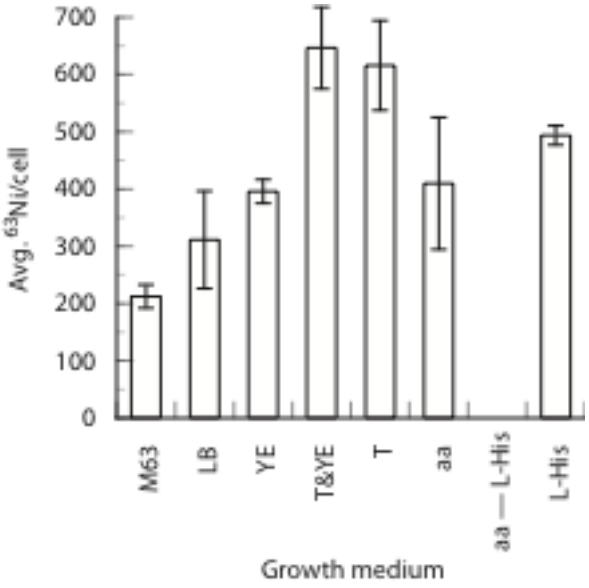
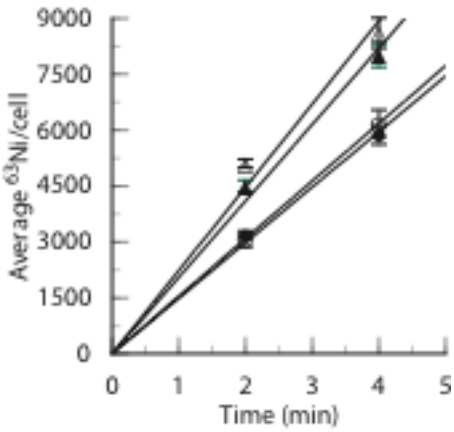
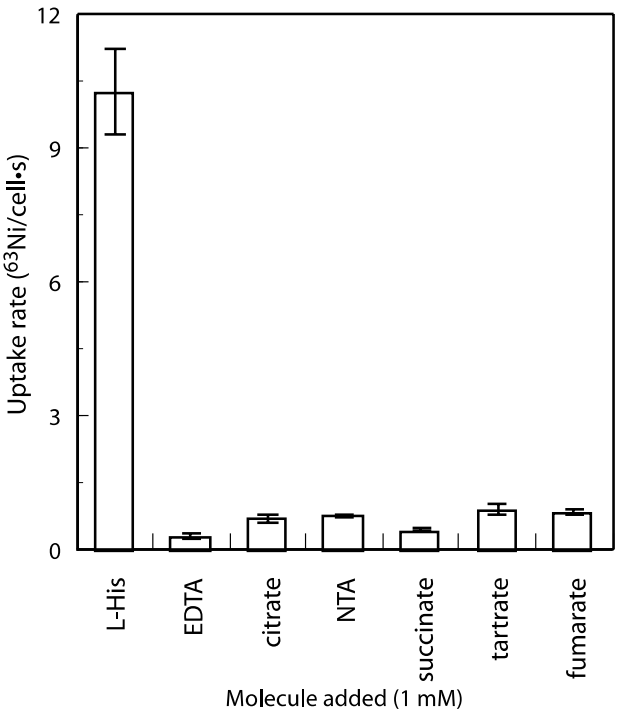


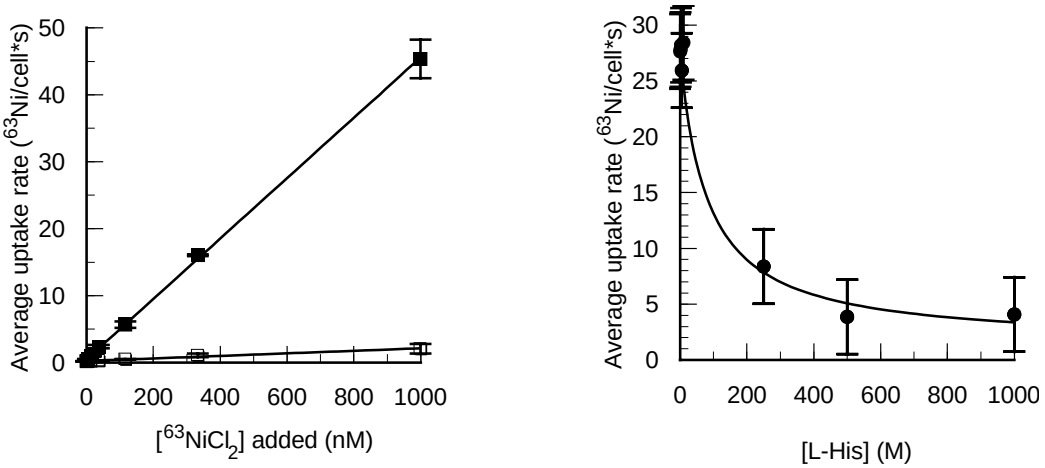
Supplementary Figure 1. **^{63}Ni accumulation in wild-type *E. coli* grown in different media.** Cells were grown anaerobically overnight (14-16 h) at 37 °C in capped microfuge tubes containing 1 nM $^{63}NiCl_2$. LB, Luria Bertani broth; YE, yeast extract (0.05% w/v added to M63 medium), T, tryptone (0.1% w/v added to M63 medium). aa, 18 amino acids except Cys and Trp added to M63. aa-His, 17 amino acids (no His, Cys, Trp).



