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REGULAR ARTICLE

DEGRADATION OF AZO DYE AND ELECTRICITY GENERATION USING YEAST MEDIATED MICROBIAL FUEL CELL

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ABSTRACT

In the present study, the efficiency of yeast mediated microbial fuel cell (MFC) was investigated towards degradation of Trypan blue (azo dye) and electricity generation. Five yeast strains viz. SC1, SC2, SCD1, SCD2 and SCD3 were isolated from different sources. The internal resistance of yeast isolates was tested using ferric oxide reduction method. To maximize the power density of MFC, NaCl was added to the medium and NaCl tolerance of yeast strains was tested. Among the five isolates, SC1 and SCD2 showed maximum ferric oxide reduction and NaCl tolerance. Initially 5 % of SC1 and SCD2 yeast culture were inoculated in wastewater containing azo dye (100 µg/ml) in a H-type MFC chamber and 250 ml conical flask used as control. Increased growth of yeast strain in MFC chamber was noted compared to conical flask culture. The data of electricity generation was taken for 15 days and electricity generation was measured using multimeter. Maximum electricity generation was noted in SC1 (950mV) followed by SCD2 (750mV). In addition, SC1 could degrade azo dye more efficiently than SCD2. Therefore, it may be concluded thatSC1 yeast mediated MFC can be used as potential technology for electricity generation and degradation of azo dye in wastewater.

Keywords: Azo dye, Electricity generation, FTIR analysis, Microbial fuel cell, Optimization

INTRODUCTION

Microbial fuel cell (MFC) has gained a great attention towards its ability in generating electricity by enhancing biodegradation of contaminants **(Zhang *et al.*, 2009)**. Several factors affect electricity generation such as pH, dissolved oxygen concentration in the cathode compartment. The highest electricity was generated at pH-7 with 6 mg/l DO supply **(Gil *et al.*, 2003)**. Due to external circuit to direct electron transfer in MFCs as a driving force, pollutant degradation could be effectively stimulated **(Bond *et a.,l* 2002; Zhang *et al.*, 2010).** It has higher capability of degrading toxic chemical or dye present in wastewater or textile industry which is very harmful to human **(Patade *et al.*, 2016)**. Azo dye is the chemical dye being used largely for textile, paper painting industries and discharge toxic effluents which cause higher pollution to the environment **(Jafari *et al.*, 2013)**. This azo dye used for textile, paper painting, dying to clothes in many industries and discharge toxic effluent which cause higher pollution to the environment **(Yueh *et al.*, 2016)**. Due to this penetration of light become less and reduce activity of photosynthetic which leads to less production of oxygen in aqueous ecosystem. It also has adverse impact of chemical oxygen demand, biological oxygen demand **(Saratale *et al.*, 2009)**. Recently, decolorized intermediate(s) of azo dye(s) were found to act as electron shuttles in dye-bearing MFC to enhance the performance of simultaneous color removal and power generation **(Chen *et al.*, 2013)**. Anodic treatment process on azo dye treatment using single chambered MFC, would be more favorable than double chamber MFCs due to non-mixing contacting reactor scheme for operation **(Miyahara *et al.*, 2015)**. MFCs considered as higher salinity for production of power after adding NaCl to it, increases the density of internal resistance of anode system and gives enhanced power output **(Kong *et al.*, 2015)**. It showed 94% degradation on acid orange 7 pollutants. In particular, Thung et al., **(2015)** applied a single-chamber up-flow membrane-less MFC for decolorization of Acid Orange 7 and proposed that both anode and cathode contribute to dye decolorization, with the latter breaking down azo bonds the received electrons and protons on its surface. Many species like *Geobacter bemidjiensis, Jishengella* sp. and *Verrucosispora* sp. reported for higher production of electricity from 200 to 954mV **(Shaikh *et al.*, 2016)**. *Bacillus circulans* NPP1 MFC showed 96% methyl red decolorization and 859 mV electricity generation in a two chambered microbial fuel cell **(Huarachi-Olivera *et al.*, 2018)**. Reports are scanty on yeast mediated microbial fuel cell.

