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# Kinetic Study of Xanthan Production Using Newly Isolated Strain by Sugarcane Molasses

Selvi V\*, Manikkandan T R and Vijayagopal V Department of Chemical Engineering, Annamalai University, Annamalai Nagar – 608002.

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# ABSTRACT

Investigations on the production of xanthan with a locally isolated strain have been emphasized. We can isolate the strain by diluting serially the extract of infected banana petioles. By considering the cost-effective aspects of the xanthan fermentation process, agro industrial substrate sugarcane molasses was exploited. The effect of initial concentration of carbon source for the substrate was studied by varying the glucose concentration in the range of 2% to 6% (glucose equivalent). The time course of cell mass, xanthan production and substrate utilization were recorded in order to facilitate the kinetics of fermentation. The substrate chosen for study, sugarcane molasses resulted the xanthan gum yield of 27.2 g/l at a fermentation period of 96 h with the initial glucose concentration of 4%. The study confirmed that the logistic equation for growth kinetics and Luedeking-Piret model for substrate utilization and product formation model fits well with the experimental data.

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### Introduction

The awareness in environmental friendly natural polysaccharide has increased in recent years due to their huge potential in varied sectors, possessing several advantages like biodegradability, nontoxicity and biocompatibility over synthetic or chemical polymers which are non-ecofriendly [1]. Xanthan is an important and natural biopolymer used in industries. It was discovered in 1963 at Northern Regional Research Centre, United States Department of Agriculture (USDA) [2]. Xanthan is non-toxic and non-sensitizing so it doesn't inhibit growth or cause eye or skin irritation. So United States Food and Drug Administration (FDA) has approved xanthan to use in food additive without any specific quantity limitations [3].

Xanthan gum is used in many applications, mainly in food industry as thickening, suspending and stabilizing agent. It is widely used in a broad range of industries, such as in toiletries, oil recovery, cosmetics, as water-based paints, etc [4].

In this present work, xanthan gum is produced by submerged aerobic fermentation by using locally isolated strain extracted from infected banana petioles. Produce xanthan gum by isolated strain using sugarcane molasses as a carbon substrate [5]. The kinetic studies for cell growth, substrate utilization and product formation were carried out with the experimental data of xanthan production using isolated strain from sugarcane molasses [9].

# **Materials and Methods**

# Isolation of strain and maintenance

Isolation of strain and maintenance as described in previous publication [7].

# **Pre-treatment of Sugarcane molasses**

Sugarcane molasses was obtained from fromN.P.K.R.Ramasamy co-operative sugar mills LTD, Thalainayar, Tamilnadu, India. Concentrated sugarcane molasses was diluted with distilled water containing sodium dihydrogen orthophosphate, at a concentration of 2 g/l at a ratio of 1:1. The solution was autoclaved at 121°C for 20min at 14.8 psi. The sterile sugarcane molasses was then filtered and used for fermentation processes. The reduced sugar was measured using DNS by colorimetric method [6].

#### **Batch Fermentations Studies**

Batch fermentation studies as described in previous publication [8].

# Determination of bacterial growth

Biomass determination was done by dry cell-weight estimations. The cells were collected after centrifugation at 5000 rpm for 10 min. After discarding the supernatant, the biomass was washed with distilled water and re-centrifuged. Cells were dried in an oven at  $65^{\circ}$ C for 2 h and weighted.

Determination of xanthan gum concentration and reducing sugars concentration

Xanthan gum and reducing sugars were determined as described in previous publication [7].

# Spectroscopy of Fourier transform infrared (FTIR)

Fourier transform infrared spectroscopic analysis was described in previous publication [8].

Kinetic studies

The logistic, Substrate consumption and Product formation kinetics can be described in previous publication [8].

