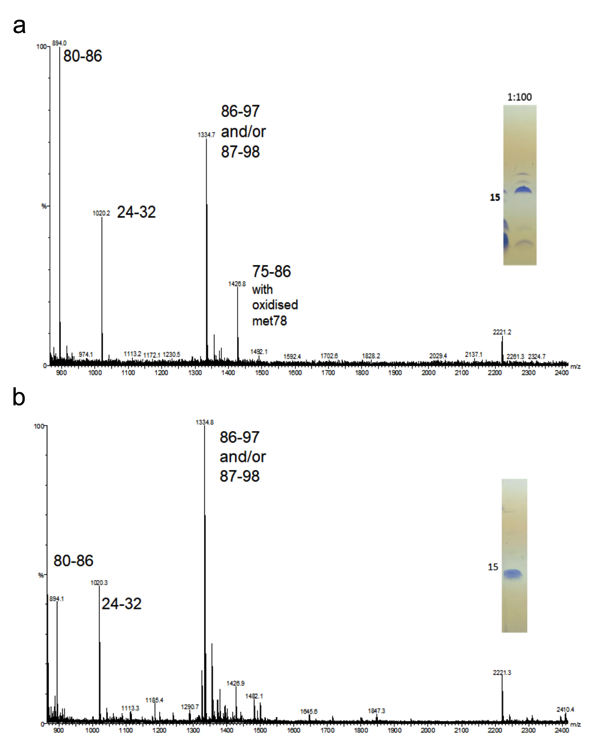
Supplementary Materials

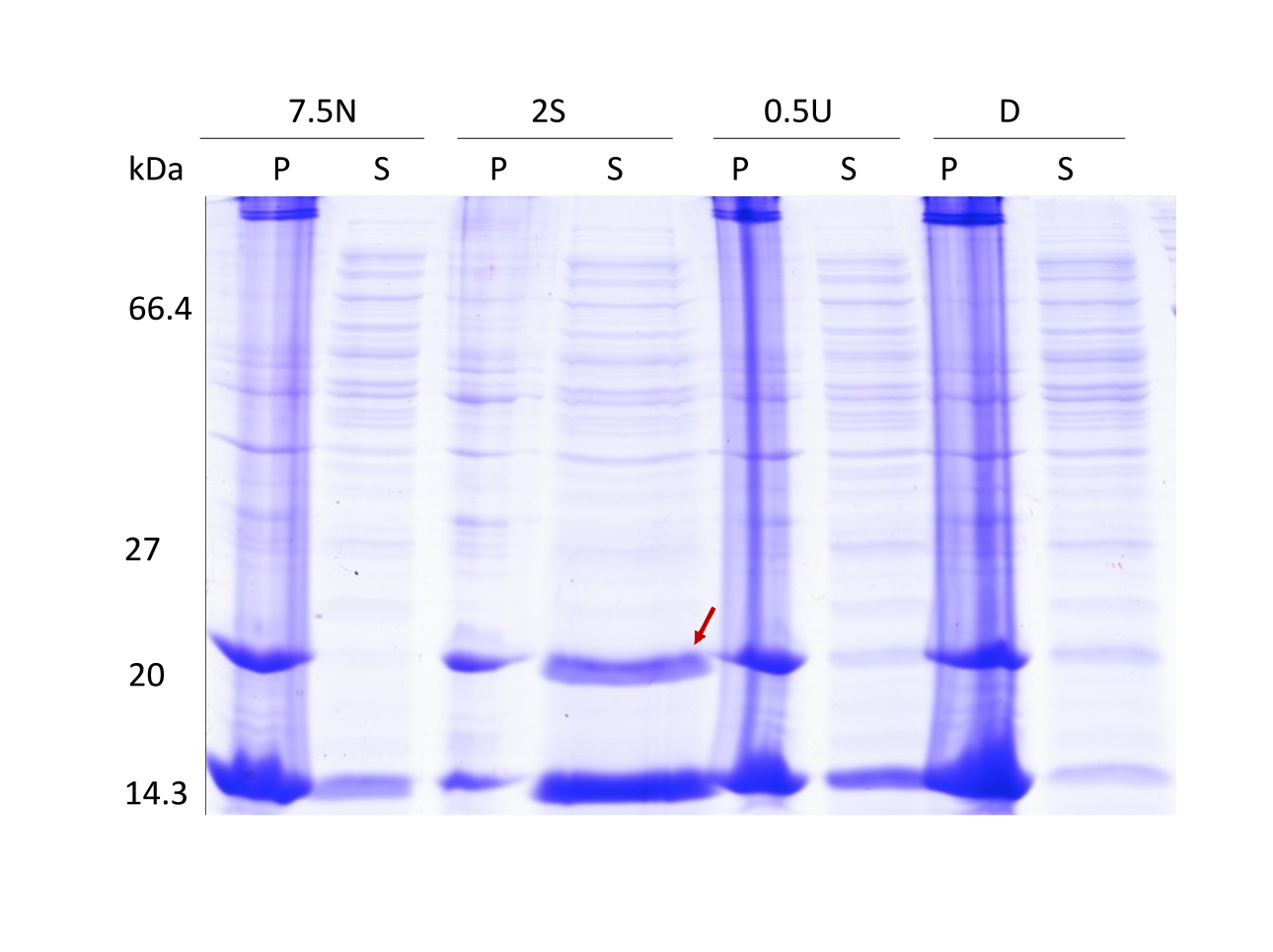
**Figure S1.** Mass-spectrometry analysis of JEV capsid protein. MS analysis of digested (**a**) and purified capsid (**b**). SDS-PAGE gels of the samples used for MS analysis are shown. Residue numbers are indicated next to the peaks.