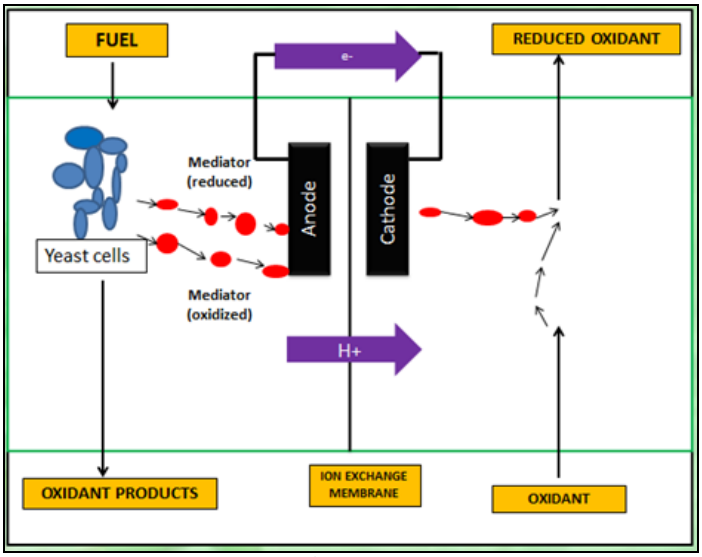

Figure 1 Yeast mediated microbial fuel cell

This study investigates the potential use of yeast mediated MFC for simultaneous electricity generation and treatment of textile effluent which is contaminated by trypan blue dye. Single chambered MFCs was used in this study and operated over three-batch cycles for 15 days (Figure 1).

MATERIALS AND METHODS

Sample collection

Sample of wastewater was collected from in and around vellore such as fresh cow dung, chicken, flower, millet root, sludge from VIT campus. Five yeast strains were isolated and screened for maximum electricity generation.

Chemicals

M9 media: $K_2HPO_4.2H_2O$ (0.25 g), K_2PO_4 (0.25 g), NaCl (1.25 g), $(NH_4)2SO_4$ (0.075 g), $MgSO_4.7H2O$ (0.075 g), $CaCl_2$ (0.005g). Trypan blue, ferric oxide, sodium thioglycollate, glucose, methylene blue, sulfuric acid, acetic acid, potassium iodide, sodium sulfite, ammonium chloride, dipotassium hydrogen phosphate, disodium hydrogen phosphate, calcium carbonate, magnesium

sulfate, ferric chloride, phosphate buffer, sodium sulfate, starch solution, glucose **(Herrero-Hernandez *et al*., 2013)**.

NaCl tolerance

Yeast isolates were inoculated on 100 ml of M9 media which contains different concentrations of NaCl (0.05 M-1.5 M) to increases internal resistance for electricity generation **(Heijne *et al*., 2007)**. Incubated at 37º C for 24-48 h. Blank media serves as a control. Samples were read at 600 nm using spectrophotometer.

Ferric oxide reduction test

Yeast isolates were inoculated on 100 ml of M9 media with 0.1% ferric oxide and 0.1% sodium thioglycollate with or without 1% glucose. Incubated at 37º C for 24-48 h. Blank media serves as a control. Ferric oxide turns into black color indicates presence of ferric oxide reducing yeast **(Ruggero Rossi, 2016).**

Respiration Test

One ml of yeast isolates was mixed with 9 ml of milk in a clean, sterilized test tube. To this 1ml of methylene blue was added and incubated on water bath at 37º C for 30 min. After 30 mins its shows reduction in milk color **(Akshay *et al*., 2016)**

Microbial fuel cell chamber

Two plastic containers, each of ~ 7-8 cm height and 8 cm breadth were taken to make the MFC design. Each container was filled with ¾ sterile soil and ¼ with yeast culture. The lid of the containers had 2 holes, one for anode and another for cathode. Kept as airtight container. Three different types of setup were made based on NaCl concentration (1%, 2% and 3 %). This setup was kept at 37ºC with pH 7 for 24-72 h. Voltage and amperes was noted by using digital multimeter and plotted a graph for electricity generation **(Patil *et al*., 2012).**

Degradation of trypan blue

Selected yeast isolates were inoculated on 100 ml trypan blue contaminated effluent and incubated at 37ºC for 24-48 h. Decolorization was noted by spectrophotometer at 600nm and degradation was calculated by Jumma Shaikh et al **(Heijne *et al*., 2007)**.

RESULTS AND DISCUSSION

Preliminary Screening of isolated yeast

Five yeast strains were isolated from different sources like fresh cow dung, chicken, millet root, waste water drainage. Yeast isolates were shown in Figure 2. Screening results reveals that SC1 and SCD2 has maximum tolerance against 3 % NaCl (Figure 3) and ferric oxide reduction (Figure 4) in M9 media.

