ABSTRACT

TITLE:	Glucose Metabolism in Acidobacterium Capsulatum Studied by 1H-NMR and In Vitro Studies of Cell Lysates and Cloning of a Putative Transaldolase Gene
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ABSTRACT: In 1991 the acidophilic bacteria, Acidobacterium capsulatum was cultured from acid mine drainage in Japan. This microorganism is of interest because there is little known about its metabolic capabilities. Ward et al. (2009) showed in their annotated genome of A. capuslatum the absence of a key enzyme, aldolase, involved in the breakdown of glucose in glycolysis. If this is true, then glucose has to be shunted between use for energy production and biomass through alternative pathways, most likely the pentose phosphate shunt (PPS) which could use transaldolase as a key controlling enzyme. A recent paper by Thompson et al. (2011) suggests that transaldolase is a key enzyme involved in shunting glucose from the Calvin cycle to the PPS when marine cyanobacteria are infected with cyanobacteria phage. To test our hypothesis that transaldolase is a key enzyme in controlling carbon flux in A. capsulatum, we have used a multipronged approach to study glucose metabolism in A. capsulatum. These experiments include 1H-NMR analysis of water soluble metabolites from cell lysates, enzyme activity assays from cell lysates and cloning a gene for a putative transaldolase. Our experiments demonstrate that A. capsulatum does indeed consume all glucose provided in growth media under aerobic conditions. Under these conditions, the cells reach log phase in a 70 to 90 hour period after inoculation. 1H-NMR of water soluble metabolites extracted from cells in log phase versus cells in stationary show distinct differences in metabolite profiles. Malate dehydrogenase assays show that we can lyse the cells and preserve enzyme activity and furthermore show that malate dehydrogenase is active in log phase. If enzyme activity is preserved during cell lysis, we plan to do more complex assays for other key enzyme activities such as aldolase and transaldolase. To further understand the role of transaldolase in A. capsulatum we are cloning the putative transaldolase gene into E. coli for expression and characterization. As a whole, we have been able to characterize A. capsulatum in aerobic conditions with glucose as the sole carbon source, as well as develop a lysis procedure that maintains enzyme activities in vitro. These methods will allow for future studies that will provide more information about glucose metabolism in A. capsulatum.