

Supplementary Material For:

Holo-Ni(II)*Hp*NikR is an asymmetric tetramer containing two different nickel binding sites

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Holo-Ni(II)*Hp*NikR Structure

Figure S1A (attached as a movie, mpg file);

The movie provides a visualization of the differences in the ligand geometries for the 4- and 5/6-coordinate sites. Two sites were aligned as shown below in figure S1B, and the intermediate models were produced using the “morph” function in UCSF Chimera.¹ The transition shown does not represent the actual transition that occurs when nickel moves between the two metal binding sites. The sole purpose of the movie is to illustrate the differences between the two nickel binding sites, which are difficult to observe when the sites are merely overlaid (as below in Figure S1B).

(1) Eric, F. P., Thomas, D. G., Conrad, C. H., Gregory, S. C., Daniel, M. G., Elaine, C. M. & Thomas, E. F. *J Comp Chem*, **2004** 25, 1605-1612.

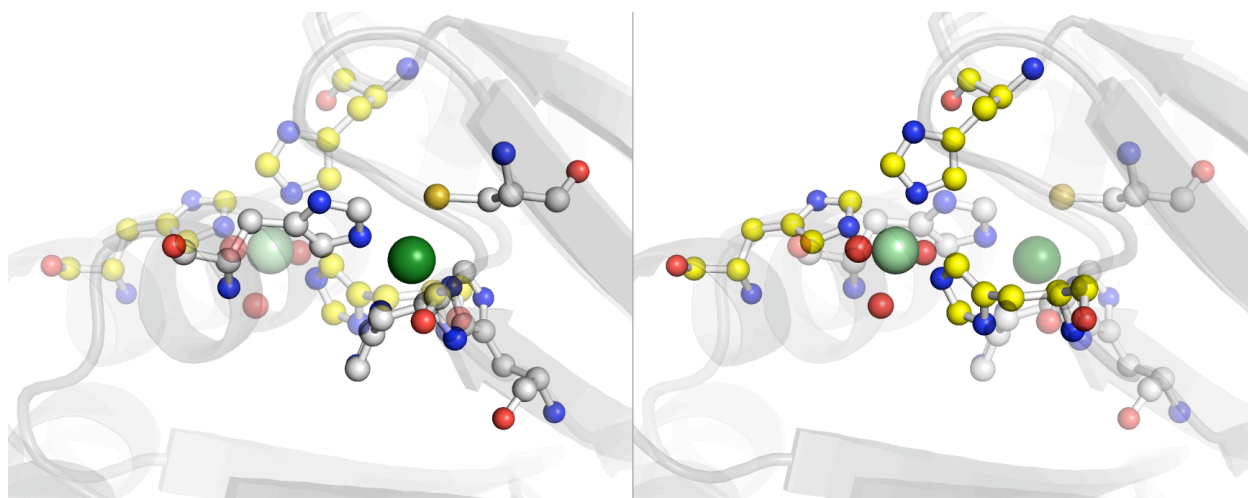


Figure S1B.

Comparison of 4- and 6-coordinate nickel binding sites. The structures were superimposed by aligning the beta-strands which contain Cys107. Residues forming the 6-coordinate site are transparent in the left panel and those forming the 4-coordinate site are transparent in the right panel.

