

Bioelectricity production by *Trichoderma viride* using corn husk, corn cob residue and banana peel as substrates

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Abstract:

Microbial fuel cell is a recent alternative for the generation of electricity. These cells (MFC's) operate on the basis of microbial metabolism and produce current from a variety of organic substrates. In this study electricity generation with corn husk, corn cob residue and banana peel by the fungus *Trichoderma viride* has been undertaken. It was found that maximum electricity was generated when corn husk was used as a substrate although electricity was generated by the use of other two substrates also.

Keywords: Bioelectricity, *Trichoderma viride*, corn husk, corn cob residue, banana peel

Introduction

Carbon dioxide emission from conversion of fossil fuels contributes to global warming and thereby demands an alternative energy production based on renewable sources like solar energy, wind, water and biomass (De Vries *et al.*, 2007) ^[1]. The production of energy from biomass is important for production of sustainable energy and minimizing emissions of CO₂. Hydrogen gas can be biologically produced by fermentation of sugars (Ginkel *et al.*, 2001; Logan *et al.*, 2002) ^[2] but fermentation process can produce only 33% of the maximum potential energy from sugar such as glucose. In contrast to production of electricity indirectly from organic materials using biologically generated Hydrogen (H₂), it is now known that the electricity can also be directly generated by the degradation of organic matter in the microbial fuel cell (MFC) (Logan *et al.*, 2004). A MFC is a bioreactor that converts the energy stored in chemical bond in organic compounds directly into current through catalytic reaction of microorganism under anaerobic condition (Bond and Lovely, 2003) ^[3]. MFC consists of two chambers – an aerobic cathode chamber and an anaerobic anode chamber partitioned by an ion conducting membrane which is usually a proton exchange membrane. At the anode, organic substrates fed are oxidized by microorganism. The electrons which are produced are transferred from anode to cathode through an external circuit. Protons produced at anode migrate through the solution of anode across the proton exchange

membrane to cathode where they react with Oxygen (O₂) and electrons to form water (H₂O) (Antonopoulo *et al.*, 2009) ^[4]. Corn husk, corn cob residue and banana peel are a rich source of cellulose (Debabandya *et al.*, 2010) ^[5] and *Trichoderma viride* is known to produce cellulases that hydrolyse that cellulose. The current paper deals with electricity generation in MFC by *T. viride* using the above said substrates.

Materials and Methods

Culture maintenance:

The strain *T. viride* TV10 was procured from Andhra Pradesh Agricultural University. The culture was grown on Saubaurd Dextrose Agar (SDA) containing Ampicillin. The plates were incubated in an environmental chamber set at 25°C ± 2°C, 90% Relative Humidity (RH) and 16:8 hours, Light Dark regime. Culture was purified by single spore method. Aqueous conidial suspensions were made used as and when required.

Substrates:

Three substrates namely corn husk, corn cob and banana peel were used for this experiment. The substrates were washed and cut into 1 inch pieces and blended by using sufficient amount of distilled water (150 gm in 200 ml distilled water). The solutions were centrifuged at 11000 rpm in sterilized tubes for 15mins to remove aggregated solids. The supernatant was taken in 500ml Erlen Mayer flask, autoclaved at 15 psi at 110°C for 15mins, and inoculated with *T. viride* TV10 and incubated at 28°C for 48hrs in an orbital shaker set at 120 rpm.

MFC construction:

The experiment was set up using a two chambered MFC consisting of two borosil bottles (200ml) capacity. Three MFC were set up for three substrates. The agar salt was used instead of proton exchange membrane (PEM). The salt bridge was used for the electrolytic contact of the solutions in the two bottles where the two electrodes of the MFC's were immersed.

The anode and the cathode electrodes were made of carbon. Both electrodes had dimensions of 12cm X

1.5cm. Single stranded copper wires (I.D. 0.8mm) were used to connect the cell to the external circuit. An agar solution was made by heating a mixture of 2-5% of agar in 1M KCl. This solution was introduced into the glass U-tube having I.D. 12mm. The glass tube was stored in 1M KCl solution until used. The anode compartment was filled with 200ml of substrate which was inoculated with *T. viride*. The cathode compartment was filled with distilled water and was continuously aerated.

Results and Discussion

Culture maintenance:

On Sabouraud's dextrose agar medium *Trichoderma viride* exhibited fast growth. White mycelial growth was characterized by compact tufts, conidiation was seen after three days, spores were glaucous to dark bluish-green. On the reverse side of the petri plate the fungal growth was white or pale yellowish. The branching of conidiophores was not very extensive. Single, paired branches were observed under the microscope. Phialides were single, paired or 3-verticillate, narrowly longiform, 8-14 μ m X 2.4-3.0 μ m. The conidia were globose to ellipsoidal, bluish-green to dark green, 4.0-4.8 X 3.5-4.0 μ m (Rifai, 1969)^[6]. Stained specimen of TV10 strain is shown in Figure 1. The mycelial growth, early sporulation and fully sporulated culture on SDA medium are shown in Figures 2, 3 and 4 respectively.

Fig 1. Microscopic image of *T.viride* TV10 under oil immersion objective



Fig 2. Mycelial growth of *T.viride* TV 10 on Sabouraud's Dextrose Agar



Fig 3. Initiation of sporulation of *T.viride* TV10 on Sabouraud's Dextrose Agar



Fig 4. Fully sporulated culture of *T.viride* TV 10 on Sabouraud's Dextrose Agar



Electricity generation:

It was observed that all three substrates were efficient in generation of electricity. The current produced by the use of all three substrates increased with time. However the maximum voltage and current was generated using the corn husk (490 mV) followed by corn cob (366.1mV) and the least current was generated by the banana peel (93.3 mV). The graphs of voltage versus time are shown in fig. 5, 6 and 7 there was a 25% increase in the voltage production by corn husk over corn cob and 80.9% increase over current produced by banana peel.

