

Progress of Bioactive Particles and Biocatalysts

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Introduction

Over the past ten years, there has been an increase in interest in the molecules produced by bioprocesses, which has fueled the growth of biocatalysis in a number of industrial fields. Enzymatically created molecules have a high added value and are regarded as natural agents. Enzymatic processes also enable waste generation to be reduced, supporting sustainable development. However, work is still needed to create biocatalysts with high stability, a longer half-life, and a lower economic impact in order to expand the use of these catalysts in industry. The field of biocatalysis is currently interested in topics such as directed evolution to enhance the properties of enzymes, enzymes to carry out novel reactions found nowhere else in nature, and the development of environmentally friendly technologies and processes, primarily through the value of renewable resources [1].

The search for novel enzyme immobilization techniques and supports, as well as the use of protein engineering, are examples of enzyme-improvement strategies that result in biocatalysts with improved properties. Immobilization can be a very effective tool to increase enzyme stability and expand the range of applications for enzymes in industrial settings when it is properly designed. The choice of the carrier matrix and immobilization technique is crucial in this regard. Protein-engineering methods make it possible to change the properties of enzymes, enhance reaction performance metrics, and broaden the use of biocatalysts in new reactions. New kinds of enzyme-catalyzed reactions, the development of affordable enzyme synthesis methods to increase the use of enzymes in bioprocesses, and the search for enzymes with extra capabilities, like catalytic properties, are the needs that will determine the future of biocatalysis [2].

Description

Ones that are sexually active and have the potential to catalyze additional reactions. The pharmaceutical, food, and dermo-cosmetic industries are particularly interested in naturally occurring compounds known as biologically active molecules. These molecules are typically single stereoisomers of chiral compounds made by living things and cannot be created artificially. In this sense, biocatalysis provides a wide variety of readily available enzymes to carry out difficult reactions with excellent yield and selectivity [3]. Because they are environmentally friendly and have positive effects on human health, bioactive molecules produced through biocatalysis can enhance quality of life by preventing and treating a variety of chronic diseases. Real advancements in sustainability demand a move toward a more circular production model using renewable biomass. The utilization of different substrates Acetoin plays a significant flavoring role in the traditional Chinese fermented soybean product known as "douchi." The aldC gene for -acetolactate decarboxylase was cloned from *Lactococcus* (*L.*) *lactis* NZ9000 and overexpressed through nisin induction. An enzyme activity of 35.16 mU was produced by the ALDC crude enzyme solution. Next, acetoin was produced in GM17 medium using whole cells of the recombinant strain NZ9000/pNZ8048-aldC as the catalyst. According to an optimization experiment, the ideal induction conditions for producing the most acetoin (106.93 mg/L) were

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an initial OD600 of 0.6, an initial pH of 7.5, a concentration of nisin of 20 ng/mL, a temperature of 37°C, and static induction for 8 hours. Finally, whole-cell biocatalytic activity of NZ9000/pNZ8048-aldC was achieved following incubation under the optimal induction conditions.

Production of acetoin using soybean as the feedstock. The highest yield of acetoin was 79.43 mg/L. To our knowledge, this is the first study in which whole cells of the recombinant *L. lactis* are used as a biocatalyst to produce acetoin in soybean [4]. The aldC gene is overexpressed in *L. lactis*. Our research thus offers a theoretical foundation for the production of fermented foods with high acetoin content as well as for the biosynthesis of acetoin in food components. A biocatalyst transforms a raw chemical compound into a structurally related, high-value-added product during the process of biotransformation, which is frequently mediated by purified enzyme and whole cells. The disadvantages of traditional chemical synthesis over biotransformation include low specificity, harsh environments, difficult purification processes, and byproduct formation. The enzyme-containing cells were taken out and used in the whole-cell biotransformation, which can do away with the need for the enzyme to be separated and purified and thus lower production costs. The structures of enzymes were more stable and the enzymes were more effective when the cell membrane was present to protect them. Cofactors that the cells already produce can also be used in whole-cell biotransformation. Due to its benefits, which include easy application, high titers, and mild reaction conditions. With the exception of the fact that one sample of the recombinant bacterium NZ9000/pNZ8048-aldC was induced by 10 ng/mL nisin and the other was not, two samples were cultured, harvested, and disrupted as previously mentioned. The supernatant was centrifuged to obtain a crude enzyme solution, and acetoin production was used to gauge the enzyme's activity. The production of 1 mol of acetoin per minute is considered to be one unit of enzyme activity [5].

Conclusion

In a nutshell, 200 liters of crude enzyme solution and 200 liters of freshly made acetolactate solution were combined and heated to 30°C for 20 minutes before reacting. At room temperature for 50 minutes, the 400 mL of acetoin produced in the solution reacted with 4.6 mL of chromogenic agent to produce a pink color. And using acetoin as a reference, its concentration was determined at 522 nm. The optical density (OD522) vs. the acetoin concentration was used to plot the acetoin standard curve. Instead of using crude enzyme solution in parallel experiments, 200 L of MES buffer (0.05 M 4-morpholineethanesulfonic acid, 0.05% Brij-35, pH 6) was used, and the resulting OD522 value was used for zeroing. For precise measurement of the acetoin production capacity of recombinant bacteria in a whole-cell biotransformation system, the substrate (-acetolactate) to bacterial biomass ratio is critical. NZ9000/pNZ8048-aldC was divided into 10 mL/tube after being incubated statically at 30°C until the OD600 reached 0.4. Each tube received a final dose of 20 ng/mL nisin before being incubated at 30°C for 4 hours. As the whole-cell biocatalyst to produce acetoin, the cells were collected by centrifugation (10,000 rpm, 10 min, and 4°C), twice washed with PBS buffer, and resuspended in 24 mL PBS. The whole-cell biocatalyst was then harvested in fractions of 1/4, 1/8, 1/16, 1/32, 1/64, and 1/128 by centrifugation. 3 mL of the previously mentioned -acetoacetate solution was combined with each cell sediment.

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Conflict of Interest

There is no conflict of interest by author.

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