



Status report for IS602

Submitted to the ISOLDE and Neutron Time-of-Flight Experiments
Committee

**Cu^I, Ag^I, Cd^{II}, Hg^{II}, and Pb^{II} binding to biomolecules
studied by Perturbed Angular Correlation of γ -rays (PAC)
spectroscopy**

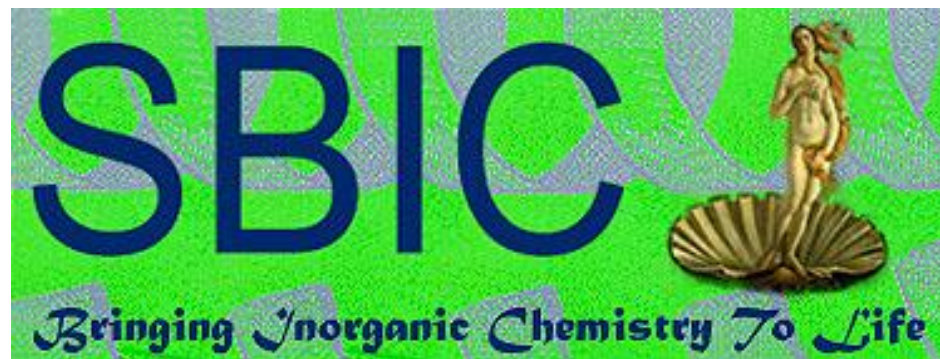
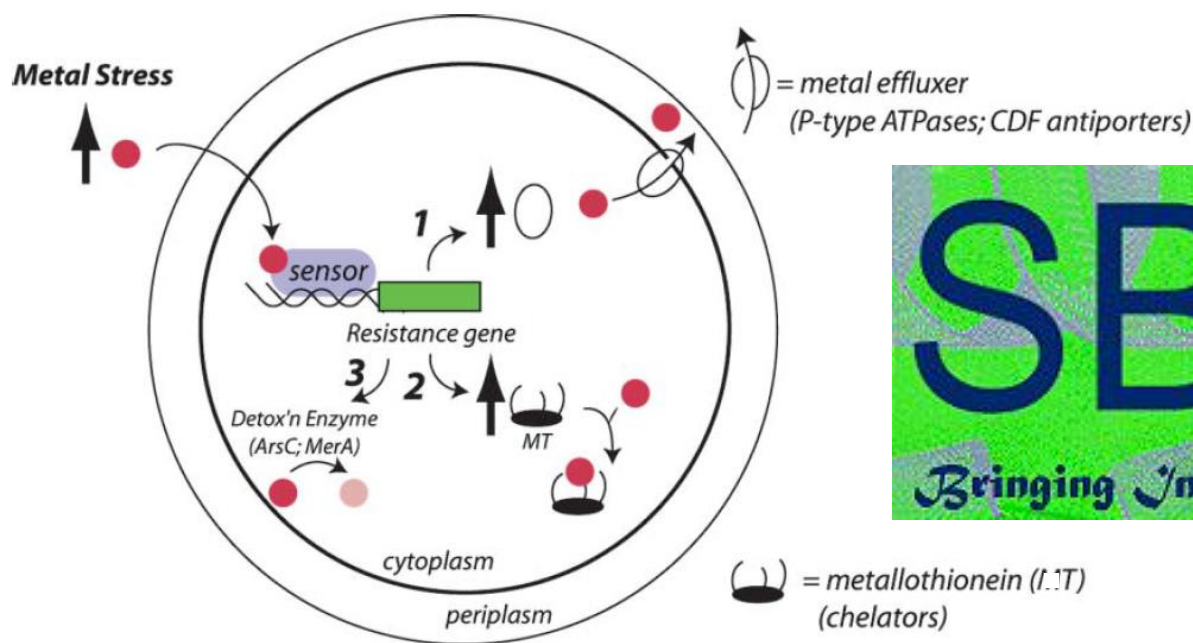
Spokesperson: Lars Hemmingsen, University of Copenhagen, Denmark

