



HAL
open science

Bulk electrocatalytic NADH cofactor regeneration with bipolar electrochemistry

Chunhua Zhang, Huiting Zhang, Junying Pi, Lin Zhang, Alexander Kuhn

► To cite this version:

Chunhua Zhang, Huiting Zhang, Junying Pi, Lin Zhang, Alexander Kuhn. Bulk electrocatalytic NADH cofactor regeneration with bipolar electrochemistry. *Angewandte Chemie International Edition*, 2021, 10.1002/anie.202111804 . hal-03516107

HAL Id: hal-03516107

<https://cnrs.hal.science/hal-03516107>

Submitted on 7 Jan 2022

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Bulk electrocatalytic NADH cofactor regeneration with bipolar electrochemistry

Chunhua Zhang[a], Huiting Zhang[a], Junying Pi[a], Lin Zhang*[a], Alexander Kuhn*[a, b]

[a] Chunhua Zhang, Huiting Zhang, Junying Pi, Prof. Dr. Lin Zhang, Prof. Dr. Alexander Kuhn

Engineering Research Center for Nanomaterials, Henan University, Kaifeng, China

E-mail: lin.zhang@henu.edu.cn

[b] Prof. Dr. Alexander Kuhn

University Bordeaux, CNRS, Bordeaux INP, ISM, UMR 5255, ENSCBP, Pessac, France

E-mail:kuhn@enscbp.fr

Supporting information for this article is given via a link at the end of the document.

Abstract: Electrochemical regeneration of reduced nicotinamide adenine dinucleotide (NADH) is an extremely important challenge for the electroenzymatic synthesis of many valuable chemicals. Although some important progress has been made with modified electrodes concerning the reduction of NAD⁺, the scale-up is difficult due to mass transport limitations inherent to large-size electrodes. Here, we propose instead to employ a dispersion of electrocatalytically active modified microparticles in the bulk of a bipolar electrochemical cell. In this way, redox reactions occur simultaneously on all of these individual microelectrodes without the need of a direct electrical connection. The concept is validated by using [Rh(Cp*)(bpy)Cl]⁺ functionalized surfaces, either of carbon felt as a reference material, or carbon microbeads acting as bipolar objects. In the latter case, enzymatically active 1,4-NADH is electroregenerated at the negatively polarized face of the particles. The efficiency of the system can be fine-tuned by controlling the electric field in the reaction compartment and the number of dispersed microelectrodes. This wireless bioelectrocatalytic approach opens up very interesting perspectives for electroenzymatic synthesis in the bulk phase.

Oxidoreductases are an important class of enzymes with great potential to be applied in industrial synthesis of a variety of chiral compounds due to their unique chemo-, regio-, and stereoselectivities compared to conventional chemical synthesis.[1–3] Among all known oxidoreductases, almost 80% of them are nicotinamide adenine dinucleotide (NAD) dependent enzymes.[4–6] During the enzymatic reduction process, the reduced form of the cofactor (NADH) is the most crucial ingredient of the redox reaction. However, the high cost of NADH is an economic roadblock, and therefore the efficient regeneration of NADH is of particular interest for both academic and industrial applications.[7] The regeneration of NADH from its oxidized form NAD⁺ has been explored by chemical,[8,9] photochemical,[10–12] enzymatic,[13] as well as electrochemical[14,15] approaches. Among these different strategies, electrochemistry is an attractive and promising route, since the price of electricity is low and the electrochemical reaction can be easily controlled via the electrode potential. In a typical direct electrochemical NADH regeneration system, the buffer solution serves as a proton source, and NAD⁺ can be reduced in a two-step reaction mechanism. However, high overpotentials are necessary and the radicals obtained after the first step prefer to react with each other, leading to the formation of biologically inactive NAD₂ dimers.[16] Therefore, different catalysts have been used to modify the electrode surface in order to achieve efficient indirect

