crystals

MDPI

*Supplementary Data*

# Structural Analysis of Xylose Isomerase from *Streptomyces avermitilis*

**Ki Hyun Nam**

College of General Education, Kookmin University, Seoul 02707, Republic of Korea; structure@kookmin.ac.kr

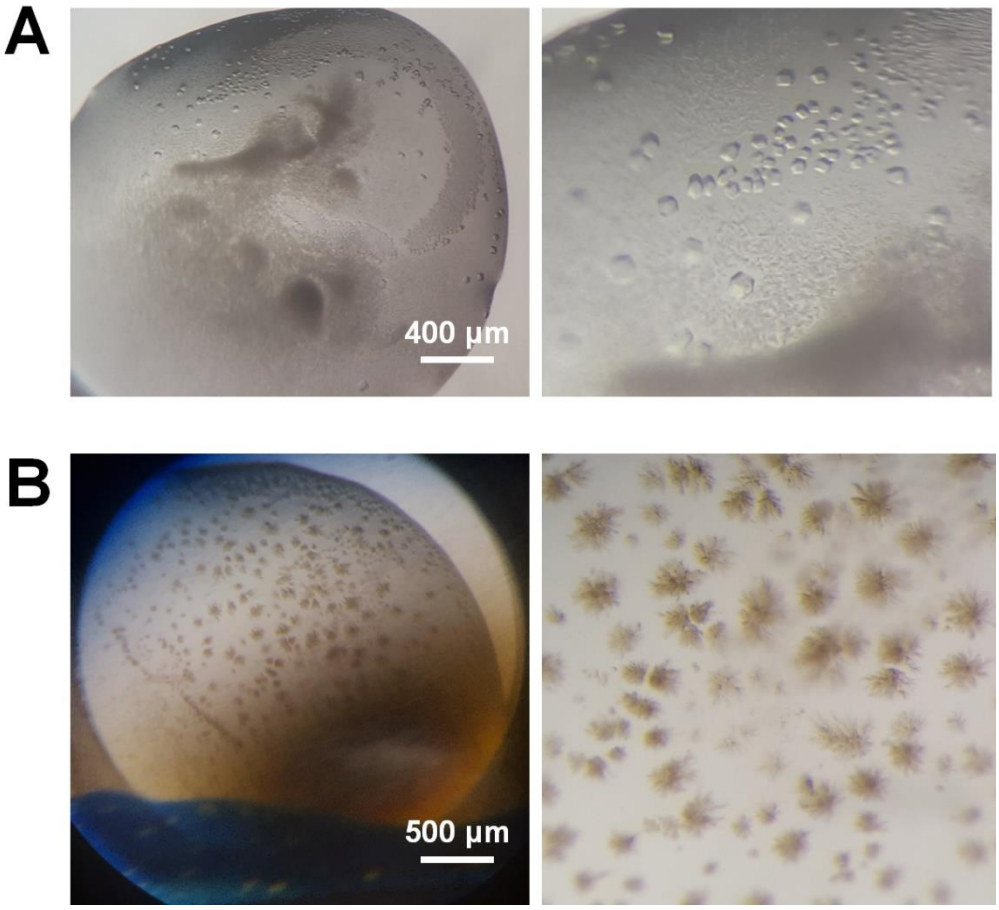


**Figure S1.** Photo of SavXI crystals grown under (**A**) 0.1 M Bis-Tris (pH 6.5) and 20% (w/v) polyethylene glycol monomethyl ether 5000 and (**B**) 0.1 M Bis-Tris (pH 6.5) and 45% (v/v) polypropylene glycol P400.

