

Recent trends in artificial photosynthetic system

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Abstract. Solar energy is the most abundant and clean energy on the earth, and developing equipment or devices with high-efficiency solar energy conversion is an effective way to alleviate the energy crisis. The majority of redox enzymes require a coenzyme to provide the hydrogen source needed for the reaction process, and the most commonly used coenzyme is nicotinamide adenine dinucleotide (NADH). However, NADH is expensive and easily decomposed, which greatly limits its application of artificial photosynthetic systems. We review recent progress in the construction of artificial photosynthetic systems that mimic natural photosynthesis.

Key word: NADH regeneration; Artificial photosynthesis; Photocatalyst.

1. Introduction

Currently, NADH regeneration is mainly classified into enzymatic, chemical, electrochemical and photochemical methods. Among them, electrochemical method is considered to be an effective method owing to its low cost. Electrochemical NADH regeneration is produced by NAD^+ accepting electrons transferred from the electrode and undergoing a redox reaction. In this case, the initial reduction step is reversible and NAD^+ is reduced to NAD^\cdot , which then proceed to a reduction reaction to accept protons to form active 1,4-NADH. However, NAD^\cdot is unstable and tends to form NAD_2 dimers, which subsequently generate further 1,6-NADH without activity.¹ To reduce the appearance of NAD_2 dimers, the researchers introduced electron mediators to assist in the transfer of electrons, which facilitated the generation of 1,4-NADH. The introduction of $[\text{Cp}^*\text{Rh}(\text{bpy})(\text{H}_2\text{O})]^{2+}$ significantly improved the reaction kinetics of 1,4-NADH and promoted the targeted regeneration of 1,4-NADH, which is of great significance in the field of coenzyme regeneration. With the gradual improvement in the understanding of optical properties of semiconductor materials, researchers mimic natural plant photosynthesis to design artificial photosynthetic systems for NADH regeneration using clean, renewable solar energy that are considered a highly potential strategy for development.²⁻⁴ The main part is composed of photocatalyst, electron mediator, electron sacrificial agent and NAD^+ . Here, we review recent progress in the construction of artificial photosynthetic systems that mimic natural photosynthesis. Among them, photocatalysts, electron sacrificial agents, electron mediators, NAD^+ /NADH and redox enzymes are mainly involved. This review first

introduces the mechanism of natural photosynthesis in plants. Then, we compare two artificial photosynthetic systems constructed to mimic natural photosynthesis. In the next section, we present the functions and recent advances of each component of artificial photosynthetic systems respectively. Finally, we summarize the obstacles encountered in the practical application of artificial photosynthetic systems and discuss the solutions.

2. Natural photosynthesis

Photosynthesis is when plants, algae and certain photosynthetic bacteria utilize light energy, CO_2 and H_2O to produce carbohydrates and release oxygen. It is divided into two main reaction stages, the light reaction and the dark reaction (Calvin cycle). The light reaction combines redox reactions between various protein components that occur on the thylakoid membrane. There are two photosynthetic systems on the thylakoid membrane, namely photosynthetic system I and photosynthetic system II, which absorb at different wavelengths. As shown in Figure 1, First, the light harvesting complex II (LHCII) absorbs light energy and transfers it to the photosynthetic reaction center II (PSII, P_{680}), which receives energy to excite photogenerated electrons, which migrate to the conduction band and leave photogenerated holes with oxidative properties in the valence band. Then, 1 mol H_2O is oxidized by the photogenerated holes to produce 1 mol O_2 , 4 mol electrons and 2 mol H^+ . Then, the electrons from H_2O oxidation obliterate the photogenerated holes produced by PSII, and the photogenerated electrons from PSII are transferred to PSI along the electron transfer chain.

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Like photosynthetic system II, LHCl absorbs light energy and transfers it to PSI, which excites photogenerated electrons to transfer to the conduction band, leaving photogenerated holes in the valence band. At this time, the photogenerated holes generated by PSI are obliterated by the photogenerated electrons from PSII (P₇₀₀), realizing the energy transfer between the two systems. Finally, the electrons are utilized by Fd-NADP reductase to reduce NAD(P)⁺ to NAD(P)H. At the same time, as the oxidation of H₂O occurs inside the thylakoid, a transmembrane proton (H⁺) potential is formed, and the transfer and release of H⁺ drive the synthesis of ATP by ATPase. The formed ATP and NAD(P)H enter the Calvin cycle to participate in the reduction of CO₂. Natural photosynthesis can be divided into hydrolysis, light collection and excitation, electron transfer, NAD(P)H and ATP regeneration and the Calvin cycle according to the energy transformation process.

