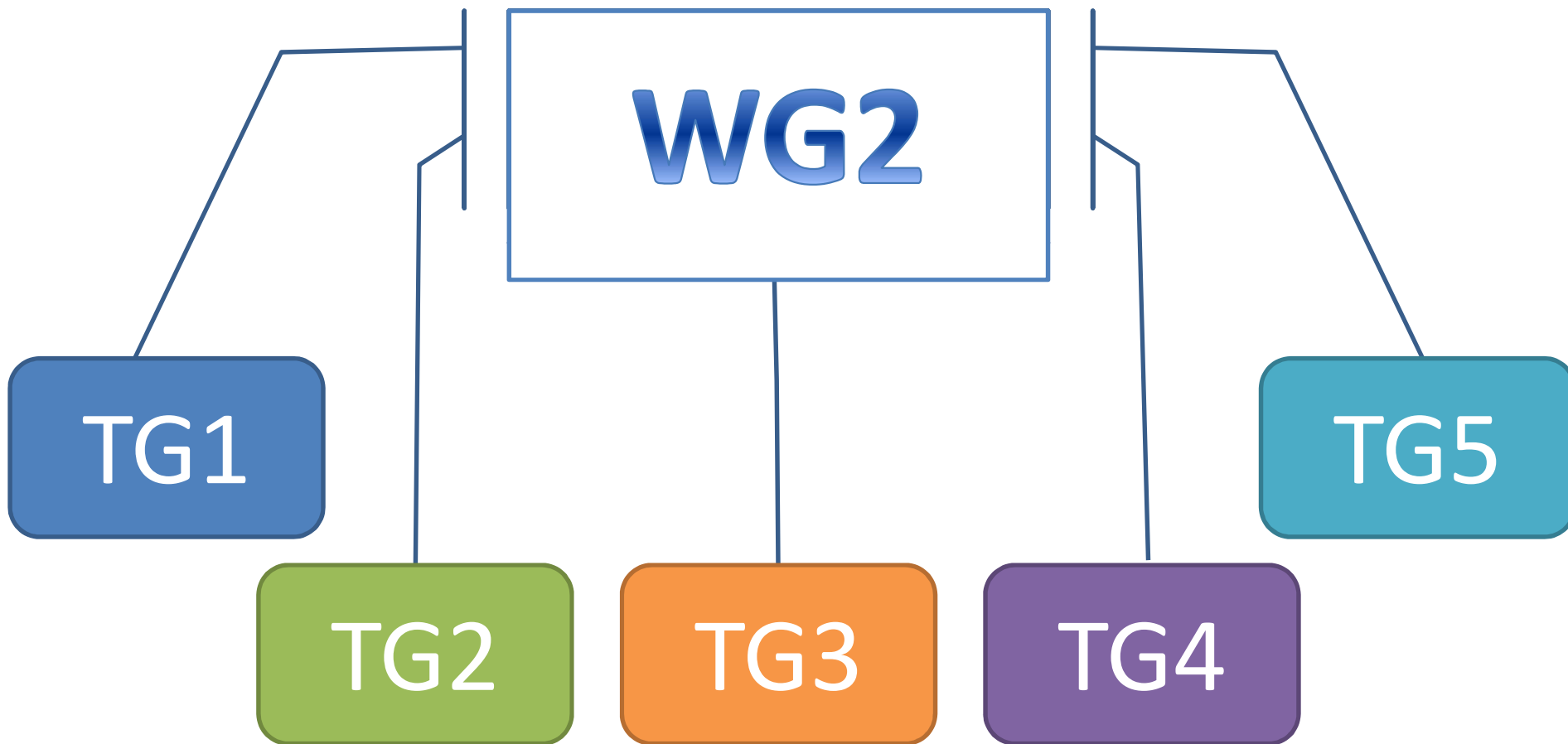




WG2

NECTAR for strong and/or multifunctional ligands, macromolecules, polyelectrolytes



TG1

Task Group 1

Complexes of polydentate ligands



Working Group 2

TASK GROUP 1

Complexes of polydentate ligands
How to deal with complexones and macrocycles ?

Potentiometric „good laboratory practice“ for titrations of polydentate ligand systems

What was taken into account

1. Materials and stock solutions:

- strong acid & base stock solutions
- ligand & metal ion purity and stock solutions
- choice of background electrolyte

2. Electrode system calibration:

- pH range of calibration titration and calibration parameters
- data at high & low pH

3. Titrations:

- detailed experimental set-up (suitable pH ranges / concentrations, M:L ratios)
- calibration-titration relations

4. Data treatment:

- data precision & titration reproducibility, a number of titration & data points
- programs for data treatment & statistical parameters
- choice of chemical model

Test systems: EDTA and EDTA-Zn²⁺

- EDTA chosen as suitable ligand – easily available, no problems with reaction kinetics, suitable necessary titration pH range, easy to deal with ...
- Chemical model with H⁺/Zn²⁺ is reasonably educative => several protonations in acid solution, protonated complexes & hydroxido species
- Conditions tested: pH ranges 1.5-11.8 & 1.8-12, EDTA concentrations 0.001-0.005 M, hydroxide concentrations, 0.1 & 0.2 M, ionic strength 0.1 & 0.2 M
- Conditions chosen: pH range at least 1.7 to at least 11.5, c(EDTA) 0.002 & 0.003 M, I = 0.1 M KCl, c(KOH) 0.1–0.2 M, Zn:L ratio 0.95–0.98:1, t = 25 °C
- Other recommended conditions: in $-\log[\text{H}^+]$, four-parameter electrode calibration, calibration followed by titration, at least 3+3 titration for each EDTA concentration, 50–60 points per titration

Data obtained & evaluation

- Four labs participated – Debrecen Messina, Prague, Turin.
- Different programs used for data treatment : BSTAC, OPIUM, PSEQUAD
- Global fitting done by each group, detailed statistical analysis of all data done by Matteo Marafante (Torino).
- Reasonable agreement between data obtained by the groups.
- Results in agreement with published data & procedure can be recommended as “good laboratory practice”.
- **However...** The most acidic EDTA protonation constant ($\log K_5 \sim 1.2$) should be determined – thus, titration should start from acidic solutions (pH 1.5–1.7). Otherwise, $\log K_4$ value is not fully correct.
- **However...** The highest attention should be paid to a correct calibration of electrode systems, four-parameter calibration is highly recommended.

Final values

From detailed statistical analysis of all data done by Matteo Marafante (Torino).

Constant	Values Interval (this work)	Average (this work)	Confidence Interval (C.I.)	NIST database	IUPAC database
$\log K_1$	10.15–10.18	10.17	± 0.08	10.19	10.12
$\log K_2$	6.12–6.23	6.17	± 0.07	6.13	6.13
$\log K_3$	2.66–2.87	2.75	± 0.14	2.69	2.77
$\log K_4$	2.02–2.25	2.11	± 0.17	2.0	2.01
$\log K_5$	(0.9–1.34)	1.15	± 0.57	1.5	(1.4)
$\log K_6$	–	–	–	–	(0.1)

- **But missing...** How to deal with systems where complexation kinetics is slow

=> “batch” / “out-of-cell” titrations.



TG2



Task Group 2

BIOSUBSTRATES

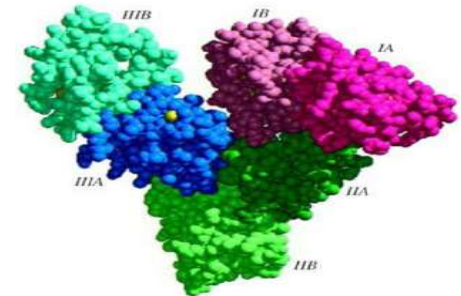
BINDING

Our task

Identify target systems for studies on biosubstrates binding to be used as validation standard. Development and testing of recommended procedures.

