ELECTRON TRANSFER MECHANISM FROM MICROBES TO ELECTRODES IN BIOELECTROCHEMICAL PROCESSES: A SHORT REVIEW

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ABSTRACT

Electron transfer mechanisms of electroactive microbes or electrogens are investigated in this research and majorly three types of electron transport mechanisms were observed such as direct electron transfer, nanowire-based electron transfer and mediator-based electron transfer. Electron transfer mechanism by electrogenic bacteria can follow multiple electron transfer pathways such as popularly known electrogens, *Geobacter sp.* and *Shewanella sp.* follows all three electron transfer mechanisms. C-type cytochrome plays an important role in direct electron transport as observed from *Geobacter sulfurreducens*, *Geobacter metallireducens*, *Shewanella oneidensis* and *Desulfovibrio desulfuricans*. Pyocyanin and pyorubrin produced by *Pseudomonas aeruginosa* acted as a mediator for electron transfer from *Pseudomonas aeruginosa* to the electrode surface. Riboflavin and flavin were observed as an excellent mediator produced by different electrogens such as *Listeria monocytogenes*, *Lactococcus lactis* and *Bacillus megaterium* etc.

INTRODUCTION

Bioelectrochemical system (BES) is a promising technology that generates electrical energy from biodegradable organic waste or synthesizes usable by-products and fuels using a biofilm on the electrode as the biocatalyst (Jadhav et al., 2017). Desalination (Pradhan and Ghangrekar, 2014), carbon sequestration (Neethu et al., 2018), valuable biomass recovery (Kaku et al., 2008) are several other emerging applications that can be possible from BES. However, in spite of excellent benefits achievable from BES, most of the BES variants have not seen the light of commercial applicability because of very low yield, i.e. energy recovery or usable by-products or fuel recovery. Additionally, successful applicability of BES is not possible without efficient conversion of chemical energy stored in a BES to electricity and also enrichment and efficient function of electroactive microorganisms. Microbial fuel cell (MFC), is the most popularly explored variant of BES technology applied for electricity recovery while treating oxidizable wastewaters in anodic chamber and trials for improvement of the efficiency of MFCs have been approached by the researchers worldwide. The overall coulombic efficiency (CE) in MFCs has been improved over the years through electrode material modifications (Das et al., 2018), genetic modification of anodic inoculum (Angelaalincy et al., 2018; Kumar et al., 2016; Li et al., 2018) or chemical (Ghadge et al., 2013) modification of microbial community based on regulation of genes in exoelectrogens, microbial ecology of the anodic biofilm (Butler and Nerenberg, 2010), adopting synthetic biology (Li et al., 2018) approaches and through syntrophic interactions among bacterial communities (Kiely et al., 2011). Electroactive or electrogenic microorganisms, alternatively termed as, electrochemically active bacteria (EAB), exoelectrogen or anode respiring bacteria (ARB) plays significant role behind the improvement in the performance of the anaerobic anodic chamber of BES to improve efficiency of the system in terms of (CE) or power density (Logan et al., 2006) and transfer of electrons from organic matter to various electrochemically active electron carriers or electrode surface. In addition, these EAB are capable of transferring electrons out of the cell membranes to the electrode surface either directly through membrane-bound protein (Ueki et al., 2018) or lipid structures (Subramanian et al., 2018), such as pili, c-type cytochrome and filaments, or using mobile electron shuttles, e.g. mediators for indirect electron transfer (Logan, 2009, 2007; Yong et al., 2012).

Performance of BES in terms of bioelectricity production can be enriched by improving mass transfer rates across electroactive microorganisms-electrolyte interfaces. Improved understanding of these EAB and microbial community, anode potential, choice of anolyte, electrode material, pH range in a MFC will aid in revealing the mechanism and will guide to increase bioelectricity generation required for intended field-scale applications of MFCs. Here in the present research we investigated the vital pathways of electron transfer mechanisms in BES and their effect on the performance.

ELECTRON TRANSFER PROCESSES

Three types of electron transfer mechanism of electrogens are commonly observed in BES (Niu et al., 2020; Santoro et al., 2017). For the first case, the surface of the cell directly attached to the electrode surface and transfer electron which is known as direct electron transfer (Gralnick and Newman, 2007). Alternatively, a cellular appendage (such as an electrically conductive pilli or nanowire) forms a connection between the cell and the electrode surface and catalyse electron transfer (Gralnick and Newman, 2007). Electron transfer can also be performed by a mediator secreted by the microorganism itself or present in the external environment which carries electron from the surface of the microbes and releases it to the electrode surface. For a particular bioelectrochemical system an electron transfer by a particular mechanism or a combination of multiple mechanisms is possible.

