C-phycocyanin as a highly attractive model system in protein crystallography, unique crystallization properties and packing-diversity screening

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### Table S1  
Crystal structures of C-phycocyanin from cyanobacteria.

| PDB ID | Source                | Max. resolution | Symmetry | Precipitant                        | Unit cell parameters |
|--------|-----------------------|-----------------|----------|------------------------------------|----------------------|
| 4ZIZ   | T. elongatus          | 1.75Å           | H32      | PEG 3350, HEPES pH 7.0             | 186, 186, 60         |
|        |                       |                 |          | Ammonium sulfate, MES pH 6.1       | 90, 90, 120          |
| 4Z8K   | T. elongatus          | 2.5Å            | P6,      |                                    | 153, 153, 39         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 4H0    | T. elongatus          | 2.2Å            | P1 2 1  | PEG 4000                           | 106, 113, 184        |
| M      | PCC7942               |                 |          |                                    | 90, 90, 120          |
| 1JBO   | T. elongatus          | 1.45Å           | H32      | Ammonium sulfate, MES pH 6.1       | 188, 188, 60         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 4N6S   | T. vulcanus           | 2.4Å            | H32      | 1.4 M phosphate buffer              | 188, 188, 60         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 4GX    | T. vulcanus           | 3.0Å            | P6,      | Ammonium sulfate, Tris pH 8.0      | 153, 153, 39         |
| E      |                       | 2.5 Å           |          |                                    | 90, 90, 120          |
|        | 4GY3                  |                 |          |                                    | 186, 186, 60         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 3O18   | T. vulcanus           | 1.35Å           | H32      | Ammonium sulfate, sucrose, Tris pH 8.0 | 186, 186, 60         |
| 3O2C   | T. vulcanus           | 1.5Å            | H32      | 1.2 M phosphate buffer              | 187, 187, 60         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 1ON7   | T. vulcanus           | 2.7Å            | P6,      | PEG4000, bis Tris pH 7.0            | 153, 153, 39         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 5TO    | Pseudanabaena sp. lw0831 | 2.04 Å  | P2 1 2 1  | PEG 4000, sodium acetate pH 5.0    | 67, 175, 194         |
|        |                       |                 |          |                                    | 90, 90, 90           |
| 4L1E   | Leptolyngbya sp. N62DM | 2.61Å   | C121     | PEG 2000, Tris pH 9.0               | 183, 107, 111        |
|        |                       |                 |          |                                    | 90, 98, 90           |
| 4F0T   | Synechocystis sp. PCC 6803 | 2.61Å | P6,      | PEG 4000, MgSO4 Tris pH 8.0        | 153, 153, 40         |
|        |                       |                 |          |                                    | 90, 90, 120          |
| 1HA7   | Arthrospira platensis | 2.2Å            | P1 2 1  | PEG 6000, 10% EtOH pH 6.8           | 107, 115, 183        |
| 1GH0   |                       |                 |          |                                    | 90, 90, 90           |
| 1CPC   | Microchaete diplosiphon | 1.66Å  | R3       | 10 to 12.5% PEG, 0.1 M phosphate pH 5.0 | 180, 180, 61         |
|        |                       |                 |          |                                    | 90, 90, 120          |
C-Phycocyanin microcrystals appear to have an advantage as a model protein for easy visualization in serial crystallography experiments. As shown in figure S1A and B, the crystal density on a fix target chip can be easily adjust. The microcrystals shown in (A, B) were grown with the batch method and appear in few hours.

**Figure S1**  (a and b) Microcrystalline material loaded on a silicon chip (Lieske et al., 2019) and used in a serial crystallography setup. The natural blue color makes the crystal density on the chip easily adjusted. Microcrystals diffracted up to 2.3 Å resolution (Meents et al., 2017). The diameter of a single hole is 30 µm and crystals do not exceed 10 µm diameter.  (c) Self-assembled crystalline material of larger particle size (formed in solution in acetate buffer pH4) loaded onto an XtalTool-HT (Feiler et al., 2019) and data were collected with the serial crystallography approach. (d) Application of C-PC microcrystals for serial crystallography on silicon chip directly at the beam. The mother liquid and nanocrystal are blotted before the data collection.
Figure S2  (a) Images of randomly picked crystals in UV-TEF and SONICC. The images show that the SHG signal is not enhanced by the chromophore present in the protein therefore there is not a false positive signal during imaging. (b) Examples of droplets were two crystal sizes are appearing, therefore in these cases, we included both sizes in the statistics shown in Figure 2(a).
Figure S3  Images of another three 96 well plates in visible (left) and the SHG imaging mode (right) in continuation of figure 3. The details on the plates are described in table 3 (protein buffered at pH 6.5) and the text. To examine the possibility of a false positive SHG signal due to the presence of the chromophore in C-PC or a higher symmetry space group, UV-TEF imaging was additionally utilized, which is based on intrinsic tryptophan fluorescence (see examples in figure S2).
Figure S4  Images of the 96 well plates in visible light (left) and the SHG imaging mode (right). The second harmonic generation imaging, as shown on the right side, is positive, i.e., light is emitted when chiral crystals are present in the droplet. To examine the possibility of a false positive due to the presence of the chromophore in C-PC or a higher symmetry space group UV-TPEF imaging was additionally utilized, which is based on intrinsic tryptophan fluorescence (see examples in figure S2).

