Short Communication

Production of ethanol from *Carica papaya* (pawpaw) agro waste: effect of saccharification and different treatments on ethanol yield

AKIN-OSANAIYE, B.C.1, NZELIBE, H.C.2* and AGBAJI, A.S.1

¹National Research Institute for Chemical Technology Private Mail Bag 1052, Zaria, Kaduna State Nigeria.

²Biochemistry Department, Ahmadu Bello University Zaria, Kaduna State, Nigeria.

Accepted 23 March, 2005

A study was carried out on yeast fermentation of *carica papaya* (pawpaw) agricultural waste using dried active baker's yeast and brewer's yeast strain (*Sacchromyces cerevisiae*). The pawpaw considered as an agricultural waste was the tapped ripe pawpaw fruit harvested after the tapping of papain. Effect of different yeast strains on the percentage yield of ethanol was investigated. The effects of yeast concentration, saccharification and different nutrient supplements as they relate to the optimization of the ethanol yield were also carried out. The fermented pawpaw yielded ethanol contents of 3.83 to 5.19% (v/v). The reducing sugar in the pawpaw was determined before and after saccharification. The reducing sugar was highest after 48 h of saccharification using *Aspergillus niger*. The value recorded was 7.6 to 13.6 g/100g. Brewers yeast gave a higher ethanol yield than bakers yeast. Saccharification for 48 h coupled with nutrient supplements significantly increased the ethanol yield.

Key words: Paw-paw agrowaste, bakers yeast, brewers yeast, fermentation, pH, temperature, saccharification.

INTRODUCTION

Carica papaya (pawpaw) is one of the fruits commonly used as food and medicine in Nigeria. It is eaten as fresh fruit or processed into deserts (Desmond, 1995). The unripe matured pawpaw fruit is used for the production of papain by the making of incisions on the back of the fruits to get the latex for the papain production. This makes the fruit unattractive to the eyes and when allowed to ripe, the taste is not like the fresh fruit from pawpaw that has not been tapped and it is often bitter.

Ethanol production by fermentation faces competition with ethanol production from petroleum-based products as feedstocks. But with the increasing value of these petrochemical feedstocks, fermentation of ethanol is bound to receive more attention (Ahmeh et al., 1988). In this regard, use of renewable materials would be more economical, since they are cheaper and easily available.

Since the large quantities of pawpaw agro waste are available from plantations cultivated for papain production, their disposal can be a problem. Here we attempt to process the fruit waste into alcohol, which will have industrial applications.

MATERIALS AND METHODS

Materials

Pawpaw fruits were obtained from National Research Institute for Chemical Technology (NARICT), Zaria papain experimental garden. This was kept in a locker and allowed to ripe and soften. The ripe fruit was peeled and the seeds removed. One kilogram of this sample was pulverised with a blender, packed in plastic container and stored in the freezer for subsequent analysis. Dried baker's yeast (*Sacchromyces cerevisiae*) is the product of GYMA CARPENTRAS TEWEX 431184, France and was purchased from Sabon-Gari Central market, Zaria; while brewer's yeast (*Sacchromyces cerevisiae*) was obtained from Nigerian Breweries PLC, Kaduna.

^{*}Corresponding Author's E-mail: zeechuks@yahoo.com.

Table I. Effect of saccharification on the amount of reducing sugar in the pawpaw waste.

Period of saccharification (h)	Amount of reducing sugar (g/100 g)
0	7.6 ± 0.03
24	8.0 ± 0.12
48	13.6 ± 0.19
72	9.2 ± 0.31

Results are means of duplicate experiment.

Table 2. Effect of saccharification and simultaneous fermentation on the percentage ethanol yield of the pawpaw waste^a

Period of	Ethanol yield (%)	
saccharification (h)	Baker's yeast	Brewer's yeast
0 _p	4.41 ± 0.05 ^d	4.58 ± 0.03
24 ^c	4.57 ± 0.03	5.01 ± 0.03

 $[^]a$ Nutrient supplements added include 0.1 g KH₂PO₄, 0.5 g CaCl₂, 0.05 g MgSO₄, 0.1 g Na₂SO₄ and 0.1 g (NH₄)₂SO₄. 5% dried active baker's yeast or 1.09 x 10⁶ yeast cell/10 ml brewer's yeast was used. *A. niger*'s concentration used was 1.96 x 10⁶ cells/5 ml.

Analytical methods

The total reducing sugar content of the syrup was determined by dinitrosalicyclic (DNS) method described by Miller (1959) with slight modification. The pH of the syrup was determined and adjusted to 4.5 with 0.5 M NaOH. Saccharification was also carried out on the sample using *Aspergillus niger* for the period of 72 h and the total reducing sugar was determined using DNS method. The values were determined after 24, 48 h and 72 h of saccharification.

