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Reducing Sugar Production from Durian Peel by Hydrochloric Acid Hydrolysis

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***Abstract*—** Agricultural waste is mainly composed of cellulose and hemicelluloses which can be converted to sugars. The inexpensive reducing sugar from durian peel was obtained by hydrolysis with HCl concentration at 0.5-2.0% (v/v). The hydrolysis range of time was for 15-60 min when the mixture was autoclaved at 121 °C. The result showed that acid hydrolysis efficiency (AHE) highest to 80.99% at condition is 2.0% concentration for 15 min. Reducing sugar highest to 56.07 g/litre at condition is 2.0% concentration for 45 min. Total sugar highest to 59.83 g/litre at condition is 2.0% concentration for 45 min, which was not significant ($p < 0.05$) with condition 2.0% concentration for 30 min and 1.5 % concentration for 45 and 60 min. The increase in concentration increased AHE, reducing sugar and total sugar. The hydrolysis time had no effect on AHE, reducing sugar and total sugar. The maximum reducing sugars of each concentration were at hydrolysis time 45 min .The hydrolysated were analysis by HPLC, the results revealed that the principle of sugar were glucose, fructose and xylose.

***Keywords*—** acid hydrolysis efficiency (AHE), reducing sugar, total sugar

I. Introduction

AGRICULTURAL waste has caused significant problems. However, most waste has been transformed into other products particularly as animal food and fertilizer. This increases the value of the waste as well as decreases the amount of waste. The major components of agricultural waste are lignin, cellulose and hemicelluloses. Hemicellulose accounts for 10 to 40 percent of natural cellulosic biomass. Hemicellulose is primarily a polymer of pentoses and hexoses. These polymers can be easily reduced to monomeric sugars, xylose and glucose by using dilute acids under mild acid condition. In recent years, it has been demonstrated [1-4] that Dilute-acid hydrolysis was proven to be a fast and cheap method for producing sugar from lignocellulosic materials [5].

Thailand is one of the primary producers as well as a world exporter of fresh and frozen durian. Beginning in May and extending through August, the durian seasonally announces its presence in Thai markets. The massive amount of the peel is disposed as waste which could lead to environmental problems [6]. Durian peel could be transformed into valuable materials to be further produced commercially; such as particleboard components used as construction panels for energy conservation in buildings [7], biosorbent for removal of dry acid [8], removal of toxic heavy metals [9], activated carbon [10], carboxy methyl cellulose [11] and polysaccharide gel (PG) isolation, purification and characterization [6].

The main objective of this research was to experiment reducing sugar production from durian peel through the hydrolyzed treatment with hydrochloric acid at different concentration and different hydrolysis time at 121°C. This process transforms durian biomass into reducing sugar that can be used to substitute ethanol production and to decrease environment problems.

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II. Methodology

A. Sample Preparation

The raw materials used in the experiment were durians (*Durio zibethinus* Murray) collected from fruit markets in Chanthaburi province of Thailand during May – June 2010. The collected materials were then washed with distilled water for several times to remove all the dirt particles. The washed materials were cut into small pieces (1-2 cm), then dried in a hot oven at 60 °C until the constant weight was shown. After that, the sample was grounded with a blender into small pieces which could pass through a 500 micron screen. Finally they were packed into a sealable plastic bag and stored in desiccators for further use.

B. Proximate Composition

After it was cut into a uniform size, the sample was then analyzed for moisture, protein, fat, ash, crude fiber by the AOAC method [12] and carbohydrate was calculated as:

$$\% \text{ carbohydrate} = 100 - (\% \text{ moisture} + \% \text{ protein} + \%\text{fat} + \%\text{fiber} + \%\text{ash}) \quad (1)$$

C. Dilute-acid Hydrolysis

The 10% (w/v) of durian peel was hydrolyzed in autoclave at 121 °C, 15-60 min at 0.5 - 2 % (v/v) of HCl. The hydrolysis sample was centrifuged at 3,000 rpm for 20 min.