February 5, 2020, INTC Meeting, CERN



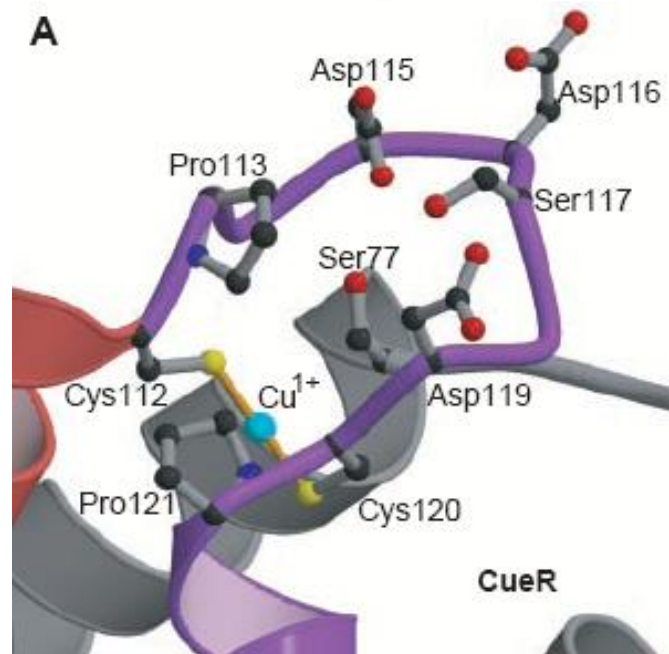
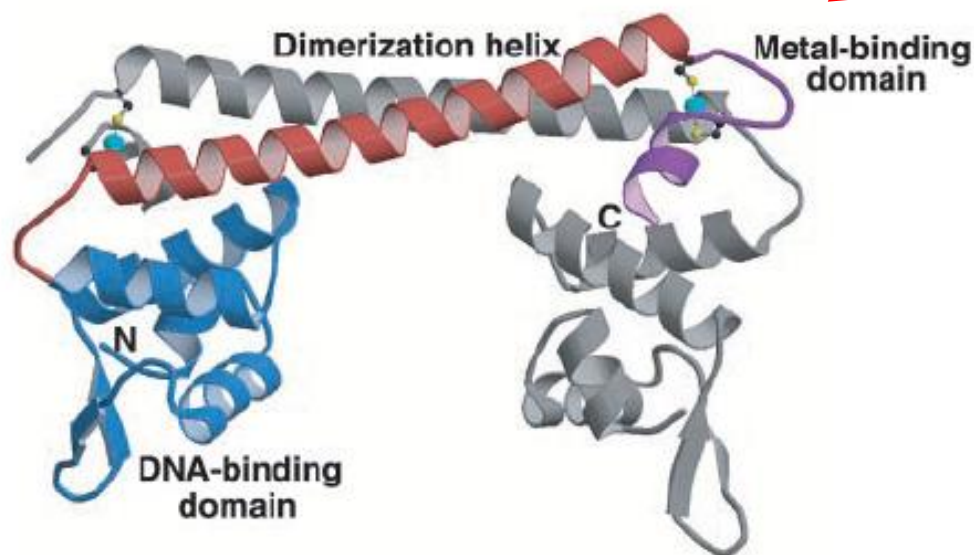
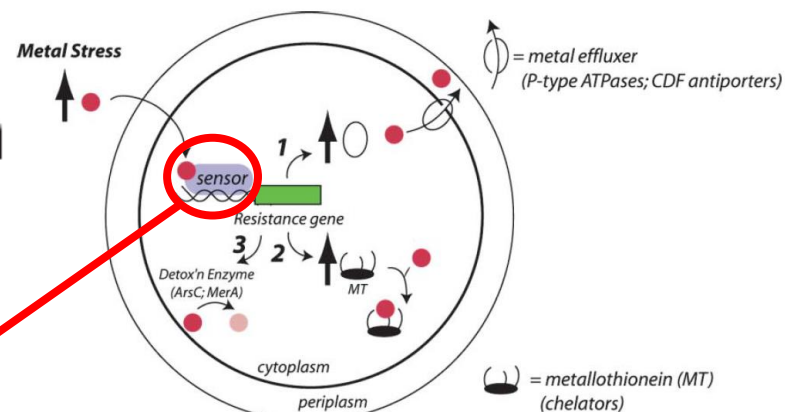
Research progress

