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Microbial Fuel Cells: Mitochondria aren't the Powerhouse of this Cell

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Professor Martin

I. Introduction

For many decades, scientists have been searching for new sources of renewable energy that can be harnessed to power our world. But one source of energy has been under our noses, or rather feet, this whole time (and it's not fossil fuels). It's DIRT! Or rather the microbial ecosystems living in the soil and marine sediments that cover our planet. How does one go about harnessing this energy? Unfortunately, it's not quite as easy as sticking a lightbulb into mud, but that's not as far off as one might think.

The natural ability of some microbes (called exoelectrogens) to release electrons as part of their metabolism can be harnessed to create an electron potential in a device called a microbial fuel cell (MFC). By placing suitable electrodes into a media that contains exoelectrogens, a small current can be generated. This happens because the microbes associate with the anode (often forming a biofilm on the material) and transfer electrons to the anode material creating a negative charge on the anode and a relative positive charge on the cathode which drives a current across these two electrodes. This energy can then be used to power an electrical device.[1-12]

This paper provides an overview of the current designs, capabilities, and uses of MFCs. The major focus throughout this paper will be the specific microbes that appear in MFCs and how their unique abilities in addition to being exoelectrogens make them ideal for use in an MFC. Initially, the innerworkings and theory behind MFCs is discussed. This is followed by an in-depth look at the diversity of exoelectrogens found in MFCs. The final portion of this paper will focus on the limitations and future uses of MFCs which could include being used to offset the cost of wastewater treatment, power small environmental monitoring equipment in remote areas, light parks, and power a variety of other small devices.

II. Making a Microbial Fuel Cell

A large and diverse group of MFC designs have been created over the past few decades in an attempt to find the most efficient and cost effective design for an MFC.[1] Some of the most common types of MFCs are sediment MFCs (SMFCs),[2,3] single chamber air cathode MFCs,[4,5] singe chamber anaerobic MFCs,[6] two chamber (H-type) MFCs,[1,3,7,8] salt bridge MFCs,[1] photoheterotrophic type MFCs,[1] and (wetland) plant MFCs (PMFCs).[9,15-18] Even though they may appear quite different from one another externally as demonstrated in Figures 1 and 2, all MFCs have several elements in common. The basic design of every MFC includes at least one chamber to hold the microbial community that produces the electrons, the substrate that the microbes utilize for their metabolic processes, an anode to accept the electrons from the microbes, and a cathode for the electrons to flow to thereby create a current.[1-12]

In two chamber MFCs, one chamber houses the anode and its associated exoelectrogens. This chamber is often anaerobic and may or may not contain chemicals to help facilitate the transfer of electrons from the microbes to the anode. The second chamber contains the cathode and is often aerobic so that the electrons moving to the cathode from the other chamber can be used to reduce oxygen to water. Often, the two chambers are separated physically by a membrane permeable to protons and impermeable to oxygen called a proton exchange membrane (PEM).[5]

One chamber MFCs have a similar design, but the PEM is often removed so the anode and its associated biofilm of exoelectrogens are no longer under completely anaerobic conditions due to the fact that the cathode must be exposed to air. While the presence of oxygen could have negative effects on anaerobic exoelectrogens, an aerobic biofilm will often form above the anode (associated with the cathode) and prevent the diffusion of oxygen into the rest of the MFC.

Given that single chamber MFCs have a simpler design, require less material, have a smaller volume, and have been shown to be more efficient, they are one of the most common MFCs used.[5]

The internal conditions of the MFC can vary greatly with respect to pH, temperature, the source of nutrients or media, the concentration of nutrients in the media, and the presence or lack of oxygen. A majority of MFCs are populated with microbial communities gathered from soil and marine sediments, so the internal temperature and pH of the MFC is often maintained in a range similar to the native conditions for those microbes.[1-12] The nutrients utilized in MFCs range from simple media like acetate,[5,7,12] butyrate,[5] glucose,[7,12] cysteine,[7,12] bovine serum albumin,[12] and ethanol[12] to complex mixtures of organic matter including marine and river sediment;[7] seawater;[7] urine;[8] plant root material;[10, 15-18] and domestic,[4,7] animal,[12] food-processing,[7,12] and meat-packing[12] wastewaters. As will be explained later, many exoelectrogens are anaerobes or facultative anaerobes that function best to produce an electron potential when oxygen is not present, so the microbes are often separated from sources of oxygen by keeping them submerged in water, having the cathode spatially and sometimes physically separated (with a barrier or in another compartment altogether) from the anode and microbes, or adding other microbes that remove oxygen from the MFC.[5]

There are a variety of anode and cathode options for MFCs.[1,6-7] The most common material used for either electrode is graphite.[3,4,6,10] However, these types of electrodes can take many forms including carbon (graphite) felt,[2,4,7] carbon cloth,[4] or graphite rods.[3,4] These materials can be further improved with the addition of coatings that increase their conductivity (by several orders of magnitude in some cases) or the ability of microbes to adherer to the material like carbon felt with polyaniline,[4,7] carbon-coated Berl saddles,[7] and graphite bound to electron mediators like Mn^{4+} (for anodes) or Fe^{3+} (for cathodes).[6] Schriliò et al. found that polyaniline coated carbon felt electrodes resulted in approximately 6 times more power production than uncoated carbon felt.[7] The microscopic difference between carbon felt, carbon felt with polyaniline, and carbon-coated Berl saddles can be seen in Figure 3. Looking at this figure, it would not be unreasonable to think that the type of microbe associated with or biofilm formed by a microbe on the anode might be affected by the morphology of the material; however, Schriliò et al. only observed planktonic microbes, so they offer no indication as to what these effects may be.[7] However, they did find that there was no significant difference between the planktonic microbial comminutes in MFCs with either of the three electrode materials.[7] Another advantage of graphite felts and cloths is that they are much less expensive than graphite rods, so MFCs designed with graphite felts and cloths are more desirable and marketable than those built with solid graphite electrodes.[4]

