

# BIOETHANOL FROM WHEY THE ELECTROHYDRODYNAMIC METHOD

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**Abstract:** Experimental investigations of the new technology for biofuel production from the whey are discussed in detail. Technological scheme of alcoholic fermentation of serum is described. It was shown that further studies of alcoholic fermentation of whey technology are necessary in order to increase the quantity and improve the quality of ethanol.

## 1. Introduction

Investigations aimed at the elaboration of high-technologies and equipment for biofuel production are among the priority tasks. It contributes to the augment of energy consumption from renewable sources and increase energy security.

The main stages of technology for production of bioethanol from whey comprise partial deproteinization of whey, fermentation with a mixture of mesophilic lactic acid bacteria and yeasts, alcohol distillation [1]. The studies aimed at establishing the optimal method for alcoholic fermentation of whey.

Modern industrial distillation columns for alcohol production have rather large overall dimensions and specific consumption of materials. Costs associated with the manufacture of these columns and construction of special buildings are very high. That is why, the elaboration of compact distillation columns with high fractionating ability, small hydraulic resistance, possibility to use liquids, including mechanical particles is an urgent scientific problem.

One of the promising methods for the intensification and control of mass transfer processes in gas-liquid systems is the application of the electric field effect. Several methods for realization of mass transfer processes in electric field conditions were elaborated and investigated. As a result, a new type of distillation columns was created and was named as electrohydrodynamic (EHD) distillation columns [2].

EHD distillation columns have been investigated for the cases of distillation of crude alcohol, wine material, ethanol-water mixtures and other liquids. The analysis of the dependencies of the intensity of mass transfer and effectiveness of distillation in an electric field, a study of the mechanisms of action of the field, and evaluation of various regimes made it possible to work out practical recommendations for designing and manufacturing an industrial-type EHD distillation columns for alcohol production. Application of the EHD distillation columns for new technology for biofuel production from the milk serum has to be estimated.

## 2. Alcoholic fermentation of milk serum

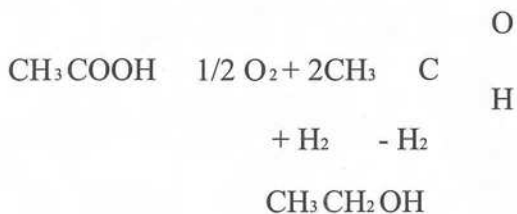
Technological scheme of alcoholic fermentation of whey includes: heating the clarified whey to 93 °C to reach pasteurization, cooling, decantation and fermentation with lactose fermenting yeast at a temperature of 32 - 34 °C. Biochemical properties of the cultures are characterized by the fact that besides the main fermentation products (ethanol and carbon dioxide), fusel oils, aldehydes and esters are accumulated in liquid media. The first phase of our research was to obtain yeast, which has a high efficiency of ethanol formation at a low percentage of fermentation by-products (aldehydes, fusel oils and esters). In addition, it is necessary to provide a minimum duration of fermentation process at the highest possible content of ethanol. We studied mixtures of yeast species *Saccharomyces* and mesophilic culture *Lactobest* (DLF-N-86DI100)

Cadorago (Co) Italy (freeze dried culture for food industry). Starter has been prepared in pasteurized clarified serum (OST 10-02-02-3-870) by the introduction of bacteria and lactose fermenting yeast in its 10% suspension and subsequent cultivation at 34 °C for 1-2 days. Adaptation of the strains was carried out by increasing the content of lactose and alcohol in the medium; thus resistance of culture gradually has been increased and selection of improved variants has been performed.

Application of *Saccharomyces cerevisiae* is promising because *Saccharomyces cerevisiae* can operate at higher ethanol concentrations in the medium. Consequently, a greater amount of lactose can be transformed in the ethanol at its pre-splitting by enzyme.

Volumes of ferment, temperature and fermentation time were varied in a wide range. Analysis of the initial and fermented serum at various stages of fermentation was carried out by nuclear magnetic resonance spectroscopy.

It should be noted that pH 4.0 - 4.6 is set during the fermentation. This PH is optimal for the reproduction of yeast.



**FIG.1 THE SCHEME OF THE ETHANOL BIOSYNTHESIS**

Vigorous stirring for 24 hours, creates favorable conditions for the accumulation of the required number of yeast cells and intensification of the alcohol production. This method allows fermenting lactose completely.