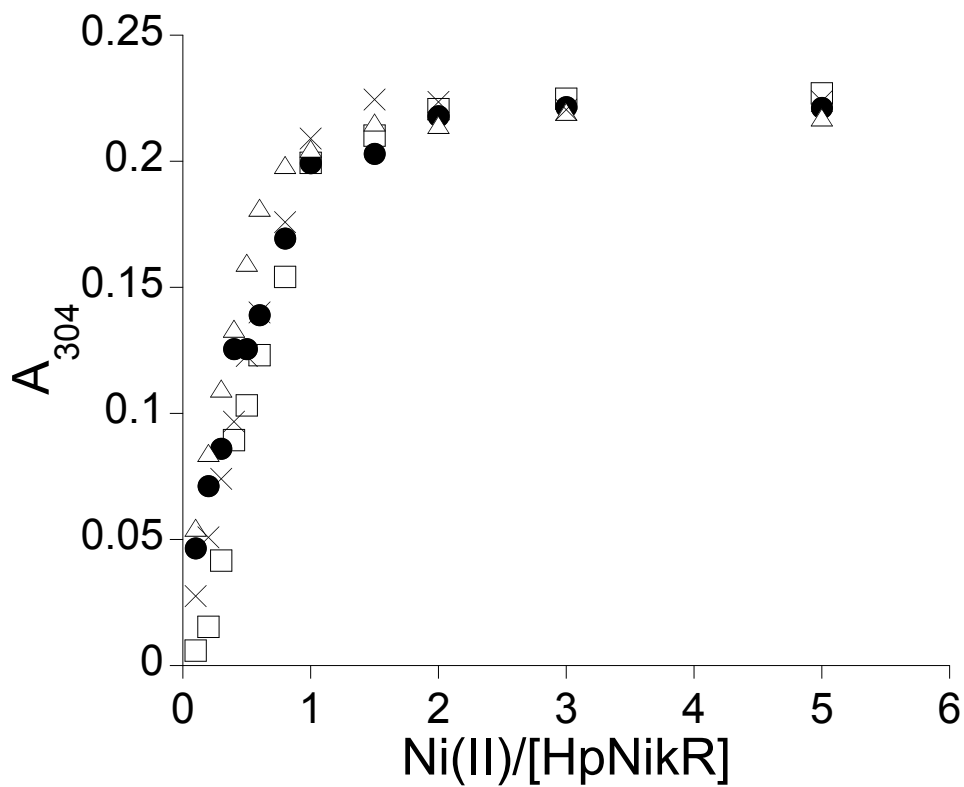


Figure S2. Plot of the absorption spectrum at 304 nm of *HpNikR* (45 μ M) as a function of added Ni(II) at pH 5.6 (\bullet), pH 6.5 (\square), pH 7.5 (\triangle), pH 8.5 (\times). Saturation occurs at 1 equivalent of Ni(II) per monomer.

