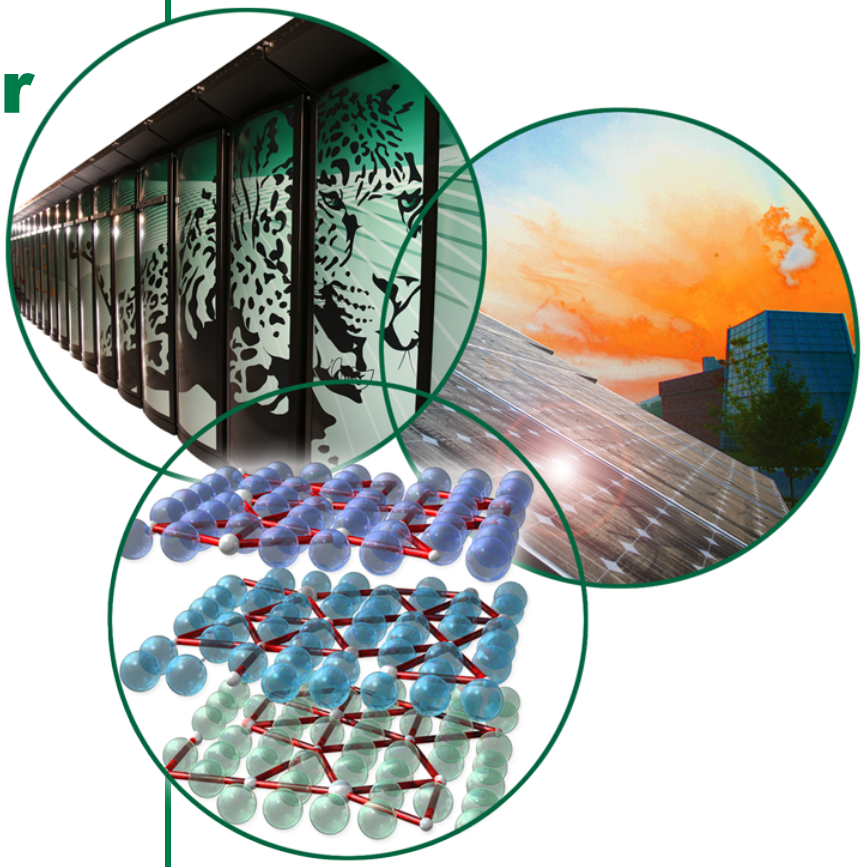
Allosteric control mechanism of the mercury-responsive transcriptional regulator MerR



Alexander Johns

ACS Fall 2009 National Meeting

U.S. DEPARTMENT OF ENERGY

OAK RIDGE NATIONAL LABORATORY
MANAGED BY UT-BATTELLE FOR THE DEPARTMENT OF ENERGY

Outline

- Introduction to the Mer system
- The MerR family of transcriptional regulators
- Experimental data by Small Angle X-ray Scattering (SAXS) on MerR
- Molecular dynamics of conformational changes
- Analysis of flexible domains
- Conclusions

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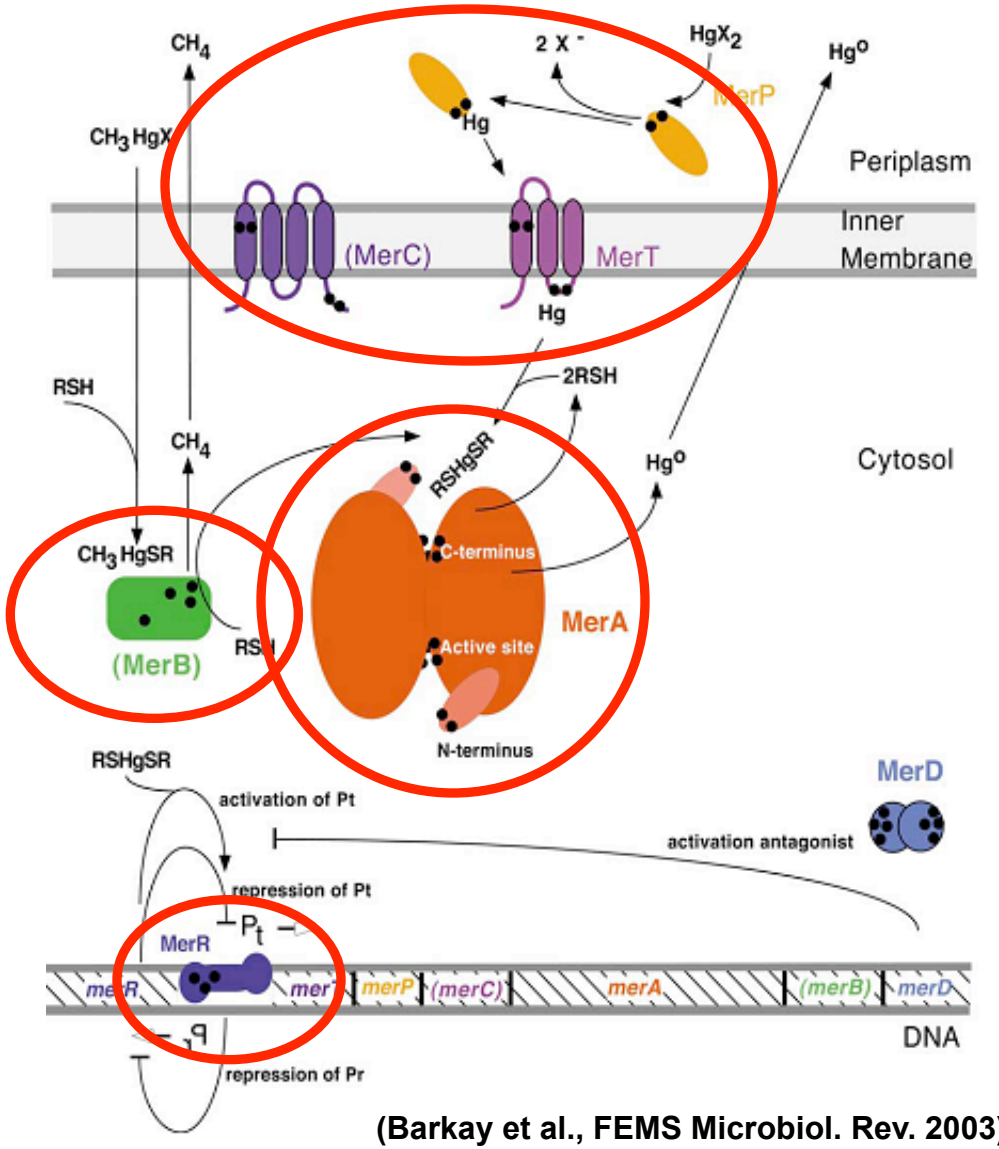
Bacterial Mercury Resistance