A wide variety of microbes can be used in MFCs. Common choices include members from the bacterial phyla *Proteobacteria*, *Firmicutes*, and *Acidobacteria* and the fungi (a yeast) *Pichia anomala*.[11] Some of the bacteria from the phylum Proteobacteria are the *α-Proteobacteria* (*Rhodopseudomonas palustris* DX-1, *Ochrobactrum anthropi* YZ-1, and *Acidiphilium* sp. 3.2Sup5), *β-Proteobacteria* (*Rhodoferax ferrireducens* and *Alcaligenes faecalis*), *γ-Proteobacteria* (*Escherichia coli, Shewanella putrefaciens, Shewanella oneidensis* DsP10, *Shewanella oneidensis* MR-1, *Proteus vulgaris, Aeromonas hydrophila, Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* L17), and *δ-Proteobacteria* (*Desulfuromonas acetoxidans, Desulfobulbus propionicus, Desulfovibrio desulfuricans,* and *Geopsychrobacter electrodiphilus*) and specifically *Geobacter* (*Geobacter sulfurreducens* and *Geobacter metallireducens*).[1-12] Some of the *Firmicutes* are *Brevibacillus agri, Clostridium butyricum,* *Enterococcus faecium*, and *Thermincola* sp. strain JR.[1-12] The *Acidobacteria* is *Geothrix fermentans*.[11] Each of these bacteria (even little *E. coli*) have some unique characteristics as exoelectrogens that make them viable for use in an MFC. Some of these bacteria like *Alcaligenes faecalis, Enterococcus faecium,* and *Pseudomonas aeruginosa* produce electron shuttles which aid in the transfer of electrons from the bacterial cells to the anode.[5] Other bacteria like *Geobacter sulfurreducens, Geobacter metallireducens, Shewanella putrefaciens, Clostridium butyricum, Rhodoferax ferrireducens,* and *Aeromonas hydrophila* are metal reducers that live their lives searching for metals to reduce with their excess electrons.[5]

Some noteworthy microbes that have been used in MFCs are *Shewanella putrefaciens* who's outer membrane is covered with iron-containing cytochromes that aid in the transfer of electrons gained from metabolizing lactate to the anode (woven graphite)[6] and *Pseudomonas aeruginosa* who's pili act as conductive nanowires to transfer electrons from the cell to the anode a very long distance away from the cell (from the perspective of the cell).[8,12] The myriad ways that other bacteria generate electricity will be discussed in the next section.

One more thing to note about the bacterial communities found in MFCs is that they are very rarely composed of a single species. Just as many bacteria will not grow well alone on a plate, exoelectrogens will often not perform at peak efficiency when they are cultured in an MFC alone.[12] Figures 4 and 5 show the relative numbers of various bacterial phyla in different MFCs under different conditions. As can be seen, the microbial communities are extremely diverse and complex containing both exoelectrogens and nonexoelectrogens.[2,9] Often, MFCs are cultured from samples taken directly from the environment, and exoelectrogens are selected for but often are not the only microbes present in the MFC.[1-12] While it is true that there is often a dominant phyla for a given set of conditions (*δ-Proteobacteria* in SMFCs), similar MFCs built under similar circumstances can look vastly different (river sediment MFCs inoculated with river water select for *β-Proteobacteria* while those inoculated with a glucose–glutamic acid mixture select for *α-Proteobacteria*).[12]

III. Generation of Electricity in MFCs

Anyone who has ever taken a biology course, has likely heard that "the mitochondria are the powerhouse of the cell." However, only eukaryotic cells have mitochondria, so how do prokaryotes get their power? Well, in a wide variety of ways; but in the end, generating energy in a cell to power its functions come down to moving around electrons just like humans move electrons through wires to power the appliances in their homes.[13] A wide range of microbes (exoelectrogens) respire by exporting electrons from their cells and releasing them to the environment either by putting the electrons on small molecules called mediators, transferring the electrons to other microbes (often other species), or by transferring the electrons to metals like iron in their environment.[1-12] Microbes like *Pseudomonas aeruginosa* and *Shewanella oneidensis* can even make tiny "nanowires" out of modified pili to move the electrons from their cells to an electron acceptor in the environment.[8,12-14] This process of transferring electrons is what is used to create power in a microbial fuel cell (MFC).[1-12,15-18]

In their natural habitats, many of the exoelectrogens used in MFCs live in environments with low levels of or no oxygen.[1-12] Because of this, they have to respire using another electron acceptor other than oxygen (the choice of eukaryotes and some other bacteria and archaea for their metabolism), or they must find a way to get their electron to a source of oxygen.[13] The electron acceptors used by exoelectrogens can be organic or inorganic ranging from simple molecules like those found in urine[8] to particles of iron (Fe^{3+}).[13,14] In the end, a microbe will utilize just about any material that can be reduced as an electron acceptor at the end of its metabolic pathway.

This ability of transferring electrons out of the cell and onto an external electron acceptor in the environment is exploited in MFCs to drive a current as the microbes metabolize whatever source of nutrients have been added to the MFC and use the anode of the MFC as their terminal electron acceptor instead of an organic compound or metal like they naturally would.[1-12] After the electron reaches the anode, it is drawn to the positively charged cathode where free protons and oxygen are waiting to join them to produce water; and as the electrons flow from the cathode to anode, this produces a current that can be harvested to run electrical devices.[1-12] Some examples of how this process is carried out can be seen visually in Figures 6 and 7.

As stated above, exoelectrogens have a variety of method for moving electrons out of their cells and onto the anode; some of the fundamental and most interesting means of carrying out this process (each of which can be seen in Figure 7) follow.

The formation of biofilms on the anode of the MFC (an example of which can be seen in Figure 8) is an easy way for microbes to directly transfer electrons from their cells to the anode.[11] Certain microbes like iron-reducing bacteria have special respiratory enzymes in their membranes that allow the bacteria to directly transfer electrons to metals like Fe^{3+} or Mn^{4+} to reduce them or artificially transfer those electrons to the anode material.[4] This ability can be enhanced when the cells are kept in close contact to the anode inside of a biofilm.[4] According to Schreeram et al., "An ideal biofilm is generated when bacteria bind tenaciously to the electrode at high densities in an open, porous structure that allows for free nutrient entry."[8] One microbe that is capable of creating such a biofilm is the facultatively anaerobic Gram-negative *γ-Proteobacteria Pseudomonas aeruginosa* and a mutant strain called *pilT*.[8] *P. aeruginosa* has been shown to have a very diverse range of metabolic pathways available for respiration, and it readily forms biofilms.[8] The *pilT* mutant is hyperpiliated and incapable of the normal twitching motion of the wild type cells.[8] As such, the cells can form very close and stable ties with one another and the anode in the biofilm in order to efficiently transfer electrons to the anode.[8] When Shreeram et al. tested *P. aeruginosa* and the *pilT* mutant in identical two chamber MFCs with a urine analogue media, they found that the *pliT* mutant produced 2.7 times more power than the wild type strain.[8] This indicates that the increased stability of the biofilm was able to improve the efficiency of the MFC.[8] However, there is another factor that must be considered in the case of the *pilT* mutant: its hyperpiliation.