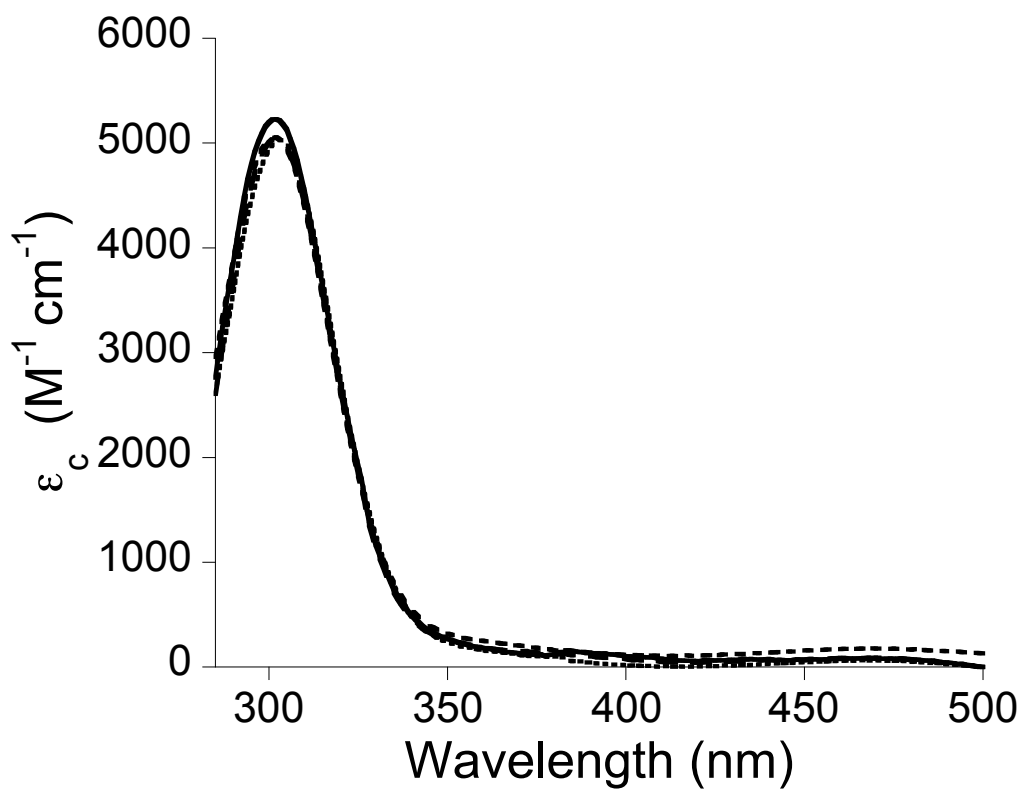


Figure S3. Overlay of the difference UV visible spectra of Ni(II)-*HpNikR* at pH 5.6 (solid line), pH 6.5 (long dashed line), pH 7.5 (medium dashed line), pH 8.5 (short dashed line). Conditions [45 μM Ni(II)*HpNikR* in 20 mM phosphate, (20 mM glycine at pH 5.6)]

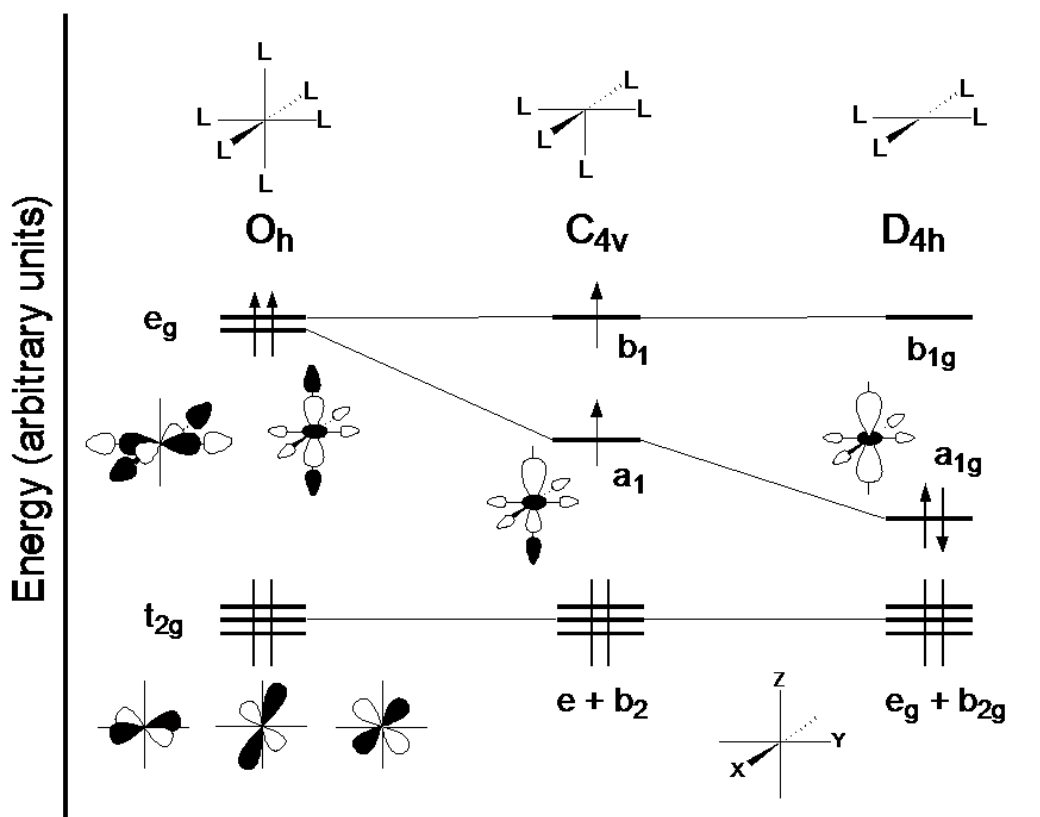


Figure S4.

Qualitative representation of the frontier orbitals of ideal octahedral (O_h), square-pyramidal (C_{4v}) and square-planar (D_{4h}) complexes. The electron configurations correspond to a d^8 metal center. For the square-pyramidal complex the ground state is represented as the triplet state, the preferred state when the contribution of the electron-electron repulsion overcomes the energy gap of the a_1 and b_1 orbitals.

