

Alcohol Production Technology: Possible Quality Improvements

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Abstract. Rectified alcohol is widely used in the food industry. Growing need for it means an increasing demand for higher-quality alcohol, which makes it competitive in the international market. One important effort to improve the quality of products consists in intensifying the processes relating to fermentation and yeast generation. This paper presents the results of researching the effects of an enzyme complex on the saccharification and yeast generation process. Research has been carried out at the Department of Fermentation Technology, North-Caucasian Mining-Metallurgical Institute (State Technical University). The following has been involved in the research: starch-containing raw materials (maize), brewing yeasts (Fermiol DY 7221), and enzyme preparations (Alfaferm 350L, Glucozyme A 400S, and Protoferm FP). Pursuant to the rules of technochemical control of alcohol production, the quality of semi-products and final products was controlled at each stage of research. The experiment was carried out in consistence with the basic stages and parameters of alcohol production: preparing the raw materials for cooking, cooling down the cooked mass, saccharification of start by means of enzyme preparations, fermenting the sugars by means of yeasts, alcohol distillation from wort, and subsequent alcohol rectification. During the experiment, we altered the concentration of Glucozyme A 400S while the concentration of the other enzyme preparations remained unaltered. Fermentation degree is one of the main indicators of the enzymatic effects. Based on the experiments, we have found the optimal amount of Glucozyme A 400S that maximizes the output of high-quality products while reducing the duration of fermentation by 10 to 12 hours (60 hours).

1. Introduction

Processing plant raw materials into ethanol is one of the most efficient uses of such materials. The main advantage of plant biomass as a raw material is that it is processed by eco-friendly technologies based on natural processes and mechanisms where conversion is done by microbial enzymes and brewing yeast.

Both in Russia and worldwide, ethanol makes for an ever-greater share of the total product volume. This is primarily due to the expanding use of this product as a biofuel.

As of today, ethanol is costly to produce. Lack of comprehensive substrate processing means no reduction in costs. It is therefore necessary to improve the production technology. One of the ways to do so consists in optimizing the enzymatic hydrolysis of the substrate by using enzyme preparations for further yeast generation and fermentation.

As it is known, yeast generation and fermentation requires nutrients to be present in the saccharified wort. The concentration of nutrients in the wort depends on the enzyme preparations in use.

Fermiol yeast is a popular choice today. It is good for fermenting maltose, sucrose, and fructose, but not for limit dextrins. Hydrolysis of limit dextrins is continued when the wort is being fermented by the mal dextrinase or glucoamilase of microbial origin. That is why the fermentation rate of starch-based wort is limited by the hydrolysis rate of limit dextrins. [1, 2, 3, 4, 5, 7, 13, 14, 18, 19]



2. Research Materials and Methods

To address this, we used:

- starch-containing raw materials (maize);
- enzyme preparations (Alfaferm 350L, Glucozyme A 400S, and Protoferm FP);
- brewing yeast (Fermiol DY 7221).

Enzyme preparation of bacterial α -amylase Alfaferm 3500L is produced by submerged cultivation of a genetically non-modified strain of *Bacillus subtilis* BF-7658.

Enzyme preparation Glucozyme A 400S is a source of glucoamilase produced by submerged cultivation of a genetically non-modified strain of *Aspergillus niger* AS3.878.

Enzyme preparation Protoferm FM is a source of acidic proteases produced by submerged cultivation of a genetically non-modified strain *Aspergillus niger* M188.

Fermiol DY 7221 is micropelletized dry yeast selected specifically for alcohol production. It has some distinctive features not found in similar preparations currently on the market: [8, 9, 10, 11, 12, 15, 16, 17]

- high content of living cells activated by reactivation;
- unlike other kinds of brewing yeast, the metabolism of this yeast generates primarily alcohol rather than carbon dioxide;
- it is highly tolerant to heat, as it is able to sustain active alcohol fermentation at 35°C; it is resistant to up to 12.5% of alcohol in the medium; it is highly resistant to lactic acid bacteria and wild-yeast infection;
- wort is fermented rapidly;
- recommended dosage is 200 to 300 g/m³.

The physical and chemical characteristics of semi-products were controlled by using state-of-the-art methods. Concentration of volatile impurities in the distilled wort was checked using an HP 6850 Agilent Series GC System gas chromatograph.

Experiment results were processed using a standard suite [20].

The goal of this research was to study how a complex of enzyme preparations would affect the quality of enzymatic hydrolysis of substrate so that we further could use Fermiol DY 7221 for fermentation.

Studies were carried out consistently with the basic stages and parameters of alcohol production: [14]

- first, the raw materials were prepared for cooking (peeled, ground, and mixed);
- then they were cooked using mechanical and enzymatic processing to destroy the cellular structure of starch and dissolve it;
- the cooked mass was cooled down, and the starch was saccharified with enzyme preparations;
- sugars were yeast-fermented;
- alcohol was distilled from the wort and rectified.

The peeled, weighed, and ground grain was mixed with water 1:3 at 40 to 45°. Before that, we added Alfaferm 3500L in a concentration appropriate for its properties.

Cooking was done at 100° over 40 minutes, stirring continuously over the first 10 minutes. The mixture was then cured for 90 minutes in an autoclave at 1.5 atmospheres (127°). The produced cooked mass was divided into 7 equal parts and then cooled down to 58-60°C for saccharification. To identify the effects of Glucozyme A400S, we added it to the cooked mass as shown in *Table 1* and cured the mass for 1 hour at the same temperature.

In order to intensify fermentation, Protoferm FP was added to the wort to ensure complete hydrolysis of starch.

The resulting wort was fermented at 28±2°C for 72 hours using Fermiol DY 7221.

Pursuant to the rules of technochemical control of alcohol production, the quality of semi-products was controlled at each stage of this study.