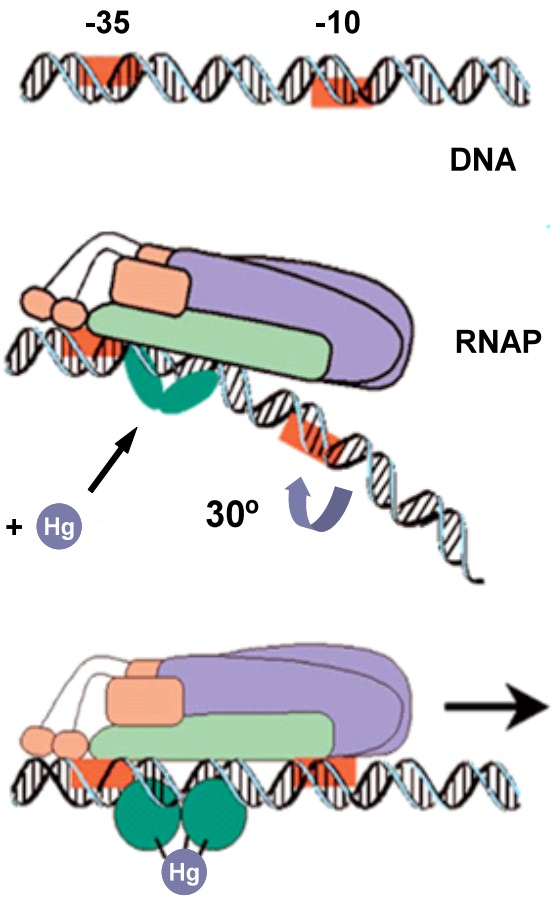


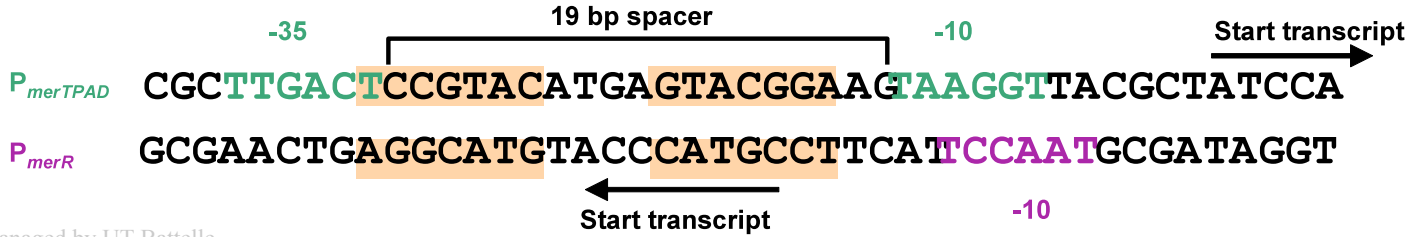
(Barkay et al., FEMS Microbiol. Rev. 2003)

- **Regulation**
  MerR, MerD
- **Uptake & Transport**
  MerC, MerT, MerP
  MerE, MerF, MerH
- **Demethylation**
  MerB
  - $CH_3\text{-}Hg(II)^+ \rightarrow Hg(II) + CH_4$
- **Reduction**
  MerA
  - $Hg(II) \rightarrow Hg(0)$

Structure and function of MerR

- MerR is a homodimer
- MerOP: dyad symmetry region between -35 and -10
- MerR forms inactive complex with RNAP
- Hg(II) initiates transcription
- Response mechanism at molecular level?





