

### **SARS-CoV Membrane (M) Protein with C-terminal Histidine Tag, Recombinant from *E. coli***

#### **Catalog No. NR-878**

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#### **Contributor:**

NIH Biodefense and Emerging Infections Research Resources Repository, NIAID, NIH.

#### **Product Description:**

The SARS-CoV<sup>1,2</sup> membrane (M) glycoprotein is the most abundant structural protein of the virus and plays a key role in virion morphogenesis and release.<sup>1</sup> During virion assembly, it is localized at specialized intracellular membrane structures. Interactions between the M protein, the envelope (E) protein and nucleocapsids [viral RNA and the nucleocapsid (N) protein] result in budding through the membrane. The spike (S) protein is incorporated into the viral envelope by interaction with the M protein and mature virions are released from smooth vesicles.

The M protein is highly hydrophobic with three membrane spanning domains in the amino-terminal half of the protein.<sup>1</sup> A highly conserved amino acid sequence, SMWSFNPE, immediately follows the third hydrophobic domain. The M protein has an NGT near its amino terminus, suggesting that it is N-glycosylated at position 4.

The SARS-CoV M protein containing a C-terminal histidine tag (NR-878) was expressed in *E. coli*<sup>3-6</sup> and purified by nickel affinity, cation exchange and size exclusion chromatography. NR-878 was extensively dialyzed against 10 mM ammonium bicarbonate and 0.3% lauryl sarcosine, aliquoted and lyophilized. NR-878 has a molecular weight of approximately 26,700 daltons. The predicted sequence, protein properties and amino acid content of SARS-CoV M protein are shown in Tables 1–3 below. Non-SARS-CoV residues are underlined in Table 1.

#### **Material Provided:**

Each vial contains approximately 1.0 mg of M protein (determined by Bradford assay) and 3 mg of lauryl sarcosine lyophilized from 1 mL of 10 mM ammonium bicarbonate and 0.3% lauryl sarcosine.

#### **Packaging and Storage:**

NR-878 was packaged aseptically in cryovials. The product is provided on dry ice and should be placed at -20°C or colder for long-term storage. Lyophilized NR-878 is stable for several weeks at 4°C.

#### **Reconstitution and Storage:**

Reconstitution of a vial of NR-878 in 1 mL of desired sample buffer will result in 1 mg/mL of M protein and 0.3% lauryl

sarcosine. The presence of 0.3% lauryl sarcosine is recommended to enhance stability and recovery. Reconstituted NR-878 should be stored at -20°C or colder.

#### **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 Membrane (M) Protein with C-terminal Histidine Tag, Recombinant from *E. coli*, NR-878.”

#### **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/bmb14/bmb14toc.htm](http://www.cdc.gov/od/ohs/biosfty/bmb14/bmb14toc.htm).

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### References:

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**Table 1 – Predicted Protein Sequence<sup>a</sup>**

1	MADNGTITVE	ELKQLLEQWN	LVIGFLFLAW	IMLLQFAYSN	RNRFLYIIKL
51	VFLWLLWPVT	LACFVLAAYV	RINWVTGGIA	IAMACIVGLM	WLSYFVASFR
101	LFARTRSMWS	FNPETNILLN	VPLRGTIVTR	PLMESELVIG	AVIIRGHLM
151	AGHSLGRCDI	KDLPKEITVA	TSRTLSYYKL	GASQRVGTDS	GFAAYNRYRI
201	GNYLNTDHA	GSNDNIALLV	QKLAAALEHH	HHHH	

<sup>a</sup>Non-SARS-CoV residues are underlined.

**Table 2 – Predicted Protein Properties**

Length	235 amino acids
Molecular weight	26707 daltons
1 microgram	39.73 pmoles
Molar extinction coefficient	51710
1 A[280]	0.49 mg/mL
A[280] of 1 mg/mL	2.05 AU
Isoelectric point	9.63
Charge at pH 7	8.00

**Table 3 – Predicted Amino Acid Content**

Amino Acids	Count	% by Weight	% by Frequency
Charged (RKHYCDE)	49	26.68	22.07
Acidic (DE)	13	6.27	5.86
Basic (KR)	21	11.97	9.46
Polar (NCQSTY)	55	24.87	24.77
Hydrophobic (AILFWV)	102	45.42	45.95
A Ala	19	5.81	8.56
C Cys	3	1.25	1.35
D Asp	6	2.74	2.70
E Glu	7	3.53	3.15
F Phe	11	6.23	4.95
G Gly	15	3.86	6.76
H His	3	1.60	1.35
I Ile	18	8.10	8.11
K Lys	6	3.01	2.70
L Leu	31	13.95	13.96
M Met	7	3.58	3.15
N Asn	13	5.89	5.86
P Pro	5	1.97	2.25
Q Gln	5	2.51	2.25
R Arg	15	8.96	6.76
S Ser	12	4.33	5.41
T Thr	13	5.31	5.86
V Val	16	6.43	7.21
W Trp	7	4.90	3.15
Y Tyr	9	5.59	4.05
B Asx	19	8.63	8.56
Z Glx	12	6.04	5.41
X Xxx	1	0.44	0.45