

Royal Society of Chemistry

Dalton 2025

University of Warwick, 1-3 Apr 2025

Book of abstracts

IBGD Oral Presentations



ROYAL SOCIETY
OF **CHEMISTRY**

DALTON
COMMUNITY

We thank the following sponsors for their support:



The logo for fluorochem., consisting of the word 'fluorochem.' in a white, sans-serif font, with 'chem.' in orange, set against a dark blue square background.



INORGANIC CHEMISTRY

FRONTIERS

Molecular MRI with paramagnetic metal complexes

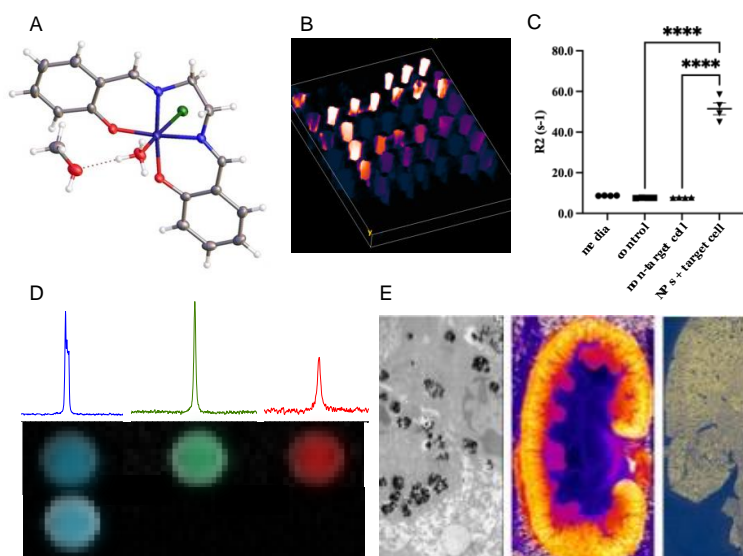
Peter Harvey^a

^aSchool of Chemistry & Sir Peter Mansfield Imaging Centre – School of Medicine, University of Nottingham

email: peter.harvey@nottingham.ac.uk

Treatments and diagnoses are severely limited by our inability to visualise the biochemical processes underlying disease. This issue is a particular challenge with inflammation, which impacts disease progression across most conditions. Our ability to monitor immune processes is often limited to fluorescence or histological methods, largely restricted to *in vitro* models that struggle to replicate the complex environment found in the body, or *ex vivo* snapshots that capture a single moment of time. New approaches are needed that combine biochemically specific *in vivo* real-time imaging with *in vitro* and *ex vivo* approaches.

Our efforts are focused on the design and application on novel contrast agents for magnetic resonance imaging (MRI) of molecular species. Key applications include detection of inflammatory biomarkers, tracking drug delivery, and small molecule agents for increased cell permeability. Our approach involves using the paramagnetic properties of lanthanides and transition metals as molecular and nanoparticle MRI contrast agents, with applications *in vitro*, *ex vivo*, and *in vivo*. We are particularly focused on the use of multimodal imaging and spectroscopy approaches towards multichannel imaging. Through this combinatorial imaging approach, we aim to use novel bioinorganic coordination chemistry to deepen understanding of biological processes across length scales - from subcellular to whole organ levels.



(A) Example of transition metal MR contrast agent scaffold. (B/C) Cell phenotype targeting with nanoparticle-based contrast agents. (D) Multichannel imaging with paramagnetic chemical shift. (E) Multimodal tissue imaging, supporting MRI with electron microscopy and mass spectrometry imaging.

1. P. Harvey, A. M. Blamire, J. I. Wilson, K N. A. Finney, A. M. Funk, P. K. Senanayake and David Parker, *Chem. Sci*, 2013, **4**, 4251-4258.
2. A. C. Sedgwick, J. T. Brewster, P. Harvey, D. A. Iovan, G. Smith, X-P. He, H. Tian, J. L. Sessler and T. D. James, *Chem. Soc. Rev.*, 2020, **49**, 2886-2915
3. S. Ghosh, P. Harvey, J. C. Simon and A. Jasanoff, *Curr. Opin. Neurobiol.*, 2018, **50**, 201-210.

Optimised diphosphine chelator platforms for ^{99m}Tc diagnostic and ^{188}Re therapeutic radiopharmaceuticals

Rachel E. Nuttall,^a Truc T. Pham,^a Ingebjørg N. Hungnes,^a Oliver W. L. Carter,^a Alex Rigby,^a Natasha Patel,^a Zilin Yu,^a Julie Cleaver,^b Jennifer D. Young,^{a,b} Jane Sosabowski,^b Paul G. Pringle^c and Michelle T. Ma^a

^a School of Biomedical Engineering & Imaging Sciences, King's College London, UK.

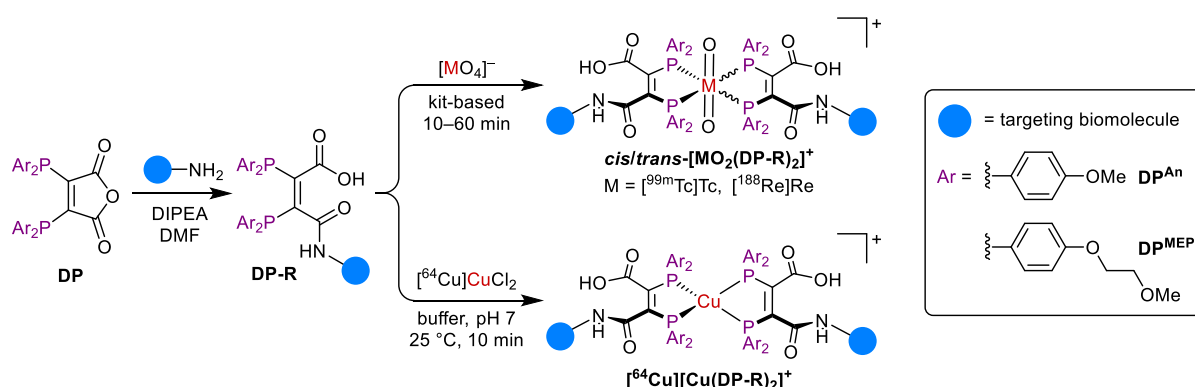
^b Centre for Cancer Biomarkers and Biotherapeutics, Barts Cancer Institute, Queen Mary University of London, UK.

^c School of Chemistry, University of Bristol, UK

email: rachel.nuttall@kcl.ac.uk

Nuclear medicine can be divided into SPECT imaging (e.g. ^{99m}Tc , γ emitter), PET imaging (e.g. ^{64}Cu , β^+ emitter) and radiotherapy (e.g. ^{188}Re , β^- emitter). By using the same ligand, analogous $^{99m}\text{Tc}/^{188}\text{Re}$ or $^{64/67}\text{Cu}$ complexes have potential for matched pair imaging and therapy. To develop receptor-targeted radiopharmaceuticals, versatile chelators that enable simple and rapid preparation of radiotracers are required. We have previously shown that diphosphines provide an adaptable platform for development of targeted ^{99m}Tc , ^{64}Cu and ^{188}Re radiopharmaceuticals.^{1–3} Our aim was to optimise this diphosphine technology.

Here we have synthesised two novel diphosphines, **DP^{An}** and **DP^{MEP}** (Scheme 1), which have been reacted with a wide range of biological targeting vectors. Exemplar diphosphine-bioconjugates were radiolabelled with ^{99m}Tc and ^{188}Re using clinically translatable procedures to form complexes of the type $[\text{MO}_2(\text{diphosphine})_2]^+$ ($\text{M} = ^{99m}\text{Tc}, ^{188}\text{Re}$). By tuning the aryl groups of the diphosphines, radiochemical yields notably increased from our first-generation diphosphine radiotracers.^{2,3} The new diphosphine-conjugates can also radiolabel ^{64}Cu , to form $[\text{Cu}][\text{Cu}(\text{diphosphine})_2]^+$. SPECT imaging and biodistribution studies of the receptor-targeted ^{99m}Tc complexes showed accumulation in tumours with low non-target uptake, high stability and favourable pharmacokinetics. This demonstrates the potential for these versatile **DP^{An}** and **DP^{MEP}** chelator platforms in the development of novel molecular imaging radiopharmaceuticals for ^{99m}Tc SPECT, ^{64}Cu PET and ^{188}Re systemic radiotherapy.



Scheme 1: Diphosphine bioconjugation and radiolabelling with ^{99m}Tc , ^{188}Re or ^{64}Cu .