electrochemical regeneration of enzymatically active 1,4-NADH. Up to now, two main families of catalysts have been studied and immobilized on electrodes for efficient NADH regeneration. The first category comprises biological catalysts such as diaphorase. Since in this case the electron transfer can only occur when the distance between the redox cofactors within diaphorase and the electrode surface is $<14 \text{ \AA}$, [17] an additional mediator (such as methyl viologen, [18] or cobaltocene [15]) needs to be combined with the enzymes in order to facilitate the electron transfer in a mediated electron transfer (MET) mode. The second family of catalysts is based on organometallic compounds, like e.g. $[\text{Rh}(\text{Cp}^*)(\text{bpy})\text{Cl}]^+$, which has been first proposed by Steckhan and co-workers. [19] Different methods to immobilize this rhodium catalyst on electrode surfaces have been explored, not only to simplify the product purification, but also to avoid the deactivation of both the organometallic complex and the enzyme. [20–23]

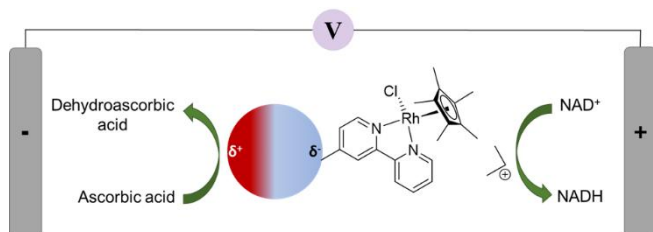
In the above mentioned studies, even though high electrocatalytic regeneration efficiencies of NADH have been achieved at the laboratory scale, the application of this electrochemical approach to industrial scale production is still limited by the intrinsic properties of classic electrodes. The two possible strategies for improving the amount of accessible catalyst and allowing a scale-up are based on the use of large-size working electrodes and an artificial increase of the active surface area by employing porous electrode structures (carbon felt, CNTs, etc.). However, functionalization of the inner pore surface with catalyst is difficult to control, and the overall electroenzymatic efficiency can also be influenced by local environmental changes (pH, concentration, etc.) inside the pores due to diffusion limitations. [24]

Therefore, a strategy to, at least partially, circumvent these problems is to employ multiple small electrodes, instead of one large porous electrode. However, in classic electrochemistry, the working electrode needs to be directly connected to a power supply through a conducting wire. This can become a limiting factor if one wants to connect a very large number of small electrodes, which in addition should be positioned ideally at different locations across the 3D reaction space. Bipolar electrochemistry provides the possibility to carry out electrochemical reactions simultaneously on multiple microelectrodes without the need of a direct electrical connection. [25–29] The basic principle of bipolar electrochemistry is based on the fact that when a conducting object is placed between two feeder electrodes in the solution, the two extremities will be polarized with respect to the surrounding solution and act simultaneously as cathode (δ^-) and anode (δ^+) when the electric field reaches a certain threshold value. In recent years, bipolar electrochemistry has been mainly studied in the context of materials science [30] and analytical chemistry. [31] Taking advantage of its inherent features, it has been used for the bulk synthesis of Janus particles by localized metal electrodeposition (Au, Pt, Cu, and AgCl). [32,33] Combined with enzymes, various wireless bioelectrochemical sensing systems have been developed, [34] and especially bulk electrochemiluminescence (ECL) could be achieved with a suspension of multiple bipolar electrodes. [35,36] This provides a solid background for carrying out electrochemical reactions on large assemblies of bipolar objects in the bulk mode.

Up to now, bipolar electrochemistry has never been explored in the frame of electroenzymatic synthesis. In this work, we demonstrate for the first time the possibility to electrochemically regenerate NADH at the δ^- extremity of multiple $[\text{Rh}(\text{Cp}^*)(\text{bpy})\text{Cl}]^+$ functionalized microparticles suspended in a bipolar electrochemical cell. Possible other reaction products, such as NAD² and 1,6-NADH, are undesirable side products of the reduction and their formation should be avoided. In this context the overall selectivity and efficiency (in terms of production rate in $\mu\text{mol}\cdot\text{m}^{-2}\text{ h}^{-1}$) of this wireless electrocatalytic system can be easily fine-tuned by adjusting the electric field strength and the number of bipolar electrodes. In this way, the polarization potential difference between the two

extremities of the bipolar object can be controlled to prevent the generation of enzymatically inactive byproducts.