First step: dye/DNA intercalation

A target (“golden standard”) system is chosen on the basis of reactants stability and cost, availability and ease of handling. The different research groups will perform tests on the evaluation of binding constant for the target system. Results will be checked to perform intercalibration exercises between laboratories.



Paper work - Deliverables

The report was refined and split into 2 deliverables.

- (i) best practices/purity issues/experimental aspects
- (ii) data treatment/equations&software comparisons

Submitted on October 2022



The people working on this part is quite ready to submit a paper on this part.



DREAMS...

Demetrio proposed to produce a deliverable where the different buffers are listed with pros and cons (in particular given the differences we noticed at the beginning).



CALIXARENE SYSTEM to test data fitting

NEW CALIXARENE SYSTEM to test data fitting

Everybody of us was asked to fit a simple 1:1 titration data kindly shared by Nuno Basilio. We have used whatever equation and software and compare the logK number we get.

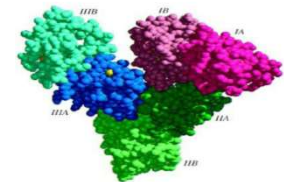
We demonstrated what we wanted (🤖 🧠): now, different people, using different data fit, obtained LogK numbers with little dispersion (± 0.1 Log units, ca. 1% error on Log K value, ca. 20% error on K value).

These data/thresholds can be used as a "general check" as for what is acceptable as an error on fluorescence titrations on more simple 1:1 systems. For DNA the error is (much) higher due to complexity of the system and use of simplified models.

PROTEIN BINDING: INTER-LABORATORY EXERCISES

ONGOING EFFORTS

- We have evaluated different systems and finally chosen ibuprofen/bovine serum albumin (IBU/BSA) and defined exactly the phosphate buffer to be used in our tests
- We have faced problems to define IBU molar extinction coefficient, but finally solved this point trough literature/measurements in different labs
- We have prepared a NEW PROTOCOL for BSA TITRATIONS
- Different groups have performed titrations
- We are now at the stage of fitting these titrations to compare our numbers on binding constant....



TG3

Task Group 3

Peptides

**COST Action CA18202
NECTAR**



Network for Equilibria and Chemical Thermodynamics Advanced Research

**4th European NECTAR Conference
and Final Action Meeting**

Milazzo, February 26th-27th, 2024



University of Wroclaw, Poland

Universitat Autoònoma de Barcelona, Spain

University of Siena, Italy

University of Balearic Islands, Spain

University of Opole, Poland

University of Ferrara, Italy

University of Sassari, Italy

University of Granada, Spain

NECTAR for strong and/or
multifunctional ligands,
macromolecules



Funded by
the European Union

Sławomir Potocki
University of Wrocław

MILAZZO February 2024

Histidine-rich C-terminal (HRCT) tail of GroEL1 chaperonins as a potential regulator of metal ions concentration in *Mycobacterium tuberculosis* – study on stoichiometry of metal-HRCT complexes



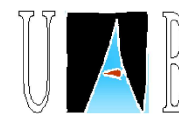
MSc Anna Rola
Origin Institution:
Faculty of Chemistry
University of Wrocław



The main aim of this STMS was to characterize the stoichiometry and preferred metal-binding sites of model peptides Cu(II)/Ni(II)-complexes in the presence of equimolar amount, as well as an excess of metal ions. For this purpose, **high-resolution mass spectrometry** was used.



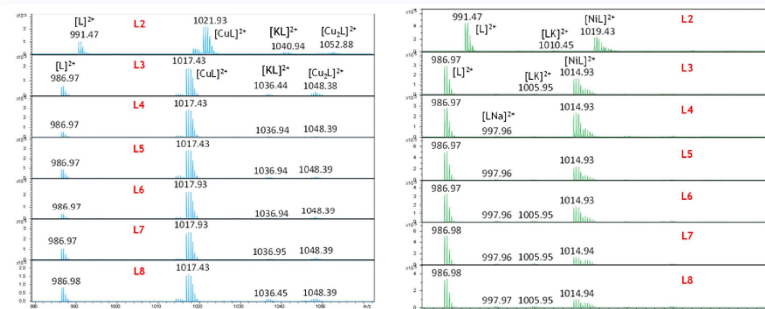
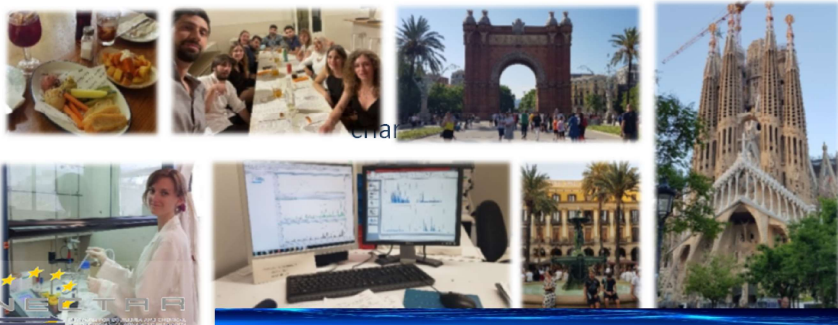
Prof. Oscar Palacios Bonilla
Host Institution:
Department of Chemistry
Universitat Autònoma de Barcelona



Universitat Autònoma de Barcelona



It was a great opportunity to learn new scientific skills, make international friendships and explore the catalan culture!



L2: Ac-DKPAKAEDHDDHHHGHAAH, L3: Ac-DKPAKAEDHQDHHHGHAAH, L4: Ac-DKPAKAEDHDQHHGHAAH
L5: Ac-DKPAKAEDHDHQHGHAAH, L6: Ac-DKPAKAEDHDDHQGHAAH, L7: Ac-DKPAKAEDHDDHHHQGHAAH
L8: Ac-DKPAKAEDHDDHHHGHAAHQ

MILAZZO February 2024

RETURN TO ISSUE | < PREV **ARTICLE** NEXT >

Histidine-Rich C-Terminal Tail of Mycobacterial GroEL1 and Its Copper Complex—The Impact of Point Mutations

Anna Rola*, Oscar Palacios, Merce Capdevila, Daniela Valensin, Elżbieta Gumienna-Kontecka, and Sławomir Potocki*

✓ **Cite this:** *Inorg. Chem.* 2023, 62, 18, 6893–6908
Publication Date: April 24, 2023
<https://doi.org/10.1021/acs.inorgchem.2c04486>

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 Supporting Info (1) »

SUBJECTS: Bacteria, Deprotonation, Metals, Monomers, Peptides and proteins

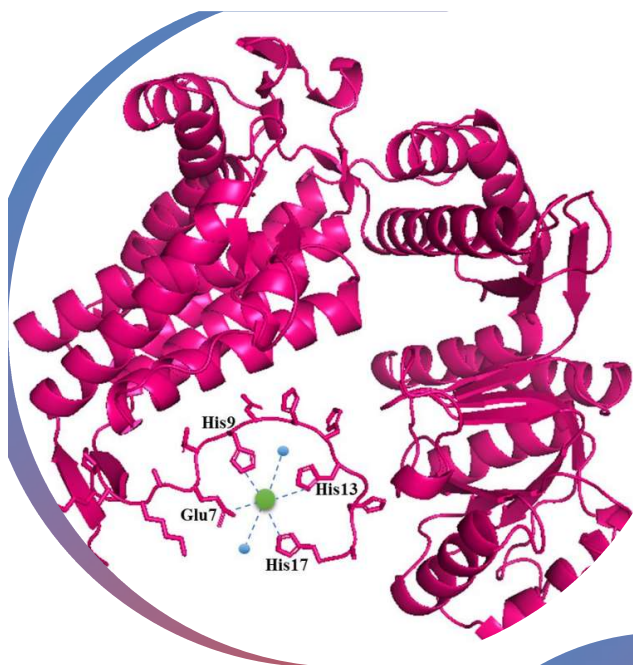


MILAZZO February 2024



**beyond Copper: Examining the Significance of His-Mutations
in Mycobacterial GroEL1 HRCT for Ni(II) Complex Stability
and Formation**