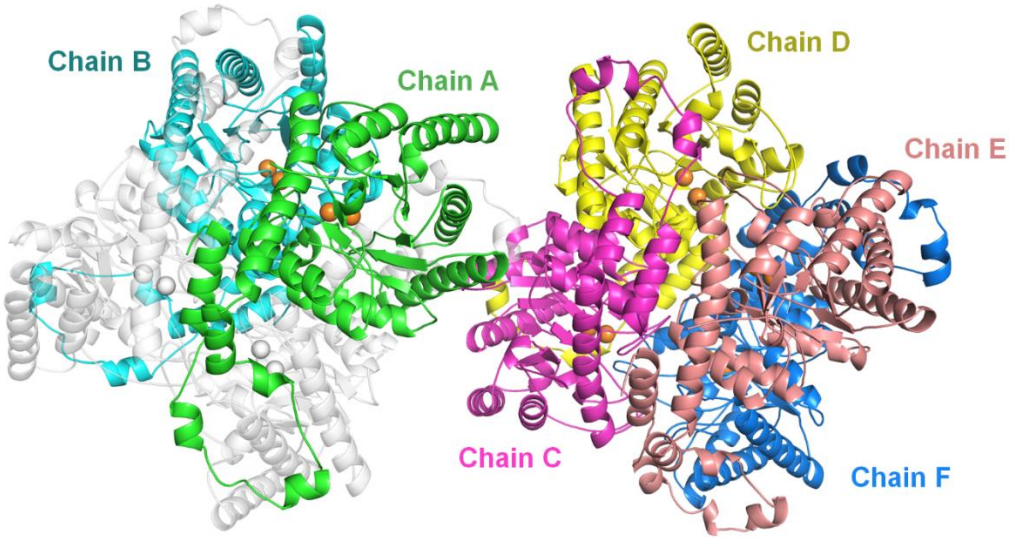


**Figure S2.** SavXI molecules (Chains A–F) in the asymmetric unit. Two SavXI molecules with symmetry coordinates in the crystal lattice are colored white.

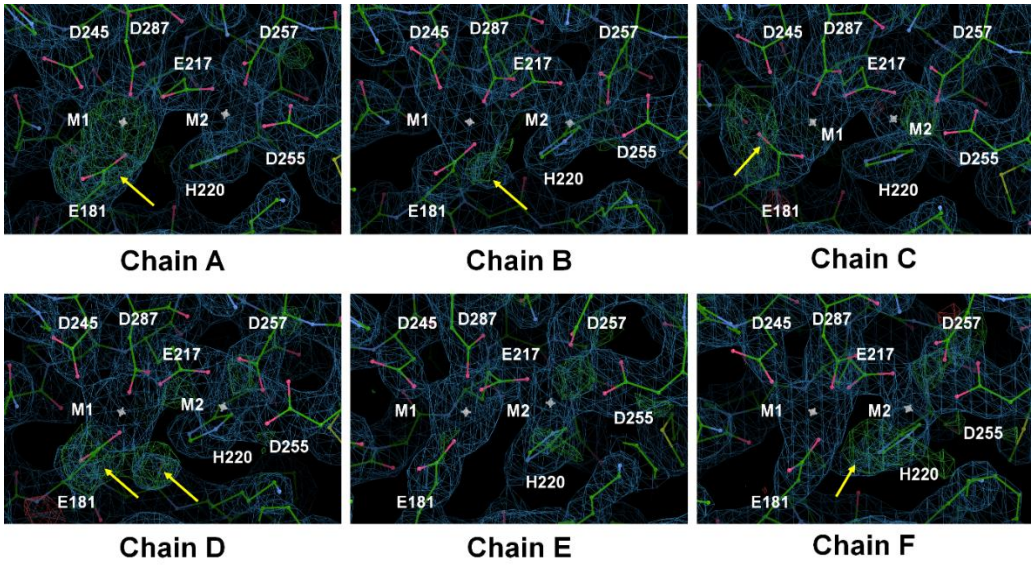


**Figure S3.** Unidentified 2Fo-Fc (blue mesh, 1σ) and Fo-Fc (green mesh, 3σ) electron density maps at the M1 site of SavXI.