![Image](image1.png)

Figure S5  The size distribution of crystals as they appear under different conditions. Naturally, within one crystallization drop, crystal sizes may vary, the results are determined by the observations of the majority of crystals. In some cases, when the crystals have two distinct size regimes, both are included, see examples in figure S2. (b) Three categories of crystal morphology in the C-PC crystallization experiments utilizing three different screens as shown in table 1, with C-PC in Tris buffer at pH 8.0. Please note that the morphologies are reported for crystals bigger than 10 µm.

![Image](image2.png)

Figure S6  A total number of 118 datasets were analysed. The data were categorized into six different resolution classes. The cumulative percentage of individual categories is reflected. The number of individual datasets populating the different maximum resolution bins in Å is provided.

![Image](image3.png)
Figure S7  Individual $\Delta G_{\text{int}}$, and $\Delta G_{\text{diss}}$, calculated in kcal/mol, indicate the solvation free energy gain upon formation of the individual assembly of the C-PC molecules and the individual assembly dissociation, respectively, $\Delta G_{\text{int}}$ is the difference in total solvation energies of isolated and assembled molecules without taking the effect of satisfied hydrogen bonds and salt bridges across the interfaces into account. The positive values of $\Delta G_{\text{diss}}$ indicate that external driving forces are needed to be applied to dissociate the given assembly. Values of $\Delta G_{\text{diss}}>0$ indicate thermodynamically stable complexes.
Figure S8  Calculated Surface areas are plotted for each observed space group and indicate the solvent-accessible protein surface area of individual assemblies. The buried area depicts the solvent-accessible surface area of monomeric units buried upon the assembly of hexameric or dodecameric structures. All surface areas are calculated in square Å.
**Figure S9**  (a) Superposition of all structural models. The average C<sub>a</sub> rmsd calculated to 0.57 Å is slightly higher than the structural coordinate error. All models are individually coloured and the positions of the phycocyanobilin cofactor are numbered throughout all panels. (b) A magnified stick representation of the cofactor and its binding region in cartoon representation is shown for each of the three ligand molecules. (c) The ligand binding pockets were calculated and are shown for each structure is superposition with the occupying ligand in stick representation. The dotted line in the middle panel indicates a 2.8 Å difference in the ligand binding position.
Table S2  Indexing parameters on data collected from crystals larger than 70 µm.

The protein was in 20 mM TRIS pH 8.0, 100 mM NaCl. The crystallization screens used is mentioned on the first column and the number from A1 to H12 corresponds to the drops on a 96 well plate MRC2. The crystallization experiments were set up manually with the mixing of 1 µl protein and 1 µl precipitant. All the plates were stored at 20 °C unless is mentioned otherwise. The crystals were randomly picked and froze in 25% PEG400 when cryoprotectant was necessary.

