THE USE OF MICROORGANISMS TO PRODUCE VITAMINS

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Introduction. Vitamins are a group of essential nutrients that are necessary to maintain normal metabolic activities and optimal health. There are wide applications of different vitamins in food, cosmetics, feed, medicine, and other areas. The increase in the global demand for vitamins has inspired great interest in novel production strategies. Chemical synthesis methods often require high temperatures or pressurized reactors and use non-renewable chemicals or toxic solvents that cause product safety concerns, pollution, and hazardous waste. Microbial cell factories for the production of vitamins are green and sustainable from both environmental and economic standpoints. Traditionally, vitamin production strains have been improved through mutagenesis and metabolic engineering, which can be conducted either through chemical or biological means [1]. The main chemical strategies include chemical mutagenesis, application of N^+ ion beam, ultraviolet radiation or laser mutagenesis. The biological methods mainly include the construction and mutagenesis of the starting strain, genetic modification, synthetic biotechnology, optimization of media and culture conditions, construction of biofilm reactors, etc [2].

The aim of the study. To study the ability of microorganisms to synthesize a number of vitamins, to consider promising directions in the development of microbiological methods for more productive vitamin production.

Methods of research. The scientific, popular science literature on the study of micro-organisms and their importance in medicine and pharmacy has been used.

Main results. At present, the fermentation method has been recognized by researchers, and it is more environment-friendly and safe than chemical methods. As the fermentation technology matures, this approach is increasingly being used in industry to increase the production of different vitamins. For example, fermentation processes for the production of vitamin B_2, vitamin B_{12}, vitamin C, and vitamin K_2 have all been industrialized successfully.

Thiamine biosynthesis results from the coupling of the pyrimidine and the thiazole moieties to form thiamine phosphate [3]. *Escherichia coli, Salmonella typhimurium*, and *Bacillus subtilis* are the most thoroughly studied thiamine production organisms.

The two most important industrial producers Vitamin B_2 are *Ashbya gossypii* and *B. subtilis*. In *A. gossypii*, malate synthase in the glyoxylate cycle is essential for riboflavin production [4].

Recent reports describe the use of recombinant *E. coli* expressing *Rhodococcus rhodochrous* nitrile hydratase for vitamin B_3 production.

Sahm and Eggeling adopted a series of methods to increase the production of pantothenic acid, including the deletion of the ilvA gene and the overexpression of the ilvBNCD and panBC genes. The pantothenic acid production of the best strain reached 1000 mg/L. Huser also used *Corynebacterium glutamicum* to produce pantothenic acid.

From large-scale screening studies of different strains, found that the Gram-negative bacterium *Sinorhizobium meliloti* is the best producer of vitamin B_6. *E. coli* and *B. subtilis* were also engineered to produce vitamin B_6.

Some microorganisms can overproduce biotin, which has been elaborated in *C. glutamicum*, *Mesorhizobium loti*, and *S. meliloti* [5].

With the continuous research progress, *A. gossypii* has attracted increasing interest as the chassis strain for folic acid production. *A. gossypii* can synthesize 0.04 mg/L of folic acid naturally, which can reach 6.59 mg/L after metabolic engineering treatment [6]. Vitamin B_{12} is synthesized by microorganisms through de novo synthesis or salvage synthesis in nature, but higher-animals and plants cannot produce it [7]. *Pseudomonas denitrificans* and *Propionibacterium freudenreichii* being widely used in industrial fermentation to produce vitamin B_{12}.

At present, L-AA is manufactured via the classic seven-step Reichstein process using d-glucose as the initial substrate. The process involves six chemical steps and one fermentation steps for the oxidation of D-sorbitol to 2-keto-l-gulonic acid by *Gluconobacter oxydans* and *Bacillus megaterium.* Sugisawa reported for the first time that *Ketogulonigenium vulgare* can produce 1.37g/L of L-AA under static culture conditions. Kim reported that the respective enzymes from *Candida albicans* and *S. cerevisiae* convert not only d-arabinose to d-arabinono-1,4-lactone but also l-galactose to l-galactono-1,4-lactone *in vitro* [8].

Conclusions. The fermentative production of vitamins using bacteria, yeasts or microalgae has many advantages over traditional chemical synthesis methods. From the aspects of safety, biological activity, absorption rate, etc., vitamins manufactured by biological methods can be more suitable for both internal and external applications [9]. Although the fermentation of B_2 and B_{12} has technologically matured and is being applied in industrial production, fermentation methods for the remaining B-group vitamins have yet to be developed or require significant yield improvement.

References

1. Vandamme E.J., Revuelta J.L. (2016) “Industrial fermentation of vitamin C”, in Industrial Biotechnology of Vitamins, Biopigments, and Antioxidants. 161-192.

2. Song J. et al. (2014) Enhanced production of vitamin K_2 from Bacillus subtilis (natto) by mutation and optimization of the fermentation medium. Braz. Arch. Biol. Technol. 57. 606-612.

3. Cea P.A. et al. (2020) Characterization of hydroxymethylpyrimidine phosphate kinase from mesophilic and thermophilic bacteria and structural insights into their differential thermal stability. Arch. Biochem. Biophys. 688:108389.

4. Schwechheimer S.K. et al. (2016) Biotechnology of riboflavin. Appl. Microbiol. Biotechnol. 100 2107-2119.

5. Lin S., Cronan J.E. (2011) Closing in on complete pathways of biotin biosynthesis. Mol. Biosyst. 7 1811-1821.

6. Serrano-Amatriain C. et al. (2016) Folic acid production by engineered *Ashbya gossypii*. Metab. Eng. 38 473–482.

7. Fang H. et al. (2017) Microbial production of vitamin B_{12}: a review and future perspectives. Microbial Cell Factories 16:15.

8. Ma Q. et al. (2019) Integrated proteomic and metabolomic analysis of a reconstructed three-species microbial consortium for one-step fermentation of 2-keto-L-gulonic acid, the precursor of vitamin C. J. Ind. Microbiol. Biotechnol. 46. 21-31.

9. Yuan P. et al. (2020) Metabolic engineering for the production of fat-soluble vitamins: advances and perspectives. Appl. Microbiol. Biotechnol. 104. 935-951.