References

1. R. E. Nuttall *et al.*, *Inorg. Chem.*, 2023, **62**, 20582–20592.
2. I. N. Hungnes *et al.*, *Inorg. Chem.*, 2023, **62**, 20608–20620.
3. T. T. Pham *et al.*, *J. Nucl. Med.*, 2024, **65**, 1087–1094.

Towards a high-relaxivity Mn(II)-based T_1 probe for MRI

O. Tyurina ^a, J. D. E. T. Wilton-Ely ^b, A. Phinikaridou ^a and G. J. Stasiuk ^a

^a School of Biomedical Engineering and Imaging Sciences, St Thomas' Hospital, Westminster Bridge Road, SE1 7EH, London, UK

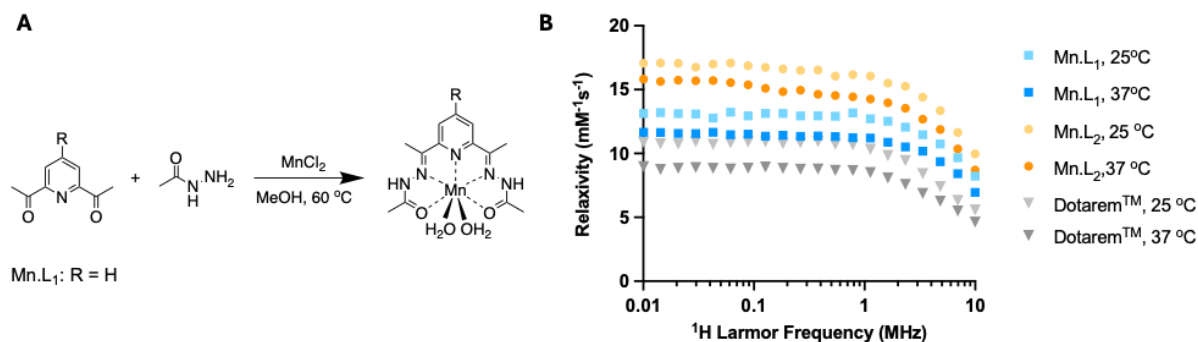
^b Department of Chemistry, Imperial College London, Molecular Sciences Research Hub, White City Campus, W12 0BZ, London, UK

email: olga.tyurina@kcl.ac.uk

Gadolinium-based magnetic resonance imaging (MRI) agents have been widely used in the clinic since the popularisation of the technique in 1980s. Over the past 30+ years there have been many developments in the field, however the clinically used agents are still non-specific and require large quantities to achieve sufficient signal enhancement. Additionally, toxicity associated with release of Gd(III) ions in patients with reduced kidney function prompted further development of transition metal-based contrast agents.¹

Metal ions such as Mn(II) and Fe(III), with 5 unpaired electrons compared to the 7 unpaired electrons of Gd(III), have been investigated as a main alternative to the first generation of MR contrast agents.² to overcome issues with toxicity.

The novel Mn(II) pentadentate complexes were synthesised *via* a template reaction that forms stable MR agents.^{3,4} This route improves relaxivity of the probe by increasing the number of inner sphere waters ($q = 2$), resulting in a relaxivity of $8.2 \text{ mM}^{-1} \text{ s}^{-1}$ compared to $5.6 \text{ mM}^{-1} \text{ s}^{-1}$ of clinical agent DotaremTM at 10 MHz (25°C). The complex was also functionalised in the 4-position of the pyridine ring, further increasing the relaxivity to $8.9 \text{ mM}^{-1} \text{ s}^{-1}$, suggesting that the number of bound water molecules remains unchanged. Further functionalisation of the chelator to achieve active targeting, along with the superior relaxivity of the complex, will provide a promising alternative to current clinical agents.



A – Template reaction for synthesis of Mn.L₁ and Mn.L₂ complexes; B – NMRD profiles of Mn.L₁ and Mn.L₂ compared to the clinical agent DotaremTM at 25 and 37 °C.

References

- 1 M. Le Fur and P. Caravan, *Metallomics*, 2019, **11**, 240–254.
- 2 E. M. Gale, I. P. Atanasova, F. Blasi, I. Ay and P. Caravan, *J Am Chem Soc*, 2015, **137**, 15548–15557.
- 3 S. Anbu, S. H. L. Hoffmann, F. Carniato, L. Kenning, T. W. Price, T. J. Prior, M. Botta, A. F. Martins and G. J. Stasiuk, *Angewandte Chemie - International Edition*, 2021, **60**, 10736–10744.
- 4 S. Anbu, L. Kenning and G. J. Stasiuk, *Chemical Communications*, 2023, **59**, 13623–13626.

Heme in Biology: Insights from Structural Biology

Eloisa Wheatley^a, Abilasha Balakrishnan^a, Jaswir Basran^a, Alistair J. Fielding^b, Emma L. Raven^c, Peter C.E. Moody^a, Hanna Kwon^a

^a Department of Molecular and Cell Biology and the Leicester Institute of Structural & Chemical Biology, University of Leicester, Leicester, LE1 7RH, United Kingdom

^b Centre for Natural Products Discovery, James Parsons Building, Liverpool John Moores University, Byrom Street, Liverpool L3 3AF, United Kingdom

^c School of Chemistry, University of Bristol, Bristol, BS8 1TS, United Kingdom.

email: hk295@le.ac.uk

Heme enzymes catalyse diverse oxidative transformations by activating molecular oxygen, a process central to metabolism, signalling, and stress responses. Structural biology provides a powerful approach to capturing the reactive intermediates that define these catalytic cycles. Using X-ray crystallography and complementary spectroscopic techniques, we have elucidated key structural snapshots of oxygen activation in heme enzymes such as cytochrome c peroxidase (CcP) and ascorbate peroxidase (APX)^{1, 2}. By integrating time-resolved structural and spectroscopic methods, we aim to bridge the gap between static structures and dynamic enzymatic processes, advancing our understanding of heme-dependent oxygen activation across biological systems.

Beyond these systems, we have expanded our investigations to heme binding proteins and their role in heme trafficking and gene regulation, investigating the wider role of heme in biology³⁻⁵. The structural insights will provide a foundation for understanding the broader biological significance of heme binding proteins and their functions in cellular metabolism and regulation

References

1. H. Kwon, et al., *Angew Chem Int Ed*, 2021, **60**, 14578-14585.
2. H. Kwon, et al., *Nat Commun*, 2016, **7**, 13445.
3. M. J. Burton, et al., *J Biol Chem*, 2020, **295**, 13277-13286.
4. S. L. Freeman, et al., *Proc Natl Acad Sci*, 2019, **116**, 19911-19916.
5. X. Yuan, et al., *Proc Natl Acad Sci*, 2016, **113**, E5144-5152.

Investigation of DNA Binding by *Yersinia Enterocolitica* IscR, Regulator of Iron-Sulfur Cluster Biogenesis

Miaomiao Gao¹, Elizabeth Gray¹, Jason. C. Crack¹, Nick E. Le Brun¹

¹ Centre for Molecular and Structural Biochemistry, School of Chemistry, Pharmacy and Pharmacology, University of East Anglia, Norwich Research Park, Norwich, NR4 7TJ, United Kingdom

Iron-sulfur (Fe-S) clusters are protein cofactors essential for life. Their assembly requires complicated intrinsic machineries which are precisely regulated according to the cellular cluster demand. IscR is a [2Fe-2S] cluster-containing protein discovered to play a regulatory role in gene expression of the ISC Fe-S cluster biogenesis pathway in *Escherichia coli* and many other bacteria. The cluster-binding EclscR functions as a repressor of the ISC operon by binding to the promoter region. The apo form has low affinity of the ISC promoter region and so allows transcription to proceed. The loss of clusters is affected by various environmental signals, such as oxidative stress and low iron levels, but the precise nature of how EclscR responds to them directly is not clear. Less is known about other IscR homologs and whether key functional features of the *E. coli* protein are broadly shared. This study focused on the IscR homolog from *Yersinia enterocolitica*, a diarrhoea-causing human pathogen, which is ~80% identical to EclscR, sharing similar structure and cluster-binding characteristics. We found the clusters in both EclscR and YelscR oxidise rapidly from a +1 state to a +2 state when exposed to O₂, which cause a slow loss of the cluster. Surface plasmon resonance (SPR) was used to investigate DNA binding. SPR data indicated that DNA recognition and binding by YelscR is dependent on the presence of its cluster, as observed for EclscR, while the effect of oxidation state of the YelscR cluster is distinct from EclscR, with a clear dependence of affinity on cluster oxidation state.

