

Research Article

Production of Ethanol using Calliandra Shrub by Hydrothermal Pretreatment Method

Sachin Tomar ^{Å*} and S. Y. Adaganti ^Å^ÅDepartment of Chemical Engineering, SDMCET, Dhrawad, Karnataka, India

Accepted 25 November 2013, Available online 01 December 2013, Vol.3, No.5 (December 2013)

Abstract

In this work the feasibility of Calliandra shrub, a lignocellulosic material, for ethanol production was investigated by using hydrothermal pre-treatment method. Lignocellulosic materials are considered as a new source to produce second generation biofuels like bio-ethanol. The aim of this study was to obtain the best condition for the hydrothermal pretreatment of lignocellulosic biomass followed by acid hydrolysis and fermentation to produce ethanol. Optimum conditions were found to be: for hydrothermal pre-treatment pressure 6 Kg/cm², temperature 165°C, time 1 hour, for hydrolysis: pressure 1-1.5 Kg/cm², time 1 hour, 5% H₂SO₄, for fermentation: pH of 6, 5% yeast (*Saccharomyces cerevisiae*), temperature 37.5°C, time 4 days in the rotary shaker incubator at a speed of 144 rpm. The biomass was characterized by scanning electron microscopy (SEM), Fourier transform infrared spectroscopy (FT-IR), Thermal gravimetric analysis (TGA), chemical methods and ethanol obtained was tested by Gas chromatography–Mass spectrometry (GC-MS).

Keywords: Biomass, Calliandra, Pretreatment, Lignocellulosic.

1. Introduction

In view of continuously rising petroleum costs and dependence upon fossil fuel resources, considerable attention has been focused on alternative energy resources. Production of ethanol from biomass is one way to reduce both the consumption of crude oil and environmental pollution. One of our greatest challenges is to reduce our nation's dependence on imported petroleum. To accomplish this, we need a variety of alternative fuels; including ethanol produced from lignocellulosic materials. Ethanol is the most widely used liquid Biofuel. It is an alcohol and is fermented from sugars, starches or from cellulosic biomass. Cellulosic materials can be used to produce ethanol, which is an important, renewable liquid fuel for motor vehicles. Various lignocellulosic biomass sources such as agricultural residues, wood, municipal solid wastes, and pulp and paper industry wastes have the potential to serve as low-cost and abundant feedstocks for production of bio-ethanol fuel (Bothast RJ et al 1997). Pretreatment techniques have been developed for various end uses of biomass feedstocks (Zheng Y et al 2009, Kumar R et al 2008). The aim of this study was to obtain the best condition for the pretreatment (Das S et al 2000) of lignocellulosic biomass to reduce the sample size, break down the hemicelluloses to sugars and open up the structure of the cellulose component followed by acid

hydrolysis to convert cellulose portion into glucose sugar that is fermented to ethanol.

2. Materials and Methods

2.1 Collection and analysis of biomass

The stem required were collected from agricultural field locally and grinded into powder in a laboratory and particles passing through 40 mesh and retained on 80 mesh were collected and used as a raw material. The material was then oven dried at 105°C until constant weight and stored in a sealed plastic bag at ambient temperature for analysis. Chemical analysis was determined according to the standard procedures of Goering and Van Soest (1970), lignin, extractives and ash content by using TAPPI Standard Test Methods T 222 om-06, T 204 cm-07, T 413 om-06 respectively.

Thermal decomposition of a raw material was checked by using Q 600, TA instruments. The structure of the biomass was observed using Hitachi S-4800 (Hitachi, Japan) instrument, Scanning Electron Microscopy (SEM) images were obtained at 400x magnification. FT-IR spectrophotometer (Tensor 27, Bruker, Germany) was used to analyse Fourier Transform Infrared spectroscopy (FT-IR) of a raw material. Scans were recorded from 500 to 4000cm⁻¹ at a resolution of 2 cm⁻¹.

*Corresponding author: Sachin Tomar

2.2 Pretreatment

10gm of sample along with 500ml of water was taken in 5 litre capacity hydrothermal unit (Fig. 1) and maintained at pressure of 6kg/cm^2 with temperature of 165°C for 60 minutes. After 60 minutes the product was exploded by depressurizing to atmospheric pressure using blow down valve. The exploded biomass was collected separately.



Fig. 1: Hydrothermal unit



Fig. 2: Hydrolysis unit

2.3 Hydrolysis Method

The pretreated material was taken in a reagent bottle with 5% H_2SO_4 and sealed. It was then digested in hydrolysis unit (Fig. 2) for 60 minutes at a pressure ranging from 1-1.5 Kg/cm^2 .