D. Analysis of Hydrolysate

The solid fraction was washed with distilled water and dried in a hot oven at 70°C until the constant weight was obtained. Then the percentage of weight determination for acid hydrolysis efficiency (AHE) was calculated using the following equation (2). The supernatant was collected and determined for reducing sugar using 3,5-dinitrosalicylic acid [13] and total sugar using phenol sulfuric acid [14]. The maximum reducing sugar from each acid concentration was determined for morphology by SEM and sugar polymers by HPLC using a YMC-Pack Polymine II column (conditions : 1.0 ml / min, 25 °C).

$$\% \text{ AHE} = \frac{\text{TS of initial hydrolysis} - \text{TS after hydrolysis}}{\text{TS after hydrolysis.}} \times 100 \quad (2)$$

whereas TS = Total solid

E. Statistical Analysis

The 4^2 factorial research design was carried out to study the effect of two factors for concentration (X_1) and hydrolysis time (X_2) on the two responses mentioned above. This experiment was designed in randomly order to minimize the effects of unexpected variability in the observed responses sixteen runs were required to cover all possible combinations of factor levels with three replicates. Experimental data were analyzed to fit the following regression model with interaction terms.

$$Y = \beta_0+\beta_1X_1+\beta_2X_2+\beta_3X_1X_1+\beta_4X_1X_2+\beta_5X_2X_2 \quad (3)$$

Where β_0, β_1, β_2, β_3, β_4 and β_5 are regression coefficients.

III. Results and Discussion

A. Proximate Composition

The durian peel in this study consisted of 6.92 % moisture content, 3.15 % protein, 4.01 % ash, 0.26 % fat, 27.81% crude fiber and 57.85% total carbohydrate.

B. Dilute-acid Hydrolysis

The results obtained from durian peel hydrolysis with HCl showed the AHE, reducing sugar and total sugar, between 70.78 to 80.99 percent, 23.39 to 56.07 g/litre and 27.14 to 59.83 g/litre respectively.

C. Analysis of Hydrolysate

Analysis of optimum condition by comparing means was employed using one-way ANOVA with Duncant by SPSS statistics 17.0. It was found the highest AHE to 80.99% at 2.0% concentration for 15 min. The highest reducing sugar for 56.07 g/litre at 2.0% concentration for 45 min. The highest total sugar for 59.83 g/litre at 2..0% concentration for 45 min. This finding was not significant ($p < 0.05$) with the condition at 2.0% concentration for 30 min and 1.5 % concentration for 45 and 60 min shown in table I.

D.Statistical Analysis

The hydrolysis with HCl regarding the increasing of concentration shows the relation to increased AHE, reducing sugar and total sugar, which is similar to that reported by Asli and Qatibi [15] who studied the hydrolysis of olive cake with concentration 0 - 4% (w/v) at 180 °C for 10 min. They found the increasing of sugar concentration when there is increasing concentration of acid. Hydrolysis time does not affect reducing sugar and total sugar shown in Table II.

TABLE I
Effects of Hydrochloric Acid Concentration and Hydrolysis Time to AHE Percentage, Reducing Sugar and Total Sugar.

Concentration (%)	Time (mins)	AHE (%)	Reducing sugar (g/litre)	Total sugar (g/litre)
0.5	15	76.41 [c]	23.39 [k]	27.14 [j]
	30	76.23 [c]	24.03 [jk]	27.60 [j]
	45	76.12 [c]	27.08 [j]	30.35 [i]
	60	70.78 [d]	31.81 [i]	31.86 [h]
1.0	15	79.20 [b]	33.24 [i]	40.47 [g]
	30	79.16 [b]	40.83 [gh]	41.30 [g]
	45	78.59 [b]	48.01 [dc]	49.52 [d]
	60	78.41 [b]	47.36 [dc]	49.64 [d]
1.5	15	79.91 [ab]	38.77 [h]	42.81 [f]
	30	79.59 [ab]	45.17 [ef]	56.58 [b]
	45	78.59 [b]	53.63 [ab]	59.56 [a]
	60	79.23 [b]	52.84 [abc]	59.36 [a]
2.0	15	80.99 [a]	42.55 [fg]	44.64 [e]
	30	79.88 [ab]	50.21 [cd]	59.51 [a]
	45	79.02 [b]	56.07 [a]	59.83 [a]
	60	79.33 [b]	52.51 [bc]	55.38 [b]

Different letters in the same low indicate significant differences, $p < 0.05$.

