Harvesting electrical current from intact plant leaves

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Abstract

Efforts to replace fossil fuels with renewable energy technologies, especially solar energy conversion, continue to improve the potential to produce useful amounts of energy without significant pollution. Utilization of photosynthetic organisms in bio-photo electrochemical cells (BPECs) are a potentially important source of clean energy. Here, we show that it is possible to harvest photocurrent directly from unprocessed plant tissues in specialized BPECs. The source of electrons are shown to originate from the Photosystem II water-oxidation reaction that results in oxygen evolution. In addition to terrestrial and crop plants, we further demonstrate the ability of the desert plant *Corpuscularia lehmannii* to produce bias-free photocurrent without the addition of an external electrolyte. Finally, we show the use of pond-grown water lilies to generate photocurrent. Different leaves produce photocurrent densities in the range of ~ 1 – 10 mA / cm^2 which is significantly higher than microorganism-based BPECs. The relatively high photocurrent and the simplicity of the plants BPEC may pave the way toward the establishment of first applicative photosynthetic based energy technologies.

Broader Context

It is no secret human society is experiencing an energy and environmental crisis due to our reliance on fossil fuels. In order to promote alternative, cleaner, and more sustainable approaches to energy production, we wish to explore the possibility of using nature's method of solar energy conversion in the simplest, least polluting, most sustainable fashion possible. Photosynthesis provides a remarkable example of molecular system for solar energy conversion to storable fuels. Many studies have strived

to merge natural photosynthesis (as isolated complexes, isolated membranes, or intact microorganisms) with a variety of electrochemical harvesting technologies. In this paper we show that we can directly couple the power of water oxidation by Photosystem II in intact plants to bio-electrochemical cells without the need to perform expensive, complicated, and polluting isolation. We show that current harvesting (up to current densities of 10 mA / cm^2) can be performed using plants of different types: plants of agricultural importance, succulents with internal water-based reservoirs and aquatic plants, used in situ in their growth ponds. We also show that with minimal external bias, hydrogen can be obtained, to be used as a clean fuel. We believe that these results can lead to the development of localized clean energy technologies, where the benefits of plant growth for any purpose can be enhanced by obtaining significant amounts of clean energy.

Introduction

World energy demands continue to increase steeply due to population growth and lifestyle requirements. It was estimated that by 2035, global energy consumption will reach an annual average of ~26 TW[1]. Current fossil fuel combustion technologies handle most energy generation requirements, however these lead to greenhouse gas emission and which may be a source of climate change[2,3]. The increasing awareness of fossil fuel pollution has led to the search for cleaner renewable energy technologies. These include utilization of biological systems, including bioelectricity generation technologies which are based on the harvesting of electrical current produced by live organisms[4–6] or by utilizing isolated enzymes[7–9] for the generation of light-induced electrical power.

The first report of use of microbes for the generation of electrical energy (microbial fuel cells, MFCs) was made by Potter et al. in 1910[10]. In this and similar technologies, bacteria are utilized as electron donors at the anode[8,11] while different bacterial species or terminal electron acceptors are linked to cathodes[8,12,13]. For the establishment of efficient electrical communication between the anode and the bacteria, direct electron transfer (DET)[14–17] and mediated electron transfer (MET)[18–21] methods have been developed. DET has been reported for the bacterial species such as *Geobacter, Sulfurreducen* and *Shewanella oneidensis* that use conductive inter-membranal protein complexes such as found in *pili* or MTR complexes to export electrons[22,23]. MET systems are more prevalent as they can be performed either by addition of exogenous mediators or by the secretion of endogenous reducing molecules by the cells[18–21,24]. To achieve enhanced electrical current generation in MFCs, artificial electron mediators such as potassium ferricyanide (FeCN), and various quinones or phenazines derivatives have been added[10,25–28]. Eukaryote yeast cells have also been electronically coupled to electrodes using Methylene Blue as the redox-active mediator[29]. MFCs typically require a

constant flow of solutions of organic molecules of specific composition optimized for the organisms long-term survival and electron transfer activity.

A different approach for electrical power generation is based on Photosynthtic organisms. Intact photosynthetic microorganisms, organelles such as chloroplasts[30], thylakoid membranes[31] or isolated photosynthetic complexes such as Photosystem II (PSII)[32–34] or Photosystem I (PSI)[34–40] have been coupled with electrodes in different bio-photo electrochemical cells configurations. Upon illumination, these photosynthetic components are able to convert absorbed light energy into electrical current collected by the anode. Similar to some MET type MFCs, the electron transfer from these photosynthetic components to the anode may be enabled or enhanced by the addition of an exogenous mediators such as FeCN or quinones or via tightly bound redox polymers or nanoparticles. Although isolated photosystems can theoretically provide a high concentration of photochemical reaction centers (and thus high current densities), their isolation is more complicated and expensive than the use of isolated photosynthetic membranes or intact organisms and therefore may be less attractive for practical applications.

