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# An innovative mixotrophic approach of distillery spent wash with sewage wastewater for biodegradation and bioelectricity generation using microbial fuel cell



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#### ABSTRACT

In the present study, H-type microbial fuel cell (H-MFC) has been used to treat distillery spent wash (DSW) diluted with sewage wastewater (SWW) at different mixing ratio. Mixture of DSW and SWW was used as substrate in anaerobic environment. The cathodic compartment was filled with BBM media and microalgae (Scenedesmus abundans). CO<sub>2</sub> generated during breakdown of organic substrates by anaerobes present in DSW was utilized by microalga Scenedesmus abundans for photosynthesis. A consortium of two bacteria (Pseudomonas aeruginosa and Bacillus cereus) was used for the 21 days period of DSW treatment. Significant reduction in chemical oxygen demand (COD) in the order of 66% to 78.66% was observed. At 50:50 ratio of DSW and SWW, total dissolved solid (TDS) of 39.66% and total suspended solid (TSS) of 97% were removed. A maximum power density (PD) of 836.81 mW/m² and open circuit voltage (OCV) of 745.13 mV were obtained along with biomass yield of 0.74 g/L d<sup>-1</sup> with above ratio of DSW and SWW. This study demonstrated that proper dilution of Distillery spent wash with sewage wastewater may leads to an efficient wastewater remediation and energy production.

#### 1. Introduction

The extensive use of fossil fuels, especially oil and gas, in recent years, has lead to the depletion of fossil fuels, environmental pollution and efficient development of renewable energy sources with higher performance [1–4]. The new alternative sustainable energy technologies are need to be eco-friendly, energy neutral and efficient [5].

Microbial fuel cells (MFCs), a hybrid biochemical device, have been tested as a promising technique for wastewater treatment with energy recovery by many researchers [6–8]. Microorganisms are the mediator to convert a large variety of biodegradable organic compounds into CO<sub>2</sub>, water and energy [5,9]. This facilitates microbial interaction to convert chemical energy to electrical energy and production of value added products through the metabolic activity of microorganisms [2,10–12]. A general design of a two chambered MFC consists of anodic compartment where microorganisms help in oxidative conversion of the substrate. Simultaneous chemical reductive conversion takes place in cathodic compartment. The electrons produced in anodic chamber, pass through the external circuit and at the same time protons passing through a proton exchange membrane (PEM) react with an oxidizing agent, such as oxygen, at the cathode surface which leads to close the

circuit [13–16,37]. With integration of multiple process, elements and experimental conditions, the setup may also have positive influence on the overall wastewater treatment efficiency. The conversion efficiency of organic wastes to bio-energy and economic viability depend on the sources of waste, its chemical composition, characteristics and concentration of the components that can be converted into bio-energy [17].

Distillery spent wash has become a major source of environmental pollution due to the presence of high organic load, dark brown color and unpleasant odor in it. It also contains considerable nutrients in terms of potassium, sulphur, nitrogen and phosphorus as well as large amount of micronutrients like Ca, Cu, Mn, and Zn [18]. Among all the conventional treatment processes, anaerobic treatment is widely accepted practice and also it has been tried at pilot and full scale operations [19].

Prior research has been carried out on the treatment of distillery spent wash in microbial fuel cells (MFCs) to utilize the high organic matters as an oxidizing agent for production of electricity. Brewery wastewater containing COD of 2000 mg/L was treated in dual chambered (rectangular) MFCs (0.2 L anode volume) to obtain power density of 305 mW/m² at 30  $\pm~2$  °C with 80% removal of COD [20]. Treatment

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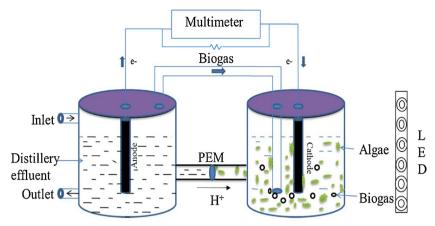


Fig 1. Schematic diagram of MFCs used in this study.

of high strength alcohol distillery spent wash in an integrated AFB-MFCs system showed 80–90% removal of COD and a maximum power density of 124.03 mW/m $^2$  was attained with an external resistance of 120  $\Omega$  [21].

Traditional aerobic or anaerobic wastewater treatment technologies are energy consuming with emission of large amount of carbon dioxide (CO<sub>2</sub>) as the major gaseous end product when either glucose or acetate was used as substrate [22]. As algae are recognized as excellent CO<sub>2</sub> sinkers and oxygen is generated simultaneously in their metabolic activities, several researchers showed interest to use algae in cathodic chamber for oxygen production which acts as a terminal electron acceptor [22–24]. Wang [22] reported a maximum voltage output of  $706 \pm 21 \, \text{mV}$  (1000  $\Omega$ ) when *Chlorella vulgaris* was grown in cathodic chamber.

In this study, sewage wastewater was mixed in a different proportion with distillery spent wash to examine the wastewater treatment, bioelectricity generation as well as utilization of  $\rm CO_2$  for biomass production using dual chambered MFC. Four different concentration of distillery effluent mixed with sewage were used as substrate. Microalgae Scenedesmus abundans was grown in cathode chamber.  $\rm CO_2$  produced in the anode chamber was supplied to the cathode for the use of microalgae for their metabolism and growth. The overall performance of the MFC was evaluated in terms of COD removal, metallic cations removal, maximum voltage production, maximum power density and biomass production along with specific growth rate and doubling time.

