primer (custom)	Rev Primer (custom)



Notes

• Everything looks OK. No major issues detected.

Required oligos

Name	Primer 5' (overlap/spacer/ANNEAL) 3'	Len	%GC	3' %GC	3' Tm	3' Ta
nsP_fwd	TGTGTGTCAGAATAGTGAATG	21	38	38	59.2	60.2
nsP_rev	GTTAGCTCACTCATTAGGC	19	47	47	59.6	60.2
sp and backbone_fwd	TGCCTAATGAGTGAGCTAAC	20	45	45	61.3	60.4
sp and backbone_rev	ATTCACTATTCTGACACACAAG	22	36	36	59.4	60.4

Build Settings

Property	Value
Product/Kit	#E5520 NEBuilder HiFi DNA Assembly Cloning Kit
Minimum Overlap	20 nt
Minimum Overlap Tm	48 °C
Circularize	Yes
PCR Polymerase/Kit	Q5 High-Fidelity DNA Polymerase
PCR Primer Conc.	500 nM
Min. Primer Length	18 nt

Assembled Sequence

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                                                          SYN 13-DEC-2021
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#ACCESSION
#VERSION
#KEYWORDS
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#SOURCE
             synthetic DNA construct
# ORGANISM
            synthetic DNA construct
#REFERENCE
             1 (bases 1 to 10082)
  AUTHORS
  TITLE
             NEBuilder-generated Construct
# JOURNAL
             Exported 13-DEC-2021 from NEBuilder https://nebuilder.neb.com
#COMMENT
             NEBuilder-generated oligos (UPPERCASE = gene-specific, lowercase = overlap)
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#COMMENT
             nsP_fwd 3'Tm: 59.2 3'Ta: 60.2
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             nsP_rev 3'Tm: 59.6 3'Ta: 60.2
#COMMENT
             sp and backbone_fwd: TGCCTAATGAGTGAGCTAAC
#COMMENT
             sp and backbone_fwd 3'Tm: 61.3 3'Ta: 60.4
#COMMENT
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