2.4 Fermentation

Media was prepared by adding 5g of yeast extract, 7.5g of ammonium sulphate, 3.5g of potassium hydrogen phosphate, 0.75g of magnesium sulphate and 1g of

$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ to 500ml distilled water taken in a 500ml conical flask. Neutralized hydrolysate and the media prepared were added to it before inoculating the yeast to the sample. At a maintained pH of 6, 5% yeast (*Saccharomyces cerevisiae*) was inoculated and kept for fermentation at 37.5°C for 4 days in the rotary shaker incubator (Fig. 3) at a speed of 144 rpm. The fermented product was centrifuged to separate the biomass. The supernatant was distilled to separate ethanol and ethanol obtained was tested by Gas chromatography–Mass spectrometry (GC-MS).



Fig. 3: Rotary shaker incubator

3. Results and Discussion

3.1 The chemical composition of biomass used as a raw material was found to have 46% cellulose, 14% hemicellulose, 26 % lignin, 12% extractives and ash accounted for 2% respectively.

3.2 Fourier Transform Infrared Analysis

The FTIR spectra of biomass in fig. 4, based on the literature (Ling-Ping Xiao *et al* 2011, Hongjie Li *et al* 2012, kacurakova M *et al* 2000) it is observed that a broad band at 3426 cm^{-1} is attributed to $-\text{OH}$ stretch of polar groups linked by inter and intermolecular bonding of hydrogen, band between $2800 - 2950\text{ cm}^{-1}$ is due to methyl and methylene of cellulose, lignin and extractives. A shoulder at 1737 cm^{-1} implies for hemicelluloses fraction. A sharp band at 1626 cm^{-1} is due to bending mode of absorbed water molecules with cellulose fibres (Haiping Yang *et al* 2007). The band between $900-1200\text{ cm}^{-1}$ results in to polysaccharides. Band near 895 cm^{-1} represents $\beta - (1 - 4)$ glycosidic linkages.

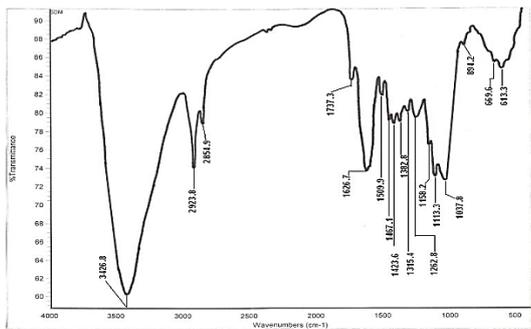


Fig.4: FT-IR spectra of biomass

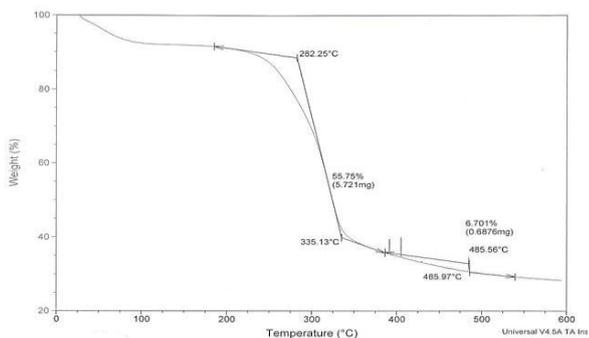


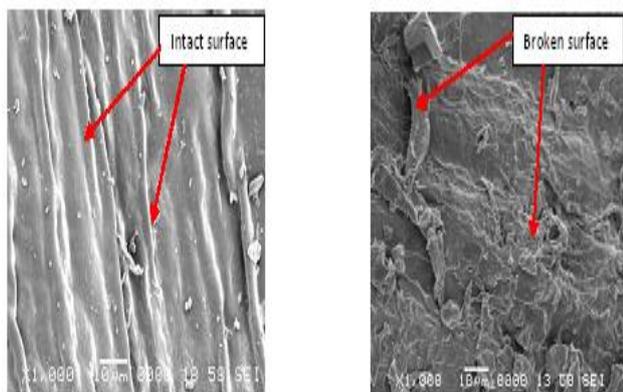
Fig. 5: TGA graph

3.3 Thermal Gravimetric Analysis (TGA)

According to TGA thermal stability temperature range is below 282°C. In TGA curve, initially the weight is 100%, as the temperature increases weight% starts decreasing slowly because it removes the moisture content. After 100°C degradation of hydrocarbon starts and at 282°C cellulose degrades completely.

3.4 Pretreatment

Figure 6(a) SEM image indicates compact and regular surface structure of biomass components (i.e. cellulose, hemicellulose, and lignin) without pores and Fig. 6(b) shows larger surface area with flexible linkages between fibres after pretreatment of lignocellulosic biomass.



