PRODUCTION OF ACETIC ACID FROM MOLASSES BY FERMENTATION PROCESS

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ABSTRACT

Acetic acid also called ethanoic acid is organic compound. Acetic acid produced via fermentation. Its pathway is conversion of glucose to ethanol and ethanol to acetic acid. In first step, Saccaromyces cerevesiae (yeast) converts fermentable sugar of molasses into ethanol and carbon dioxide. In second step, acetobacter aceti (acetic acid bacteria) converts ethanol into acetic acid and water. After completing process, the separation of product is carried out via centrifugation. Mixture of acetic acid and water is separated by distillation.

In this report, details regarding cultures (micro-organism) have been used for the process is discussed. In practical laboratory work, ethanol fermentation and acetic acid fermentation have been carried out and it's optimum parameters (pH, temperature, sugar concentration, and ethanol concentration) have been decided, which is discussed in detail. The kinetic study also have been done is mentioned.

Key words: Saccaromyces cerevesiae, acetobacter aceti, micro-organism, fermentable sugar

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1. FERMENTATION

The term "fermentation" comes from a Latin word fermentum(to ferment). Fermentation is described as the process in which chemical changes occur in an organic substrate as the result of action of microbial enzymes.
The product can either be:

- The cell itself: referred to as biomass production.
- A microorganisms own metabolite: referred to as a product from a natural or genetically improved strain.
- A microorganisms foreign product: referred to as a product from recombinant DNA technology or genetically engineered strain, i.e. recombinant strain.

All microorganisms need for their microbial activity the presence of several nutrients.

1.1 Micro-organisms involved in the fermentation

The organisms involved in production usually grow at the top of the substrate, forming a jelly like mass. This mass is known as 'mother of vinegar'. The mother is composed of both acetobacter and yeasts, which work together. The principal bacteria are Acetobacter aceti. The main yeasts are Saccharomyces cerevisiae. It is important to maintain an acidic environment to suppress the growth of undesirable organisms and to encourage the presence of desirable acetic acid producing bacteria.
Although a variety of bacteria can produce acetic acid, mostly members of Acetobacter, Gluconacetobacter, and Gluconobacter are used commercially.

1.2 Acetic acid fermentation

Production of acetic acid can be carried out by aerobic and anaerobic fermentation. Anaerobic process is one stage process carried out by Clostridium. Aerobic fermentation is two stage processes. Glucose is converted into ethanol by S.cerevesiae. And second stage is ethanol is converted into acetic acid by acetobacter aceti.
The fermentation is usually initiated by yeasts which break down glucose into ethyl alcohol with the liberation of carbon dioxide gas. Following on from the yeasts, acetobacter aceti oxidise the alcohol to acetic acid and water.
Yeast reaction:

$C_6H_{12}O_6 \rightarrow 2CO_2 + 2CH_3CH_2OH$
Glucose ethanol

Acetobacter aceti reaction:

$CH_3CH_2OH + O_2 \rightarrow CH_3COOH + H_2O$

Ethanol Acetic acid

The acetobacter aceti are dependent upon the yeasts to produce an easily oxidisable substance (ethyl alcohol). It is not possible to produce vinegar by the action of one type of micro-organism alone.

For etanol fermentation, it is essential to have sugar concentration around 20% and also pH and temperature should be maintained around 5 and 32°C respectively.

For a good fermentation, it is required to have an alcohol concentration of 10 to 13%. If the alcohol content is much higher, the alcohol is incompletely oxidised to acetic acid. If it is lower than 13%, there is a loss of vinegar because the esters and acetic acid are oxidised. In addition to acetic acid, other organic acids are formed during the fermentation which become esterified and contribute to the characteristic odour, flavour and colour of the vinegar. Also pH and temperature should be maintained around 4.5 and 30°C respectively.

In general, the yield of acetic acid from glucose is approximately 60%. That is three parts of glucose yield two parts acetic acid. For analysis of ethanol and acetic acid Gas chromatography and high performance liquid chromatography methods are used.

2. PARAMETRIC AND KINETIC STUDY

This study shows the effect of parameter on fermentation. During this study effect of sugar and ethanol concentration, effect of media pH and effect of temperature on fermentation were observed and rate of reaction and rate constant are determined.

2.1 Ethanol fermentation

Different concentrations of sugar were prepared by diluting molasses. These solutions were kept at different pH and temperature to determine the optimum condition of fermentation.

Increasing in the concentration of sugar up to 20% resulted in gradually increase in ethanol production. Beyond 20% sugar concentration growth of culture was inhibited resulting in decrease in ethanol production. Initial sugar concentration of 20% and optimum temperature of 30°C was selected. At pH 4, fermentation took place but it gave low ethanol concentration.

As pH increasing ethanol production is also increased. Best results were obtained at pH 5.2 where maximum ethanol production was noticed. Further increase in pH decrease the ethanol production. S.cerevesia can sustain up to 4 pH. In more acidic pH culture can't sustain. Better growth of culture was noticed at 5.2 pH.

Temperature is one of the major constraints that determine the ethanol production. To know the optimum temperature for ethanol fermentation, the solutions were kept at 28°C, 32°Cand 36°C with 20% initial sugar concentration and fermentation was carried out for 48 hours. A low ethanol yield of 8.23%was observed at 28°C in 48 hours. As shown at 32°C ethanol yield was maximum and turned out to be 8.81%. However, increasing the temperature up to 36°C concentration of alcohol decreased. So, 32°Cwas selected as optimum temperature for ethanol production.

Ethanol concentration was initially 0 after 32 hours concentration of ethanol was maximum. Ethanol concentration was increase gradually. In almost 32 hour reaction was completed after 32 hour ethanol concentration decreased due to decomposition.

The order is closely equal to 1. The average value of k =0.07715 (l/mol) ($hour^{-1}$).

2.2 Acetic acid fermentation

Different concentrations of ethanol were prepared by diluting molasses. These solutions were kept at different pH and temperature to determine the optimum condition of fermentation.

Increasing in the concentration of ethanol up to 10% resulted in gradually increase in ethanol production. Beyond 10% concentration growth of culture was inhibited resulting in decrease in production. Initial concentration of 10% and optimum temperature of 30°C was selected.

As pH increasing ethanol production is also increased. Best results were obtained at pH 4.5 where maximum production was noticed. Further increase in pH decreases the production. Acetobacter aceti can sustain up to 3 pH. In more acidic pH culture can't sustain. Better growth of culture was noticed at 4.5 pH.

To know the optimum temperature for ethanol fermentation, the solutions were kept at 25°C, 30°Cand 35°C with 10% initial ethanol concentration and fermentation was carried out for 5-7 days. A low yield was observed at 25°C. As shown at 30°C yield was maximized. However, increasing the temperature up to 40°C concentration of acid decreased. So, 30°Cwas selected as optimum temperature for ethanol production.

By using this parameters concentration up to 7-8% can be achieved.

Ethanol concentration was increase gradually. The order is closely equal to 2.

3. RESULT AND CONCLUSIONS

For ethanol concentration, best result obtains at 20% sugar concentration, 32°C temperature and 5.2 pH. And for acetic acid fermentation, 10% ethanol concentration, 30°C temperature and 4.5 pH are considered as optimum temperature. Saccaromyces cerevesia and, acetobacter aceti are used respectively.

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