The basic principle of this work is illustrated in Scheme 1. $[\text{Rh}(\text{Cp}^*)(\text{bpy})\text{Cl}]^+$ functionalized spherical carbon particles are positioned between two platinum feeder electrodes. The carbon spheres will be polarized under the influence of a constant electric field, and the two extremities will act as polarized cathode δ^- and anode δ^+ , respectively. The δ^- extremity will be responsible for the NAD^+ reduction, catalyzed by the immobilized rhodium complex, while ascorbic acid is used as a sacrificial redox couple for the oxidation at the δ^+ extremity, in order to avoid the re-oxidation of NADH due to the fact that ascorbic acid is easier to be oxidized than NADH (Supporting information, Figure S1).



Scheme 1. Schematic illustration of the bipolar electrochemical NADH regeneration.

Before directly going to multiple bipolar carbon electrodes, a first experiment concerning electrochemical NADH regeneration was carried with a classic electrochemical set-up using a piece of $[\text{Cp}^*\text{Rh}(\text{bpy})\text{Cl}]^+$ functionalized carbon felt (CF-Rh) as the working electrode. The covalent immobilization of the rhodium complex on the electrode was confirmed by NMR and XPS (Supporting information, Figure S2, S3), and its electrocatalytic response in the presence of NAD^+ was also validated (Figure S4). Then a piece of the CF-Rh electrode ($h * l * w, 10\text{mm} * 20\text{mm} * 3\text{mm}$) was placed in the bipolar cell and wireless NADH regeneration was attempted at its δ^- side. Proton and anion exchange membranes were used to design the bipolar electrochemical cell (see supporting information). In the reaction compartment, a low concentration of 50mM PBS was used, while in the two feeder electrode compartments, a high concentration of 250mM PBS was used in order to decrease the potential drop. The membranes eliminate, or at least decrease, the influence of parasitic phenomena occurring in the two feeder electrode compartments, such as bubble formation from water electrolysis or pH changes. The electric field in the reaction compartment was controlled by adjusting the external potential between the two feeder electrodes. According to the principle of bipolar electrochemistry, the potential difference between the two extremities of a bipolar electrode is proportional to its size d . As the length of CF-Rh is 3mm, the driving force ΔV can be estimated using the formula

$$\Delta V = \epsilon * d$$

with ϵ being the electric field in the cell. The oxidation potential of ascorbic acid is around +0.05V, the electrocatalytic NAD^+ reduction occurs at -0.65V, so theoretically the minimum threshold ΔV is 0.7V, corresponding to an electric field of 2.3V/cm.

Bipolar electrocatalytic NADH regeneration with the CF-Rh electrode was tested for different electric field values. The obtained enzymatically active 1,4-NADH was quantitatively measured after 60 min using an enzymatic method based on UV-vis spectroscopy[37] (see Figure S5 for the calibration curve of 1,4-NADH). Figure 1A illustrates the basic principle for testing the concentration of 1,4-NADH by addition of a NAD-dependent dehydrogenase after the bipolar NADH regeneration experiment. Since

1,4-NADH has a unique absorption peak at 340 nm, its concentration and enzymatic activity can be estimated by following the decrease of this absorption peak after addition of NAD-dependent alcohol dehydrogenase and acetaldehyde substrate. The progress of 1,4-NADH consumption by the enzymatic reaction is illustrated in Figure 1B.

