ELECTROCATALYSIS STUDIES RELATED TO [FeFe]-HYDROGENASE

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FACULTY OF SCIENCE
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Abstract

This research is concerned with building structures related to the active site of [FeFe] hydrogenase, the H-cluster (a) within surface confined electropolymers (b) in proteins by chemical modification of ferredoxin centres and (c) as catalytic moieties on light harvesting nanoparticles.

The study begins by laying out the synthesis of materials used in the studies detailed in this thesis. Components including the cubane cluster, the {2Fe-2S} sub-site centre and the synthesis of functionalised ligands for modification of electrode surfaces and proteins, as well as synthesis of key monomers which can be electropolymerised for electrode surface modification. An improved synthetic route to a key diiron sub-site target molecule is described.

Some first steps towards constructing structure related to [FeFe]-hydrogenases on electrode surface are reported. The incorporation of a sub-site unit into an ionic polymer scaffold clearly contributes to the stability of the protonated sub-site, where the presence of the sub-site hydride form is confirmed through infrared spectroscopy. Construction of the active site H-cluster on a modified electrode is described. Electrochemical and spectroscopic characterisations show that a redox active cubane assembly, covalently bound within a cysteinyl-alkylammonium functionalised polypyrrole, can be modified with a diiron dithiolate carbonyl unit to give an artificial hydrogenase H-cluster framework, confined within the polymer matrix.

An attempt to construct an H-cluster based on a mutant ferredoxin protein from hyperthermophile *Pyroccocus furiosus* led to the conclusion that mutant system containing is very robust. It is not easily to unfold it with guanidine hydrochloride, but at low pH, it seems possible that chemistry at one of the iron can be carried out. This is potential very interesting.

Preliminary work on constructing a photoelectrochemical platform for hydrogen production is described. This based on the modification of indium phosphide nanocrystals and the incorporation of an $Fe_2(\mu\text{-}S_2)(CO)_6$, a procatalyst with a framework nominally related to the catalytic diiron sub-site of hydrogenase. Immobilisation of the components on a gold substrate provides a photocathode which can produce dihydrogen.

Finally, a summary of the key findings of this work and an outline of possibilities for future work are provided.

Abstrak

Kajian ini tertumpu kepada pemahaman tapak aktif [FeFe]-hidrogenesis iaitu H-kluster yang menjalankan penurunan berbalik proton kepada hidrogen. H-kluster mengandungi {4Fe-4S} kubika yang disambung kepada dwiferum yang turut terikat kepada kumpulan dwisufurlat, CO dan CN^- melalui ikatan sistina. Interpretasi kimia ini telah membawa kepada ciptaan mangkin elektro yang membolehkan penjanaan hidrogen evolusi yang penting kepada tenaga transduksi.

Kajian ini bermula dengan penjelasan ke atas sintesis bahan yang berkaitan dengan H-kluster seperti {4Fe-4S} kluster kubika, sub-tapak {2Fe-2S} dan sintesis ligan berfungsi untuk modifikasi permukaan elektrod dan protein, serta monomer yang boleh dielektropolimerasasi dalam konteks modifikasi permukaan elektrod. Pengubahsuaian keatas sintesis tri-podal yang bertanggungjawab untuk pembentukkan molekul sasaran, dwiferum sub-tapak telah berjaya ditingkatkan hasil bahan sebanyak 41%.

Beberapa langkah pertama dalam pembinaan struktur berhubung dengan [FeFe]-hidrogenesis diatas tapak elektrod telah dibincangkan. Pengkoperasian sub-tapak kedalam polimer ionik telah menyumbang kepada stabiliti protonasi sub-tapak dimana kewujudan hydrit sub-tapak telah dikenalpasti menerusi spektroskopi infra merah. Pembinaan tapak aktif, H-kluster diatas permukaan elektrod juga telah dibincang. Pencirian elektrokimia dan spektroskopi menunjukan kejayaan pemasangan kubika redok aktif $\{Fe_4S_4\}^{2+}$ yang terikat secara kovalen dengan *polypyrrole* berfungsi sistina-

alkylammonium yang boleh diubahsuai dengan dwiferum dwisulfulat. Ini telah menghasilkan rekaan H-kluster yang terbina dalam rangkaian matrik polimer.

Percubaan untuk pembinaan H-kluster berdasarkan *ferredoxin* yang diambil dari *Pyroccocus furiosus* mendapati bahawa sistem mengandungi {4Fe-4S} adalah sangat tegap. Ini menyebabkan ia tidak senang diuraikan dengan *guanidine hydrochloride*. Akan tetapi pada pH rendah, ada kemungkinan tindak balas kimia keatas salah satu atom ferum boleh dilaksanakan.

