ABSTRACT
Microbial fuel cells (MFCs) have been a potential green energy source for a long time but one of the problems is that either the technology must be used on a large scale or special equipment have been necessary to keep the fuel cells running such as syringe pumps. Paper-based microbial fuel cells do not need to have a syringe pump to run and can run entirely by themselves when placed in contact with the fluids that are necessary for it to run. Paper-based microbial fuel cells are also more compact than traditional MFCs since the device doesn’t need any external equipment to run.

The goal of this paper is to develop a microbial fuel cell that does not require a syringe pump to function. This is done by layering chromatography paper with wax design printed onto it. This restricts the fluids to a specific flow path allowing it to act like the tubes in a typical microbial fuel cell device by delivering the fluids to the chamber. The fluids are picked up by tabs that sit in the fluid and use capillary attraction to flow up the tab and into the device. The fluids are directed to the chambers where the chemical and biological processes take place. These flows are then directed out of the device so that they are taken to a waste container and out of the system.

Our microliter scale paper-based microbial fuel cell creates a significant current that is sustained for a period of time and can be repeated. A paper-based microbial fuel cell also has a fast response time. These results mean that it could be possible for a set of paper-based microbial fuel cells to create a power density capable of powering small, low power circuits when used in series or parallel.

In this paper, we discuss the fabrication and experimental results of our paper-based microbial fuel cell. Also there will be a discussion of how paper-based microbial fuels cells compare to the traditional microbial fuel cells and how they could be used in the future.

INTRODUCTION
With the current search for new energy sources, there is a need for small, inexpensive, and efficient sources. Many people are looking at biorenewables because they mimic the way that nature creates energy, which should mean that they would be less harmful to the ecosystem. One specific type of biorenewable technology that has a lot of potential but has yet to be widely adopted is microbial fuel cell (MFC) technology [1-3].

The future will require more power than is currently required and current battery technology is already pushing its limits [4]. Fuel cells generally have a higher power density than batteries do and because of this, they are believed to be a likely source of electrical power in the future. Fuel cells are likely to be used in consumer devices such as cell phones, laptop computers, and analytical devices [1, 4-6]. Abiotic fuel cells such as hydrogen and methanol fuel cells are more well-known than MFCs are because they are currently able to produce the power that is needed to put them in many applications [7]. The problem with these devices is that they have several factors that make them less desirable to use in many applications, these factors include: high operating temperatures, expensive catalysts and very pure fuels required to function properly. The
fuels that are used in these devices also are not very safe since they are generally either highly explosive or toxic chemicals. These are large problems that just come with these kinds of fuel cells [7]. MFCs on the other hand could theoretically operate at any temperature that the microorganisms can withstand, can use fuels that are things that are usually thought to have very little value, such as wastewater, organic waste or several other similar types of fuels. MFCs are also generally easier to deal with since there are no special things that need to be done to deliver the fuel to the fuel cell. One large drawback to MFCs is that they have not been able to be used on a portable scale without the use of an outside pump to move the fluids through the device. This causes MFCs to be impractical for most uses. Currently MFCs provide enough power to be used in low power applications in remote regions of the world or in developing countries but they are not able to be effectively used in this way because they are tied to some outside source of a flow such as a syringe pump. If MFCs could be created that don’t require syringe pumps, they could be employed in these applications.

Some of these tasks can be currently accomplished by using three-dimensional microfluidic paper-based analytical devices (3D µPADs). These devices have been shown to be able to produce an electrical current and act as a battery [8]. This opens up opportunities to run small devices such as low power analytical devices. When these devices are placed in series or parallel with each other they can be used for higher power applications. A big advantage to using 3D µPADs is their ability to split the flow into several flows in a small device footprint [9]. This allows the device to do have a faster distribution of fluid due to the shorter travel distance required in the vertical direction; it also allows the device to do several different things from the same flow, this could mean that a device might be conceived that would have several different cells inside of a small device. Other advantages include the small volume of fluid needed to function, the lack of dependence on external devices or power. This is due to the fluid movement mainly being caused by capillarity and evaporation. Another application that µPADs are currently used in is point-of-care testing [10]. Point-of-care testing could also be done using MFCs if they didn’t need outside devices.