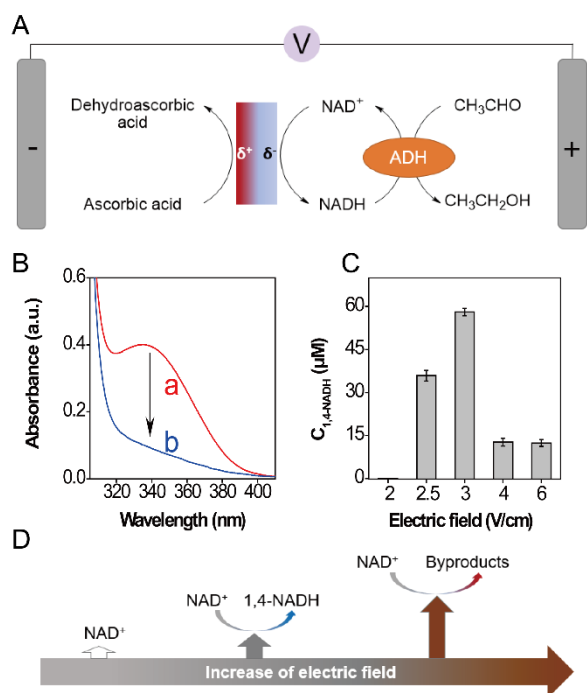


Figure 1. (A) Schematic illustration of the reactions involved for testing the concentration of enzymatically active 1,4-NADH. (B) UV-Vis absorption spectra of the bipolar electrochemical product obtained with an electric field of 3 V/cm after 60 min, before (curve a) and after (curve b) the addition of 100U alcohol dehydrogenase and 50mM acetaldehyde. (C) Concentration of produced 1,4-NADH as a function of the electric field in the bipolar cell after a period of 90 min. Experiments were carried out with cell 1 (see supporting information for details), a CF-Rh electrode (10mm*20mm*3mm) in 10 mL PBS pH 7.0 in the presence of 5 mM ascorbic acid and 1 mM NAD+ under nitrogen atmosphere. Error bars refer to the standard deviations of three measurements. (D) Scheme of different electrochemical reactions occurring with increasing electric field.

From Figure 1C it becomes clear that at electric field values below 2 V/cm no 1,4-NADH is produced since the driving potential hasn't reached the threshold value for the redox reactions to occur. However, when applying an electric field of 2.5V/cm, which is slightly higher than the theoretical threshold value, the transformation of 1,4-NADH, catalyzed by [Cp*Rh(bpy)Cl]⁺, can be achieved. The concentration of 1,4-NADH increases with increasing electric field, and an optimum transformation of 70 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ is observed at 3V/cm. The amount of enzymatically active 1,4-NADH starts to decrease when the electric field reaches 4V/cm. This decrease is caused by the direct electrochemical reduction of NAD⁺ at high overpotentials, leading to the formation of enzymatically inactive byproducts (NAD₂ and 1,6- NADH). This series of experiments with CF-Rh illustrates that the wireless electrochemical regeneration of 1,4-NADH can be achieved by carefully controlling the electric field in the bipolar cell (Figure 1D).

After this first proof-of-principle experiment with a single bipolar electrode, multiple bipolar electrodes were tested in the next step. The experiments were carried out with carbon microbeads

with a diameter of around $960 \pm 20 \mu\text{m}$. Spherical electrodes have been chosen because, due to their isotropic properties, the effective dimension exposed to the electric field does not depend on the orientation of the electrode. The same synthetic protocol as for the carbon felt was applied to modify the carbon beads with $[\text{Cp}^*\text{Rh}(\text{bpy})\text{Cl}]^+$ catalyst (CB-Rh). Elemental mapping of CB-Rh (Figure 2A) shows an efficient and uniform modification of the carbon beads. Then the production of 1,4-NADH has been monitored for different electric field values with forty CB-Rh positioned at the bottom of the bipolar cell. Theoretically, a decrease in electrode size by a factor of three requires a threefold increase in electric field in order to obtain the same driving force ΔV ($\Delta V = \epsilon \cdot d$). Figure 2B indicates that the optimum electric field for NADH regeneration is around 12.5 V/cm, which is slightly higher than the expected value. This might be attributed to differences in materials properties between carbon felt and carbon beads, resulting in variations in conductivity and overpotential with respect to the involved redox reactions.