Insight into the Electronic Structure of [FeFe] Hydrogenase Revealed by Two-Dimensional Infrared Spectroscopy

Cornelius Bernitzky,^a Manon Lachmann,^b Mathesh Vaithyanathan,^a Yvonne Rippers,^a Denise Poire,^{a,c} Solomon Wrathall,^d Barbara Procacci,^d Igor Sazanovich,^e Gregory Greetham,^e Neil Hunt,^c Patricia Rodríguez Maciá,^b Marius Horch,^a James Birrell^f

^a*Freie Universität Berlin, Department of Physics, Arnimallee 14, 14195 Berlin, Germany*

^b*School of Chemistry, University of Leicester, Leicester, LE1 7RH, UK*

^c*Technische Universität Berlin, Straße des 17. Juni 135, 10623 Berlin, Germany*

^d*Department of Chemistry, University of York, York, YO10 5DD, UK*

^e*STFC Central Laser Facility, Research Complex at Harwell, Didcot, OX11 0QX, UK*

^f*School of Life Sciences, University of Essex, Wivenhoe Park, Colchester, CO4 3SQ, UK*

email: james.birrell@essex.ac.uk

[FeFe] hydrogenases are highly active catalysts for the cleavage and evolution of molecular hydrogen. Key aspects of the catalytic cycle and the underlying geometrical and electronic properties of the active-site cofactor, the H-cluster, are not fully understood. Spectroscopic techniques have played a central role in establishing the current state of knowledge on [FeFe] hydrogenases, and further advances in the field depend critically on novel techniques that yield so-far inaccessible insights into structural and mechanistic aspects. Infrared (IR) absorption spectroscopy is a versatile technique for characterizing active and inactive states of the H-cluster by means of structurally sensitive and spectrally isolated CO and CN stretching vibrations. However, the information that can be extracted from these linear experiments is inherently limited. Here, we introduce experimental and computational two-dimensional (2D-)IR spectroscopy for the characterization of [FeFe] hydrogenases. Utilizing the H_{inact} state of the H-cluster as a model system, we demonstrate that this nonlinear technique yields direct insights into the nature and interactions of the CO and CN stretching vibrations. These insights allow a quantitative description of the character of these widely used reporter vibrations, their spatial localization, and the way they change upon structural variation of the H-cluster. The strength of this approach is demonstrated by correctly identifying the proposed structure of the H_{inact} state, in solution at ambient temperature. We have also studied the controversial $H_{\text{red}}H^+$ and $H_{\text{sred}}H^+$ catalytic intermediates, providing conclusive evidence for the presence of a bridging CO ligand in these states. Finally, we used this approach to study conformational dynamics of an oxadithiolate version of the [FeFe] hydrogenase active site.

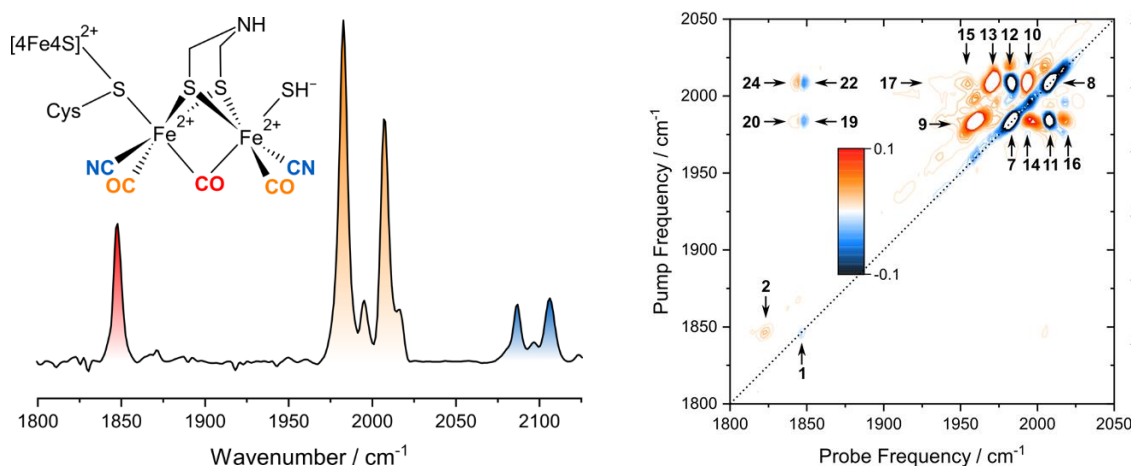


Figure 1. Left) Schematic representation and linear IR absorption spectrum of the H-cluster in the H_{inact} state. Right) 2D-IR spectrum of *DdHydAB* enriched in the H_{inact} state.

Rewiring metalloenzymes for new and improved C-H functionalisation chemistry

Jack S. Rowbotham^a

^aManchester Institute of Biotechnology, Department of Chemistry, University of Manchester, M1 7DN, United Kingdom

email: jack.rowbotham@manchester.ac.uk

Metalloenzymes catalyse a diverse set of reactions in the natural world to sustain and proliferate life. These proteins are highly evolved to perform challenging chemistry in a selective manner under mild conditions, and therefore have considerable potential for wide biotechnological applications. For example, cytochromes P450 have the ability to perform O-atom insertion on inactive C-H bonds directly from O₂, which is highly desirable for clean chemical synthesis. Unfortunately, the native enzymes often fail to deliver sufficient productivity for industrial applications. P450s contain a heme centre which is the heart of the catalytic activity, facilitating C-H hydroxylation *via* a high valent Fe-oxo species. This mechanism, however, relies on the transfer of electrons from redox partner proteins to the heme active site (Figure 1), which is often a limiting step for catalysis. In this talk, I will present results related to two key aspects of P450 catalysis: (i) How electron transfer from the redox partners (fused together as a single reductase domain) can be optimised to improve turnover, and (ii) How metals other than Fe can be incorporated into the heme domain to facilitate alternative reaction pathways, potentially offering routes to desirable transformations such as C-H fluorination.

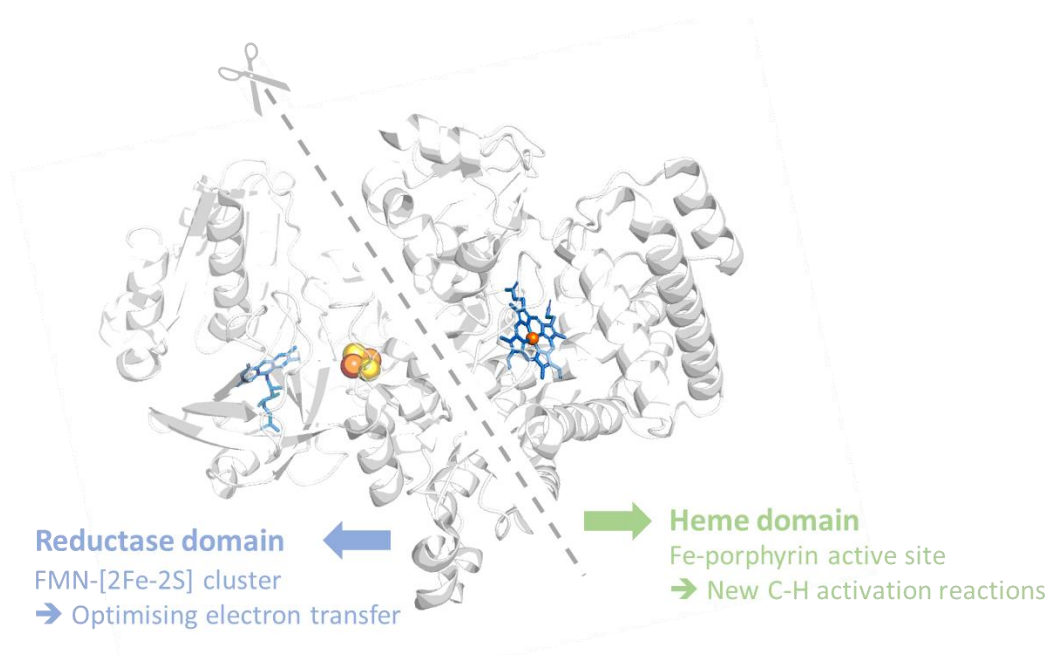


Figure 1: Crystal structure of P450 from *Tepidiphilus thermophiles*.¹ The two domains form part of a single protein chain, but can be isolated and investigated individually to study, and ultimately optimise, key aspects of P450 catalysis.