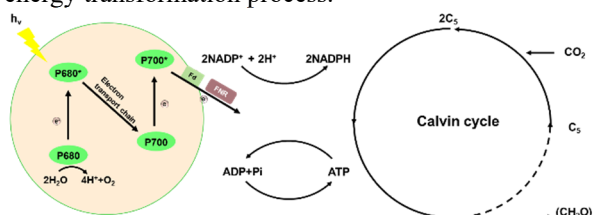


Figure 1. Schematic of the charge separation process and mass transfer of natural photosynthesis

3. Artificial photosynthesis

Artificial photosynthesis is a process inspired by natural photosynthesis, mimicking the function of each functional component in the cystoid to realize the conversion of light energy into chemical energy, which mainly consists of five components: photocatalyst, electron sacrificial agent, NAD⁺/NADH, electron mediator and oxidoreductase. Among them, the photocatalytic reaction is mainly divided into three sequential processes: (1) The photocatalyst absorbs light energy and excites electrons to migrate from the valence band to the conduction band, producing photogenerated electrons and holes. (2) Separation and migration of the excited photogenerated electrons to the active sites on the surface for accumulation. (3) The strongly oxidizing photogenerated holes obliterate the photogenerated holes by redox reactions with electron sacrificial agents.

According to the different mimicking plant photosynthetic systems, artificial photosynthetic systems can be divided into two types: PSII water-splitting artificial photosynthetic systems and PSI light-enzyme coupled artificial photosynthetic systems. The roles played by photocatalysts are the same in both systems. PSII water-splitting artificial photosynthetic systems is actually the photocatalytic splitting of water molecules for hydrogen production. In photocatalytic water splitting, the energy value of the difference between the conduction band and the valence band is called the band gap energy (E_g). When photocatalyst is irradiated by ultraviolet light, if photocatalyst absorbs light energy ($h\nu > E_g$), photocatalyst will generate photogenerated electrons, which will be excited from the valence band to the

conduction band within femtoseconds, generating photogenerated electron-hole pairs.⁵ According to Figure 2, to achieve photocatalytic water splitting, the reduction potential of the conduction band minimum of the photocatalyst must be less than the reduction potential of H⁺ to H₂ (-0.41 V vs NHE, pH 7). The oxidation potential of the valence band must be greater than the Oxidation potential of H₂O to O₂ (+0.82 V vs NHE, pH 7). Therefore, the minimum photon energy required by photosensitizers for water splitting is 1.23 V, which severely limits the application of various photocatalysts in photocatalytic water splitting.

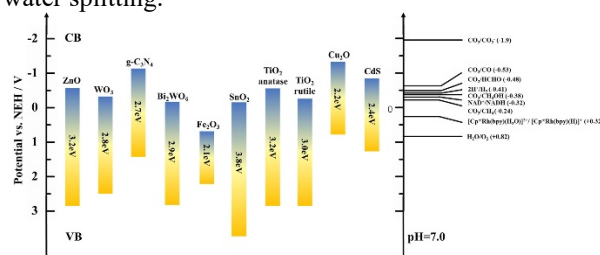


Figure 2. The band gap of some photocatalysts and the redox potential of some chemical species at pH of 7.0

Compared with the PSII water-splitting artificial photosynthetic system, the PSI light-enzyme coupling artificial photosynthetic system is more sophisticated, involving multiple factors such as photocatalyst, electron sacrificial agent, electron mediator, coenzyme factor and redox enzyme. As shown in Figure 3, the process is divided into three reactions: photocatalytic, NAD(P)H regeneration, and redox. The specific process is slightly different from the photo-splitting of water. The photocatalyst absorbs light energy and excites the photogenerated electrons, which are transferred to the electron mediator, reducing NAD(P)⁺ to NAD(P)H. NAD(P)H participates in oxidoreductase reactions and generates products with stable chemical energy. The electron holes generated by photocatalyst excitation are annihilated by electron sacrificial agents such as H₂O and triethanolamine (TEOA).

