Supplemental Data

Supplemental Table 1. Oligonucleotide primers used in this study.

Primer	Sequence
IMP-C221X-top	5' - GAAAATATTATTCGGTGGTNNNTTTATTAAACCGTACGG - 3'
IMP-C221X-bot	5' - CCGTACGGTTTAATAAANNNACCACCGAATAATATTTTC - 3'
pTP123 top	5' - CATCCGGCTCGTATAATGTGTGGAATTGTG - 3'
pTP123 bot	5' - CAGACCGCTTCTGCGTTCTGATTTAATCTG - 3'
S121X IMP-1 F	5' - CCTCTCATTTTCATAGCGACNNNACGGG - 3'
S121X IMP-1 R	5' - CCCGTNNNGTCGCTATGAAAATGAGAGG - 3'

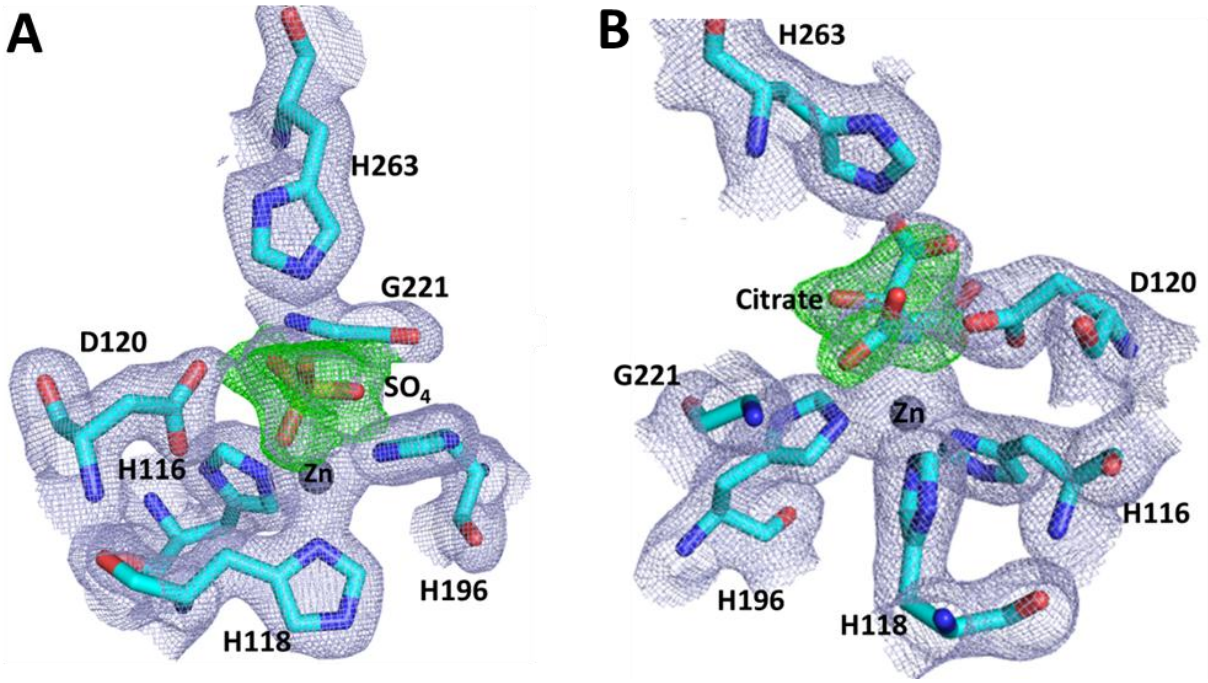
Supplemental Table 2. IMP-1 β-lactamase zinc content as determined by the PAR assay.

Sample	Raw zinc concentration (□M)	Relative zinc content (per enzyme)
Buffer	0.16 ± 0.02	N/A
WT	10.2 ± 0.01	2.04
WT (DPA)	5.3 ± 0.10	1.06
C221G	5.0 ± 0.10	1.01
C221G (DPA)	4.8 ± 0.10	0.96

Supplemental Table 3. Data collection and refinement statistics for the IMP-1 C221G structures.

Parameter	Sulfate-bound	Citrate-bound
Space Group	$P2_12_12_1$	$P2_12_12_1$
Cell dimensions *a,b,c* (Å)	50.02, 60.5, 84.29	50.02, 59.77, 83.55
Cell dimensions α, β, γ (°)	90, 90, 90	90, 90, 90
Resolution Range (Å)	30.25-1.74 (1.81-1.74)*	25.24-2.00 (2.07-2.00)
Molecules per asymmetric unit	1	1
Number of unique reflections	26059	15635
Completeness (%)	97.5 (93.4)	89.2 (93.0)
Redundancy	3.62 (3.43)	4.04 (3.86)
I/σ	13.7 (2.3)	11.2 (3.1)
R_{merge} (%)	3.6 (30.6)	5.7 (31.1)
R_{work} (%) / R_{free}(%)	19.6/22.6	19.9/24.9
Number of protein atoms	1946	1852
R.m.s. deviations		
Bond lengths (Å)	0.013	0.015
Bond angles (°)	0.987	1.044
Ramachandran analysis		
Preferred (%)	100	100
Allowed (%)	0	0

* Values in parentheses are for the highest resolution shell.



Supplemental Figure 1. Structure of IMP-1 C221G enzyme with electron density.
Electron density maps around the Zn binding sites: the 2Fo –Fc electron density maps (blue) contoured at 1 σ for the residues composing the zinc coordinating sites. The Fo-Fc electron density maps (green) countered at 2.8 σ used for modeling the sulfate ion (A) and citrate ion (B) is also represented. The residues and ions are labeled and shown as stick representation with carbon atoms in cyan, nitrogen in dark blue, oxygen in red and sulfur in yellow. Zn ion is represented as grey sphere.