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# Exploring Esterification and Pyrolysis Pathways for Sustainable Biofuel and Biomass Production

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***Abstract:*** ***The recent Years Increase the demand of the biofuels along with need of reduce the production cost of Improving the environmental sustainability of the biomass production or the biofuels. Biomass is on area of importance to reduce the overall uses of fossil fuels. In which the focusing the kinetic properties or the optical property of the production of the Biofuel or Biomass production. Process optimization and reaction kinetic model development were carried out for two stage esterification transesterifications... reactions of waste biomass. This study focused on these traditional processes due to their techno- economic feasibility- Which is an important factor before deciding on a type of Feedstock for Industrilisation.Since esterification reactions are acid catalyzed the synthesis of an efficient acidic catalyst will be very helpful for pretreatment. esterification of acetic acid which was used model reaction in Stabilization of biomass. The esterification is a potential route to remove the organic acid in biomass reacting them with alcohol Present in biomass with added alcohol. Water is a product of the esterification reaction. The concentration of acetic acid used to acidification; acetic acid is a common chemical species in biomass & was used as the model compound. The research compared the quality of slow pyrolysis produced. biomass collected. by condensing pyrolysis vapor using atomized ethanol (EtOH).Were also carried out for the analysis of biomass produced.***
***Keywords: optimization biofuels, kinetic, esterification, transesterifications, techno- economic feasibility, ethanol (EtOH).***

## I. INTRODUCTION

Alternative fuels developed form renewable feedstocks are gaining market share recently. Biofuel is recognized as a green fuel is the one of the alternatives of the vegetable fuel. The demand for renewable energy sources has made biofuels on attractive alternative that can reduce the consumption of the traditional fossils fuel.

Development of second-generation bioethanol processes will not enable replacement of diesel or jet fuels. For these applications new processes, or third generation biofuels must be developed to which we will here refer to as advanced biofuels. Presently the most widely used biofuel is ethanol produced from sugarcane, corn, wheat but the use of feedstock that can be used for food production prevents this process form expanding further. Bioenergy represents the utilization of biomass as starting material to produce sustainable fuels.

The comprehensive idea on the developing technologies for bioenergy production which includes cell surface engineering of yeast to produce bioethanol from various biomass resources and the enzymatic and whole cell catalyzed transesterification of plant oils to biodiesel fuel.

Biofuels have a closed loop for the CO2 that is the main green house gas and beside that they can contribute to the reduction in the emission of toxic gases such as SO2, SO3 and carbon. Biomass has considered as a green fuel however is production process as resulting in high environmental impact.

The yeast saccharomyces cerevisiae is workhorse in the current biofuel industry as it is used for production of esterified.

### *A. Types Lignocellulosic Biomass*

Examples- Maize, straw, rice, wheat straw, corn stalks, sugarcane, corn stover

For biodiesel production, lignocellulosic biomass constitute is largest renewable resources.

Ethanol and esterified production through bioconversion of lignocellulosic biomass, have been already successfully achieved by many research worldwide laboratory scale, industry scale. The amount of residue is obtained from agricultural, industrial, forest sources.

Table 1: List of materials

| Material's | Quantity |
|---|---|
| Potato | 200gm |

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| | |
|---|---|
| Agar | 4gm |
| soil | 0.5gm |
| *Sacchromyces Cerevisiae* broth | 19ml |
| Corn | 10gm |
| Honey | 19ml |
| Sugarcane Juice | 6ml |
| Molasses | 40ml |
| Gram Straw | 42gm |
| Jaggery | 160gm |
| Distilled Water | 250ml |

*B. Equipment*

Water Bath[11], Incubator[12], PH meter[13], Autoclave[14], Calorimeter[15], Weighing Balance[16].

## II. METHOD

*A. Culture Media*

Preparation of culture media

Potato Dextrose Agar it is a frequently used microbial growth media for cultivation of malfeasants and other fungi. Take 200gm of potato for 1litre of PDA media preparation. wash the potato to remove dirt. Peel of the skin and dice them. Add the pieces to 1Litre of distilled water. Boil for 20-25min on hot plate. Filter / collect the extract through the muslin cloth. Then add 3-4 agar. Then autoclave 121°C for 15 PCI for 15 min. Then Cool It. Pour into 2 Petri plate then Solidify. Add yeast in one Petri dish and another add pinch of soil. Incubate, The Petri dish at 37°C for 3-4 days.

*B. Detection of mold*

Preparation of potato dextrose Broth PDB Take 200 gm of potato for 1 Liter of PDB media preparation wash the potato remove dirt. Peel off the skin. and dice them Add the pieces to 1 Liter of water. Boil for 20-25 min on Hot plate. Filter the extract through the muslin cloth. Then autoclave 121 °C for 15 pci for 15 min. Then cool it. Then, add 250 mL of PDB into conical Flask. Then Inoculate yeast into PDB by using sterile inoculating loop and another flask inoculate yeast which are present
in soil. Incubate the conical flask at 37°C for 3-4 days. Then detection of mold.

*C. Acid Fast staining*

The most Common staining technique used to identify acid fast bacteria is the Ziehl Neelsen Stain in which the acid-fast species are stained bright red. Firstly Air-dry Heat fix a thin film of microorganism on slide. Flood the slide with carbolfuchsin dye. Dry Heat for 2 min. cool and rinse with water wash the top and bottom of slide with water and clean the slide bottom well. Counterstain with methylene Blue for 30 sec to 1 min. Then observe under the microscope, Then High content of my colic acid in their cell walls. So Acid fast bacteria will be red in color. Acid fast bacteria was found.

