Supplementary Table 1. Effect of metals on ZinT thermal denaturation profiles tested by differential scanning fluorimetry.

| Metal | Counter-ion | [Metal] (µM) | Δ <i>T</i> m (°C) | |
|--------------------|--------------------------------------|------------------|--|--------------------------------------|
| | | | 1ºC/min | 1°C/10 sec |
| Ba ²⁺ | Cl | 10 | | $+0.2\pm0.6$ |
| Ca ²⁺ | Cl | 10 500 | | +0.4±0.7 +0.5+0.1 |
| Cd^{2+} | Cl | 10 500 | | +4.5±0.2 +7.0±0.3 |
| Co ²⁺ | Cl | 10 500 | | +4.2±0.0 +8.3±0.1 |
| Cr ³⁺ | Cl | 10 500 | | +0.1±0.3 +0.7±0.3 |
| Cu ²⁺ | Cl | 10 100 500 | +9.6±0.4 +5.7±0.1 +4.2+0.5 | +5.2±0.2 |
| Fe ³⁺ | Cl | 10 500 | 17.2±0.3 | +0.6±0.3 +0.5±0.2 |
| Hg ²⁺ | Cl | 10 100 500 | $+6.1\pm0.3$ +9.7 ±0.1 +7.4 ±0.1 | |
| Mg ²⁺ | Cl | 10 500 | | +0.7±0.5 +0.6±0.1 |
| Mn ²⁺ | Cl | 10 100 500 | $+2.7\pm0.0$ +2.8±0.1 +2.6±0.1 | +0.5±0.0 +0.9+0.0 |
| Ni ²⁺ | SO ₄ ²⁻ | 10 100 500 | $+8.5\pm0.3$ +11.3±0.1 | +4.7±0.3 |
| Pr ³⁺ | CH ₃ COO ⁻ | 10 500 | +10.0±0.1 | $+8.7\pm0.0$ +0.5±0.7 +0.6±0.1 |
| Sr ²⁺ | Cl | 10 500 | | +0.6±0.0 +8.7±0.0 |
| Y ³⁺ | Cl | 10 500 | | +0.5±0.3 -0.1±0.1 |
| Zn ²⁺ | SO4 ²⁻ | 10 100 500 | $+13.2\pm0.2+13.7\pm0.1+2.4\pm0.4+12.4\pm0.2$ | +10.8±0.3 +11.8±0.3 |

| Metal | Concentration (µM) |
|-----------------------------|--|
| Cobalt (Co ²⁺) | 100 250* 400 500* 1000 |
| Mercury (Hg ²⁺) | 1 2.5 10 15* 20* 25 40 50 |
| Cadmium (Cd ²⁺) | 25 100 250 300* 500 * |
| Copper (Cu ²⁺) | 100 500 1000 2000 3000* 5000 |
| Nickel (Ni ²⁺) | 100 500 1000* 1250 2000 |
| Zinc (Zn ²⁺) | 100 200 400 500 600* 800* 1000 |

Supplementary Table 2 - Metal concentrations supplemented into growth medium of $\Delta zinT$ and $\Delta galT$ Escherichia coli strains.

* - metal concentrations presented in metal sensitivity assays (Figures 2-4 and Supplementary Figures S2-S4).

| Supplementary | Table 3. Data | collection and | processing | statistics |
|---------------|---------------|----------------|------------|------------|
|---------------|---------------|----------------|------------|------------|

| Beamline | ESRF ID 29 | | |
|------------------------------------|---------------------------|--|--|
| Detector | PILATUS 6M | | |
| Wavelength (Å) | 1.0000 | | |
| Data Processing | XDS | | |
| Space Group | $P 4_1 2_1 2$ | | |
| Unit cell parameters (Å) | a=62.01, c=149.72 | | |
| Resolution (Å) | 45.8 - 1.79 (1.85 - 1.79) | | |
| Nr. Observations | 191493 (11611) | | |
| Unique reflections | 28379 (2634) | | |
| Completeness (%) | 99.6 (97.0) | | |
| Multiplicity | 6.7 (4.4) | | |
| R-merge $(\%)^{a}$ | 7.4 (57.6) | | |
| R-pim (%) ^b | 3.0 (27.3) | | |
| R-meas (%) c | 8.0 (64.3) | | |
| <i o(i)=""></i> | 15.6 (1.7) | | |
| $\operatorname{CC}^{\frac{1}{2}d}$ | 0.999 (0.824) | | |
| Wilson Plot B | 24.6 | | |
| Z ^e | 2 | | |
| \mathbf{V}_{m} | 2.3 | | |
| Estimated Solvent Content (%) | 46 | | |

