Metalloenzyme (electro)catalysis for hydrogen and ammonia production

R. D. Milton[1,2]

[1]Department of Inorganic and Analytical Chemistry, University of Geneva, Sciences II, Quai Ernest-Ansermet 30, 1205 Geneva, Switzerland, [2]National Centre of Competence in Research (NCCR) Catalysis, University of Geneva

Molecular hydrogen (H_2) and ammonia (NH_3) are key global chemical commodities, largely produced in centralized industrial processes that are heavily reliant on fossil fuels. One example is the Haber-Bosch process, whose high overall efficiency for dinitrogen (N_2) reduction to NH_3 requires (i) high temperatures and pressures, as well as (ii) H_2 that is primarily obtained by the steam-reformation of natural gas. The decentralization of these processes has the potential to improve environmental sustainability globally.

Nature produces metal-containing enzymes that catalyze H_2 formation (hydrogenases) and N_2 reduction (nitrogenases) under comparatively mild conditions and with high selectivity. Understanding precisely how these metalloenzymes catalyze substrate reduction is central to the deployment of these enzymes in new biotechnologies, as well as to the bio-inspired design of new synthetic catalysts. One such prospective biotechnological application is in enzymatic electrocatalysis, where electrodes provide the electrons that these metalloenzymes require for selective substrate reduction.

Recent research in the Milton group has reported the immobilization of [FeFe]-hydrogenase on nanostructured electrode surfaces for H_2 production for relatively prolonged periods of time in near-neutral pH solutions [1]. [FeFe]-hydrogenase spontaneously adsorbs to indium:tin oxide nanoparticles with no additional engineering required to coerce the enzyme into heterogeneous electron transfer. After 120 h of continuous potentiostatic operation, this enzyme electron was observed to retain ~92% of its electrocatalytic H_2 formation activity. We have also taken steps towards employing nitrogenases in NH_3-producing systems. Using a mutant nitrogenase MoFe protein that carries a single solvent-exposed cysteine residue, a bifunctional steric-inhibitor/affinity purification peptide was introduced to prevent electron delivery to one half of the heterotetrameric MoFe protein and enable purification of the inhibited conjugates. Our research confirms that activity in both catalytic halves of the MoFe protein is not strictly required for N_2 fixation, paving the way towards enzymatic electrocatalysis and/or enzyme minimization by engineering [2].

[1] Yongpeng Liu, Sophie Webb, Pavel Moreno-García, Amogh Kulkarni, Plinio Maroni, Peter Broekmann, Ross D Milton, *JACS Au*, **2023**, 3 (1), 124-130.
[2] Cécile Cadoux, Daniel Ratcliff, Nevena Maslać, Wenyu Gu, Ioannis Tsakoumagkos, Sascha Hoogendoorn, Tristan Wagner, Ross D Milton, *JACS Au*, **2023**, 3 (5), 1521-1533.