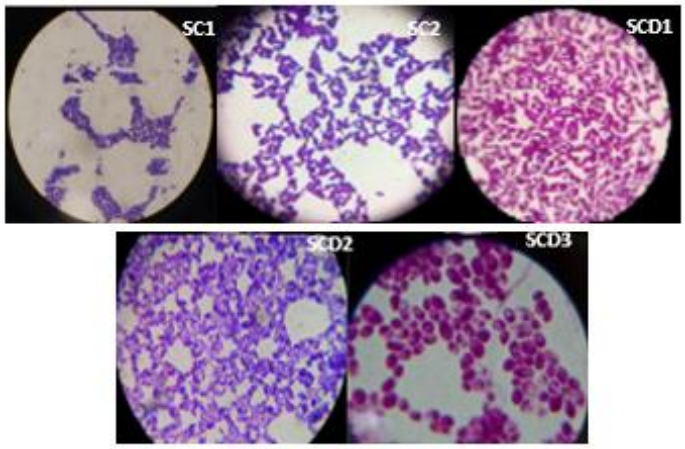

Figure 2 Microscopic images of yeast isolates

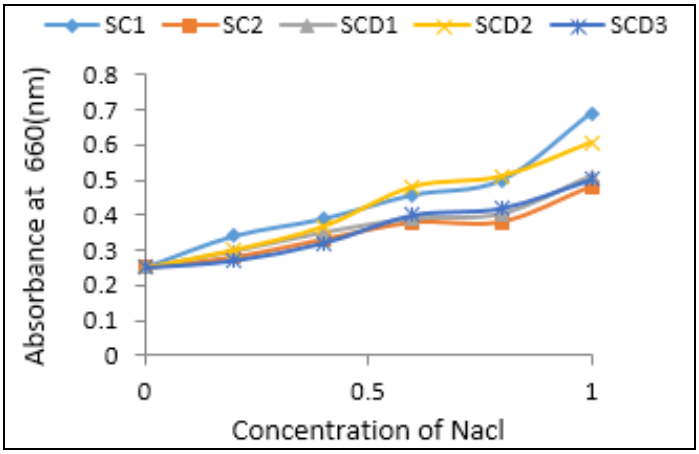

Figure 3 NaCl tolerance

Figure 4 Ferric oxide reduction test

Respiration test

Respiration of selected yeast isolates were analyzed and results showed decoloration within 30mins (Figure 5). Faster respiration rate in microbes speeds up an electron transport system which leads to maximum electricity generation.

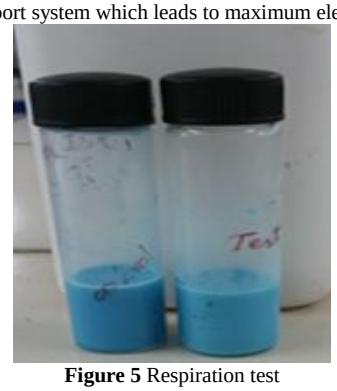

Figure 5 Respiration test

Electricity generation

MFC was designed for two isolates and initially electricity was measured for 15 days by using multivoltmeter. SC1 and SCD2 showed maximum electricity generation 720 mV, 560 mV on 7th day culture without trypan blue (Figure 6a). MFC chamber with trypan blue showed increased power density 820mV and 620mV for SC1, SCD2 respectively (Figure 6b). In both the case SC1 showed higher electricity generation compare to SCD2.

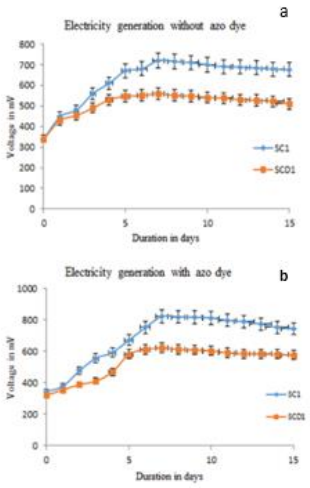

Figure 6 Electricity generation

Electricity generation after dye treatment

Electricity generation was taken by multivoltmeter after treating with trypan blue dye. SC1 showed increased electricity (670-720 mv) than SC2 (580-880 mv) on 7th day and 15thday respectively (Figure 7). Further SC1 was chosen for degradation studies.

