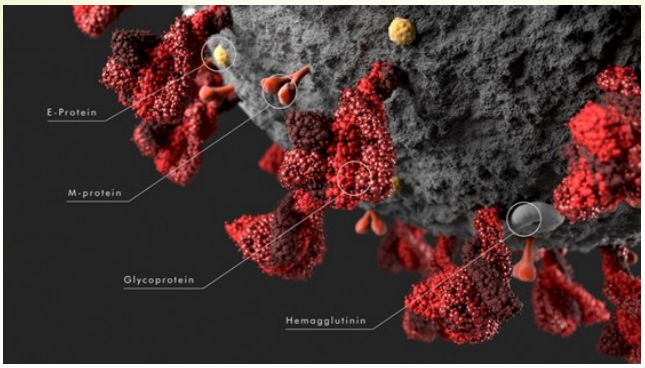
Complete Proteome of COVID-19



The genome of SARS-CoV-2 counts 29,811 nucleotides, encoding for 29 different proteins. The translation of the linear single-stranded RNA conducts to the generation of the following proteome:

- 4 are structural proteins, S, N, M, and E
- 16 proteins are non-structural proteins or NSP: the first 11 are encoded in ORF1a whereas the last 5 are encoded in ORF1b
- 9 are accessory proteins named ORF3a, ORF3b, ORF6, ORF7a, ORF7b, ORF8, ORF9b, ORF9c, and ORF10.

Figure 1. Genome architecture of SARS-CoV-2. SARS-CoV-2 contains a positive single-stranded mRNA as genetic material. The 5' capped mRNA has a leader sequence (LS), poly-A tail at 3' end, and 5' and 3' UTR. It contains the following genes: ORF1a, ORF1b, Spike (S), ORF3a, ORF3b, Envelope (E), Membrane (M), ORF6, ORF7a, ORF7b, ORF8, ORF9b, ORF14, Nucleocapsid (N), and ORF10. Dark Proteome of Newly Emerged SARS-CoV-2 in Comparison with Human and Bat Coronaviruses. Rajanish Giri, Taniya Bhardwaj, Meenakshi Shegane, Bhuvaneshwari R. Gehi, Prateek Kumar, Kundlik Gadhave.

1. Structural proteins of SARS-COV-2

- Spike (S). Trimeric transmembrane spike glycoprotein (1,273 a.a.) precursor is cleaved into S1 and S2 segments in order to bind to ACE2 receptors. A mutation in the SARS-CoV-2 S protein allows the human enzyme called furin to do the proteolytic cleavage into the sequence (PRRARS|V). Since bat CoV doesn't present this sequence, its apparition may be an explanation for the virus ability to infect humans (1).

- Nucleocapsid (N). Dimer (419 a.a.) that binds viral genome to membrane forming a shell and demonstrates non-specific nucleic acid binding capability (2). Although the overall structure is similar to other reported coronavirus nucleocapsids, the surface electrostatic potential characteristics are unique (3).

- Membrane (M). Matrix protein (222 a.a.) is the most abundant structural component of the virion and defines the shape of the envelope. M protein of SARS-CoV-2 has a triple helix bundle, forms a single 3-transmembrane domain (TM), and is homologous to the prokaryotic sugar transport protein semiSWEET. The advantage and role of sugar transporter-like structures in viruses are still unknown (4). M protein cooperates with Spike during the cell attachment and entry (5).

- Envelope (E). This small membrane protein (75 a.a.) is involved in viral assembly, budding, and pathogenesis. Envelope protein is identical to its counterparts from CoV MP798, CoVZXC21, CoVZC45 and RaTG13. However, a substitution at position 69, and a deletion in position 70 were found (5).

2. Non-structural proteins of SARS-COV-2

Non-Structural Proteins (nsp) are expressed as two long polyproteins (pp1a and pp1ab) then are cleaved into 16 mature smaller proteins by the papain-like protease (PLpro) and the 3-chymotrypsinlike protease (3CLpro, also known as the main protease-Mpro).

- Nsp1 (180 aa): 28 inserts or deletions occurs along its amino-acids sequence, compared to SARS-Cov. The role of nsp1 is believed to be very similar in SARS-Cov and SARS-Cov-2, suppressing type I IFN expression (6).

- Nsp2 (638 aa): nsp 2 interacts with a host protein complex of PHB1 and PHB2 involved in mitochondrial biogenesis (7). Position 321 of this methyltransferase sequence has a polar amino acid while Bat SARS-like coronavirus has a non-polar amino acid. It can be speculated, that due to this polarity, and potential to form H-bonds, the glutamine amino acid may confer higher stability to the protein (8).

- Nsp3 (1,945 aa): Nsp3 shuts down host enzymes called PARPs, which prevent viruses from replicating (9). The destabilizing mutation in nsp3 proteins could suggest a potential mechanism differentiating COVID-2019 from SARS (8).

- Nsp4 (500 aa): Nsp4 critically interacts with nsp3 to rearrange host cell membrane. Only their synergy is able to perform the job (10).