Fig. 5. Graph of voltage versus Time with corn husk as substrate

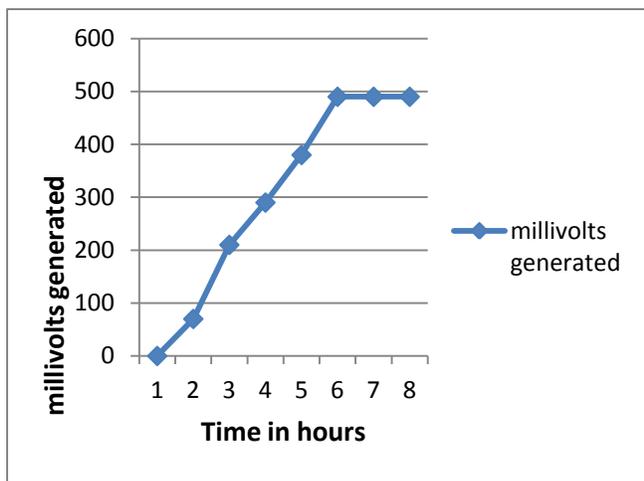


Fig. 6. Graph of voltage versus Time with corn cob residue as substrate

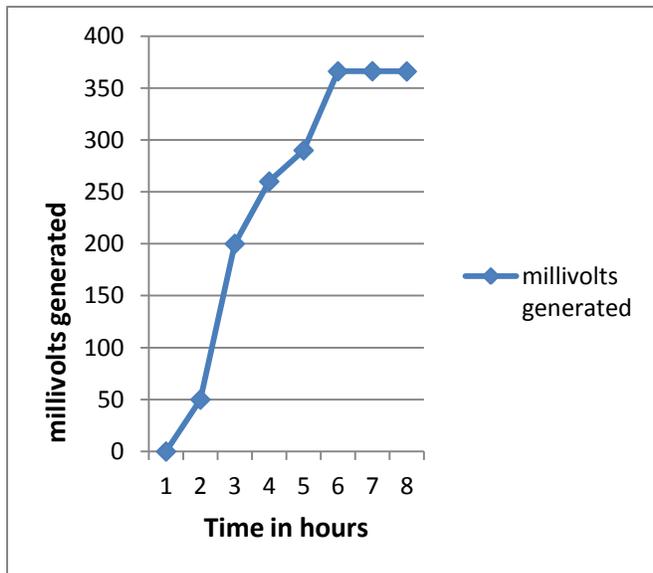
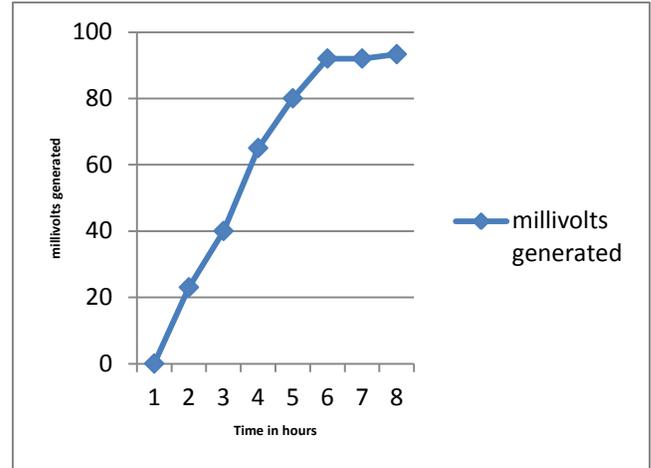


Fig. 7. Graph of voltage versus Time with banana peel as substrate



The results suggest that *Trichoderma viride* can be used to generate electrode potential using variety of substrates. However, corn husk has proven to be most efficient. Several factors influence the quantum of electricity generated in a microbial fuel cell and the knowledge of optimized parameters ensures maximized production of bioelectricity. Since the reactions in a microbial fuel cell are carried out by microorganisms, the primary requirement in any MFC is optimized conditions for the growth of the culture. In a previous experiment carried out in our laboratory, we optimized the conditions for growth of *T.viride* TV10 strain and ideal conditions were found to be pH 5, a temperature of incubation 25°C and incubation time of 72 hours. Glucose when used as a supplementary sugar, proved to be efficient in growth of several microbes as glucose is a respiratory sugar (Brannon, 1923)^[7]. Hence it becomes obvious that substrates rich in glucose might increase the rate of respiration thereby the electricity generated. However sources of glucose are on a demand and there is not enough substrate available for generation of electricity. On the other hand cellulose, when hydrolysed yields glucose. Thus a cellulose rich substrate can be inoculated with a high cellulase yielding microorganism to generate a large amount of electricity. The optimized conditions for cellulase production by *T.viride* (pH 5, temperature 25°C) can be used for generation of electricity as well. Corn cob residue contains other sugars besides cellulose such as lignin. But the content of cellulose is high in corn husk, which is probably responsible for the high bioelectricity generated.

Conclusion

Cellulose rich substrates such as corn husk can be efficiently utilized by *T.viride* for bioelectricity production. This preliminary work has a wide scope as various substrates can be explored with an aim to minimize the use of fossil fuels.

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