Fermentation

Fermentation was carried out on the sample by using two different yeasts (baker's and brewer's). 100 g of the sample was weighed into 250 ml conical flask and 100 ml of distilled water was added. The pH was adjusted to 4.5 and then pasteurized. The inoculum was added. Fermentation was allowed to proceed for 3 days at 30°C. At the end of fermentation, the alcohol was recovered by simple batch distillation using laboratory distillation unit. The alcohol content was determined by the determination of the specific gravity using a standard alcohol density table (AOAC, 1970).

The effect of the concentration of the yeast on the percentage alcohol yield was also investigated. Yeast supplements were added in the form of (NH₄)₂SO₄, KH₂PO₄, Na₂SO₄, CaCl₂ and MgSO₄ and their effects in relation to the percentage ethanol yield were determined. Also, the samples were saccharified using A. niger and then fermented. Simultaneous saccharification and fermentation was also carried out for the period of 72 h.

Table 3. Effect of different treatments of the pawpaw waste on the ethanol yield (v/v).

Treatment	% Ethanol yield (v/v)
Α	4.35 ± 0.08
В	4.61 ± 0.03
С	3.78 ± 0.06
D	4.02 ± 0.03
E	4.89 ± 0.07
F	4.47 ± 0.02
G	5.19 ± 0.03
Н	4.93 ± 0.02

- A = Substrate + Brewer's yeast $(3.56 \times 10^6 \text{ yeast cell})$ without nutrient supplement.
- B = Substrate + Brewer's yeast $(3.56 \times 10^6 \text{ yeast cell})$ + nutrient supplement listed in Table 2.
- C = Substrate + 10 g Baker's yeast , without nutrient supplements.
- D = Substrate + 10 g Baker's Yeast + nutrient supplement.
- E = Substrate + Aspergillus niger (mixed culture) at 0 hour + Brewer's yeast + nutrient supplement.
- F = Substrate + 10 g Baker's yeast + *Aspergillus* (1.09 x 10⁶ Yeast cell/5 ml).
- G = Substrate + Aspergillus niger $(1.09 \times 10^6 \text{ Yeast cell/5 ml})$ + Brewer's yeast $(3.56 \times 10^6 \text{ Yeast cell/10 ml})$ after 48 h of saccharification.
- H = Substrate + *Aspergillus niger* (1.09 x 10⁶ Yeast cell) + 10 g Baker's yeast after 48 h of saccharification.

RESULTS AND DISCUSSION

The values of the reducing sugars obtained from the agro-wastes in this study are shown in Table I. The fresh pawpaw waste has the lowest value (7.6 g/100 g) while the value significantly (P < 0.05) increased to 13.6 g/100 g after 48 h of saccharification. The ethanol yield when brewer's yeast was used is significantly higher (P < 0.05) than when baker's yeast was used. The value recorded was 4.18% and 3.83% (v/v) when 1.09 x 10^6 yeast cell/10 ml brewer's yeast and 5% baker's yeast were used, respectively. The percentage yield was increased when the concentration of the yeast was increased to 3.56 x 10^6 yeast cell/10 ml and 10%, respectively. This observation is consistent with the report of Morris and Sarad (1990), which indicated that the amount of yeast influenced ethanol production.

Table 2 recorded the alcohol yield from the simultaneous saccharification and fermentation of the pawpaw waste. It was noted that the percentage yield of the ethanol increased significantly (P < 0.05) after 24 h saccharification in both yeast strains. Also when different treatments were carried out in the pawpaw waste (Table 3). The highest ethanol yield was recorded when brewer's yeast was used for the fermentation after 48 h of saccharification with different yeast nutrient supplements. The ethanol yield increased significantly to a value of

^b0 h means the *A. niger* and the *S. cerevisiae* were added simultaneously.

 $^{^{\}circ}$ 24 h represent the period of saccharification before the *S. cerevisiae* was added.

dResults are means of duplicate experiment.

5.19% (v/v).

The results of this work have shown that paw-paw agro waste could serve as raw material for the production of biofuel, alcohol. Futher work will focus on the use of proteolytic enzymes and acid hydrolysis to release more fermentable sugars from the agro waste to optimize the ethanol production. The findings of this work recommend that alcohol can be produced from pawpaw waste obtained as papain by-product to maximize profit.

REFRENCES

Ameh JB, Okagbue RN, Ahmadm AA, Ikediobi CO (1988). Ethanol Production from Corn-cob Wastes and Grass-straw. Nig. J. Biotechnol. 6: 110 - 112.

- Association of Official Analytical Chemists (A.O.A.C.) (1970 and 1980). Official Methods of Analysis 13th Ed. Horwits, W., Ed. A.O.A.C., Washington D.C.
- Desmond R Layne (1995). The Pawpaw (*Asimina triloba* (L.) Dunal). In New Crop Fact Sheet, pp. 1-5.
- Miller GC (1959). Use of the Dinitrosalicylic Acid Reagent for the Determination of Reducing Sugar. Analytical Chemists. 31: 420 428.
- Morris W, Sarad RP (1990). Biotechnol. of Biomass Conversion: Fuels and Chemicals from Renewable Resources. p. 235.