Shining light on metalloenzyme catalysis: photochemistry coupled with electrochemistry as a mechanistic tool

Highlights

• Development of a combined photochemical-electrochemical methodology for time-resolved studies of metalloenzyme mechanism
• Identification of unknown, short-lived catalytic intermediates
• Determine influence of fundamental coordination chemistry on reactivity of metalloenzymes

Overview

The activation and redox chemistry of small molecules such as CO, CO₂, H₂, formate, O₂, NO, N₂, and NH₃ by metalloenzymes play important roles in vital processes such as the global carbon and nitrogen cycles, and as sources of energy or low-potential reducing equivalents in cellular environments (for example certain bacteria are capable of using CO₂ as their sole C source, and H₂ as their sole energy source). Despite the current urgent need for sustainable fuels and ‘green’ routes to chemical synthesis, detailed mechanistic understanding of how small molecule activation reactions are carried out in nature is often lacking. In part this is due to the high turnover frequencies achieved by the metalloenzymes which carry out these reactions, such as carbon monoxide dehydrogenase (CO₂/CO, >2000 s⁻¹), formate dehydrogenase (CO₂/HCOO⁻, >112 s⁻¹), and hydrogenase (H⁺/H₂, >9000 s⁻¹). New experimental tools are therefore required, capable of probing reactivity on fast, sub-turnover frequency timescales (ca. ps – ms).

Electrochemistry provides convenient control over the oxidation state of metalloenzymes and makes it possible to regulate enzymatic turnover. However, electrochemistry is most commonly applied to studies of steady-state kinetics, and is most suited to characterisation of long-lived catalytic intermediates. In contrast, photochemical triggers are widely used for sub-turnover mechanistic studies, offering valuable insight if catalysis can be initiated by light without triggering unwanted photochemical side reactions.

During this project we will develop a combined ‘photobioelectrochemical’ methodology coupled with a range of spectroscopic techniques in order to facilitate sub-turnover frequency studies of redox metalloenzyme mechanisms. Using this new approach we will identify and characterise short-lived catalytic intermediates and probe the
fundamental coordination chemistry that equips metalloenzymes, Nature’s ‘machines’, with such remarkable efficiency.

Methodology

Work on this project will be carried out both in Leicester and using existing collaborations of Ash at national facilities: Diamond Light Source for infrared microspectroscopy and X-ray spectroscopy, and the Central Laser Facility for ultrafast pump-probe spectroscopy.

This project will develop ‘caged’ electron sources with stable and reproducible photochemical properties, suitable for photoinitiation of both reductive and oxidative redox processes on subturnover frequency timescales. These methods will then be used to study mechanistic details of a range of redox enzymes. Activity of the photosensitized enzyme systems will be confirmed using standard spectroscopic and analytical techniques such as HPLC, GC, UV-visible and IR spectroscopy.

Further Reading


Funding

This research project is fully funded.

Home/EU Applicants

This project is eligible for a fully funded EPSRC studentship which includes:

- A full UK/EU fee waiver for 3.5 years
- An annual tax free stipend of £15,285 (2020/21)
- Research Training Support Grant (RTSG)

If you have any queries about the studentship packages please email csepgr@le.ac.uk

Application Instructions

The online application and supporting documents are due by Thursday 27th February 2020.
Applicants are advised to apply well in advance of the deadline, so that we can let you know if anything is missing from your application.

Required Materials

1. Online application form
2. Two academic references
3. Transcripts
4. Degree certificate/s (if awarded)
5. Curriculum Vitae
6. EPSRC Studentship Form
7. English language qualification *(If English is not your first language)*

All applications must be submitted online, along with the supporting documents as per the instructions on the website.

Please ensure that all email addresses, for yourself and your referees, are correct on the application form.

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<th>Online application form</th>
<th>Deadline : 27th February 2020</th>
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<td>Please refer to the application advice and link to the Apply Button on the <a href="#">EPSRC Studentships webpage</a>.</td>
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<td>Research Proposal/Proposal Statement</td>
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<td>If you are applying for more than one project, please email your additional EPSRC Studentship form to <a href="mailto:Csepgr@leicester.ac.uk">Csepgr@leicester.ac.uk</a></td>
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**Application Timeline**

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<tr>
<td>27th February 2020</td>
<td>Deadline for online application and supporting documents</td>
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<tr>
<td>13th March 2020</td>
<td>Interview invitations to be sent out</td>
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<td>25th – 27th March 2020</td>
<td>Interview days</td>
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<td>31st March 2020</td>
<td>Informal offers to be made</td>
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<td>15th April 2020</td>
<td>Deadline for acceptance of informal offers</td>
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**Application Tips and Advice/Application Status**

**Applying Early**
Although the deadline is 27th February, we advise you to apply early so we can advise you of any delay with your application such as missing documents etc.

**Online Application Advice**
- Check all the email addresses you have entered are correct before submitting.
• Check you have uploaded the CSE Studentship Form in place of your personal statement.

Once You Apply
Once you have submitted your online application form, and uploaded your supporting documents, a copy will be sent to the school for review.