While the harvesting of direct electron transfer is an attractive alternative energy source, an energy-rich compound such as H_2 has advantages as it can be stored and used for many existing technologies. Indeed, recent research has shown the potential of bio-photo electrochemical cells which are based on photosynthetic components to produce molecular H_2[34,35]. Hydrogen can be utilized for pollution-free electricity production in H_2/O_2 fuel cells[41,42]. H_2 generation may be conducted by utilizing platinum cathodes to catalyse the reduction of H^+ into H_2[31] or by utilizing enzymes such as hydrogenase or nitrogenase as electron acceptors[34,42–44].

BPECs based on intact, live photosynthetic microorganisms such as cyanobacteria or microalgae can transfer photoexcited electrons to the anode to generate electrical current in a fashion similar to non-photosynthetic organism based MFCs[40,45–47]. These photosynthetic microorganisms can utilize atmospheric CO_2 to synthesize their carbon source and therefore, are not dependent on a constant external source of carbon [48]. Moreover, species such as *Dunalliela salina*[49,50] or *Arthrospira platensis* (*Spirulina*)[51] are already cultivated in industrial facilities around the world for the production of food additives and cosmetics. These well-established methodologies are an excellent platform to integrate bioelectricity or hydrogen production technologies in the existing cultivation facilities. Recently it has been shown that the source of the external electron transport in cyanobacteria derives from a combination of the respiratory and photosynthetic pathways[46]. It was further shown that cyanobacterial cells in association with a BPEC anode, secrete NADPH which acts as the major electron mediator[45]. Upon illumination, more NADPH is formed by the photosynthesis pathway, leading to an enhanced secretion process which subsequently leads to higher currents. The $NADP^+$ can be uptaken by the cells and can be re-reduced by photosynthetic ferredoxin-$NADP^+$ reductase.

Furthermore, the addition of exogenous $NADP^+$ (or NAD^+) was reported to significantly enhance the photocurrent intensity and duration[45]. Using a different approach, enhanced photocurrents were gained by cyanobacterial biofilms grown directly on an anode surface[52].

We have recently shown that bio-photo electrochemical cells are not limited to microorganisms, and that macroalgae (seaweeds) can be utilized for bioelectricity production as well[53]. The finding that photocurrent can be generated from bulk marine organisms such as seaweeds led us to explore the bioelectricity potential of terrestrial plants. Here, we show that terrestrial plants can generate photocurrents in BPECs using different tissues such as leaves and stems. Furthermore, we show the ability of a succulent desert plant to generate continuous bias-free photocurrent for more than 1 day from its internal tissues without the addition of exogenous electrolyte and demonstrate electrical current generation directly from a lily pond under natural sunlight irradiation.

Results and Discussion

***Spinacia oleracea* (Spinach) produces photocurrent and hydrogen in a BPEC that originates from PSII.** In our previous studies, we describe a BPEC designed to generate an electric current directly from seaweeds via MET[53]. This BPEC was consisted of a transparent glass container, stainless steel anode clips that holds the leaf, a platinum cathode wire and an Ag/AgCl 3M NaCl as the reference electrode. As mentioned above[45,46] we have also shown that cyanobacterial cells can generate currents under light irradiation, with only minor currents in the absence of light. Unlike cyanobacteria, seaweeds generate electrical currents in both dark and light conditions[53]. We wanted to assess whether plants leaves can be connected to a BPEC under similar conditions as the seaweeds (in the presence of 0.5M NaCl as the electrolyte and an anodic bias of 0.5V). Chronoamperometric (CA) measurement were performed by directly connecting a round segment of leaf from spinach purchased at local markets with a diameter of 1 cm to the stainless-steel clip either in dark or light. The measured current in the BPEC system was allowed to stabilize in the dark (Fig. S1) which takes 5-10 min. Upon initiation of illumination, the BPEC produced a maximal photocurrent of ~ 6.5 mA / cm^2 (above the stable dark baseline) under illumination after 10 min, while no significant additional electrical currents were generated in dark (Fig. 1). CA measurements at different temperatures (20-28 ^oC) did not show any significant effect on the current density. As the response to illumination suggests that the photosynthetic apparatus is the source of the photocurrent, we wished to identify whether PSII is the source of the reducing MET molecules. CA measurements performed under light following the addition of the PSII inhibitory herbicide 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU)[54], showed efficient inhibition of light dependent MET (Fig. 1). DCMU inhibition of PSII dependent current was previously seen in plant membranes[31] and seaweeds[53], but not in BPECs containing intact cyanobacteria[46]. We predicted that addition of 0.5V bias to the BPEC would be sufficient for