**Figure S2.** Hydrogen bonds at the JEV capsid dimer interface. Hydrogen bonds at the α1-α1’, α2-α2’, and α3-α3’ are shown with dashed lines in figure (**a**), (**b**), and (**c**), respectively. One monomer is in orange. Another is in green.



**Figure S3.** DENV capsid α1-α3 bundle. Residues invlove in side chain packing of DENV (PDB: 1R6R) capsid α1-α3 bundle are labeled. Each monomer coloured by the 1-4 α helices in orange, green, purple, and yellow, respectively.

**Figure S4.** JEV capsid protein lysis buffer screening. Lysis buffer screening demonstrated that capsid protein is soluble in 2 M salt buffer indicating by red arrow and partially soluble in buffer containing urea and detergent. Pellet fractions and suspension fractions were labelled P and S, respectively. Four lysis buffers were the buffer added with the following additives: no additive (7.5N), 2 M NaCl (2S), 0.5 M urea (0.5U), and 0.2% Triton X 100 (D).

**Table S1.** Data collection and refinement statistics.

|  |  |
| --- | --- |
|  | **JEV Capsid** |
| **Data Collection** |  |
| Space group | P212121 |
| Cell dimensions |  |
| *a*, *b*, *c* (Å) | 46.31, 49.78, 68.25 |
| α,β,γ(°) | 90, 90, 90 |
| Resolution (Å) | 38.32-1.98(2.03-1.98)\* |
| *R*merge  *R*pim | 0.058(0.862)  0.044(0.686) |
| *I* / σ*I*  CC half | 12.1(1.4)  0.999(0.551) |
| Completeness (%)  Redundancy | 99.6(99)  4.5(4.3) |
|  |  |
| **Refinement** |  |
| Resolution (Å) | 38.32-1.98 |
| No. reflections | 10871 |
| *R*work / *R*free | 0.188/0.237 |
| No. atoms  Protein  Water  Ethylene glycol  Citrate ion  *B-*factors  Protein  Water  Ethylene glycol  2-propanol  Citrate ion | 1229  1144  56  16  13  42.77  53.26  52.91  -  66.22 |
| R.m.s. deviations |  |
| Bond lengths (Å) | 0.014 |
| Bond angles (°) | 1.428 |

**Table S2.** Hydrogen bonds between JEV capsid dimer interfacing residues and the distance.

|  |  |  |  |
| --- | --- | --- | --- |
| **Number** | **Structure 1** | **Distance (Å)1** | **Structure 2** |
| 1 | Lys 31 [NZ] | 2.64 | Asp 39 [OD1]2 |
| 2 | Arg 45 [NE] | 3.11 | Phe 56 [O] |
| 3 | Arg 45 [NH2] | 3.13 | Phe 56 [O] |
| 4 | Arg 98 [NH2] | 3.36 | Trp 69 [O] |
| 5 | Arg 98 [NH1} | 2.94 | Val 72 [O] |
| 6 | Lys 85 [NZ] | 3.87 | Lys 85 [O] |
| 7 | Lys 55 [NZ} | 3.03 | Glu 87 [O} |
| 8 | Lys 74 [NZ] | 2.77 | Arg 98 [O] |
| 9 | Asp 39 [OD1] | 2.69 | Lys 31 [NZ] |
| 10 | Phe 56 [O] | 3.16 | Arg 45 [NH2] |
| 11 | Phe 56 [O] | 3.07 | Arg 45 [NE] |
| 12 | Glu 87 [O] | 3.00 | Lys 55 [NZ] |
| 13 | Arg 98 [O] | 2.98 | Lys 74 [NZ] |

1 Assembly analysis in the program PISA [[50](#_ENREF_50)].

**Table S3.** Salt bridges between JEV capsid dimer interfacing residues.

|  |  |  |  |
| --- | --- | --- | --- |
| **Number** | **Structure 1** | **Distance (Å)1** | **Structure 2** |
| 1 | Lys 31 [NZ] | 2.64 | Asp 39 [OD1] |
| 2 | Lys 74 [NZ] | 2.77 | Arg 98 [O] |
| 3 | Asp 39 [OD1] | 2.69 | Lys 31 [NZ] |
| 4 | Arg 98 [O] | 2.98 | Lys 74 [NZ] |

1 Assembly analysis in the program PISA.

**Table 4.** JEV capsid dimer interfacing residues reported with accessible (ASA) and buried surface area (BSA), solvation energy effect (ΔG) and conservation score.