Several studies have shown that the pili produced by many gram-negative bacteria are capable of conducting electricity and are likely an integral part of the process of respiration for many microorganisms.[8,12-14] These specialized pili are called nanowires and are used by a variety of exoelectrogens including *P. aeruginosa* (and the *pilT* mutant), the facultatively anaerobic Gram-negative *γ-Proteobacteria Shewanella oneidensis* MR-1 (imaged in in Figure 9), and obligately anaerobic Gram-negative *δ-Proteobacteria Geobacter sulfurreducens*.[8.12-14] In these microbes' environments, the pili serve to transfer electrons from the cell to electron acceptors like Fe^{3+} particles or sulfur compounds located at great distances (in excess of 10000 cell lengths or about 12 mm) away from the cell.[13] This process of electron transfer likely begins in the periplasm or outer membrane of the Gram-negative bacteria where the pili is anchored, and the pili acts as a conduit between the proteins located just inside and on the outside of the cell that release the electrons and the electron acceptor (be it a metal like Fe^{3+} or the anode of the MFC).[14] Nanowires likely have a great impact on the overall efficiency of MFCs by allowing a greater number of cells to interact with and move electrons onto the anode of the MFC than would be possible if only the cells in the first few layers of a biofilm on the anode were able to interact with the anode in any significant way.[8]

Another method of long distance electron transfer for exoelectrogens is through the use of mediators.[4,12,14] Again, due to the nature of the terminal electron acceptors of many exoelectrogens in their native habitats being distributed quite far from where the cells are located, the cell can produce small organic molecules that can carry an electron that they then expel in hopes of the mediator coming into contact with a terminal electron acceptor or another microbe that can get it to a terminal electron acceptor.[4,12,14] Some common mediators are phenazine produced by *P. aeruginosa*,[12] certain cytochromes like cyt *c* produced by *S. oneidensis* MR-1,[4] and anthraquinone-2,6-disulfonate (AQDS) in *G. sulfurreducens.*[14] Much like nanowires, exoelectrogens can use their naturally produced mediators to transfer electrons to the anode of the MFC increasing the effeminacy of the MFC. It is also possible to use artificial mediators like humic acids,[4] 2-hydroxy-1,4-naphthoquinone (HNQ), or thionin to induce exoelectrogenic behavior in microbes that would normally have very low potentials like *Escherichia coli* or *Proteus vulgaris*, However, these compounds are often toxic to the microbes preventing the possibility of long-term stability for these MFCs.[6]

IV. Uses for MFCs

Many plans and potential uses have been put forward for MFCs, but very few have reached fruition at this time. At the moment, the uses for MFCs is limited to powering small objects such as LEDs and environmental monitoring equipment that require low levels of power and receive very little maintenance do to being located in remote areas.[1-12] To spite the current lack of adoption by the larger electronics community of MFCs, many researchers have put forward some rather lofty goals for MFC technology in the future.

One potential use that has gained a large backing by MFC researchers is their potential use in wastewater treatment.[1,4] Specifically, MFCs could be used to both process wastewater to make it suitable for reuse for irrigation or simply safe to release into the environment while also producing electricity to run any necessary equipment in the process.[1,4] While this goal may not be achieved in the near future, MFCs do have a real potential to offset the cost of wastewater treatment. The United States alone will use an estimated $2 trillion between 2000 and 2020 to keep its water infrastructure running and expand its infrastructure to accommodate increasing demand with an annual cost as of the early 2000s of over $25 billion.[4] If just a portion of this figure could be reduced through the implementation of MFCs, it would have a major effect on the wastewater treatment industry. There are, however, some drawbacks to this plan; MFCs are not yet efficient enough to make any significant impact on the overall power usage rate during wastewater processing, and the installation of MFC technology in wastewater treatment facilities would be a very large investment.[4,7]

A more achievable goal for MFCs is to increase their energy density and use them to power mini-devices. Another goal would be to utilize MFCs to produce both power and hydrogen gas though the use of exoelectrogens that are capable of simultaneously producing an electron potential and hydrogen gas through their metabolism of glucose into hydrogen and acetate.[5]

A final and very exciting potential use for MFCs is in greenbelts, parks, and crop field to produce electricity for lighting and general power production.[9] Plant MFCs (PMFCs) are similar in design to most other marine sediment MFCs, but their microbiomes are tailored to break down plant matter released from the roots of plants.[9] PMFCs could be installed in areas with large amounts of plant biomass to both produce electricity, aid in the breakdown and recycling of plant matter, and assist with bioremediation of the area as an added bonus.[9] Their use as a source of electricity for lighting is so exciting because they would be a renewable source of energy like solar or wind power, but they would not require large aboveground surface area like solar panels and would not be reliant on the weather like solar or wild power.[9] It is also noteworthy to mention that the peak time for the use of any such lighting would be during the evening and at night when solar cells would be useless, but PMFCs would be fully active. There is currently a company in the Netherlands called Plant-e that is creating modular cells to install in parks, roundabouts, on roofs, in wetland areas, and rice paddies that make electricity with very little maintenance or upkeep required.[15-17] Plant-e's PMFCs utilize a grass that is grown hydroponically and carbon electrodes separated by a membrane that is permeable to protons to generate electricity as microbes in the water breakdown mainly glucose released from the roots of the plants (Figure 10).[18] Currently, a 50 m^2 area of modules can produce enough power to charge a cell phone, but Plant-e hopes to be able to satisfy the energy requirements of an average Dutch home with the area provided on the home's roof in a few years.[16,17] While this goal is lofty, the current trend in rapid advancements in MFC efficiency may allow for this goal to be met.

V. Limitations of MFCs

The energy output of MFCs is currently limited by several factors which must be overcome for the technology to become commercially viable. Current MFCs produce very little power and have rather low energy densities.[1-12] By some estimates, a cubic meter sized SMFC could only run one or two compound fluorescent lightbulbs.[7] Similarly, current PMFCs can only produce around 100 mA/m^2.[9] This is quite a far leap from Plant-e's goal of powering an entire household with only a couple hundred square meters of roof area. However, there are some solutions emerging that may help to improve the design of MFCs in the near future making them more efficient.