In this paper we attempt to use these ideas to create an MFC that can be applied in similar ways to the 3D µPAD devices. In order to accomplish this we needed to (1) develop a way to transfer the fluids through the device like the µPADs and make the flow function in a similar way to MFCs, without the use of a syringe pump; (2) make the device compact like the µPADs and (3) make the device easily expandable to allow several cells to draw from the same reservoir of fluids and be placed in series or parallel with each other in order to provide more power.

MATERIALS
The materials used in this paper include Nafion® membrane (Nafion® 115, Ion Power, New Castle, DE), carbon cloth (Fuel Cell Earth, Stoneham, MA), 0.01” titanium wire (Alfa Aesar, Ward Hill, MA), trypticase soy broth (Sigma Aldrich, St. Louis, MO), phosphate buffered saline (Sigma Aldrich, St. Louis, MO), and Chromatography paper (Grade 1 CHR, Whatman plc, Maidstone, Kent, UK), 10 kΩ resistor (RadioShack, Fort Worth, TX). Shewanella oneidensis MR-1 was cultured aerobically in trypticase soy broth (TSB) for use in this paper. Once the culture was matured a day it was put into the refrigerator for preservation throughout the course of the experiments. The bacteria was then soaked in TSB and transferred to a container of TSB for use by the device.

![Fig. 1: Design of the paper-based microfluidic microbial fuel cell.](http://proceedings.asmedigitalcollection.asme.org/)

The numbers tell which layer is depicted, 1 is the top and 4 is the bottom. Layers 1 and 2 are the anodic side of the device, Layers 3 and 4 are the cathodic side. Layers 2 and 3 have carbon cloth electrodes in the center of the device, and squeeze a PEM between them.

**DESIGN**
The device uses ideas from the 3D µPADs to create an MFC that can be used in applications similar to 3D µPAD applications. The device (design depicted in Fig. 1) is made from hydrophilic chromatography paper with a design printed on it with wax. The wax is melted into the paper creating areas that are hydrophobic, allowing us to direct the flow around the device [9]. The device consists of 4 separate layers of chromatography paper that sandwich 2 carbon cloths and a proton exchange membrane (PEM) at the center to form the basis of the MFC. The top 2 layers provide a flow to the anodic chamber. The 2nd layer begins with the tab that is placed in a reservoir of medium to draw it up and into the device, once the medium has travelled up the tab it enters the device through the 2nd layer and is transferred through the 2nd layer as well as up to the 1st layer. The medium travels along this path until in the 2nd layer it reaches the chamber and in the 1st layer until it reaches the line of hydrophobic paper that runs through the top layer just above the anodic chamber; this was placed here to force the medium to go through the chamber in order to proceed through the device. Once the medium has filled the chamber it proceeds through the device and out the other side. These 2 layers are actually the same design but the 1st layer has the tab cut off and the 2nd layer has the chamber area cut out for the carbon cloth which cuts the hydrophobic line out. The top layers are separated from the bottom layers by a PEM creating an anodic side and cathodic side. The bottom 2 layers are the exact same as the top 2 layers, just turned 90° so that they don’t
interfere with each other. They also function the same way except using ferricyanide to push through the cathodic chamber. The device functions by inoculating the anodic chamber with Shewanella Oneidensis MR-1 by placing it in TSB medium and running it through the top tab and top layers of the device to the chamber. In the anodic chamber the bacteria metabolize the TSB medium and produce electrons as byproducts, which are sent out through the titanium wire that is connected to the carbon cloth electrode and acts as a lead to connect to. At the same time the ferricyanide is introduced into the cathodic chamber through the bottom tab and through the bottom layers. The ferricyanide is continuously run through the device throughout the entirety of the testing. This allows the device to have constant amount of negative ions to accept the H⁺ that are exchanged through the PEM.

**EXPERIMENTAL SETUP**

In the current generation experiment (results shown in Fig. 2) the device was connected to a 10 kΩ resistor and the potential drop across the resistor was measured and recorded at 1 minute intervals. The current was calculated using Ohm’s Law:

\[ I = \frac{V}{R} \]  

(1)

With I being the current through the resistor, V being the potential drop across the resistor and R the resistance of the resistor. The device was inoculated by running the TSB medium through the anodic side of the device until the chamber was full and then removing the tab from the medium. The device was allowed to run as a batch culture until the current returned to a predetermined baseline level. The baseline level was determined by running ferricyanide through the cathodic side and TSB medium without bacteria through the anodic side. Once the current returned to the baseline the tab was replaced into the medium and the process started over.