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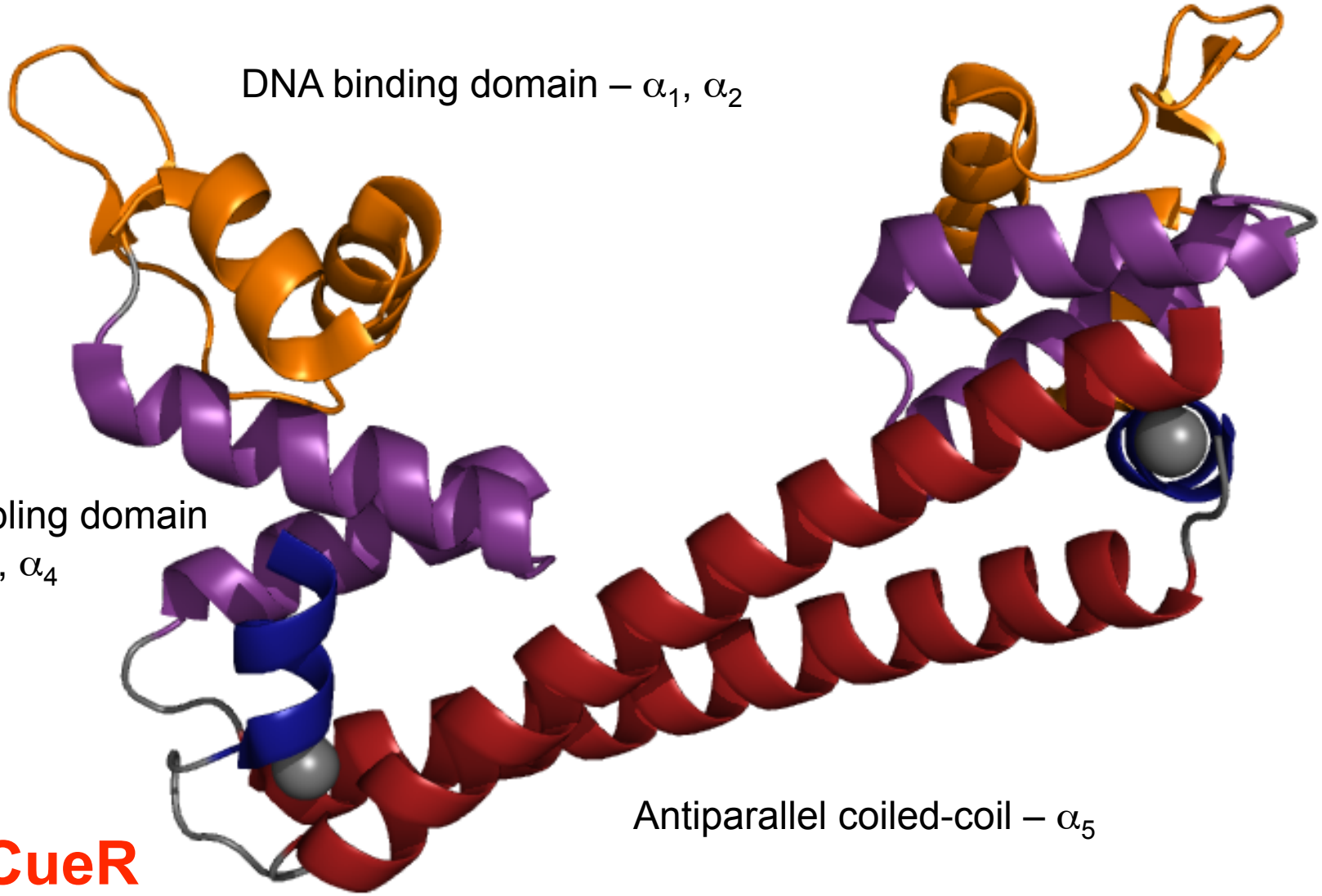
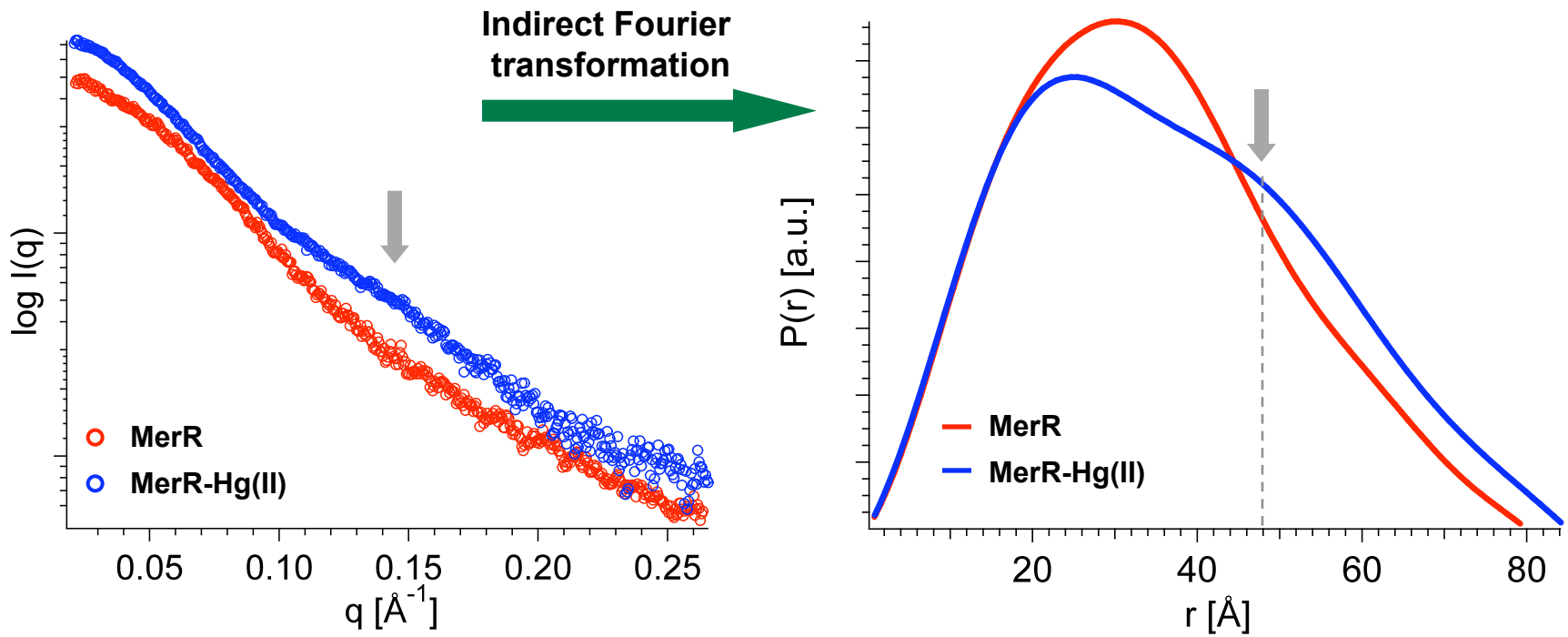
MerR family of transcriptional activators/ repressors