TABLE II
Correlation Coefficient of Durian Peel Hydrolysis by Hydrochloric Acid.

Factor	AHE	Reducing sugar	Total sugar
Concentration	0.730**	0.809**	0.828**
Time	*-0.345*	0.435	0.331

** Correlation is significant at $p < 0.01$
* Correlation is significant at $p < 0.05$

The decrease in hydrolysis time increased AHE with regard to durian peel can be absorb acid solution with increasing hydrolysis time, some part of hydrochloric acid in solution can be volatile.

The experimental data were analyzed through the factorial design to fit the regression models mentioned by Statistica 7 for AHE, reducing sugar and total sugar at hydrolysis with HCl which are given by Eqs. (4), (5) and (6) respectively:

$$Y_1 = 73.027 + 10.081X_1 - 0.043X_2 - 3.48X_1X_1 + 0.045X_1X_2 - 0.008X_2X_2 \quad (R^2 = 0.829) \quad (4)$$

$$Y_2 = -9.575 + 46.757X_1 + 0.705X_2 - 13.050 X_1X_1 + 0.031X_1X_2 - 0.006 X_2X_2 \quad (R^2 = 0.958) \quad (5)$$

$$Y_3 = -10.155 + 53.372X_1 + 0.810X_2 - 15.733X_1X_1 + 0.085X_1X_2 - 0.009X_2X_2 \quad (R^2 = 0.945) \quad (6)$$

The response surface plots for AHE, reducing sugar and total sugar of hydrolysis with HCl using Eqs. (4), (5) and (6) are given in Fig 1