Since the degree of fermentation is one of the main indicators of the enzyme effects, we measured the parameters of the spirit wort after fermentation was complete. *Table 2* presents the alcohol content data.

Table 1. Ratio of enzyme preparations in samples

Sample #	Gluczyme A400S	Alfaferm 3500L	Protoferm FP
1	2.53	3.5	0.19
2	2.84	3.5	0.19
3	3.16	3.5	0.19
4	3.48	3.5	0.19
5	3.79	3.5	0.19
6	4.11	3.5	0.19
7	4.43	3.5	0.19

Table 2. Concentration of alcohol in the spirit wort

Sample #	FP amount, ml.	Alcohol vol., %
1	2.53	8.1
2	2.84	8.3
3	3.16	8.8
4	3.48	9.2
5	3.79	8.9
6	4.11	9.1
7	4.43	9.0

Apparently, Sample #4 had the highest concentration of alcohol in the wort. Studies also showed that further increase in the concentration of Gluczyme A400S did not affect the alcohol output, meaning that it would be a waste of the enzyme preparation.

Figure 1 presents the correlation of alcohol concentration in the spirit wort and Gluczyme A400S concentration in the enzyme complex.

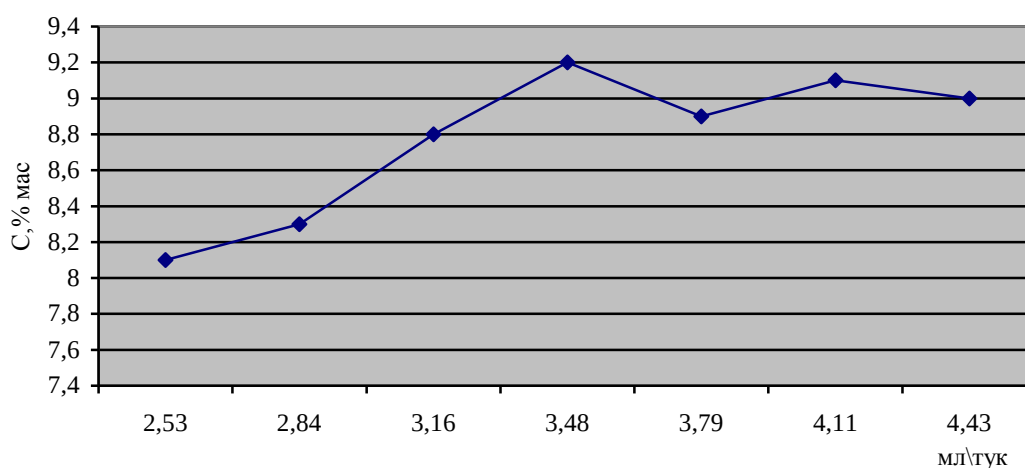


Figure 1. Correlation of alcohol concentration in the spirit wort and Gluczyme A400S concentration in the enzyme complex.

For further research, we used Sample #4 that had the highest alcohol content (9.2%). Sample #4 parameters are shown in *Table 3*.

Table 3. Sample #4 analysis results.

Indicator	Value for maize wort
	Fermiol DY 7221
Apparent concentration of solids in the wort filtrate, %	-0.4
Wort-filtrate acidity, deg	0.32
Alcohol mass concentration, %	9.2

Table 4. Fermentation effects on pH

Maize wort	pH	Alcohol concentration, %
Before fermentation	4.9	0
In 12 h	4.72	3.7
24 h	4.34	4.9
36 h	4.38	7.7
48 h	4.19	8.5
60 h	4.05	8.8
72 h	3.98	9.2

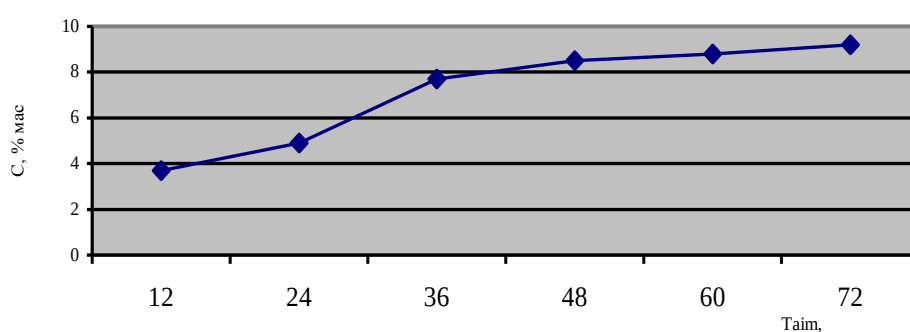


Figure 2. Correlation of alcohol concentration and fermentation duration.

Concentration of volatile impurities in the distilled spirit was checked using an HP 6850 Agilent Series GC System gas chromatograph, see *Table 5*.

Gas chromatography showed that Sample #4 was the closest to Grade 1 alcohol in terms of physics and chemistry.

Table 5. Gas chromatography

Indicators	Sample results (raw)	Normative values (raw)
acetaldehyde	6.25	10 ml/l
methyl acetate	-	30 mg/l
ethyl acetate	28.0	30 mg/l
methanol	0.048	0.05
2-propanol	-	35 mg/l
1-propanol	29.7	35 mg/l
izoamilol	16.57	35 mg/l
1-butanol	7.73	35 mg/l
furfural	-	-

Conclusion

It has been found out that using an enzymatic complex consisting of 3.5 ml Alfaferm 350, 3.48 ml Glucozyme A 400S, and 0.19 ml Protoferm FP enables one to:

- produce a wort containing 9.2% alcohol (mass, 11.7% vol.);
- reduce the duration of fermentation by 10 to 12 hours (60 hours).

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