Journal:	<i>Dalton Transactions</i>
Manuscript ID:	DT-ART-01-2024-000011
Article Type:	Paper
Date Submitted by the Author:	02-Jan-2024
Complete List of Authors:	Rola, Anna; University of Wrocław, Faculty of Chemistry Kola, Arian; University of Siena, Department of Biotechnology, Chemistry and Pharmacy Valensin, Daniela; University of Siena, Department of Chemistry Palacios, Oscar; Universitat Autònoma de Barcelona, Chemistry Capdevila, Mercè; Universitat Autònoma de Barcelona, Departament Químic Guimienna-Kontecka, Elzbieta; University of Wrocław, Faculty of Chemistry Potocki, Slawomir; University of Wrocław, Department of Chem'



Coordination sphere for a Ni(II)-L2
(L2: Ac- DKPAKAEDHDHGHGHAH)
complex at pH around 7.1.

The presence of Lys5 residue significantly increases the stability of the system. His9 and His13 are the crucial residues for Ni(II) binding, whereas His12 has minimal relevance in complex formation.

STSM COST Nectar -

Exploiting NMR spectroscopy to determine coordination mode of the Zn(II), Ni(II), and Cu(II) complexes with metalloproteinases.



Origin Institution: University of Wrocław,
Poland, Department of Chemistry
MSc Paulina Potok



CA18202 STSM
31 days

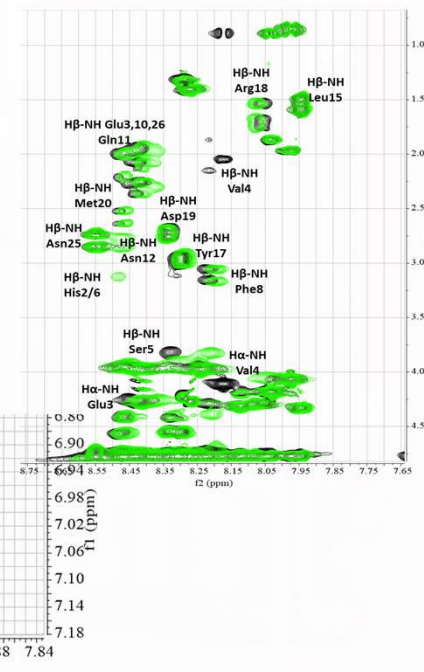
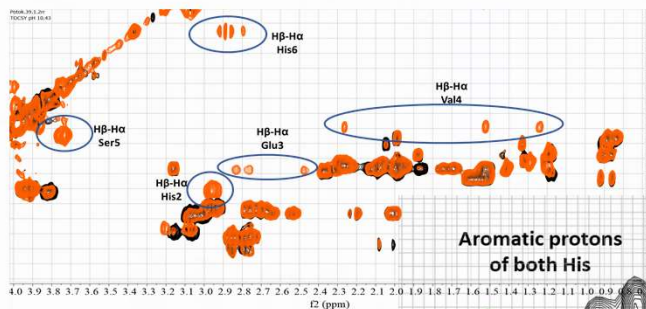


Host Institution: University of Siena,
Italy Department of Biotechnology,
Chemistry and Pharmacy
Prof. Daniela Valensin



DIPARTIMENTO DI BIOTECNOLOGIE,
CHIMICA E FARMACIA

The main goal of the proposed STSM project was to characterize the coordination mode of metalloproteinases from pathogenic bacteria with transition metal ions Zn(II), Ni(II), and Cu(II) by Nuclear Magnetic Resonance Spectroscopy.



MILAZZO February 2024

Copper Forms a PPII Helix-Like Structure with the Catalytic Domains of Bacterial Zinc Metalloproteases

Paulina Potok, Arian Kola, Daniela Valensin, Merce Capdevila, and Sławomir Potocki*

📄 Cite this: *Inorg. Chem.* 2023, 62, 45, 18425–18439

Publication Date: November 1, 2023

<https://doi.org/10.1021/acs.inorgchem.3c02391>

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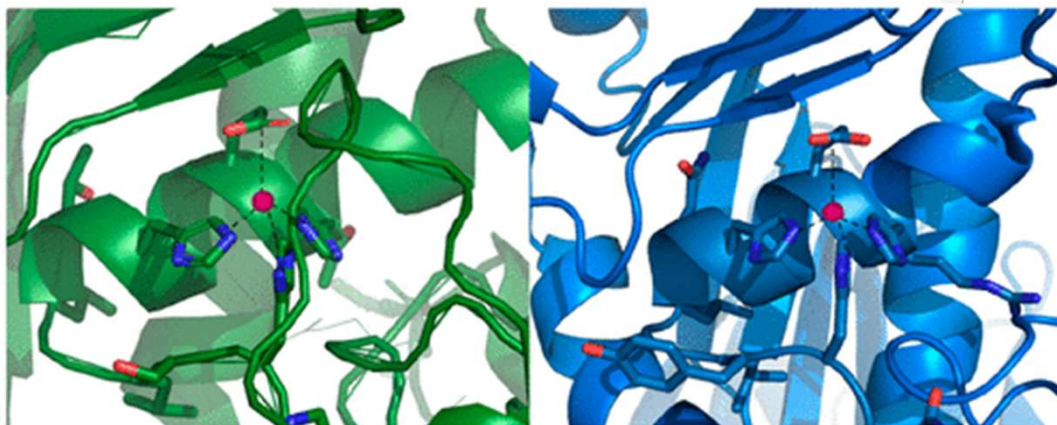
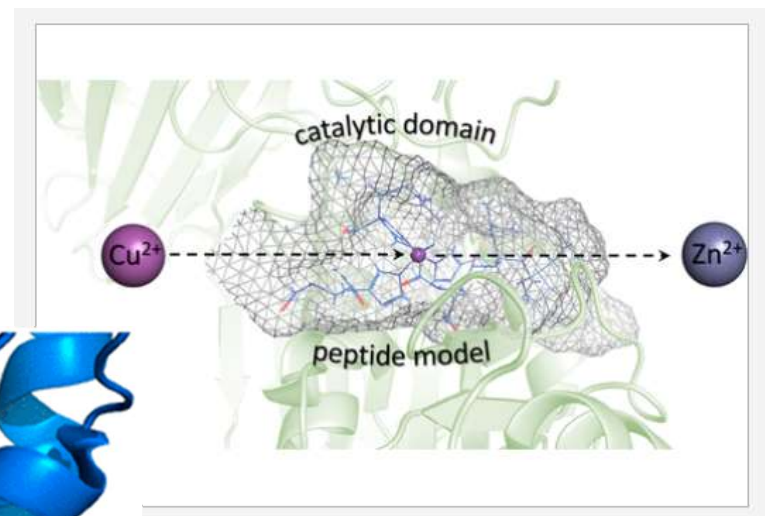
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**CH vs. HC-Promiscuous
Metal Sponges in
Antimicrobial Peptides and
Metallophores**