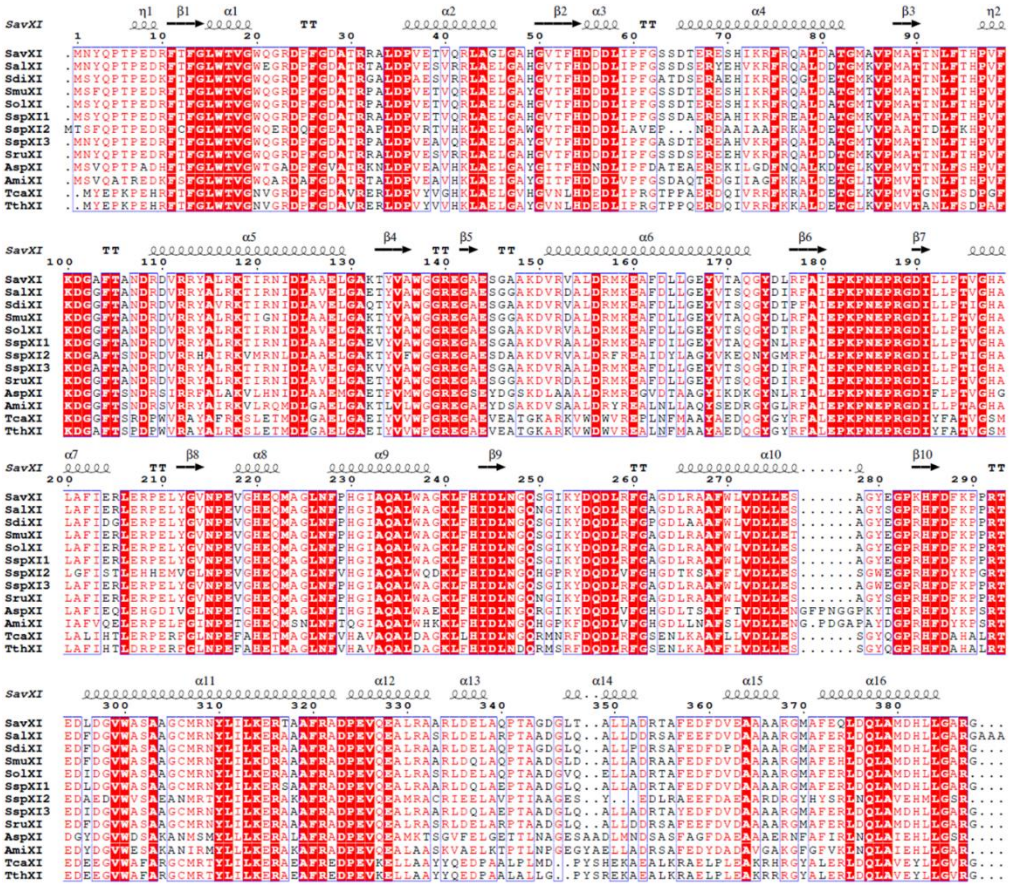

**Figure S4.** Structure-based amino acid sequence alignment of SavXI (UniProt: Q93HF3) with XI from *S. albus* (SalXI, P24299), *S. diastaticus* (SdiXI, P50910), *S. murinus* (SmuXI, P37031), *S. olivochromogenes* (SolXI, P15587), *Streptomyces* sp. F-1 (SspXI1, A0A1K2FKX8), *Streptomyces* sp. F-1 (SspXI2, A0A1K2FZ20), *Streptomyces* sp. SK (SspXI3, Q9ZAI3), *S. rubiginosus* (SruXI, P24300), *Arthrobacter* sp. strain NRRL B3728 (AspXI, P12070), *A. missouriensis* (AmiXI, P12851), *T. caldophilus* (TcaXI, P56681), and *T. thermophilus* HB8 (TthXI, P26997).