CueR

Cu(I)-dependent transcriptional regulator

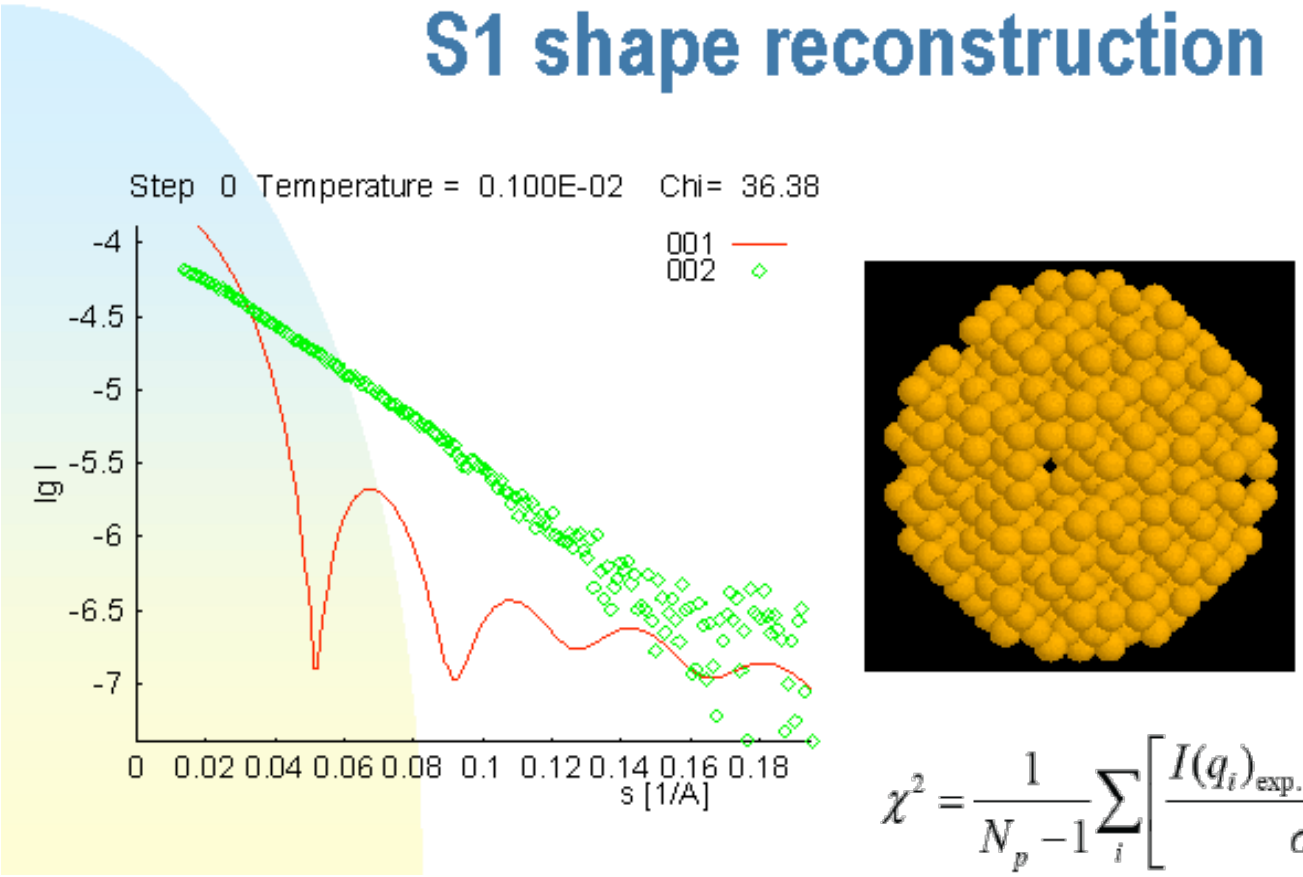
Experimental data - SAXS



- Hg(II) induces <u>conformational change</u> from a compact structure to a configuration with two distinct centers of mass
- Radius of gyration R_G:
  Apo-MerR: 24.7 ± 0.4 Å, Hg(II)-MerR: 28.6 ± 0.3 Å, ΔR_G = 3.9 ± 0.7 Å.