#### **Results and Discussion**

The effect of initial concentration of sugarcane molasses was studied by varying the glucose concentrations ranging 2%, 3%, 4% 5% & 6% (w/v) (glucose equivalent). The experiments are carried out in flasks for 96 hours with an initial pH of 7 at a constant temperature of  $30^{\circ}$ C and inoculums size 10 % (v/v). The time course of cell mass, xanthan production and substrate utilization were recorded in order to facilitate the kinetics of fermentation. Among the substrate concentration chosen for study, 4% (w/v) (glucose equivalent) resulted in the highest xanthan yield. The results are given in Table 1 Xanthan productions from sugarcane molasses using an isolated strain in batch cultures were performed at concentrations ranging 4% (w/v) (glucose equivalent) at different time intervals. The maximum Xanthan production of 27.2 g/l was achieved at 96 h. The results are given in Table 2

# Table 1.Effect of sugarcane molasses concentration on xanthan production using newly isolated strainin batch cultures

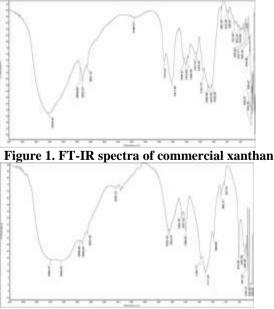
substrate concentration (Glucose) (g/L)	Cell mass (g/L)	Xanthan (g/L)	Substrate (g/L)
20	4.3	15.1	0.8
30	4.8	18	1.4
40	7.5	27.2	1.5
50	4.1	18.4	4.2
60	3.8	15.5	7.5

Table 2. Effect of fermentation time on xanthan production using sugarcane molasses by newly isolated isolated strain in batch cultures.

isolated strain in batch cultures.					
Time	Cell mass	Xanthan	Substrate		
(hours)	(g/L)	(g/L)	(g/L)		
0	0.2	0	40		
24	1.4	4.9	33.6		
48	4.4	13.7	17.7		
72	7.2	22.9	3.8		
96	7.5	27.2	1.5		
120	7.5	26.4	1.1		

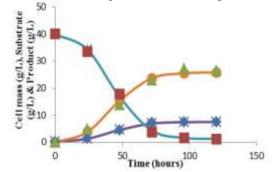
#### FTIR Spectroscopic Analysis of Xanthan

A FT-IR spectrum of commercial xanthan was used as a reference and this was compared with the spectra of produced xanthan obtained from sugarcane molasses as a substrate. Figure 1 show that the infrared spectrum of the commercial xanthan is very similar to that obtained for the produced xanthan (Figure 2) using the isolated strain by the substrate. Based on the results obtained from FTIR, the remote polysaccharide was found to follow the same spectral behavior as the standard



# Figure 2. FT-IR spectra of produced xanthan Kinetic Studies

The capability of substrate utilization, growth kinetics and product formation fitted into various models, namely, Logistic, Monod, Andrew, Herbert, Verlhurst, Shehata& Marr, Moser, Tessier, Contois and Haldane models in representing the batch kinetic data of the present work were analyzed, The substrate utilization data best fits with substrate utilization kinetics with yield coefficient of 0.188, the growth kinetic data fit with the Logistic model with a correlation coefficient of 0.9975 and Luedeking-Piret model fits for the product formation with yield coefficient of 3.6. Figures 3, illustrate the experimental, predicted values and error percentage for cell mass, substrate utilized and product formed at a given time.



#### Figure 3. Comparative chart showing experimental and predicted values of cell mass concentration, substrate utilization and product formation of xanthan.

#### Conclusion

The present study is aimed at fermentative production of xanthan using isolated strain. The substrate sugarcane molasses has resulted in the xanthan concentration (27.2 g/l) with a cell mass concentration of 7.5 g/L at 96 h of fermentationunder the optimal conditions. FTIR spectroscopic analysis was also performed to check out the functional groups of xanthan. The characteristic peaks denoting functional groups apparently confirmed the samples as xanthan. The kinetics of cell growth, substrate utilization and product formation were studied for the strain utilizing the substrate. Results indicated that logistic model befitted the cell growth kinetics and Luedeking-Piret model suited substrate consumption and product formation kinetics well.

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