Ewing et al. found that there is not a linear relationship between the surface area of an electrode and the power generated by an MFC.[2] In fact, electrode must be made nearly 100 times larger to double the power output of an average SMFC one time.[2] One can imagine that simply making the current MFC technology larger to produce a useful amount of electricity would quickly become unwieldy if not impossible. This phenomenon of voltage reduction due to increased size happens because of the increase in internal resistance of the MFC as the electrodes are made larger.[1] Because of this inherent physical property of MFCs, large scale MFCs will likely never be feasible. However, Ewing et al. theorized that this problem could be solved by combining several smaller MFCs together to act as one large MFC (Figure 11).[2] They quickly realized, however, that simply placing several MFCs in parallel (like one might do with batteries) would not work because this would mathematically be identical to increasing the electrode size. The real solution is to combine the output of several small MFCs into one location without allowing the MFCs to electrically communicate with one another thereby keeping each MFC isolated from the others but still allowing for a substantial amount of power (65% more than equivalent MFCs in parallel) to be produced.[2] Ewing et al. also noted (as can be seen in Figure 4) that there was no significant difference in the microbial community found in the scaled-up (non-parallel) MFCs and the single-equivalent (parallel) MFCs meaning that the microbes are not significantly affected by the relative size of the electrodes, and the microbes were not responsible for the difference in power output observed.[2]

Another major drawback to using microbes to generate electricity is that you will not get out as much energy as you put in; there will always be an inherent loss of energy through the microbes' metabolisms. If the MFC utilizes a facultative anaerobe like *Pseudomonas aeruginosa*, a significant portion of the electron potential could be lost to aerobic respiration if oxygen reaches the microbes on the anode; therefore, MFC designers must do their best to limit the ability of oxygen to diffuse from the cathode to the anode by means of increasing cathode efficiency or coating the cathode with a material that will increase proton transfer and decrease oxygen transfer. The redox potential between the substrate being consumed by the microbe and the anode of the MFC must also be kept as small as possible to get the greatest voltage from the MFC; however, if the redox potential is made too small, the microbe will find another metabolic path to follow that is more favorable and will provide more energy for the microbe.[12] This means a delicate balance must be maintained between the energy being used by the microbes and the energy being harvested from the MFC in order to have it function successfully for an extended period of time. One other problem related to microbial metabolisms is the growth of methanogens (bacteria that produce methane by reducing carbon dioxide with the free protons on the cathode) that remove protons from the cathode and reduce the electron potential and power output of an MFC. However, methanogens can be controlled for controlling the pH of the MFC or by adding material to the cell that will prevent or discourage their growth and methanogenesis.[5]

Maintenance of the diversity and health of the microbial community in an MFC is also of major concern to researchers attempting to improve the efficiency of MFCs.[3,9] Holmes et al. studied the effects of predation of protozoans like *Trepomonas agilisstrain, Breviata anathema, Hexamita inflatastrain*, and *Heteromita* strain DH-1 on exoelectrogenic organisms by using *Geobacter sulfurreducens* as a model organism in a marine sediment MFC. Holmes et al. found that there was a significant negative impact on the efficiency of an MFC when protozoans were allowed to graze on the exoelectrogens in the MFC.[3] With the addition of *Heteromita* strain DH-1 to the MFC, the researchers observed a reduction in cathode biofilm thickness of nearly 20 μm which was reflected by a 91% reduction in current output of the MFC as compared to an MFC with no protozoans.[3] Clearly, predation of the exoelectrogens in an MFC is a real problem that could be significantly reducing the efficiency of many modern MFCs that could be theoretically achieving much more power output. Holmes et al. propose that MFCs that will be placed in the environment be outfitted with meshes that allow only the (on average) smaller exoelectrogenic bacteria into the cell to associate with the anode while excluding the (on average) larger eukaryotic organisms from entering the MFC and preying upon the biofilms formed by the exoelectrogens.[3] The researchers also theorize that phage activity could be a factor in the low efficiency of many SMFCs;[3] this is not unreasonable given the enormous number of viruses that are present in soil and marine habitats where MFCs would likely be deployed. However, Holmes et al. does not propose any means by which phage could be excluded from an MFC.[3] Increasing the efficiency of an MFC though its design is an ongoing task that may take several more years before MFC efficiency is increased to a point where they are deemed commercially viable.

VI. Conclusion

MFC come in a variety of shapes and sizes; but at their heart, they all have one thing in common: exoelectrogenic microbes. These microbes with the assistance of electron shuttles, their mediator molecules, membrane proteins, and nanowires have not only made themselves ideal for living in near to complete anaerobic conditions but have inadvertently made themselves perfect for use in MFCs. Though the current uses for MFCs are limited because they only produce very small amounts of power, recent advancements in anode material, the microbial community contained in the MFC, and cell design are increasing the potential usefulness and efficiency of MFCs. Future MFCs could be used to offset the cost of wastewater treatment, power small environmental monitoring equipment in remote areas, light parks, and power a variety of other electronic devices. MFCs have allowed scientist to harness a new form of energy that they have been dreaming about for decades. This source is clean, renewable, and abundant. Harnessing the power of microbes really is *almost* as easy as going into one's backyard and shoving a lightbulb into the ground.

