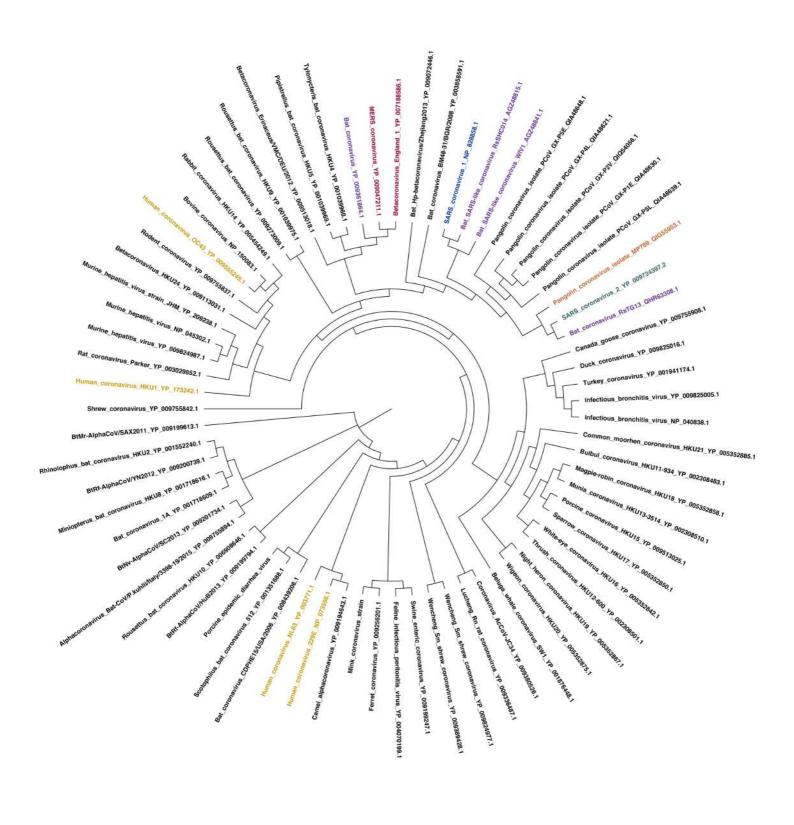
## High-resolution structure and biophysical characterization of the nucleocapsid phosphoprotein dimerization domain from the Covid-19 severe acute respiratory syndrome coronavirus 2

Luca Zinzula a, \*, Jerome Basquin b, Stefan Bohn a, c, Florian Beck a, Sven Klumpe a, Günter Pfeifer a, István Nagy a, Andreas Bracher d, F. Ulrich Hartl d, Wolfgang Baumeister a, \*

<sup>a</sup> The Max-Planck Institute of Biochemistry, Department of Molecular Structural Biology, Am Klopferspitz 18, 82152 Martinsried, Germany
<sup>b</sup> The Max-Planck Institute of Biochemistry, Department of Structural Cell Biology, Am Klopferspitz 18, 82152 Martinsried, Germany
<sup>c</sup> The Max-Planck Institute of Biochemistry, Department of Cellular Machines and Signaling, Am Klopferspitz 18, 82152 Martinsried, Germany
<sup>d</sup> The Max-Planck Institute of Biochemistry, Department of Cellular Biochemistry, Am Klopferspitz 18, 82152 Martinsried, Germany
Corresponding authors: zinzula@biochem.mpg.de; baumeist@biochem.mpg.de

Supplementary Material



**Fig. S1. Phylogenetic analysis of the SARS-CoV-2 N.** Maximum-likelihood phylogenetic tree of the CoV N protein based on reference sequences from members of the *Coronaviridae* family. Sequences from human CoVs N are highlighted in ochre. Sequences from MERS-CoV, SARS-CoV and SARS-CoV-2 N are highlighted in purple, blue and dark green, respectively. Sequences of closely-related viral species of bat and pangolin origin are highlighted in violet and orange, respectively.

