A326 - METABOLIC PROFILING OF SECRETED FERMENTATIVE BYPRODUCTS BY SALMONELLA TYPHIMURIUM

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Abstract:

Salmonella typhimurium is a pathogenic bacterium that has been studied to be used as a platform for vaccine development as a live bacterial vector (LBV). It is closely

related to other enterobacteria like E. coli, but presents some metabolic differences regarding the efficiency of biomass and energy production. This work presents a metabolic profile of the main byproducts excreted by S. typhimurium aiming to evaluate the distribution of carbon between respiratory and fermentative pathways for this bacterium. A genome scale metabolic model (GSMM) of S. typhimurium was also used to simulate the phenotypic behavior of this bacterium under the studied conditions and compare the experimental results to the model predictions. Simulations were performed using the free software Optflux and biomass maximization was used as the objective function for Flux Balance Analysis (FBA). Experimental cultivations were conducted in 2 L bioreactors with a working volume of 0.8 L under varying dilution rates (D) of 0.1, 0.24, 0.5, 0.58 and 0.67 h-1, using 10.0 g/L of glucose as the sole carbon source and M9 minimum medium. The dissolved oxygen concentration was kept at 30 % of its saturation, and pH and temperature were kept constant at 7.0 and 37°C, respectively. Supernatant samples were analyzed by HPLC to measure glucose consumption rate and fermentative products formation. Our initial results showed an intense production of organic acids by S. typhimurium, mainly acetic acid, at increasing amounts for higher dilution rates, and starting from the lower dilution rate studied. From D of 0.5 to D = 0.67 h-1 formate was also produced, and the secretion rates of both by-products increased linearly with the growth rate. These results may reflect some metabolic limitations of the species under investigation concerning its respiratory capacity, as the fermentative pathways seem to be active even under low growth rates and aerobic conditions, what is not observed for E. coli. It is also noteworthy that a considerable fraction of the carbon consumed is directed to the formation of these metabolites, compromising the biomass formation. Simulation results employing the free software Optflux 3.0.7 and the GSMM STM_v1.0 predicted some aspects of the fermentative behavior observed in the experiments, but with low accuracy, indicating that the model employed (that was constructed based on an E. coli model) does not reflect the differences between the metabolism of these two closed related bacteria. Results from this work will be incorporated in the model to increase its predictive capacity regarding the organic acids formation by Salmonella cells.

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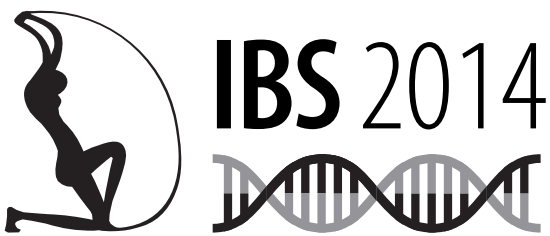

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