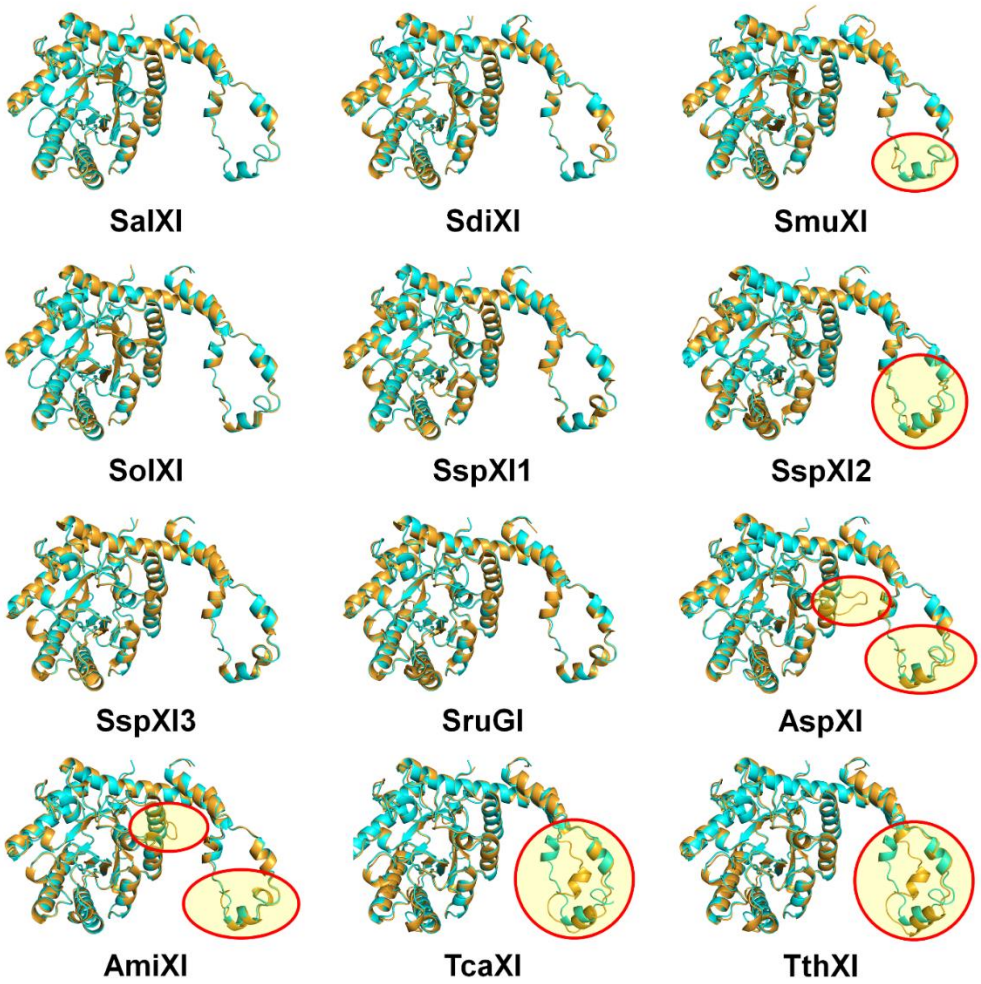


**Figure S5.** Superimposition of SavXI (cyan, Molecule A) with SalXI (PDB code: 6XIA), SdiXI (1CLK), SmuXI (1DXI), SolXI (1XYC), SspXI1 (6N99), SspXI2 (6N98), SspXI3 (4HHL), SruGI (5I7G), AspXI (1DID), AmiXI (1BHW), TcaXI (1BXC), and TthXI (1BXB). Significant regions with conformational differences are indicated by red circles.

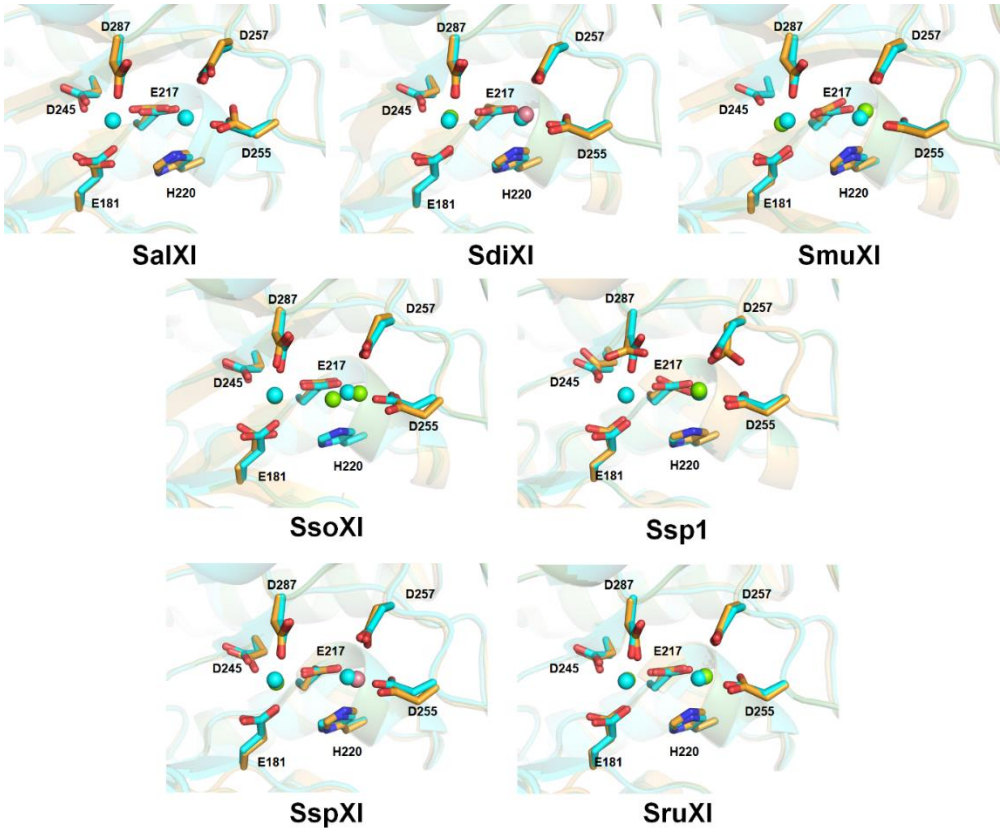


**Figure S6.** Superimposition of the metal binding site of SavXI (cyan) with SalXI (PDB code: 6XIA), SdiXI (1CLK), SmuXI (1DXI), SolXI (1XYC), SspXI1 (6N99), Ssp3XI (4HHL) and SruXI (5I7G).