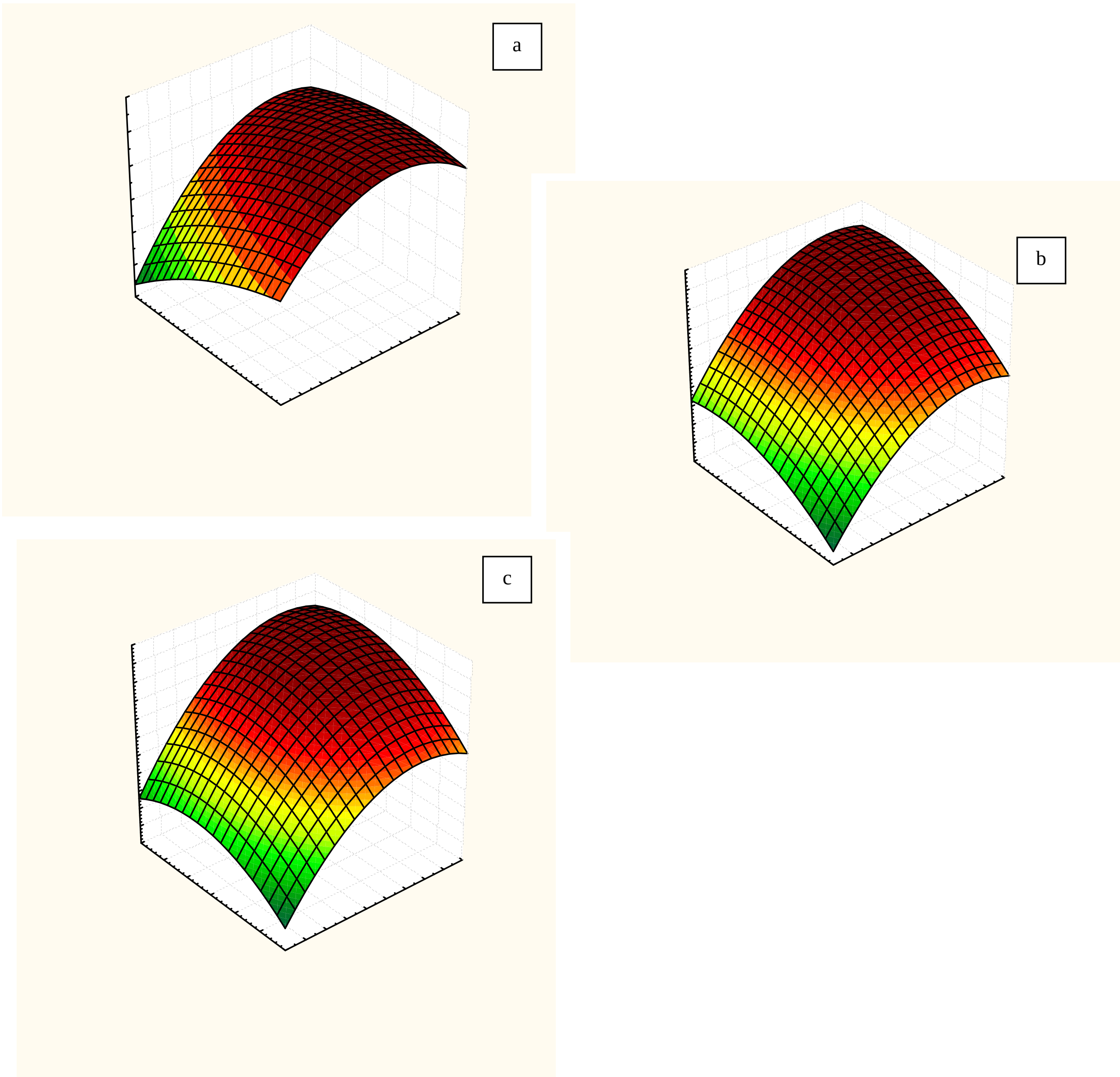

Fig. 1 Response surface plots for AHE (a), reducing sugar (b) and total sugar (c) of hydrolysis with HCl

E. Scanning Electron Micrographs (SEM)

The maximum reducing sugars of each concentration at hydrolysis time 45 min from hydrolysate was selected for morphology by SEM shown in Fig. 2 and sugar polymers by HPLC shown in Fig. 4 and Table III.

