Acta Crystallographica Section D

Volume 70 (2014)

Supporting information for article:

Structural basis for the recognition of muramyltripeptide by *Helicobacter pylori* Csd4, a D,L-carboxypeptidase controlling the helical cell shape

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Table S1 Statistics on data collection and refinement

A. Data collection		
Data set	Zn1	Zn2
Space group	$P2_12_12_1$	$P2_12_12_1$
Unit cell lengths (Å)	45.5, 66.1, 142.0	53.3, 66.9, 144.8
X-ray wavelength (Å)	1.2823	1.2823
Resolution range[a,b] (Å)	50.0–2.60 (2.64–2.60)	50.0–2.40 (2.44–2.40)
No. total reflections[a,b]	176,100 (8,669)	315,647 (15,050)
No. unique reflections[a,b]	27,639 (1,376)	38,851 (1,905)
Completeness[a,b] (%)	100.0 (100.0)	99.8 (99.8)
$\langle I \rangle / \langle \sigma_I \rangle$ [a,b]	23.2 (3.5)	43.2 (10.1)
Wilson B factor (Å^2)	55.9	46.6
R_{merge}[a–c] (%)	12.2 (73.7)	13.0 (60.1)
B. Model refinement		
PDB code	4Q6P	4Q6Q
Resolution range (Å)	30.0–2.62	30.0–2.40
R_{work} / R_{free}[d] (%)	18.4 / 25.1	16.7 / 20.5
No. of non-hydrogen atoms / average B-factor (Å^2)		
Total	3,533 / 45.2	3,606 / 33.0
Protein	3,366 / 45.2	3,366 / 32.7
Water oxygen	145 / 42.6	223 / 36.9
mDAP	13 / 58.7	13 / 36.9
Zinc ion	8 / 79.1	2 / 67.9
Calcium ion	1 / 49.5	2 / 40.7
R.m.s. deviations from ideal geometry		
Bond lengths (Å)	0.010	0.009

Bond angles (°)	1.30	1.34
Ramachandran plot (%)[e]		
Favored / Outliers	94.9 / 0.0	96.1 / 0.0
Poor rotamers (%)[e]	3.25	0.54

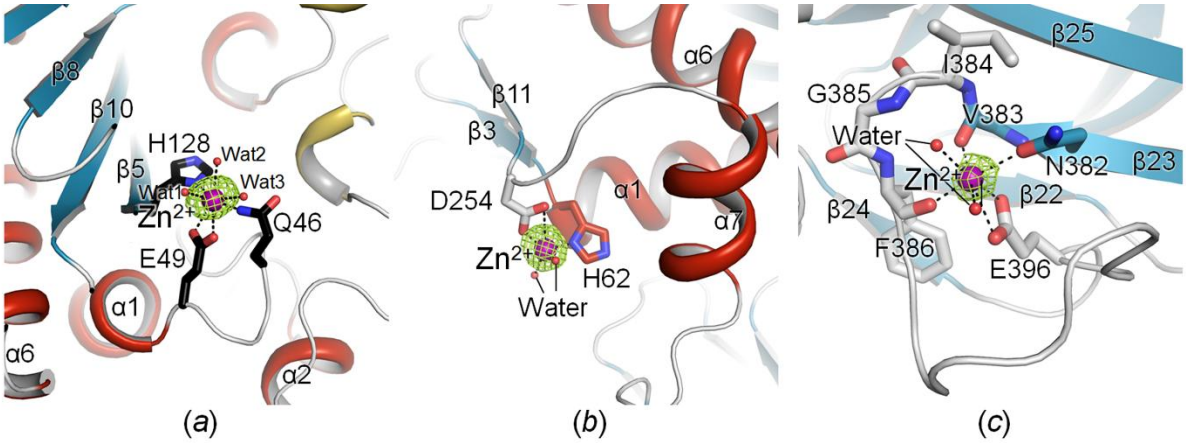
[a] Values in parentheses refer to the highest resolution shell.

[b] Friedel pairs were treated as separate observations.

[c] $R_{merge} = \Sigma_h\Sigma_i \mid I(h)_i - \langle I(h)\rangle \mid / \Sigma_h\Sigma_i I(h)_i$, where $I(h)$ is the intensity of reflection h, Σ_h is the sum over all reflections, and Σ_i is the sum over i measurements of reflection h.

[d] $R_{work} = \Sigma \mid |F_{obs}| - |F_{calc}| \mid / \Sigma |F_{obs}|$, where R_{free} is calculated for a randomly chosen 5% of reflections, which were not used for structure refinement and R_{work} is calculated for the remaining reflections.

[e] Values obtained using *MolProbity*.



~~Figure S1~~ Anomalous difference Fourier maps (contoured at 5σ and colored in green) for Zn^{2+}-replaced metal sites. (*a*) The active site metal ion for D,L-carboxypeptidation by the CPase domain from the Zn1 data set (Table S1). (*b*) The metal ion that stabilizes the β11-α7 loop in the CPase domain from the Zn1 data set (Table S1). (*c*) The metal ion in the calcium-binding site of the C-terminal Ig-like domain from the Zn2 data set (Table S1). The ribbon diagram of metal-binding sites are colored as in Figure 1(*a*).