References

1. L. Zhang, Z. Xie, Z. Liu et al., *Nat Commun.*, 2020, **11**, 2676

Unlocking new insights in metalloprotein redox chemistry

N. G. Baranska,^a H. O. Lloyd-Laney,^{a,b} H. Ford,^c D. Gavaghan,^b J. L. R. Anderson^c
and A. Parkin^a

^a Chemistry Department, University of York, Heslington, York YO10 5DD, UK; ^b Department of Computer Science, University of Oxford, Parks Road, Oxford OX1 3QD, UK; ^c School of Biochemistry, University of Bristol, University Walk, Bristol, BS8 1TD, UK.

email: natalia.baranska@york.ac.uk

The redox chemistry of metalloproteins is fundamental to life. The electron transport pathways of these metalloproteins underpin essential biological processes, sustaining the biochemistry of living organisms. Beyond their biological roles, metalloproteins are harnessed for their catalytic activity in the production of valuable industrial products.

Protein film voltammetry (PFV) is used to probe the electron transfer parameters of redox proteins.¹ Traditionally, direct current voltammetry (dcV) has been utilised for such investigations. However, low signal-to-background ratio of the Faradaic output, limits the precision of the technique. Square-wave voltammetry (SWV) offers improved accuracy by minimising background current contributions, although multiple experiments are required to achieve this. This poses challenges for proteins with limited stability on electrode surfaces.

We therefore introduce Fourier-transformed alternating current voltammetry (FTacV) as a powerful addition to the electrochemical toolkit for investigating electron transfer rates. We present methodological advancements that enable FTacV to be performed using commercial potentiostats, making the technique widely accessible.² Through a case study on a *de novo* haem redox protein, we demonstrate the ability of FTacV to accurately determine electron transfer parameters within a single experiment, supported by both experimental data and computational simulations. Furthermore, we showcase how a careful selection of input parameters, such as frequency, facilitates the study of reversible electron transfers which reach a Nernstian limit under standard voltammetric conditions.

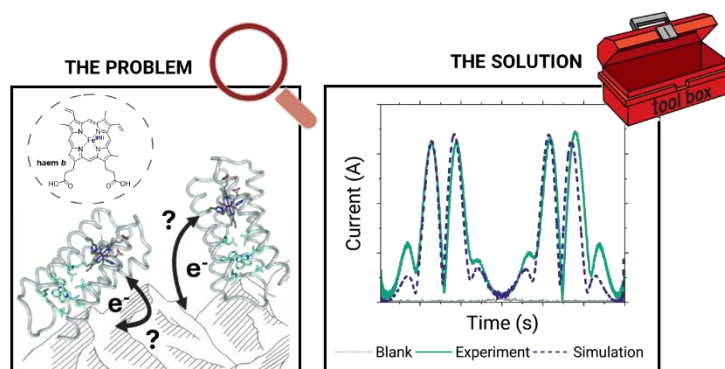


Figure 1 (*left*) Schematic representation of a metalloprotein immobilised on a rough surface of an electrode. (*right*) A current-time output response obtained from a FTacV experiment of a metalloprotein.

References

1. J. N. Butt, L. J. C. Jeuken, H. Zhang, J. A. J. Burton and A. L. Sutton-Cook, *Nat. Rev. Methods Primers*, 2023, **5**, 77.
2. N. G. Baranska, B. Jones, M. R. Dowsett, C. Rhodes, D. M. Elton, J. Zhang, A. M. Bond, D. Gavaghan, H. O. Lloyd-Laney and A. Parkin, *ACS Meas. Sci. Au*, 2024, **4**, 418–431.

***De novo* designed β -hairpin peptides as LPMO mimics**

Enrico Falcone,^{at} Rosemary Tomey,^a Emma Turley,^a David Cannella,^b David Robinson^c and Luisa Ciano^a

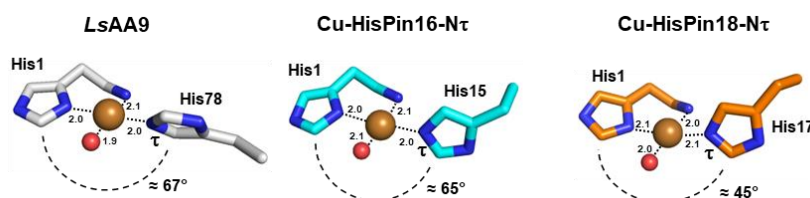
^a School of Chemistry, University of Nottingham, Nottingham, NG7 2RD, UK; ² PhotoBiocatalysis Unit, Biomass Transformation Lab and Crop Production Biostimulation Lab, Université Libre de Brussels, Brussels, Belgium; ³ Department of Chemistry and Forensics, School of Science and Technology, Nottingham Trent University, Nottingham, NG11 8NS, UK; [†] current address: LCC-CNRS, Université de Toulouse, Toulouse, France

email: luisa.ciano@nottingham.ac.uk

The effective use of waste lignocellulosic biomass is crucial in addressing the global need for sustainable energy, being a relevant renewable source of biofuels. However, the recalcitrance of polysaccharides to degradation challenges their effective utilisation as a feedstock. Among carbohydrate-active enzymes, Lytic Polysaccharide MonoOxygenases (LPMOs) stand out as potential biocatalysts for industrial biomass conversion.¹ LPMOs catalyse the oxidative cleavage of β -1,4-glycosidic bonds in polysaccharides, thus boosting the action of other polysaccharide-active enzymes such as glycoside hydrolases.

The active site of LPMOs contains a copper ion coordinated in a 3N T-shaped site by the so-called histidine brace (His-brace) motif, which includes the bidentate N-terminal His, bound via both amine and imidazole groups, and the side-chain of another His.² The development of LPMO mimics could be beneficial to understand the catalytic mechanism, modulate substrate specificity and allow a controlled catalyst immobilisation.

Hence, we designed *de novo* peptides folding into β -hairpins and binding Cu in an LPMO-like His-brace motif. ATR-FTIR and CD spectroscopy showed that the peptides secondary structure is dominated by antiparallel β -strands and turns, commensurate with the formation of β -hairpins. EPR spectroscopy revealed the presence of a single axial Cu^{II} species with parameters comparable to those of LPMOs. Lastly, catalytic assays using p-nitrophenyl- β -D-glucopyranoside as a model substrate demonstrated that the Cu-hairpins can perform LPMO-like activity. Furthermore, the Cu-hairpins are able to perform light-driven oxidation of PASC in the presence of melanin, similarly to some LPMOs, an activity that is unreported for any LPMO mimic so far. This work is the first example of a β -hairpin LPMO-mimic, and these data will be presented highlighting their implication in LPMO-mimic development.



Structure and key metric parameters for Cu-binding sites in LsAA9 (7PYL), Cu-HisPin16 and Cu-HisPin18.

References

1. G. Hemsworth *et al.*, *Trends Biotechnol.*, 2015, **33**, 747-761.
2. L. Ciano *et al.*, *Nature Catal.*, 2018, **1**, 571-577.

Electrostatic Fields Induce Accelerated Proton Coupled Electron Transfer Rates in a Mg-Porphyrin P680^{•+} Mimic

Oscar Reid Kelly, Brendan Twamley and Aidan R. McDonald.^a

^a School of Chemistry, Trinity College Dublin, The University of Dublin, Dublin 2
email: kellyos@tcd.ie

Photosynthetic water oxidation is initiated in Photosystem II (PSII) by the 1-electron photo-oxidation of a chlorophyll-a tetramer named P680. The product of this reaction is P680^{•+}, a π -cation radical complex with an exceptionally high redox potential of 1.1 - 1.3 V vs SHE. This species drives water oxidation by oxidising the oxygen evolving complex via a tyrosine residue.¹ The extreme redox potential of P680^{•+} stands in contrast to the potentials of monomeric chlorophyll-a and other related photosynthetic pigments (0.45 - 0.78 V vs SHE) for reasons that are not yet understood;^{1,2} the electrostatic/dielectric medium of PSII has been calculated to be a significant factor, but experimental support for these calculations is lacking.² To this end, the present work demonstrates that electrostatic fields can be used to tune the redox and reactivity properties of chlorophyll model compounds. We have synthesised and characterised crown ether appended Mg-porphyrins, their adducts with redox-inactive metal cations and their π -cation radical complexes in the presence/absence of bound cations. The electrostatic fields imposed by the proximally-bound cations were found to induce anodic shifts in the redox potential of the porphyrin ligand and to accelerate the rate of PCET from a phenolic substrate — a reaction that mimicks the reaction between P680^{•+} and tyrosine in PSII. These results therefore have implications for our understanding of water oxidation in PSII and photosynthetic electron transport more generally.

