

Date: 29th September-2026

**EFFECT OF DIFFERENT LIPASES ON THE REDUCTION OF DRY
MATTER DURING ALCOHOLIC FERMENTATION**

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INTRODUCTION

Alcoholic fermentation is one of the key technological stages in the production of ethanol. During this process, fermentable carbohydrates are converted by yeast mainly into ethanol and carbon dioxide. The efficiency of fermentation depends on substrate composition, sugar concentration, temperature, pH, fermentation duration, yeast physiological state and other technological factors [1–3]. Previous studies have shown that optimization of fermentation conditions can significantly influence ethanol productivity and the overall efficiency of the process [1,2].

The reduction of dry matter content is one of the important technological indicators for evaluating the progress of alcoholic fermentation. As fermentable carbohydrates and other components of the wort are consumed or transformed by microorganisms, the dry matter content generally decreases over time. Therefore, monitoring the dynamics of dry matter reduction can provide information about the intensity and approximate completion of fermentation [1,3].

Lipolytic enzymes are primarily involved in the hydrolysis of lipids and can influence the composition and physicochemical properties of fermentation media. Changes in lipid metabolism are also associated with yeast physiology and cellular adaptation during fermentation [4]. In addition, enzyme-mediated transformations may influence the formation of fermentation metabolites and the utilization of components present in the fermentation medium [5].

The application of enzyme preparations in alcoholic fermentation is therefore of practical interest for improving fermentation kinetics. The present study aimed to investigate the effect of different lipases on the dynamics of dry matter reduction during alcoholic fermentation and to compare the fermentation behavior of control and enzyme-treated samples.

Materials and Methods

The experiment was conducted using fermentation wort prepared under identical technological conditions. Four experimental variants were studied: a control sample without additional lipase treatment, a sample containing pancreatic lipase, a sample containing rice bran lipase, and a sample treated with lipase produced by *Aspergillus niger*.

At the beginning of the experiment, the dry matter content in all variants was standardized at 20.1%. Equal initial dry matter content allowed the influence of the different



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lipase preparations on the subsequent fermentation dynamics to be compared under similar experimental conditions.

The fermentation process was carried out for 72 h. Samples were analyzed at different time intervals, and the dry matter content was determined throughout the fermentation period. The obtained data were used to evaluate the rate of dry matter reduction and to identify differences among the experimental variants.

The dynamics of dry matter reduction were presented graphically. The control sample was compared with the three enzyme-treated variants. The main criterion for comparison was the time required for a substantial reduction of dry matter and the approximate completion of substrate utilization.

Results and Discussion

The dynamics of dry matter reduction during alcoholic fermentation demonstrated clear differences between the experimental variants (Figure 1). At the beginning of fermentation (0 h), the dry matter content was identical in all samples and amounted to 20.1%. This confirms that the experimental variants were established under comparable initial conditions.

As fermentation progressed, a gradual reduction in dry matter content was observed in all variants. This decrease can primarily be attributed to the utilization of fermentable carbohydrates by yeast and their conversion into ethanol and carbon dioxide. Similar relationships between substrate utilization and fermentation efficiency have been described in studies of industrial ethanol fermentation. Yeast fermentation performance is strongly influenced by substrate availability and physiological conditions, and these factors determine the rate and duration of ethanol production.

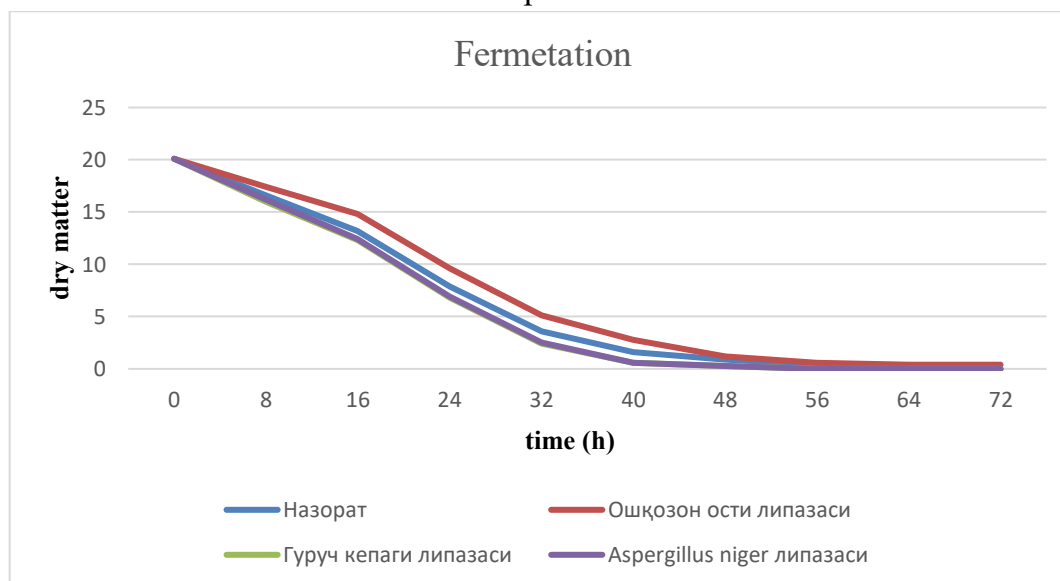


Figure 1. Time-dependent changes in dry matter content during alcoholic fermentation