Bacterial metal ion sensor proteins

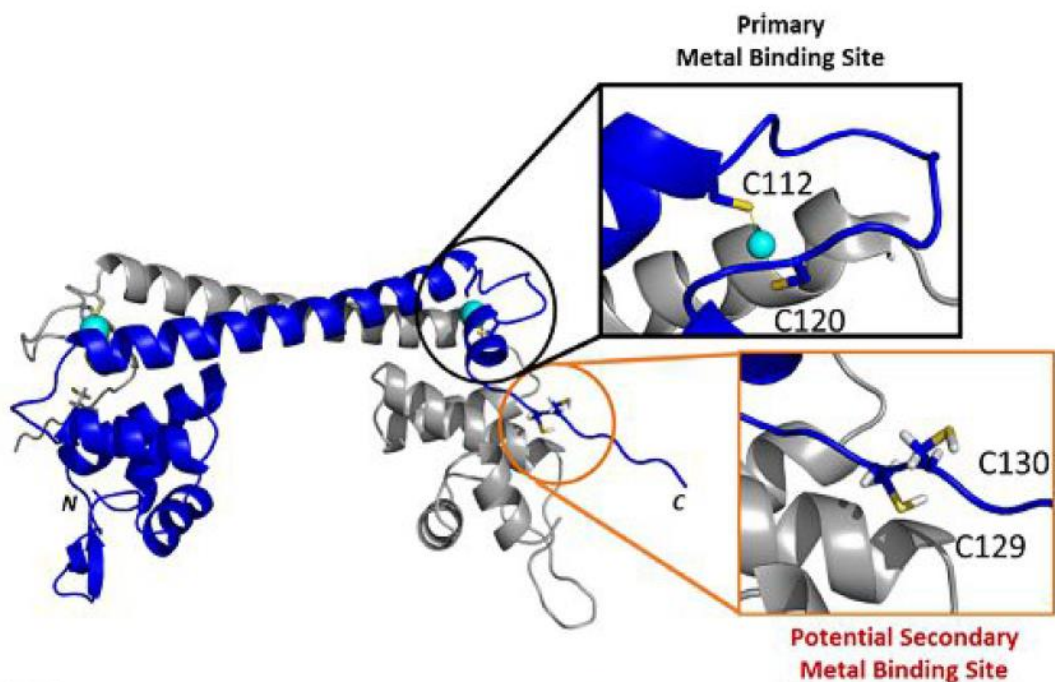


In *E. coli* Cu^{I} and Ag^{I} is controlled by CueR

Molecular Basis of Metal-Ion Selectivity and Zeptomolar Sensitivity by CueR

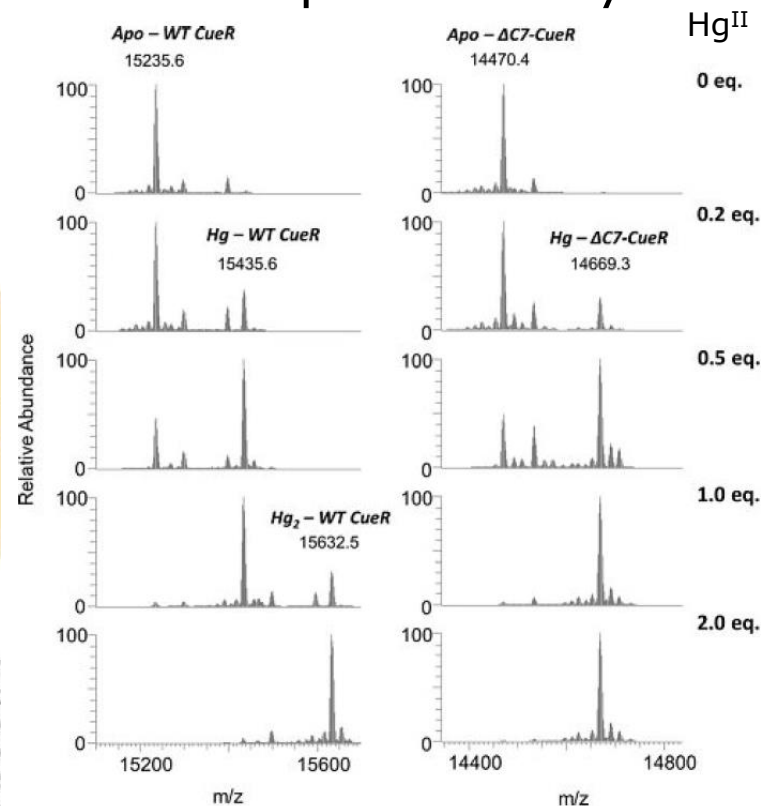


CueR metal binding: How are mono- and divalent metal ions discriminated?



CueR						
<i>Pseudomonas fluorescens</i>	100	LRDTLQDLVEH	C	NGDHRPD	C	PILKELASGCCAHPARA-- 13
<i>Pseudomonas aeruginosa</i>	100	LRDTLQDLVEH	C	QGDRHPDC	P	ILKDLASGCCH----- 13
<i>Klebsiella pneumoniae</i>	100	MRAHLLALAES	C	PGDSDAEC	P	IIDNL-SGCCHRKAQA-- 13