VII. Figures

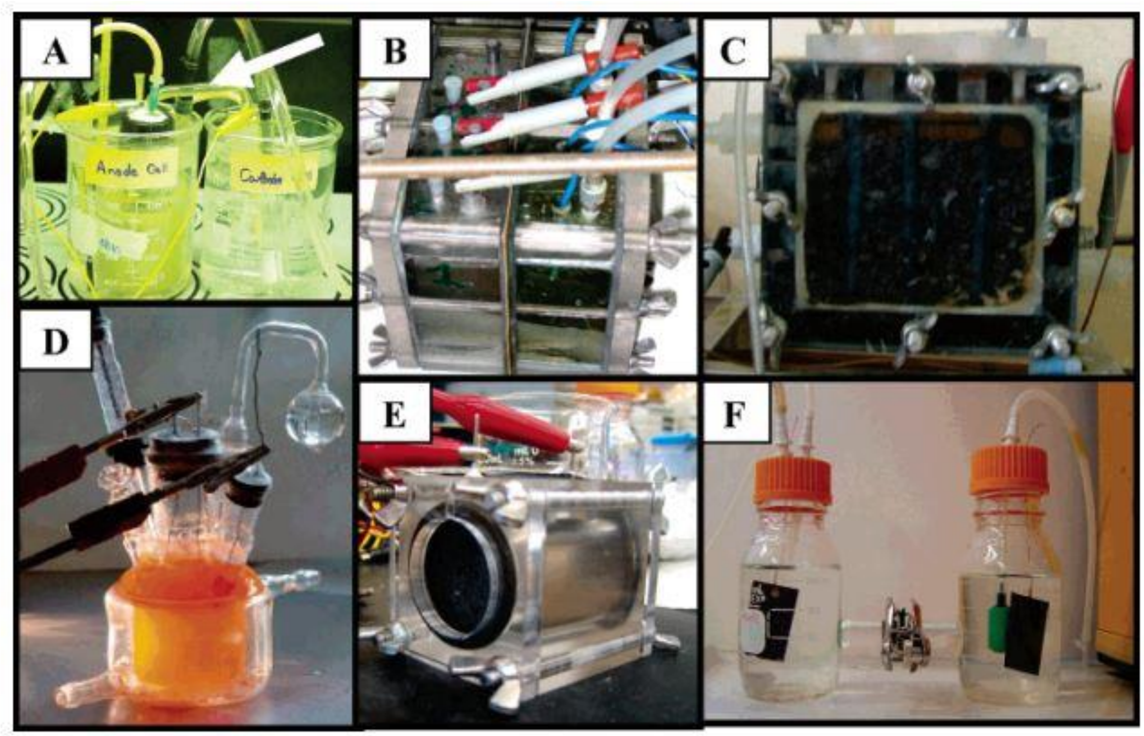

Figure 1. "Types of MFCs used in studies: (A) easily constructed system containing a salt bridge (shown by arrow); (B) four batch-type MFCs where the chambers are separated by the membrane (without a tube) and held together by bolts; (C) same as B but with a continuous flow-through anode (granular graphite matrix) and close anode-cathode placement; (D) photoheterotrophic type MFC; (E) single-chamber, air-cathode system in a simple "tube" arrangement; (F) two-chamber H-type system showing anode and cathode chambers equipped for gas sparging."[1] [citations removed]

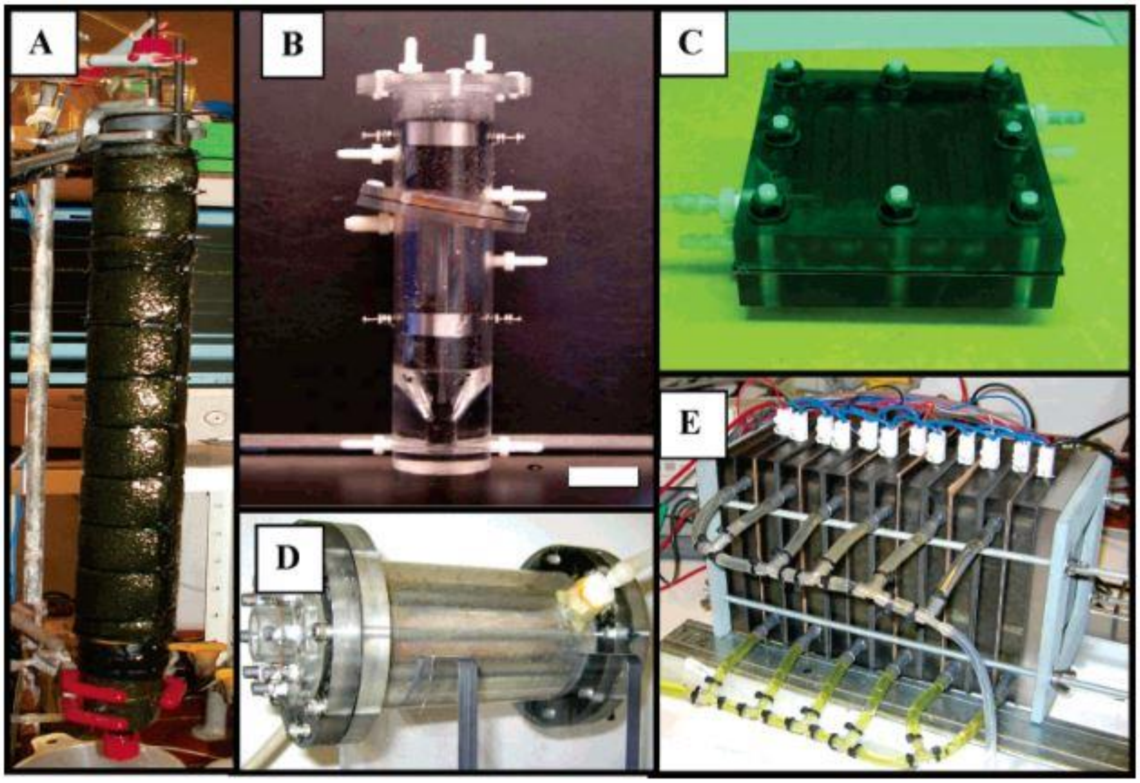

Figure 2. "MFCs used for continuous operation: (A) upflow, tubular type MFC with inner graphite bed anode and outer cathode; (B) upflow, tubular type MFC with anode below and cathode above, the membrane is inclinated; (C) flat plate design where a channel is cut in the blocks so that liquid can flow in a serpentine pattern across the electrode; (D) single-chamber system with an inner concentric air cathode surrounded by a chamber containing graphite rods as anode; (E) stacked MFC, in which 6 separate MFCs are joined in one reactor block."[1] [citations removed]

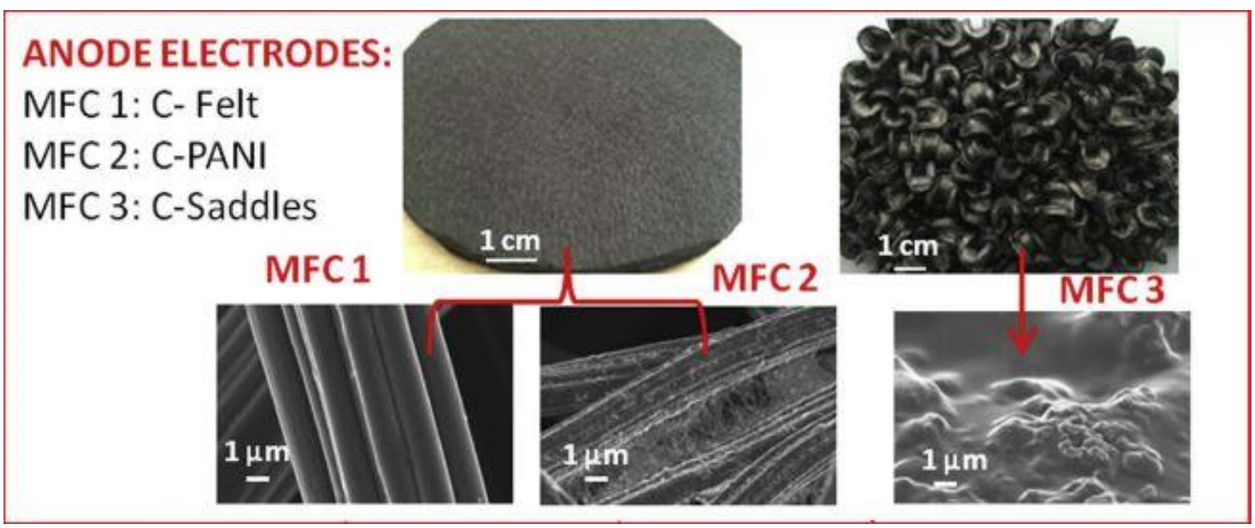

Figure 3. A macroscopic and microscopic view of carbon felt, carbon felt with polyaniline, and carbon-coated Berl saddles. Note the difference in appearance and texture.[7]