Garstka Kinga



Dzyhovskiy Valentyn



Wątył Joanna



Stokowa-Sołtys Kamila



Kozłowski Henryk

**Świątek-
Kozłowska
Jolanta**

University of Wrocław



University of Ferrara



**University of
the Balearic Islands**



Barceló-Oliver Miquel

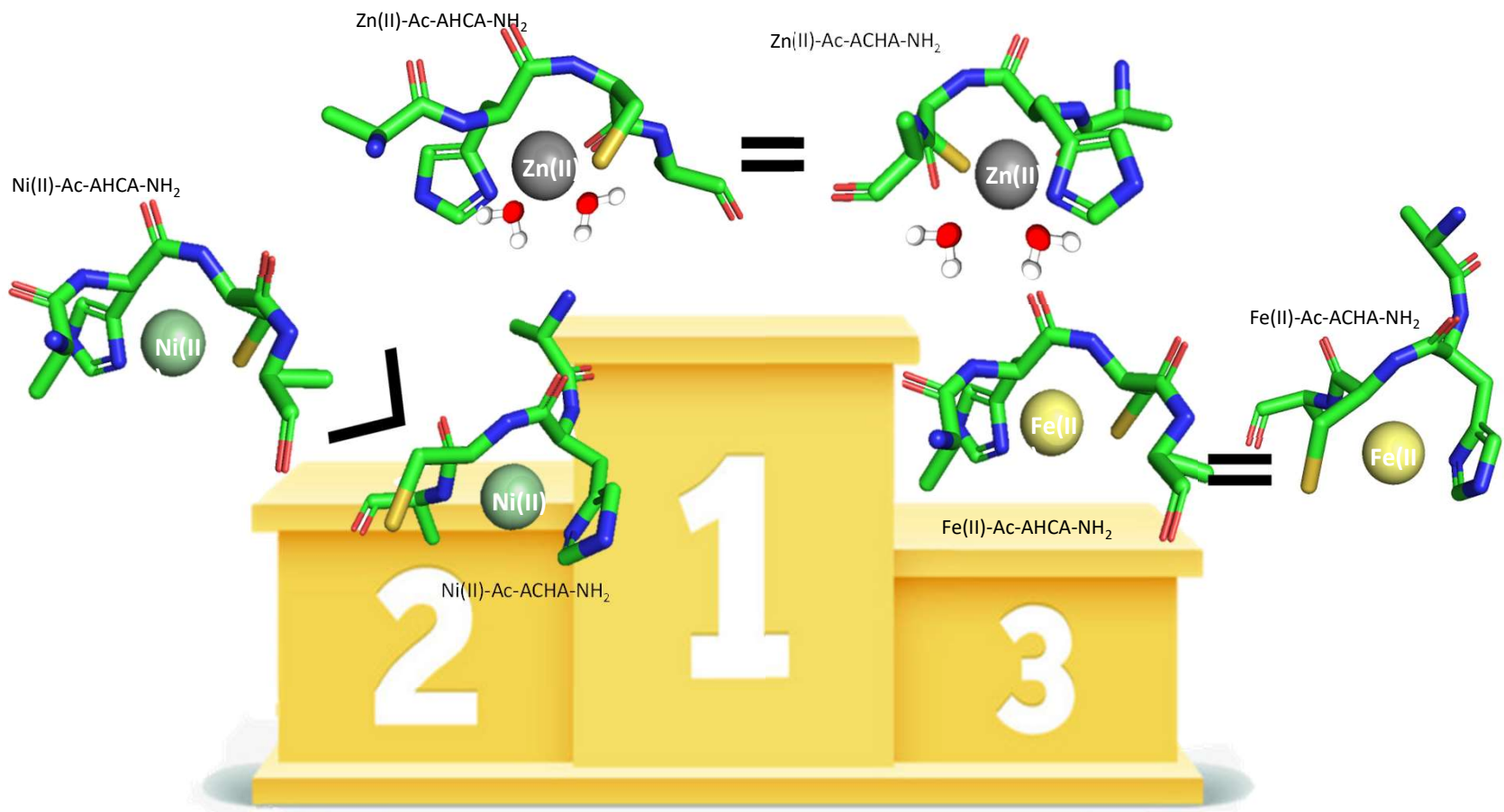


Bellotti Denise



Rowińska-Żyrek Magdalena

**Molecules, 2023, 28, 3985/1-3985/14
DOI:10.3390/molecules28103985**



Garstka, K.; Dzyhovskyi, V.; Wąty, J.; Stokowa-Sołtys, K.; Świątek-Kozłowska, J.; Kozłowski, H.; Barceló-Oliver, M.; Bellotti, D.; Rowińska-Żyrek, M. CH vs. HC—Promiscuous Metal Sponges in Antimicrobial Peptides and Metallophores, *Molecules*, 2023, **28**, 3985/1-3985/14, DOI:10.3390/molecules28103985

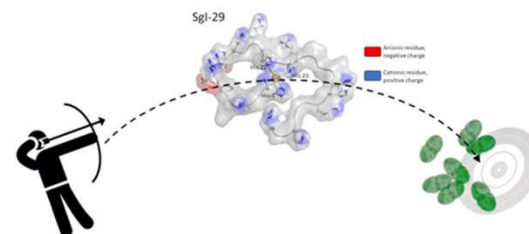
Semenogelins Armed in Zn(II) and Cu(II): May Bioinorganic Chemistry Help Nature to Cope with *Enterococcus faecalis*?

Dorota Dudek, Adriana Miller, Aleksandra Hecel, Arian Kola, Daniela Valensin, Aleksandra Mikołajczyk, Miquel Barcelo-Oliver, Agnieszka Matera-Witkiewicz, and Magdalena Rowińska-Żyrek*

[Cite this:](#) *Inorg. Chem.* 2023, 62, 34, 14103–14115
 Publication Date: August 15, 2023
<https://doi.org/10.1021/acs.inorgchem.3c02390>
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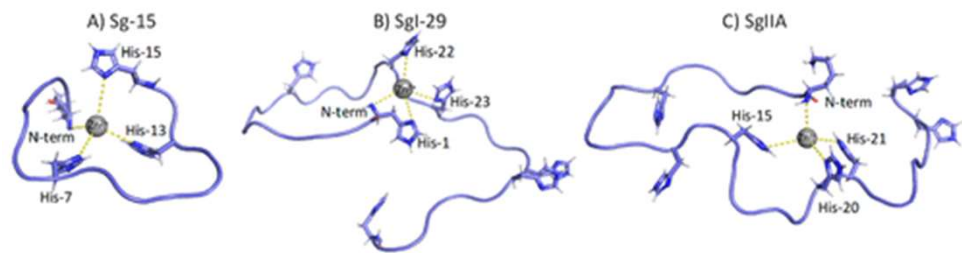


The Cu(II)-induced antimicrobial activity of Sg-15 against *Enterococcus faecalis* is shown.

The most abundant proteins from human semen, results in the formation of 26- and 29-residue peptides (Sg15 and Sg29, respectively), which share a common 15-residue region.



Sg1-29	HNKQEGRDHDKSKGFHHRVVIHHKGGKAH
Sg11A	KQEGRDHDKSKGFHMIVIHKGGQAHHG
Sg15 (common region)	KQEGRDHDKSKGFH



MILAZZO February 2024

Joint work on the characterization of Fe(II) bacterial transporters



Bartosz Orzeł
MSc



Prof. Elżbieta Gumienna-
Kontecka



Uniwersytet
Wrocławski



Prof. Massimiliano
Francesco Peana



Università degli Studi di Sassari



MILAZZO February 2024

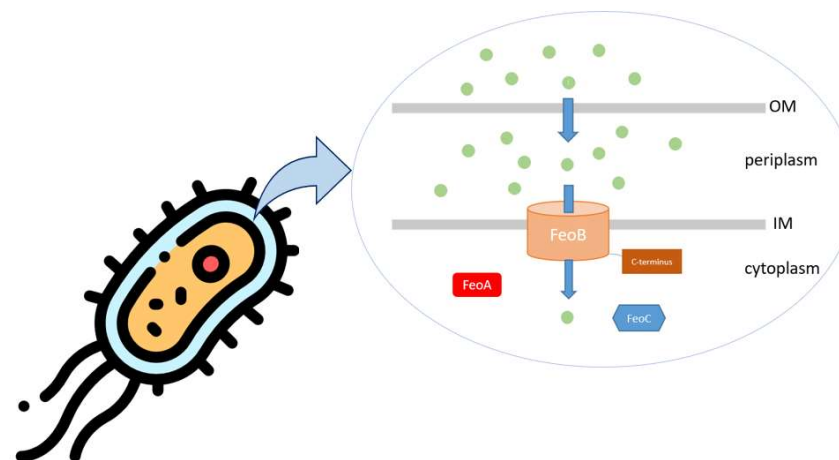
Fe(II), Mn(II), and Zn(II) Binding to the C-Terminal Region of FeoB Protein: An Insight into the Coordination Chemistry and Specificity of the *Escherichia coli* Fe(II) Transporter