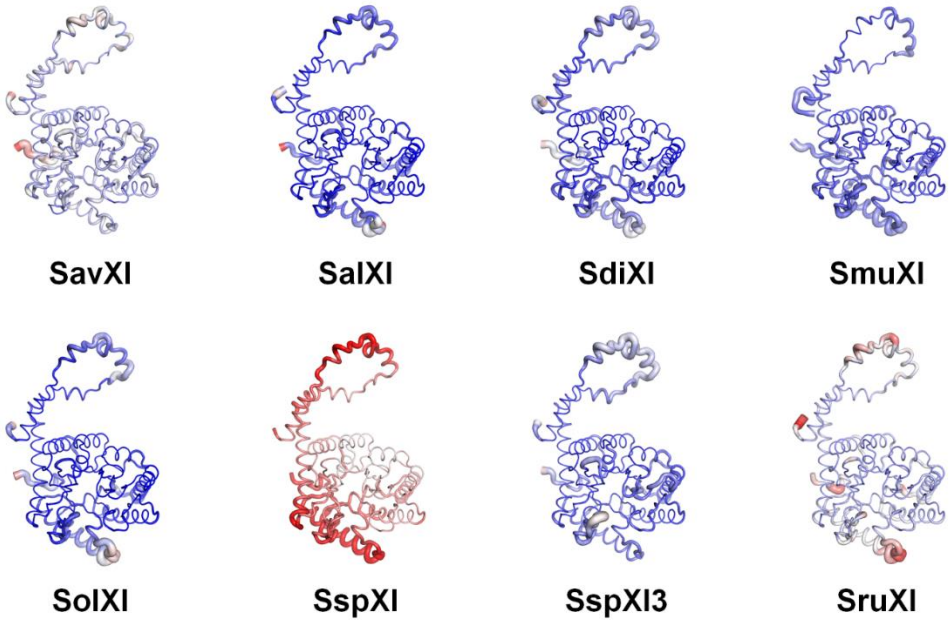


**Figure S7.** B-factor putty representaion of SavXI (cyan), SalXI (PDB code: 6XIA), SdiXI (1CLK), SmuXI (1DXI), SolXI (1XYC), SspXI1 (6N99), Ssp3XI (4HHL) and SruXI (5I7G).