Low resolution shape reconstruction from SANS/SAXS data

Press Shift-F5 to view animation



$$\chi^2 = \frac{1}{N_p - 1} \sum_i \left[\frac{I(q_i)_{\text{exp.}} - cI(q_i)_{calc.}}{\sigma(q_i)} \right]^2$$

- ***Ab initio* shape reconstruction by DAMMIN/DAMMIF**
  D. Svergun, J. Appl. Cryst. 42: 342-346, 2009

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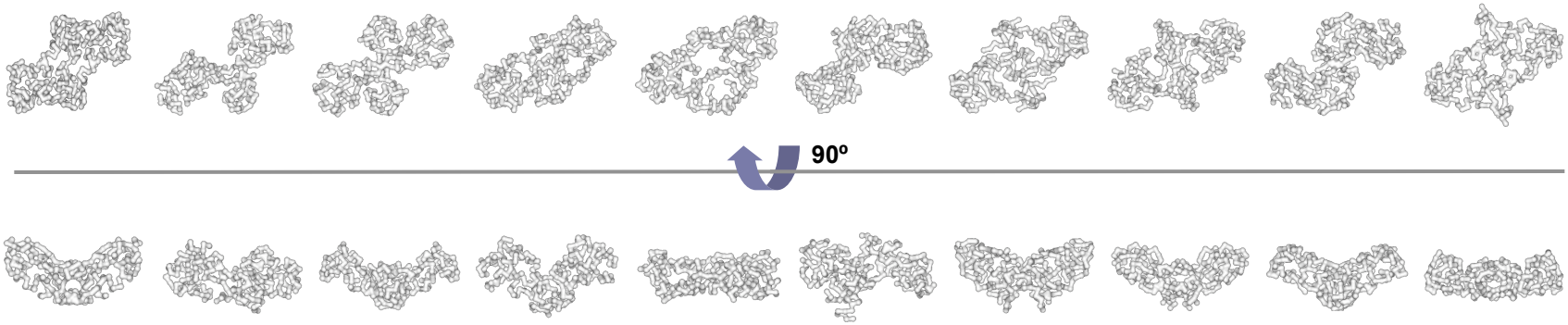
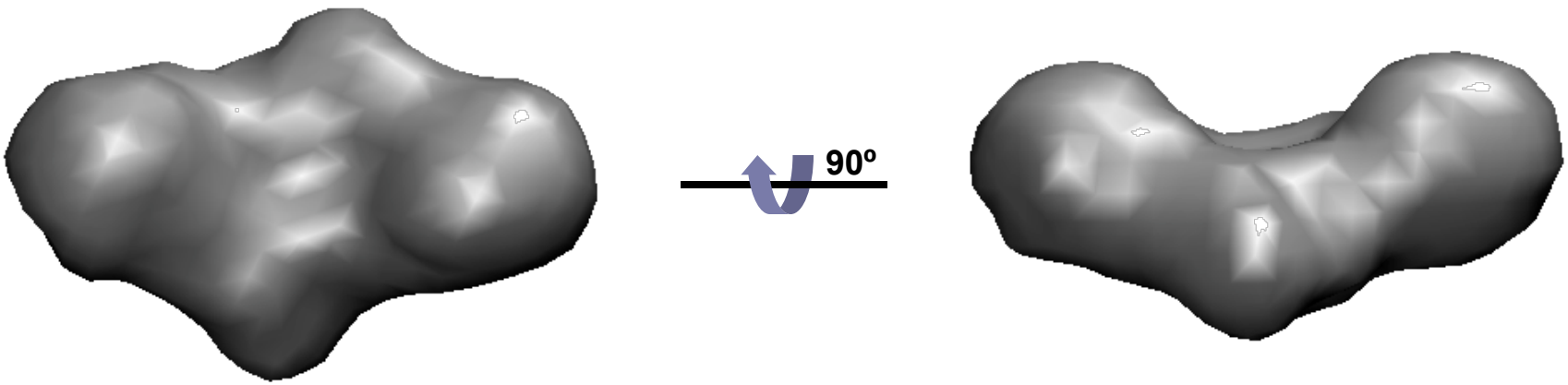
Shape reconstruction of apo-MerR

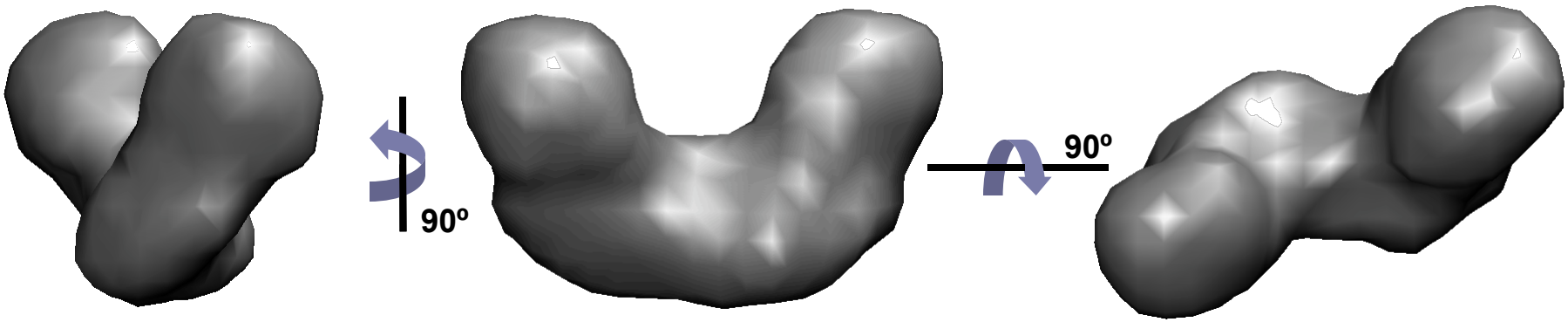


Independent single models obtained SAXS data using the program GASBOR.