Bartosz Orzel, Alessio Pelucelli, Malgorzata Ostrowska, Slawomir Potocki, Henryk Kozlowski, Massimiliano Peana, and Elzbieta Gumienna-Kontecka*

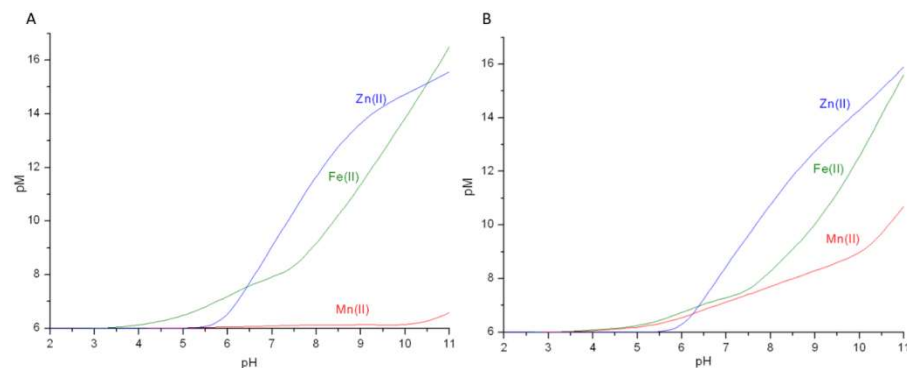
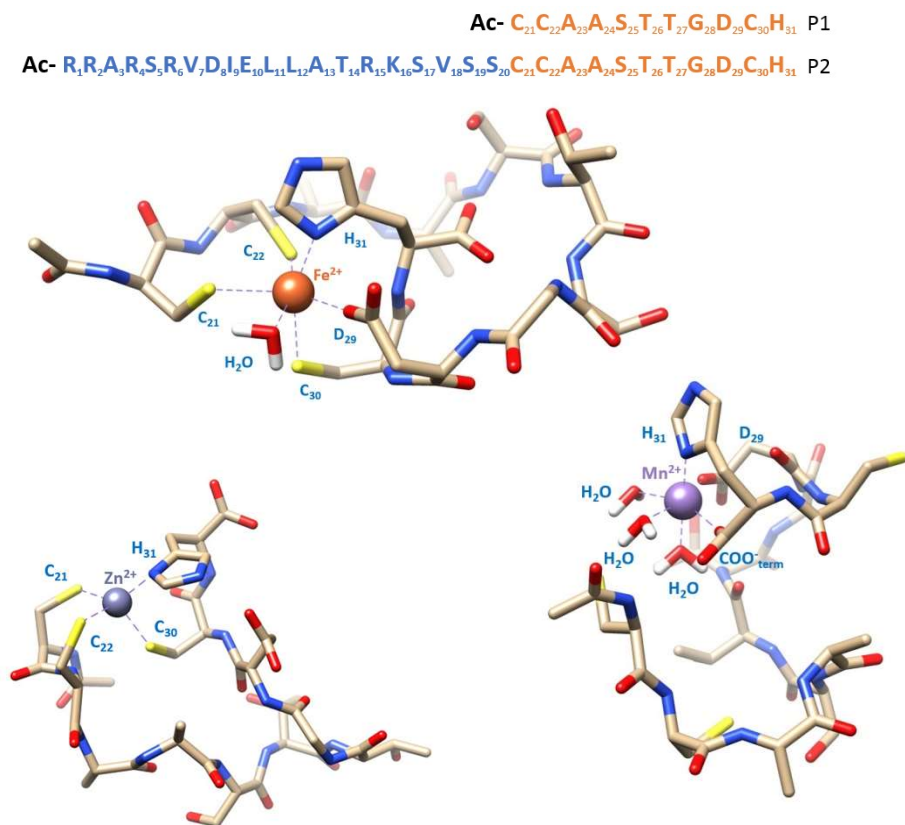
Cite This: *Inorg. Chem.* 2023, 62, 18607–18624

Read Online

Iron(II) bacterial transporters, such as **Feo system**, are crucial for bacterial survival and pathogenicity.



B. Orzeł, A. Pelucelli, M. Ostrowska, S. Potocki, H. Kozłowski, M. Peana, E. Gumienna-Kontecka, *Inorganic Chemistry* **2023**, 62(45), 18607–18624.



Comparison of K_d values for studied and biological ligands for Fe(II), Zn(II) and Mn(II).^a

Ligand	Fe(II)	Mn(II)	Zn(II)	Ref.
P2	$4.75 \cdot 10^{-7}$	$7.02 \cdot 10^{-7}$	$6.31 \cdot 10^{-8}$	This work
<i>E. coli</i> Fur	$1.2 \cdot 10^{-6}$	$2.4 \cdot 10^{-5}$	$1.4 \cdot 10^{-10}$	78
<i>S.pyogenes</i> MtsA	$4.3 \cdot 10^{-6}$	-	-	79
<i>B.subtilis</i> MntR	-	$0.2 \cdot 10^{-6} - 2 \cdot 10^{-6}$	-	80
<i>Y.pestis</i> YfeA	-	$1.78 \cdot 10^{-8}$	$6.6 \cdot 10^{-9}$	81
<i>T.pallidum</i> TroA	-	$7.1 \cdot 10^{-9}$	$2.25 \cdot 10^{-8}$	82
<i>D.radiodurans</i> MntH	-	$1.9 \cdot 10^{-4}$	-	83
<i>Synechocystis</i> ZnuA	-	-	$7.3 \cdot 10^{-9}$	84

^a K_d values calculated for our systems as: $K_d = \frac{[M][L]}{[ML]}$ at pH=7.0.

“Histidine-Rich C-Terminal Tail of Mycobacterial GroEL1 and Its Copper Complex—The Impact of Point Mutations.”

A. Rola, O. Palacios, M. Capdevila, D. Valensin, E. Gumienna-Kontecka & S. Potocki; *Inorganic Chemistry* (2023), **62**, 6893–6908. DOI: [10.1021/acs.inorgchem.2c04486](https://doi.org/10.1021/acs.inorgchem.2c04486)

(collaboration: University of Wrocław, Poland + Universitat Autònoma de Barcelona, Spain + University of Siena, Italy)

“Semenogelins Armed in Zn(II) and Cu(II): May Bioinorganic Chemistry Help Nature to Cope with Enterococcus faecalis?.”

D. Dudek, A. Miller, A. Hecel, A. Kola, D. Valensin, A. Mikołajczyk, M. Barcelo-Oliver, A. Matera-Witkiewicz & M. Rowińska-Żyrek; *Inorganic Chemistry* (2023), **62**, 14103–14115.

DOI: [10.1021/acs.inorgchem.3c02390](https://doi.org/10.1021/acs.inorgchem.3c02390)

(collaboration: University of Wrocław and Wrocław Medical University, Poland + University of Siena, Italy + University of Balearic Islands, Spain)

“CH vs. HC—Promiscuous Metal Sponges in Antimicrobial Peptides and Metallophores.”

