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Study of Biodegradation of waste printed paper by enzymes and Conversion of sugar(s) derived from paper degradation into alcohol

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Abstract--We treated laser printed waste paper in which, we degraded paper using enzymes like cellulase and xylenes and this process is safer and ecofriendly as compare to chemical degradation of paper (Woodward, J.L., Stephan, M., Koran, L.J., Wong, K.K.Y. and Saddler, J.N.,1994). In addition, enzymes used during process generated monosaccharide and other reducing sugar. The sugar can be further subjected to bioconversion applying microorganisms, e.g. Saccharomyces cerevisiae provided value added by product (Bio-fuel).

I. INTRODUCTION

Paper is usually made from wood pulp and it is mainly consisted of cellulaseand Xylanase.Papers are widely used everywhere, it poses great environmental threat, so recycling of printed paper or conversion of waste printed paper into value added commodities, is a need of time (Lee Chee, K., Darah, I. and Ibrahim.C., 2011).The processes of recycling and use of waste paper must be environmental friendly. We used waste printed paper and subjected it to enzymatic degradation for production of sugar(s) which might further be utilized for different microbial products like food, yeast, and Organic acid and fuel production. The industries produce lots of waste of difference types, papers and textiles industries during their processes produce a large quantity of chemical waste these compounds add to the C.O.D. of the waste water and such water can't be directly disposed in to the natural resources. We performed experiments of recycling of paper in which we degraded paper using enzymes like cellulase and xylanase and this process is safer and ecofriendly as compare to chemical degradation of paper. In addition, enzymes used during process generate monosaccharide and other reducing sugar this contributes less C.O.D. as compare to the chemical processes. The sugar can be further subjected to bioconversion applying microorganisms, e.g. *Saccharomyces cerevisiae.*

II. MATERIAL

Acetate buffer (pH = 5.6) = 100 ml(Robert, C.editor.,1974.)

Enzymes[Xylanase(S.R.L.)]

Cellulase(Cellusoft Maps India) Printed papers.

Abressive (porcelain pieces)

Detection of sugar:-Detection of reducing sugar by Benedict test and Detection of xylose sugar by Bial's test.

Sugar(s) derived from degraded paper. Degraded paper system was filtered. Nitrogen source.

$NaNO_3$Culture of *S.cerevisiae* producing alcohol. Glucose yeast extract Broth (GYE). Sterilizable polycarbonate container of 50ml. Distillation unit

III. METHODOLOGY

Printed papers were cut (0.5g) and placed into 25ml,0.2M acetate buffer(pH 5.6) flask(flask E) with 0.2g xylanase (1200 units) enzyme and abrasive. In second flask (flask T) printed papers were cut (0.5g) and placed into 25ml,0.2M acetate buffer(pH 5.6) flask– with 0.2g cellulase(1200 units) enzyme and abrasive. Third control flask (flask C) was prepared with printed papers (0.5g) containing 25ml, 0.2M acetate buffer (Ph-5.6) without enzyme. These three flasks (flask – C, T and E) were placed on rotary shaker (80 rpm) at35°C±5°Cfor one hour. Content of these three flasks – E,T,C were filtered through nylon filter and centrifuged at 5000 rpm

for 10 mins. The supernatant of – E,T,C were tested by Benedict's test & Bial's test(Benedict, S. R.,2009). Sugar(s) solution derived from degradation of paper transferred into 50ml polycarbonate sterile containers. To this medium 0.5% nitrogen source $NaNO_3$ was added .*S.cerevisiae* culture was isolated from grapes was inoculated into G.Y.E. broth and incubated for 24 hours at 25±5ºC temperature.10^6cells/ml was inoculated into the sugar solution derived from paper degradation and added with the nitrogen source. The containers were closed tightly. They were closed with polythene plastic pieces. After 7th day's fermentation medium was distillated into distillation unit at 78ºC for two hours (Collins, Ed., Excel *International GCSE* Biology, *Student Book)*. Ethanol was qualitatively confirmed by F.T.I.R.

VI.RESULT

Results of control flask (flask – C):-

No sugar could be detected by Benedict's test and result of bial's test was negative which clearly show that paper was not degraded. The evidence are given below:

Figure:1

Benedict's test of control flask

Figure:2

Bial's test of control flask

Result of flask – A :-

In Benedict's test, development of brick red precipitation were observed which clearly indicated the production of reducing sugar.

In bial's test, development of bluish black precipitation are for this experiment are as given below:

Figure:3

Benedict's test of flask – A

Figure:4

Bial's test of flask – A

Result of flask – B:-

In Benedict's test, development of brick red precipitation were observed which clearly indicated the production of reducing sugar.