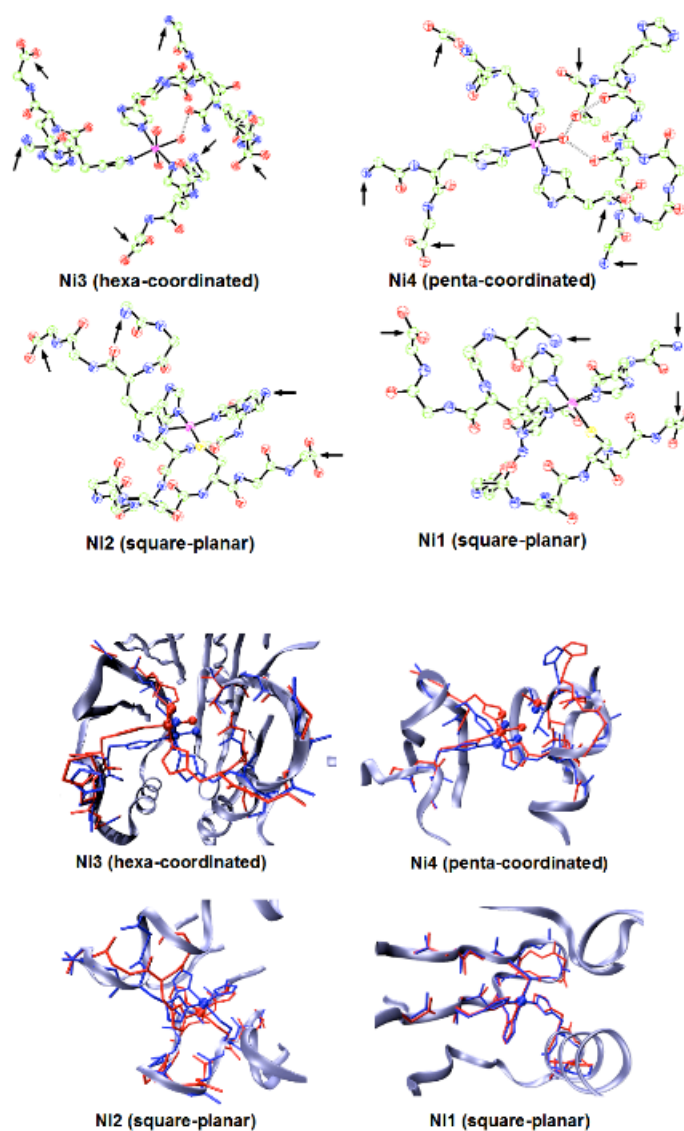


Figure S5. Illustration of the large clusters alone (top) and superimposed on the experimental protein structure (bottom). Optimized clusters are represented in red and the original coordinates (x-ray) are in blue. Arrows indicate atoms that were kept frozen during the geometry optimization.

