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Alkaline Peroxide Mediated Pre-Treatment of Corn Stover to Enhance Bioethanol Production

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Introduction:

Cellulosic ethanol technology is one of the most commonly discussed second generation biofuel Technologies worldwide. Cellulosic bio fuels are derived from the cellulose in plants, some of which are being developed specifically as energy crops rather than for food production Ethanol. Now a days, around 35% of total ethanol production is been produced by cellulose (Kumar andTewari, 2014).

There are four stages in the production of lignocellulosic-based ethanol: Pretreatment, Hydrolysis, Fermentation and Distillation.

Present research is focused to study an alternate pre-treatment approach to enhance bioethanol production by corn Stover. At alkaline condition, hydrogen peroxide produces superoxide radicals which have capacity to destruct lignocellulose sheath at room temperature. The objectives are aimed design the effective concentration of alkaline peroxide to treat corn Stover so as to improve bioethanol production through simple enzymatic hydrolysis followed by fermentation (SHF) process.

Review of literature:

History of Bio-fuel

The concept of bio-fuel was invented by Rudolf Diesel, whose invention now bears, envisioned vegetable oil as fuel source. Similarly, Henry Ford expected his had modal T to run on ethanol, a corn product. Eventually, in both Diesel's and Ford's case, petroleum entered the pictured and to prove be the most logical fuel source. Fuel involves bio-fuels which is a hydrocarbon that is made from living organism that humans can used to power something. Biofuel can also be made through chemical reaction, carried out in a laboratory or in an industrial setting, that use organic matter (biomass) to make fuel. (Galbe and Zacchi, 2009).

Research area:

- Bioethanol production from corn Stover using Saccharomyces Cerevisiae.
- Corn Stover hemicellulose acid hydrolyse has been utilized as a substrate for ethanol production using Saccharomyces Cerevisiae.
- Production of Ethanol fermented from renewable for fuel additives are known as biofuel.
- Bioethanol is being widely investigated as a renewable fuel source because in many respects it is superior to gasoline fuel.
- Ethanol provides energy that is renewable and less carbon intensive than oil.
- Bioethanol production.
- Agricultural waste.
- Process optimization.
- Fermentation process.

Applications of ethanol:

- Ethanol can be used as a transport fuel to replace gasoline.
- Ethanol can be used as a fuel for power generation by thermal combustion.
- It used as a fuel for fuel cells by thermo chemical reaction.
- Ethanol used as a fuel in co-generation system.
- Ethanol used as a feed stock in the as an additive for petrol.
- Over 80% of the words ethanol production is used in the fuel sector.
- It is also used in the practical lab as a sterilizer.

Aims and objective:

- To study the utility of hydrogen peroxide in lignocellulose pre-treatment and to discuss its effect on enzymatic hydrolysis and bioethanol production by corn Stover, objective set is designed as follows:
- Optimize the concentration of hydrogen peroxide in combination with sodium hydroxide for corn Stover treatment
- Enzymatic hydrolysis of prior treated corn Stover by commercial celluloses
- Fermentative performance of enzymatic hydrolysate by using Saccharomyces cerevisiae.

Methodology:

- **Pre-treatment to corn Stover by alkaline peroxide solution**
- Hence, 1 gm. of prior dried corn Stover biomass was added to 10 ml of alkaline peroxide with the varying concentrations of peroxides as 0.1 %, 1% and 10% (v/v) for 24 hrs. at room

temperature.

- After 24 hrs., the biomass was washed with distilled water, filtered and oven dried at 1050C.
- The weight losses of residual samples were recorded after treatment. Also the total carbohydrates content was recorded by phenol sulphuric acid assay explained elsewhere.
- The treated aliquots were subjected to record the released reducing sugar after treatment by DNS method explained elsewhere.
- The structural changes in the biomass were also recorded by FTIR method (SHIMADZU IR Affinity 1) by comparing the treated biomass residues with the native samples which reflected the complex polymer destruction (Singni and Negi, 2014).

Corn Stover biomass after ball illing and sieving

Enzymatic hydrolysis:

To discuss the effectiveness of peroxide treatment, it is necessary to hydrolyse the biomass through enzymes which detect the elevated yield of monosaccharide's.

Peroxide treated 0.5 gm. of biomass were added to 50 ml of 0.05 mm citrate buffer and around 100 µl of 12 FPU commercial cellulose was added to same.

Then the biomass was incubated at 500C at 120 rpm for 24 hrs.The released sugar was detected