hydrogen production at the cathode[46]. To measure the hydrogen generation, the top of the reaction vessel was sealed with a thick layer of parafilm, and CA measurements were performed for 10 min in dark or light, with or without the addition of 100 µM DCMU. A sample of the head space of the BPEC was removed with an air-tight syringe and the amount of hydrogen was quantified by gas chromatography analysis. We found that the system evolved 4.19 µmol (+/- 0.53, n=3) hydrogen after irradiation without DCMU. No hydrogen was detected after CA measurements in dark or in the presence of DCMU. See Methods for additional details on the hydrogen evolution measurements. To evaluate the effective distance in the leaf that influence the photocurrent production under irradiation, a piece of spinach leaf was cut into different sizes of with a fixed width of 0.2 cm (the width of the anode) and a length of 0.2 – 0.5 cm. The smallest piece had the same area of the anode and was thus not exposed to light. The light exposed area of the other pieces was 0.04, 0.08 and 0.12 cm^2. (Fig. S2 a and b). The maximal effective distance in the leaf that can contributes to photocurrent production was evaluated to be 0.1 cm. (Fig. S2c).

These experiments show the usefulness of using intact leaves as this allows the use of elevated ionic strength buffers as the electrolyte solution. The elevation of the ionic strength has shown to promote orders of magnitude increases in current densities in microbial fuel cells [55,56] and in photosynthetic organism based BPECs[53,57].

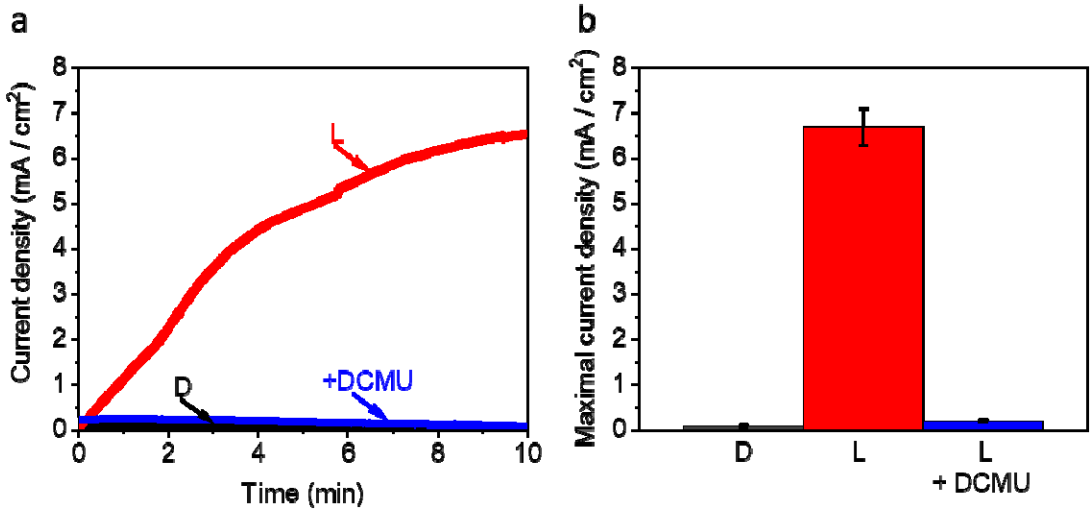

Fig. 1. Spinach produces photocurrent in a BPEC that originates from PSII. a. CA measured using a leaf disk. A maximal photocurrent of 6.5 mA / cm^2 was obtained when the leaf disk was illuminated with white light (red). No significant current was obtained in either dark (D, black) or in the presence of DCMU (blue). The onset of the light irradiation was at t = 0, however ~10 minutes of pre-incubation are necessary for system stabilization (Fig. S1) **b.** Maximal current production in the CA of spinach leaves in dark (D, black), light (L, red), and light + DCMU (blue). The error bars represent the standard deviation over 3 independent measurements.

Photocurrent production of different plants. The obtained results led us to explore if leaves of other plants can be coupled in a BPECs configuration and generate photocurrents. In addition, do

other photosynthetic plant tissues such as stems can generate photocurrent? To elucidate these questions, CA measurements were performed using leaves of various plants from different native habitats. We explored *Salvia officinalis* (Sage), *Pinus* trees (Pine), *Cistaceae* (Cistus), *Salvia Rosmarinus* (Rosemary), *Vitis* (Grapevine), *Bryophyta* (Moss), stems of *Rosa* (Rose) and the *Cacti* plant *Opuntia Ficus-indica*. Our results show that upon illumination, most leaves are able to generate an electric current in the BPEC described above (Fig 2). These include leaves from trees, bushes and the miniature leaf like structure of a moss. Planar leaves with soft textures produced ~ 6 – 9 mA / cm^2 while photosynthetic tissue (stems or leaves) with hard textures produced lower currents of ~ 1– 2.5 mA / cm^2. No significant photocurrent could be obtained from intact *Cacti* plant *Opuntia ficus-indica* phylloclade (that consists of short photosynthetically competent stems). However, removal of its external rough layer enabled the harvesting of a photocurrent of ~ 11 mA / cm^2. A photo of *Opuntia ficus-indica* in a BPEC configuration before and after removal of its external layer is shown in Fig. S3. The results show that the ability of photosynthetic leaves and stems to produce photocurrent is dependent on the toughness of their external cuticle. Based on these results, we suggest that plant leaves that are more permeable (lack a cuticle) enable a higher flux of reducing molecules to exit the cells.