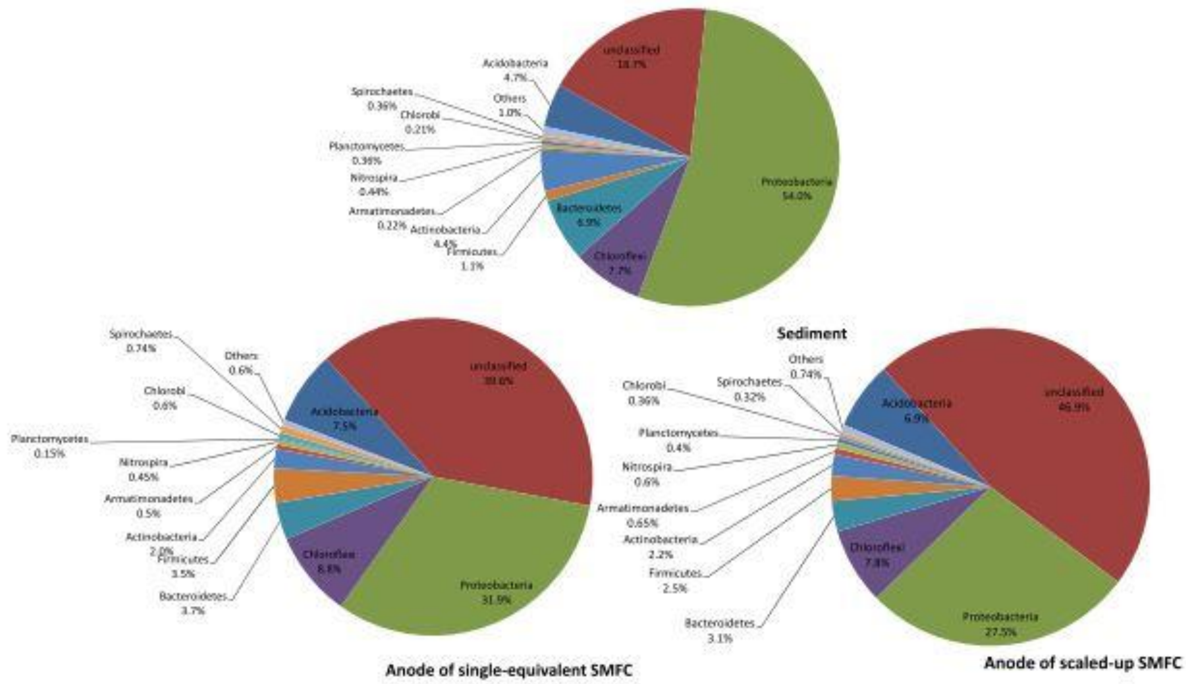

Figure 4. "Microbial community structure at the phyla level on anodes of the single-equivalent SMFC and the scaled-up SMFCs compared with the original sediment community."[2]

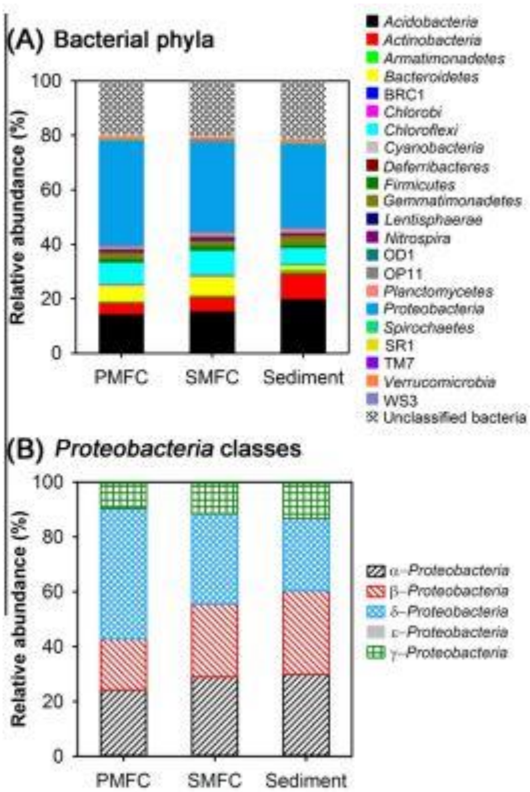

Figure 5. "Taxonomic classification of bacterial DNA sequences from communities of PMFC, SMFC control and original sediment at the (A) phylum level, and the (B) class level distribution of the most dominant phylum of Proteobacteria. The families that are less than 1% of total composition in all libraries were not included."[9]

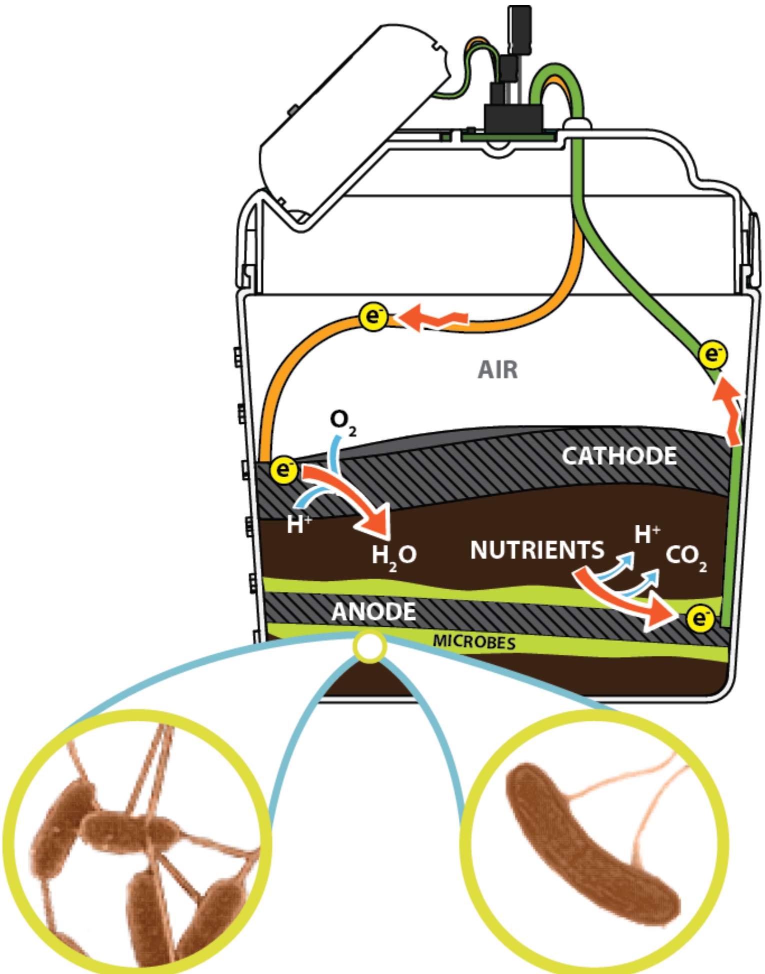

Figure 6. Schematic of a basic single-chamber air-cathode SMFC. Image from: http://www.fuelcellstore.com/mudwatt-microbial-fuel-cell-kit