**Table S1.** Interaction between Molecule A and B (Chain C and Chain D).

| **Hydrogen bonds** | | | | | |
|---|---|---|---|---|---|
| **Chain C Residue [atom]** | **Distance (Å)** | **Chain D Residue [atom]** | **Chain C Residue [atom]** | **Distance (Å)** | **Chain D Residue [atom]** |
| Asn107 [O] | 3.53 | Leu338 [N] | Leu338 [N] | 3.31 | Asn107 [O] |
| Asp108 [OD1] | 2.82 | Arg334 [NE] | Arg368 [NH1] | 3.76 | Asp108 [OD1] |
| Asp108 [OD1] | 2.63 | Arg334 [NH2] | Arg334 [NE] | 2.82 | Asp108 [OD1] |
| Asp110 [OD1] | 3.08 | Arg368 [NH2] | Trp237 [NE1] | 2.77 | Asp156 [OD1] |
| Asp156 [OD1] | 2.84 | Trp237 [NE1] | Gly344 [N] | 3.72 | Glu160 [OE2] |
| Glu160 [OE1] | 2.94 | Thr342 [N] | Thr342 [N] | 2.85 | Glu160 [OE2] |
| Glu160 [OE2] | 3.01 | Ala343 [N] | Ala343 [N] | 2.65 | Glu160 [OE2] |
| Glu160 [OE2] | 3.15 | Thr342 [N] | Asn227 [ND2] | 2.81 | Asp190 [OD1] |
| Asp190 [OD1] | 2.73 | Asn227 [ND2] | Gln234 [NE2] | 2.94 | His198 [O] |
| Thr195 [OG1] | 3.60 | Thr195 [OG1] | Arg205 [NH1] | 2.64 | Glu204 [OE1] |
| His198 [O] | 2.70 | Gln234 [NE2] | Asn107 [ND2] | 3.62 | Ala333 [O] |
| Ala238 [O] | 3.36 | Arg205 [NH1] | Arg112 [NH2] | 2.80 | Glu337 [O] |
| Ala333 [O] | 3.16 | Asn107 [ND2] | Arg109 [N] | 2.79 | Glu337 [OE2] |
| Glu337 [O] | 2.96 | Arg112 [NH2] | Arg157 [NH1] | 3.02 | Leu338 [O] |
| Glu337 [O] | 3.32 | Arg112 [NH1] | Arg112 [NH2] | 2.76 | Gln340 [O] |
| Glu337 [OE2] | 2.44 | Arg109 [N] | Arg109 [NE] | 2.70 | Pro341 [O] |
| Leu338 [O] | 2.90 | Arg157 [NH1] | Arg109 [NH2] | 3.14 | Thr342 [O] |
| Gln340 [O] | 2.67 | Arg112 [NH2] | Arg113 [NH1] | 2.66 | Thr342 [O] |
| Thr342 [O] | 2.92 | Arg113 [NH1] | Arg112 [NE] | 2.72 | Thr342 [OG1] |
| Thr342 [O] | 3.31 | Arg109 [NE] | Arg113 [NH1] | 3.08 | Asp345 [OD1] |
| Thr342 [OG1] | 2.86 | Arg112 [NE] | Arg113 [NH2] | 2.71 | Asp345 [OD1] |
| Asp345 [OD1] | 3.22 | Arg113 [NH1] | Arg117 [NH1] | 2.86 | Leu350 [O] |
| Asp345 [OD1] | 3.30 | Arg113 [NH2] | Arg117 [NH1] | 2.46 | Asp353 [O] |
| Leu350 [O] | 2.73 | Arg117 [NH1] | Arg113 [NH2] | 3.66 | Asp353 [OD1] |
| Asp353 [O] | 2.71 | Arg117 [NH1] | Arg117 [NH2] | 3.55 | Glu358 [OE1] |
| Asp353 [OD1] | 3.01 | Arg113 [NH2] | His96 [NE2] | 3.19 | Glu363 [OE2] |
| Glu358 [OE2] | 3.26 | Arg117 [NH2] | Lys100 [NZ] | 3.32 | Ala366 [O] |
| Glu363 [OE2] | 3.10 | His96 [NE2] | Lys100 [NZ] | 2.59 | Arg368 [O] |
| Arg368 [O] | 2.96 | Lys100 [NZ] | | | |
| **Salt bridges** | | | | | |
| **Chain C Residue [atom]** | **Distance (Å)** | **Chain D Residue [atom]** | **Chain C Residue [atom]** | **Distance (Å)** | **Chain D Residue [atom]** |
| Asp108 [OD1] | 2.82 | Arg334 [NE] | Arg368 [NH1] | 3.76 | Asp108 [OD1] |
| Asp108 [OD1] | 3.66 | Arg334 [NH2] | Arg334 [NE] | 2.82 | Asp108 [OD1] |
| Asp108 [OD1] | 3.39 | Arg334 [NE] | Arg334 [NH2] | 3.21 | Asp108 [OD1] |
| Asp108 [OD1] | 2.63 | Arg334 [NH2] | Arg334 [NH2] | 3.89 | Asp108 [OD1] |
| Asp110 [OD1] | 3.08 | Arg368 [NH2] | Arg205 [NH1] | 2.64 | Glu204 [OE1] |
| Glu204 [OE1] | 3.44 | Arg205 [NH2] | Arg205 [NH2] | 3.08 | Glu204 [OE1] |
| Glu204 [OE2] | 3.54 | Arg205 [NH2] | Arg205 [NH2] | 3.03 | Glu204 [OE2] |
| Asp345 [OD1] | 3.22 | Arg113 [NH1] | Arg113 [NH1] | 3.08 | Asp345 [OD1] |
| Asp345 [OD1] | 3.64 | Arg113 [NH1] | Arg113 [NH2] | 3.94 | Asp345 [OD1] |
| Asp345 [OD1] | 3.30 | Arg113 [NH2] | Arg113 [NH1] | 3.25 | Asp345 [OD1] |
| Asp353 [OD1] | 3.95 | Arg113 [NE] | Arg113 [NH2] | 2.71 | Asp345 [OD1] |
| Asp353 [OD1] | 3.01 | Arg113 [NH2] | Arg113 [NE] | 3.96 | Asp353 [OD1] |
| Glu358 [OE2] | 3.26 | Arg117 [NH2] | Arg113 [NH2] | 3.66 | Asp353 [OD1] |
| Glu363 [OE1] | 3.93 | His96 [NE2] | Arg117 [NH2] | 3.55 | Glu358 [OE1] |
| Glu363 [OE2] | 3.10 | His96 [NE2] | His96 [NE2] | 3.19 | Glu363 [OE2] |