#### 2. Materials and methods

#### 2.1. Culture collection and cultivation

Active bacterial cultures of Pseudomonas aeruginosa (MCC No. 2622) and Bacillus cereus (MCC No. 2240) were procured from National Center for Cell Science, Pune, India. Both the bacterial cultures were cultivated in nutrient agar with sodium chloride media for 48 h at 25  $\pm$  1  $^{\circ}$ C in incubator. After incubation period of 48 h, cultures were transferred in a single media to develop an efficient bacterial consortium. Scenedesmus abundans (NCIM No. 2897) was obtained from National chemical Laboratory, Pune, India and cultivated in BBM medium with the following components per liter: NaNO<sub>3</sub>, 0.25 g; MgSO<sub>4</sub>. 7H<sub>2</sub>O, 0.075 g; NaCl, 0.025 g;: K<sub>2</sub>HPO<sub>4</sub>, 0.075 g; KH<sub>2</sub>PO<sub>4</sub>, 0.175 g; CaCl<sub>2</sub> 2H<sub>2</sub>O, 0.025 g; H<sub>3</sub>BO<sub>3</sub>, 0.011 g; EDTA, 0.05 g; KOH, 0.031 g; FeSO<sub>4</sub>·7H<sub>2</sub>O 0.0049 g; H<sub>2</sub>SO<sub>4</sub>, 0.001 mL and trace elements (ZnSO<sub>4</sub>·7H<sub>2</sub>O, 0.00882 g MnCl<sub>2</sub>·4H<sub>2</sub>O 0.00144 g, MoO<sub>3</sub>, 0.0071 CuSO<sub>4</sub>·5H<sub>2</sub>O, 0.00157 g Co (NO<sub>3</sub>)<sub>2</sub>·6H<sub>2</sub>O 0.00049 g). The microalga Scenedesmus abundans was cultivated in 1L Erlenmeyer flasks with 750 mL of autoclaved BBM medium at 25  $\pm$  1  $^{\circ}$ C under continues illumination (22W, LED tube light) with light intensity of  $94.6 \,\mu$  mol m<sup>-2</sup> s<sup>-1</sup>. There was no supply of CO2 externally except the availability in the atmosphere.

#### 2.2. Wastewater collection and characterization

The raw distillery spent wash (DSW) was collected from the biogas production unit of ethanol distillation plant and the sewage wastewater (SWW) was collected from domestic sewage treatment plant. Both the samples were preserved at 4 °C prior using. Heavy metals were characterized using inductively coupled plasma optical emission spectrometry (ICP-OES) (Teledyne Leeman Labs, Prodigy SPEC, USA). COD, TDS, TSS and TS were determined using method provided by APHA (1992). Electrical conductivity and pH was measured using digital meter (HI 3512, Hanna Instruments). Samples were prepared by mixing distillery spent wash and sewage wastewater in the ratios of 100:0, 75:25, 50:50 and 25:75 by v/v and were denoted as  $DSW_{100} + SWW_{00}$ ,  $DSW_{75} + SWW_{25}$ ,  $DSW_{50} + SWW_{50}$ ,  $DSW_{25} + SWW_{75}$ , respectively.

#### 2.3. Experimental setup

An 'H' type dual chambered microbial fuel cell (Fig. 1) was designed and fabricated (Fig. 2) using transparent Plexiglas material of thickness 0.5 cm which consists two equal volume cylindrical chambers (each with working volume of 900 mL) separated by proton exchange membrane (PEM). The height and diameter of each chamber were 15 cm and 10 cm, respectively. Graphite rod with 17 cm length and 1.2 cm diameter were used as electrodes in both the chambers. The electrodes were connected to an external resistor of  $100\,\Omega$  in a loop configuration and the voltage generated was monitored by multi-meter (Mashtech India pvt.ltd.). In this experiment pretreated Nafion-117 from DuPont was used as proton exchange membrane. Pretreatment of nafion-117 membrane was done by dipping the membrane first in a solution of  $H_2O_2$  (3% (v/v) for 1 h at 80 °C and then in  $H_2SO_4$  (0.5 M) for 1 h at 80 °C. The membrane was washed with boiling water after each solution treatment [25].

Pretreated (autoclaved) wastewater sample was fed in batch mode for 21 days in the MFC at 25  $\pm$  1 °C. Anaerobic condition was maintained in anodic chamber by purging nitrogen gas was for around 15 mins. The gasses produced in anodic chamber including CO $_2$  were transferred to cathodic chamber through an outlet duct. The gas was utilized by microalgae in cathodic chamber. BBM medium was used in the cathodic chamber for the growth of Scenedesmus abundans. Cathodic chamber was kept under temperature controlled environment and illumination using 4 LED strips with light intensity of 94.6  $\mu$ mol m $^{-2}$ s $^{-1}$ . The light and dark cycle maintained was 16 h: 8 h. A sample of 2 mL of sample was collected every day for test of different parameter test.

#### 2.4. Electrochemical and chemical analysis

Open circuit voltage was monitored and recorded at regular interval of 1hr for 21 days by digital multi-meter connected to  $100\,\Omega$  external

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