<i>Salmonella enterica</i>	100	MRLHLLALAES	C	PGDSDAEC	P	IIMDNL-SGCCCHHKAKA-- 13
<i>Escherichia coli</i>	100	MRDQLLALANA	C	PGDSDADC	P	IIEIENL-SGCCCHHRAG-- 13
<i>Enterobacter cloacae</i>	100	MREQLQLAES	C	PGDSDAGC	P	IIDNL-SGCCCHRKTHA-- 13
<i>Citrobacter freundii</i>	112	MRLHLLALADS	C	PGDSDAEC	P	IIMDNL-SGCCCHHKAKA-- 148
<i>Pantoea ananatis</i>	100	MRARLLALAAA	C	PGNDSADC	P	IINNLA-GCCSQVGGTRD 138
<i>Klebsiella oxytoca</i>	100	MRTQLLALADS	C	PGDSDADC	P	IIDNL-SGCCCHHRKAQA-- 136

Mass spectrometry

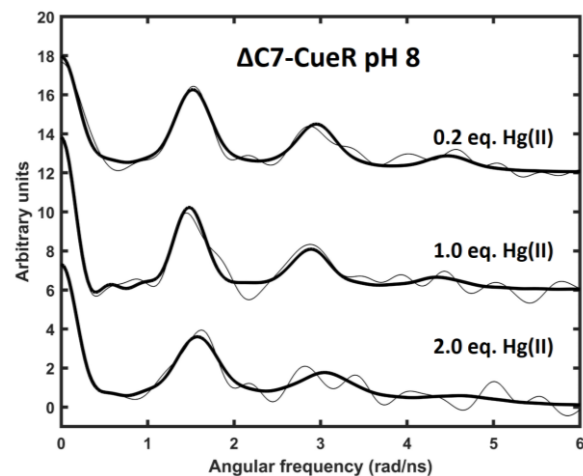
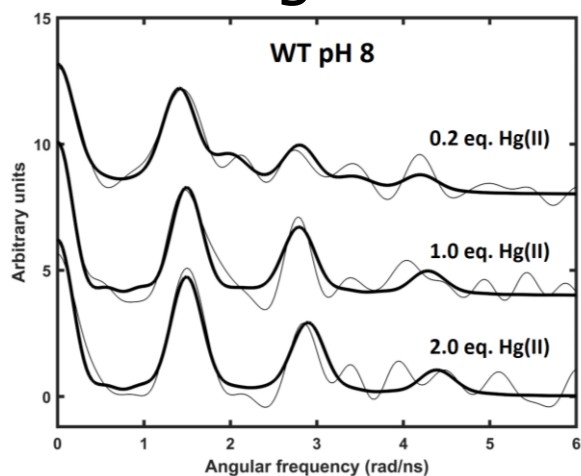