**Table S2.** Interaction between Molecule A and D (Chain C and Chain F).

| Hydrogen bonds | | | | | |
|---|---|---|---|---|---|
| **Chain C** | **Distance** | **Chain E** | **Chain E** | **Distance** | **Chain E** |
| **Residue [atom]** | **(Å)** | **Residue [atom]** | **Residue [atom]** | **(Å)** | **Residue [atom]** |
| Thr293 [O] | 3.13 | Met370 [N] | Phe372 [N] | 3.13 | Glu294 [OE1] |
| Glu294 [OE1] | 3.26 | Phe372 [N] | Phe372 [N] | 3.34 | Glu294 [OE2] |
| Glu294 [OE2] | 3.48 | Phe372 [N] | Gln377 [NE2] | 2.77 | Asn309 [OD1] |
| Asn309 [OD1] | 2.66 | Gln377 [NE2] | Ser302 [OG ] | 2.81 | Glu373 [OE1] |
| Glu373 [OE2] | 3.35 | Ser302 [OG] | Arg259 [NH1] | 3.00 | Glu373 [OE1] |
| Glu373 [OE2] | 2.66 | Arg259 [NH1] | Arg259 [NH2] | 2.85 | Asp376 [OD1] |
| Asp376 [OD1] | 2.23 | Arg259 [NH2] | Arg259 [NH1] | 2.97 | Asp376 [OD1] |
| Gln377 [OE1] | 3.10 | Arg259 [NE] | Arg259 [NE] | 2.65 | Gln377 [OE1] |
| Gln377 [OE1] | 3.32 | Asn309 [ND2] | Asn309 [ND2] | 3.32 | Gln377 [OE1] |
| Asp381 [OD1] | 2.93 | Arg308 [NE] | Asn309 [ND2] | 3.82 | Met380 [SD] |
| Leu384 [O] | 2.79 | Arg316 [NH1] | Arg308 [NE] | 3.23 | Asp381 [OD1] |
| Ala386 [O] | 2.77 | Arg308 [NH2] | Arg316 [NH1] | 2.61 | Leu384 [O] |
| Met370 [N] | 2.97 | Thr293 [O] | Arg308 [NH2] | 2.55 | Ala386 [O] |
| **Salt bridges** | | | | | |
| **Chain C** | **Distance** | **Chain E** | **Chain C** | **Distance** | **Chain E** |
| **Residue [atom]** | **(Å)** | **Residue [atom]** | **Residue [atom]** | **(Å)** | **Residue [atom]** |
| Glu373 [OE1] | 3.80 | Arg259 [NH1] | Arg259 [NH1] | 3.00 | Glu373 [OE1] |
| Glu373 [OE2] | 2.66 | Arg259 [NH1] | Arg259 [NH1] | 3.59 | Glu373 [OE2] |
| Asp376 [OD1] | 3.94 | Arg259 [NH1] | Arg259 [NH2] | 2.85 | Asp376 [OD1] |
| Asp376 [OD1] | 2.23 | Arg259 [NH2] | Arg259 [NH1] | 2.97 | Asp376 [OD1] |
| Asp381 [OD1] | 2.93 | Arg308 [NE] | Arg308 [NE] | 3.23 | Asp381 [OD1] |
| Asp381 [OD1] | 3.36 | Arg308 [NH2] | Arg308 [NH2] | 3.81 | Asp381 [OD1] |
| Asp381 [OD1] | 3.35 | Arg308 [NE] | Arg308 [NE] | 3.43 | Asp381 [OD1] |
| Asp381 [OD1] | 3.62 | Arg308 [NH2] | Arg308 [NH2] | 3.84 | Asp381 [OD1] |