| No. | Screen       | SPG no | a [Å]       | b [Å]       | c [Å]       | α [deg] | β [deg] | γ [deg] | Maxres [Å] | pH | Crystallization solution                                                                 |
|-----|--------------|--------|-------------|-------------|-------------|---------|---------|---------|------------|----|----------------------------------------------------------------------------------------|
| 1   | Morpheus3 B4 | 155    | 186.8       | 186.8       | 59.96       | 90      | 90      | 120     | 1.05       | 6.5| 1.5% Vitamins Mix. 0.1 M Buffer System1. 50% Precipitant Mix 4                          |
| 2   | Morpheus3 H1 | 155    | 187.31      | 187.31      | 59.92       | 90      | 90      | 120     | 1.05       | 6.5| 0.8% Anaesthetic alkaloids mix. 0.1 M Buffer System1. 50% Precipitant mix 1            |
| 3   | Morpheus3 D1 | 155    | 187.23      | 187.23      | 59.7        | 90      | 90      | 120     | 1.07       | 6.5| 0.35% Phytochemicals 1 mix; 0.1 M Buffer System 1; 50% Precipitant Mix 1               |
| 4   | Morpheus3 E1 | 155    | 187.22      | 187.22      | 59.85       | 90      | 90      | 120     | 1.07       | 6.5| 0.25% Phytochemicals 2 mix; 0.1 M Buffer System 1; 50% Precipitant Mix 1               |
| 5   | Morpheus3 A1 | 155    | 186.98      | 186.98      | 59.74       | 90      | 90      | 120     | 1.08       | 6.5| 1.6% Dipeptide Mix. 0.1 M Buffer System1. 50% Precipitant Mix 1                        |
| 6   | JCSG C11     | 155    | 187.28      | 187.28      | 59.84       | 90      | 90      | 120     | 1.10       | 4.6| 2.0 M Ammonium sulfate; 0.1 M Sodium acetate                                           |
| 7   | JCSG E10     | 155    | 187.38      | 187.38      | 60.11       | 90      | 90      | 120     | 1.10       | 9  | 0.1 M Bicine; 10% w/v PEG 6000                                                        |
| 8   | JCSG H2      | 155    | 187.02      | 187.02      | 59.97       | 90      | 90      | 120     | 1.10       | -  | 1.0 M Ammonium sulfate; 0.1 M BIS-Tris; 1% w/v PEG 3350                              |
| 9   | JCSG A3      | 155    | 187.01      | 187.01      | 59.94       | 90      | 90      | 120     | 1.12       | -  | 0.2 M Ammonium citrate dibasic; 20% w/v PEG 3350                                      |
| 10  | PGA H8       | 155    | 187.64      | 187.64      | 59.87       | 90      | 90      | 120     | 1.12       | 7  | 0.1 M Ammonium sulfate; 0.3 M Sodium formate; 0.1M Tris; 3% w/v γ-PGA (Na+ form. LM); 10% w/v PEG 2000 MME |
| 11  | JCSG C4      | 155    | 186.89      | 186.89      | 59.72       | 90      | 90      | 120     | 1.13       | 7  | 0.1M HEPES; 10% w/v PEG 6000                                                         |
| 12 | MIDAS H11 | 155 | 187.08 | 187.08 | 59.69 | 90 | 90 | 120 | 1.13 | - | 0.2 M Ammonium formate; 10% w/v Polyvinylpyrrolidone; 20% w/v PEG 4000
| 13 | Morpheus3 F1 | 155 | 187.19 | 187.19 | 60.12 | 90 | 90 | 120 | 1.13 | 6.5 | 0.6% Antibiotics mix; 0.1M Buffer System 1; 50% Precipitant Mix 1
| 14 | Morpheus3 F4 | 155 | 186.63 | 186.63 | 60.13 | 90 | 90 | 120 | 1.14 | 6.5 | 0.6% Antibiotics Mix. 0.1M Buffer System1. 50% Precipitant Mix4
| 15 | JCSG G9 | 155 | 187.27 | 187.27 | 60 | 90 | 90 | 120 | 1.15 | - | 0.1 M potassium thiocyanate. 30% PEG2000MME