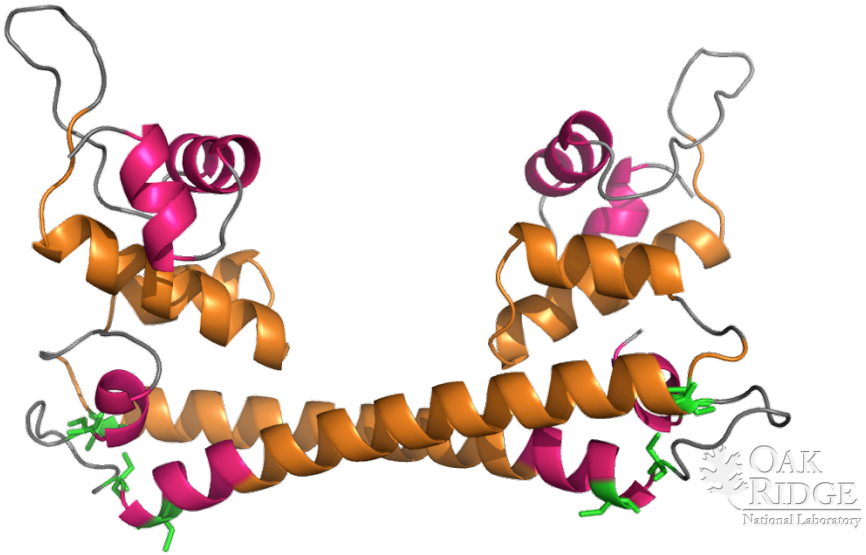
Envelope shape obtained from averaging of 20 GASBOR runs.

Hg(II)-MerR: Envelope shape and homology model

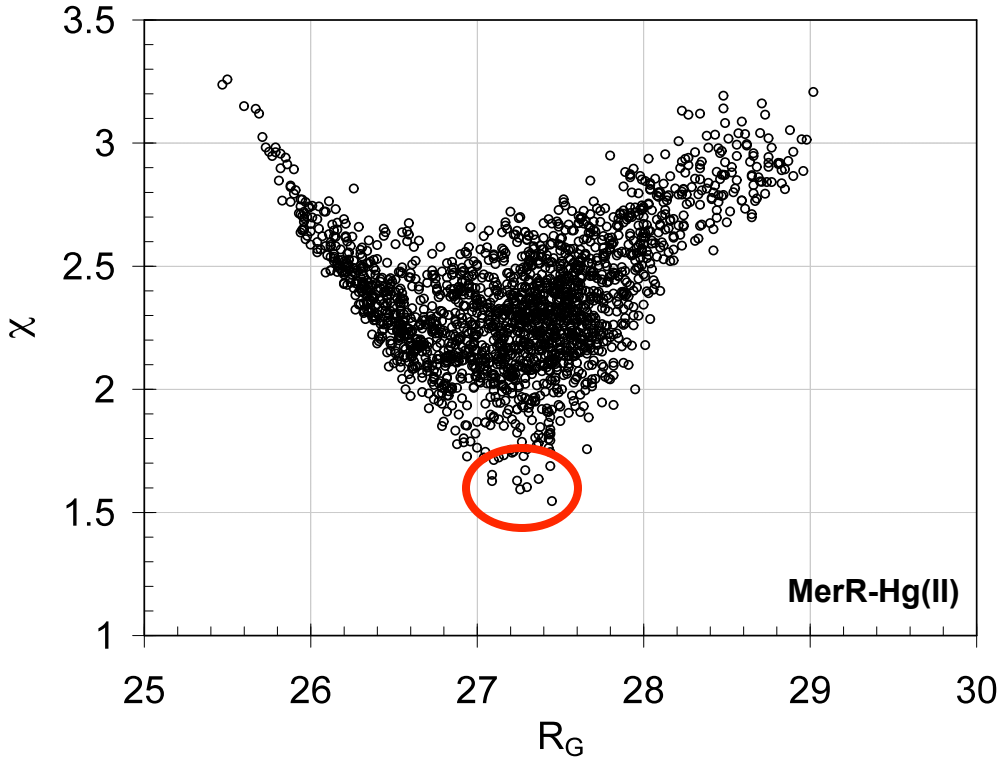


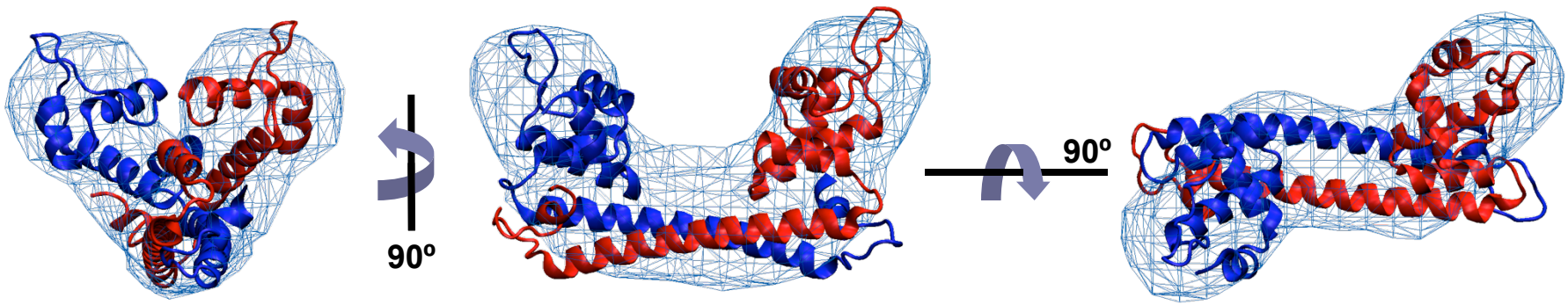
Envelope shape obtained from averaging of 20 GASBOR runs.