Kajian awal keatas binaan platform fotoelektrokimia untuk penghasilan hidrogen yang berasaskan modifikasi *indium phosphide* dan pengkoperasian promangkin, $Fe_2(\mu\text{-}S_2)(CO)_6$ yang berkait dengan mangkin dwiferum hidrogenesis telah berjaya menghasilkan gas hidrogen yang dianggarkan sebanyak 60%.

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Lastly, love and hugs to my family and those unnamed “soldiers” for keeping my motivation up until the finish line of my Ph.D. study.

Nothing shocks me. I'm a scientist.

Harrison Ford, as *Indiana Jones*

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Abbreviations

AIREs	abnormal infrared effects
Asp	aspartic acid
atm	atmospheric pressure
b, br	broad
C	coulomb
CB	conduction band
CD	circular dichroism
cm	centimetre
CNMe	methyl cyanide
Cp1	*Clostridium pasteurianum*
CV	cyclic voltammetry
Cys	cysteine
d	doublet
D	deuterium
Da	*Desulfuricans africanus*
DCM	dichloromethane
Ddh	*Desulfovibrio desulfuricans*
deg	degree
DFT	density functional theory
DMF	dimethylformamide
Dppe	1,2-bis(diphenylphosphino)ethane
DTT	DL-dithiothreitol
δ	chemical shift
e	electron
ENDOR	electron nuclear double resonance
EPR	Electron paramagnetic resonance
eq.	equilibrium, equivalent
ES-MS	Electron Spray Mass Spectroscopy
Et	ethyl, CH_2CH_3
Et_2O	diethyl ether
EtOAc	ethylacetate
EtS	thiol-ethyl
E_p	peak potential of a redox process
E_p^{ox}	potential of the oxidation peak
E_p^{red}	potential of the reduction peak
ΔE_p	$E_p^{ox} - E_p^{red}$
$E_{1/2}$	$(E_p^{ox} + E_p^{red})/2$
E^o	Standard electrode potential
Fd	ferredoxin
Fd_{ox}	oxidised ferredoxin
Fd_{red}	reduced ferredoxin
FRET	Förster Resonance Energy Transfer

FTIR	Fourier transform infrared spectroscopy
FWHM	full width at half maximum
GdnHCl	guanidine hydrochloride
h	hour(s)
H	proton
HDA	hexadecylamine
HEPES	4-(2-Hydroxyethyl)piperazine-1-ethanesulfonic acid
HiPIP	high potential iron protein
HOMO	highest occupied molecular orbital
Hmd	H_2-forming methylenetetrahydromethanopterin dehydrogenase
HP	high potential
HP_{ox}	oxidised high potential
HP_{red}	reduced high potential
HYSCORE	hyperfine sublevel correlation spectroscopy
Hz	hertz
I_p^{ox}	maximum currents in oxidation
I_p^{red}	maximum currents in reduction
J	coupling constant
k	kilo
kDa	kilo Dalton
L	litre
LED	light emitting devices
LMCT	ligand-to-metal charge-transfer transition
$LiAlH_4$	lithium aluminium hydride
LUMO	lowest unoccupied molecular orbital
λ	wavelength
m	multiplet, milli
M	mega, molar
max	maximum
MeCN	acetonitrile
Mes	2-[N-Morpholino]ethanesulfonic acid
min	minute, minimum
m/z	mass per charge
μ	micro
n	nano
NHC	N-heterocyclic carbene
NMR	Nuclear magnetic resonance
ODE	methyl myristate
ORTEP	oak ridge thermal ellipsoid plot program
Ω	ohm
υ	frequency
pdt	propanedithiolate
Pf Fd	*Pyrococcus furiosus* ferredoxin
PFV	protein film voltammetry
PGE	pyrolytic graphite edge

Ph	Phenyl
PMe_3	Trimethylphosphine
$P(OMe_3)$	Trimethylphosphane
ppm	part per million
Pr	Propyl
Pt	Platinum
q	quartet, quaternary
RT	room temperature
s	singlet, sharp
S	spin quantum number
SCE	saturated calomel electrode
SHE	standard hydrogen electrode
S-R	thiol-alkyl
SRCD	synchrotron radiation circular chroism
t	triplet
Taps	N-[Tris(hydroxymethyl)methyl]-3-aminopropanesulfonic acid
t-BuOK	potassium *tert*-butoxide
TCD	thermal conductivity detector
TFA	trifluoroacetic acid
THF	tetrahydrofuran
Tris	tris(hydroxymethyl)methylamine
TsCl	tosyl chloride
UV	ultra-violet
V	volt
VB	valence band
Vis	visible
vs	*versus*