Balogh *et al.*, Chem. Eur. J., 2019, 25, 15030 – 15035

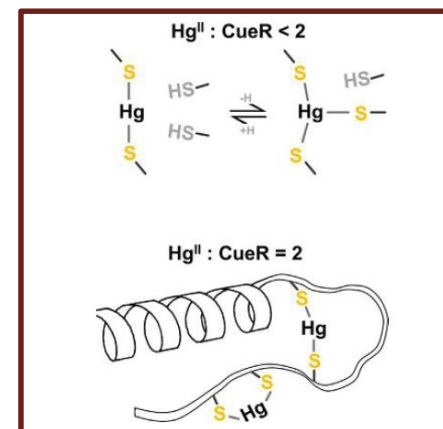
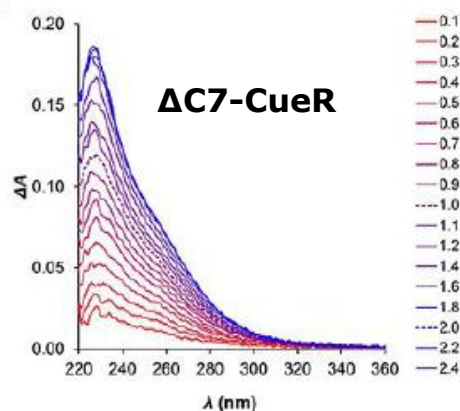
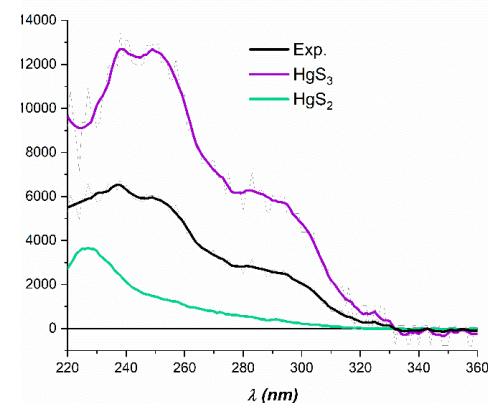
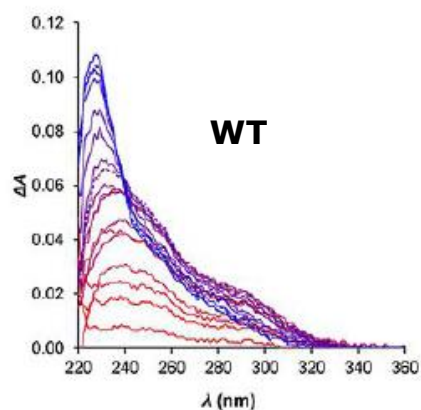


CueR metal binding: How are mono- and divalent metal ions discriminated?

