Organic acid production by Aspergillus niger

The environmental impetus to move away from traditional chemical techniques towards biologically based production has revealed Aspergilli as very attractive cell factories. Aspergillus niger is particularly interesting for the production of organic acids, as it tolerates low pH, may ensure high conversion rates of the substrate to the metabolite of interest, can give high conversion yields and is already used extensively for production of organic acids (e.g. production of citric acid). All these factors should make Aspergillus niger an ideal candidate for metabolic engineering. However, the application of metabolic engineering for production of high concentrations of different organic acids has been limited by the lack of detailed knowledge on the central carbon metabolism and its regulation. The aim of this Ph.D. was therefore to develop and apply the necessary metabolic engineering tools for the study, and eventual manipulation, of the central carbon metabolism towards enhanced organic acid production. Several techniques related to enhanced genomic manipulation, intracellular metabolite profiling and fermentation were either developed specifically in this work, or transferred from work on other organisms. The application of metabolic engineering to A. niger allowed for the development of several strains with enhanced citrate production capabilities. The mechanisms underlying the increased productivities were also carefully investigated and described in detail. Finally, a proof-of-concept application of a novel reversible auxotrophic marker system was demonstrated through the deletion of a global acetate regulatory gene (acuB). The effects of this deletion on organic acid production are also discussed.