Title
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Publication Date
2010-09-01
Fumarobacterium G20 and the Effect of Formate

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Abstract

The anaerobic sulfite-reducing bacteria (SRB) of the genus Desulfovibrio are found in a remarkable variety of habitats, including hot springs and salt water environments. MSB3061 allows them to metabolize formic acid, a common, though volatile, precursor and through changing the redox state of the metal and thus its volatility. In studying the native products of Desulfovibrio G20 during growth on various media, we discovered a potential outcome under the strain's less known metabolic pathways. When Desulfovibrio G20 anaerobic growth on fumarate appears to occur in a distribution with the primary end products being acetic and malic acids at approximately the theoretical ratio of 2:1, respectively. The initial growth with formate was inhibited with additions of 13 H2 and 5 mM formate. In wildtype G20, formate may inhibit growth directly by serving as an energy pathway or by downregulating the genes encoding the enzymes involved in the formate growth. Therefore, the inability of the cytoplasm to grow on fumarate might result from an interruption in another level at the level of CO2 fixation. To determine if the presence of a regulatory effect on this pathway, quantitative RT-PCR experiments were performed in N.25 PCR experiments was performed to determine if the presence of the gene is that observed.

Figure 3: Growth on Fumarate Medium of Desulfovibrio G20 and Three Formate Dehydrogenases Transposon Mutants

A. Subcultured from Lactate/Sulfate (60/30mM)

B. Subcultured from Fumarate (60mM)

Transposon mutants exhibited longer lag time versus wildtype before initiating growth on fumarate, regardless of the source of the inoculum. Ultimately, the wildtype and mutants exhibited final growth yields within 19% of each other. No unique trend in the end products was observed in the mutants compared to wildtype.

Figure 4: Inhibition of Growth of Desulfovibrio G20 on Fumarate by Formate

The presence of three formate dehydrogenase transposon mutants is tested in the study (Figure 3A, B, C and D). The 203 and 3.4.2 (Fig. 3D), respectively, were identified in the fumarate (Figure 4) and 3.4.2.2 (Fig. 3D) of the complex. Owing to the gene annotations are from http://www.ncbi.nlm.nih.gov.

Figure 5: Desulfovibrio Growth on Fumarate (60mM) with H2, H2, or CO2 in Headspace

A. Formate (0mM, 2mM, 5mM, and 15mM) added at Time 0

B. Formate (0mM, 2mM, 5mM, and 15mM) added at 15.5 hours

C. CO2 added at 0.5 gl/min

D. Formate added at 0.5 gl/min

Figure 6: Proposed Model for the Growth of Desulfovibrio on Fumarate (60mM) and possible Inhibitions

Summary

- G20 grows by dominance of formate producing the theoretical ratio of 2.5 acetate/malate endproducts.
- Formate dehydrogenase mutants grow more slowly than wildtype G20 on fumarate with Fed-A (Fig. 4C, D). Ising delayed.
- Growth of G20 on fumarate is inhibited by formate, H2, and CO2.
- G20 was produced during growth of CO2 on fumarate.
- The H2 inhibition may be due to inability to retain ferredoxin or a possible blockage of pyruvate pim惺ing allowing only the eight reduction of ferredoxin to occur.

Future Plans

- Compare (b) growth to wildtype on fumarate with formate, H2, and CO2
- Perform RT-PCR on genes in fumarate and formate dehydrogenase operons during different growth conditions
- Microarray analysis and proteomic analysis on fumarate-grown G20 cells are currently underway.

Acknowledgements

This work was supported by U. S. Department of Energy, Office of Science, Office of Biological and Environmental Research grant DE-AC02-05CH11231, and DOE (DE-AC02-05CH11231) and as a result of the Great Lakes Program supported by U. S. Department of Energy, Office of Science, Office of Biological and Environmental Research grant DE-AC02-05CH11231, Lawrence Berkeley National Lab. and Oak Ridge National Lab.