^{199m}Hg PAC



UV absorption



Research progress - ongoing projects

- **Sensor proteins**
CueR, ArsR, Attila Jancso, Uni of Szeged, Hungary
- **Zinc-hooks & DNA repair**
Artur Krezel, Marek Luczkowski, Uni of Wroclaw, Poland
- **γ -Crystallins and cataract of the eye**
Liliana Quintanar, Cinvestav, Mexico
- **De novo designed proteins**
Vincent L. Pecoraro, Uni of Michigan, USA
- **The HAH1 Cu(I) transporter**
David Hufmann, Uni of Western Michigan, USA



Difficulties encountered

- ^{111}Ag
 - Implantation into ice failed, activity mainly on sample holder
 - Implantation into and extraction from polyethylene failed (activity remained in polyethylene)
 - Alternative production (at ILL) worked

- $^{68\text{m}}\text{Cu}$ (not done within IS602)
 - Implantation into Cu_2O demonstrated that the electric quadrupole moment of the relevant nuclear state is small [Fenta et al. EPL 115 (2016) 62002] combined with short half life of intermediate state
 - => measurement of NQIs in biomolecules will be difficult



Conclusions

- Metal ion specificity for the CueR metal ion sensor protein explored & published ($^{199\text{m}}\text{Hg}$ PAC)
- Several ongoing projects, some of which will be completed with the remaining 9½ shifts
- ^{111}Ag and $^{68\text{m}}\text{Cu}$ abandoned (for now)
- Focus for remaining 9½ shifts mainly on $^{199\text{m}}\text{Hg}$ PAC spectroscopy, with $^{111\text{m}}\text{Cd}$ and $^{204\text{m}}\text{Pb}$ as alternative isotopes (reply to TAC question)



