# **Nucleocapsid-Fc**

# Soluble SARS-CoV-2 nucleocapsid protein fused to a human IgG1 Fc tag

Catalog code: fc-sars2-n

https://www.invivogen.com/sars2-nucleocapsid-proteins

For research use only, not for diagnostic or therapeutic use Version 20119-NJ

#### PRODUCT INFORMATION

#### Contents:

• 50 µg of lyophilized Nucleocapsid-Fc protein

1 5 ml endotoxin-free water

#### Protein construction:

Full-length Nucleocapsid [M1-A419] with a human IgG1 Fc tag in C-terminus

Accession sequence: PODTC9

Species: SARS-CoV-2 (2019-nCoV); Wuhan-Hu-1 (D614) isolate

Tag: C-terminal human IgG1 Fc

Total protein size: 669 a.a. (secreted form)

Molecular weight: ~79 kDa (SDS-PAGE)

**Purification:** Protein A affinity chromatography

Purity: >95% (SDS PAGE)

#### Formulation:

0.2 µm filtered solution in a sodium phosphate buffer with glycine, saccharose, and stabilizing agents

#### Storage:

- Product is shipped at room temperature. Store lyophilized product at -20 °C. Lyophilized product is stable for at least 1 year.
- Reconstituted protein is stable for 1 month when stored at  $4^{\circ}$ C and for 1 year when aliquoted and stored at -20 °C. Avoid repeated freezethaw cycles.

#### Quality control:

- The size and purity of the protein has been confirmed by SDS-PAGE.
- Nucleocapsid-Fc has been functionally validated by ELISA using an Anti-SARS Nucleocapsid antibody.
- Absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and TLR4 cellular assays.

#### **BACKGROUND**

The SARS-CoV-2 Nucleocapsid (N) is an important structural protein that plays important roles in the viral life cycle including replication, transcription, and genome packaging<sup>1</sup>. The SARS-CoV-2 N features two important NTD and CTD functional domains in N-terminal and C-terminal, respectively<sup>1-6</sup>. NTD interacts with both the RNA genome and M proteins to form virion particles. The N protein interaction with the RNA forms the virus ribonucleoprotein core which is packed as a helical "beads-on-a-string" conformation. CTD allows RNA synthesis through binding of the replication-transcription complexes (RTCs), oligomerization of multiple N proteins through its dimerization domain, and genome incorporation into the new virion. N is a major immunogen of SARS-CoV-2. Indeed, elevated Anti-SARS-CoV-2 N IgG and IgM antibody titers have been reported in COVID-19 patients' sera<sup>7-9</sup>. These observations make SARS-CoV-2 N an attractive tool for early diagnosis<sup>7-9</sup> and a potential therapeutic drug-target<sup>3</sup>.

# PRODUCT DESCRIPTION

Nucleocapsid-Fc is a soluble protein generated by fusing the full-length SARS-CoV-2 nucleocapsid [M1-A419] to a C-terminal human IgG1 Fc tag with a TEV (Tobacco Etch Virus) sequence linker. This fusion protein has a molecular weight of ~79 kDa on a SDS PAGE gel. Nucleocapsid-Fc has been generated by recombinant DNA technology, produced in HEK293 cells, and purified by protein G affinity chromatography.

#### **APPLICATIONS**

- Vaccination studies: using combinations of Nucleocapsid protein antigens and adjuvants.
- Antibody screening: finding anti-Nucleocaspid antibodies in COVID-19 patients' sera.
- Inhibitor screening: finding small molecules able to block the SARS-CoV-2 nucleocapsid interaction with replication-transcrption complexes (RTCs).

#### **METHODS**

#### Nucleocapsid-Fc resuspension (100 µg/ml)

Note: Ensure you see the lyophilized pellet before resuspension.

- Add 500  $\mu l$  of endotoxin-free water to the vial and gently pipette until completely resuspended.
- Prepare aliquots and store at -20 °C or 4°C.