K. Garstka, V. Dzyhovskiy, J. Wąty, K. Stokowa-Sołtys, J. Świątek-Kozłowska, H. Kozłowski, M. Barceló-Oliver, D. Bellotti & M. Rowińska-Żyrek; *Molecules* (2023), **28**, 3985/1–3985/14.

DOI: [10.3390/molecules28103985](https://doi.org/10.3390/molecules28103985)

(collaboration: University of Wrocław and University of Opole, Poland + University of Balearic Islands, Spain + University of Ferrara, Italy)

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P. Potok, A. Kola, D. Valensin, M. Capdevila & S. Potocki; *Inorganic Chemistry* (2023), **62**, 18425–18439. DOI: [10.1021/acs.inorgchem.3c02391](https://doi.org/10.1021/acs.inorgchem.3c02391)

(collaboration: University of Wrocław, Poland + University of Siena, Italy + Universitat Autònoma de Barcelona, Spain)

“Fe(II), Mn(II), and Zn(II) Binding to the C-Terminal Region of FeoB Protein: An Insight into the Coordination Chemistry and Specificity of the Escherichia coli Fe(II) Transporter.”

B. Orzel, A. Pelucelli, M. Ostrowska, S. Potocki, H. Kozłowski, M. Peana & E. Gumienna-Kontecka; *Inorganic Chemistry* (2023), **62**, 18607–18624. DOI: [10.1021/acs.inorgchem.3c02910](https://doi.org/10.1021/acs.inorgchem.3c02910)

(collaboration: University of Wrocław and University of Opole, Poland + University of Sassari, Italy)

“The N-terminal domain of Helicobacter Pylori’s Hpn protein: the role of multiple histidine residues.”

D. Bellotti, A. Sinigaglia, R. Guerrini, E. Marzola, M. Rowińska-Żyrek, M. Remelli; *J. Inorg. Biochem.* (2021), **214**, 111304. DOI: [10.1016/j.jinorgbio.2020.111304](https://doi.org/10.1016/j.jinorgbio.2020.111304)

(collaboration: University of Ferrara, Italy + University of Wrocław, Poland)

“Novel insights into the metal binding ability of ZinT periplasmic protein from Escherichia coli and Salmonella enterica.”

D. Bellotti, M. Rowińska-Żyrek, M. Remelli; *Dalton Trans.* (2020), **49**, 9393–9403. DOI: [10.1039/D0DT01626H](https://doi.org/10.1039/D0DT01626H)

(collaboration: University of Ferrara, Italy + University of Wrocław, Poland)

“Exploring the specificity of rationally designed peptides reconstituted from the cell-free extract of Deinococcus radiodurans toward Mn(II) and Cu(II).”

M. Peana, E. Gumienna-Kontecka, F. Piras, M. Ostrowska, K. Piasta, K. Krzywoszyńska, S. Medici, M. A. Zoroddu; *Inorg. Chem.* (2020), **59**, 4661–4684. DOI: [10.1021/acs.inorgchem.9b03737](https://doi.org/10.1021/acs.inorgchem.9b03737)

(collaboration: University of Wrocław, Poland + University of Sassari, Italy + Public Higher Medical Professional School, Poland)

“Zinc(II) – the overlooked éminence grise of chloroquine’s fight against COVID-19?”

A. Hecel, M. Ostrowska, K. Stokowa-Sołtys, J. Wąty, D. Dudek, A. Miller, S. Potocki, A. Matera-Witkiewicz, A. Domínguez-Martin, H. Kozłowski, M. Rowińska-Żyrek; *Pharmaceuticals* (2020), **13**, 228.

DOI: [10.3390/ph13090228](https://doi.org/10.3390/ph13090228)

(collaboration: University of Granada, Spain + University of Wrocław, Poland + Public Higher Medical Professional School, Poland)





TG4

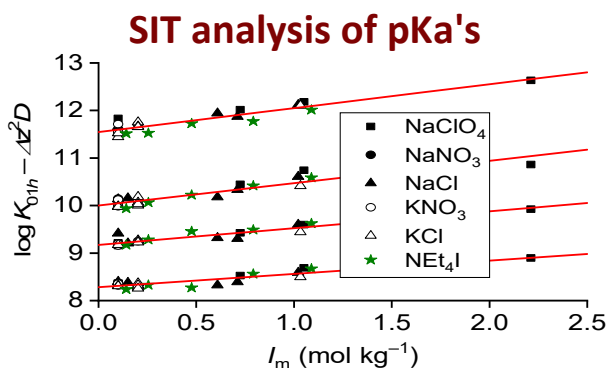
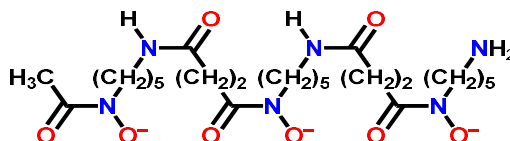
Task Group 4

Metallophores

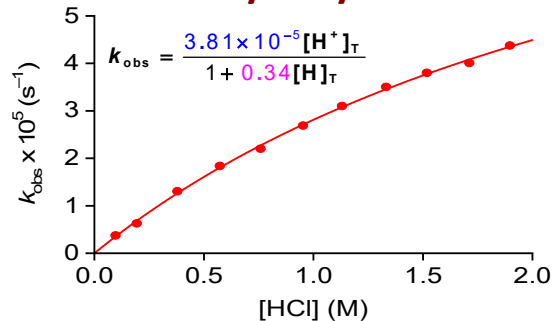
New Insights into the Acid-Base Properties of DFO



Streptomyces sp.



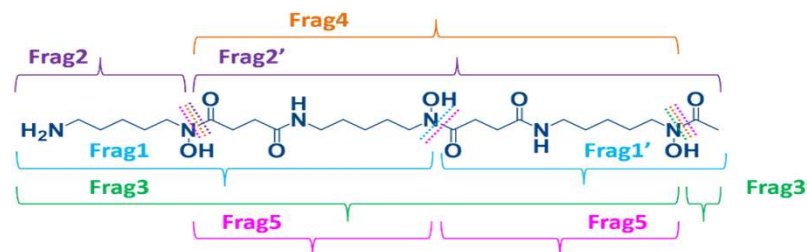
H⁺-assisted hydrolysis kinetics



New pKa measurements in NET₄I

<i>I</i> (M)	log <i>K</i> ₁ ± 2σ	log <i>K</i> ₂ ± 2σ	log <i>K</i> ₃ ± 2σ	log <i>K</i> ₄ ± 2σ
0.138	10.79 ± 0.03	9.46 ± 0.02	8.94 ± 0.02	8.25 ± 0.03
0.243	10.67 ± 0.02	9.50 ± 0.02	9.01 ± 0.02	8.35 ± 0.02
0.436	10.73 ± 0.02	9.57 ± 0.01	9.15 ± 0.01	8.31 ± 0.02
0.689	10.67 ± 0.03	9.70 ± 0.01	9.16 ± 0.01	8.62 ± 0.02
0.905	10.85 ± 0.04	9.84 ± 0.01	9.29 ± 0.02	8.75 ± 0.03

LCMS analysis of the fragmentation pattern



Metal-complexes as Potential drugs

Journal of Molecular Liquids 396 (2024) 124027



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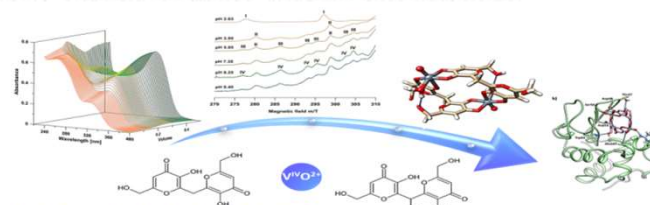
Journal of Molecular Liquids

