**Acta Cryst**

# D

## STRUCTURAL
## BIOLOGY

**Supporting information for article:**

# Determination of crystal structures of proteins of unknown identity using a marathon molecular-replacement procedure: structure of *Stenotrophomonas maltophilia* phosphate-binding protein

**Kaushik Hatti, Ashutosh Gulati, Narayanaswamy Srinivasan and M. R. N. Murthy**

**Table S1**    Top 4 homologues of SCOPIDsd4m1va_ and d3w9va_ from DALI search which performed the best when used as phasing model.

| Sl. No. | PDBID | Top LLG | Top TFZ |
|---|---|---|---|
| 1 | 2Q9T | 176.109 | 6.2 |
| 2 | 2V3Q | 156.693 | 6.8 |
| 3 | 4F1V | 176.462 | 6.0 |
| 4 | 4M1V | 159.218 | 7.5 |
| 5 | Ensemble* | 199.769 | 11.2 |

*Ensemble was generated using these 4 homologs (as discussed in Results section) and used as phasing model

**Table S2**    Comparative structural analysis of *Sm*PBP against other known PBPs

| Protein, Source Organism | Sequence Identity (%) | Number of aligned residues | RMSD | PDBID |
|---|---|---|---|---|
| PBP, *Escherichia coli* | 25.5 | 270 | 1.98 | 1IXH(Wang *et al.*, 1997) |
| PstS1, *Mycobacterium tuberculosis* | 19.5 | 194 | 2.58 | 1PC3 (Vyas *et al.*, 2003) |
| PstS, *Vibrio cholera* | 13.5 | 192 | 3.07 | 1TWY (unpublished) |
| ModA, *Escherichia coli* | 25.5 | 270 | 1.98 | 2ONK (Hollenstein *et al.*, 2007) |
| Psts, *Yersinia pestis* | 25.1 | 270 | 1.96 | 2Z22(Tanabe *et al.*, 2007) |
| ModA, tungstate, *Pyrococcus furiosus* | 8.3 | 192 | 3.21 | 3CG1 (Hollenstein *et al.*, 2009) |
| PstS, *Lactobacillus brevis* | 20.5 | 204 | 1.65 | 4ECF (unpublished) |
| PstS, *Streptococcus pneumoniae* | 15.6 | 198 | 2.81 | 4EXL (unpublished) |
| DING, *Pseudomonas fluorescens* | 52.0 | 334 | 1.28 | 4F1U(Elias *et al.*, 2012) |
| PBP, *Clostridium perfringens* | 19.0 | 215 | 1.73 | 4GD5 (Gonzalez, Richet *et al.*, 2014) |
| PstS, *Borreliella burgdorferi* | 9.5 | 168 | 3.86 | 4N13 (Brautigam *et al.*, 2014) |