| 16 | PGA A1 | 155 | 186.97 | 186.97 | 59.85 | 90 | 90 | 120 | 1.16 | 5 | 0.3 M Potassium bromide; 0.1 M Sodium acetate; 8% w/v γ-PGA (Na+ form. LM)
| 17 | PGA E10 | 155 | 187.5 | 187.5 | 60 | 90 | 90 | 120 | 1.16 | 6.5 | 0.1M Ammonium sulfate; 0.3 M Sodium formate; 0.1M Sodium cacodylate; 3% w/v γ-PGA (Na+ form. LM); 20% v/v PEG 500 MME
| 18 | PGA A2 | 155 | 187.36 | 187.36 | 59.73 | 90 | 90 | 120 | 1.17 | 5 | 0.2 M Magnesium chloride; 0.1 M Sodium acetate; 8% w/v γ-PGA (Na+ form. LM)
| 19 | JCSG C6 | 155 | 153.8 | 153.8 | 39.61 | 90 | 90 | 120 | 1.18 | 4.5 | 40% PEG300 100mM Phosphate/citrate
| 20 | JCSG A9 | 155 | 187.73 | 187.73 | 60.08 | 90 | 90 | 120 | 1.19 | - | 0.2 M Ammonium chloride; 20% w/v PEG 3350
| 21 | JCSG H8 | 155 | 187.03 | 187.03 | 59.63 | 90 | 90 | 120 | 1.19 | 5.5 | 0.2 M Sodium chloride; 0.1 M BIS-Tris; 25% w/v PEG 3350
| 22 | JCSG D2 | 155 | 186.79 | 186.79 | 59.73 | 90 | 90 | 120 | 1.19 | 7.5 | 0.2 M Magnesium chloride hexahydrate; 0.1 M Sodium HEPES; 30% v/v PEG 400
| 23 | MIDAS A5 | 155 | 187.35 | 187.35 | 60.1 | 90 | 90 | 120 | 1.19 | - | 0.5 M Ammonium phosphatemonobasic; 12.5% w/v Poly (acryl acid sodium salt) 2100
| 24 | MIDAS F8 | 155 | 186.76 | 186.76 | 59.49 | 90 | 90 | 120 | 1.19 | 5.5 | 0.2 M Magnesium chloride hexahydrate; 0.1 M MES; 14% v/v Pentaerythritol propoxylate (17/8 PO/OH)
|   | Method | Bijvoet | Crystallographic Information | Reason for Change | Details |
|---|---|---|---|---|---|
| 25 | MIDAS H7 | 155 | 187.41 | 187.41 | 60.14 | 90 | 90 | 120 | 1.22 | - | 0.2M Potassium citrate tribasic monohydrate; 15% w/v SOKALAN CP 42 |
| 26 | MIDAS E11 | 155 | 187.29 | 187.29 | 60.28 | 90 | 90 | 120 | 1.23 | 6.5 | 0.1 M Lithium sulfate; 0.1 M HEPES; 25% w/v Poly (acrylic acid sodium salt) 2100 |
| 27 | MIDAS H6 | 155 | 187.6 | 187.6 | 60.37 | 90 | 90 | 120 | 1.25 | 6 | 0.1 M MES; 30% w/v Poly (acrylic acid sodium salt) 5100. 10% Ethanol |
| 28 | JCSG C7 | 155 | 186.62 | 186.62 | 59.76 | 90 | 90 | 120 | 1.26 | 4.5 | 0.2 M Zinc acetate dihydrate; 0.1 M Sodium acetate; 10% w/v PEG 3000 |
| 29 | JCSG H9 | 155 | 187.61 | 187.61 | 60.05 | 90 | 90 | 120 | 1.26 | 5.5 | 0.2 M Lithium sulfate; 0.1 M BIS-Tris; 25% w/v PEG 3350 |
| 30 | JCSG B2 | 155 | 186.79 | 186.79 | 59.71 | 90 | 90 | 120 | 1.26 | - | 0.2 M Sodium thiocyanate; 20% w/v PEG 3350 |
| 31 | PGA A12 | 155 | 187.53 | 187.53 | 60.16 | 90 | 90 | 120 | 1.26 | 5 | 0.1 M Sodium acetate; 5% w/v γ-PGA (Na+ form. LM); 20% w/v PEG 2000 MME |
| 32 | PGA G5 | 155 | 186.81 | 186.81 | 59.54 | 90 | 90 | 120 | 1.26 | 7.8 | 0.1 M Tris; 5% w/v γ-PGA (Na+ form. LM); 20% w/v PEG 3350 |
| 33 | PGA B3 | 155 | 186.93 | 186.93 | 59.95 | 90 | 90 | 120 | 1.29 | 5 | 0.1M Sodium acetate; 5% w/v γ-PGA (Na+ form. LM); 12% w/v PEG 8000 |
| 34 | PGA D9 | 155 | 187.31 | 187.31 | 60.03 | 90 | 90 | 120 | 1.31 | 6.5 | 0.1 M Sodium cacodylate; 5% w/v γ-PGA (Na+ form. LM); 20% w/v PEG 3350 |
| 35 | Morpheus3 F7 | 173 | 154.24 | 154.24 | 39.59 | 90 | 90 | 120 | 1.33 | 7.5 | 0.6% antibiotics mix; 0.1M Buffer System 2; 50% precipitant mix 3 |
| 36 | JCSG A2 | 155 | 187.25 | 187.25 | 59.83 | 90 | 90 | 120 | 1.35 | 5.5 | 0.1 M Sodium citrate; 20% w/v PEG 3000 |
| 37 | MIDAS F4 | 155 | 187.5 | 187.5 | 60.18 | 90 | 90 | 120 | 1.37 | 9 | 0.2 M Sodium chloride; 0.1 M BICINE; 20% w/v Poly(acrylic acid sodium salt) 2100 |
| 38 | JCSG C5 | 155 | 187.24 | 187.24 | 60.13 | 90 | 90 | 120 | 1.39 | 7.5 | 0.8 M Sodium phosphate monobasic monohydrate; 0.1 M Sodium HEPES |