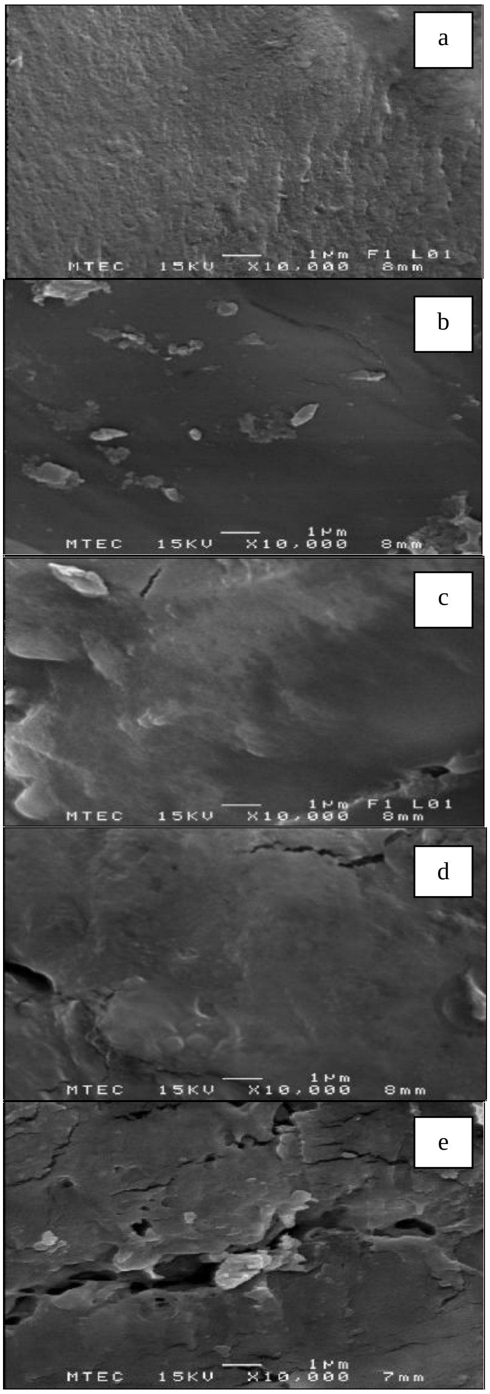

Fig. 3 SEM of effect from concentration of acid (a) powder of durian peel, hydrolysis with HCl concentration at 0.5% (b), 1.0% (c), 1.5% (d) and 2.0% (e) for 45 min

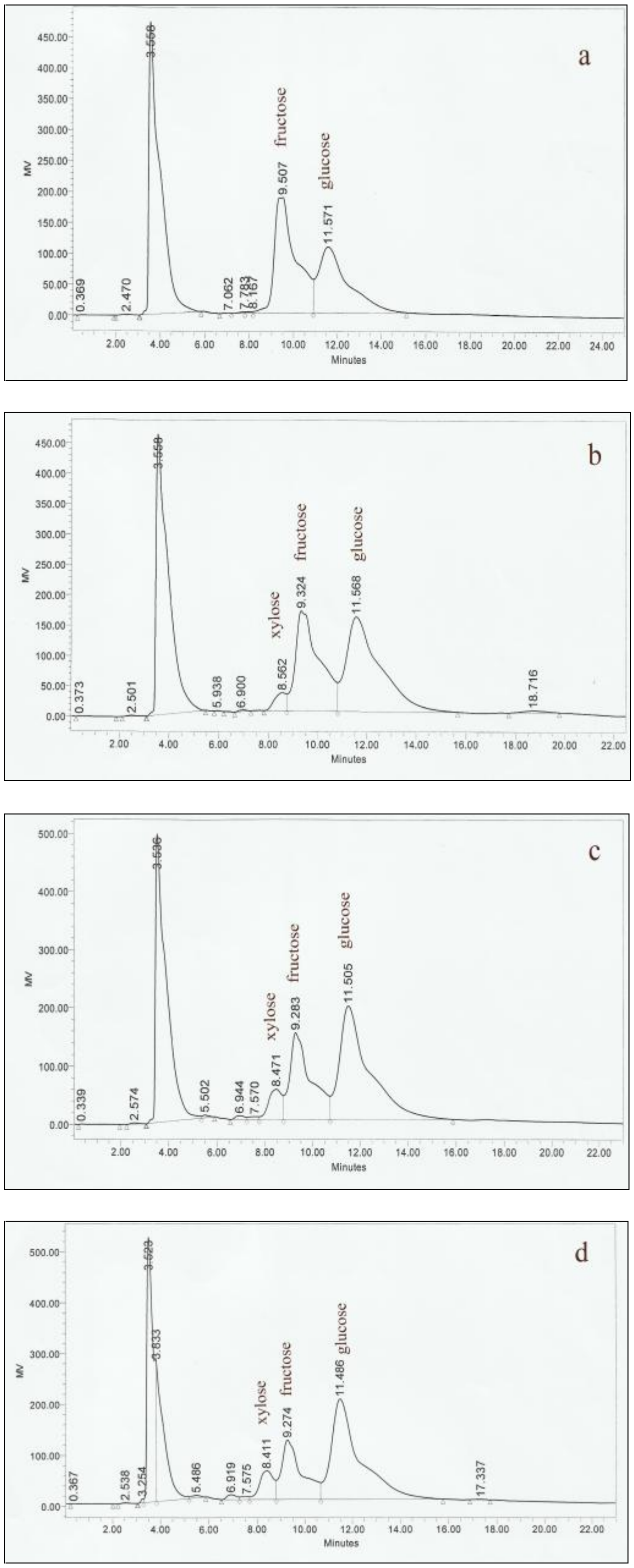

Fig. 4 HPLC chromatograms show peak of sugars in the durian peel hydrolysis with HCl concentration at 0.5% (a), 1.0% (b), 1.5% (c) and 2.0% (d) for 45 min