University of Szeged, Hungary

Collaborations



Attila Jancso



Béla Gyurcsik,



Ria K. Balogh



Artur Krezel



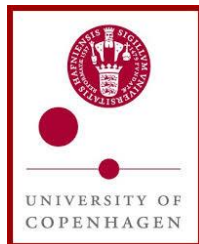
David Hufmann



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Christensen



Karl
Johnston



Joao
G.M. Correia



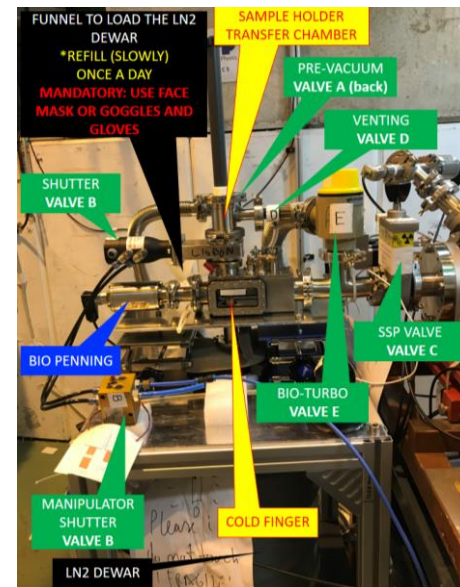
Juliana
Schell



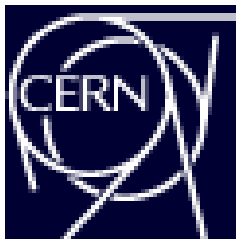
Vincent L. Pecoraro



Liliana Quintanar



Funding



Bundesministerium
für Bildung
und Forschung



**DET FRIE
FORSKNINGSRÅD**
DANISH COUNCIL
FOR INDEPENDENT
RESEARCH

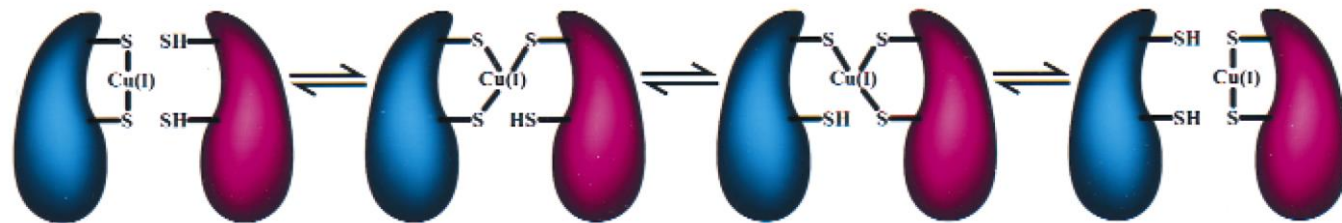
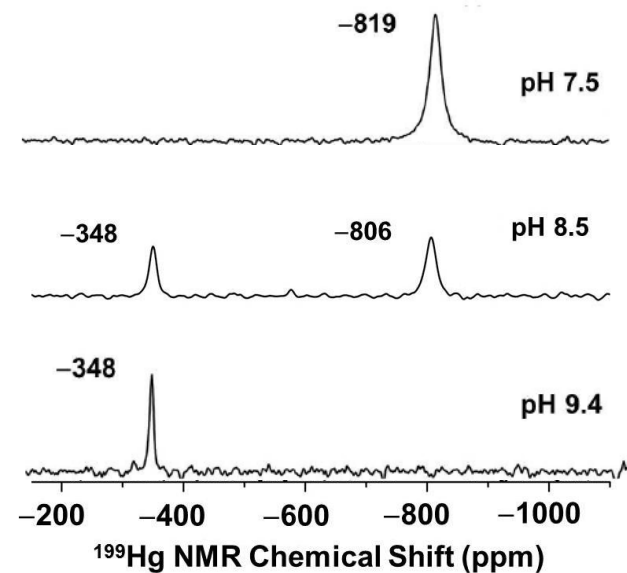
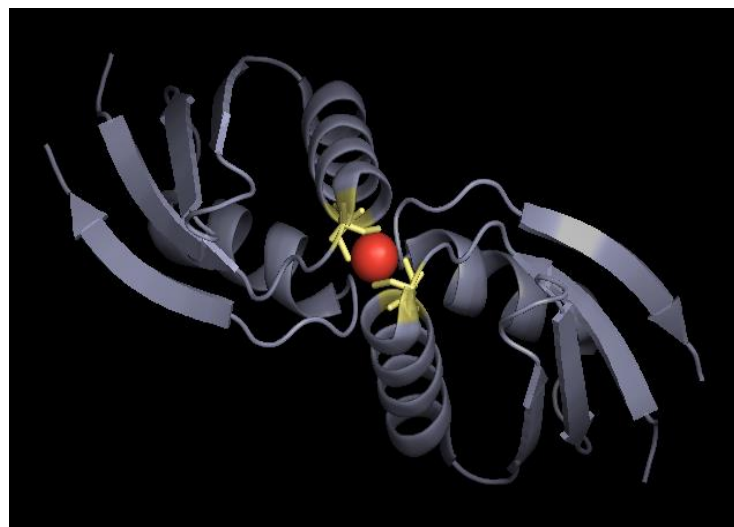
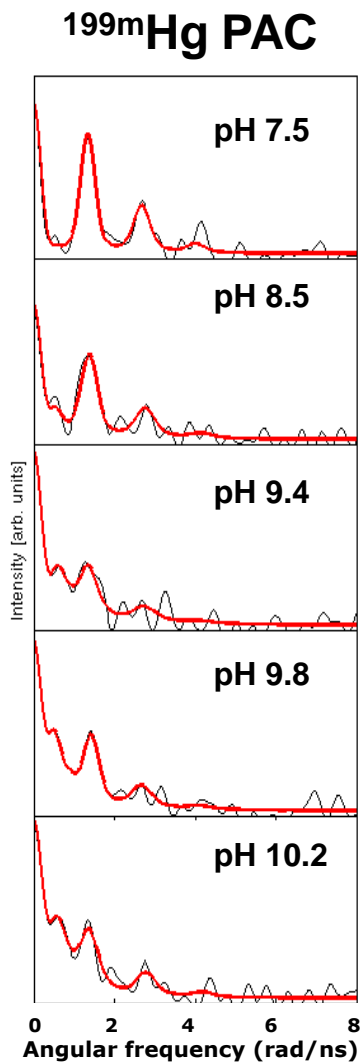


