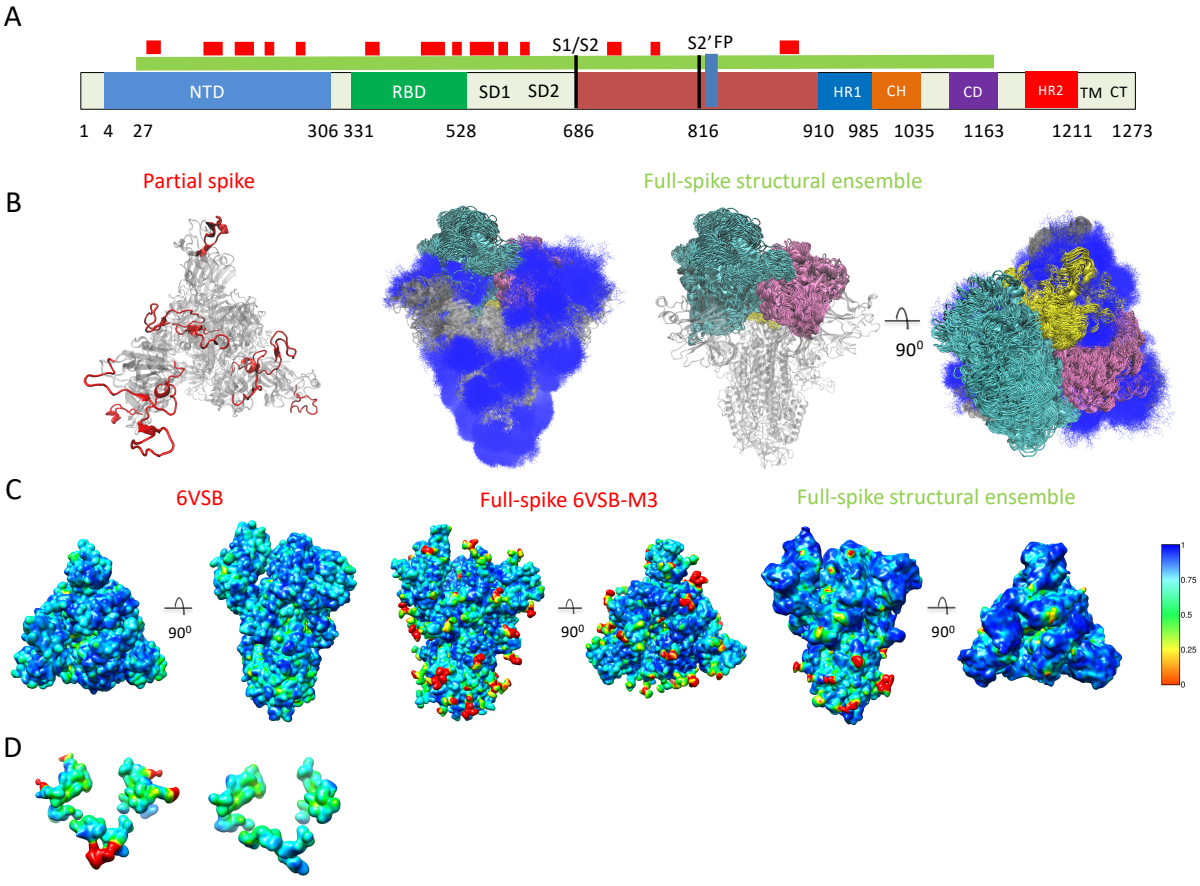
Electronic Supplementary Material (ESI) for Chemical Science.