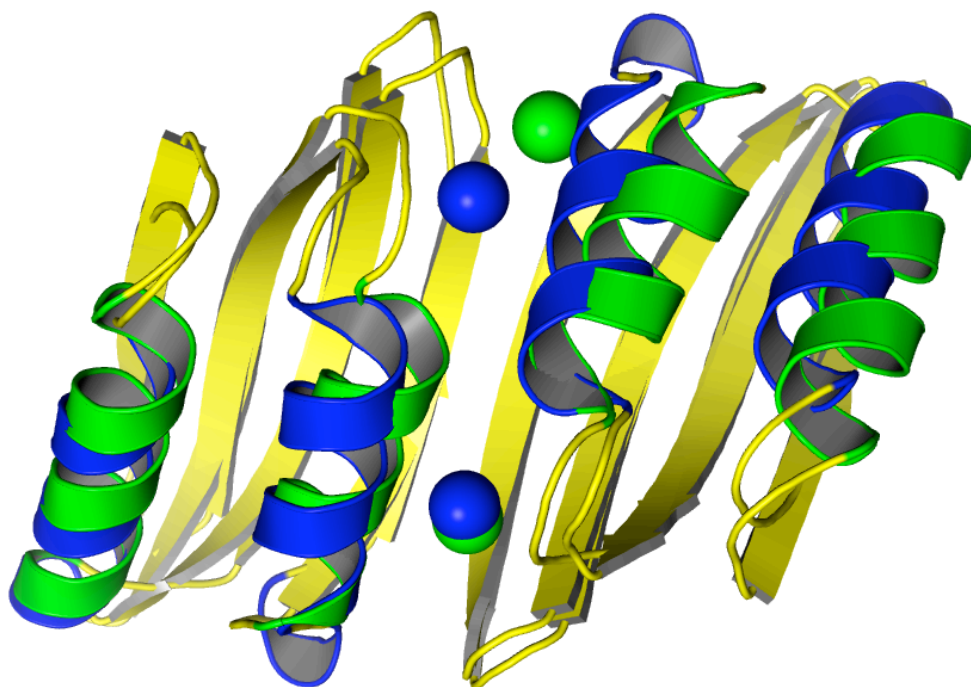


Figure S6. MBD of holo-Ni(II)*Hp*NikR (α helices are colored green) overlaid with MBD of holo-Ni(II)*Ph*NikR (α -helices are colored blue). Structural alignment was based on the C_{α} atoms in the β -sheets.

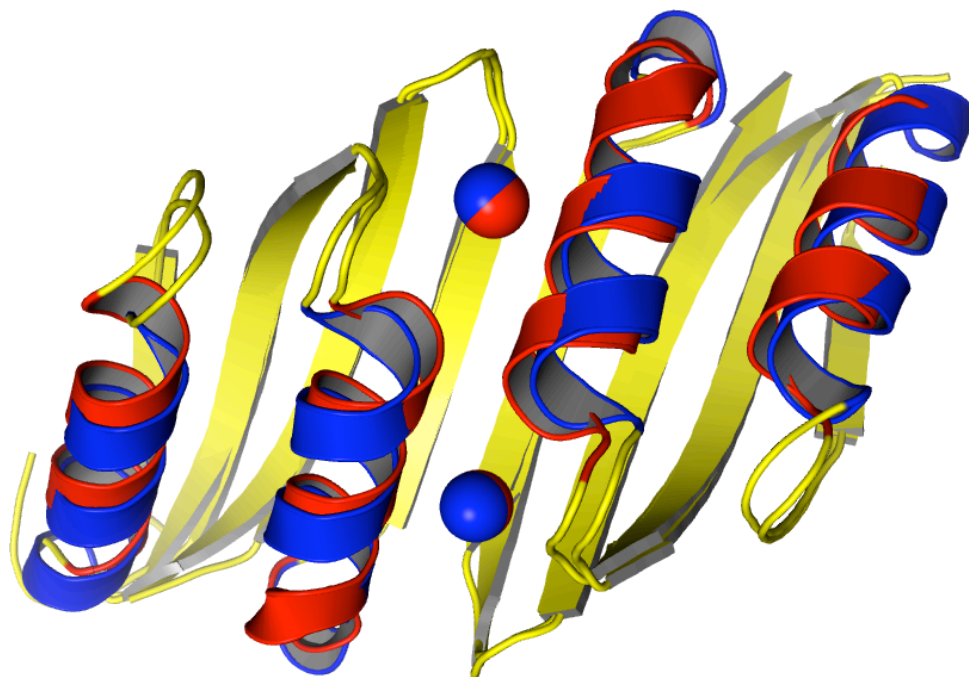


Figure S7. MBD of holo-Ni(II)*Ec*NikR (α helices are colored red) overlaid with MBD of holo-Ni(II)*Ph*NikR (α -helices are colored blue). Structural alignment was based on the C_{α} atoms in the β -sheets.