|   |   |   |   |   |   |   |   |   |
|---|---|---|---|---|---|---|---|---|
|39| JCSG E8 | 155 | 186.35 | 186.35 | 59.88 | 90 | 90 | 120 | 1.41 | 4.5 | 1.0 M Ammonium phosphate dibasic; 0.1 M Sodium acetate
|40| JCSG G6 | 155 | 187.18 | 187.18 | 59.72 | 90 | 90 | 120 | 1.42 | - | 0.2 M Sodium malonate dibasic monohydrate; 20% w/v PEG 3350
|41| JCSG A8 | 155 | 187.16 | 187.16 | 59.58 | 90 | 90 | 120 | 1.43 | - | 0.2 M Ammonium formate; 20% w/v PEG 3350
|42| Morpheus3 H6 | 173 | 108.12 | 108.21 | 66.05 | 90 | 90 | 120 | 1.45 | 7.5 | 0.8% Anaesthetic alkaloids mix. 0.1 M Buffer System2 .50% Precipitant mix2
|43| JCSG E9 | 155 | 187.42 | 187.42 | 60.69 | 90 | 90 | 120 | 1.46 | 6.5 | 1.6 M Magnesium sulfate heptahydrate; 0.1 M MES
|44| JCSG G8 | 155 | 187.26 | 187.26 | 60.25 | 90 | 90 | 120 | 1.47 | - | 0.15 M d-l malic acid. 20% PEG3350
|45| JCSG E4 | 155 | 187.19 | 187.19 | 60.44 | 90 | 90 | 120 | 1.49 | 8.5 | 1.26 M Ammonium sulfate; 0.1 M Tris
|46| JCSG H1 | 155 | 187.08 | 187.08 | 59.97 | 90 | 90 | 120 | 1.54 | 5.5 | 0.3 M Magnesium formate dihydrate; 0.1M BIS-Tris
|47| MIDAS A2 | 155 | 186.06 | 186.06 | 59.82 | 90 | 90 | 120 | 1.54 | 5.5 | 0.1 M MES. 12% polyvinylpyrrolidone
|48| Morpheus3 C11 | 173 | 153.51 | 153.51 | 39.36 | 90 | 90 | 120 | 1.82 | 8.5 | 1% Nucleosides Mix. 0.1 M Buffer System3. 50% Precipitant Mix3
|49| Morph3 G6 | 4 | 151.64 | 39.01 | 157.87 | 90 | 116.9 | 90 | 1.87 | 7.5 | 1.2% Cholic Acid derivatives mix. 0.1 M Buffer System2. 50% precipitant Mix2
|50| JCSG F10 | 155 | 184.11 | 184.11 | 58.59 | 90 | 90 | 120 | 1.89 | 7 | 1.1 M Sodium malonate dibasic monohydrate; 0.1 M HEPES; 0.5% v/v Jeffamine ED-2003
|51| Morpheus3 F6 | 173 | 152.85 | 152.85 | 39.28 | 90 | 90 | 120 | 1.91 | 7.5 | 0.6% Antibiotics mix; 0.1 M Buffer System 2; 50% Precipitant Mix 2
|52| Morpheus3 F10 | 173 | 153.8 | 153.8 | 39.61 | 90 | 90 | 120 | 1.94 | 8.5 | 0.6% antibiotics mix; 0.1 M Buffer System 2; 50%precipitant mix 3
|53| Morpheus3 H7 | 173 | 153.58 | 153.58 | 39.44 | 90 | 90 | 120 | 2.03 | 7.5 | 0.8% Anesthetic alkaloids mix. 0.1M Buffer System2. 50% Precipitant mix3
|54| Morpheus3 A8 | 173 | 152.49 | 152.49 | 39.26 | 90 | 90 | 120 | 2.04 | 7.5 | 1.6% Dipeptide Mix. 0.1 M Buffer System2. 50% Precipitant Mix 4
|   | Morph 3 | 173 | 153.69 | 153.69 | 39.32 | 90 | 90 | 120 | 2.10 | 6.5 | 1.5% Vitamins Mix. 0.1 M Buffer System1. 50% Precipitant Mix2  |
|---|---------|-----|--------|--------|-------|----|----|-----|-----|-----|---------------------------------------------------------------|
| 55| B2      | 173 | 152.54 | 152.54 | 39.19 | 90 | 90 | 120 | 2.11 | 8.5 | 1.5% Vitamins Mix 0.1 M Buffer System3. 50% Precipitant Mix4  |
| 56| B12     | 173 | 152.65 | 152.65 | 39.22 | 90 | 90 | 120 | 2.23 | 6.5 | 1% Nucleosides mix; 0.1 M Buffer System 1; 50% Precipitant Mix3 |
| 57| C3      | 173 | 154.43 | 154.43 | 39.61 | 90 | 90 | 120 | 2.24 | 6.5 | 0.6% Antibiotics Mix. 0.1M Buffer System1. 50% Precipitant Mix2  |
| 58| F2      | 173 | 59.82  | 109.93 | 122.04| 112.98| 96.29| 100.12| 2.66| 5.5 | 0.1 M BIS-Tris; 25% w/v PEG 3350                              |
| 59| JCSG H3 | 1   | 154.17 | 154.17 | 39.25 | 90 | 90 | 120 | 2.68 | 6.5 | 0.8% Anesthetic alkaloids mix; 0.1 M Buffer System 1; 50% Precipitant Mix2 |
Table S3  Indexing parameters on data collected from crystals larger than 70 µm.

