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Fed-batch process development using metabolically efficient CHO cells

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FED-BATCH PROCESS DEVELOPMENT USING METABOLICALLY EFFICIENT CHO CELLS

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The development and application of fed-batch strategies have greatly contributed to the improvement of the productivity of cell culture processes. However, in such processes, lactate and ammonia accumulation in the culture medium over time can have detrimental impacts on cell growth and product quality. Continuous cell lines typically exhibit an inefficient metabolism whereby most of the pyruvate derived from glucose is diverted to lactate and only a small percentage is incorporated into the TCA cycle. Thus, further process improvements can be expected by combining the application of rational fed-batch strategies with targeted metabolic engineering of cells to reduce waste metabolite accumulation.

We have previously reported the establishment of a PYC2-expressing CHO cell line that exhibits a significantly altered lactate metabolism compared to its parental cell line; the ensuing reduction in waste metabolite accumulation is associated with a prolonged exponential growth phase, leading to significant increases in both maximum cell density and product titer in batch culture.

In this work, we demonstrate further process enhancements by cultivating the PYC2-expressing cells in fed-batch mode. Unlike the parental cell line, even under high nutrient levels, the engineered cells maintained their highly efficient metabolism characterized by low lactate\glucose and ammonia\glutamine molar ratios. This effectively alleviates the need to control substrates at low levels to avoid waste accumulation, a strategy which can negatively impact product quality. And unlike nutrient substitutions which are often accompanied by cell growth reduction, the decrease in lactate accumulation is associated with an increased in maximum cell density translating into a net gain (20%) in final volumetric productivity. To further characterize the process, isotopic tracer studies were conducted to analyze the fate of the main nutrients and various product characterizations were performed to assess the impact of this cellular engineering approach on antibody quality.