(a) Untreated

(b) Pretreated

Fig.6: SEM images of biomass

3.4.1 Heating Curves of Hydrothermal Pretreatment

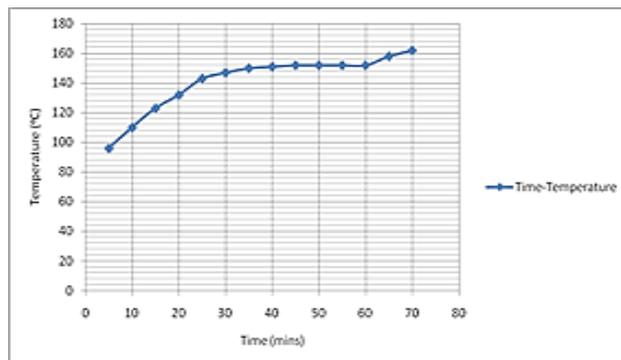


Fig.7: Time v/s Temperature graph

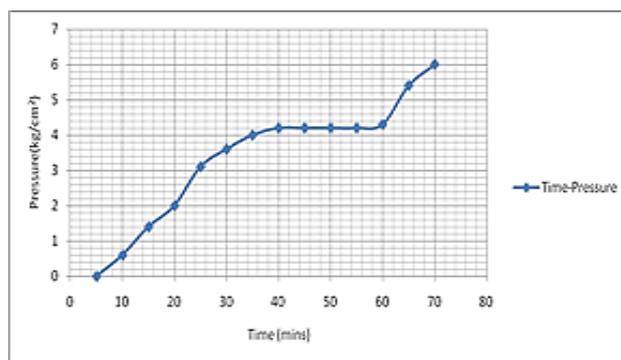


Fig.8: Time v/s Pressure graph

3.5 Gas Chromatography–Mass Spectrometry (GC-MS)

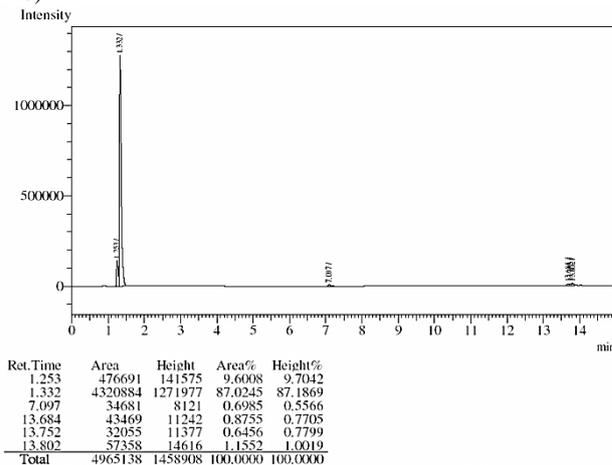


Fig. 9 GC-MS of distillate showing ethanol peak at retention time of 1.332 min.

4. Conclusion

This study suggests that Calliandra shrub can be used as raw material to obtain ethanol, which can be used as a fuel. The chemical analysis of biomass indicated rich cellulose and lignin content and less hemicellulose. Hydrothermal is found to be promising pretreatment

method for lignocellulosic biomass. Microscopy studies shows intact surface structure with strong network of fibrous polymers in a biomass. From the observation of FT-IR spectra, it was illustrated that cellulose, hemicelluloses and lignin gave characteristic bands. Optimum conditions were found to be: for hydrothermal pretreatment pressure 6 Kg/cm², temperature 165°C, time 1 hour, for hydrolysis: pressure 1-1.5 Kg/cm², time 1 hour, 5% H₂SO₄ and for fermentation: pH of 6, 5% yeast (*Saccharomyces cerevisiae*), temperature 37.5°C, time 4 days in the rotary shaker incubator at a speed of 144 rpm.

References

- Bothast RJ, Saha BC (1997), Ethanol production from agricultural biomass substrates. *Adv. Appl. Microbiol.*, 44, 261-286.
- Zheng Y, Pan Z, and Zhang R (2009), Overview of biomass pretreatment for cellulosic production. *International Journal of Agricultural and Biological Engineering*, 2, 51-68
- Kumar R, Singh S, Singh OV (2008), Bioconversion of lignocellulosic biomass: Biochemical and molecular perspectives. *J. Ind. Microbiol. Biotechnol.* 35, 377-391.
- Das S, Saha A K, Choudary P H, Basak R K, Mitra B C, Todd T, Lang S and Rowell R M (2000), Effect of Steam pretreatment of jute fiber on dimensional stability of jute composite, *Journal of Applied Polymer Science*, 76, 1652-1661.
- Ling-Ping Xiao, Zhao-Jun Sun, Zheng-Jun Shi, FengXu and Run-Cang Sun (2011), "Impact of hot compressed water pretreatment on the structural changes of woody biomass for bioethanol production *BioResources*, 6(2), 1576-1598.
- Hongjie Li, Jirong Lu, and Jianchu Mo (2012), Physiochemical lignocellulose modification by the formosan subterranean termite *Coptotermes formosanus shiraki* and its potential uses in the production of biofuels, *Bioresources*, 7(1), 675-685.
- Kacurakova M, Capek P, SasinKova V, Wellner N, Ebringerova A (2000), FT-IR study of plant cell wall model compounds: pectic polysaccharides and hemicellulose, *Carbohydrate Polymer*, 43, 195-203.
- Haiping Yang, Rong Yan, Hanping Chen, Dong Ho Lee, Chuguang Zheng (2007), Characteristics of hemicelluloses, cellulose and lignin pyrolysis, *Fuel*, 86(12), 1781-1788.