The protein was in 20 mM MES pH 6.5 100 mM NaCl. The crystallization screens used is mentioned on the first column and the number from A1 to H12 corresponds to the drops on a 96 well plate MRC2. The crystallization experiments were set up manually with the mixing of 1 µl protein and 1 µl precipitant. All the plates were stored at 20 °C unless is mentioned otherwise. The crystals were randomly picked and froze in 25% PEG 400 when cryoprotectant was necessary.

| No | Screen        | No | a [Å] | b [Å] | c [Å] | α [deg] | β [deg] | γ [deg] | Maxres [Å] | pH | Crystallization solution                                                                 |
|----|---------------|----|-------|-------|-------|---------|---------|---------|------------|----|----------------------------------------------------------------------------------------|
| 1  | JCSG B1       | 155| 187.65| 187.65| 60.1  | 90      | 90      | 120     | 1.07       | 4.0| 0.8 M Ammonium Sulfate. 1 M Citrate                                                    |
| 2  | Morph3 E1     | 155| 187.22| 187.22| 59.85 | 90      | 90      | 120     | 1.07       | 6.5| 0.25% Phytochemicals 2 mix. 0.1 M Buffer System 1. 50% precipitant Mix1                  |
| 3  | JCSG C4       | 155| 187.01| 187.01| 60.11 | 90      | 90      | 120     | 1.1        | 7.0| 0.1 M HEPES; 10% w/v PEG 6000                                                          |
| 4  | JCSG F1       | 155| 187.36| 187.36| 60.08 | 90      | 90      | 120     | 1.1        | 6.5| 0.05 M Cesium Chloride. 0.1 M MES. 30% Jeffamine 600                                    |
| 5  | JCSG G1       | 155| 186.91| 186.91| 60.03 | 90      | 90      | 120     | 1.1        | 7.5| 0.1 M HEPES. 30% Jeffamine ED2003                                                      |
| 6  | MIDAS G7      | 155| 186.95| 486.95| 60.14 | 90      | 90      | 120     | 1.1        | 6.5| 0.2 M Ammonium acetate; 0.1 M MES. 30% v/v Glycerol ethoxylate                          |
| 7  | Morph3 F1     | 155| 187.41| 187.41| 59.94 | 90      | 90      | 120     | 1.1        | 6.5| 0.6% antibiotics mix; 0.1 M Buffer System 1; 50% precipitant mix1                       |
| 8  | Morph3 H1     | 155| 187.21| 187.21| 59.74 | 90      | 90      | 120     | 1.1        | 6.5| 0.8% anesthetic alkaloid mix; 0.1 M Buffer System 1; 50% precipitant mix1                |
| 9  | MIDAS C2      | 155| 187.07| 187.07| 59.98 | 90      | 90      | 120     | 1.12       | 6.0| 0.2 M Sodium chloride; 0.1 M MES; 30% v/v Jeffamine ED-2003                            |
| 10 | Morpheus H1   | 155| 187.17| 187.17| 59.93 | 90      | 90      | 120     | 1.13       | 6.5| 0.1 M Amino acids. 0.1 M buffer system 1. 30% v/v P500MME_P20K                         |
| 11 | Morpheus H5   | 155| 187.16| 187.16| 59.94 | 90      | 90      | 120     | 1.15       | 7.5| 0.1 M Amino acids. 0.1 M buffer system 2. 30% v/v P500MME_P20K                         |
| 12 | Morph3 H5     | 155| 187.28| 187.28| 59.98 | 90      | 90      | 120     | 1.15       | 7.5| 0.8% anesthetic alkaloid mix; 0.1 M buffer system 2; 50% precipitant mix1                |
| 13 | PGA B2        | 155| 186.85| 186.85| 59.78 | 90      | 90      | 120     | 1.16       | 5.0| 0.1 M Sodium acetate; 5% w/v γ-PGA (Na+ form. LM); 15% w/v PEG 4000                     |
| ID  | Name          | P | Q | R | S | T | U | V | W | X | Y | Z | Notes                                                                 |
|-----|---------------|---|---|---|---|---|---|---|---|---|---|---|------------------------------------------------------------------------|
