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Screening of Yeasts for Fermentation of Jerusalem Artichoke Tubers Juice and Selection of the Active Strains for Ethanol Production

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Abstract: Strains *K. marxianus* Y-303, *Oenoferm credo*, *Oenoferm rouge*, *Sacch. vini* Rkaseteli-6, *Sacch. cerevisiae* XII and *Sacch. uvarum* 73 were used for fermentation of juice, extracted from Jerusalem artichoke tubers grown in Uzbekistan. The fermentation process was carried out in laboratory conditions. Among these yeasts *Oenoferm credo*, *Oenoferm rouge* and *Sacch. uvarum* 73 showed higher performance than *K. marxianus* Y-303 and the ethanol yields were 92%, 90% and 88%, respectively. After 72 h fermentation with *Sacch. vini* Rkaseteli-6 and *Sacch. cerevisiae* XII the yield of ethanol was 82% and 74% of maximum theoretical yield, respectively.

Key words: Jerusalem artichoke, *K. marxianus* Y-303, *Oenoferm credo*, *Oenoferm rouge*, fermentation, ethanol yield.

1. Introduction

Nowadays, the ethanol manufacture is one of the large and scientific, technical developed sectors in the national economy of Uzbekistan.

The main processes, such as a conversion of starch into sugar and sugar into ethanol, were carried out by yeast enzymes. Ethanol is widely used in a distillery production, fruit and berry wines and vinegar production, food flavorings and perfumes. In the microbiological and medical industries, ethanol is used to precipitate enzyme preparations from a yeast fluid or an extract from a solid-phase yeast, in vitamin production, various medicines, as a disinfectant and a substance that prevents infection and spoilage of therapeutic extracts. As well as ethanol is used in the chemical, engineering and other industries and a veterinary [1].

The cost of raw materials takes more than 60% of the total costs of ethanol manufacture. Therefore, replacing grain with non-traditional, cheaper raw materials will increase production profitability and will save valuable products.

It is known that traditional agricultural crops for ethanol production are starch-containing raw materials—grain and potato, the sugar-containing raw material—molasses [2]. In many countries of the world, as well in Uzbekistan grain is used as the main agricultural crop for ethanol production. However, as a non-traditional raw material for the ethanol production, any material, containing sugars or polysaccharides can be used, which, as a result of hydrolysis, converts into fermentable sugars.

Jerusalem artichoke (*Helianthus tuberosus* L.) tubers have a very low coefficient of accumulation of toxic substances (nitrates, heavy metals, radio nuclides), compared to other agricultural crops and in addition it has no specialized disease pests [3]. It has a number of advantageous characteristics over traditionally agricultural crops, including high growth rate, good tolerance to frost, drought and poor soil,

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strong resistance to pests and plant diseases, with minimal to zero fertilizer requirements [3-5].

According to researchers, Jerusalem artichoke is the cheapest and most promising raw material for ethanol production. The yield of ethanol from this non-traditional agricultural crop is higher than that from grain and potatoes. In many countries, including Uzbekistan, *Saccharomyces cerevisiae* is used in ethanol production. However, recently, *Kluyveromyces* yeasts have been used in ethanol manufacture from Jerusalem artichoke tubers [6-8].

Jerusalem artichoke tubers are rich in inulin, which can be easily hydrolyzed and then converted into ethanol using biocatalysts. The ethanol yield is equivalent to that of sugarcane and twice that obtained from corn. These characteristics make Jerusalem artichoke an outstanding substrate for ethanol production [6, 9].

Russian scientists [10], by investigating the effect of different yeasts on the biosynthesis of ethanol from topinambur, found that *Kluyveromyces marxianus* Y-303 is the most effective yeast strains for fermentation of Jerusalem artichoke mash.

The aim of this research is screening different yeasts which can actively ferment mash of Jerusalem artichoke tubers with the high ethanol yield. Moreover, the ethanol yield is the most important techno-economic factor for ethanol production in industry.

The objects of research were different strains of alcohol, wine, brewer yeast and *Kluyveromyces marxianus* Y-303 was used as a control strain.

2. Materials and Methods

2.1 Strains and Media

K. marxianus Y-303 [8], *S. vini* Rekasiteli-6, *Oenoferm credo*, *Oenoferm rouge*, *S. cerevisiae* sp., *S. uvarum* 731 were cultured in YPD (Yeast Extract-Peptone-Dextrose) medium composed of 20 g/L glucose, 10 g/L yeast extract, and 20 g/L peptone at 30 °C and 150 rpm. All yeast strains were bred by the Department of Food Technology, Tashkent Chemical-Technological Institute, Uzbekistan.