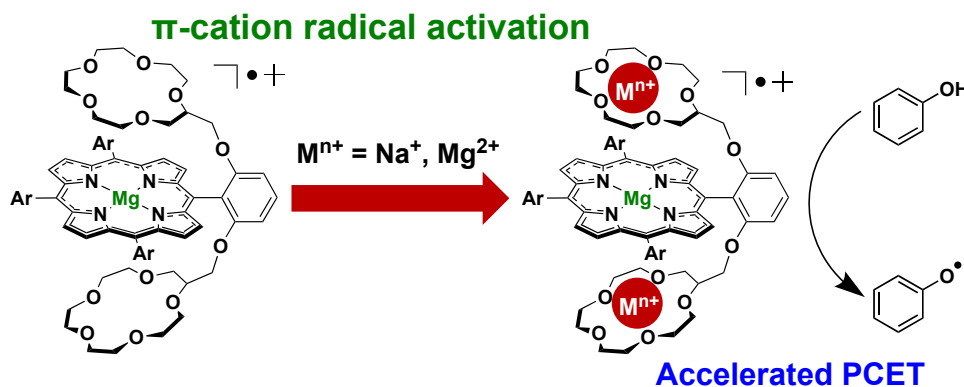


Figure 1. Summary of the present work.

References

1. J. M. Kargul, J. Barber, Chapter 5, Structure and Function of Photosynthetic Reaction Centres, *RSC Energy and Environment Series*, 2012, 107-142.
2. a) H. Ishikita, W. Saenger, J. Biesiadka, B. Loll, E.-W. Knapp, *Proc. Natl. Acad. Sci.* 2006, **103**, 9855- 9860. b) Ishikita, H.; Loll, B.; Biesiadka, J.; Saenger, W.; Knapp, E.-W. *Biochemistry*, 2005, **44** (10), 4118-4124.

Artificial metalloenzymes based on anchoring of piano stool complexes to alcohol dehydrogenase

Henry A. W. Padley,^a Floriane L. Martins,^a Mattias Basle,^a Simone Morra,^a Christof M. Jäger,^a Ingrid Dreveny,^b Anca Pordea^{a,*}

^a Faculty of Engineering, University of Nottingham, Nottingham, UK.

^b Biodiscovery Institute, School of Pharmacy, University of Nottingham, Nottingham, UK

email: anca.pordea@nottingham.ac.uk

Artificial metalloenzymes (ArMs) are assembled by incorporation of a catalytic metal moiety into a protein scaffold. This enables the exertion of control over the chemical environment of the catalyst via genetic manipulation of the scaffold, providing one of the key advantages of ArMs over small-molecule catalysts. When natural enzymes are used as starting points for ArM design, their naturally evolved architectures can provide the advantage of substrate binding and orientation.

Our group developed ArMs based on the attachment of rhodium and iridium piano-stool complexes to alcohol dehydrogenase (ADH). The aim was to exploit the ability of this enzyme to bind both a hydrophobic ketone / alcohol substrate and a nicotinamide cofactor, therefore providing different opportunities to anchor an artificial catalyst. Here, we present and discuss two approaches we used for the creation of these ArMs (Figure 1).

In a first approach, covalent attachment of rhodium complexes to a single cysteine in the substrate binding site delivered artificial formate dehydrogenases for the recycling of nicotinamide cofactors.^{1,2} Changing the location of the cysteine influenced the binding affinity of the cofactors, leading to an improved Michaelis constant. In a second approach, a molecular docking workflow enabled the design and optimisation of iridium catalysts with high affinity for the cofactor binding site within ADH.³ The creation of an active ArM was demonstrated for the transfer hydrogenation of salsolidine. A combination of competition experiments (indicating binding affinity) and catalytic activity was used to evidence that the catalyst is bound to a defined site that can also accommodate the substrate.

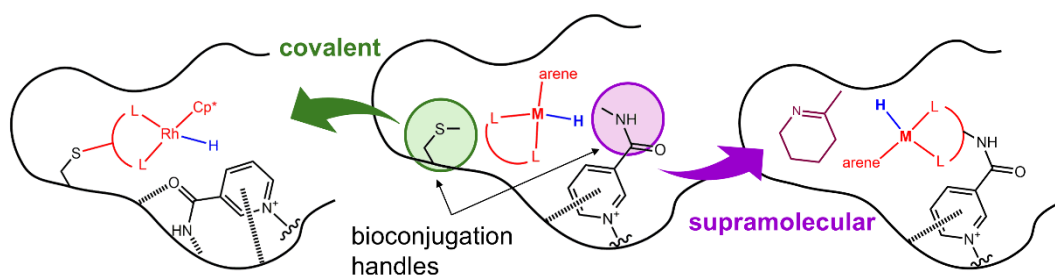


Figure 1. Two approaches to create artificial metalloenzymes from alcohol dehydrogenase.

References

1. S. Morra & A. Pordea, *Chem. Sci.*, 2018, **9**, 7447–7454.
2. M. Basle, H. A. W. Padley, F. L. Martins, G. S. Winkler, C. M. Jäger, A. Pordea, *J. Inorg. Biochem.*, 2021, **220**, 111446.
3. F. L. Martins, A. Pordea, C. M. Jäger, *Faraday Discuss.*, 2022, **234**, 315.

Approaches to Design Artificial Metalloenzymes for Sustainable Chemistry

Evelina Venckutė^a, M. T. Lachman^b, Z. Duan^c, J. A. Birrell^d, S. B. Carr^c Amanda G. Jarvis^a
and Patricia Rodríguez Maciá^{*b}

^a*EaStCHEM School of Chemistry, Joseph Black Building, University of Edinburgh, David Brewster Road, Edinburgh EH9 3FJ, UK*

^b*School of Chemistry and Leicester Institute of Structural and Chemical Biology, University of Leicester, University Road, Leicester, LE1 7RH, UK*

^c*Department of Chemistry, University of Oxford, Inorganic Chemistry laboratory, South Parks Road, Oxford, OX1 3QR, UK*

^d*School of Life Sciences, University of Essex, Wivenhoe Park, Colchester CO4 3SQ, UK*

email: prm28@leicester.ac.uk

Designing efficient catalysts for energy-conversion reactions (e.g., reduction of CO₂ into simple carbon-building blocks, reduction of N₂ to ammonia and production or oxidation of H₂) is essential to solve many major environmental and energy-related problems confronting our planet. In this respect, small-molecule activation (SMA) reactions are crucial for sustainable energy but the chemistry of these reactions is extremely complex and represents huge challenges for industry.¹ Metalloenzymes have evolved in nature for billions of years to be able to activate small molecules such as CO₂, H₂ and N₂ with spectacular efficiency and selectivity only by using earth-abundant metals in their active sites.² However, their large size, tedious and high cost of preparation hinder their widespread use in biotechnology. Semi-synthetic and artificial metalloenzymes (ArMs) combine the best of two worlds: state-of-the-art transition-metal catalysis with the selectivity and efficiency of natural metalloenzymes, offering an attractive means to develop sustainable biohybrid catalysts for energy-relevant reactions. However, the design and engineering of ArMs is complex.³ Therefore, approaches providing mechanistic insight to inform design of enhanced ArMs are needed. For example, direct electrochemistry tools, such as protein film electrochemistry (PFE), offer valuable insight into the redox properties of metalloenzymes. However, such characterisation methods remain scarce in ArM design campaigns. In this talk, I will show how a suite of structural, spectroscopic and electrochemical techniques combined with computational methods has been used to mechanistically characterised newly developed semi-synthetic and artificial metalloenzymes and feed it into the development of design criteria for the generation of tailored ArMs with target applications in electrocatalysis. Overall, this work provides a tool-box of mechanistically-driven approaches for design of enhanced biohybrid catalysts for small-molecule activation and new-to-nature reactions.

