Hg^{+2} Transfer Between Flexibly Linked Domains of Mercuric Reductase MerA

Contact: Alex Johs (johsa@ornl.gov, 865-574-7444)

DOE/Office of Science/Biological & Environmental Research

Objective

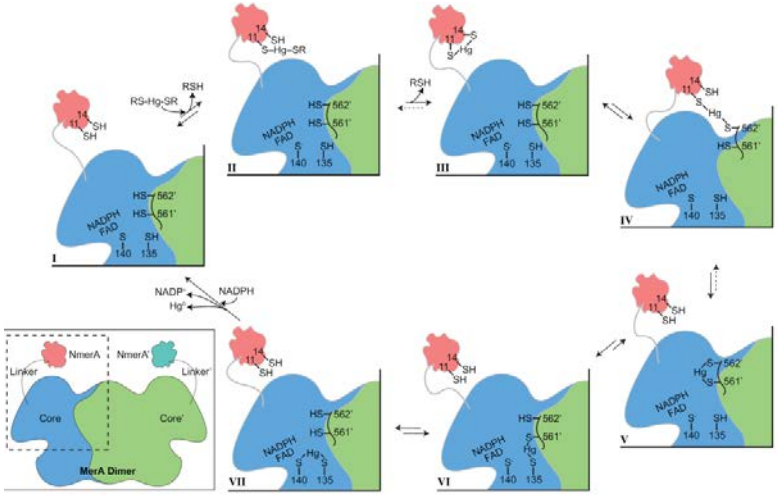
- Demonstrate how reductase MerA transfers Hg^{2+} to its catalytic core domain for reduction to relatively benign Hg^0.

New Science

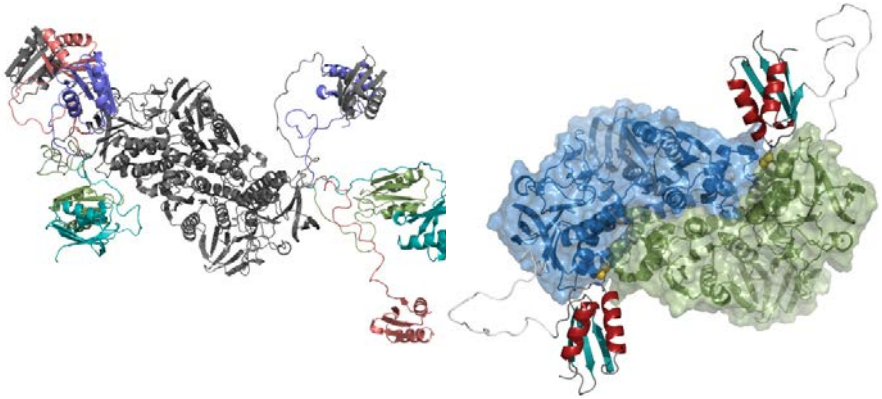
- By combining experimental neutron and X-ray scattering data with computational methods, as well as using specific mutations in MerA, we determined the position and relative orientation of NmerA interacting with its core during Hg^{2+} handoff.

Significance

- The mechanism by which MerA transfers Hg^{2+} from a flexibly tethered metallochaperone domain to its catalytic core for reduction to Hg^0 has general applications as many bacteria in Hg-contaminated environments are resistant to toxic levels of mercury.



Pathway for Hg^{2+}-ligand exchange and reduction in MerA. NmerA (red), dimeric catalytic core (blue/green).



Left: Superposition of the five models representing conformational ensemble. Right: Conformation during Hg^{2+} handoff.

Johs, A., I.M. Harwood, J.M. Parks, R. Nauss, J.C. Smith, L. Liang and S.M. Miller. 2011. Structural characterization of intramolecular Hg^{2+} transfer between flexibly-linked domains of mercuric ion reductase. J. Mol. Biol. 413:639-656 (doi:10.1016/j.jmb.2011.08.042).

OAK RIDGE National Laboratory

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The enzyme mercuric ion reductase, MerA, is the central component of bacterial mercury resistance encoded by the mer operon. Many MerA proteins possess metallochaperone-like N-terminal domains, NmerA, that can transfer Hg^{2+} to the catalytic core domain (Core) for reduction to Hg^0. These domains are tethered to the homodimeric Core by ~30-residue linkers that are susceptible to proteolysis which has prevented characterization of the interactions of NmerA and Core in the full-length protein. Here, we report purification of homogeneous full-length MerA from the Tn21 mer operon using a fusion protein construct and combine small-angle X-ray and neutron scattering with molecular dynamics simulation to characterize the structure of full-length wild-type and mutant MerA proteins that mimic the system before and during handoff of Hg^{2+} from NmerA to the Core. The radii of gyration, distance distribution functions and Kratky plots derived from small-angle X-ray scattering data are consistent with full-length MerA adopting elongated conformations as a result of flexibility in the linkers to the NmerA domains. The scattering profiles are best reproduced using an ensemble of linker conformations. This flexible attachment of NmerA may facilitate fast and efficient removal of Hg^{2+} from diverse protein substrates. Using a specific mutant of MerA allowed formation of a metal-mediated interaction between NmerA and Core and determination of the position and relative orientation of NmerA to the Core during Hg^{2+} handoff.

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