During the period from 8 to 24 h, the lipase-treated samples demonstrated a faster reduction in dry matter compared with the control sample. The most pronounced changes were observed in the samples containing rice bran lipase and *A. niger* lipase. In these

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variants, the dry matter content decreased to approximately 6.8–6.9%, whereas the corresponding value in the control sample was approximately 7.9%.

The observed differences indicate that the presence of lipase preparations was associated with more intensive changes in the fermentation medium during the early and middle stages of fermentation. Lipid metabolism is an important component of yeast physiology, and changes in lipid composition can affect cellular processes and adaptation to fermentation conditions. Therefore, the observed effect of lipases may be related to modification of lipid-containing components of the substrate and changes in the physicochemical environment of the fermenting wort.

During the 32–48 h period, the enzyme-treated variants continued to demonstrate a rapid decrease in dry matter content. Particularly pronounced changes were observed in the samples containing rice bran lipase and *A. niger* lipase. By approximately 56 h, the dry matter content in these two variants reached 0.0%, indicating that the readily available fermentable substrate had been practically exhausted.

In contrast, the control sample reached a comparable endpoint only during the 64–72 h period. Thus, the application of rice bran lipase and *A. niger* lipase was associated with an approximately 8–16 h earlier reduction of dry matter to the final level compared with the untreated control.

The pancreatic lipase variant demonstrated a comparatively slower fermentation pattern. Even after 72 h of fermentation, the dry matter content remained at approximately 0.4%. This result indicates that, under the experimental conditions, pancreatic lipase had a weaker effect on the dynamics of dry matter reduction than rice bran lipase and *A. niger* lipase.

The differences among the experimental variants can also be considered in relation to the general kinetics of alcoholic fermentation. Previous research has demonstrated that fermentation efficiency is affected by substrate concentration, fermentation time, temperature, pH, yeast strain and other technological parameters. Therefore, the faster reduction in dry matter observed in the rice bran lipase and *A. niger* lipase variants may indicate more intensive utilization of available substrates under the experimental conditions.

At the same time, dry matter reduction should not be interpreted as a direct equivalent of ethanol production. Dry matter includes not only fermentable sugars but also proteins, lipids, minerals and other soluble or suspended components. Consequently, a reduction in dry matter can reflect several simultaneous biochemical and physicochemical processes.

The potential relationship between lipolytic activity and fermentation metabolites is also of technological interest. Recent research has demonstrated that lipid-derived compounds and enzymatic processes can participate in the formation of fatty acid ethyl esters and other fermentation-related compounds. This indicates that changes in the lipid fraction of a fermentation medium may influence not only fermentation kinetics but also the composition of secondary products.

The results obtained in the present study therefore suggest that rice bran lipase and *A. niger* lipase may have potential for further optimization of alcoholic fermentation. However,



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additional experiments are necessary to determine the optimum enzyme dosage and to establish whether the accelerated reduction in dry matter is accompanied by increased ethanol yield and desirable changes in fermentation by-products.

Conclusion

The experimental results demonstrated that different lipase preparations affected the dynamics of dry matter reduction during alcoholic fermentation.

All experimental variants showed a gradual decrease in dry matter content from the initial value of 20.1%. However, the rate of reduction differed considerably among the treatments. The most pronounced effect was observed in the variants containing rice bran lipase and *Aspergillus niger* lipase.

During 8–24 h of fermentation, dry matter content in the rice bran lipase and *A. niger* lipase variants decreased to approximately 6.8–6.9%, compared with 7.9% in the control sample. By approximately 56 h, dry matter content in these two enzyme-treated variants reached 0.0%, whereas a comparable level was observed in the control only during 64–72 h.

The pancreatic lipase variant demonstrated a slower fermentation pattern, with approximately 0.4% dry matter remaining after 72 h.

Thus, under the studied experimental conditions, rice bran lipase and *A. niger* lipase were associated with faster reduction of dry matter and earlier completion of the fermentation process. These findings indicate the potential of selected lipase preparations for further investigation as tools for improving fermentation kinetics and reducing fermentation duration.

Further studies should evaluate the optimum enzyme concentration and investigate the relationship between dry matter reduction, ethanol concentration, yeast biomass, residual sugars, pH, acidity and volatile fermentation by-products.

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