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參展科別 微生物學

作品名稱 **A Novel Procedure to Identify Genes involved in Electron Transfer of Exoelectrogens**

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Abstract

Purpose of research. Microbial fuel cells (MFCs) are bioelectrochemical systems that generate electrical energy by exploiting the extracellular electron transport (EET) capabilities of electrochemically active bacteria (EAB) (Logan 2009). This investigation aims to identify genes involved in driving bacterial EET with a new procedure that enables rapid screening of a side array of genes. These insights may lead to improved MFC performance through enhancing reactor design or genetic engineering EABs (Alfonta 2009).

Procedures.

MFC metagenomic analysis. Twelve MFCs incubated with four different bacterial samples were operated for approximately one year. The bacterial DNA from before and after incubation was extracted and the 16S rRNA regions were PCR amplified and sequenced. The bacterial community changes were analyzed using the QIIME program to identify bacteria that were being selected.

Fosmid Clone Isolation. An E. coli fosmid library (Mewis et al. 2013) that contained genes from EAB inferred in the previous step was incubated in three MFCs. After a 48 hour enrichment period, biofilm samples from the MFCs were extracted and individual clones were isolated and screened in the MFCs individually. An E. coli DH5α strain with no insert DNA was incubated separately as the control.

DNA sequencing. Fosmid insert DNA from high-performing clones were extracted, purified using gel electrophoresis, constructed into sequencing libraries and sequenced.

Bioinformatics Analysis. The sequences were constructed into larger contigs using the Velvet algorithm package. The open reading frames (ORFs) were inferred and translated

into amino acid sequences and annotated with proteins identified from the KEGG, and SEEDs databases using Metapathways 2.5.

Results. The changes in bacterial communities from the metagenomic analysis revealed increases in relative abundance in numerous genera from Firmicutes and Bacteroidetes. The MFCs incubated with the fosmid clones generated about 4 times more peak power than the MFCs incubated with the E. coli DH5α. Polarization curves generated for the MFCs demonstrated that the fosmid clones were able to sustain a higher current.

Incubation of pure cultures of individual clones yielded four clones with significant performance improvements over the control strain. Protein data from Metapathways outputs reveled both novel and previously reported EET genes encoding for Type IV pilus structures, c-type cytochromes, soluble cytochromes, flavoproteins, and porins. Taxonomy inferences of the gene inserts by the Green Genes database reveal the genes most likely came from the same EABs that were inferred from the metagenomic analysis.

Conclusions. The increased performance of the fosmid clone-powered MFCs suggest that the clones carried genes that enhanced their performance in the MFCs. This is further confirmed by polarization curves generated for the MFCs. The results of the taxonomy inferences suggest that the bacteria being selected for in the environmental samples carried genes that enhanced their performance in the MFCs, and that these genes were successfully identified in the subsequent steps. The results of this study demonstrate that using a gain of function approach to rapidly screen a wide array of genes in a gene library may be an efficient method to identify genes that enhance power generation of EABs in MFCs.

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Many novel genes that can enhance the performance in the MFCs have been identified in this study. The author should give a potential mechanisn to explain

Why the proteins encoded by the identified genes can promote the performance in the MFCs.

If is interesting to know whether the different Carbon source used for that enhance the performance in the MFCs.