^{*a*} R-merge = merging R-factor, $(\Sigma_{hkl} \Sigma_i | I_i(hkl) - \langle I(hkl) \rangle) / (\Sigma_{hkl} \Sigma_i I(hkl)) \times 100\%$.

^{*b*} R-pim = precision independent R-factor, $\Sigma_{hkl} [1/(N_{hkl}-1)]^{1/2} \Sigma_i |I_i(hkl) - \langle I(hkl) \rangle | \Sigma_{hkl} \Sigma_i I_i (hkl)$, where *I* is the observed intensity, $\langle I \rangle$ is the average intensity of multiple observations from symmetry-related reflections, and N_{hkl} is their redundancy. (Diederichs and Karplus, 1997);

^c R-meas = redundancy independent R-factor, $\Sigma_{h} [N_{hkl}/(N_{hkl}-1)]^{1/2} \Sigma_{i} |I_{i}(hkl) - \langle I(hkl) \rangle | / \Sigma_{hkl} \Sigma_{i} I_{i}(hkl) \times 100\%$. (Diederichs and Karplus, 1997);

^d CC^{1/2} is the correlation coefficient between two randomly calculated half-sets (Karplus et al, 2012)

^e Nr. monomers in the asymmetric unit according to Matthews coefficient (Matthews, 1968)

References

(Diederichs and Karplus, 1997) - Diederichs, Kay, and P. Andrew Karplus. "Improved R-factors for diffraction data analysis in macromolecular crystallography." *Nature structural biology* 1997, 4(4): 269-275.

(Karplus et al, 2012) - Karplus, P. Andrew, and Kay Diederichs. "Linking crystallographic model and data quality." *Science* 2012, 336(6084): 1030-1033.

(Matthews, 1968) - Matthews, Brian W. "Solvent content of protein crystals." *Journal of molecular biology* 1968, 33(2): 491-497.

| Resolution limits (Å) | 47.75 - 1.79 (1.85 - 1.79) |
|--|----------------------------|
| R-factor (%) a | 17.4 (26.7) |
| nr.reflections | 28294 (2536) |
| Free R-factor (%) ^b | 20.8 (32.6) |
| nr. reflections | 1433 (141) |
| Overall coordinate error estimate (Å) c | 0.17 |
| Model composition | |
| non-hydrogen protein atoms | 1775 |
| Nº mol in asymmetric unit | 1 |
| Zinc atoms | 8 |
| Solvent molecules | 181 |
| Model r.m.s. deviations from ideality | |
| Bond lengths (Å) | 0.012 |
| Bond angles (°) | 1.318 |
| Chiral centers (Å ³) | 0.051 |
| Planar groups (Å) | 0.006 |
| Ramachandran plot statistics. Residues in: | |
| most favored regions (%) | 97.4 |
| allowed regions (%) | 2.6 |
| disallowed regions (%) | 0 |
| Rotamers ouliers (%) | 2.23 |
| C^{β} outliers | 0 |
| Clash score | 1.3 |
| Mean B values $(\mathring{A}^2)^d$ | Chain A |
| protein main-chain | 31.92 |
| protein side-chain | 40.34 |
| Zinc (acetate) | 43.03 (38.17) |
| solvent | 38.36 |

Supplementary Table 4. Final refinement statistics.

^{*a*} R-factor = $\Sigma_{hkl} ||F_o| - |F_c|| / \Sigma_{hkl} |F_o|$, where $|F_o|$ and $|F_c|$ are the observed and calculated structure factor amplitudes, respectively; ^{*b*} Free R-factor is the cross-validation R-factor computed from a randomly chosen subset of 5% of the total number of reflections, which were not used during the refinement.; ^{*c*} Maximum-likelihood estimate with PHENIX; ^{*d*} Calculated from equivalent isotropic B values, including the TLS contribution for the protein atoms.