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Supplementary Information

The electrochemical ion membranes system (EIMs) enhanced light reactions of photosynthesis with intermittent electrical stimulation

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Experimental

1.1 Strains and Cultural Conditions

The microalgae strain *Dunaliella salina* HTBS used in present study was screened and maintained in our lab[1]. For seed cultivation, cells were cultured to logarithmic phase in sea-water BG11 medium under 80 μmol.m^{-2}.s^{-1} at 25 °C. Then the cells were centrifuged with 6000 rpm for 5 min for further use. The initial cell density in EIMs was kept at OD680=1 using a UV-vis spectrophotometer (UV-1800PC, Shanghai Mapada Instruments) and subsequently maintained at 25 °C with a light intensity of 20 μmol m^{-2} s^{-1}. Additionally, the EIMs microalgae champer was equipped with a rotor that continuously stirred the culture upon.

1.2 Design and Optimization of EIMs

To mitigate the effects of carbon supply and pH on photosynthesis within the microalgae chamber, the saturated $NaHCO_3$ solution in the cathode chamber was replaced with ddH_2O. Meanwhile, to facilitate the collection of byproducts, the EIMs system was modified into a sealed structure, enclosing the tops of both the cathode and anode chambers while incorporating an opening for gas collection.

1.3 Measurement of Chloroplast Fluorescence Parameters

For chloroplast fluorescence parameters analysis, 2 mL of microalgae culture was dark-adapted for 15 min before measuring Fv/Fm, Y(II), and R_{fd} using a Palm Water Chlorophyll Fluorometer (FluorPen AP110-C, Czech Republic).

1.4 Measurement of ATP and NADPH

Intracellular ATP and NADPH contents were detected using the ATP Content Assay Kit (Solarbio, UK) and $NADP^+$/NADPH Assay Kit with WST-8 (Beyotime,

China) for extraction, respectively. Briefly, 1 mL of microalgae culture was centrifuged at 6,000 rpm for 5 min, followed by the addition of 1 mL ATP extraction buffer, mixing, and ultrasonic disruption (200 W, 1 min). After centrifugation at 12,000 rpm for 10 min at 4°C, the supernatant was mixed with 500 μL chloroform, and centrifuged again at 12,000 rpm for 5 min at 4°C. The absorbance at 340 nm was measured for 10 s, followed by a 3-minute incubation at 25°C before a second absorbance measurement for ATP detection. For NADPH measurement, a similar extraction method was used with 200 μL of NADPH extraction buffer. After centrifugation, 100 μL of the supernatant was incubated at 60 °C for 30 minutes and then allowed to cool to room temperature. Subsequently, 50 μL of the supernatant was mixed with 100 μL of the NADPH standard solution and incubated at 37°C in the dark for 10 min. After adding 10 μL of color developing solution, the plate was incubated for an additional 10 minutes, and the absorbance was measured at 450 nm. NADPH content in each well was calculated based on the standard curve.

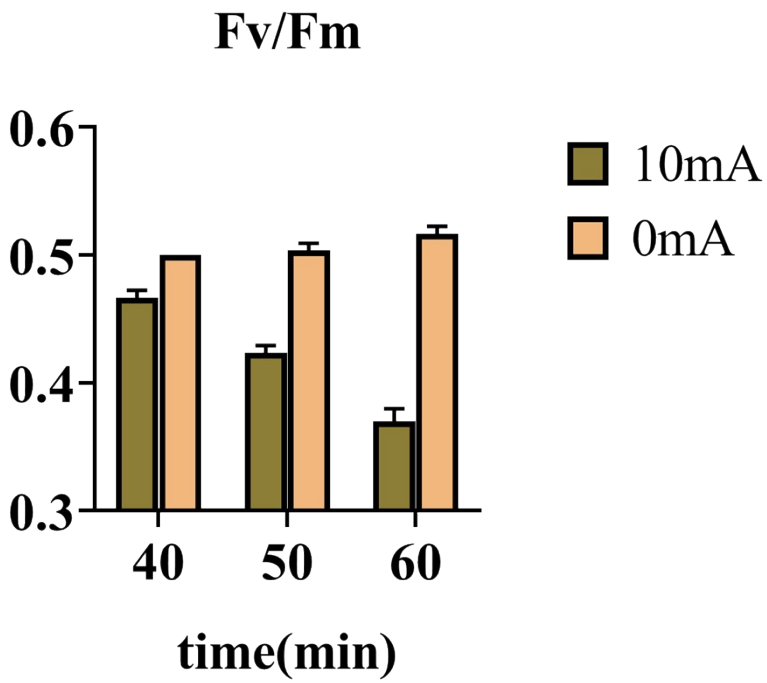

Figure S1. The Fv/Fm value of cultured cells in EIMs and control group at 40, 50 and 60min.

References

1. Y. Hou, T. Han, R. Wu, Z. Liu, Y. Ma, Z. Guo, N. Hao, W. Wang, X. Ji, Z. Zhu, F. Chen and L. Zhao, Green Chemistry, 2023, 25, 7273-7282.