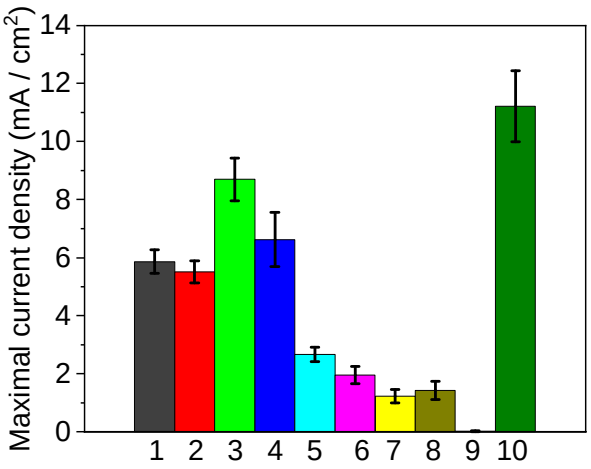

Fig. 2. Photocurrent production of different plants. CA of leaves and the stem of ten plants were determined. Maximal current production of Sage (1), Origanum (2), Moss (3), Cistus (4), Pine (5), Grapevine (6), Rose stem (7) Banana (8), *Opuntia ficus-indica* (9) and *Opuntia ficus-indica* after removal of its cuticle (10). The error bars represent the standard deviation over 3 independent measurements.

Greener leaves produce more photocurrent. In addition to outer surface permeability, we expected that the level of produced current in the BPEC would be proportional to the relative concentration of photosynthetic complexes in the leaf which might change due to leaf growth. In order to ascertain the correlation between leaf content and current, we examined *Cercis siliquastrum* leaves of different

shades of green, indicating different amount of chlorophyll (Chl). The leaves that were chosen were of similar size and location in the tree, but had different intensities of green, perhaps due to a bleaching/senescence process. We performed simultaneous CA and dissolved oxygen (DO) measurements on 1 cm round segments of the different leaves (Fig. 3), followed by measurement of the Chl concentration (see Methods section for details). The results show good correlation between the harvested photocurrents, the Chl concentration, and the DO values (which indicate the magnitude of photosynthetic activity from the entire illuminated leaf area). The chlorophyl *a/b* ratio in all three leaf types was 3.5, indicating that the pigment intensity is related to the concentration of photosynthetic units and not to changes in the composition of the complexes (with respect to the ratio of reaction centres to antenna proteins) These results support our initial hypothesis that the photosynthesis derived electrons are the source of current generation in the BPEC.