- Nsp5 (306 aa): the main protease of 306 aa cleaves at 11 sites. COVID-19 NSP5 is also highly homologous to SARS NSP5 (96% identity, 98% similarity) (11).

- Nsp6 (290 aa): It interacts with nsp3 and nsp4. Nsp6 presents 7 putative trans-membrane helices like in other coronaviruses. The presence of two mutations affecting the Non-Structural Protein 6 has been recently found (12).

- Nsp7 (83 aa) & Nsp 8 (198 aa): The SARS-coronavirus nsp7+nsp8 primase complex is capable of both de novo initiation and primer extension (13). The complex triggers RNA-synthesizing activity of nsp12 (14). NS7b and NS8, were exclusively conserved among 2019-nCoV, BetaCoV_RaTG, and BatSARS-like Cov. Functional changes in the NS7b and NS8 proteins during evolution may provide important information to explore the human infective property of 2019-nCoV (15).

- Nsp9 (113 aa): Current understanding suggests that Nsp9 is involved in viral genomic RNA reproduction. The structure of the SARS-CoV-2 Nsp9 revealed the high level of structural conservation within the Nsp9 family (16).

- Nsp10 (139 aa): forms a complex with NSP16 to cap viral mRNA transcripts for efficient translation and to evade immune surveillance (17).

- Nsp11 (13 aa): overlapping sequence with nsp10, nsp11 short peptide function is still unknown.

- Nsp12 (932 aa): RNA polymerase complex consisting of the viral RdRp (nsp12) and associated cofactors (nsp7 and nsp8). Alignment of nsp12 for the whole CoV family indicates that the SARS-CoV-2 nsp12 is almost identical to that of the SARS-CoV (96% identity, 98% similarity) (18).

- Nsp13 (601 aa): Like SARS and MERS-Nsp13, the overall structure of SARS-CoV-2 nsp13 adopted a triangular pyramid shape comprising five domains (19). Nsp13 is a helicase that unpacks viral genome material to make it more accessible.

- Nsp14 (527 aa): Nsp14 is the proofreading non-structural protein for normal CoV recombination. Thus, nsp14-ExoN is a key determinant of both high fidelity CoV replication and recombination, and thereby represents a highly-conserved and vulnerable target for virus inhibition and attenuation (20).

- Nsp15 (346 aa): nidoviral RNA uridylate-specific endoribonuclease (NendoU). While initially Nsp15 was thought to directly participate in viral replication, it was later shown that Nsp15-deficient coronaviruses were viable and replicating. More recently, it was proposed that NendoU activity of Nsp15 is responsible for the protein interference with the innate immune response, though other studies indicate that the process is independent of the endonuclease activity. There are also suggestions that Nsp15 de grades viral RNA to hide it from the host defences (21).

- Nsp16 (298 aa): forms a complex with NSP10 to cap viral mRNA transcripts for efficient translation and to evade immune surveillance (17).

3. Accessory proteins of SARS-COV-2

- ORF3a (275 aa): the 3a protein is unique to SARS-CoV and is essential for virulence, infectivity, ion channel formation, and virus release (22).

- ORF3b (22 aa): overlapping sequence with 3a, the 3b short peptide is a potent IFN-1 antagonist but allegations have yet to be confirmed. The ORF3b sequences of SARS-CoV-2 are considerably shorter than those of their SARS-CoV orthologs (153.2 ± 0.47 amino acids on average) (23).

- ORF6 (61 aa): IFN-1 antagonist. ORF6 has been shown to be an IFN antagonist that disrupts karyopherin transportation of transcriptions factors like STAT1 (24, 25).

- ORF7a (121 aa): SARS-CoV ORF7a directly binds to BST-2 and inhibits its activity by blocking the glycosylation of BST-2 (26).

- ORF7b (43 aa): overlapping sequence with 7a, ORF7b protein is not only an accessory protein but a structural component of the SARS-CoV virion (27). We haven't found any reference in SARS-CoV-2 so far.

- ORF8 (121 aa): ORF8 is the most different protein compared to SARS-Cov (30% homology). A 382-nt deletion covering the ORF8 of SARS-CoV-2 has been reported. The deletion also removes the ORF8 transcription-regulatory sequence (TRS), which in turn enhances the downstream transcription of the N gene. (28)

- ORF9b (97 aa): coded within the N gene, ORF9b interacts with a mitochondrial import receptor, Tom70, which acts as an essential adaptor linking MAVS to TBK1/IRF3, resulting in the activation of IRF-3 (29).

- ORF9c (XX aa): coded within the N gene, sigma 2 receptors are hijacked by ORF9c. ORF9c protein was found to interact with multiple proteins that modulate IkB kinase and NF-kB signalling pathway including NLRX1, F2RL1, NDFIP2 (29).

- ORF10 (38 aa): ORF10 does not have any similar proteins in the NCBI repository for SARS-CoV (ORF10 had a premature stop codon in both SARSCoV and BM48-31) and seems unique to SARS-CoV-2 (30).

And you, which one of these proteins are you working on?

Which one would you like to see developed and validated?

You can now contact Diaclone to discuss these topics!

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