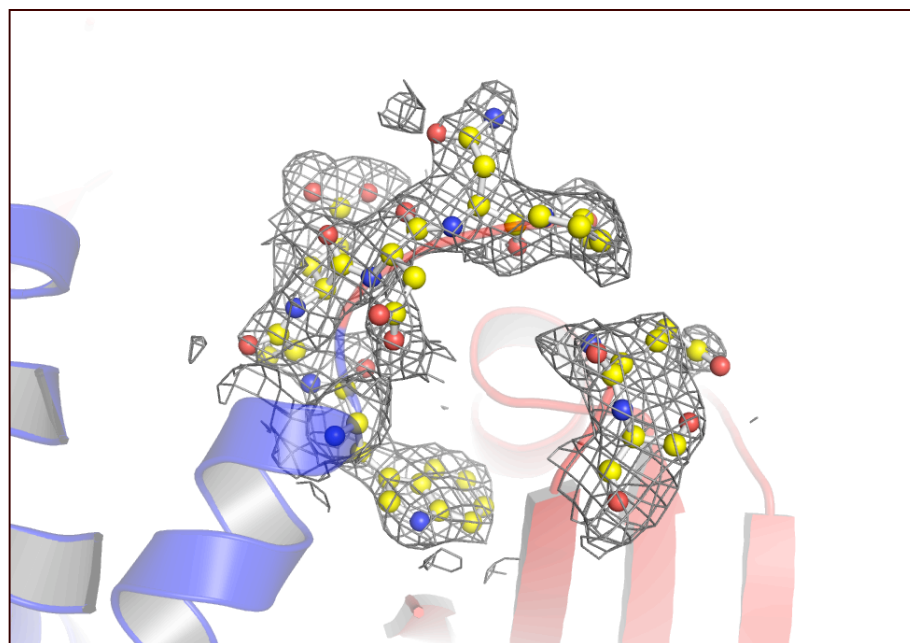
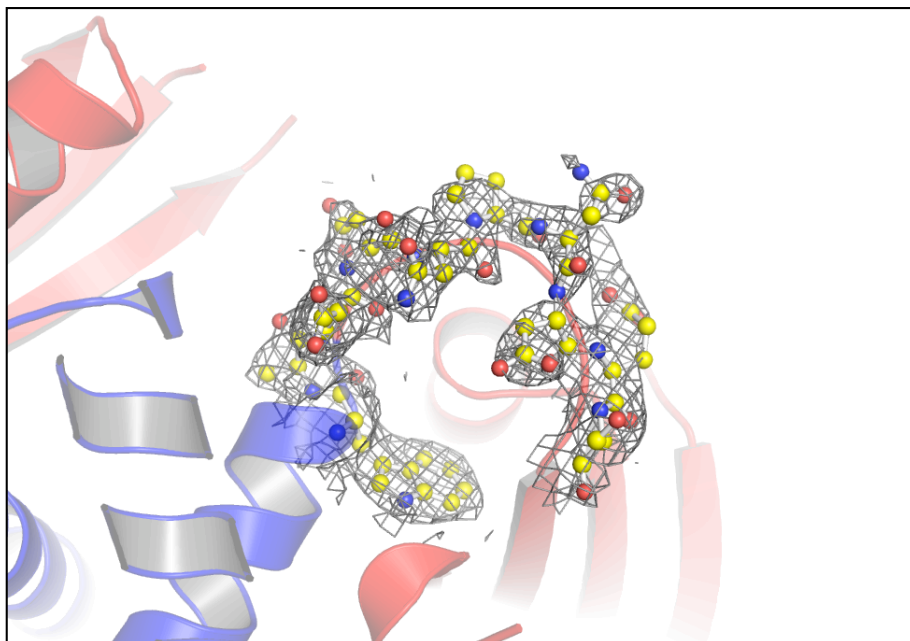


Figure S8. Electron density observed at the interface between the MBD and DBD in the asymmetric unit cell.

Full Citation for Reference 39:

E. J. Bylaska, W. A. de Jong, N. Govind, K. Kowalski, T. P. Straatsma, M. Valiev, D. Wang, E. Apra, T. L. Windus, J. Hammond, P. Nichols, S. Hirata, M. T. Hackler, Y. Zhao, P.-D. Fan, R. J. Harrison, M. Dupuis, D. M. A. Smith, J. Nieplocha, V. Tipparaju, M. Krishnan, Q. Wu, T. Van Voorhis, A. A. Auer, M. Nooijen, E. Brown, G. Cisneros, G. I. Fann, H. Fruchtl, J. Garza, K. Hirao, R. Kendall, J. A. Nichols, K. Tsemekhman, K. Wolinski, J. Anchell, D. Bernholdt, P. Borowski, T. Clark, D. Clerc, H. Dachsel, M. Deegan, K. Dyall, D. Elwood, E. Glendening, M. Gutowski, A. Hess, J. Jaffe, B. Johnson, J. Ju, R. Kobayashi, R. Kutteh, Z. Lin, R. Littlefield, X. Long, B. Meng, T. Nakajima, S. Niu, L. Pollack, M. Rosing, G. Sandrone, M. Stave, H. Taylor, G. Thomas, J. van Lenthe, A. Wong, and Z. Zhang, "NWChem, A Computational Chemistry Package for Parallel Computers, Version 5.1" (2007), Pacific Northwest National Laboratory, Richland, Washington 99352-0999, USA.