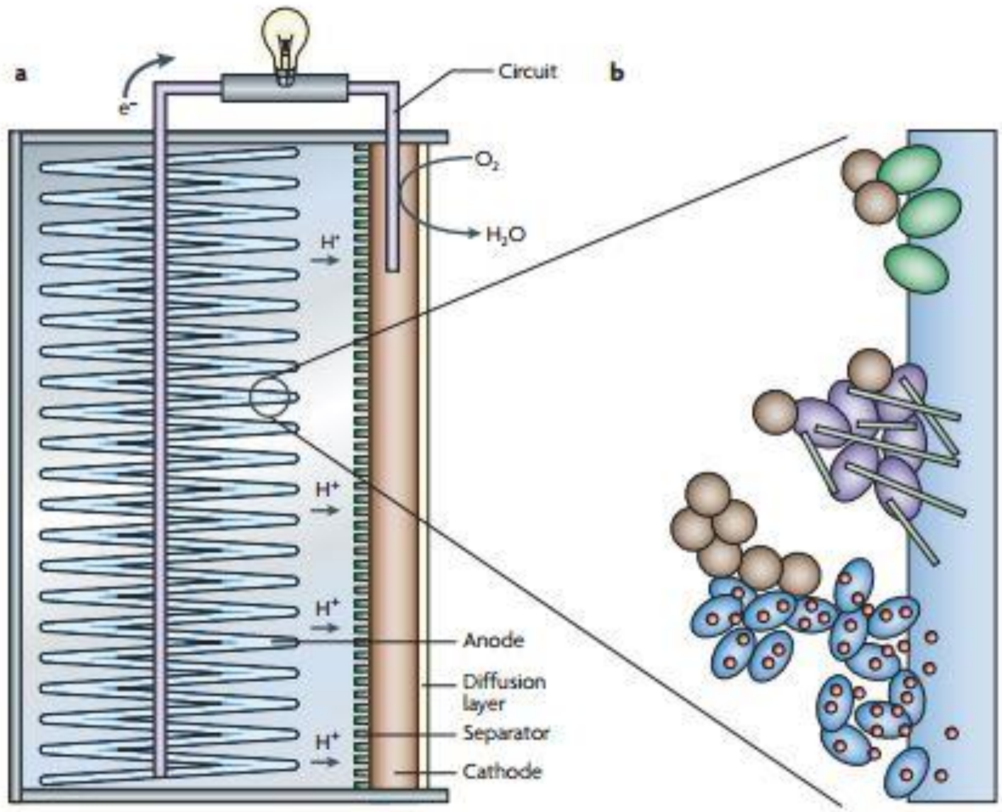

Figure 7. "Schematic of a microbial fuel cell (MFC) that contains an electrically conductive graphite fiber brush anode as the surface for bacterial growth and a flat carbon cloth cathode coated with a catalyst on the water-facing side. A diffusion layer, such as PTFE (polytetrafluoroethylene), is placed on the air-facing side to reduce water leakage, and a separator (or ion exchange membrane) is sometimes placed between the electrodes to allow charge transfer. O_2 is reduced to H_2O through a combination of electrons from the circuit and protons in the water. b| shows the different types of microorganisms in an anodic biofilm, including exoelectrogens that transfer electrons by direct contact (green), produce nanowires (purple) and use endogenous (and therefore self-produced) mediators (blue). other non-exoelectrogenic bacteria (brown) that live off the products produced by other bacteria or possibly use mediators or nanowires produced by other microorganisms can also be present."[11]

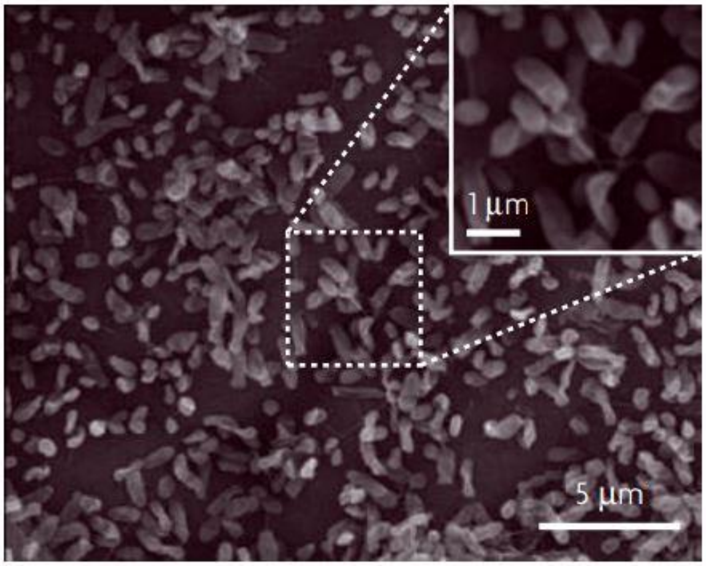

Figure 8. "Scanning electron micrograph of *Rhodopseudomonas palustrison* a carbon paper anode."[11]

