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Perspective

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Electrical Conductivity in Anaerobic Sludge from full-Scale Wastewater Treatment Reactors

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Introduction

Granular biofilms are common in anaerobic wastewater treatment reactors. In anaerobic granules, distances between microbial species involved in sequential degradation of organic matter are small (micron scale), which facilitates substrate degradation. Interspecies hydrogen transfer (IHT) is a key mechanism in anaerobic degradation, as hydrogen is formed from the oxidation of organic intermediates by fermentative microorganisms. The hydrogen is transferred to hydrogen-consuming microorganisms, in particular methanogens and sulfate reducing bacteria, thereby maintaining the hydrogen partial pressure below the critical thermodynamic limit for fermentative degradation of substrates. In addition to hydrogen, formate has also been reported as an electron carrier in anaerobic communities. Alternatively to interspecies H2/formate transfer, electrons can be transferred directly between microbial species by redox-mediating proteins or conductive appendages. Although most research on direct interspecies electron transfer (DIET) focused on defined co-cultures of Geobacter spp or Geobacter spp. with methanogenic archaea this mechanism has also been proposed to be relevant for granules in anaerobic digesters, especially granules grown on brewery wastewater. For example, Rotaru and co-workers suggested DIET between Geobacter and Methanosaeta spp. occurs in aggregates from a brewery wastewater digester and further demonstrated that Methanosaeta harundinacea is capable of directly accepting electrons from Geobacter spp. for the reduction of carbon dioxide to methane in a defined ethanol degrading co-culture.

Ability of Microorganisms

Electrical conductivity i.e. the ability to conduct electric current has been found for microbial cells and biofilm. Determining the electrical conductivity of microbial biomass can reveal the ability of microorganisms to transport electrons, which could make these microorganisms useful for technologies such as microbial fuel cells (MFCs) but also anaerobic digestion, with potential direct interspecies electron transfer within granular microbial aggregates. Electrical conductivity in methanogenic aggregates was measured for the first time by Morita and co-workers, who found a value of 6.1 µS cm-1 for anaerobic granules obtained from a brewery UASB reactor. Shrestha and co-workers reported electrical conductivities in granules from fourteen brewery UASB reactors, with values between 0.8 and 36.7 µS cm-1. These two studies correlated the measured electrical conductivity with DIET, and the authors suggested that DIET is a widespread phenomenon in granules grown in reactors treating brewery wastewater. Other studies proposed DIET as the predominant electron transfer mechanism in laboratory anaerobic digesters that manifested an enhanced methanogens rate when conductive materials (like carbon felt, carbon cloth or graphite) were added. These studies also showed that higher rates usually coincided with a shift of the microbial community and an increased electrical conductivity of the microbial aggregates.

To the best of our knowledge, electrical conductivity has only been studied and reported for anaerobic granular sludges treating brewery wastewater. However, it remains obscure how widespread this characteristic is in anaerobic granular sludges from other industries. In this study, we measured the electrical conductivity of anaerobic granular sludge obtained from a wide variety of industries: food and beverage, brewery, alcohol production, paper and textile, petrochemical and chemical. These sludges were predominantly methanogenic except one, which was predominantly sulfidogenic. Granules were also examined for their microbial community composition and other chemical and physical properties, such as their elemental composition or surface morphology. Before each test, carried out at room temperature, the electrode slide was cleaned with ethanol (70 %) and demi-water. Before conductance measurement, a layer of granules was gently flattened to form a homogeneous and confluent layer on top of the electrode slide, covering the whole surface area including the non-conductive gap. Two independent measurements (duplicates) were performed for each granular sludge. Media, the aqueous phase of the bioreactor from which the sludge originated, were also analyzed for electrical conductance and served as control (one measurement per medium). Media were centrifuged twice to eliminate suspended sludge particles.

Anaerobic Reactors

Chronoamperometry was used to measure conductance of granular sludges and media in a two-electrode set up. In this way, the potential applied during measurements corresponds to a cell voltage difference between one gold electrode serving as working electrode and the other gold electrode serving as counter electrode. This cell voltage can drive an electron flow from one electrode to another only when the sample crossing the non-conductive gap is electrically conductive. The measured current is then related to the applied voltage and can reveal the electrical conductance of the sample. Therefore, when a sample is not electrically conductive, there should be no current dissolved salts in media should only result in ionic conductivity. To ensure that current reached a steady-state after the decay of current transients due to surface charging effects. Additionally, an equilibration time was applied prior to each measurement in order to stabilize the initial current. The steady-state current obtained after each voltage step was used to plot current as a function of voltage. The slope corresponding to a linear regression line of this plot was used to calculate conductance, i.e. the measured current within a certain voltage range. Electrical conductance is used for the discussion of results within this study. However, in order to compare the results with those in the literature, it is necessary to use normalized values and convert conductance (in S) to conductivity (in S cm-1). This was done by using the Schwarz-Christoffel transformation which corrects for the non-uniformity in current distribution occurring in the sludge that is placed on the gold electrode slide. A paired-samples t-test was conducted to evaluate the differences in electrical conductance between duplicates. Additionally, the correlation between electrical conductance and both the relative abundance of microbial community (at genus level) and the concentration of chemical elements in granules was investigated by using Pearson product-moment correlation coefficients.

The other two sludges originated from anaerobic reactors at an alcohol distillery and an industry carrying out a propylene epoxidation process. Finally, one granular sludge showed a high conductance of 578 µS, corresponding to a conductivity of 171 µS cm-1, which is more than 4 times higher than the maximum reported for other industrial samples. This latter conductivity value is within the range of magnetite but lower than stainless steel or granular activated carbon all conductive materials that could possibly substitute biological connections in DIET. No significant differences were found between the measured duplicates, i.e. between the first groups of samples. No relation was found between electrical conductance of granules and the type of industry they originated from. The granular sludge with the highest conductance originated from a bioreactor at a fruit canning and juice production industry, while the second most conductive sludge was taken from a bioreactor fed with wastewater primarily containing benzoate and acetate. Granular sludge from only 4 out of 13 ethanol-related industries showed high electrical conductance, revealing that wastewater from such industries does not per se result in electrical conductance. Additionally, a wide range of electrical conductance of brewery granules was found, similar to a previous report. To conclude, our results showed that electrical conductance is not a generic characteristic of anaerobic granules, it is also not exclusively related to granules from the brewery industry, and it varies greatly between granules from bioreactors used in the same industry. As previously reported but also in other granule types such as those found in the food and beverage industry or the alcohol producing industry. However, no other correlation was found to be significant. These results are only indicative, as elucidating the mechanisms of DIET within anerobic granules is complex and may vary under different environmental conditions.