Биосинтез этанола осуществлялся по схеме, представленной на FIG.1. The end of alcoholic fermentation is determined by the residual lactose content (%). In some experiments, the high levels of lactic acid is obtained, which exceeds the content of ethanol.

Increased alkalinity of whey on the first day of fermentation caused by yeast during growth consume acidic component: lactic acid, citrates, soluble proteins. Alcoholic fermentation is more intense on the third day of fermentation. As the depletion of nutrient medium and increasing the content of ethanol in it, the intensity of the alcoholic fermentation is reduced.

Time day	pH	Composition, %			
		Ethanol	Acetic acid	Lactic acid	Sugar
1	4,9	19,9	5,5	34,6	36,6
2	4,6	16,8	5,0	28,2	46,5
3	4,6	22,6	5,3	27,8	41,7

**TABLE 1. THE MAIN FERMENTATION PRODUCTS OF THE WHEY. The initial whey: pH-4,7; Ethanol - 10,0 %; Acetic acid-6,8 %; Milk acid-31,6 %; 2,3 Butandiol - 2,3 %; Sugar - 49,3 %.**

In experiments with yeast and lactic bacteria (Table 2) both alcohol and lactic acid fermentations took place. Joint using of yeast and lactic acid bacteria has the following advantages: acidification of the medium leads to a splitting of complex nitrogen-containing compounds, thus favoring the nutrition of yeast. However, in order not to inhibit the growth of yeast the amount of lactic acid bacteria should not be large (not more than 1%).

Among the secondary alcoholic fermentation products other than lactic acid there are acetic acid, acetaldehyde, 2,3 butylene glycol, succinic acid and acetoin. Acetaldehyde is generated from glucose during the alcohol fermentation. It can also be formed by the oxidation of ethanol. In general, in our experiments the contents of acetaldehyde were from 3 to 5% of fermented whey.

**Table 2. THE MAIN FERMENTATION PRODUCTS OF THE WHEY. The initial whey: pH - 4,75; Ethanol - 4,5%; Acetic acid - 4,7 %; Milk acid - 34,9 %; 2,3 Butandiol - 2,3 %; Sugar - 54,8 %.**

Time day	pH	Composition, %			
		Ethanol	Acetic acid	Lactic acid	Sugar
1	4,0	11,3	5,2	32,6	48,2
3	4,1	24,5	22,2	--	35,9
4	4,3	19,9	17,9	--	49,3
6	4,6	0	20,4	65,1	0

Formation of acetic acid is due to oxidation of acetaldehyde oxygenated water. Subsequently, succinic acid is formed as the result of two molecules condensation of acetic acid (Thunberg reaction). Content of secondary fermentation products depends on the aeration, pH medium, temperature, composition of the initial whey. Concentration of succinic acid is increased at aeration and is reduced under anaerobic conditions.

At the above mentioned conditions, an analysis of whey samples were carried out on the second, third, sixth day of fermentation. Chromatographic peaks were identified for ethanol, lactic acid, acetic acid, succinic acid. pH 4.0 - 4,6 coincides with the isoelectric point of lactoalbumin fraction of proteins and provides them with a quick coagulation. Whey as secondary product from milk factory was used as raw materials for the investigation.

In separate investigation the preliminary thickening of initial whey by evaporation was carried out. Thickening makes it possible to increase the concentration of lactose, and hence the concentration of ethanol. Osmosis is more preferable method for thickening of whey than the evaporation in vacuum.

The most economically advantageous to conduct the process of whey conversion in ethanol is the separation of protein from whey, its concentration by reverse osmosis and then performing fermentation.

### 3. Conclusions

The results of the investigation allow us to optimize the operational parameters of technology for ethanol production from the whey. Process steps for the pre-treatment whey are defined: the separation of protein from whey, concentration of the whey by reverse osmosis and fermentation.

Further studies of alcoholic fermentation of whey technology are necessary in order to increase the quantity and improve the quality of ethanol.

The obtained results of the study make it possible to proceed the elaboration of the next stage of technology, namely, the EHD distillation of ethanol.

### 4. References

- [1] Zalashko, M.V., Biotechnology of the milk serum reprocessing, Agropromizdat, Moscow, 1990, 190.
- [2] Maximuk, E.P., About new type of distillation columns // Vision 2020: Distillation Priorities Topical Symposium proceedings (AIChE 2000 Spring National Meeting in Atlanta, GA, USA), ISBN#0-8169-9889-2, Session 4: Distillation Modeling and Equipment, 201-208, 2000.