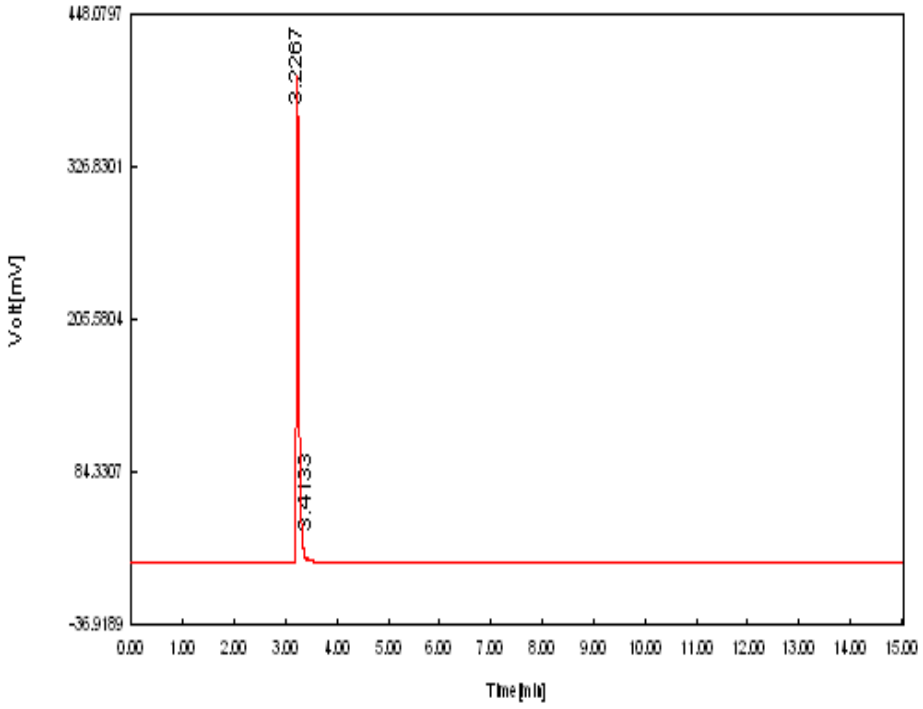
in aliquot as the available reducing sugar content detected by DNS method.
After treatment, the residual solid fraction also subjected to analyse the residual total carbohydrate by phenol sulphuric acid method and structural changes were recorded by FTIR method (Alya et al., 2013).

Fermentation process:
Prior treated hydrolyses from each treatment were subjected for fermentation by using Saccharomyces cerevisiae.
Approximately 1% ammonium sulphate was added to each treatment flask, and pH was adjusted to 4.5.
After sterilization around 4% of prior grown Saccharamyces cerevisiae culture was added to each flask and all samples were incubated for 72 hrs. at 370C at rotary shaker with 120 rpm.
After 72 hrs., fermented broths were filtered and ethanol was detected by spectrophotometer (Shimadzu UV 200) by dichromate assay and confirmed by Gas chromatography (Ricke, 2012).

The list chemicals used for analysis given as follows:
- Hydrogen peroxide (30 %): Further diluted in distilled water as per the requirement of experiments
- Celluloses
- Citric Acid (AR grade)
- Sodium Hydroxide (AR Grade)
- Malt Extract
- Glucose
- Yeast extract
- Peptone
- Hydrochloric Acid
- Dinitrosalicyclic Reagent
- Dichromate Reagent

Result and Conclusion:

1) Analysis of ethanol by Gas Chromatography:



	Pretreatment	of corn Stover	biomass by al	kaline peroxid	e
Alkaline peroxide	**Initial weight**	**Residual weight**	**Weight loss**	**Total carbohydrat e**	**Reducing sugar**
0.1%	1gm	0.7gm	0.3gm	141.61µg/ml	25µg/ml
1%	1gm	0.7gm	0.3gm	201.17µg/ml	23µg/ml
10%	1gm	0.7gm	0.3gm	230.88µg/ml	10µg/ml

Conclusion:

Alkaline peroxide is one of the strong oxidising agent has potential to solubilise lignocellulosic biomass viz. Corn Stover.

However, the lignocellulose destruction was depends on the concentration of peroxide in the reagent, as the concentration of peroxide increased, the yield at enzymatic hydrolysis and fermentation were found to be increased.

The increased yield of hydrolysis and fermentation with increased peroxide concentration also

suggested that the inhibitory actions of phenolic derivatives were found absent during enzymatic and yeast activity. It also proved the advantage of the method over acid and temperature based pretreatment methods.

The structural changes detected by FTIR methods described the target functional groups of the alkaline peroxide reagents vize. alcohol, ketone, esters and carbon –carbon stretch. So irrespective of specificity of substrate, the reagent can be useful for pretreatment of other lignocellulosic biomass viz. Cotton stalk, sorghum stalk.

Advantages:

Exhaust gases of ethanol are much clearer.

It burns more cleanly as a result of more complete combustion. Any plant can be used for production of bioethanol it only has to contain sugar and starch.

Bioethanol is a much cleaner fuel than petrol.

Ethanol can be produced from renewable biomass resource. Ethanol can be blended into normal gasoline up to 10% without any engine modification.

Flexible fuel cars can run on any mixture of gasoline and ethanol from

i.e. 0 to 85% ethanol.

Ethanol is renewal resources.

Challenges:

It initiates the release of more carbon emission into the surroundings, utilizes more water to irrigate the plants, uses more fertilizer for farm fields, and destroys more wildlife habitats.

For bioethanol to become more sustainable to replace petrol, production process has to be more efficient.

Currently most of the ethanol being produced uses either sugarcane on rice as its raw material.

The current economic downturn, difficulty in obtaining financing, insufficient government support, lack of security of long term biomass supply, and technological limitations**.**

Future directions:

The traditional strains of S.cerevisiae uses in the production of 1GE continue to be studied to increase yield, productivity, and tolerance to stress.

New cultivation techniques are also being developed with the implementation of S. cerevisiae flocculent strains.

The use of these engineered strains allows the fermentation to continue because the microorganism has the capacity of auto flocculation, setting at the bottom of the tank, and

allowing higher productivity of ethanol.

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