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Design of new strategy for green algal photo-hydrogen production: Spectral-selective photosystem I activation and photosystem II deactivation

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HIGHLIGHTS

- ▶ A new strategy in photo-H_2 production from green algae was successfully demonstrated.
- ▶ The new strategy is convenient because it only requires turning on and off PSI light.
- ▶ The PSI light (692 nm) achieved initial photo-H_2 production rate of 0.055 mL H_2 mg^{-1} Chl h^{-1}.
- ▶ The PSI light (692 nm) achieved maximal total hydrogen production of 0.108 mL H_2 mg^{-1} Chl.
- ▶ The new strategy shows promise of improved photo-H_2 production through further optimization.

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ABSTRACT

A new strategy in photosynthetic hydrogen (photo-H_2) production from green algae was developed based on theory and successfully demonstrated. The new strategy applied a spectral-selective photosystem I (PSI) activating/photosystem II (PSII) deactivating radiation (or PSI light) that would drive a steady flow of electrons in the electron transport chain for delivery to hydrogenase for photo-H_2 production, but would reduce oxygen production through water photolysis below the respiratory oxygen consumption so that an anoxic condition would be maintained as required by hydrogenase. Implementing the strategy by using a PSI light (692 nm peak, 680–700 nm) on *Chlamydomonas reinhardtii* cells resulted in relatively sustained photo-H_2 production (total of 0.108 mL H_2 mg^{-1} Chl, exceeding 0.066 mL H_2 mg^{-1} Chl under white light). The strategy also proved successful and convenient in allowing cells to alternately switch between photo-H_2 production and a recovery period by simply turning on or off the PSI light.

1. Introduction

Light-driven hydrogen production (photo-H_2) in green algae, such as *Chlamydomonas reinhardtii*, issues from the light-dependent reactions of photosynthesis as an alternate pathway when conditions are sufficiently anoxic, giving rise to the synthesis of the enzyme hydrogenase, which catalyzes the production of hydrogen. In regular photosynthesis, the radiation activation of Photosystems II (PSII) incites photolysis, which splits water molecules into electrons, unbound hydrogen and oxygen. In this aerobic condition, the photosynthetic light-dependent reactions' electron transport chain terminates with $NADP^+$ accepting the electrons and unbound hydrogen, yielding the reduced form NADPH. Under anoxic conditions, however, it is hydrogenase that accepts the electrons and, using the available protons, catalyzes the production of hydrogen molecules (Melis et al., 2000).

The green alga *C. reinhardtii* has been the model organism used in recent studies on photo-H_2 production. Photo-H_2 production is catalyzed by hydrogenase in a number of green algal species (Kessler, 1974). Given its high sensitivity to oxygen, hydrogenase is immediately inhibited in the presence of even a small amount of oxygen (Erbes et al., 1979). Thus, photo-H_2 production does not last long under illumination on account of the immediate inhibition of hydrogenase by oxygen which is generated by photolysis (Ghirardi et al., 1997).

Among the strategies developed for photo-H_2 production (Gfeller and Gibbs, 1984; Miura et al., 1982; Melis et al., 2000; Posewitz et al., 2004; Oh et al., 2011; Hallenbeck et al., 2012), the sulfur-deprivation method applied to *C. reinhardtii* cell culture has, thus far, shown the most promise (Esquivel et al., 2011). In this method, as sulfur is deprived, the synthesis of structural proteins in the PSII complex is inhibited, degrading PSII and resulting in the suppression of the oxygen-generating photolysis. With the ensuing anaerobic condition, hydrogenase is synthesized and its activity is sustained over long periods. The implementation of

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