**Danish Agency for Science
Technology and Innovation**

Ministry of Science
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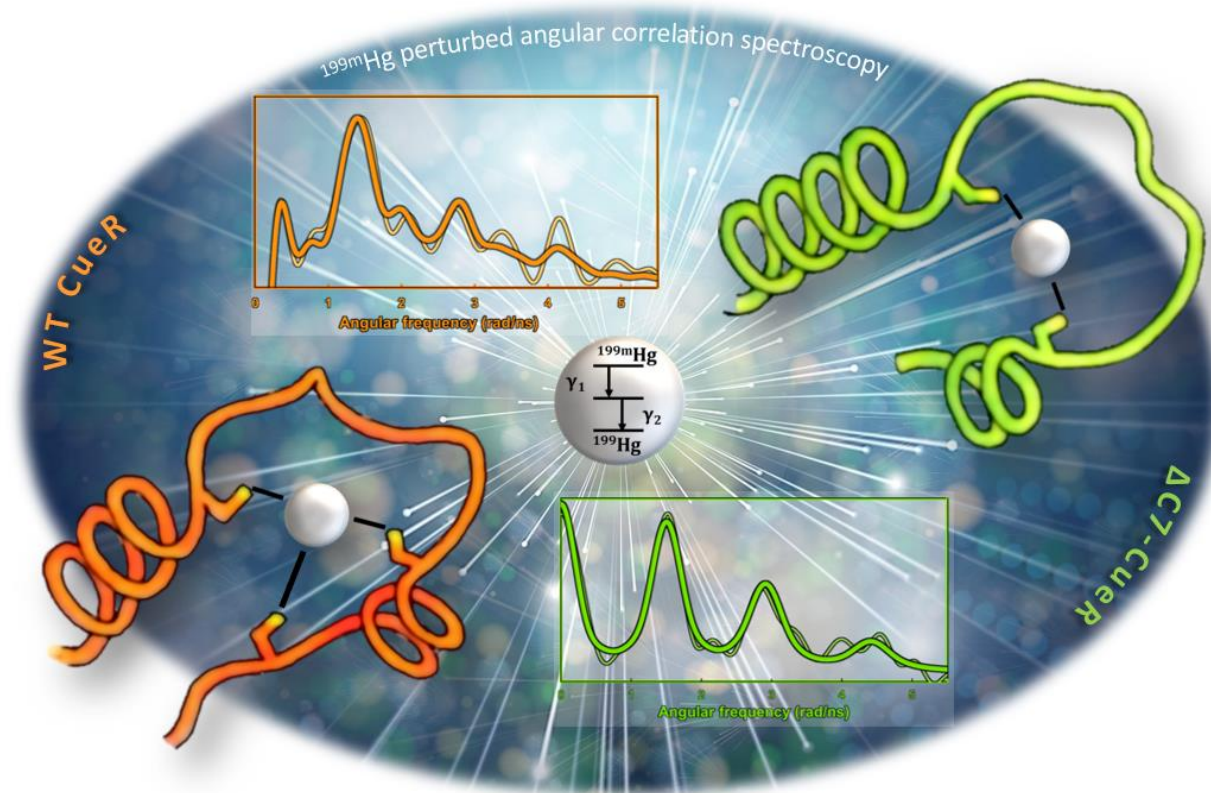
Metal ion transfer between proteins: The Cu(I) binding protein HAH1



Wernimont et al. Nat. Struct. Biol., 2008, 102, 114; Luczkowski et al., Chem. Eur. J. 2013, 19, 9042



CueR metal binding: How are mono- and divalent metal ions discriminated?



By recruiting auxiliary ligands for divalent metal ions, disrupting the functional protein structure