A    37.32/42.44
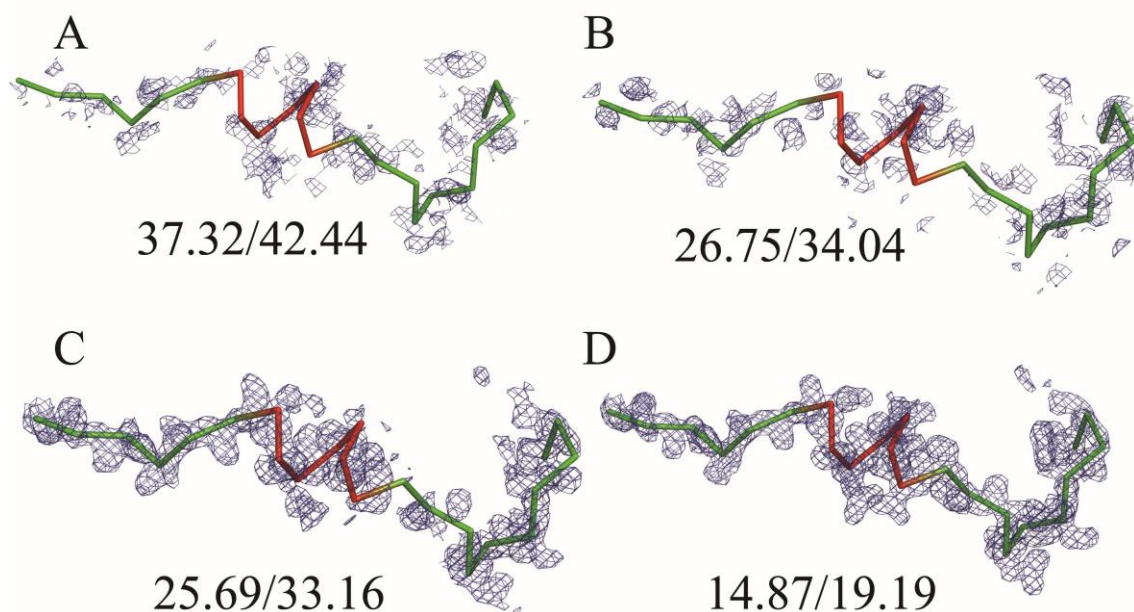
B    26.75/34.04

C    25.69/33.16

D    14.87/19.19

**Figure S1** Electron density corresponding to a peripheral region (residues 241-263) rendered as ribbon (green-loop; red-helix) illustrating the progressive improvement as model building and refinement progressed. A) to D) represents different stages of model building. Electron density (2mFo-DFc) map is contoured at 1 σ. Respective $R_{work}/R_{free}$ in percentages are written at the bottom of each panel.
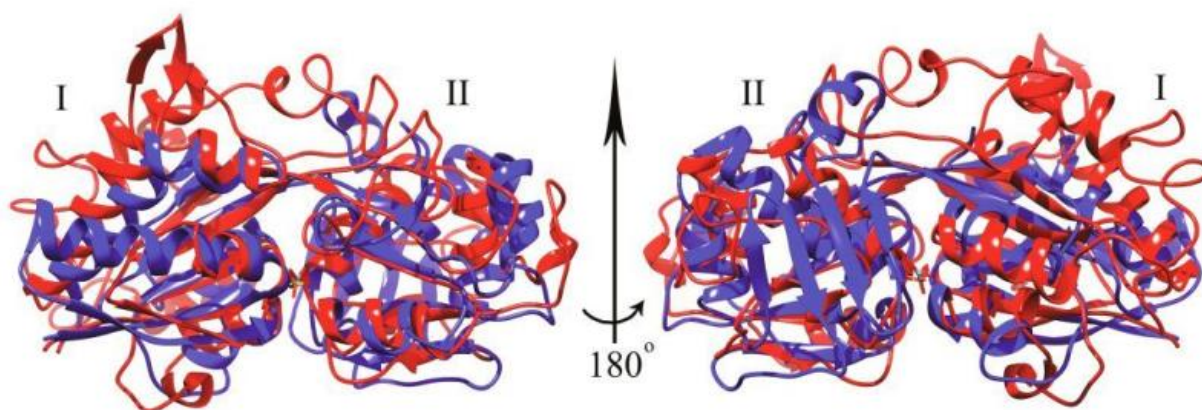


