Electrochemistry and microbial ecology approach of anodic biofilms for the study of scaled-up microbial fuel cells

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Since 1911 when Potter described bacteria that could produce electrical currents, interest in this ability and associated technologies like microbial fuel cells (MFCs) has grown specially in the last 20 years. Many MFCs have an anaerobic chamber filled with the feeding solution where an anode and a cathode are immerged although the cathode can be in contact with air for the supply of oxygen. Biofilms develop on the anode and cathode after inoculation with bacteria (pure bacterial culture or from environmental sources such as wastewater). Some bacterial strains (*e.g.*, *Geobacter* species) are able to use the anode as their terminal electron acceptor. In the anodic biofilm, the stepwise and complex electron pathway from bacteria to the anode results in charge transfer resistance that can induce potential energy losses. The electrons travel via an external electric circuit to the cathode where the oxygen reduction occurs with or without bacterial participation.

The application of MFCs for the production of energy from wastewater requires the scale-up that does not yet produce an electrical yield comparable to lab-scale MFCs. Our work was based on the hypothesis that typical scale-up efforts modify specific MFC parameters (substrate diffusion, geometry, hydraulic forces, etc.) that in turn affect the anodic biofilm and the efficiency of electron transfer to the anode. We investigated the microbial ecology and electricity production of three MFCs of different volumes: 10 mL, 500 mL, 4 L operated under similar conditions and identical MFC parameters.

Electrical production, electrochemical characterization and microbial community structure (16S rRNA gene sequencing to evaluate the enrichment of electroactive bacteria species like *Geobacter sulfurreducens* and *Shewanella oneidensis MR-1*) and function (to examine electron exchange functions) were monitored during the MFC start-up and operation. In parallel, the physical structure of the biofilms (such as density, thickness, bacterial distribution and nanowire (conductive pili) presence) was studied by microscopy techniques and proteomics analysis.

In addition, we electrochemically characterized several MFCs with different architectures and electrode material by electrochemical impedance spectroscopy and voltammetry techniques. The aim was to model electrical behaviour and equivalent circuit of MFCs as batteries in relation with the identification of redox proteins involved in electron transfer in order to understand the limiting step of extracellular electron transfer to electrodes in anodic biofilms.