References

1. R. S. C. Dalton and I. B. DeeGee, *Abbr. J. Title*, 2025, **4**, 1-3.
2. A. A. Salamatian, K. L. Bren, *FEBS Letters* 2023, **597**, 174-190
3. J. M. Le, K. L. Bren, *ACS Energy Letters* 2019, **4**, 2168-2180
4. C. Van Stappen, Y. Deng, Y. Liu, H. Heidari, J.-X. Wang, Y. Zhou, A. P. Ledray, Y. Lu, *Chem. Rev.* 2022, **122**, 11974-12045

New hybrid hydroxypyridinone-aza-crown-macrocyclic chelators for molecular imaging and radiotherapy

Rory A. J. Kenrick^a, Alex Rigby^a, Rachel E. Nuttall^a, Truc Pham^a, Natasha Patel^a, Charlotte Rivas^a, Saul M. Cooper^b, Oliver W. L. Carter^a, Thomas Hicks^c, Nicholas J. Long^b and Michelle T. Ma^a

^a Division of Biomedical Engineering and Imaging Science, King's College London, St Thomas' Hospital, SE1 7EH ^b Molecular Sciences Research Hub, White City Campus, Imperial College London, W12 0BZ ^c Department of Chemistry, King's College London, 7 Trinity Street, SE1 1DB

email: rory.kenrick@kcl.ac.uk

Macrocyclic chelators incorporating aza-18-crown-6 rings have shown promise for stable encapsulation of f-block metal ions, notably medically useful radionuclides of Tb³⁺, Lu³⁺, Ac³⁺ and Th⁴⁺.^{1, 2} Multidentate ligands incorporating hydroxypyridinone (HOPO) motifs have demonstrated versatility in complexation of many radionuclides, including [⁶⁸Ga]Ga³⁺ and [²²⁷Th]Th⁴⁺.^{3, 4} Given this previous success, with notable affinity of both families toward electronically 'hard' metal ions, three novel hybrid chelators, **2C-DIHOPO**, **3C-DIHOPO** and **3C-TRIHOPO** have been synthesised and characterised by NMR spectroscopy and high-resolution ESI-MS. Each contains an aza-18-crown-6 crown ether macrocycle functionalised with pendent 3,2-HOPO groups for additional coordination of metal ions.

Radionuclides of the lanthanide ion, Tb³⁺, have utility in nuclear medicine in both diagnosis and therapy. The binding behaviour of Tb³⁺ was therefore studied with each ligand. Radiolabelling studies with radiotherapeutic, β⁻-emitting [¹⁶¹Tb]Tb³⁺ (β⁻, t_{1/2} = 6.9 d) indicated that all three ligands bind [¹⁶¹Tb]Tb³⁺. Out of the three new radiolabelled chelators, [¹⁶¹Tb]Tb-**3C-TRIHOPO** was prepared in highest radiochemical yield. The stability of [¹⁶¹Tb]Tb-**3C-TRIHOPO** was assessed by incubating the radiolabelled complex in human serum at 37 °C, indicating the complex was 66% and 23% intact after 1 h and 24 h, respectively. The Tb-labelled complex was also studied *in vivo* by SPECT-CT imaging up to 1 h post-injection, and by obtainment of an *ex-vivo* biodistribution profile. These data are consistent with serum studies, suggesting initial stability of the radiolabelled complex, but with complex dissociation over time. These findings explore the potential of **3C-TRIHOPO** as a novel ligand for ¹⁶¹Tb-based chelation and therapy.

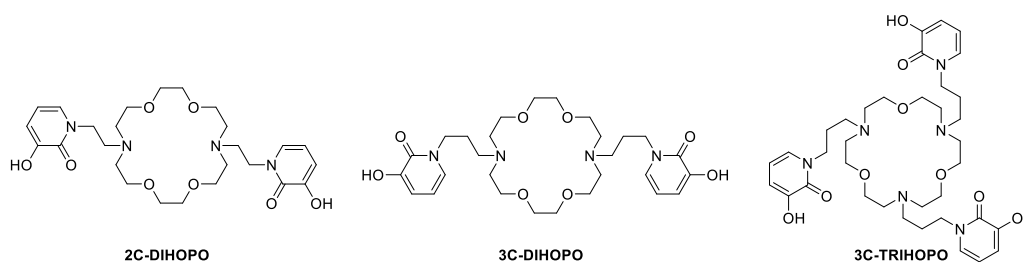


Figure 1. Structures of aza-18-crown-6 chelators discussed in this work.

References

1. A. Hu et al., *Inorg. Chem.*, 2022, **61**, 12847-12855.
2. N. A. Thiele et al., *Angew. Chem., Int. Ed.*, 2017, **56**, 14712-14717.
3. T. Ramdahl et al., *Bioorg. Med. Chem. Lett.*, 2016, **26**, 4318-4321.
4. J. D. Young et al., *J. Nucl. Med.*, 2017, **58**, 1270-1277.

Dithiocarbamates for Molecular Imaging with Technetium-99m Labelled Peptides

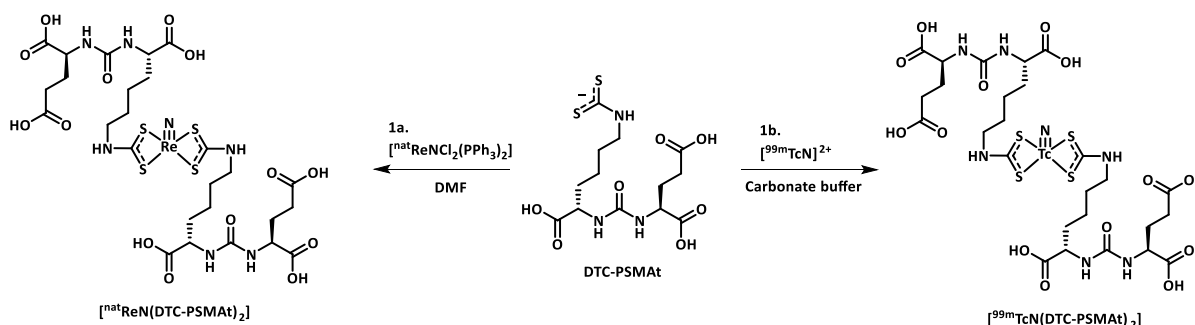
Authors: Jung Sik Shin^a, Rachel E. Nuttall^a, Oliver W. L. Carter^a, Truc T. Pham^a, Rafael T. M. de Rosales^a, Graeme Hogarth^a and Michelle T. Ma^a

^a *King's College London, Division of Imaging Sciences and Biomedical Engineering, St Thomas' Hospital, London SE1 7EH, UK*

email: jung_sik99m.shin@kcl.ac.uk

The radionuclide ^{99m}Tc (t_{1/2} = 6h, 89% abundance, E_γ = 141 keV) is a γ-emitter used in radiotracers for diagnostic SPECT (Single Photon Emission Computed Tomography) and γ-scintigraphy imaging. Existing ^{99m}Tc-labelled radiopharmaceuticals for perfusion imaging are commonly produced using kits.¹ Novel ^{99m}Tc chelators are required to enable efficient incorporation of ^{99m}Tc into a receptor-targeting biomolecule, for molecular SPECT/γ-scintigraphy imaging. Dithiocarbamate (DTC) chelators are attractive candidates for this purpose, as they are highly adaptable and versatile in their binding to a wide range of metals in various oxidation states.^{2,3} In addition, synthesis of DTCs is simple and cost-efficient, which could increase access to molecular imaging of diseases.

Our aim is to develop a molecular imaging agent for imaging the Prostate Specific Membrane Antigen (PSMA) over-expressed in prostate cancer. Here, a DTC-PSMA conjugate was prepared from a primary amine derivative of a PSMA-targeted dipeptide (PSMA^t), and its coordination chemistry was investigated with non-radioactive ^{nat}Re and radioactive ^{99m}Tc (Scheme 1). DTC-PSMA^t was reacted with [^{nat}ReCl₂(PPh₃)₂] to obtain [^{nat}ReN(DTC-PSMA^t)₂] (Scheme 1a), which has been isolated and fully characterised. The isostructural ^{99m}Tc complex [^{99m}TcN(DTC-PSMA^t)₂] was then prepared via two-steps: [^{99m}TcO₄]⁻ was reduced to a [^{99m}TcN]²⁺ intermediate, followed by the addition of DTC-PSMA^t in carbonate buffer, to yield [^{99m}TcN(DTC-PSMA^t)₂] (Scheme 1b) in consistently high radiochemical yield (>95%). The radiotracers showed high metabolic stability in human serum (>90% after 24 hours). Preliminary *in vitro* studies showed that [^{99m}TcN(DTC-PSMA^t)₂] exhibit specific uptake in PSMA-expressing prostate cancer cells.



Scheme 1. Preparation of a) [^{nat}ReN(DTC-PSMA^t)₂] and b) [^{99m}TcN(DTC-PSMA^t)₂].

References

1. PY. Marie et al., *Journal of Nuclear Medicine*, 1995, **36**(6), 936-943.
2. G. Hogarth, *Progress in Inorganic Chemistry*, 2005, 71–561.
3. J. K. Bordoloi, D. Berry, I. U. Khan, K. Sunassee, R. T. de Rosales, C. Shanahan and P. J. Blower, *Dalton Transactions*, 2015, **44**, 4963–4975.

