

## THE BATCH ETHANOL FERMENTATION OF JERUSALEM ARTICHOKE USING *SACCHAROMYCES CEREVISIAE*

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### ABSTRACT

The potential of using Jerusalem artichoke (*Helianthus tuberosus* L.) grown in Thailand as substrate for fuel ethanol production was evaluated in this study. Chemical composition analysis of the plant juices extracted from its tubers was investigated and the results reveal that its contained 256.3 g/l total sugars, 74% of which is inulin. The plant juices also contained several minerals essential for growth and ethanol fermentation by ethanol-producing microorganisms. The highest mineral contents were nitrogen (2,431.5 mg/l) and potassium (2,491.8 mg/l). The batch ethanol fermentation of the acid hydrolyzed plant juices using *Saccharomyces cerevisiae* was determined and the maximum ethanol concentration, ethanol yield, and ethanol productivity of 88.1 g/l, 0.45 g/g, and 1.84 g/l.h, respectively, were obtained under the optimal conditions: 250 g/l initial sugar concentration, pH 5.0-5.5 and 10<sup>8</sup> cell/ml initial yeast cell. The conversion efficiency of the fermentation of Jerusalem artichoke juices was approximately 88% of the theoretical ethanol yield.

**KEYWORDS:** Jerusalem artichoke, ethanol fermentation, *Saccharomyces cerevisiae*

### 1. INTRODUCTION

The Jerusalem artichoke (*Helianthus tuberosus* L.) is a plant native of temperate North America, and has been proposed for many years to be a possible the material for ethanol fermentation. Great attention is focused on renewable sources in bioethanol production. Among unconventional materials, Jerusalem artichoke (*Helianthus tuberosus* L.) is one of the most interesting substrates. This plant is a rich source of carbohydrates (11-20%), where 70-90% of them are inulin and inulids. Inulin consists of linear poly fructose chains in the  $\beta$  (2 $\rightarrow$ 1) position with a terminal glucose

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residue which is linked to fructose by an  $\alpha(1\rightarrow2)$  bond. Yeasts with an inulinase are able to directly ferment inulin and inulids in this material, whereas yeast or bacterium which has no inulinase activity requires a hydrolysis step before ethanol fermentation process. The interest in Jerusalem artichoke as a raw material for fuel ethanol results from its high tuber yield up to 90 t/ha with a carbohydrates yield of 5-14 t/ha [1-2]. In this research, acid and enzymatic hydrolyses of inulin and inulids in Jerusalem artichoke tubers for fuel ethanol production were investigated. Some factors influencing ethanol production from Jerusalem artichoke juices by selected *Saccharomyces cerevisiae* were also described.

## 2. MATERIALS AND METHODS

### 2.1 Jerusalem artichoke

Jerusalem artichoke tubers collected from different places in Northeastern part of Thailand were used in this study. The tubers were cleaned, crushed and pressed. Juices were collected after pressing and were either acid or enzymatic hydrolysis before using as raw materials for ethanol production in classical batch fermentation process.

### 2.2 Microorganism and medium

*Saccharomyces cerevisiae* was grown in YM medium containing : glucose 10 g; yeast extract 3 g; peptone 5 g and malt extract 3 g per l for 18 h at 30° C with shaking at 150 rpm.

### 2.3 Acid hydrolysis

Acid hydrolysis of inulin and inulids into fermentable sugars was conducted at pH 2.0 adjusted with sulphuric acid ( $H_2SO_4$ ) and heated at 80° C for 40 min. The pH was adjusted to 5.0 before fermentation.

### 2.4 Batch fermentation

Batch fermentation of Jerusalem artichoke juices was conducted in 500 ml Erlenmeyer flasks with 350 ml working volume. The effect of initial pH of fermentation medium was studied at 4.5, 5.0, 5.5 and 6.0. The effects of sugar concentration and initial cell number on ethanol production were also tested; initial sugar concentration at 250, 300 and 350 g/l ; initial cell number at  $10^6$ ,  $10^7$  and  $10^8$  cell/ml. Fermentation was statically incubated at 30°C for 72 h by in temperature controlled incubator.