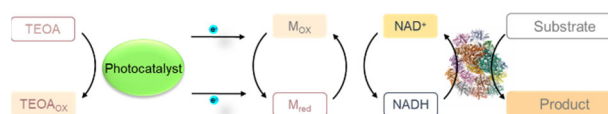


Figure 3. Illustration of the PSI light-enzyme coupling artificial photosynthetic system

4. NAD(P)⁺/NAD(P)H

In the artificial photosynthetic system, the continuous and efficient generation of NAD(P)H with enzymatic activity is the key to ensuring the high efficiency of the artificial photosynthetic system. Similar to the photo-splitting of water, achieving NAD(P)H regeneration requires a photosensitizer that satisfies the redox potential of the NAD(P)⁺/NAD(P)H (-0.32 V vs NHE, pH 7). In addition, in the process of coenzyme regeneration, not only 1,4-NAD(P)H with enzymatic activity will be produced, but also isomers such as 1,2-NAD(P)H, 1,6-NAD(P)H,

irreversibly changes the structure of the coenzyme factor, resulting in the waste of the coenzyme factor.^{6,7} In order to reduce the appearance of isomers, researchers usually introduce electron mediator and electron sacrificial agents when constructing artificial photosynthetic systems to ensure the efficient generation of 1,4-NAD(P)H.

5. Electron mediator (M)

In the NADH regeneration system, the most commonly used M is $[\text{Cp}^*\text{Rh}(\text{bpy})\text{H}_2\text{O}]^{2+}$, because $[\text{Cp}^*\text{Rh}(\text{bpy})\text{H}_2\text{O}]^{2+}$ has a high area for the regeneration of 1,4-NADH selective. $[\text{Cp}^*\text{Rh}(\text{bpy})\text{H}_2\text{O}]^{2+}$ first accepts electrons to form $[\text{Cp}^*\text{Rh}(\text{bpy})]$, and then H^+ in aqueous solution combines with the Rh metal center to form $[\text{Cp}^*\text{Rh}(\text{bpy})\text{H}]^+$ with catalytic activity. Finally, it binds to NAD^+ to drive the enzymatic production of 1,4-NADH.^{8,9} Although the electron mediator $[\text{Cp}^*\text{Rh}(\text{bpy})\text{H}_2\text{O}]^{2+}$ performs exceptionally well in the regeneration reaction of 1,4-NADH, it is expensive. It is in a free state in the NADH regeneration system, making it difficult to recover and reuse. The formation of $[\text{Cp}^*\text{Rh}(\text{bpy})\text{H}_2\text{O}]^{2+}$ is a coordination reaction between the pentamethylcyclopentadienyl rhodium(III) chloride dimer and the bipyridine moiety. According to Figure 4a, Chen et al. used bipyridine containing compounds as monomers for synthesising COF materials. The electron mediator M was modified on the COF skeleton through a coordination reaction, significantly promoting NADH regeneration, 1, 4-NADH achieved a regeneration rate of 80% with 20 min, which was 4 times higher than that of the immobilised M material. The reason may be that M acts as a reactive site to facilitate the separation of photogenerated charges.¹⁰ Therefore, research on more efficient, sustainable, and low-cost M immobilization will help to improve the reaction rate and efficiency of artificial photosynthetic systems.

6. Electron sacrificial agent

The role of the electron sacrificial agent is to annihilate the electron holes generated by the photocatalyst and inhibit the recombination of photogenerated electrons and holes. Typical electron sacrificial agents are triethanolamine (TEOA), ethylenediaminetetraacetic acid (EDTA), organic acids (formic acid, ascorbic acid), sulfur compounds (methionine), and zwitterionic buffers.¹¹ They can stably donate electrons to photocatalysts at a suitable pH value and are widely used in NADH regeneration. However, using electron sacrificial agents usually introduces impurities, making separation and recovery difficult. In the natural photosynthetic reaction of plants, H_2O acts as an electron sacrificial agent to provide electrons to the photosynthetic center, and 1 mol of water molecules are completely decomposed to produce 4 mol electrons, 2 mol H^+ and 1 mol O_2 , which is clean and pollution-free. However, because H_2O is extremely stable, it has higher requirements for the performance of photocatalysts. Therefore, using H_2O as an electron sacrificial agent is one of the difficulties to be solved in the future.

