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Development of a sustainable bioprocess to produce Vitamin B_9

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Abstract:

The demand for natural derived products is growing. Nutraceuticals such as pre- and post-biotics, antioxidants molecules and vitamins are prominent examples in this scenario.

Folate (Vitamin B_9) is a water-soluble B vitamin with important roles in nucleic acid synthesis, repair and methylation, naturally produced only by green plants and some microorganisms: for these reasons it represents an essential nutritional component for humans. Vitamin B_9 commercially available is chemically synthesized in folic acid form; the production of natural folates by microbial fermentation is therefore becoming a sustainable alternative for human supplementation.

In the present work, *Saccharomyces cerevisiae* was engineered in the anabolic pathways of the two main building blocks of folate, para-aminobenzoic acid (pABA) and dihydropteridine, to identify the impact of these modifications on the production of intra- and extracellular levels of free and poly-glutamate folates. In parallel, we evaluated for the first time the production of folate in shake flasks exploiting another natural producer, the yeast *Scheffersomyces stipites*. We optimize the growth on minimal (Verduyn) synthetic medium to obtain a direct comparison between the folate productions of the 2 yeasts.

S. stipitis was able to produce higher amounts of folate, if compared to the well-known yeast cell factory *S. cerevisiae*. Moreover, interesting results were obtained on Verduyn minimal medium with co-presence of glucose and xylose, proving that this unconventional yeast has a greater flow towards the shikimate pathway than *S. cerevisiae*. These results provide a solid starting point for setting up bioreactor fermentations to increase and exceed the production titers reported in literature to date.