In a following set of experiments, the bipolar electrochemical regeneration of NADH was performed with the optimum electric field of 12.5 V/cm, but by systematically varying the number of CB-Rh electrodes. A linear increase of 1,4-NADH concentration as a function of the number of beads was observed (Figure 2C). For example, the transformation rate was four times higher when increasing the number of microelectrodes from 10 to 40, indicating that each bipolar microelectrode operates independently in the reaction compartment. Even though the generated concentration of 1,4-NADH is limited during this experiment, when the production rate of NADH is renormalized with respect to the specific electrode surface area, a value of $630 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ is obtained with 40 beads. This value is significantly higher compared to the carbon felt. It shows promise with respect to a controlled increase in the efficiency of the system when adding more microelectrodes. However, the limited surface area at the bottom of the bipolar cell restricts the number of microbeads that can be employed. Therefore it would be interesting that beads occupy not only a 2D reaction plane, but are also present in the bulk phase of the set-up. Such a 3D reaction space should allow a further increase of the reaction efficiency.

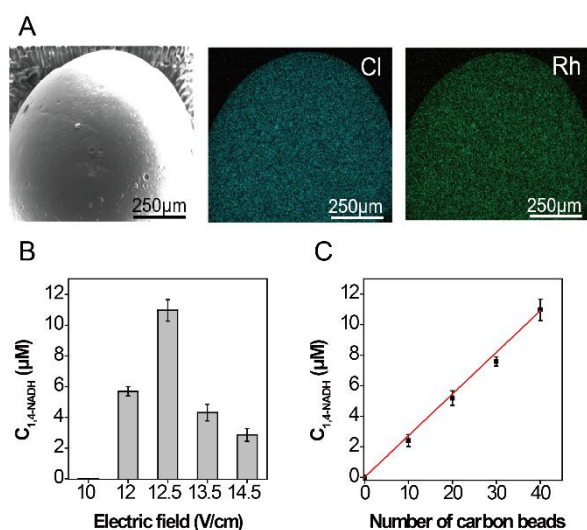


Figure 2. (A) SEM image of a CB-Rh bipolar electrode and the corresponding elemental mapping. Scale bar is 250 μm . Bipolar electrochemically produced NADH concentration (B) as a function of the applied electric field with 40 carbon beads and (C) as a function of the number of CB-Rh beads at an electric field of 12.5V/cm. Experiments were carried out with cell 1 (see supporting information for details), CB-Rh electrodes in 10mL PBS pH 7.0 in the presence of 5 mM ascorbic acid and 1 mM NAD⁺

under nitrogen atmosphere for 90 min. Error bars refer to the standard deviations of three measurements.

In order to maintain the carbon beads suspended in the bulk solution it is necessary to increase its viscosity. In previous work it has been demonstrated that bipolar electrochemistry can be carried out with gelified electrolytes.[32] Carboxymethyl cellulose (CMC) was reported as efficient additive for increasing the viscosity of aqueous solutions without changing their electrochemical performance.[38] Therefore, CMC has been added to the bipolar reaction compartment for increasing the viscosity of the electrolyte so that the microbeads do not sediment during the bipolar electrochemical process (Figure 3A). After testing different concentrations, 2% of CMC has been chosen as an optimal value (Figure S6). In order to be consistent with the previous experiments, CB-Rh particles of the same size were used and the same optimum electric field of 12.5V/cm was applied. Different concentrations of CB-Rh beads in the reaction compartment were employed (see Figure 3A for the configuration of the bipolar cell), and the produced NADH was measured after 90 min by UV-Vis spectroscopy. Figure 3B shows a photograph of the reaction compartment containing a suspension of CB-Rh electrodes. As illustrated in Figure 3C, a linear increase of the absorption peak at 340 nm was observed as a function of the concentration of suspended bipolar electrodes.

