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Permalink
https://escholarship.org/uc/item/3x8390xx

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Publication Date
2007-05-14
Energy Conservation Mechanisms for Syntrophic Growth of Desulfovibrio vulgaris and Methanococcus maripaludis


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ABSTRACT

In the absence of electron acceptors, many Desulfovibrio species grow on non-fermentable substrates via syntrophic association with hydrogen consuming methanogens. We examined the phylogeny of 168 Desulfovibrio Hildenborough syntrophically associated with Methanococcus maripaludis. We describe a novel mechanism of transcriptional up-regulation of electron transport genes in response to syntrophic growth. Global transcriptomic analyses were performed on cocultures grown on lactate/sulfate (wildtype) and lactate/sulfate with acetate (mutant). Genes involved in energy conservation of syntrophic growth were up-regulated and a putative carbon-monoxide induced hydrogenase (Coo, DVU2286-93) and a high-molecular weight cytochrome (Hmc, DVU0531-6) in energy conservation during syntrophic growth. Mutant monocultures grew to the same density on lactate/sulfate as the wildtype. The putative carbon-monoxide induced hydrogenase (cooL, DVU2286-93) and a high-molecular weight cytochrome (Hmc, DVU0531-6) in energy conservation during syntrophic growth. Mutant monocultures grew to the same density on lactate/sulfate as the wildtype. The putative carbon-monoxide induced hydrogenase (Coo, DVU2286-93) and a high-molecular weight cytochrome (Hmc, DVU0531-6) in energy conservation during syntrophic growth. Mutant monocultures grew to the same density on lactate/sulfate as the wildtype. The putative carbon-monoxide induced hydrogenase (Coo, DVU2286-93) and a high-molecular weight cytochrome (Hmc, DVU0531-6) in energy conservation during syntrophic growth. Mutant monocultures grew to the same density on lactate/sulfate as the wildtype.

CHEMOSTAT CONFIGURATION

BiOMASS & HEADSPACE GAS MEASUREMENTS

Chromatots are run using a 24 h rotation time at 37 °C and a stirring speed of 250 rpm. The headspace of the chemostat is flushed with a mixture of N₂ (90:10) at a rate of 0.20 - 0.50 ml/min. The headspace gas composition is sampled in 15 min. intervals using a Hiden QIC-20 mass spectrometer.

TRANSCRIPTIONAL ANALYSIS

Triplicate biological replicates of cocultures and sulfate-limited D. vulgaris monocultures were analyzed by the ESPP Functional Genomics Core using custom-designed whole-genome microarrays. Microarray slides were designed with duplicate spots for each open-reading frame (ORF) for both organisms. At least three slides were used for each biological replicate. The ESPP Computational Core calculated RNAseq expression ratios for each ORF and the log ratio comparing the coculture versus sulfate-limited monoculture growth conditions was determined. Z-scores for each ORF were calculated to determine statistical significance. Operon-based estimates of local connectivity indicate an absolute Z-score of 1.0 accurately predicts expression changes between the two conditions. Using this value, 189 ORFs displayed significant up-regulation. 254 ORFs were statistically down-regulated. Genes were assigned classes of orthorhombic groups (COG) functional codes based on previous genome annotations.

ENERGETIC OVERVIEW

Lactate $\rightarrow$ pyruvate + H₂
- Energetically unfavorable unless pyruvate and/or H₂ kept at low concentrations.
- Can acts to produce hydrogen (likely consumed by the methanogen) and translocate H₂ or Na⁺.
- H₂ serves to keep pyruvate concentration low, allowing continued lactate oxidation.

FUTURE WORK

- Developing synergetic growth pathway on other compounds, such as ethanol.
- Refine metabolic interaction model between D. vulgaris and M. maripaludis, using transcriptional, genomic and phenotypic analyses.
- Explore salt, nitrate, pH, etc., stress response of coculture.
- Compare responses with other organisms, especially other Desulfovibrio strains.

ACKNOWLEDGMENT

ESPP is part of the Virtual Institute for Microbial Stress and Survival supported by the U.S. Department of Energy, Office of Science, Office of Biological and Environmental Research, Genomics-GRS Program through contract DE-AC05-96R12981 between Lawrence Berkeley National Laboratory and the U.S. Department of Energy. The box of Rf and Rm mutants were kindly donated by Dr. G. Voordouw, U of Calgary. M. maripaludis was kindly donated by Dr. J. Leigh, U of Washington.