To determine the open circuit voltage the device was prepared, the tabs placed in their respective fluids and the device was allowed to run unconnected to a circuit. The potential difference between the anode and cathode was measured by directly connecting the leads of the multimeter to the leads of the device. The ferricyanide and TSB medium with bacteria were both allowed to run continuously and the potential difference was recorded in 1 minute intervals. The reported open circuit voltage is the average of the values which were all within 5% of each other.

**RESULTS AND DISCUSSION**

This device reached a maximum current of 0.7 µA through a 10 kΩ resistor 5 minutes after the medium was reintroduced into the chamber, the current fell off over the next 5 minutes to around 0.25 µA, and then slowly fell back to the baseline over a period of half an hour. The subsequent spikes were lower and more consistent. These reached peak in just over 5 minutes and gradually fell back to baseline over a period of about 40 or 50 minutes. The run time was longer for lower peaks but keeping a relatively consistent average running current between 0.1 and 0.2 µA. The current vs. time graph has the predicted shape for each spike, including the slight bump about halfway down as the current drops back to baseline. This shape has been shown before for MFC current generation [11].
The maximum current density of the device was calculated to be 54 µA/cm², this is less than 40% of other MFC devices as shown in table 1. This is most likely attributed to a slower flow rate through the cathodic chamber, which is limiting how quickly the electrons can be transferred by limiting how quickly the protons can be transferred through the PEM because of less ions passing by the electrode per unit of time. We also obtained an open circuit voltage for both continuous culture and batch culture setups. This was calculated by measuring and recording the potential difference between the electrodes at 1 minute intervals and averaging the values that were recorded once it had reached a roughly constant value. For the continuous culture the constant value of 0.34 V was obtained shortly after the tab had initially been placed in the medium, the batch culture was measured by removing the tab from the medium. The result was a consistent exponentially decreasing curve, as the medium was metabolized, until the voltage leveled out at about 0.24 V. This level was retained for several days of just catholyte running through the device. We conclude that the bacteria provide a 0.1 V increase to the overall voltage of this device translating into about 30% of the most similar device [11], which agrees with the medium and the remaining 70% being contributed by the catholyte. The open circuit voltage of this device is about 40 % of that of the most similar device [11], this is less than 40% of other MFC devices as shown in table 1. This is most likely attributed to a slower flow rate through the cathodic chamber, which is limiting how quickly the electrons can be transferred by limiting how quickly the protons can be transferred through the PEM because of less ions passing by the electrode per unit of time.

We also obtained an open circuit voltage for both continuous culture and batch culture setups. This was calculated by measuring and recording the potential difference between the electrodes at 1 minute intervals and averaging the values that were recorded once it had reached a roughly constant value. For the continuous culture the constant value of 0.34 V was obtained shortly after the tab had initially been placed in the medium, the batch culture was measured by removing the tab from the medium. The result was a consistent exponentially decreasing curve, as the medium was metabolized, until the voltage leveled out at about 0.24 V. This level was retained for several days of just catholyte running through the device. We conclude that the bacteria provide a 0.1 V increase to the overall voltage of this device translating into about 30% of the voltage being contributed by the bacteria metabolizing the TSB medium and the remaining 70% being contributed by the catholyte. The open circuit voltage of this device is about 40 % of that of the most similar device [11], which agrees with the lower current density that we found previously. This could mean that we are just not operating at the optimum power output or it more likely this is due to the infancy of this device.

Further research will explore these possibilities. This device is also operating on a much shorter time scale than similar devices are. The explanation to this is most likely the fact that the chamber volume is not completely filled before the medium starts to flow out of the chamber and it cannot be filled to a greater extent. This can be fixed by tweaking the design of the chamber to not allow a flow to go directly through it in two dimensions but rather require that it be filled in the third dimension before the medium reaches a place that it is able to flow out.

**REFERENCES**