The renormalized production rate of NADH per surface area should be higher for the 3D-set-up (Figure 3D) compared to the 2D-set-up (Figure 2C). In the 3D-set-up, the beads are positioned further away from each other, thus the interference in terms of overlapping diffusion profiles and electric field distribution is expected to be less. In addition, the 3D arrangement allows more efficient hemispherical diffusion of educt and product, compared to the beads positioned at the bottom of the 2D-set-up, for which diffusion is restricted to the upper part of the beads. This should theoretically lead to a conversion that is roughly twice as efficient when using the 3D set-up instead of the 2D configuration. The measurements indicate an increase slightly larger than two times, with a maximum NADH production rate changing from 630 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ (2D case) to 1500 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{h}^{-1}$ for the 3D set-up. This demonstrates that the cofactor recycling efficiency can be significantly improved by the bulk approach, and the overall production can be further enhanced by increasing the concentration of beads. However, an appropriate distance should be kept between the CB-Rh beads in the bulk gel, in order to ensure that they are not touching each other during the reaction or generate a shadow effect of the electric field.

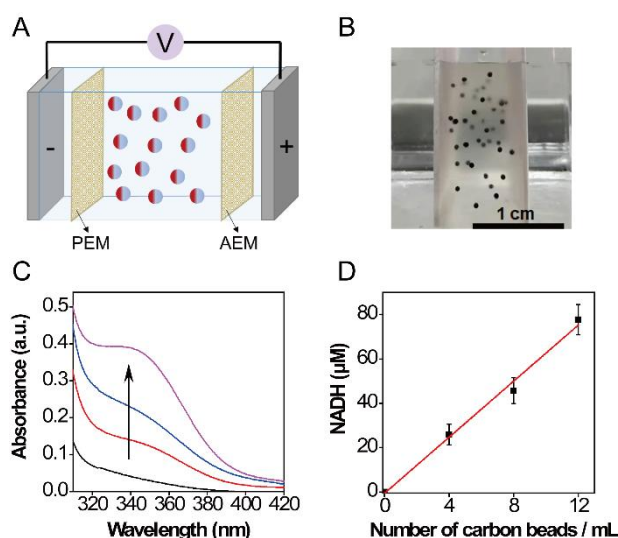


Figure 3. (A) Schematic representation of the bulk bipolar electrocatalytic NADH regeneration. PEM: proton exchange membrane (Nafion-117); AEM: Anion exchange membrane (FAB-PK-130). (B) A photograph of CB-Rh electrodes suspended in the reaction compartment. (C) UV-Vis spectra of bipolar products obtained with different concentrations of CB-Rh beads in 2% CMC gel in the presence of 5 mM ascorbic acid and 1 mM NAD⁺ at an electric field of 12.5V/cm under nitrogen. Black, red, blue, and purple lines correspond to 0, 20, 40, and 60 beads, respectively. (D) The concentration of produced NADH versus concentration of CB-Rh electrodes in 2% CMC gel. Experiments were carried out with cell 2 (see supporting information for details).

In conclusion, we propose a concept to carry out electrocatalytic NADH regeneration in a wireless way in the bulk volume of an electrochemical cell. Taking advantage of bipolar electrochemistry, the catalytic reaction can occur simultaneously at many [Cp*Rh(bpy)Cl]⁺ functionalized carbon beads. By controlling the electric field in the reaction compartment, enzymatically active 1,4-NADH can be selectively regenerated. The total efficiency can be improved by suspending the particles in a viscous electrolyte, allowing the bulk regeneration of the cofactor NADH. This method might be extended to achieve cofactor regeneration also with some other, non-molecular catalyst-functionalized, bipolar particles. It allows carrying out electroenzymatic reactions with a 3D ensemble of microelectrodes, operating without direct electrical connection. Even though the co-immobilization of both catalysts and enzymes on the microelectrodes is still a challenge, electroenzymatic systems with high efficiency are expected to be obtained by the rational design of the functionalized microobjects. This should open up interesting perspectives for scaling-up the present laboratory scale experiments to the level of large-scale bulk electroenzymatic synthesis in reactors, but obviously the usual problems like efficient heat dissipation, inhomogeneities and recycling of the catalyst will have to be studied in more detail.