The hunt for neutral pH radiolabeling with gallium-68, a study in acyclic chelators

Thomas W. Price¹, Laurène Wagner,¹ Veronika Rosecker,¹ Ivan Hawala,¹ Graeme J. Stasiuk¹

1. Department of Imaging Chemistry and Biology, School of Biomedical Engineering and Imaging Sciences, King's College London, London, SE1 7EH, UK

email: graeme.stasiuk@kcl.ac.uk

Positron emission tomography (PET) is a powerful diagnostic tool in nuclear medicine. Typically, a positron emitting radiometal is labelled with a ligand in the (sub)micromolar range within minutes in high chemical and radiochemical yield. Gallium 68 ($t_{1/2} = 68$ min) is the most widely used radiometal for PET due to its good availability and short half life. Macrocyclic ligands are currently used in clinic but have the disadvantage of slow reaction kinetics. Therefore, there is an interest in developing acyclic ligands with more favourable reaction kinetics. We present a series of acyclic ligands for gallium-68, dpaa,¹ tpa², bispidine³ and Bn₂DT3A.⁴ Dpaa and tpa radiolabel efficiently with gallium-68, at pH 7.4 but do not form stable complexes. Bispidine shows high stability but only radiolabelling at pH 4. Bn₂DT3A gives good radiochemical yields, but different products depending on the pH. At pH 4 [⁶⁸Ga][Ga(Bn₂DT3A)] is formed and is unstable. At pH 7.4 [⁶⁸Ga][Ga(Bn₂DT3A)(OH)]⁻ is formed and is stable in serum. *In vivo* studies show rapid renal clearance and negligible unspecific uptake. Bn₂DT3A has been modified to give 1-naphthyl₂DT3A and 2-naphthyl₂DT3A, incorporating naphthyl groups to evaluate the increase in steric hindrance on radiolabelling and stability, giving rise to stable ⁶⁸Ga complexes at labelled at both pH 4 and 7 with different clearance pathways *in vivo*. Bn₂DT3A has been modified with PSMA, [⁶⁸Ga][Ga(Bn₂DT3A-PSMA)] and shows targeted imaging of prostate cancer.

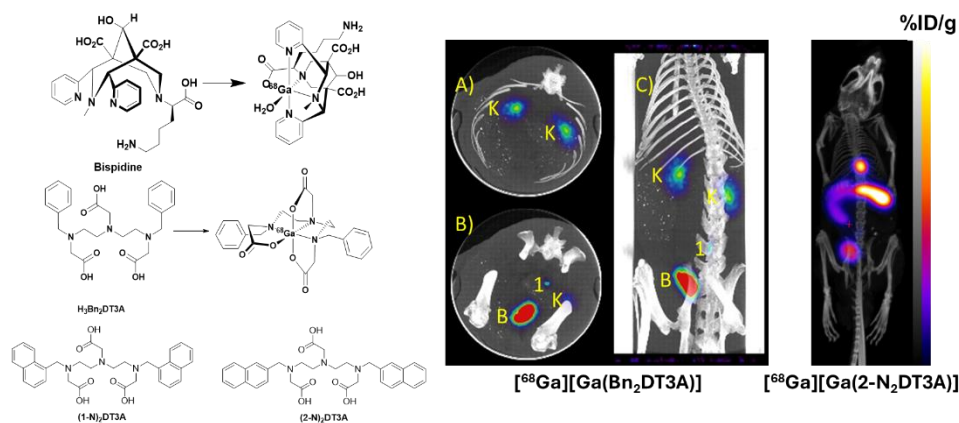


Figure 1. Ga.bispidine, Ga.Bn₂DT3A, 2N₂DT3A, 1N₂DT3A and PET image of [⁶⁸Ga][Ga(Bn₂DT3A)(OH)]⁻ [⁶⁸Ga][Ga(2-Naphthyl₂DT3A)(OH)]⁻

References

1. T. W. Price, J. Gallo, V. Kubíček, Z. Böhmová, T. J. Prior, J. Greenman, P. Hermann, and G. J. Stasiuk*, Dalton Trans., 2017,46, 16973-16982
2. T. W. Price, L. Wagner, V. Rosecker, J. Havlíčková, T. J. Prior, V. Kubíček, P. Hermann, G. J. Stasiuk, Inorganic Chemistry, 2023, 62, 50, 20769–20776T.
3. W. Price, S. Y. Yap, R. Gillet, H. Savoie, L. J. Charbonnière, R. W. Boyle, A.M. Nonat, and G. J. Stasiuk, Chem. Eur. J., 2020, 26, 7602 – 7608.
4. T. W. Price, I. Renard, T. J. Prior, V. Kubicek, D. Benoit, S. J. Archibald, A.-M. Seymour, P. Hermann, G. J. Stasiuk*, Inorganic Chemistry, 2022, 61, 43, 17059-17067.

Delineating the interactions of cytotoxic pyrrole-based metal chelates with human serum albumin

Sheldon Sookai^a, and Orde Q. Munro^{a,b}

^aSchool of Chemistry, University of the Witwatersrand, 1 Jan Smuts Avenue, Johannesburg, PO WITS 2050, South Africa

^bSchool of Chemistry, University of Leeds, Woodhouse Lane, LS2 9JT, Leeds, UK

email: O.Munro@leeds.ac.uk

Human serum albumin (HSA) efficiently transports drugs *in vivo*: most are organic, and their binding site(s) are known from X-ray crystallography. To better understand how metallodrug candidates are bound and transported by the protein, we have studied how a series of related d⁸ metal chelates of bis(pyrrolide-imine) ligands interact with HSA. The binding affinity and site specificity are shown to depend on (i) the identity of the d⁸ metal ion in Ni^{II}, Pd^{II} and Pt^{II} chelates of the ligand,¹ (ii) the substitution pattern and polarity of the chelating ligands in several Pt^{II} analogues,² and (iii) the chirality of the chelating ligand in a pair of enantiopure Au^{III} derivatives.³ Fluorescence quenching data for native and probe-bound HSA showed binding sites close to Trp-214 (subdomain IIA) are targeted. The Stern-Volmer constants, K_{SV} , typically range from 10⁴ M⁻¹ to 10⁵ M⁻¹ while the affinity constants, K_a , range from $\sim 3.5 \times 10^3$ M⁻¹ to $\sim 1 \times 10^6$ M⁻¹ at 37 °C, following the order Pd(PrPyrr) > Pt(PrPyrr) > Ni(PrPyrr) > H₂PrPyrr when the ligand is held constant. Ligand uptake is usually enthalpically driven, though some exceptions occur. Induced CD spectra for the protein-bound metal chelates can be simulated by hybrid QM:MM TD-DFT methods, proving that the metal complexes neither decompose nor demetallate after uptake by HSA.^{1–4}

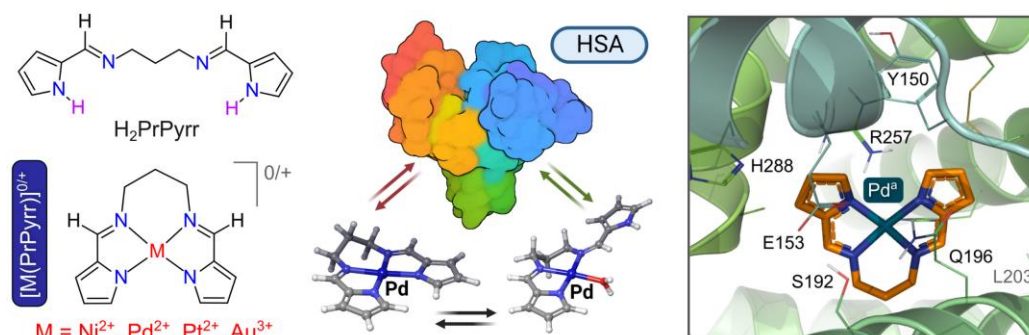


Fig. 1. Bis(pyrrolide-imine) metal complexes bind as intact metal chelates to human serum albumin.

The broader picture emerging from this work is that transport and delivery of the metal chelates, including Au(III) derivatives that target human topoisomerase II,⁴ by HSA *in vivo* could be feasible for suitably redox-stabilized cytotoxic or bactericidal metal chelates within this class of compounds.

References

1. S. Sookai and O. Q. Munro, ChemistryEurope, 2023, 1, e202300012.
2. S. Sookai and O. Q. Munro, Dalton Trans., 2023, 52, 14774–14789.
3. S. Sookai, M. P. Akerman and O. Q. Munro, Dalton Trans., 2024, 53, 5089–5104.
4. S. Sookai, M. Akerman, M. Færch, Y. Sayed and O. Q. Munro, Eur. J. Med. Chem., 2025, 287, 117330.