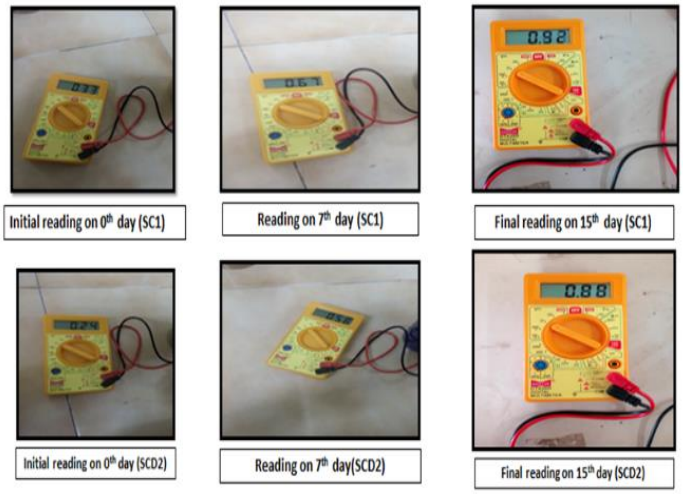

Figure 7 Electricity generation by multivoltmeter using MFC

Decoloration of dye by MFC

Maximum decoloration rate was achieved on 4th day (Figure 8) and 6th day (not shown) in SC1, SC2 respectively. MFC with trypan blue contaminated textile effluent showed 76 % and 64 % degradation rate in SC1, SCD2 respectively. Maximum degradation was achieved by yeast mediated MFC compared to conventional method (Figure 9).

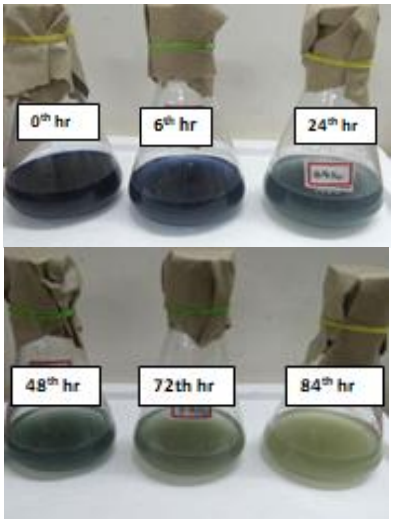

Figure 8 Decoloration of dye by MFC

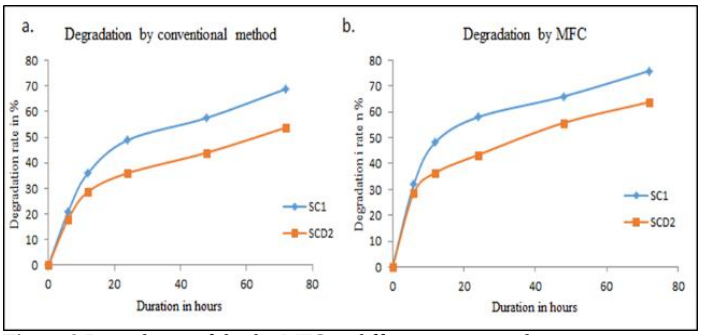

Figure 9 Degradation of dye by MFC in different time period

FTIR Analysis of trypan blue

FTIR analysis shows five different peaks of trypan blue without the sample and SC1 shows two peaks which indicates degardation of trypan blue at 3329.14cm^{-1} alkanes group (O-H bond) and 1635.64cm^{-1} (C-H bond). SCD2 shows peaks at 3331.07cm^{-1} (O-H bond) ,2768.20cm^{-1} (C-H bond) and 1635.64cm^{-1}(N=N) which indicates partial degrdation of trypan blue (Figure 10).

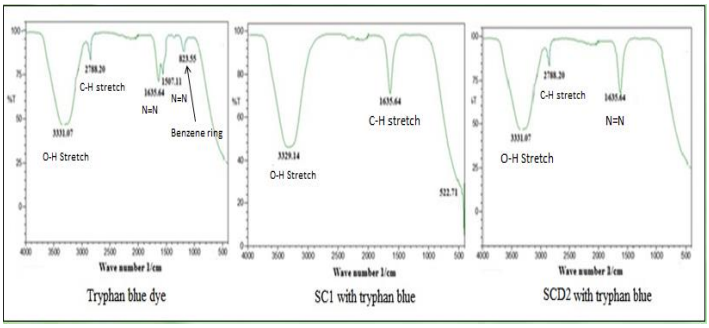

Figure 10 FTIR Analysis of trypan blue with two yeast isolates

DISCUSSION

In the present study, five yeasts were isolated from different sources and screened for ferric oxide reduction and NaCl tolerance to improve the quantity of electricity generation. Among the five isolates, SC1 and SCD 2 showed maximum electricity (~720 and 880 mV respectively) from a single container. The most interesting observation was that the setup could give almost same electricity output for nearly 10 days without adding anything. Further, yeast isolate SC1 and SCD2 chosen for trypan blue degradation in waste water. After treatment significant degradation of trypan blue was noted in MFC mediated degradation compared to conventional method of degradation. SC1 degraded trypan blue, azo dye more efficiently than SCD2. Therefore, it may be concluded that SC1 yeast mediated MFC can be used as potential technology for electricity generation and degradation of azo dyes in wastewater.

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