| 14  | JCSG B7       | 155| 187.11| 187.11| 59.89| 90 | 90 | 120| 1.16| 4.6 | 0.1 M Sodium acetate; 8% w/v PEG 4000                                      |
| 15  | JCSG E1       | 155| 187.79| 187.79| 60.28| 90 | 90 | 120| 1.19| 6.5 | 1 M Sodium Citrate tribasic dehydrate; 0.1 M MES                     |
| 16  | PACT A6       | 155| 187.37| 187.37| 60.14| 90 | 90 | 120| 1.19| 9.0 | 0.1 M SPG; 25% w/v PEG 1500                                           |
| 17  | PACT H2       | 155| 186.81| 186.81| 59.88| 90 | 90 | 120| 1.19| 8.5 | 0.2 M Sodium bromide; 0.1M Bis-Tris propane; 20% w/v PEG 3350         |
| 18  | Morpheus B1   | 155| 186.9 | 186.9 | 60.08| 90 | 90 | 120| 1.21| 6.5 | 0.09 M Halogens. 0.1 M buffer system1. 30% w/v P500MME_P20K           |
| 19  | Morph3 F5     | 155| 187.28| 187.28| 60.24| 90 | 90 | 120| 1.22| 7.5 | 0.6% antibiotics mix; 0.1M buffer system 2; 50% precipitant mix1      |
| 20  | JCSG F9       | 155| 187.24| 187.24| 60.21| 90 | 90 | 120| 1.23| 7.0 | 2.4 M Sodium Malonate dibasic monohydrate                              |
| 21  | Morph3 F4     | 155| 186.87| 186.87| 60.03| 90 | 90 | 120| 1.23| 6.5 | 0.6% antibiotics mix; 0.1 M Buffer System 1; 50% precipitant mix4     |
| 22  | Morpheus C1   | 155| 187.07| 187.07| 60  | 90 | 90 | 120| 1.24| 6.5 | 0.09 NPS. 0.1 M Buffer System1. 30% v/v P500MME_P20K                  |
| 23  | Morpheus B5   | 155| 187.53| 187.53| 60.14| 90 | 90 | 120| 1.25| 7.5 | 0.09 M Halogens. 0.1 M buffer System2. 30% v/v P500MME_P20K           |
| 24  | PGA G5        | 155| 186.88| 186.88| 59.96| 90 | 90 | 120| 1.25| 7.8 | 0.1 M Tris; 5% w/v γ-PGA (Na+ form. LM); 20% w/v PEG 3350             |
| 25  | PACT C11      | 155| 186.88| 186.88| 60.03| 90 | 90 | 120| 1.25| 7.0 | 0.2 M Calcium chloride dihydrate; 0.1 M HEPES; 20% w/v PEG 6000        |
| 26  | Morpheus A11  | 155| 187.07| 187.07| 60.19| 90 | 90 | 120| 1.31| 8.5 | 0.06 M Divalent. 0.1M Buffer system 3. 30% v/v GOL_P4K                 |
| 27  | MIDAS A1      | 155| 186.8| 59.88| 59.95| 90 | 90 | 120| 1.31| 6.0 | 0.1 M HEPES; 50% v/v Polypropylene glycol 400                          |
| 28  | PACT C3       | 155| 187.08| 187.08| 59.83| 90 | 90 | 120| 1.34| 6.0 | 0.1 M PCTP; 25% w/v PEG 1500                                           |
| 29  | PGA A1        | 155| 187.06| 187.06| 60.2 | 90 | 90 | 120| 1.37| 5.0 | 0.3 M Potassium bromide; 0.1 M Sodium acetate; 8% w/v-PGA (Na+ form. LM) |
| 30  | MIDAS D3      | 155| 186.93| 186.93| 59.92| 90 | 90 | 120| 1.37|  -  | 45% v/v Polypropylene glycol 400. 10% Ethanol                          |
| 31  | JCSG A8       | 155| 187.15| 187.15| 59.9 | 90 | 90 | 120| 1.39|  -  | 0.2 M Ammonium formate; 20% w/v PEG 3350                                |
|   |   |   |   |   |   |   |   |   |   |   |   |   |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 32 | JCSG F5 | 155 | 188.3 | 188.3 | 60.45 | 90 | 90 | 120 | 1.4 | 8.5 | 0.2 M Magnesium Chloride hexahydrate; 0.1M Tris, 50% Ethylene glycol |
| 33 | MIDAS A9 | 155 | 187.01 | 187.01 | 60.04 | 90 | 90 | 120 | 1.4 | 6.0 | 0.1 M MES; 25% v/v Pentaerythritol propoxylate (5/4 PO/OH) |
| 34 | PGA D12 | 155 | 186.47 | 186.47 | 59.72 | 90 | 90 | 120 | 1.42 | 6.5 | 0.1 M Sodium cacodylate; 5% w/v γ-PGA (Na+ form. LM); 8% w/v PEG 20000 |
| 35 | JCSG G12 | 155 | 186.93 | 186.93 | 59.98 | 90 | 90 | 120 | 1.42 | 5.5 | 3 M NaCl, 0.1M Bis Tris |
| 36 | PACT B8 | 155 | 187.14 | 187.14 | 60.17 | 90 | 90 | 120 | 1.43 | 6.0 | 0.2 M Ammonium chloride; 0.1 M MES; 20% w/v PEG 6000 |