DIRECT ELECTRON TRANSFER

Direct electron transfer mechanism follows the pathway of electron transfer from the electrogens to electrode surface through direct contact between cell wall and electrode. Multiple instances were observed regarding the direct electron transfer of exoelectrogens. Popularly known electrogenic microbes *Geobacter sulfurreducens*, *Geobacter metallireducens* and *Shewanella oneidensis* was observed to transfer electrons through direct electron transfer mechanism via C-type cytochromes (Carmona-Martínez et al., 2013; Liu et al., 2014; Meitl et al., 2009; Smith et al., 2013). C-type cytochrome also plays an important role in electron transport by *Desulfovibrio desulfuricans* (Kang et al., 2014). Arnold et al. observed *Pseudomonas aeruginosa* primarily follows direct electron transfer mechanism (Arnold et al., 1988). Direct electron transfer through C-type cytochrome was also observed from *Thermincola potens strain* JR although this is a gram-positive anaerobic bacterium having thick cell wall (Wrighton et al., 2011).

ELECTRON TRANSFER THROUGH NANOWIRE OR PILI

Microbial nanowires play an important role in extracellular electron transfer and increasing nanowire production enhances current densities in bioelectrochemical systems like microbial fuel cells (Malvankar and Lovley, 2014). An efficient electron transfer mechanism was observed by

Kluyvera georgiana MCC 3673 by using pili (pil Q gene) and it also uses flagella (fli P gene) to move towards electrode based on electrical potential gradient and thus responsible for bio-electricity generation in bio-electrochemical systems (Thapa and Chandra, 2020). Other than direct electron transfer mechanism, some *Geobacter* and *Shewanella* strains can develop electrically conductive molecular pili (e-pili) to enable distant electron transfer (Qiao et al., 2008; Reguera et al., 2005). Electron hopping and tunneling between cytochromes surrounded by the filaments or nanowire is electron transfer pathway for *Shewanella oneidensis* nanowires however *Geobacter sulfurreducens* nanowires are comprised of pili having high conductivity because of the presence of overlapping pi–pi orbitals of aromatic amino acids (Malvankar and Lovley, 2014). Poorly conductive native pili of *Pseudomonas aeruginosa* were modified genetically by Liu et al. to a highly conductive e-pili and excellent electron transfer behaviour was observed (Liu et al., 2019).

ELECTRON TRANSFER THROUGH MEDIATOR

The direct transfer mechanism (Gralnick and Newman, 2007) and “nanowire” or “pili” based transfer mechanism (Reguera et al., 2005) primarily depends upon a single layer of microorganisms or biofilm uniformly distributed on the surface of the anode electrode, so that the electrons transfer ability is limited. However, during the mediator dependent electron transfer mechanism, mediator (chemical compound internally secreted by the microbes or exogenous small molecules having redox functions) acts as a charge carrier and transfer electrons from intracellular space of microbes to the electrode surface (Gralnick and Newman, 2007). The *Klebsiella oxytoca* d7 exhibited both direct or nanowire based electron transfer and mediator based electron transfer and almost 60% of the total electron transfer was observed by the mediator secreted by the microorganisms during closed circuit condition of a bioelectrochemical system (Niu et al., 2020). Although initially direct electron transfer mechanism was detected from *Pseudomonas aeruginosa*; however mediator based electron transfer by using pyocyanin and pyorubrin was also observed (Gao et al., 2017; Hernandez and Newman, 2001)**.** A novel electrogenic bacterium, *Dietzia sp*. RNV-4 was investigated by Sacco et al. and mediated electron transfer mechanism was reported (Sacco et al., 2017). Other than direct electron transfer, *Shewanella oneidensis* MR-1 excreted riboflavin and flavin mononucleotide mediators and exhibited mediator based electron transfer (Marsili et al., 2008b, 2008a). *Listeria monocytogenes*, *Lactococcus lactis* and *Bacillus megaterium* also depends on flavin based mediator for electron transport (Light et al., 2018; Masuda et al., 2010; You et al., 2018). Electrical signal propagation through potassium channel was observed for *Bacillus subtilis* which plays significant role in biofilm formation (Prindle et al., 2015). *E. coli* is not capable for direct electron transfer or distant electron transfer. However, hydroquinone type endogenous compound produced by *E.coli* cells can act as a mediator for electron transfer (Qiao et al., 2008). 2,6-di-tert-butyl-p-benzoquinon (2,6-DTBBQ) produced from *Klebsiella pneumoniae* was observed as a mediator between electrode surface and *Klebsiella pneumoniae* (Deng et al., 2010).

CONCLUSION

Direct electron transfer, distant or nanowire based electron transfer and mediator based electron transfer are three major electron transfer procedures. Electrogens can follow a single pathway or a combination of multiple pathways to transfer electrons from microbe to electrode surface.

Geobacter sp. and *Shewanella sp.* are popularly known electrogens which follows all three types of electron transfer mechanisms. Although several other electrogens or electroactive microbes are also investigated such as *Desulfovibrio desulfuricans*, *Kluyvera georgiana* etc. However a significant research is still required to accurately identify the electron transfer mechanisms and electrogens to improve the performance of BES to use it for sustainable applications.

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