

**SARS-CoV Nucleocapsid (N) Protein, Recombinant from *Escherichia coli***

**Catalog No. NR-48761**

This reagent is the tangible property of the U.S. Government.

**For research use only. Not for human use.**

**Contributor:**

BEI Resources

**Manufacturer:**

LifeSensors, Inc., Malvern, Pennsylvania, USA

**Product Description:**

The SARS-CoV nucleocapsid (N) protein is a phosphoprotein that binds the viral RNA inside the virion. The N protein bears a high similarity to the nucleocapsid proteins of other coronaviruses. However, it contains a short unique lysine-rich region (KTFPPTPEPKDKKKKTDEAQ) that is not found in any other viruses. The function of this region is not known; however, it is speculated that it may be involved in pathogenesis.<sup>1</sup> The highly basic nature of this peptide is characteristic of an RNA-binding protein. The N protein is known to have B and T cell epitopes and to elicit host protective immune responses.

NR-48761 was expressed and purified using a SUMO fusion system.<sup>2-4</sup> An N-terminal histidine-tagged SUMO-nucleocapsid fusion was expressed in *Escherichia coli* and purified by nickel affinity chromatography. After the fusion was cleaved by the SUMO protease, the SUMO tag and the SUMO protease (both histidine-tagged) were subtracted from the nucleocapsid by nickel affinity chromatography. The nucleocapsid was further purified by cation exchange chromatography, dialyzed against 10 mM ammonium bicarbonate, aliquoted and lyophilized. NR-48761 has a molecular weight of approximately 46,000 daltons. The predicted sequence, protein properties and amino acid content of SARS-CoV nucleocapsid are shown in Tables 1-3 below.

**Material Provided:**

Each vial contains approximately 100 µg of NR-48761 lyophilized in 10 mM ammonium bicarbonate.

**Packaging/Storage:**

NR-48761 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-48761 is stable for several weeks at 4°C.

**Functional Activity:**

In western blot assays, NR-48761 reacts with rabbit

polyclonal sera prepared against the SUMO-SARS-CoV-N protein, but not with rabbit polyclonal sera prepared against a SUMO-3CL protease.<sup>2</sup>

**Citation:**

Acknowledgment for publications should read “The following reagent was obtained through BEI Resources, NIAID, NIH: SARS-CoV Nucleocapsid (N) Protein, Recombinant from *Escherichia coli*, NR-48761.”

**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. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see [www.cdc.gov/biosafety/publications/bmbl5/index.htm](http://www.cdc.gov/biosafety/publications/bmbl5/index.htm).

**Disclaimers:**

You are authorized to use this product for research use only. It is not intended for human use.

Use of this product is subject to the terms and conditions of the BEI Resources Material Transfer Agreement (MTA). The MTA is available on our Web site at [www.beiresources.org](http://www.beiresources.org).

While BEI Resources uses reasonable efforts to include accurate and up-to-date information on this product sheet, neither ATCC® nor the U.S. Government makes any warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. Neither ATCC® nor the U.S. Government warrants that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, use and disposal. ATCC® and the U.S. Government are not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to ensure authenticity and reliability of materials on deposit, the U.S. Government, ATCC®, their suppliers and contributors to BEI Resources are not liable for damages arising from the misidentification or misrepresentation of products.

**Use Restrictions:**

**This material is distributed for internal research, non-commercial purposes only.** This material, its product or its derivatives may not be distributed to third parties. Except as performed under a U.S. Government contract, individuals contemplating commercial use of the material, its products or its derivatives must contact the contributor to determine if a license is required. U.S. Government contractors may need a license before first commercial sale.

**References:**

1. Marra, M. A., et al. "The Genome Sequence of the SARS-Associated Coronavirus." *Science* 300 (2003): 1399–1404. PubMed: 12730501.
2. Zuo, X., et al. "Expression and Purification of SARS Coronavirus Proteins using SUMO-fusions." *Protein Expr. Purif.* 42 (2005): 100–110. PubMed: 15939295.
3. Butt, T. R., S. C. Edavettal, J. P. Hall, and M. R. Mattern. "SUMO Fusion Technology for Difficult-to-express Proteins." *Protein Expr. Purif.* 43 (2005): 1–9. PubMed: 16084395.
4. Malakhov, M. P., et al. "SUMO Fusions and SUMO-specific Protease for Efficient Expression and Purification of Proteins." *J. Struct. Funct. Genomics* 5 (2004): 75–86. PubMed: 15263846.

ATCC® is a trademark of the American Type Culture Collection.



**Table 1 – Predicted Protein Sequence**

1	MSDNGPQSNQ	RSAPRITFGG	PTDSTDNNQN	GGRNGARPKQ	RRPQGLPNNT
51	ASWFTALTQH	GKEELRFPRG	QGVPINTNSG	PDDQIGYYRR	ATRRVRGGDG
101	KMKELSPRWY	FYYLGTGPEA	SLPYGANKEG	IVWVATEGAL	NTPKDHIGTR
151	NPNNNAATVL	QLPQGTTLPK	GFYAEGSRGG	SQASSRSSSR	SRGNSRNSTP
201	GSSRGNSPAR	MASGGGETAL	ALLLLDRLNQ	LESKVSGKGQ	QQQGQTVTKK
251	SAAEASKKPR	QKRTATKQYN	VTQAFGRRGP	EQTQGNFGDQ	DLIRQGTDYK
301	HWPQIAQFAP	SASAFFGMSR	IGMEVTPSGT	WLTYHGAIKL	DDKDPQFKDN
351	VILLNKHIDA	YKTFPTEPK	KDKKKKTDEA	QPLPQRQKKQ	PTVTLLPAAD
401	MDDFSRQLQN	SMSGASADST	QA		

**Table 2 – Predicted Protein Properties**

Length	422 amino acids
Molecular weight	46022 daltons
1 microgram	21.7 pmoles
Molar extinction coefficient	42530
1 A[280]	1.08 mg/mL
A[280] of 1 mg/mL	0.92 AU
Isoelectric point	10.11
Charge at pH 7	24.22

Table 3 – Predicted Amino Acid Content			
Amino Acids	Count	% by Weight	% by Frequency
Charged (RKHYCDE)	112	32.45	26.54
Acidic (DE)	36	9.30	8.53
Basic (KR)	60	17.98	14.22
Polar (NCQSTY)	138	33.34	32.70
Hydrophobic (AILFWV)	100	23.02	23.70
A Ala	34	5.65	8.06
C Cys	0	0.00	0.00
D Asp	22	5.46	5.21
E Glu	14	3.84	3.32
F Phe	13	4.01	3.08
G Gly	45	6.30	10.66
H His	5	1.45	1.18
I Ile	11	2.69	2.61
K Lys	29	7.91	6.87
L Leu	26	6.36	6.16
M Met	7	1.95	1.66
N Asn	25	6.16	5.92
P Pro	31	6.66	7.35
Q Gln	34	9.27	8.06
R Arg	31	10.07	7.35
S Ser	35	6.86	8.29
T Thr	33	7.33	7.82
V Val	11	2.40	2.61
W Trp	5	1.90	1.18
Y Tyr	11	3.72	2.61
B Asx	47	11.62	11.14
Z Glx	48	13.11	11.37