**Figure S2** *Sm*PBP (red) is superposed on the tungstate transporter protein (blue; PDB ID: 3CG1), ModA of the ABC transporter complex. I and II represent the two globular subdomains. Phosphate and tungstate (rendered in stick model) superpose within 1 Å though significant differences are observed in domain II.
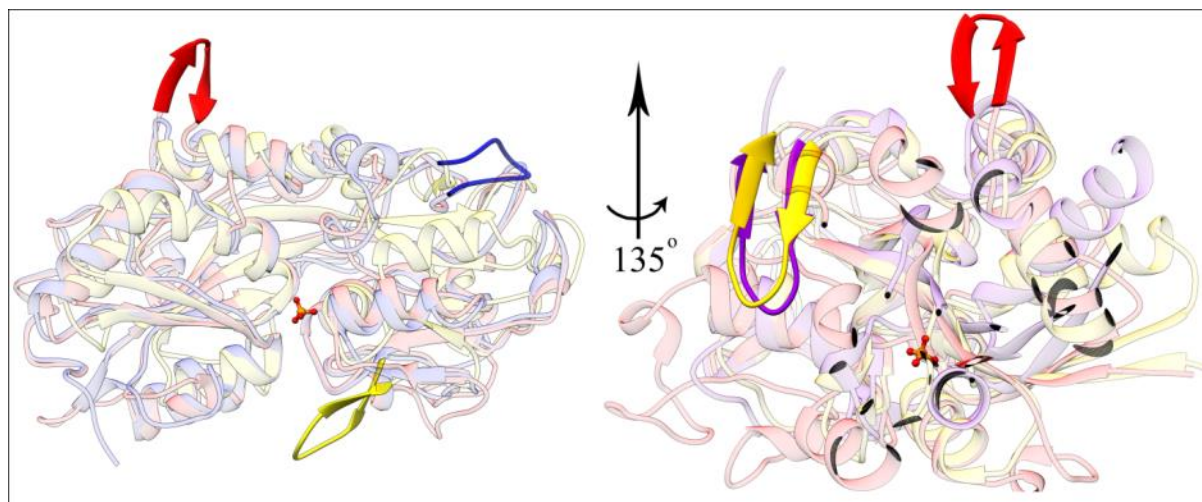
**Figure S3** Left-panel: β-hairpin like substructure of *Sm*PBP (red) occurs on domain II of *Pflu*DING (blue) and *V. cholerae*_PstS (yellow). While it is on the surface on *Sm*PDB, the same is present on near binding cleft in *Pflu*DING . Right panel: The orientation of the β-hairpin is similar in *S. pneumoniae*_PstS (violet) and *L. brevis*_PstS (golden yellow). Some parts of the protein is masked and regions other than the β-hairpin is coloured in light-shades for ease of visualisation
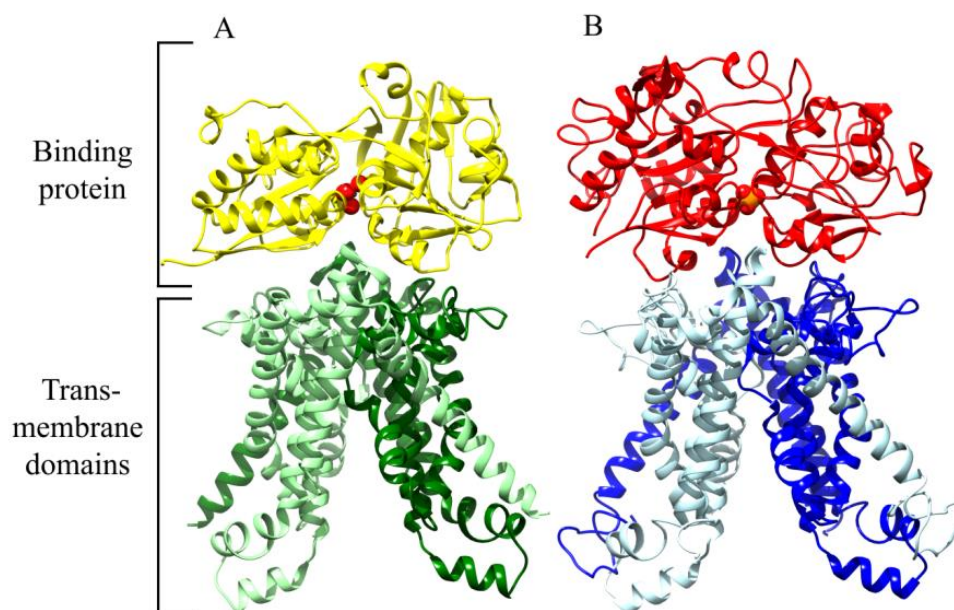


**Figure S4** Periplasmic binding protein bound to transmembrane domains of the ABC transporter complex is shown. A) ModA (yellow) is shown in complex with ModB2 (green). B) *Sm*PBP (red) forms a similar complex with modelled PstC (blue). Two chains of transmembrane domains are shown in dark and light shades and the bound ligand rendered in space-filled model.

## S1.  Homology modelling

Homology model of PstC protein, the transmembrane region of the ABC transporter complex (NCBI Reference Sequence: WP_005408790.1.) was modelled using I-Tasser (Yang *et al.*, 2014).  ModB of the ModBC complex (PDB ID: 3D31) was used as template.  TMscore of $0.58 \pm 0.14$ and C-score of -1.09 suggested that the generated model was reliable.