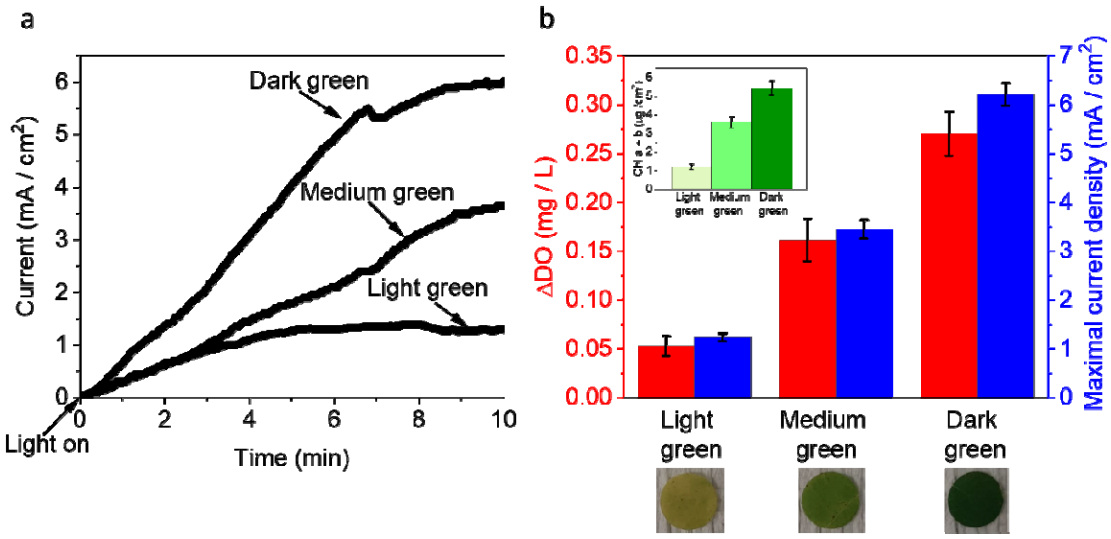

Fig. 3. Greener leaves produce more photocurrent. CA, dissolved oxygen (DO) and total chlorophyll (Chl) determination were conducted for *Cercis siliquastrum* leaves with different levels of greenness. **a.** Representative CA and DO measurements of the dark green, medium green, and light green leaves. DO values are indicated above each curve. **b.** Maximal current and DO values of the low, medium and dark green leaves. Blue bars indicate the maximal current density (right scale). Red bars indicate the DO values (left scale). Chl *a* + *b* concentrations are shown in the inset. The error bars represent the standard deviation over 3 independent measurements.

Bias-free photocurrent generation using water-preserving plants. Many plant families have members that can accumulate significant amount of water in their leaves and are colloquially named succulents. We postulated that the elevated content of internal solution reservoirs (a water-based gel) can be utilized as an electrolyte in the BPEC. Unlike the BPEC configuration described above, in this type of plant we measured the current in a bias-free configuration, using a two-electrode mode in either white-light illuminated or in the dark. An iron nail anode and a platinum wire cathode were

inserted into a live leaf of *Corpuscularia lehmannii* (CL), without disconnecting the leaf from the plant (Fig. 4). Maximal currents of 0.001 and 0.0045 mA / cm^2 were obtained after 24 h under dark and light conditions, respectively (Fig. 4). As with our previous study of macroalgae, we suggest that the source of the dark current is reduced molecules (NADPH/NADH) exported from the mitochondria and released from the leaf to the BPEC. Injection of DCMU into the internal leaf solution (500 μM, final concentration) completely inhibited current production (Fig. S4), indicating that PSII is the source of the electrons.

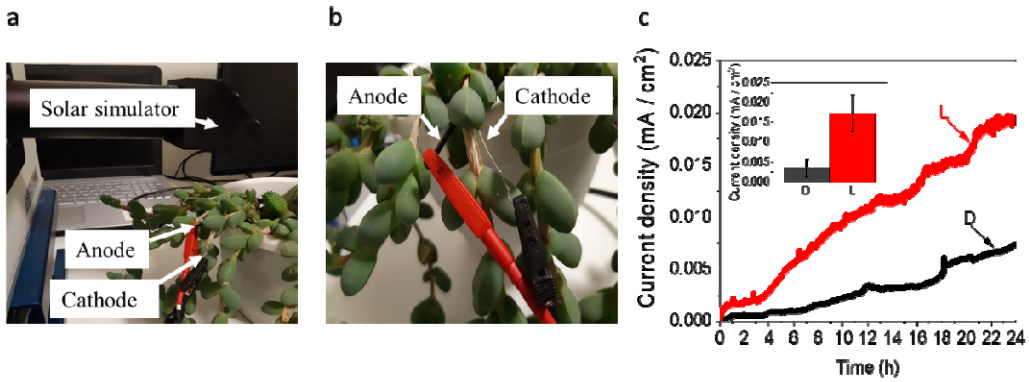

Fig. 4. Liquid containing leaves produce bias-free current without the addition of exogenous electrolyte. Bias-free CA measurements were measured for 24 h in dark and light. The iron nail anode and platinum wire cathode were inserted into the leaf using the internal solution of the leaf as the electrolyte. **a.** A picture of the *Corpuscularia lehmannii* (CL) BPEC setup. **b.** enlargement of the anode and cathode that are inserted into the CL leaf. white arrows point at the anode, cathode, and the solar simulator. **c.** Representative CA measurements in dark (black) and light (red). The insert shows the maximal current production in dark (black) and light (red). The error bars represent the standard deviation over 3 independent measurements.

CA measurements were conducted for 10 min under dark or light irradiation while 0.5 V was applied. (Fig. 5b). The headspace of each measurement was analysed, using gas chromatograph with a thermal conductivity detector (GC-TCD, Agilent 8860) (Fig. 5c).