### 2.5 Analytical methods

Reducing sugars were estimated with 3,5-dinitrosalicylic acid (DNS), using fructose as the standard [3]. Total sugars were assayed by the Phenol-sulfuric acid method [4]. Minerals in the raw material was measured by atomic absorption. Ethanol concentration was analyzed by gas chromatography with flame ionization detector.

## 3. RESULTS AND DISCUSSIONS

The chemical compositions of Jerusalem artichoke (*Helianthus tuberosus* L.) tuber juices were shown in Table 1. The plant juices contained approximately 256.3 g/l total sugars, 74% of which was inulin. It also contained several minerals which were essential for growth and ethanol fermentation by ethanol-producing microorganisms. The highest mineral contents were found to be nitrogen (2,431.5 mg/l) and potassium (2,491.8 mg/l).

**Table 1** Major components in Jerusalem artichoke tuber juices

Composition	Amount	Composition	Amount
Moisture (%)	77.7	Minerals	
Total soluble solid (°Brix)	19.5	- nitrogen (mg/l)	2,431.3
Total sugar (g/l)	256.3	- phosphorus (mg/l)	2,491.8
Reducing sugar (g/l)	21.0	- magnesium (mg/l)	273.8
Inulin (g/l)	189.0	- sodium (mg/l)	182.7
Phenolic compound (g/l)	1.3	- zinc (mg/l)	1.9

The batch ethanol fermentation of acid hydrolyzed plant juices by *S. cerevisiae* was investigated. Table 2 summarized the kinetic parameters for the batch fermentation of Jerusalem artichoke juices by *S. cerevisiae* at 48 h after fermentation. Ethanol yield decreased when initial sugar concentration increased. In contrast, the rate of ethanol fermentation increased when initial cell number increased. The maximum ethanol concentration, ethanol yield and productivity of 88.1 g/l, 0.45 g/g and 1.84 g/l.h respectively, were obtained at initial cell number of  $10^8$  cells/ml and initial sugar concentration of 250 g/l.

**Table 2** Fermentation parameters of ethanol production from Jerusalem artichoke juices by *S. cerevisiae* under various initial cell number and sugar concentration

Cell number (cells/ml)	Sugar conc. (g/l)	Ethanol conc. (g/l)	Ethanol yield (g/l)	Productivity (g/l.h)
$10^6$	250	60.22	0.42	1.26
	300	44.24	0.40	0.92
	350	46.21	0.18	0.96
$10^7$	250	79.81	0.42	1.66
	300	83.78	0.40	1.75
	350	85.35	0.31	1.78
$10^8$	250	88.08	0.45	1.84
	300	83.59	0.35	1.74
	350	82.75	0.32	1.72

Table 3 showed the kinetic parameters for ethanol production from Jerusalem artichoke tuber juices at different initial pHs. No significant difference in ethanol concentration, ethanol yield and productivity was observed at all pH, tested. However, pH 5.0 tended to give higher ethanol concentration than that in other pH, therefore, pH 5.0 was chosen for ethanol fermentation.

**Table 3** Fermentation parameters for ethanol production from Jerusalem artichoke juices by *S. cerevisiae* under different initial pHs

Initial pH	Ethanol conc. (g/l)	Ethanol yield (g/l)	Productivity (g/l.h)
4.5	74.89	0.39	1.56
5.0	76.06	0.41	1.58
5.5	76.42	0.41	1.59
6.0	74.50	0.44	1.55

#### 4. CONCLUSIONS

The batch ethanol fermentation of the acid hydrolyzed plant juices using *S. cerevisiae* was determined. The hydrolysis of inulins and inulids in Jerusalem artichoke juices was necessary process for ethanol fermentation. Acid hydrolysis with concentrated sulfuric acid ( $H_2SO_4$ ) at 80 °C for 40 min gave highest reducing sugar content as well as ethanol yield. The maximum ethanol concentration, ethanol yield, and ethanol productivity of 88.1 g/l, 0.45 g/g, and 1.84 g/l.h, respectively, were obtained. The optimal conditions for fermentation were as follows: 250 g/l initial sugar concentration, pH 5.0 and  $10^8$  cell/ml initial yeast cell.

#### 5. ACKNOWLEDGEMENTS

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