Mass spectrometric imaging and quantitative analysis of the *in vivo* biodistribution of trastuzumab using a rhodium(III) sarcophagine complex functionalized with dibromopyridazinedione

Natasha Patel^a, Truc T. Pham^a, Alexander Griffiths^a, Brett M. Paterson^b, George Firth^a, Alexander Morrell^a, Cliona McMahon^c, Nicholas J. Long^d, James R. Baker^d, Vijay Chudasama^d, Michelle T. Ma^a

^aKing's College London, United Kingdom; ^bThe University of Queensland, Australia; ^cUniversity College London, United Kingdom; ^dImperial College London, United Kingdom

email: michelle.ma@kcl.ac.uk

Mass cytometry with antibodies labelled with stable metal isotopes enables both sensitive imaging and the quantification of protein expression in biological samples. Typically, these specimens are exposed to a panel of labelled antibodies *ex vivo*, after sample collection. Here, we aimed to study the *in vivo* biodistribution of a rhodium-labelled immunoconjugate of a HER2-targeted therapeutic IgG1 antibody, trastuzumab, using mass cytometry techniques.

A Rh³⁺ complex of a macrobicyclic “sarcophagine” chelator was appended with a dibromopyridazinedione (DBPD), to produce a novel disulfide bond labelling molecule, “Rh-sar-DBPD”. Rh-sar-DBPD was site-specifically conjugated to trastuzumab *via* its four native solvent-accessible disulfide bonds to yield a highly homogenous immunoconjugate, Rh-sar-PD-trastuzumab. In biological studies, inductively coupled plasma mass spectrometry (ICP-MS) and laser ablation (LA) ICP-MS were applied to measure ¹⁰³Rh content, as a proxy for Rh-sar-PD-trastuzumab accumulation. Female NSG mice bearing orthotopic HCC1954 breast cancer tumors were administered Rh-sar-PD-trastuzumab. Quantitative ICP-MS of ¹⁰³Rh signal in dissected tissues indicated receptor-specific HER2-mediated uptake in tumors. LA ICP-MS imaging analysis of tumor and ovary tissue sections showed heterogeneous uptake in HER2-expressing HCC1954 tumor cells and ovarian granulosa cells.

To the best of our knowledge, this is the first report in which both ICP-MS and LA-ICP-MS have been used on tissue exposed to a metal-tagged antibody *in vivo*, enabling quantification of the biodistribution of the novel immunoconjugate, Rh-sar-PD-trastuzumab, in a murine model of breast cancer.

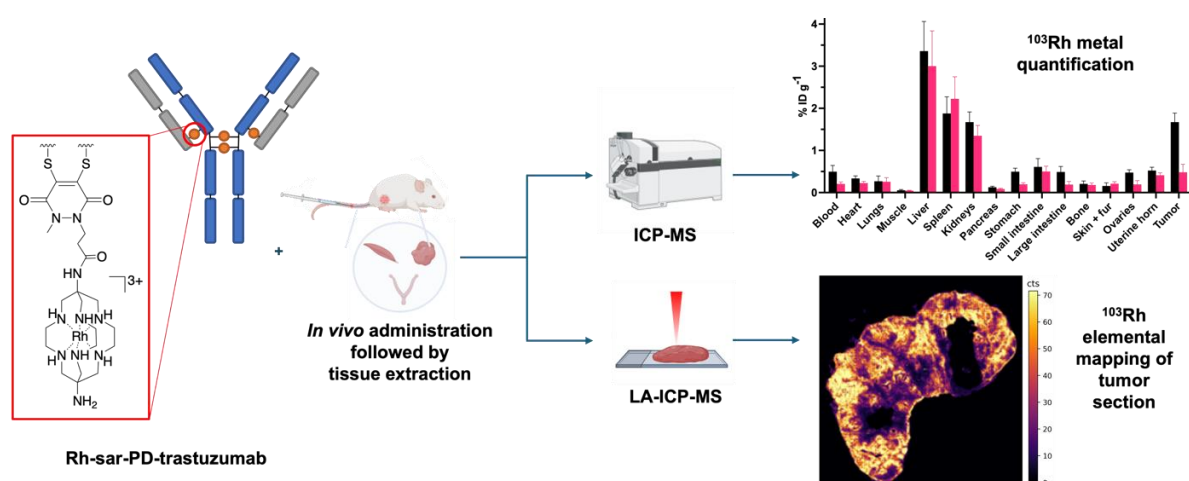


Figure: A Rh³⁺ complex of a macrobicyclic sarcophagine chelator enables mass spectrometric imaging and analysis of the biodistribution of a cancer-targeted antibody in a mouse model of breast cancer.

Design and development of gold(III)-glycoconjugates as antiviral agents against SARS-CoV-2

F. Brescia,^a J. Ott,^b I. Ott,^b L. Ronconi^{*a}

^a University of Galway, School of Biological and Chemical Sciences, Galway (Ireland)

^b Technische Universität Braunschweig, Institute of Medicinal and Pharmaceutical Chemistry, Braunschweig (Germany)

email: luca.ronconi@universityofgalway.ie

First identified in Wuhan (China) in December 2019, the COVID-19 pandemic caused a worldwide health and socio-economic emergency, exposing the vulnerability of our globalized society. COVID-19 is the third major coronavirus outbreak following SARS-CoV (2002) and MERS-CoV (2012), which worryingly highlights the urgent need for large-spectrum antiviral treatments,¹ whose approval rate is dramatically low.² Despite extensive drug design and repurposing efforts, only a few drugs are currently approved for COVID-19 patients, mostly recommended to treat patients at high risk.³

When it comes to antiviral drug development, virus proteases are well-established targets since selective inhibitors of viral proteases can interrupt virus lifecycle without affecting host cells.⁴ In this context, metal-based compounds are under-represented in drug discovery, despite growing evidence of their therapeutic potential. Two lead gold(III)-dithiocarbamate glucoconjugates developed by the Ronconi group have shown exceptionally high and selective activity against SARS-CoV-2 papain-like protease (PL^{pro}), with IC₅₀ values of 1.04–1.15 µM, as well as the capability to block completely viral replication in infected Caco-2 cells at the non-toxic concentration of 500 µM.⁵

Building upon such findings, we have designed a library of gold(III) glycoconjugates (**Figure 1**) all sharing the {Au^{III}–dithiocarbamate} scaffold, which is believed to interact with the zinc-finger domain of PL^{pro}. This design aims to retain PL^{pro} inhibition and achieve viral suppression at lower, more effective concentrations through structural variations of the monosaccharide scaffold and linker, thus potentially broadening the therapeutic applicability of this approach.

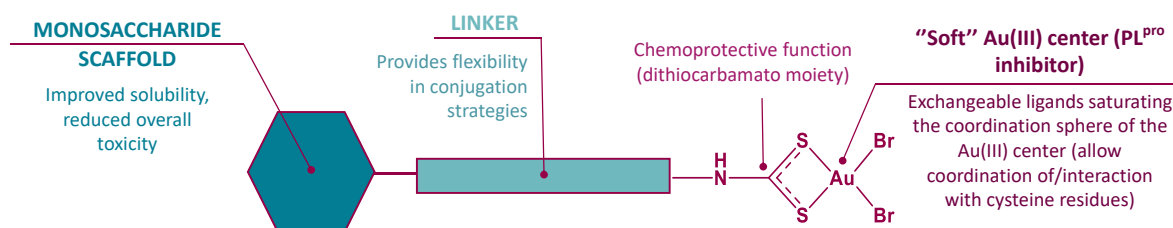


Figure 1. General design of the gold(III)-dithiocarbamate glycoconjugates here reported.

References

1. A.K. Ghosh, M. Brindisi, D. Shahabi *et al.*, *ChemMedChem*, 2020, **15**, 907-932.
2. A. Batta, B.S. Kalra and R. Khirasaria, *J. Family Med. Prim. Care*, 2020, **9**, 105-114.
3. European Medicines Agency - COVID-19 Medicines (<https://www.ema.europa.eu/en/human-regulatory/overview/public-health-threats/coronavirus-disease-covid-19/treatments-vaccines/covid-19-treatments>), last accessed February 2025).
4. S. Gevorgyan, H. Khachatryan, A. Shavina *et al.*, *Viol. J.*, 2024, **21**, 330.
5. M. Gil-Moles, S. Türc, U. Basu *et al.*, *Chem. Eur. J.*, 2021, **27**, 17928-17940.