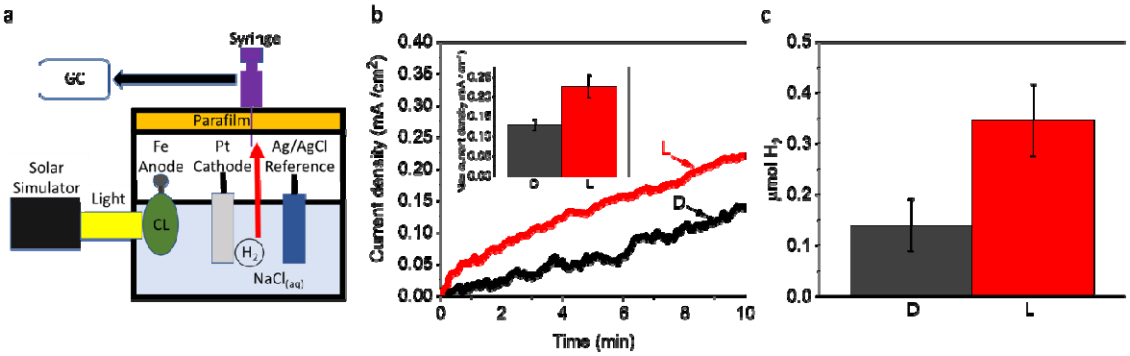

Fig. 5. H_2 gas production from *Corpuscularia lehmannii*. a. A schematic drawing of the BPEC. An iron nail anode is inserted into a CL leaf. The leaf, a platinum cathode, and an Ag/AgCl refence electrode are dipped inside the BPECs 0.5 M NaCl solution. The top of the glass vessel is sealed with a thick layer of parafilm. A solar simulator illuminates the CL leaf

with the intensity of 1 Sun. Upon illumination, H_2 is formed at the cathode and is released to the space above the solution. At the end of the CA measurement the H_2 gas is removed by a syringe and quantitated by GC-TCD analysis. **b.** CA measurements in dark (D, black) and light (L, red). The insert shows the maximal measured currents in dark and light. **c** GC measurements of H_2 concentration. In all panels, the error bars represent the standard deviation over 3 independent measurements.

H_2 gas production using *Corpuscularia lehmannii.* We next examined whether CL can be used for H_2 gas generation. To achieve that, we designed a modified BPEC. For these measurements, a rectangular glass vessel with an iron nail anode was inserted into the leaf of the CL which was held in an 0.5 M NaCl aqueous solution. H_2 generation by the BPEC requires an applied bias and proton reduction catalyst. Therefore, the anode was coupled with Ag/AgCl reference electrode and a platinum cathode. The glass vessel was sealed to allow H_2 accumulation, (Fig. 5a).

Towards applicative renewable energy technologies. In order to produce electricity from the leaves of trees or bushes in applicative technologies, the leaves must be harvested, transported, and inserted into an aqueous solution. Unlike terrestrial plants, the native habitat of water plants can serve as an electrolyte, therefore may enable direct electricity generation without harming the plants. In order to demonstrate the potential of utilization of water plant based BPECs we examined the ability of Water Lilies (*Nymphaeaceae*) in their native pond to generate photocurrents under the sunlight. CA measurements were conducted in the pond under the naturally changing sunlight intensity using an applied bias of 0.5 V. A stainless-steel anode clip was dipped in the pool held one of the leaves in the pond. A platinum wire cathode and an Ag/AgCl KCl 3M were held by a sponge, to float on the pond surface (Fig. 6a, b). Sunlight intensities of 200 – 900 μE / m^2 /s and water temperatures of 13 -17 ^{0}C were measured during the duration of the CA measurement (Fig. S5). Current densities of 6.5 mA /cm^2 was obtained in this configuration. If the anode is not associated with a leaf only a small current (~0.5mA/cm^2) was obtained, most likely originating from reducing molecules that exist in the pond water. The pond water is of course a highly complex mixture of tap water, organic molecules, and a variety of insects and animals that inhabit the immediate environment.

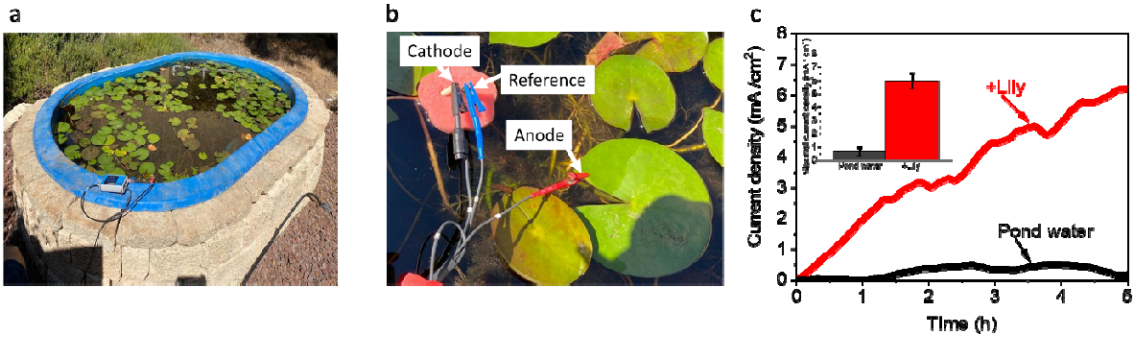

Fig. 6. Towards applicative renewable energy technologies. CA of a Lily leaf was measured directly from its native growth environment using the water in the Lily pond as the electrolyte of the BPEC. **a.** Photo of the lily pond with the measurement setup. **b.** An enlargement of the active area of the pond where the CA measurements were conducted. A

stainless-steel clip anode grabs the Lily leaf. The cathode and reference electrodes are held by a sponge that floats on the water surface. **c** CA measurements in the Lily pond water connected (red) or floating in the pond water (black) between the anode and the leaf. The insert displays the maximal obtained currents over a 5 hr measurement. The error bars represent the standard deviation over 3 independent measurements.