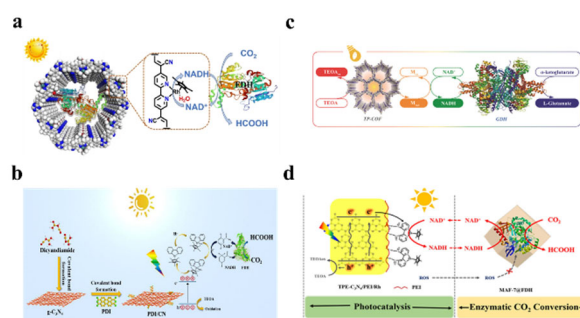


Figure 4. a The artificial photocatalytic system using COF-based photocatalyst and FDH. b Illustration of the artificial photocatalytic system for coenzyme regeneration and l-glutamate by l-glutamate dehydrogenase (GDH). c PDI/CN/FDH artificial photocatalytic system for CO_2 reduction to HCOOH under visible light irradiation. d TPE-C3N4/PEI/Rh/FDH artificial photocatalytic system for CO_2 reduction to HCOOH under visible light irradiation.

7. Photocatalyst

The absorption of light energy by photocatalysts to excite electrons is the basis of artificial photosynthesis. In this process, if the electrons cannot be utilized in time, the photogenerated electrons and photogenerated holes will recombine and be released in the form of heat and fluorescence, resulting in a waste of electrons, which affects the electron excitation efficiency and transfer efficiency of the artificial photosynthetic system.¹² Therefore, searching for stable and efficient new photosensitizers has attracted many researchers research. According to the current development of photocatalysts, they can be classified into organic dyes, metal oxides, covalent organic frameworks, carbon-based materials, quantum dots, etc.

With the in-depth study of photocatalysts, advanced synthesis processes have enabled the manufacturing of more complex nanostructured materials. By designing and controlling the process of photocatalyst synthesis, nano photocatalysts with various morphologies have been developed. Carbon nitride is a new type of inorganic conjugated semiconductor photocatalyst discovered by Wang et al., and it is used in photocatalytic hydrogen production.¹³ Not only are their abundant sources of raw materials, simple synthesis, and low cost, but they also have excellent physical and chemical properties, such as flexible electronic structures, strong heat resistance, and strong stability. Carbon nitride with different morphologies can be synthesized by designing and controlling the synthesis route, such as carbon nitride nanosheets (CNS), mesoporous carbon nitride (CNM), carbon nitride nanorods, carbon nitride prisms, carbon nitride lines etc.¹⁴⁻²⁰ However, some limitations remain, such as limited light absorption ability, poor charge mobility, and poor crystallinity. In order to improve the optical properties of carbon nitride, some studies have attempted to form heterojunctions between carbon nitride and other nanomaterials to improve photocatalytic efficiency. According to Figure 4b, Li et al. prepared a metal-free heterojunction PDI/CN by constructing an amide bond between pyromellitic diimide (PDI) and

carbon nitride (CN). This resulted in an NADH regeneration rate of 75% and formic acid generation. The rate is $1.269 \text{ mmol g}^{-1} \text{ h}^{-1}$, which is 2.6 times higher than that of pristine CN.²¹ This is due to the more vital visible light absorption ability of the heterojunction PDI/CN, which is beneficial to the NADH regeneration reaction and photocatalytic CO_2 reduction.