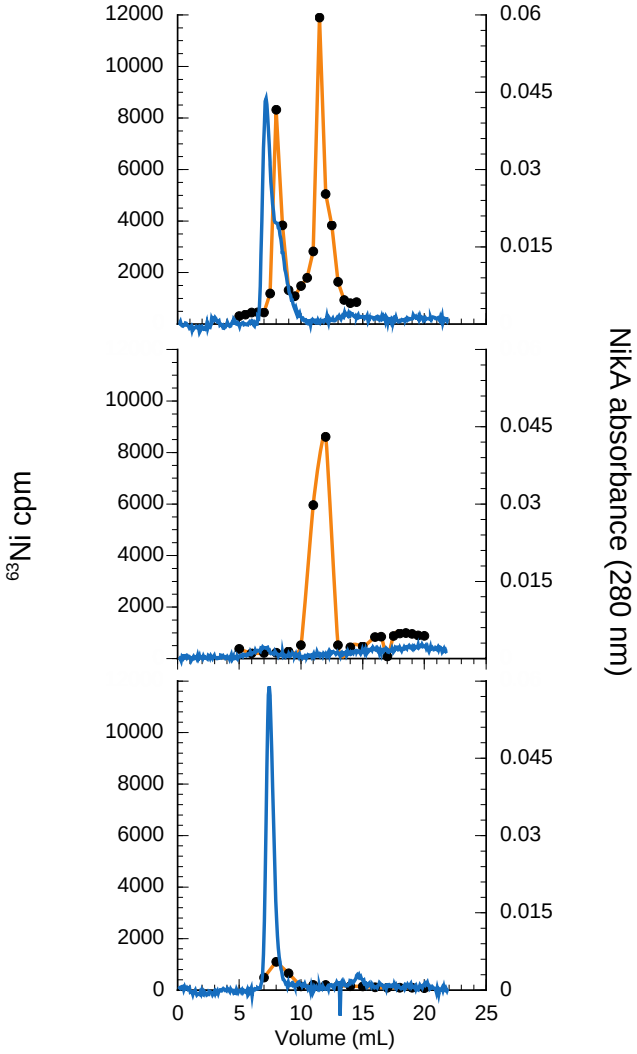
Supplementary Figure S2. *tonB* is not required for NikABCDE-dependent Ni-uptake in *E. coli*. Nickel uptake assays were performed on wild-type (filled and open circles)and Δ*tonB* (filled and open triangles) *E. coli* grown as described in Material and Methods. $^{63}NiCl_2$ (20 nM) and L-His (400 μM) were added to the tube prior to the addition cells. The two reagents were either premixed to allow Ni-L-His complex formation (filled symbols), or mixed only upon addition of cells (open symbols). Each point is the average of two independent samples and standard deviation is shown by the error bars.