Models for electron transport in plant-based MET based BPECs. Based on the results we present here, we propose a mechanism that explains the generation of photocurrents from intact leaves (Fig. 7a), or liquid-containing leaves based BPECs whose inner solution content is utilized as an electrolyte (Fig. 7b). For the exogenous electrolyte configuration (Fig. 7a), we suggest that the main source of electrons originates from the natural photosynthetic linear electron pathway from water to NADPH. NADPH can then diffuse out the chloroplasts and the cell membrane to reduce the anode. The external electron transport occurs only under illumination and is inhibited by the addition of DCMU. In the case of plants with hard cuticles, electron transfer mediators are not freely released to the anode. Therefore, negligible currents are generated to the external anode or electrolyte solution. This can be overcome by inserting the electrodes directly into the gel-like interior of the leaf. Various electron transfer molecules (including NADPH) in the native electrolyte are the source of current generation.

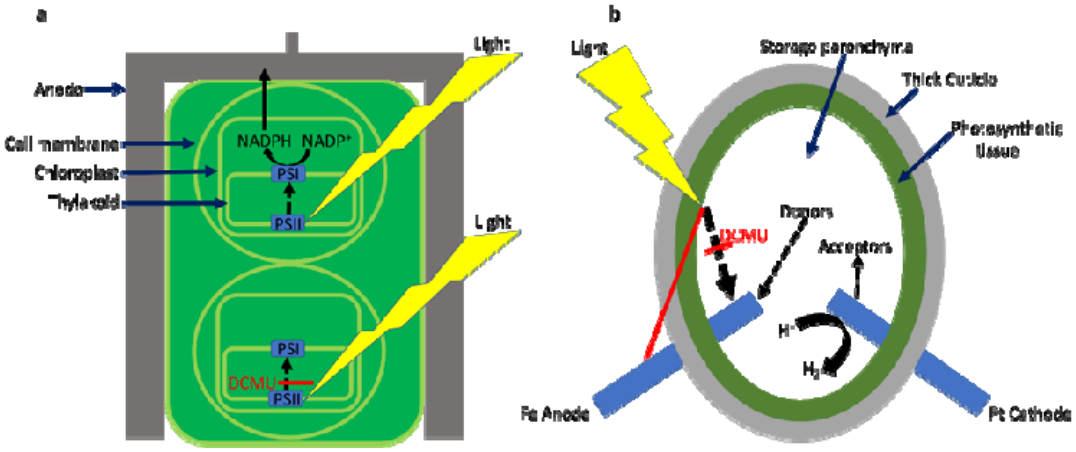

Fig. 7. Models for the electron transport mechanisms in plants-based BPECs. Schematic models for the suggested electron transfer mechanisms in BPECs which utilize flat-shaped leaves with exogenous electrolytes or Succulents with endogenous electrolytes. **a.** Flat leaves BPEC. The electron source is PSII which conducts water splitting under sunlight illumination. The electrons are being transferred by the multistep reduction reactions of the photosynthetic pathway through PSI to reduce $NADP^+$ to NADPH. Under association with the electrochemical component of the BPEC, NADPH can exit the Chloroplast and the outer cell membrane to reduce the anode and produce current. The addition of DCMU blocks the electron transport from PSII and thus abrogates photocurrent generation. Yellow stripes indicate the light illumination, dark blue arrows mark the anode, cell membrane chloroplast, and thylakoid. Dashed arrows indicate electron transport. Whole black arrows indicate the trafficking of NADPH. A red line indicates the inhibition of the electron transport by DCMU. **b.** Succulents-based BPECs. The thick Cuticle of the Succulent leaf prevents external electron transport and therefore disables the possibility for current production by an external connection of the leaf with the anode. In order to produce current, the anode and the cathode are inserted into the leaf utilizing its internal solution as an endogenous electrolyte. The electron

source for the current production mostly originates from the photosynthetic pathway but may also result from electron donors that exist in the internal solution. The terminal acceptor may be H^+ ions that are being reduced by the platinum cathode to form H_2 or other electron acceptors that exist in the electrolyte. Addition of DCMU significantly eliminates the photocurrent production. a yellow stripe indicates the light illumination. Blue arrows mark the thick Cuticle, photosynthetic tissue, and storage parenchyma. Blue rectangular shapes mark the anode and the cathode. Black dashed arrows indicate electron transport. red lines indicates the blockage of electron transport.