2.2 Ethanol Fermentation from Jerusalem Artichoke Tubers

Jerusalem artichoke was grown in Tashkent Province, North-East Uzbekistan. Tubers were washed, mashes were obtained in squeezer and were used as a substrate for ethanol fermentation. Due to the slow growth of yeast under ethanol fermentation conditions, an inoculation level of 10% was adopted for ethanol fermentation. A seed culture of 10 mL was inoculated into a 250 mL flask containing 90 mL mash, and ethanol fermentation was carried out at 30 °C and 150 rpm under non-aeration conditions by plugging the flask with a water trap to prevent oxygen in air from dissolving into the broth during fermentation. All media were sterilized at 121 °C for 20 min, and duplicates were performed for all experiments [11].

2.3 Analytical Methods

Ethanol was analyzed by distillation method. At 20 °C, a 100 mL fermented mash filtrate, neutralized with NaOH at 1 mol/dm^3 in advance, poured into flask, of which the volume is 200-250 mL. The flask, which is used for measuring, is rinsed with 50 mL of distilled water and placed in the same distillation flask. The distillation flask is connected to a distillation unit. Then 10-15 mL of distilled water is poured into the receiving flask and placed in an ice-cold container. The distillation unit must be hermetic.

The process stops after about 4/5 of filtrate volume is distillated. After cooling up to 20 °C, the volume was made to the mark with distilled water and was mixed. The distillate in the flask was poured into a dry measuring cylinder and the concentration of alcohol was determined by alcoholmeter at 20 °C [12].

3. Results

3.1 Ethanol Production from Jerusalem Artichoke Tubers

Ethanol production from Jerusalem artichoke tubers was evaluated for several yeast strains and compared with *Kluyveromyces marxianus* Y-303. The content of

Table 1 Ethanol production from Jerusalem artichoke tubers by several yeast strains.

No.	Yeast	Fermentation conditions	Ethanol concentration, % (*v*/*v*)	The percentage of theoretical yield on the basis of total sugar, %
1	*Sacch. vini* Rkaseteli-6	28 °C, 72 h	7.9 ± 0.2	82
2	*Oenoferm credo*	28 °C, 72 h	8.8 ± 0.2	92
3	*Oenoferm rouge*	28 °C, 72 h	8.6 ± 0.2	90
4	*Sacch. cerevisiae* XII	28 °C, 72 h	7.1 ± 0.2	74
5	*Sacch. uvarum* 731	28 °C, 72 h	8.4 ± 0.2	88
6	*Kluyveromyces marxianus* Y-303	28 °C, 72 h	8.3 ± 0.2	87

mash for each fermentation process was 16% of fermentable sugar. The ethanol fermentation performance was evaluated by ethanol accumulation and ethanol fermentation conditions, as illustrated in Table 1.

Ethanol was produced from Jerusalem artichoke tubers by several yeast strains. The data were collected from three independent experiments.

4. Discussion

As can be seen in Table 1, culture of *Oenoferm credo* yeast gave the best results under the authors' research scope and the yield of ethanol was 92% of maximum theoretical yield and the ethanol concentration was 8.8% (*v*/*v*) for 72 h fermentation processing. In contrast, only 8.3% (*v*/*v*) ethanol was produced by the *Kluyveromyces marxianus* Y-303 in the same fermentation time which is 87% of ethanol yield. Less ethanol productivity was observed for the *Oenoferm rouge* and *Sacch. uvarum* 731, than the above mentioned *Oenoferm credo* yeast strain, correspondingly 8.6% (*v*/*v*) and 8.4% (*v*/*v*). Their ethanol yields were 90% and 88% respectively. Compared to the *Kluyveromyces marxianus* Y-303, these strains represented an increase of 3.5% and 1.2%, respectively. After 72 h fermentation, the ethanol concentration was achieved in the fermentation broth by the *Sacch. vini* Rkaseteli-6 and *Sacch. cerevisiae* XII by 7.9% (*v*/*v*) and 7.1% (*v*/*v*) and the yields of ethanol were 82% and 74% of maximum theoretical yield respectively, which are acceptable from the viewpoint of industrial application.

5. Conclusion

The ethanol productivity and yield were improved by the *Oenoferm credo*, *Oenoferm rouge* and *Sacch. uvarum* 731, and less unfermented sugar remained, addressing important techno-economic considerations in industrial ethanol production from Jerusalem artichoke tubers.

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