- **Homology model of Hg(II)-bound MerR based on**
  - **Structure of Cu(I)-bound CueR**
  - **Sequence of MerR (48% conserved residues)**
  - **Swiss-model http://swissmodel.expasy.org**

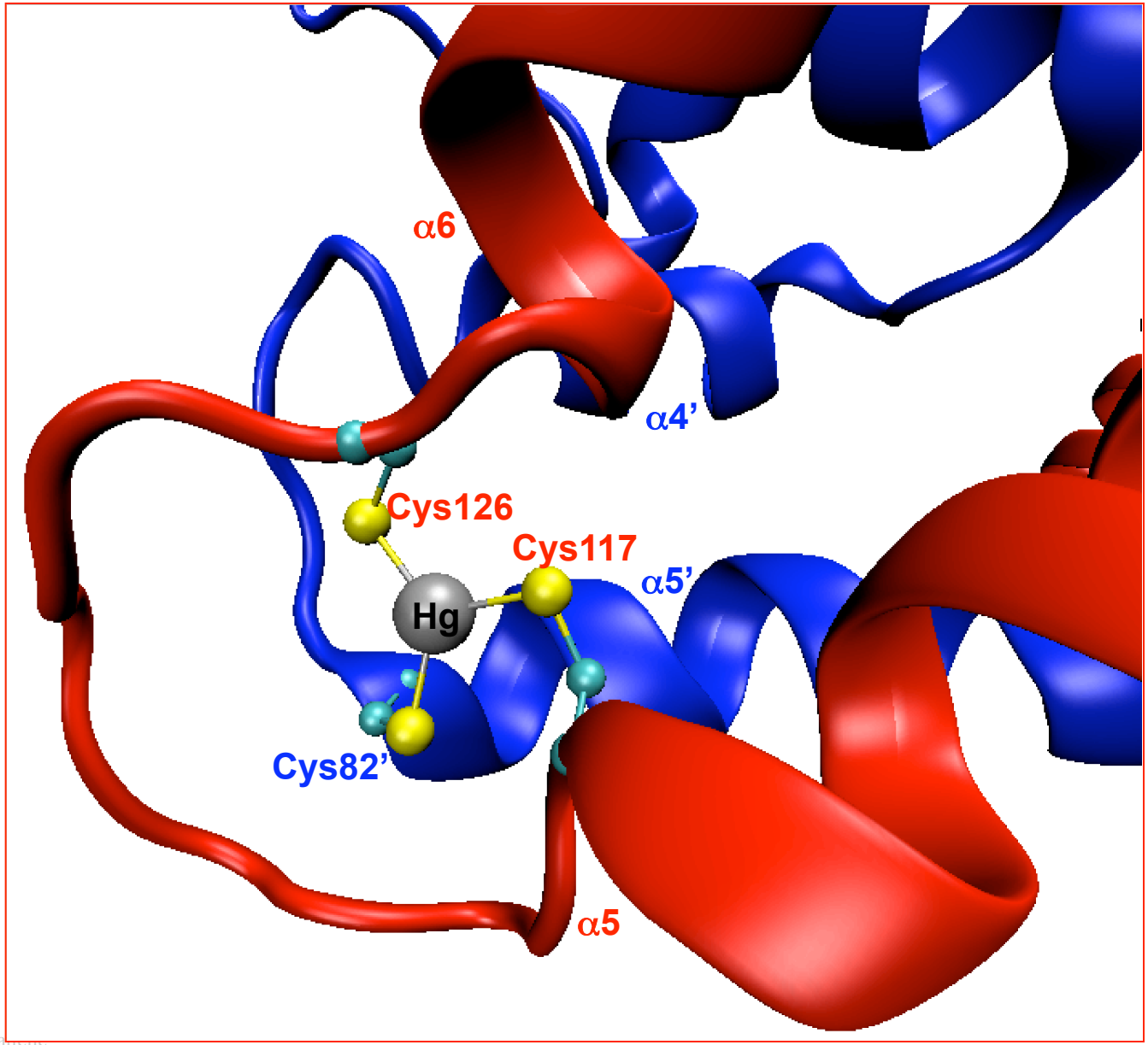


MD & Envelope Shapes of Hg(II)-MerR





Trigonal Planar Coordination/bonding of Hg(II) in MerR



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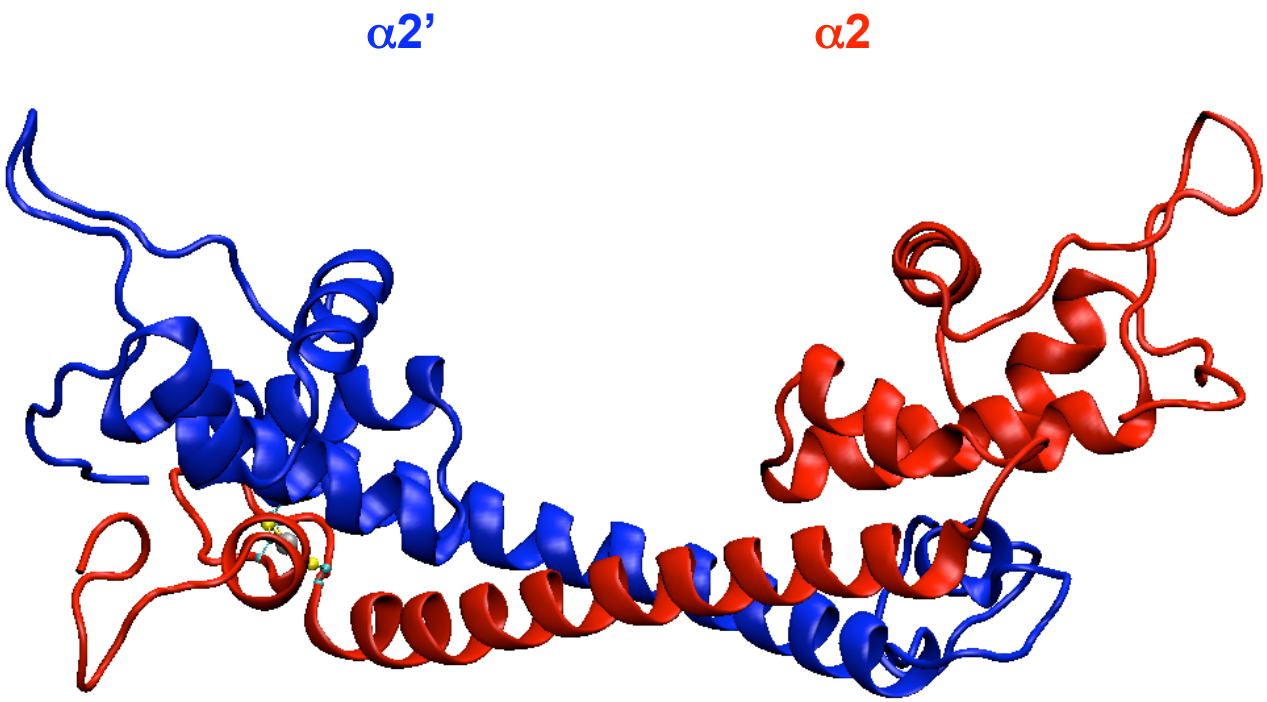
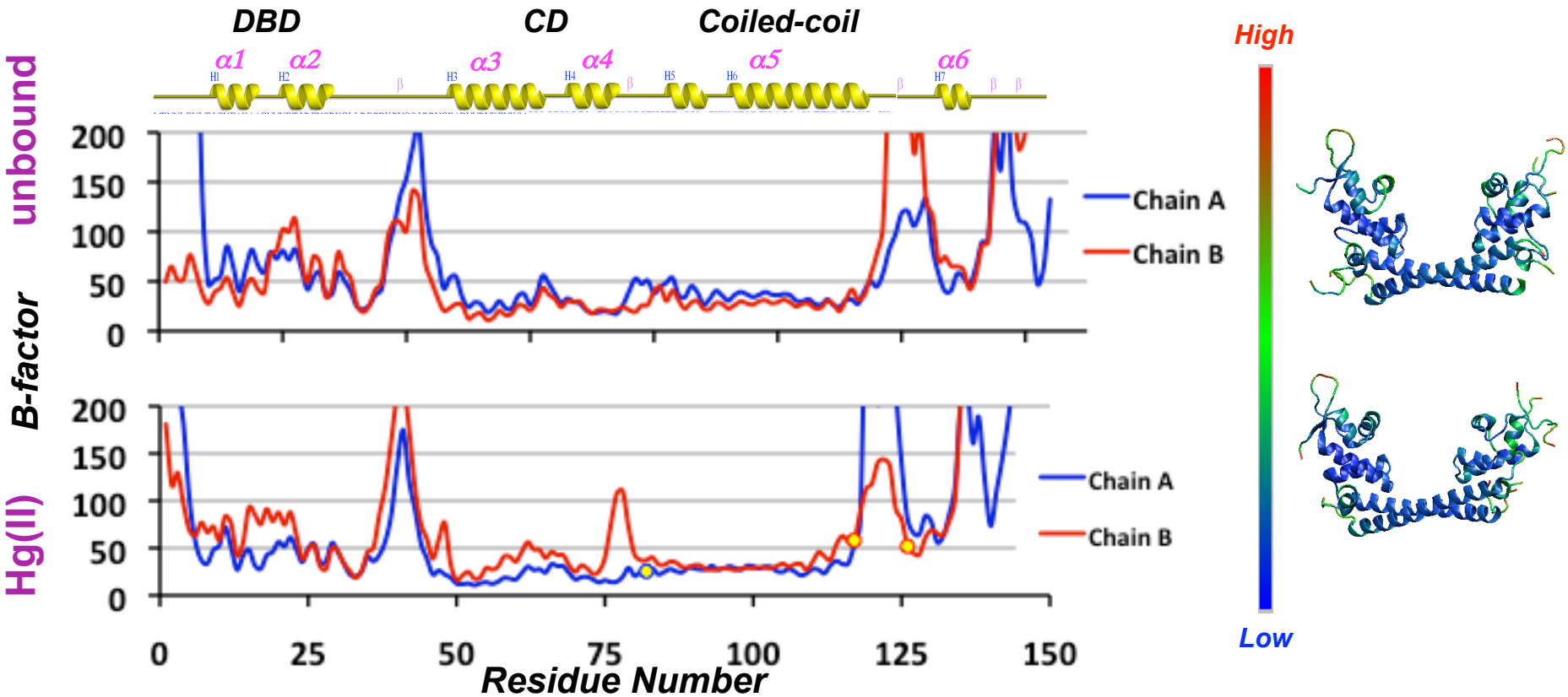
Molecular Dynamics Simulations



Largest amplitude motions are related to approaching/ departing of DNA binding domains.

B-factors of MerR



Top: unbound-MerR (MD); Bottom: Hg(II)-MerR (MD)

Conclusion

- SAXS shows a compact to extended conformational change upong binding of Hg(II)
- Obtained low resolution envelope shapes of unbound and Hg(II)-bound MerR
- Molecular dynamics on the homology model of MerR shows a closed to open conformational change upon binding of Hg(II)
- Identified flexible domain linkers
- Future:
  MerR / DNA / RNAP complex by SANS

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http://*www.esd.ornl.gov/programs/rsfa*