Conclusions

In this study, we show for the first time the ability of intact terrestrial plants to generate electrical currents in BPEC configurations. We further show that while leaves with softer texture can generate higher photocurrents, currents can be generated from other photosynthetic tissues of almost any species. Our results show that the electron source for the generated photocurrents derive from PSII as it is DCMU sensitive. We further show that a plant that can thrive in desert environments such as CL can produce bias-free photocurrent without the addition of an external electrolyte. Finally, we demonstrate the ability of water Lily ponds to generate photocurrents in their native habitat without the need to harvest them. As there is no need to process the leaves for generating photocurrents and the fact that almost any kind of leaves may be used, the economic cost of using plants as an electron source should be very low. Moreover, it may be integrated with crops that are grown for biodiesel production. Therefore, the ability of plants to directly generate electricity in BPECs without any processing holds great promise and potential for designing a “greener” future for energy technologies.

Experimental

Materials. All chemicals were purchased from Merck. ***Spinacia oleracea*** leaves were purchased at local markets in Haifa. *Corpuscularia lehmannii* (CL) was purchased at a local nursery. All other leaves were harvested directly from trees or bushes in Haifa.

Indoor CA measurements of flat leaves. The indoor CA measurements of flat leaves were done in the same setup which was described in our previous work[53]. The measurements of all leaves were done in a small rectangular transparent glass vessel with dimensions of 4.5 cm^3. A solar simulator (Abet, AM1.5G) was placed horizontally to illuminate the leaves with a solar intensity of 1 Sun (1000 W/m^2). Determination of the light intensity at the surface of the leaves was done as a function of distance from the light source in an empty vessel neglecting small intensity losses caused by the glass and ~ 0.5 cm of the electrolyte solution. The measurements were conducted in 3 electrode mode (unless otherwise mentioned) using the stainless-steel clip as anode, a platinum wire as a cathode, and Ag/AgCl 3M NaCl as a reference electrode (RE-1B, CH Instruments, USA) with an applied electric

potential bias of 0.5 V on the anode in 0.5 M NaCl solution. In all measurements, the current density was calculated based on the contact area between the anode and the leaves of 0.08 cm^2. When applied, the addition of DCMU was done prior to the measurements (5 min). The BPEC system was allowed to achieve a stable baseline prior to beginning the measurements (in the dark) which usually took about 10 min (Fig. S1). Values described in the Results section were measured using the stabilized current density as 0 mA/cm^2.

Hydrogen production and quantification. The BPEC was covered by a thick parafilm layer. The volume of the electrolyte solution was 50 mL and the headspace volume was 40 mL. Following 10 min of CA measurements, 1 mL of air was removed from the top of the reaction vessel and injected into vials (1.8 mL). 50 µL samples were injected into a gas chromatograph system coupled with a thermal conductivity detector (GC-TCD, Agilent 8860) with a 5-Å column (Agilent, 25m x 0.25mm x 30µm). Hydrogen that evolved during the BPEC stabilization stage (see previous section) were subtracted from the values of hydrogen obtained during the actual experiment.

CA measurements of CL leaf. The CA was measured in two electrodes mode without application of potential bias, using an iron nail as the anode and a platinum wire as a cathode. The two electrodes were inserted into internal liquid in the CL leaf. The surface area of the iron nail which was inside the leaf was 0.5 cm^2. Solar illumination of 1 Sun was applied on the top of the leaf. Addition of DCMU was done by a direct injection of 10 µl of 100 mM DCMU in ethanol into the internal volume of the leaf (~ 2 cm^3) to obtain a final concentration of ~ 500 µM.

Direct CA measurements from the water lily pond. CA measurements were done directly from the pools using a stainless-steel clip anode, a Pt wire as a cathode, and Ag/AgCl 3M NaCl as a reference electrode. The anode clip was grabbing a waterlily leaf. The anode and reference electrode were inserted into a sponge that was floating on the pool surface. An applied electric potential of 0.5 V on the anode under the sunlight. Light intensity was measured at the water surface height of the pond with an app-based portable light meter (name of app). The temperature was monitored manually during the duration of the measurement.

Dissolved oxygen measurements. DO measurements were conducted in the same system of the CA measurements with the addition of a small magnetic stirrer bar. The top of the reaction vessel was tightly covered with a thick layer of parafilm. A DO meter probe (Hanna Instruments, HI-5421 research grade DO and BOD bench meter) was inserted into the BPEC liquid phase (50 ml) to quantify the accumulated DO concentration after 10 min.

Chlorophyll concentration determination. Chlorophyll determination was done by grinding of the leaves followed by acetone extraction followed by absorption measurements as previously described[58,59].

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Conflicts of interest

There are no conflicts to declare.

Author contributions

YS and NA conceived the idea. YS, OY and NA designed the experiments. YS performed the main experiments. MM assisted in performing parts of different experiments. YS, OY, GS and NA wrote the paper. NA and GS supervise the entire research project and provide funding.