The figures from SEM showed that the structure of durian peel was destroyed by hydrolysis with HCl. Hydrolysis reactions of sugar polymers in a dilute-acid medium are very complex. The substrate is in a solid phase and the catalyst in a liquid phase. The mechanism of the hydrolysis reaction includes [16-18] : (i) diffusion of protons through the wet lignocellulosic matrix; (ii) protonation of the oxygen of heterocyclic ether bond between the sugar monomers; (iii) breaking of the ether bond; (iv) generation of the carbocation as intermediate; (v) solvation of the carbocation with water; (vi) regeneration of the proton with cogeneration of the sugar minomer, oligomer or polymer depending on the position of the ether bond; (vii) diffusion of the reaction products in the liquid phase if it permit for their form and size; (viii) restarting of the second step.

TABLE III
SUGAR ANALYSIS BY HPLC OF DURIAN PEEL HYDROLYSIS WITH HYDROCHLORIC ACID

HCl Conc. (%)	Time (min)	Glucose(G) (g/litre)	Fructose(F) (g/litre)	Xylose(X) (g/litre)	G+F (g/litre)	G+F+X (g/litre)
0.5	45	10.59[d]	13.01[a]	0.04[c]	23.60[b]	23.64[c]
1.0	45	15.52[c]	11.59[b]	2.60[b]	27.11[a]	29.70[b]
1.5	45	17.86[a]	9.62[c]	4.98[a]	27.48[a]	32.46[a]
2.0	45	16.94[b]	7.09[d]	5.43[a]	24.03[b]	29.46[b]

Different letters in the same low indicate significant differences, $p < 0.05$.

From reducing sugar analysis by DNS method found that almost every concentration at hydrolysis time 45 min have highest reducing sugar, exclude 0.5% concentration, must use the hydrolysis time long ago go up to 60 min because of low concentration make durian peel still don't hydrolyzed is like 1.0-2.0% concentration then selected every concentration at hydrolysis time 45 min and have AHE between 78-79% (~80%) was analysis sugar by HPLC showed that hydrolysis with HCl components are identical to glucose, fructose and xylose by glucose found amount most, next be fructose and xylose respectively showed in table III which differ from the hydrolysis of agricultural residues such as corn hull, rice straw, corn cob, sugarcane bagasse and rice husk which found glucose and cellobiose [19].

From the experiment reducing sugar production from durian peel through the hydrolyzed treatment with hydrochloric acid at 0.5-2.0% concentration for 15-60 min at 121 ◦C found that 1.5% concentration for 45 min have glucose, fructose and xylose at 17.86, 9.62 and 4.98 g/litre respectively by highest glucose and reducing sugar (glucose, fructose and xylose). Glucose is a main product obtained in the hydrolysated of durian peel, like the acid hydrolysated of sugar cane bagasse during the hydrolysis of sugar cane bagasse, other sugars are released to liquors, mainly glucose. This sugar can be proceeded from both hemicellulosic heteropolymers and cellulose [3].

This process transforms durian biomass into reducing sugar that can be used as substrate for ethanol production and to decrease environment problems.

IV. CONCLUSION

The effect of concentration of hydrochloric acid and hydrolysis time on reducing sugar production of durian peel found that glucose, fructose and xylose is a main product obtained in the hydrolysated durian peel. The model was found to predicting experimental conversion data for the hydrolysis of agricultural residue for ranges of acid concentration and hydrolysis time studied.

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