In bial's test formation of precipitation was not observed which clearly indicated that the cellulose was not able to produce pentose sugar. The evidence for the same as given below in photographs.

Figure:5

Benedict's test of flask – B

Figure:6

Bial's test of flask – B

In figure -1, 2 the system for paper degradation has been shown in below figure

Figure:7
Flask-1 Enzyme

Figure:8
Flask -2 Control

Reducing sugar was confirmed by Benidict's test as shown in the figure no-3,4

Figure:9
Positive Benidict's test

Figure:10
Control

Paper treated with xylanase showed The presence of xylose was detected by Bial's test figure- 5,6

Figure:11

Positive Bial's test

Figure:12

Control

Fermentation of paper waste sugar for ethanol production.

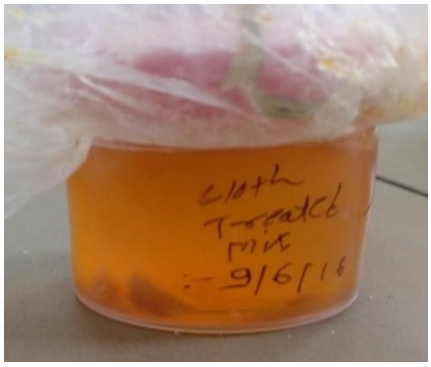


Figure:13

Polycarbonate fermenter for ethanol production

Figure:14 Distillation unit for ethanol recovery

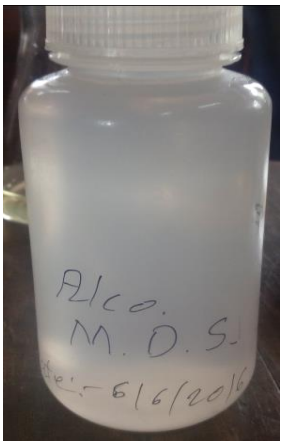
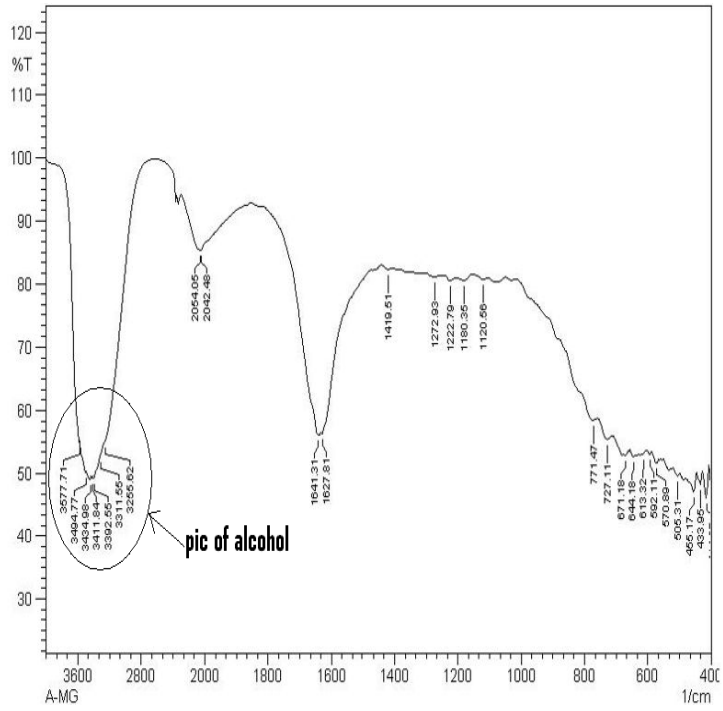


Figure:16 Recovered alcohol



Graph:1 F.T.I.R. of product (Ethanol).

VI. CONCLUSION

Untreated papers were not degraded.

In flask-E, printed papers were found degraded which indicated presence of xylan (hemicellulose) in paper.

In flask-T, printed papers were also found degraded which indicated the presence of cellulose in paper.

Thus, both xylan and cellulose were present in paper and after degradation, yielded the reducing pentose sugar.

This reducing pentose sugar could be exploited as a raw material for industrial products.

Paper could be degraded to sugars both by cellulase and xylanase.

The paper was degraded to reducing sugar was detected by Benidict's test. Xylanase reduced the paper to xylose which was detected by Bial's test. The sugar produced from the waste was converted to value added fuel –ethanol using yeast*: Sachharomyces cerevisiae.*

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