In addition, the approach might become especially interesting when taking into account the current trend in the scientific community to reconsider chemical processes and explore more sustainable concepts based on electroorganic synthesis.[39]

Acknowledgements

The work has been funded by the National Natural Science Foundation of China (No. 21902045). The project has also been supported by the European Research Council (ERC) under the European Union's Horizon 2020 research and innovation program (grant agreement no 741251, ERC Advanced grant ELECTRA).

Keywords: NADH regeneration □ bipolar electrochemistry □ wireless electrocatalysis □ electroenzymatic synthesis □ electroorganic synthesis

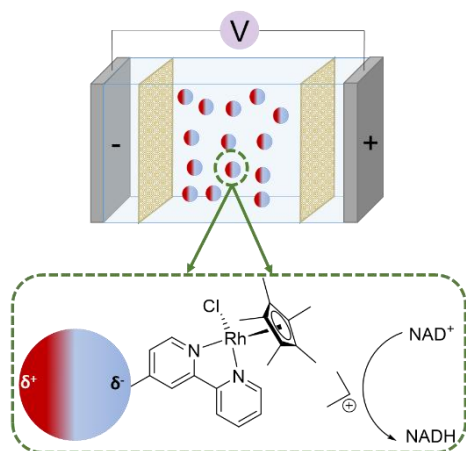
- [1] L. M. Schmitz, K. Rosenthal, S. Lütz, *Adv. Biochem. Eng. Biotechnol.* 2019, 167, 87–134.
- [2] L. Sellés Vidal, C. L. Kelly, P. M. Mordaka, J. T. Heap, *Biochim. Biophys. Acta - Proteins Proteomics* 2018, 1866, 327–347.
- [3] W. Andrea, G. Harald, H. Werner, *Adv Biochem Eng Biotechnol.* 2010, 120, 195–242.
- [4] X. Wang, T. Saba, H. H. P. Yiu, R. F. Howe, J. A. Anderson, J. Shi, *Chem* 2017, 2, 621–654.
- [5] L. Gorton, E. Dominguez, in *Encycl. Electrochem.*, 2002, pp. 67–143.

- [6] E. Simon, P. N. Bartlett, in *Biomol. Film.* (Ed.: J.F. Rusling), CRC Press, New York, 2003, p. 42.
- [7] H. Wu, C. Tian, X. Song, C. Liu, D. Yang, Z. Jiang, *Green Chem.* 2013, 15, 1773–1789.
- [8] M. M. Grau, M. Poizat, I. W. C. E. Arends, F. Hollmann, *Appl. Organomet. Chem.* 2010, 24, 380–385.
- [9] Y. Maenaka, T. Suenobu, S. Fukuzumi, *J. Am. Chem. Soc.* 2012, 134, 367–374.
- [10] X. Ji, J. Wang, L. Mei, W. Tao, A. Barrett, Z. Su, S. Wang, G. Ma, J. Shi, S. Zhang, *Adv. Funct. Mater.* 2018, 28, 1–14.
- [11] J. Huang, M. Antonietti, J. Liu, *J. Mater. Chem. A* 2014, 2, 7686–7693.
- [12] J. Kim, S. H. Lee, F. Tieves, D. S. Choi, F. Hollmann, C. E. Paul, C. B. Park, *Angew. Chemie - Int. Ed.* 2018, 57, 13825–13828.
- [13] F. Hollmann, I. W. C. E. Arends, D. Holtmann, *Green Chem.* 2011, 13, 2285–2314.
- [14] H. K. Song, S. H. Lee, K. Won, J. H. Park, J. K. Kim, H. Lee, S. J. Moon, D. K. Kim, C. B. Park, *Angew. Chemie - Int. Ed.* 2008, 47, 1749–1752.
- [15] M. Yuan, M. J. Kummer, R. D. Milton, T. Quah, S. D. Minter, *ACS Catal.* 2019, 9, 5486–5495.
- [16] A. Walcarius, R. Nasraoui, Z. Wang, F. Qu, V. Urbanova, M. Etienne, M. Göllü, A. S. Demir, J. Gajdzik, R. Hempelmann, *Bioelectrochemistry* 2011, 82, 46–54.
- [17] C. C. Page, C. C. Moser, X. Chen, P. L. Dutton, *Nature* 1999, 402, 47–52.
- [18] H. Chen, R. Cai, J. Patel, F. Dong, H. Chen, S. D. Minter, *J. Am. Chem. Soc.* 2019, 141, 4963–4971.
- [19] E. Steckhan, S. Herrmann, R. Ruppert, E. Dietz, M. Frede, *Organometallics* 1991, 10, 1568–1577.
- [20] L. Zhang, N. Vilà, G.-W. Kohring, A. Walcarius, M. Etienne, *ACS Catal.* 2017, 7, 4386–4394.
- [21] B. Tan, D. P. Hickey, R. D. Milton, F. Giroud, S. D. Minter, *J. Electrochem. Soc.* 2015, 162, H102–H107.
- [22] L. Zhang, N. Vilà, A. Walcarius, M. Etienne, *ChemElectroChem* 2018, 5, 2208–2217.
- [23] L. Zhang, M. Etienne, N. Vila, T. X. H. Le, G.-W. Kohring, A. Walcarius, *ChemCatChem* 2018, 10, 4067–4073.
- [24] L. Zhang, C. Carucci, S. Reculosa, B. Goudeau, P. Lefrançois, S. Gounel, N. Mano, A. Kuhn, *ChemElectroChem* 2019, 6, 4980–4984.
- [25] J. R. Backhurst, J. M. Coulson, F. Goodridge, R. E. Plimley, M. Fleischmann, *J. Electrochem. Soc.* 1969, 116, 1600.
- [26] C. A. C. Sequeira, D. S. P. Cardoso, M. L. F. Gameiro, *Chem. Eng. Commun.* 2016, 203, 1001–1008.
- [27] L. Koefoed, S. U. Pedersen, K. Daasbjerg, *Curr. Opin. Electrochem.* 2017, 2, 13–17.
- [28] S. E. Fosdick, K. N. Knust, K. Scida, R. M. Crooks, *Angew. Chem. Int. Ed.* 2013, 52, 10438–10456.