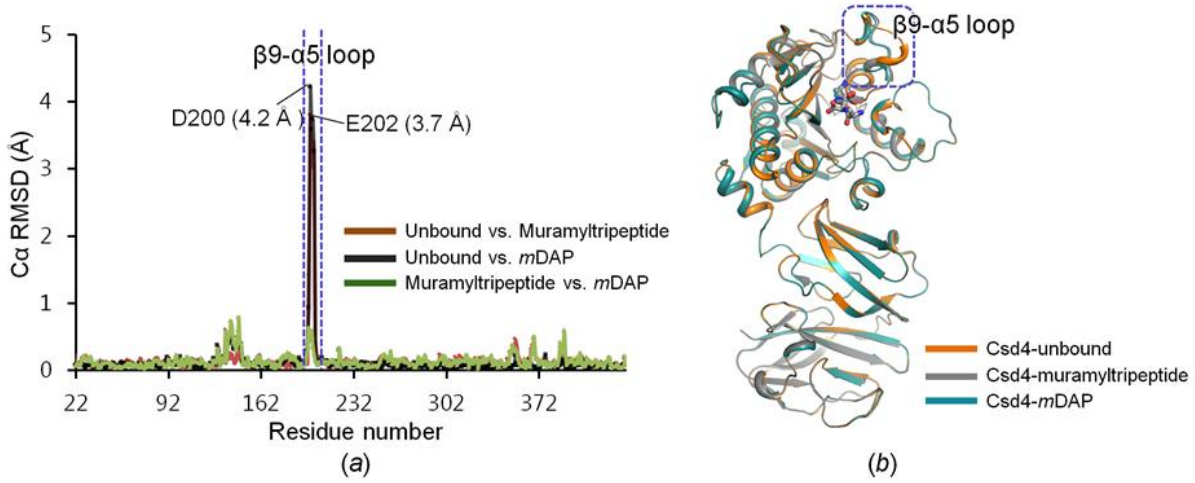


Figure S2 Comparisons of three Csd4 structures. (*a*) A plot of the Cα r.m.s. deviations for pairwise comparisons among Csd4-unbound, Csd4-muramyltripeptide, and Csd4-*m*DAP structures. (*b*) A superposition of three Csd4 models. The Csd4-unbound, Csd4-muramyltripeptide, and Csd4-*m*DAP structures are colored in orange, gray, and cyan, respectively. The β9-α5 loop region is highlighted by the dotted blue box, showing a significant conformational change.

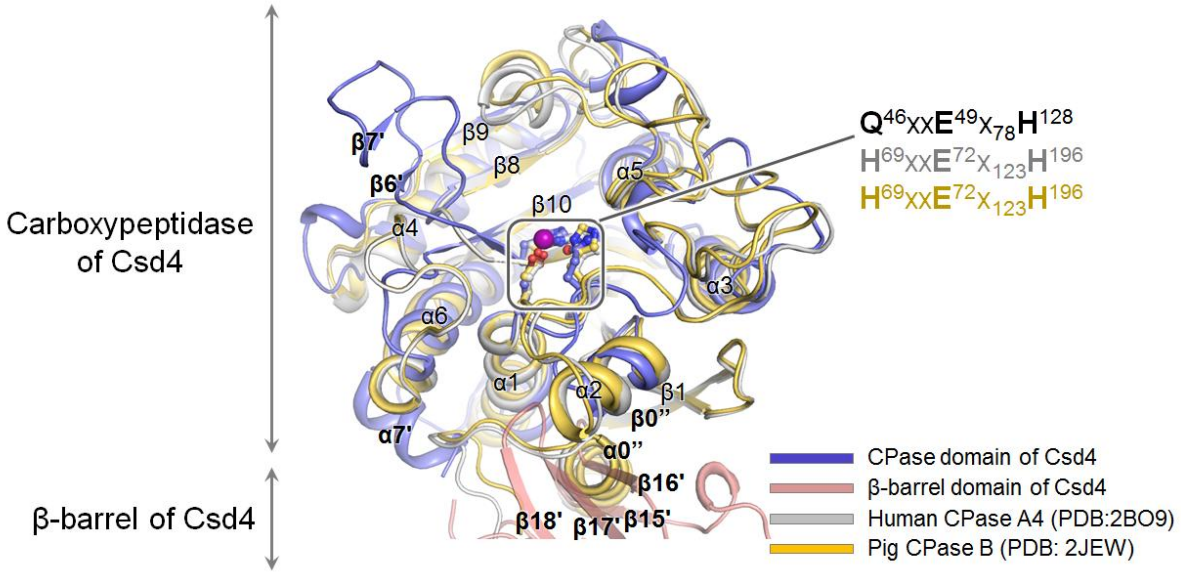


Figure S3 Superposition of carboxypeptidase (CPase) domains in three M14 metallopeptidases. The CPase domains of *H. pylori* Csd4 (bluish-violet), human CPase A4 (gray; PDB code: 2BO9), and pig CPase B (yellow; PDB: 2JEW) are superimposed and shown in ribbon diagrams. The β-barrel domain of *H. pylori* Csd4 is colored in pink. The regions present only in *H. pylori* Csd4 are labeled with a single quote (α7' and β15–β18'), whereas the regions absent only in *H. pylori* Csd4 are labeled with a double quote (α0" and β0").