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Solution chemistry of oxidovanadium(IV) complexes with two bis-kojic acid derivatives

Rosita Cappai ^{a,b,*}, Alessandra Fantasia ^b, Giuseppe Sciortino ^c, Daniele Sanna ^d, Federico Pisanu ^e, Eugenio Garribba ^e, M. Amélia Santos ^f, Guido Crisponi ^b, Valeria M. Nurchi ^b



Solution chemistry and molecular simulation studies on the oxidovanadium(IV) complexes with two bis-kojic acid derivatives



Article

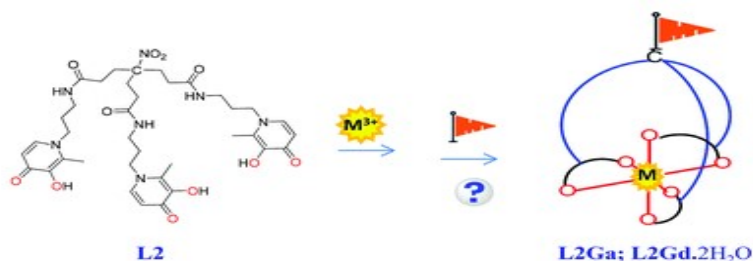
A Multi-Technique Investigation of the Complex Formation Equilibria between Bis-Deferiprone Derivatives and Oxidovanadium (IV)

Rosita Cappai ¹, Alessandra Fantasia ¹, Guido Crisponi ¹, Eugenio Garribba ², M. Amélia Santos ³ and Valeria Marina Nurchi ^{1,*}

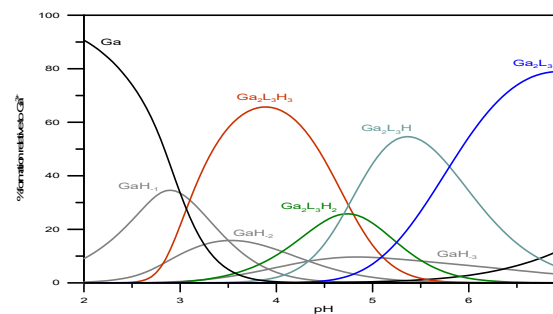
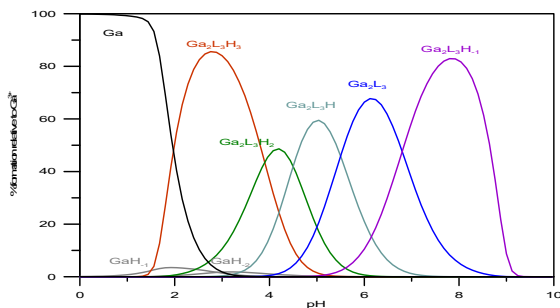
Metal-complexes as Potential drugs

Gd(III) and Ga(III) complexes with a new tris-3,4-HOPO ligand as new imaging probes: complex stability, magnetic properties and biodistribution

S. Chaves, K. Gwizdała, K. Chand, a L.Gano, A Pallier, É. Tóth, M. A. Santos, | *Dalton Trans.*, 2022, 51, 6436–6447, <https://doi.org/10.1039/D2DT00066K> (coll. Portugal, France , Poland)



Ga(III) complexes with biskojic acids as new metallophores: complex stability, and biological properties. D. Griffith, R. Cappai , M.A.Santos *et al.* (work in progress) (coll. Ireland , Italy , Portugal)

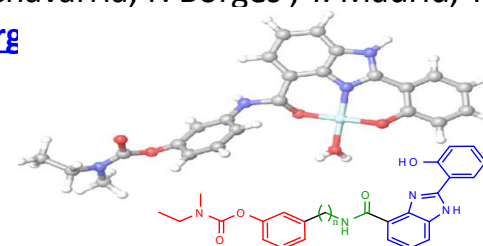
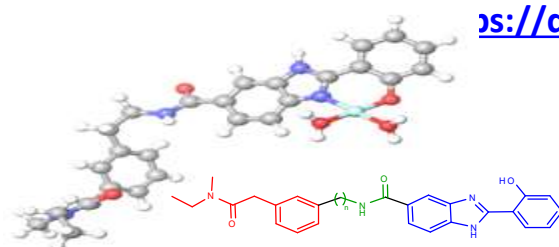
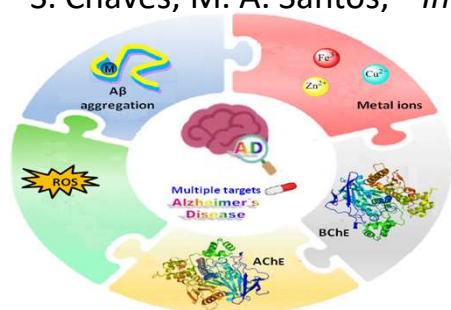


Multitarget metal chelators for potential therapy of Alzheimer's Disease (AD)

Rivastigmine–Benzimidazole Hybrids as Promising Multitarget Metal-Modulating Compounds for Potential Treatment of Neurodegenerative Diseases” ,

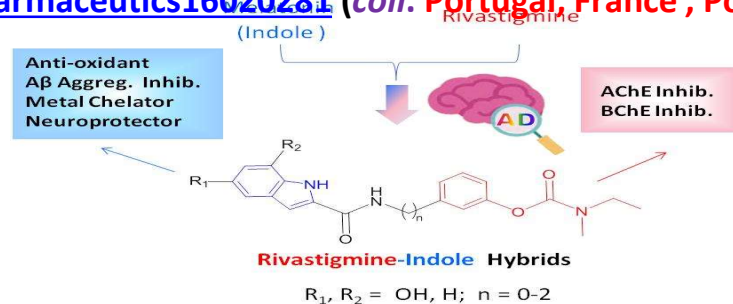
D. Vicente-Zurdo, L. Brunetti, L. Piemontese, B. Guedes, S.M Cardoso, D. Chavarria, F. Borges , Y. Madrid, Y. Madrid, S. Chaves, M. A. Santos, “ *Int. J.*

<https://doi.org>



New Multitarget Rivastigmine-Indole Hybrids as Potential Drug Candidates for Alzheimer's Disease” ,

L. Bon , A. Banás , I. Dias , I Melo-Marques , S. M. Cardoso, S. Chaves, M. A. Santos, *Pharmaceutics* **2024**, 16(2), 281; <https://doi.org/10.3390/pharmaceutics16020281> (coll. Portugal, France , Poland)



TG5

Task Group 5

Calorimetry



Milazzo, Feb
26-27 2024

WG2 Task Group n.5 on Calorimetry

**ITC study of multiple host-guest complex
formation:
an interlaboratory exercise**
Research groups from:

Catania, Ferrara, Firenze, Ljubljana,
Messina, Strasbourg, Udine, Wroclaw



MILAZZO February 2024

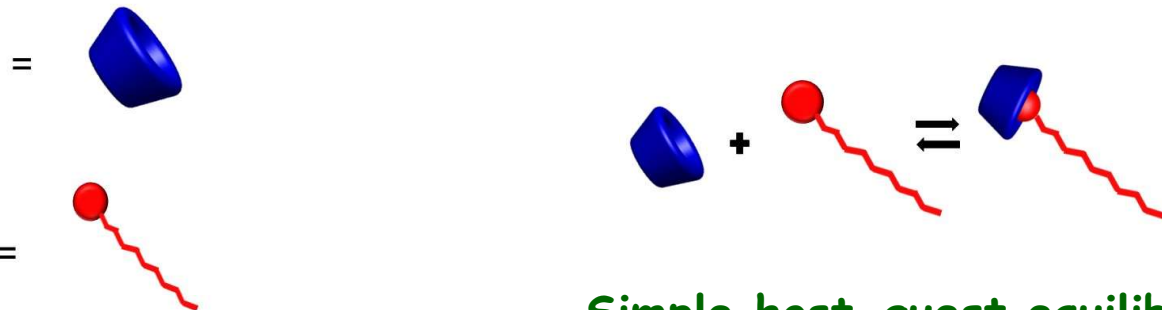
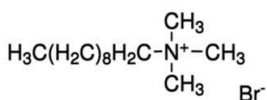
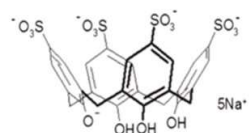
Main goals of WG2 - TG5