- [29] G. Loget, D. Zigah, L. Bouffier, N. Sojic, A. Kuhn, *Acc. Chem. Res.* 2013, 46, 2513–2523.
- [30] N. Shida, Y. Zhou, S. Inagi, *Acc. Chem. Res.* 2019, 52, 2598–2608.
- [31] K. L. Rahn, R. K. Anand, *Anal. Chem.* 2021, 93, 103–123.
- [32] G. Loget, J. Roche, A. Kuhn, *Adv. Mater.* 2012, 24, 5111–5116.
- [33] G. Loget, A. Kuhn, *J. Mater. Chem.* 2012, 22, 15457–15474.
- [34] L. Bouffier, D. Zigah, N. Sojic, A. Kuhn, *Annu. Rev. Anal. Chem.* 2021, 14, 65–86.
- [35] M. Sentic, S. Arbault, L. Bouffier, D. Manojlovic, A. Kuhn, N. Sojic, *Chem. Sci.* 2015, 6, 4433–4437.
- [36] A. De Poulpiquet, B. Diez-Buitrago, M. Dumont Milutinovic, M. Sentic, S. Arbault, L. Bouffier, A. Kuhn, N. Sojic, *Anal. Chem.* 2016, 88, 6585–6592.
- [37] T. Saba, J. W. H. Burnett, J. Li, P. N. Kechagiopoulos, X. Wang, *Chem. Commun.* 2020, 56, 1231–1234.
- [38] P. Galek, A. Slesinski, K. Fic, J. Menzel, *J. Mater. Chem. A* 2021, 9, 8644–8654.
- [39] D. Pollok, S. R. Waldvogel, *Chem. Sci.* 2020, 11, 12386–12400.

Entry for the Table of Contents

Insert graphic for Table of Contents here.



Wireless electrocatalytic regeneration of the enzymatically active cofactor NADH is achieved on functionalized carbon beads suspended in the bulk phase of a bipolar electrochemical cell. The

catalytic efficiency can be fine-tuned by the concentration of suspended particles and the applied external electric field.