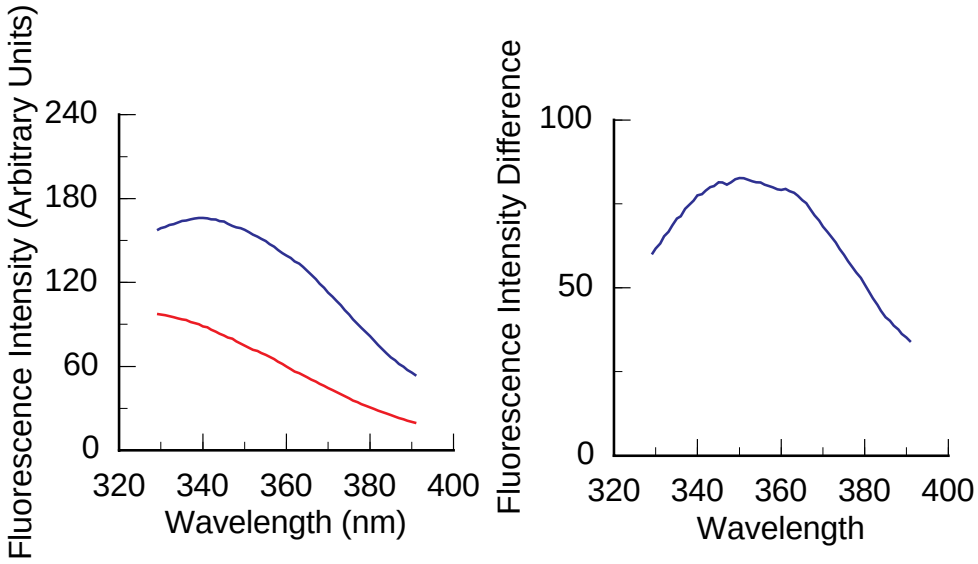
Supplementary Figure S3 ^{63}Ni uptake rates for various polycarboxylic acid molecules. Each point is the average of three samples and the error is reported as the standard deviation. EDTA, ethylenediaminetetraacetic acid; NTA, nitrilotriacetic acid.



Supplementary Figure S4. **Comparison of the kinetic properties of L-His dependent and independent Ni-uptake pathways. A**, $^{63}NiCl_2$ was added to cells grown anaerobically overnight (37 °C) containing no added L-His (wild-type strain, filled squares) or 1 mM added L-His (Δ*nikABCDE* strain, open squares). Each point is the average of two individual samples and error is reported as standard deviation. The data were fit to y=mx+b (R^2 = 0.9996, no L-His; R^2 = 0.9794, 400 μM L-His). The goodness of the linear fits indicates that this process was non-saturable and represented non-specific entry of nickel ions into the cell down a concentration gradient. B, The concentration dependence of L-His inhibition of non-specific Ni-uptake. Half-maximal inhibition occurred at ~80 μM L-His, which is consistent with the concentration dependence of Ni-$(L\text{-}His)_2$ formation (See Figure 2).



Supplementary Figure S5. Co-elution of ^{63}Ni and NikA from size-exclusion chromatography in the presence of L-His. Size exclusion chromatography (BioRad Bio-Silect SEC 125-5 column (300 mm x 7.8 mm) of 200 □L samples (NikA, 50 μM; $^{63}NiCl_2$, 1 μM; and L-His, 1 mM)**.** The blue trace represents the continuous A_{280} reading and the orange trace with black circles indicates ^{63}Ni cpm for each 1 mL fraction collected. A, all three components present; B, no NikA present; C, no L-His present. The traces shown above are representative of multiple runs. The presence of all three components appears to slow the elution of the NikA protein, noted by the shoulder in the blue trace in panel A. This is likely due to the conformational change known to occur with solute binding proteins in the presence of their ligand. In the absence of L-His, ^{63}Ni reproducibly elutes after the fraction collection window (panel C).



Supplementary Figure S6. Ni-His dependent quenching of intrinsic NikA tryptophan fluorescence. Left, NikA (2 μM in 20 mM Hepes (pH 7.5) and 200 mM NaCl) was incubated at 22 °C for 30 min with 400 μM L-His and with (red line) or without (blue line) $NiCl_2$ (2 μM). Right, difference spectrum from left panel (blue-red traces).

Table S1 Metal-ligand distances in Me-(L-His)$_2$ crystal structures

	Metal			
Ligand atom	Ni(II)[a]	Co(II)[b]	Cu(II)[c]	Zn(II)[d]
Imid-Nδ	2.106	2.172	1.984	2.039
NH_2	2.092	2.172	2.003/1.966	2.106
COO^-	2.113	2.121	2.277	*2.788*

a, *trans*-imidazole; Fraser, K. A. and M. M. Harding *J. Chem Soc A* 415 (1967)
b, *trans*-imidazole; Harding, M. M. and H. A. Long *J. Chem Soc A* 2554 (1968)
c, square-pyramidal, only one imid coordinated; Deschamps, P, Kulkarni, P. P., and B. Sarkar *Inorg Chem* **43**, 3338—3340 (2004).
d, tetrahedral, no COO^-–coordination; Kretsinger, R, H., Cotton, F. A., and R. F. Bryan *Acta Crystallogr* **16**, 651 (1963).