- Experimental design/conditions for the analysis of multiple equilibria
- Data refinement: use proper chemical models and software
- Critical comparison of data/results obtained by different labs, instruments, software
- Guidelines on the correct use of ITC data for the study of solution equilibria
- Training people: invite researchers to deal/interact with trained people, STSM

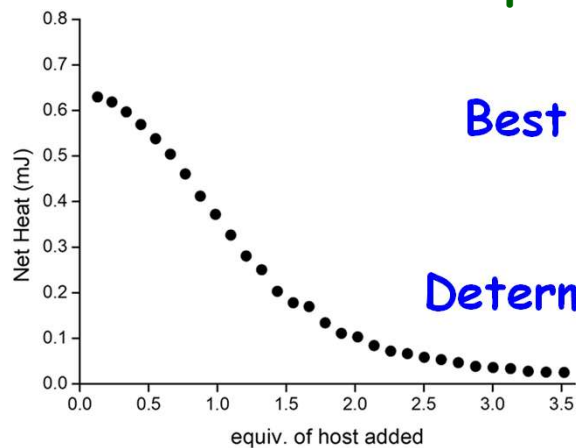
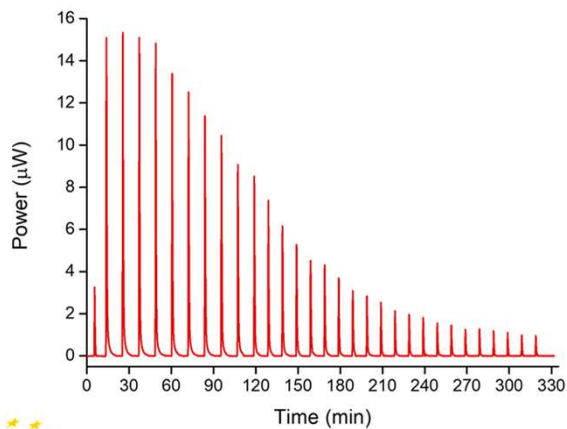
inter-laboratory exercise

«best practices» protocol

ITC titration for a host-guest 1:1 complex in buffered aqueous solution



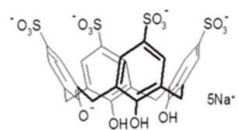
Simple host-guest equilibrium



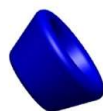
Best experimental design and data analysis model

Determination of accurate K and ΔH values

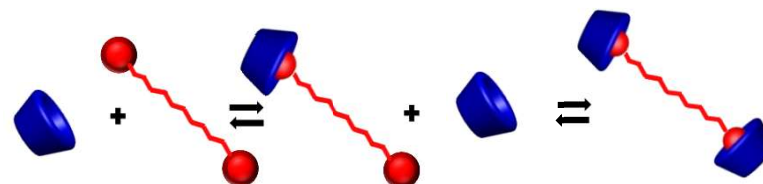
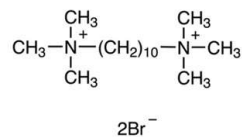
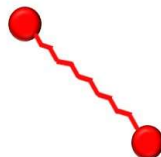
ITC titrations for host-guest 1:1 and 2:1 complexes in buffered aqueous solution



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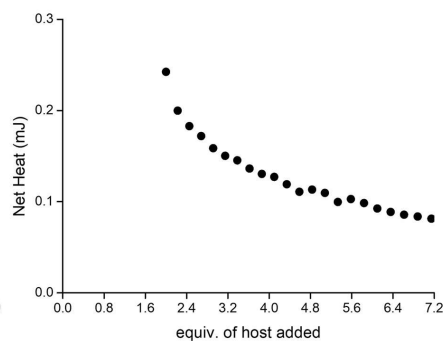
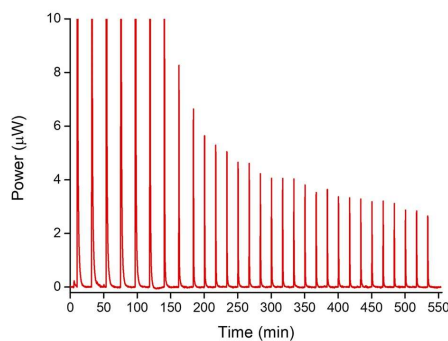
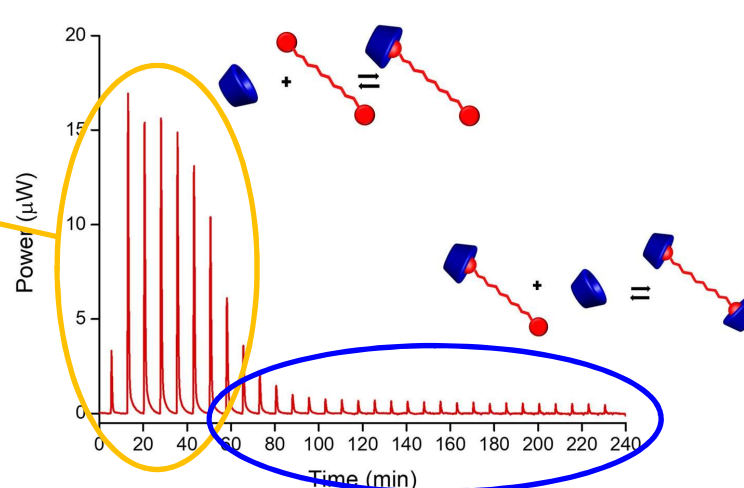
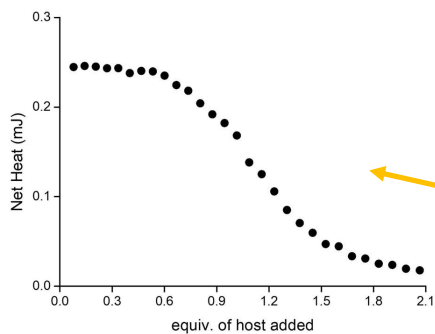
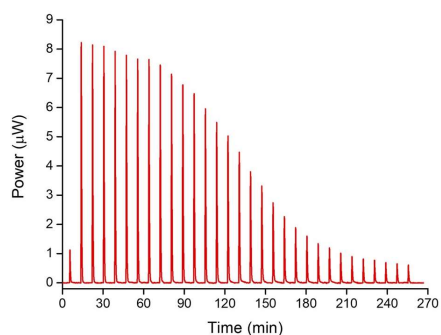


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Multiple host-guest equilibria

ITC titration for host-guest 1:1 and 2:1 complexes in buffered aqueous solution



Issues in the heat measurement!!

Careful experimental design needed for the determination of accurate K and ΔH values

WG2

Task Group n.5 on Calorimetry

- Proper design of ITC experiments to measure reliable heat values for the different complexation steps of multiple equilibria.
- The appropriate data refinement should include all the equilibria occurring in solution and the exact stoichiometry of the species rather than “obscure” binding models used by most software.
- The results from this exercise will contribute to prepare new **guidelines on correct experimental design and data analysis** in ITC titrations.



**THANK YOU FOR THE GREAT TIME!!
LET'S KEEP IN TOUCH**