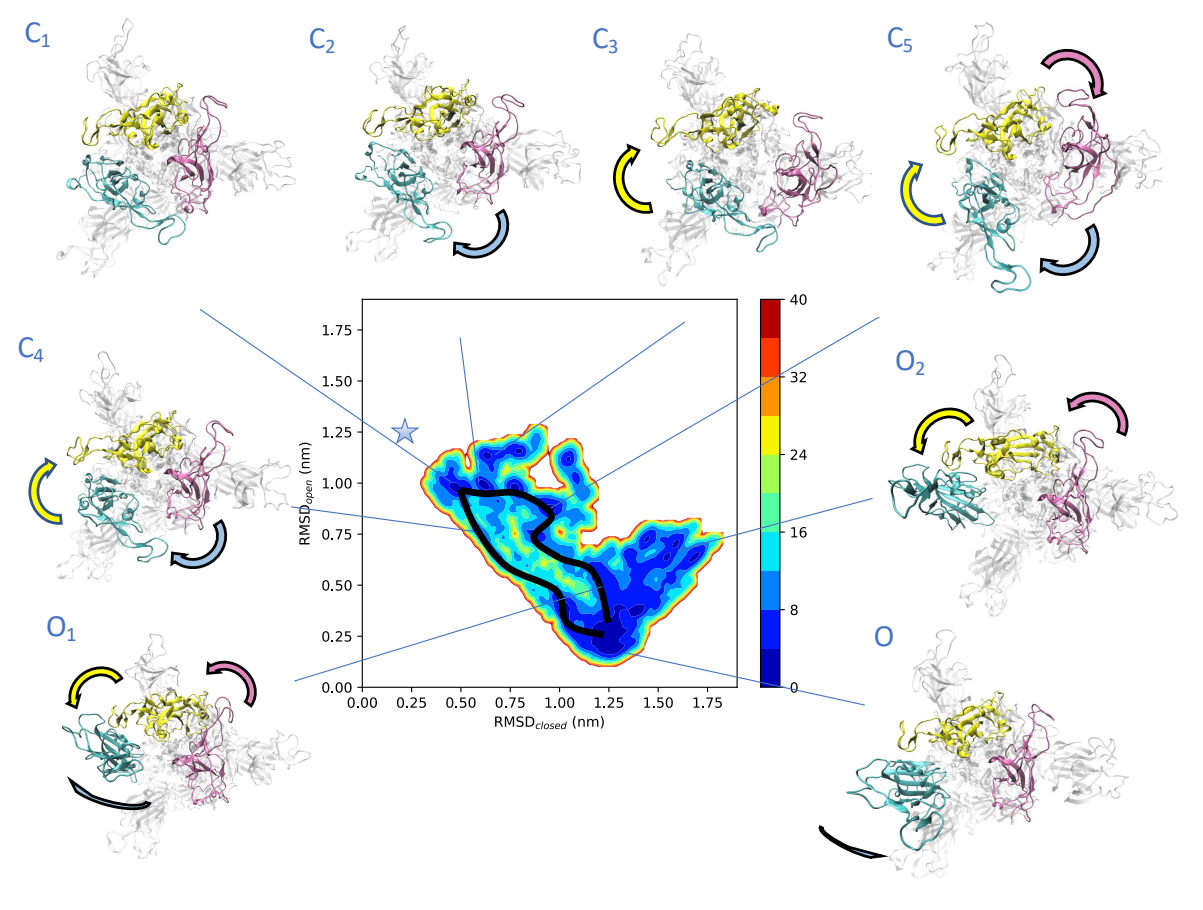
Figure Captions

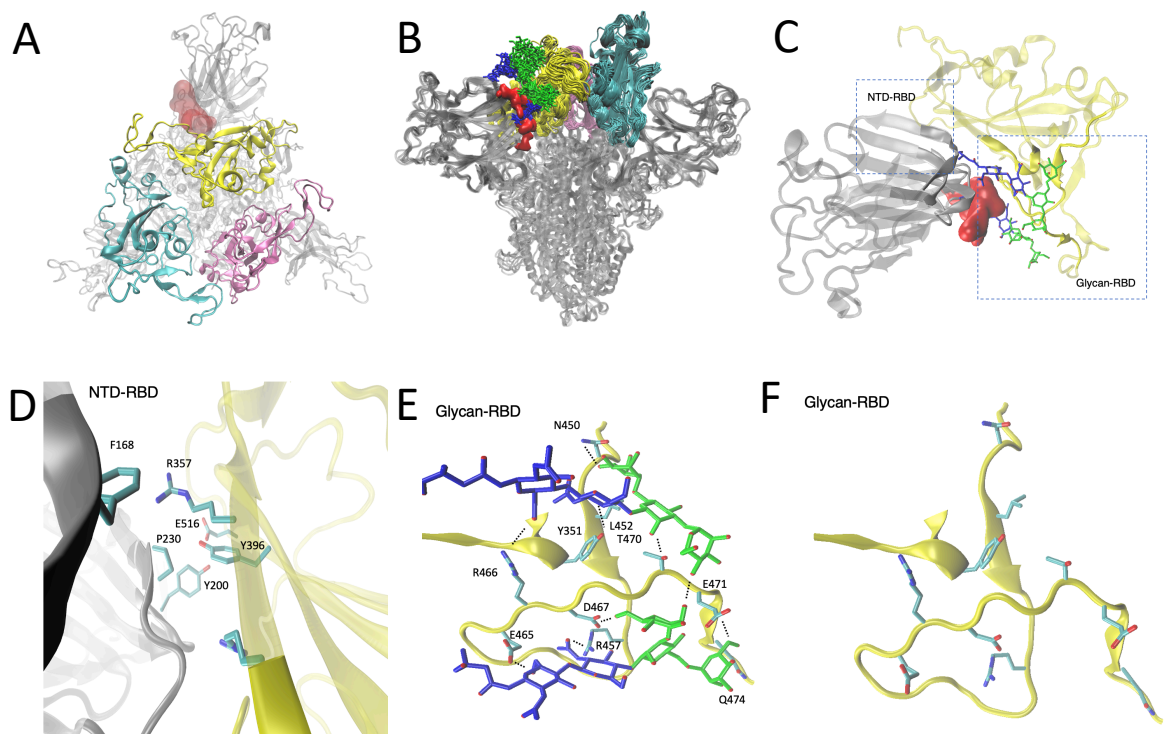
Fig. 1 Simultaneous determination of the structure and dynamics of SARS-CoV-2 spike from a cryo-EM electron density map of the open state. (A) Cartoon representation of the primary sequence and respective functional domains of spike. Regions that were not previously determined (PDB: 6VSB) are shown in red. (B) Open state structural ensemble determined in this study; the different RBD protomers RBD1, RBD2 and RBD3 are coloured in cyan, purple and yellow, respectively. The conformationally heterogeneous regions shown in red in panel A are also represented. (C) Local correlation of the experimental electron density map (EMD-21375) with a map generated using the model 6VSB (left), a map derived from the full-spike 6VSB-M3 model for the open state (middle), and with a structural ensemble average map (right). (D) Local correlation of the experimental electron density map with the average electron density map corresponding to the structural ensemble for the region of glycans N717, N1098 and N1094, showing that the correlation is improved if the glycans are absent.

Fig. 2 Determination of intermediate states in the opening transition pathways of spike. The structural ensemble of spike in the open state (Fig. 1) is represented here as a free energy landscape as a function of the structural distances (in terms of RMSD) from previously reported closed state (PDB: 6VXX) and open state (PDB: 6VSB) structures. Cyan, purple and yellow arrows indicate the typical motions of RBD1, RBD2 and RBD3, respectively. Two distinct opening pathways are drawn by black lines, while with asterisk the position of the closed state (PDB: 6VXX). The energy is given in kBT units.

Fig. 3 Identification of a cryptic pocket in an intermediate state of spike populated in the opening transition. (A) Position (shown in pink) of the cryptic pocket between the NTD and the RBD, which is present in the intermediate state C2. (B) Structural ensemble of the glycans in the NTD and RBD in the intermediate state C2, illustrating their protective role of the cryptic pocket. (C) Position of the cryptic pocket in the intermediate state C2 with the glycans highlighted. (D) Interatomic interactions between the NTD and the RBD. (E) Interatomic interactions between the glycans and the RBD. (F) Representation of RBD residues interacting with the glycans.


A
S1/S2
S2'
FP
NTD
RBD
SD1
SD2
HR1
CH
CD
HR2
TM
CT
1
4
27
306
331
528
686
816
910
985
1035
1163
1211
1273
B
Partial spike
Full-spike structural ensemble
90⁰
C
6VSB
Full-spike 6VSB-M3
Full-spike structural ensemble
90⁰
90⁰
90⁰
1
0.75
0.5
0.25
0
D


C1
C2
C3
C5
C4
O2
O1
O
RMSDopen (nm)
RMSDclosed (nm)
0.00
0.25
0.50
0.75
1.00
1.25
1.50
1.75
0
8
16
24
32
40


A
B
C
NTD-RBD
Glycan-RBD
D
NTD-RBD
F168
R357
E516
P230
Y396
Y200
E
Glycan-RBD
N450
Y351
L452
T470
R466
E471
D467
E465
R457
Q474
F
Glycan-RBD