SARS-CoV Envelope (E) Protein, Recombinant from *Escherichia coli*

Catalog No. NR-4284

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Product Description:

The major role of the SARS-CoV[1,2] envelope (E) protein is to participate in the formation of viral particles[3], although it is known that some coronaviruses also play a part in replication[4,5] and apoptosis[6]. Co-expression of the E protein and membrane protein (M) is sufficient for release of virus-like particles[7] and interactions between the E and M proteins and nucleocapsids [viral RNA and the nucleocapsid (N) protein] result in budding through the membrane[8].

The SARS-CoV E protein is hydrophobic, with one predicted transmembrane helix (amino acid residues 12 to 34). The first 11 amino acids of the N-terminus do not have any specific structural elements and are expected to be in the virion. The hydrophilic C-terminus is exposed to the cytoplasmic side of the membrane and folds into 2 β-sheets. β-sheet 1 is expected to interact with the surface of the lipid bilayer through hydrogen bonding[3].

The SARS-CoV E–SUMO fusion protein containing an N-terminal histidine tag was expressed in *Escherichia coli* and purified by nickel affinity, ion exchange and size exclusion chromatography[9-12]. After the fusion was cleaved by the SUMO protease, the SUMO tag and the SUMO protease (both histidine-tagged) were subtracted from the SARS-CoV E protein by nickel affinity chromatography. The SARS-CoV E protein was dialyzed against 10 mM ammonium bicarbonate and aliquoted into 10 mM sodium carbonate and 10 mM sodium phosphate, vacuum dried and stored at -20°C. The predicted sequence, protein properties and amino acid content of SARS-CoV E protein are shown in Tables 1–3 below.

Material Provided:

Each vial contains approximately 0.1 mg of E protein (determined by Bradford assay with BSA as a standard), 20 µg sodium carbonate and 31 µg sodium phosphate.

Packaging and Storage:

NR-4284 was packaged aseptically in cryovials. The product is shipped frozen on dry ice. Lyophilized NR-4284 is stable for several weeks at 4°C but should be kept at -20°C or colder for long-term storage.

Reconstitution and Storage:

Reconstitution of a vial of NR-4284 in 1 mL of buffer containing 10 mM sodium carbonate (pH 11) is recommended to keep the protein soluble. The addition of 0.3% lauryl sarcosine is recommended to enhance protein stability. For analysis by SDS-PAGE, the sample buffer (pH 11) should contain 5 M urea, 1 M thiourea, 1 mM DTT, 0.5% CHAPS, 100 mM sodium carbonate and 25 mM Tris-HCl.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through the NIH Biodefense and Emerging Infections Research Resources Repository, NIAID, NIH: SARS-CoV Envelope (E) Protein, Recombinant from *Escherichia coli*, NR-4284."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories. 4th ed. Washington, DC: U.S. Government Printing Office, 1999. HHS Publication No. (CDC) 93-8395. This text is available online at www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4toc.htm.

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Table 1 – Predicted Protein Sequence					
1	MYSFVSEETG	TLIVNSVLLF	LAFVVFLLVT	LAILTALRLC	AYCCNIVNVS
51	LVKPTVYVYS	RVKNLNSSEG	VPDLLV		

Table 2 – Predicted Protein Properties	
Length	76 amino acids
Molecular weight	8361 daltons
1 microgram	119.6 pmoles
Molar extinction coefficient	5240
1 A[280]	1.56 mg/mL
A[280] of 1 mg/mL	0.64 AU
Isoelectric point	6.01

Table 3 – Predicted Amino Acid Content			
Amino Acids		**Count**	**% by Frequency**
Charged (RKHYCDE)		15	19.74
Acidic (DE)		4	5.26
Basic (KR)		4	5.26
Polar (NCQSTY)		24	31.58
Hydrophobic (AILFWV)		39	51.32
A	Ala	4	5.30
C	Cys	3	3.90
D	Asp	1	1.30
E	Glu	3	3.90
F	Phe	4	5.30
G	Gly	2	2.60
H	His	0	0.00
I	Ile	3	3.90
K	Lys	2	2.60
L	Leu	14	18.40
M	Met	1	1.30
N	Asn	5	6.60
P	Pro	2	2.60
Q	Gln	0	0.00
R	Arg	2	2.60
S	Ser	7	9.20
T	Thr	5	6.60
V	Val	14	18.40
W	Trp	0	0.00
Y	Tyr	4	5.30

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