| 37 | Morpheus E2 | 155 | 187.21 | 187.21 | 60.12 | 90 | 90 | 120 | 1.47 | 6.5 | 0.12 M Ethylene Glycols. 0.1 M Buffer System 1. 30%w/v EDO_P8K |
| 38 | Morpheus D5 | 155 | 186.8 | 186.8 | 60 | 90 | 90 | 120 | 1.49 | 7.5 | 0.12 M Alcohols. 0.1 M Buffer System 2. 30%w/v P500MME_P20K |
| 39 | JCSG F10 | 155 | 186.93 | 186.93 | 60.21 | 90 | 90 | 120 | 1.51 | 7.0 | 1.1 M Sodium Malonate 0.1 M HEPES. 0.5% Jeﬀamine |
| 40 | Morpheus A10 | 155 | 186.68 | 186.68 | 59.8 | 90 | 90 | 120 | 1.54 | 8.5 | 0.06 M Divalent. 0.1M Buffer System 3. 30%w/v EDO_P8K |
| 41 | PGA E12 | 155 | 187.3 | 187.3 | 60.27 | 90 | 90 | 120 | 1.54 | 6.5 | 0.1 M Ammonium sulfate; 0.3 M Sodium formate; 0.1M Sodium cacodylate; 3% w/v γ-PGA (Na+ form. LM); 10% w/v PEG 2000 MME |
| 42 | Morpheus A1 | 155 | 187.21 | 187.21 | 60.28 | 90 | 90 | 120 | 1.57 | 6.5 | 0.06 M Divalent. 0.1 M Buffer System 1. 30%w/v P500MME_P20K |
| 43 | Morpheus A2 | 155 | 187.43 | 187.43 | 60.32 | 90 | 90 | 120 | 1.58 | 6.5 | 0.06 M Divalent. 0.1 M Buffer System 1. 30%w/v EDO_P8K |
| 44 | PACT B3 | 155 | 187.2 | 187.2 | 60.22 | 90 | 90 | 120 | 1.58 | 6.0 | 0.1 M MIB; 25% w/v PEG 1500 |
| 45 | PGA B10 | 155 | 187.21 | 187.21 | 60.35 | 90 | 90 | 120 | 1.62 | 5.0 | 0.2 M Potassium bromide; 0.2M Potassium thiocyanate; 0.1 M Sodium acetate; 3% w/v γ-PGA (Na+ form. LM); 5% w/v PEG 4000 |
| No. | Name     | Neut. | cell edge [Å] | Space Group |
|-----|----------|-------|---------------|-------------|
| 46  | Morpheus A3 | 155   | 187.16        | 187.16      | 60.07 | 90   | 90   | 120   | 1.67  | 6.5  |
| 47  | Morpheus H8 | 155   | 186.56        | 186.56      | 59.71 | 90   | 90   | 120   | 1.69  | 7.5  |
| 48  | Morpheus B4 | 155   | 186.41        | 186.41      | 59.67 | 90   | 90   | 120   | 1.7   | 6.5  |
| 49  | JCSG F7   | 155   | 189.51        | 189.51      | 60.92 | 90   | 90   | 120   | 1.73  | 7.0  |
| 50  | PACT H8   | 155   | 186.71        | 186.71      | 59.85 | 90   | 90   | 120   | 1.75  | 8.5  |
| 51  | Morph3 F6 | 173   | 153.76        | 153.76      | 39.09 | 90   | 90   | 120   | 1.85  | 7.5  |
| 52  | MIDAS B6  | 155   | 185.96        | 185.96      | 60.78 | 90   | 90   | 120   | 1.89  | 7.5  |
| 53  | JCSG E8   | 155   | 186.88        | 186.88      | 60.59 | 90   | 90   | 120   | 1.96  | 4.5  |
| 54  | PACT E3   | 173   | 152.91        | 152.91      | 39.28 | 90   | 90   | 120   | 2.1   |     |
| 55  | MIDAS B1  | 173   | 152.07        | 152.07      | 39.12 | 90   | 90   | 120   | 2.11  | 5.5  |
| 56  | Morph3 G1 | 18    | 118.36        | 98.23       | 104.6 | 90   | 90   | 90    | 2.16  | 6.5  |
| 57  | PGA B4    | 155   | 180.83        | 180.83      | 57.61 | 90   | 90   | 120   | 2.51  | 5.0  |
| 58  | PGA B8    | 5     | 113.79        | 179.79      | 58.86 | 90   | 122  | 90    | 3     | 5.0  |

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Table S4  Structural comparison of all superposed protein models.

The RMSDs are plotted against the different models and calculated on the bases 4958 atoms.

| Structural C<sub>a</sub>-rmsd (Å) | P6$_1$-large | P6$_3$-small | R32  | P2,2,2 |
|-----------------------------------|--------------|--------------|------|--------|
| P6$_1$-large                      | --           | 0.663        | 0.632| 0.547  |
| P6$_3$-small                      | 0.663        | --           | 1.129| 1.044  |
| R32                               | 0.632        | 1.129        | --   | 0.326  |
| P2,2,2                            | 0.547        | 1.044        | 0.326| --     |