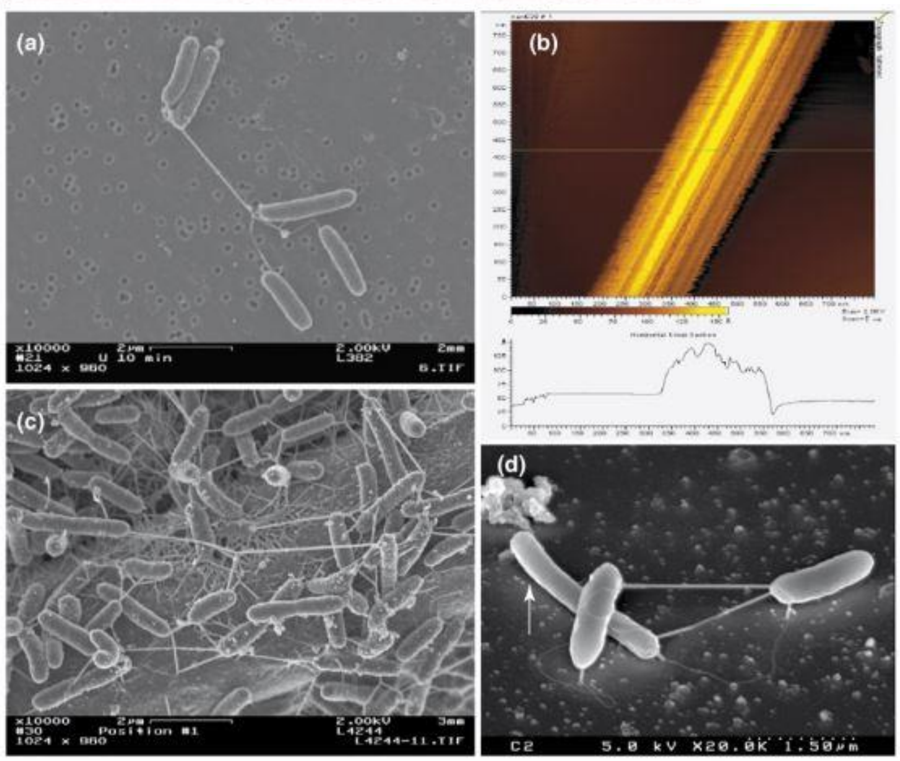

Figure 9. "(a) A scanning electron micrograph (SEM) of wild-type *Shewanella oneidensis* MR-1 grown under electron-acceptor-limited conditions, showing pilus-type nanowires that connect to other cells. (b) STM image of a single pilus-type nanowire from wild-type MR-1 (lateral diameter of 100 nm, topographic height of 5–10 nm) showing ridges and troughs running along the long axis of the structures consistent with a bundle of wires. The corresponding conductivity of the pilus as the tip moves over the indicated surface is shown beneath the STM image. (c) The anode from an MFC colonized by *S. oneidensis* MR-1. (d) An SEM image of *Pelotomaculum thermopropionicum* and *Methanothermobacter thermautotrophicus* (arrow) in methanogenic co-cultures showing pili connecting the two genera. Subsequent STM imaging has shown that the pili are conductive."[12]

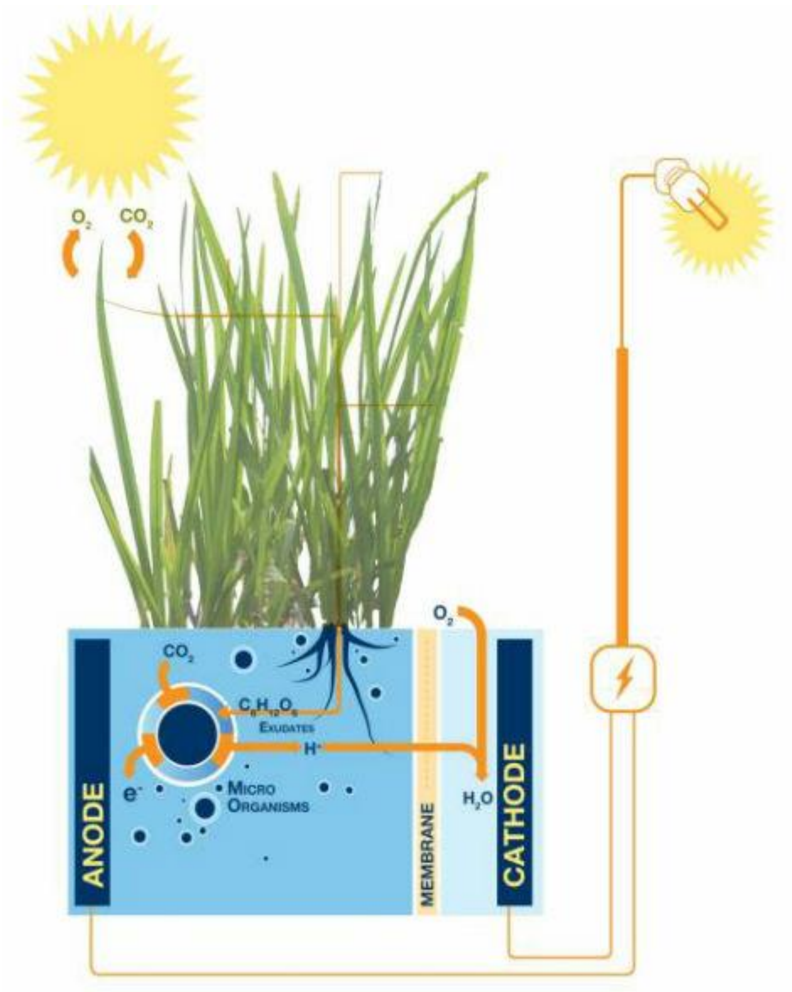

Figure 10. Schematic of the Plant-e PMFC. Image from: http://factor-tech.com/green-energy/1569-plant-power-the-new-technology-turning-green-roofs-into-living-power-plants/

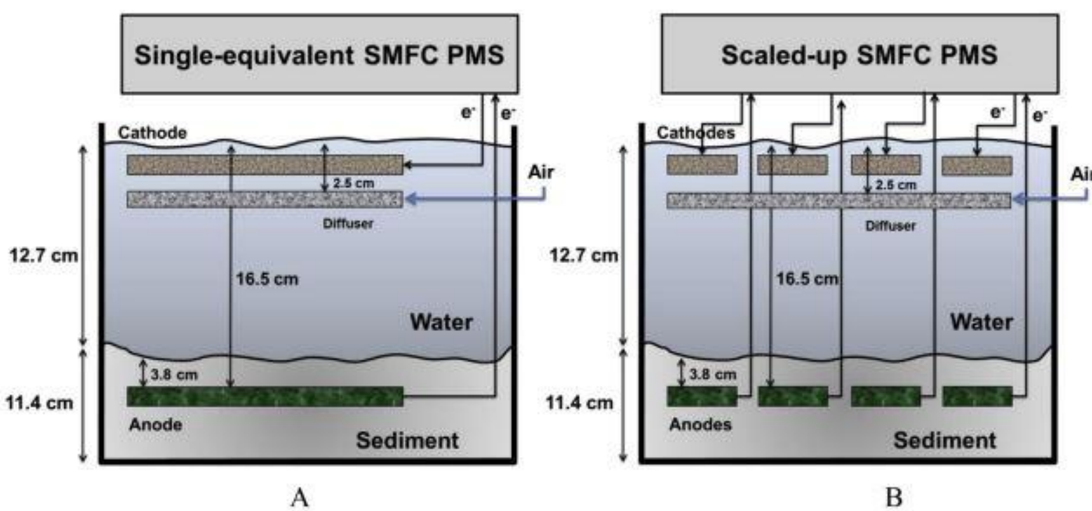

Figure 11. Schematic of (A) a single-equivalent SMFC and (B) a scaled-up SMFC.[2]

VIII. Bibliography

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