# PROTEIN SEQUENCE

M E I K V L F A L I C Î A V A E A K P T E L E M S D N G P Q N Q R N A P R I T F G G P S D S T G S N Q N G E R S G A R S K Q R R P Q G L P N N T A S W F T A L T Q H G K E D L K F P R G Q G V P I N T N S S P D D Q I G Y Y R R A T R R I R G G D G K M K D L S P R W Y F Y Y L G TGPEAGLPYGANKDGIIWVATEGALNTPKDHIGT RNPANNAAIVLQLPQGTTLPKGFYAEGSRGGSQA S S R S S S R S R N S S R N S T P G S S R G T S P A R M A G N G G D A A L A L L L D R L N Q L E S K M S G K G Q Q Q G Q T V T K K S AAEASKKPRQKRTATKAYNVTQAFGRRGPEQTQG N F G D Q E L I R Q G T D Y K H W P Q I A Q F A P S A S A F F G M S RIGMEVTPSGTWLTYTGAIKLDDKDPNFKDQVILL NKHIDAYKTFPPTEPKKDKKKADETQALPQRQK KQQTVTLLPAADLDDFSKQLQQSMSSADSTQART ENLYFQGSGSEPKSSDKTHTCPPCPAPEAEGGPSV FLFPPKPKDQLMISRTPEVTCVVVDVSHEDPEVKF N W Y V D G V E V H N A K T K P R E E Q Y N S T Y R V V S V L T V L H Q D W L N G K E Y K C K V S N K A L P A S I E K T I S K A K G Q P REPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIA VEWESNG QPENNYKTTPPVLDSDGSFFLYSKLTVD K S R W Q Q G N V F S C S V L H E A L H N H Y T Q K S L S L S P G K

Green: signal sequence

Purple: stabilizing amino acid sequence

Blue: Nucleocapsid sequence Black: TEV cleavage sequence Red: human IgG1 Fc sequence

#### REFERENCES

1. Mu, J. et al., 2020. SARS-CoV-2-encoded nucleocapsid protein acts as a viral suppressor of RNA interference in cells. Sci China Life Sci 63, 1-4, 2. Chang C. et al., 2006. Modular organization of SARS coronavirus nucleocapsid protein. J. Biom. Sci. 13:59-72. 3. Krokhin O. et al., 2003. Mass spectrometric characterization of proteins from the SARS virus. Mol. & Cell. Prot. 2:346-356. 4. Cubuk, J. et al., 2020. The SARS-CoV-2 nucleocapsid protein is dynamic, disordered, and phase separates with RNA. bioRxiv. doi:10.1101/2020.06.17.158121. 5. Kang. S. et al., 2020. Crystal structure of SARS-CoV-2 nucleocapsid protein RNA binding domain reveals potential unique drug targeting sites. Acta Pharm Sin B. doi:10.1016/j. apsb.2020.04.09. 6. Khan, M.T. et al., 2020. SARS-CoV-2 nucleocapsid and Nsp3 binding: an in silico study. Arch Microbiol. doi: 10.1007/s00203-020-01998-6. 7. Liu, W. et al., 2020. Evaluation of Nucleocapsid and Spike Protein-Based Enzyme-Linked Immunosorbent Assays for Detecting Antibodies against SARS-CoV-2. J Clin Microbiol 58. 8. Guo L. et al., 2020. Profiling Early Humoral Response to Diagnose Novel Coronavirus Diseases (COVID-19). Clinical Infectious Diseases. 71(15):778-785. 9. To K. K-W. et al., 2020. Temporal profiles of viral load in posterior oropharyngeal saliva samples and serum antibody responses during infection by SARS-CoV-2: an observational cohort study. The Lancet Infectious Diseases. 20(5):565-574.

### **RELATED PRODUCTS**

Product	Catalog Code
Nucleocapsid-His	his-sars2-n
Spike-S1-Fc	fc-sars2-s1
Spike-S1-His	his-sars2-s1
Spike-RBD-Fc	fc-sars2-srbd
Spike-RBD-His	his-sars2-srbd

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