(The values are reported from one monomer)

| **Number** | **Residue** | **ASA (Å2)** | **BSA (Å2)** | **ΔG (kcal/mol)** | **Conservation1** |
| --- | --- | --- | --- | --- | --- |
| 1 | Leu27 | 83.65 | 21.41 | 0.34 | 1 |
| 2 | Val30 | 117.35 | 93.60 | 1.5 | 2\* |
| 3 | Lys31 | 144.46 | 90.81 | 0.19 | 8 |
| 4 | Val33 | 69.18 | 18.08 | 0.29 | 3\* |
| 5 | Val34 | 64.36 | 56.65 | 0.91 | 6\* |
| 6 | Met35 | 100.45 | 54.43 | 1.29 | 1 |
| 7 | Leu37 | 10.72 | 9.71 | 0.16 | 5\* |
| 8 | Leu38 | 49.69 | 37.96 | 0.59 | 3\* |
| 9 | Asp39 | 74.39 | 20.45 | -0.36 | 2\* |
| 10 | Arg45 | 84.22 | 18.52 | -0.08 | 6\* |
| 11 | Phe46 | 93.98 | 43.04 | 0.67 | 2\* |
| 12 | Ala49 | 41.95 | 41.95 | 0.56 | 7 |
| 13 | Leu50 | 24.80 | 24.80 | 0.38 | 2\* |
| 14 | Ile51 | 19.52 | 7.19 | 0.12 | 5\* |
| 15 | Thr52 | 28.95 | 28.56 | 0.17 | 5\* |
| 16 | Phe53 | 92.37 | 91.73 | 1.47 | 7\* |
| 17 | Phe54 | 22.78 | 16.86 | 0.27 | 5\* |
| 18 | Lys55 | 164.75 | 92.62 | -1.11 | 7\* |
| 19 | Phe56 | 146.69 | 144.04 | 1.49 | 6\* |
| 20 | Thr57 | 60.84 | 42.49 | 0.44 | 6\* |
| 21 | Leu59 | 102.50 | 49.22 | 0.79 | 7\* |
| 22 | Ala60 | 89.25 | 18.81 | -0.10 | 5\* |
| 23 | Thr62 | 52.30 | 27.04 | 0.43 | 8 |
| 24 | Trp69 | 87.57 | 51.05 | 0.60 | 8 |
| 25 | Lys70 | 155.76 | 6.41 | 0.02 |  |
| 26 | Lys72 | 25.04 | 11.51 | -0.11 |  |
| 27 | Lys74 | 158.12 | 83.67 | -0.29 | 6\* |
| 28 | Ala77 | 16.40 | 16.40 | 0.26 | 9 |
| 29 | Met78 | 106.53 | 38.83 | 0.97 | 7\* |
| 30 | Leu81 | 56.97 | 56.80 | 0.77 | 8 |
| 31 | Thr82 | 69.35 | 21.75 | 0.35 | 2\* |
| 32 | Phe84 | 36.14 | 36.14 | 0.51 | 7\* |
| 33 | Lys85 | 156.49 | 85.08 | -0.30 | 7 |
| 34 | Glu87 | 97.01 | 47.74 | 0.13 | 6\* |
| 35 | Leu88 | 79.21 | 79.04 | 1.24 | 7 |
| 36 | Gly89 | 30.81 | 14.57 | 0.21 | 5\* |
| 37 | Thr90 | 93.35 | 9.88 | 0.16 | 4\* |
| 38 | Leu91 | 108.63 | 90.54 | 1.44 | 7 |
| 39 | Ile92 | 91.95 | 74.87 | 1.19 | 6\* |
| 40 | Val95 | 102.29 | 87.78 | 1.23 | 4\* |
| 41 | Asn96 | 114.80 | 45.24 | -0.49 | 8 |
| 42 | Lys97 | 165.54 | 0.48 | 0.01 | 3\* |
| 43 | Arg98 | 130.94 | 29.07 | 0.03 | 9 |

1 Amino acid conservation scores are given by Consurf. [[51](#_ENREF_51)] (9 = conserved and 1 = variable) The following UniProtKB were submitted for conservation score calculation: W0LHC1, A0A1B2CW17, A0A0A7AAT7, W0LLY6, Q32ZE1, P12823, P14335, Q89277, P07720, and P27395.

* Below the confidence cut-off [[51](#_ENREF_51)].

**References**

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