Covalent organic frameworks (COFs) are a class of highly crystalline porous polymers composed of hydrophobic organic molecules.²² Its structure and photochemical function are realized by regulating the modular design of small organic molecules, which has a high degree of structural adjustability. Basic units such as triazine rings, thiophenes, and porphyrins are constructed by designing covalent bonds to form COFs with two-dimensional or three-dimensional structures. Therefore, COFs can form nanoscale, uniform and adjustable pores, which endow materials with special optical properties, and have great application potential in photocatalysis, energy and the environment. According to Figure 4c, Zhao et al. synthesized TP-COFs with high crystallinity and porosity using monomeric TFPT with triazine-based structural units and monomeric PDAN. The regeneration rate of NADH reached 90.35%, showing a record NADH reduction efficiency. After coupling with oxidoreductase, the conversion rate of L-glutamic acid reached 97 % within 12 min.²³ The main reason may be that the olefin ($-\text{C}=\text{C}-$) linkage is used instead of the traditional imide ($-\text{C}=\text{N}-$) linkage, and the sp^2 carbon bridge provides TP-COF with stronger electron delocalization characteristics and stability.

8. Oxidoreductase

At present, the oxidoreductases coupled with the photocatalytic NADH regeneration system mainly include L-glutamate dehydrogenase (GDH), formate dehydrogenase (FDH) and old yellow enzyme (OYE). GDH uses NADH to convert α -ketoglutarate into L-glutamic acid as a model enzyme to detect whether the coenzyme regeneration system produces active NADH. FDH is a utility enzyme that converts CO_2 into formic acid with the assistance of NADH. In some studies, FDH, alcohol dehydrogenase (ADH) and formaldehyde dehydrogenase (ALDH) are also used to form an enzyme-catalyzed cascade system coupled with a photocatalytic NADH regeneration system to catalyze the conversion of CO_2 into methanol. Transfer of light energy to stable chemical energy and conversion of CO_2 to important fuels and renewable chemicals. However, the oxidoreductase is in a free state in the catalytic system, which is not conducive to separation and recovery, so it is not easy to meet the needs of industrialization.

In order to solve the recyclability problem of oxidoreductases, researchers began to pay attention to immobilized enzyme technology. Basic methods for immobilizing enzymes include adsorption, encapsulation, covalent binding, entrapment, and cross-linking. When constructing a light-enzyme coupled artificial photosynthetic system, avoiding the loss of oxidoreductase activity caused by the photocatalytic

NADH regeneration process is necessary. The photogenerated holes generated by photocatalysis have oxidative solid properties, and superoxide radicals are easily generated in contact with oxygen or water, which is one of the critical factors for the loss of enzyme activity. According to Figure 4d, Song et al. encapsulated FDH into a metal-organic framework (MOF, MAF-7) to reduce CO_2 to formic acid by combining an enzyme-catalyzed reaction and regeneration of NADH.²⁴ This encapsulation strategy isolates FDH from the external environment, provides a stable microenvironment for FDH, and enhances the stability of the enzyme. Therefore, the immobilization of oxidoreductases can not only reduce the number of enzymes used, and avoid the separation and recovery of products, but also have the advantage of maintaining the stability and activity of enzymes, which is very important for the practical application of artificial photosynthetic systems in the future.

9. Summary and Outlook

Solar energy is the most abundant and clean energy on the earth, and developing equipment or devices with high-efficiency solar energy conversion is an effective way to alleviate the energy crisis. The natural photosynthesis of plants is a common way in nature to utilize solar energy efficiently. By simulating this process, an artificial photosynthetic system is constructed to realize the co-catalysis of photocatalyst and oxidoreductase to produce high-value-added products. The obstacle that limits the practical application of artificial photosynthetic systems is that the functional components are in a free state, which is challenging to meet the needs of industrialization. One is that the free functional components and products are difficult to recover and separate. Second, the oxidoreductase activity in the free state is easily lost. Third, using an electron sacrificial agent is easy to introduce new impurities. In this review, we discuss some related work on immobilizing functional components. Compared with free functionalized components, immobilized electron mediators and oxidoreductases showed excellent reproducibility, and shortened distance between functionalized components facilitated the mass transfer efficiency of electrons and substances. In order to realize the practical application of future artificial photosynthetic systems, the immobilization of multifunctional components is the future development trend. At the same time, in the artificial photosynthetic system, H_2O is the cleanest electron sacrificial agent. However, due to the high energy required to achieve water splitting, most photocatalysts are challenging to meet their requirements, and the performance of photocatalysts needs to be improved in the future. Exploring the above challenges will facilitate the development of artificial photosynthetic systems and their applications.

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