**Table S3.** Interaction between Molecule A and B (Chain C and Chain D).

| Hydrogen bonds | | | | | |
|---|---|---|---|---|---|
| **Chain C** | **Distance** | **Chain F** | **Chain C** | **Distance** | **Chain F** |
| **Residue [atom]** | **(Å)** | **Residue [atom]** | **Residue [atom]** | **(Å)** | **Residue [atom]** |
| Thr95 [O] | 2.89 | Asp28 [N] | Arg140 [NH2] | 3.05 | Asp24 [OD1] |
| Phe 26 [O] | 3.56 | Thr95 [OG1] | Arg140 [NH1] | 2.73 | Asp24 [OD1] |
| Thr293 [O] | 3.37 | Lys100 [NZ] | Thr95 [OG1] | 2.86 | Phe26 [O] |
| Asp24 [OD1] | 2.60 | Arg140 [NH1] | Asp28 [N] | 3.16 | Thr95 [O] |
| Asp24 [OD1] | 3.09 | Arg140 [NH2] | Arg292 [NH2] | 3.85 | Thr95 [O] |
| Phe 26 [O] | 2.75 | Arg140 [NH2] | Tyr254 [OH] | 3.35 | Pro187 [O] |
| Asp190 [OD1] | 3.50 | Lys253 [NZ] | Lys253 [NZ] | 2.73 | Gly189 [O] |
| Gly189 [O] | 3.25 | Lys253 [NZ] | Lys253 [NZ] | 3.49 | Asp190 [OD1] |
| Pro187 [O] | 3.68 | Tyr254 [OH] | Lys100 [NZ] | 3.41 | Thr293 [O] |
| **Salt bridges** | | | | | |
| **Chain C** | **Distance** | **Chain F** | **Chain C** | **Distance** | **Chain F** |
| **Residue [atom]** | **(Å)** | **Residue [atom]** | **Residue [atom]** | **(Å)** | **Residue [atom]** |
| Asp24 [OD1] | 3.97 | Arg140 [NH1] | Arg140 [NH2] | 3.05 | Asp24 [OD1] |
| Asp24 [OD1] | 2.60 | Arg140 [NH1] | Arg140 [NH1] | 2.73 | Asp24 [OD1] |
| Asp24 [OD1] | 3.09 | Arg140 [NH2] | Arg140 [NH2] | 3.08 | Asp24 [OD1] |
| Asp24 [OD1] | 3.08 | Arg140 [NH2] | Lys253[ NZ ] | 3.49 | Asp190 [OD1] |
| Asp190 [OD1] | 3.50 | Lys253 [NZ] | | | |

**Table S4.** Distance between metal-binding sites and key residues.

| Metal | Residue | Molecule | | | | | |
|---|---|---|---|---|---|---|---|
| | | **A** | **B** | **C** | **D** | **E** | **F** |
| **M1** | Glu181 | 1.90 | 1.84 | 2.14 | 1.95 | 1.76 | 2.07 |
| | Glu217 | 2.45 | 2.55 | 2.28 | 2.31 | 1.69 | 2.29 |
| | Asp245 | 2.62 | 2.18 | 3.08 | 2.69 | 2.77 | 2.30 |
| | Asp287 | 1.80 | 2.06 | 1.73 | 1.78 | 2.21 | 2.07 |
| | His45 | 6.85 | 7.08 | 6.71 | 6.95 | 7.11 | 6.92 |
| **M2** | Glu217 | 1.89 | 2.56 | 2.45 | 1.95 | 2.31 | 2.71 |
| | His220 | 2.83 | 2.80 | 3.46 | 2.55 | 2.59 | 2.91 |
| | Asp255 | 1.86 | 2.57 | 2.47 | 2.10 | 2.52 | 2.39 |
| | Asp257 | 2.55 | 2.65 | 2.79 | 2.67 | 3.16 | 3.17 |
| | Lys183 | 4.62 | 4.54 | 4.52 | 4.59 | 4.15 | 4.68 |

**Table S5.** Coordination angle for metal-binding sites.

| Metal | Residue | Molecule (°) | | | | | |
|---|---|---|---|---|---|---|---|
| | | **A** | **B** | **C** | **D** | **E** | **F** |
| **M1** | Glu181-M1-Asp245 | 104.7 | 97.8 | 99.6 | 85.7 | 80.2 | 108.6 |
| | Glu181-M1-Glu217 | 90.0 | 76.7 | 96.8 | 84.5 | 97.1 | 77.3 |
| | Asp245-M1-Asp287 | 88.2 | 100.6 | 72.3 | 99.3 | 88.9 | 97.9 |
| | Glu217-M1-Asp287 | 84.3 | 87.7 | 89.4 | 87.0 | 93.0 | 81.3 |
| **M2** | Glu217-M2-His220 | 80.3 | 61.1 | 59.4 | 78.6 | 66.5 | 62.2 |
| | Glu217-M2-Asp257 | 79.7 | 69.5 | 70.8 | 74.3 | 64.6 | 65.0 |
| | His220-M2-Asp255 | 94.9 | 75.3 | 69.2 | 94.7 | 81.2 | 76.8 |
| | Asp255-M2-Asp257 | 82.6 | 75.7 | 70.6 | 87.8 | 61.0 | 56.2 |