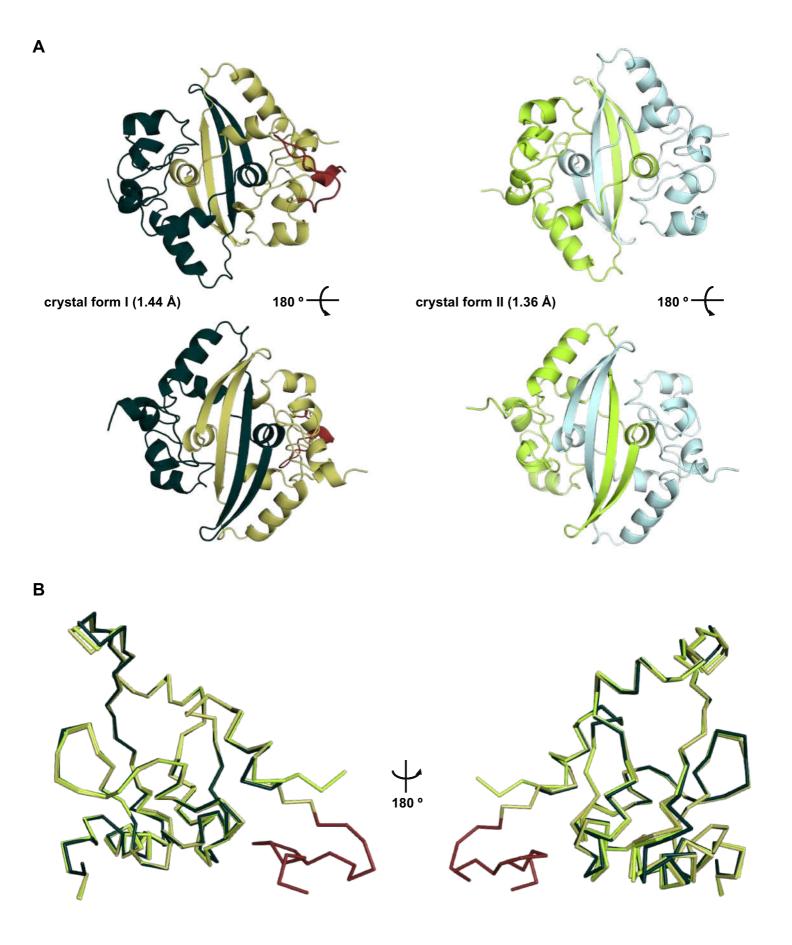
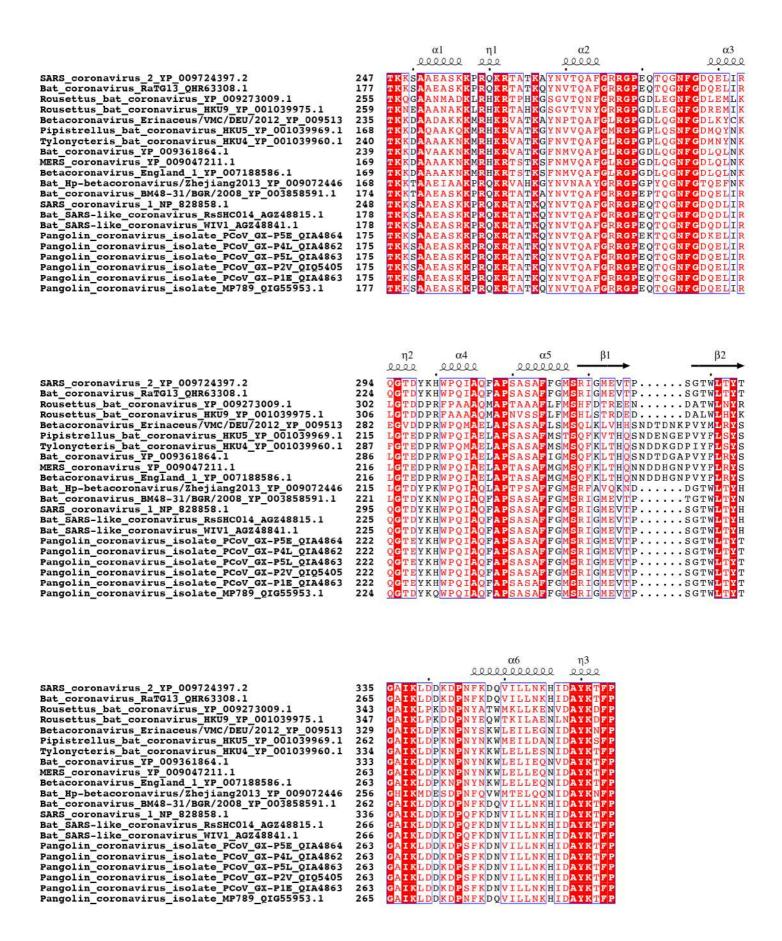
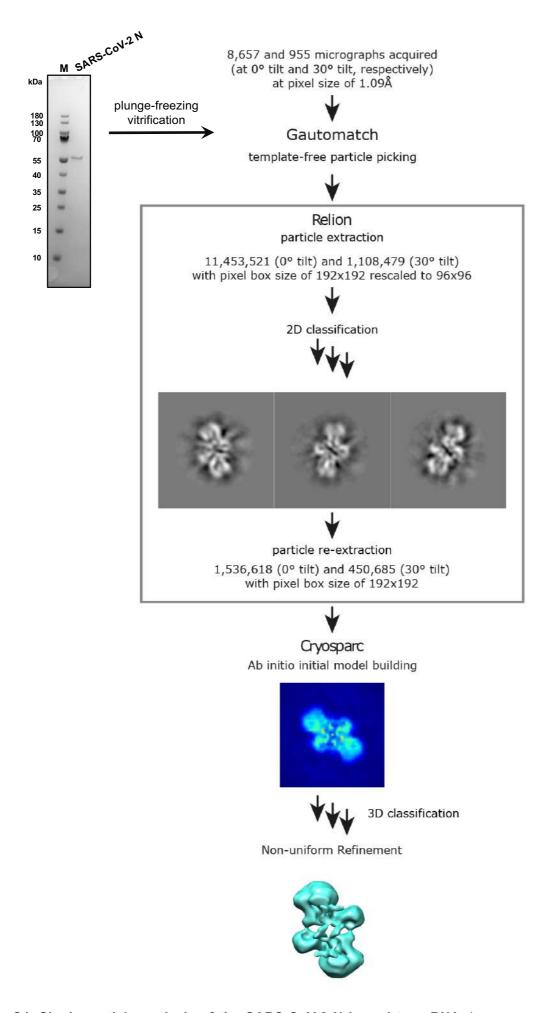


Fig. S2. Crystal structures of the SARS-CoV-2 N CTD. (A) Ribbon representation of the SARS-CoV-2 N CTD crystal form I (PDB: 6YUN, left) and crystal form II (PDB: 6ZCO, right). (B) Cα-backbone trace representation of superimposed (r.m.s.d.s.  $\approx$  0.3 Å) SARS-CoV-2 N CTD monomers from crystal forms I (dark and light green) and II (lime). Structured His<sub>6</sub>-tag in crystal form I is highlighted in ruby.



**Fig. S3. Consensus sequences and topological organization of SARS-related CoV N CTD.** Multiple sequence alignment showing the N CTD amino acid consensus among the SARS-related members of the *Sarbecovirus* subgenus of the *Betacoronavirus* genus in the *Coronaviridae* family. Row above the sequence alignment indicates amino acid topological organization relative to the N CTD structure.



**Fig. S4. Single particle analysis of the SARS-CoV-2 N bound to ssRNA.** Image processing workflow of the cryo-EM dataset of the full-length SARS-CoV-2 N (SDS-PAGE analysis shown in the upper left: M, molecular weight marker) pre-incubated with a heptameric SARS-CoV-2 ssRNA genomic fragment.