*D. Fermentation Tank*

A fermentation tank is the vessel used to house work and yeast in the production of beer fermentation is. based on the principle of Anaerobic respiration for deriving energy from the breakdown of carbohydrates Such as glucose. In this process. glucose is first broken to pyruvate by glycolysis. The pyruvate is then converted to alcohol or lactic acid along with the regeneration of NAD. Fermentation of sugar by saccharomyces cerevisiae for production of ester fire in a batch experiment was Conducted to Improve the performance. of the Fermentation process. The thermotolerant ability of *saccharomyces cerevisiae* to grow and ferment glucose at elevated temperature, like the optima for Saccharification was Investigated.

The two essential raw material required for ferment carbon sources for ex. starch, cane molasses, beet molasses etc. Nitrogen sources ex. corn, Steep liquor, urea These are raw material extracted from venous Sources.

fermentation of biomass by Saccharomyces cerevisiae for production OF biofuel to improve the performance of the fermentation process the thermotolerant ability of *saccharomyces cervices* to grow and ferment glucose at elevated temperature Base of Fermentation tank is compose with wheat grain, corn, sugarcane Juice, yeast, molasses, Honey, Jaggery and water respectively.

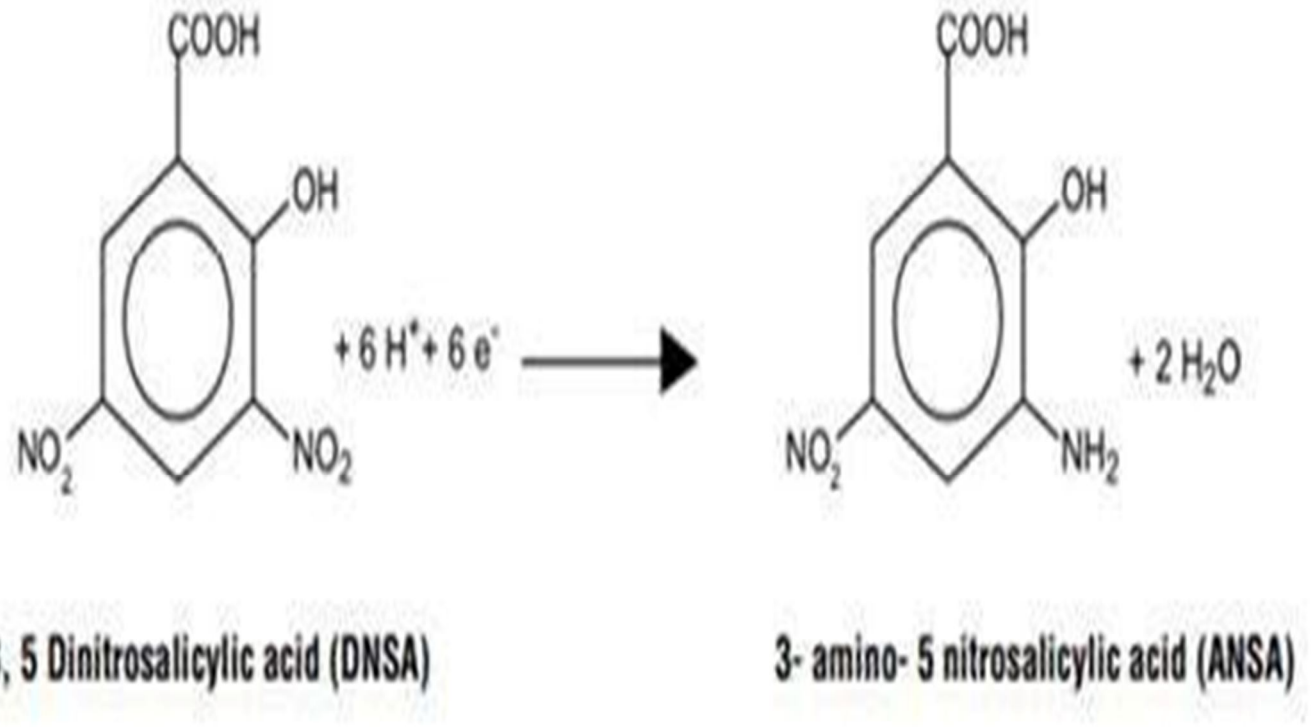
Wheat is composed of 70% Starch, 10% crude protein and remaining part composed with water, fat, ash and fiber, soaking and fermenting grain improve their nutritional benefits corn is a primary biomass source being used for producing cellulose ethanol. Sugar cane juice used for production of biofuel like ethanol molasses is mainly used as a Source of sugar for the fermentation. It is a used as a microbiological energy source. Honey can work to. Feed yeast. It helping in to produce the neutral by product of carbon. dioxide and alcohol Jaggery contain. carbon sources as compared with Sucrose It provide feed to yeast. *Saccharomyses cerevisiae* & seethe production of acetyl co A derived form n-butane, fermentation were performed in 250 mL borosilicate glass bottle, Containing wheat grain, sugar cane Juice, Jaggery and water. yeast, Honey

The maximum biofuel production rate were observe between 30 to 45℃ with difference. Initial glucose concentration increase Substrate supply did not. improve the specific ethanol production rate when pH was not controlled. PH 4.0-5.0 was optimal. range for biofuel production process a changes in the main fermentation pathway was observe with various PH range. The Highest specific biofuel production rate at pH 4.0 formation of acetic acid increase when PH was below 4. When temp was increase to us of the System still show High cell growth and production of Biofuel.

*E. DNSA Test*

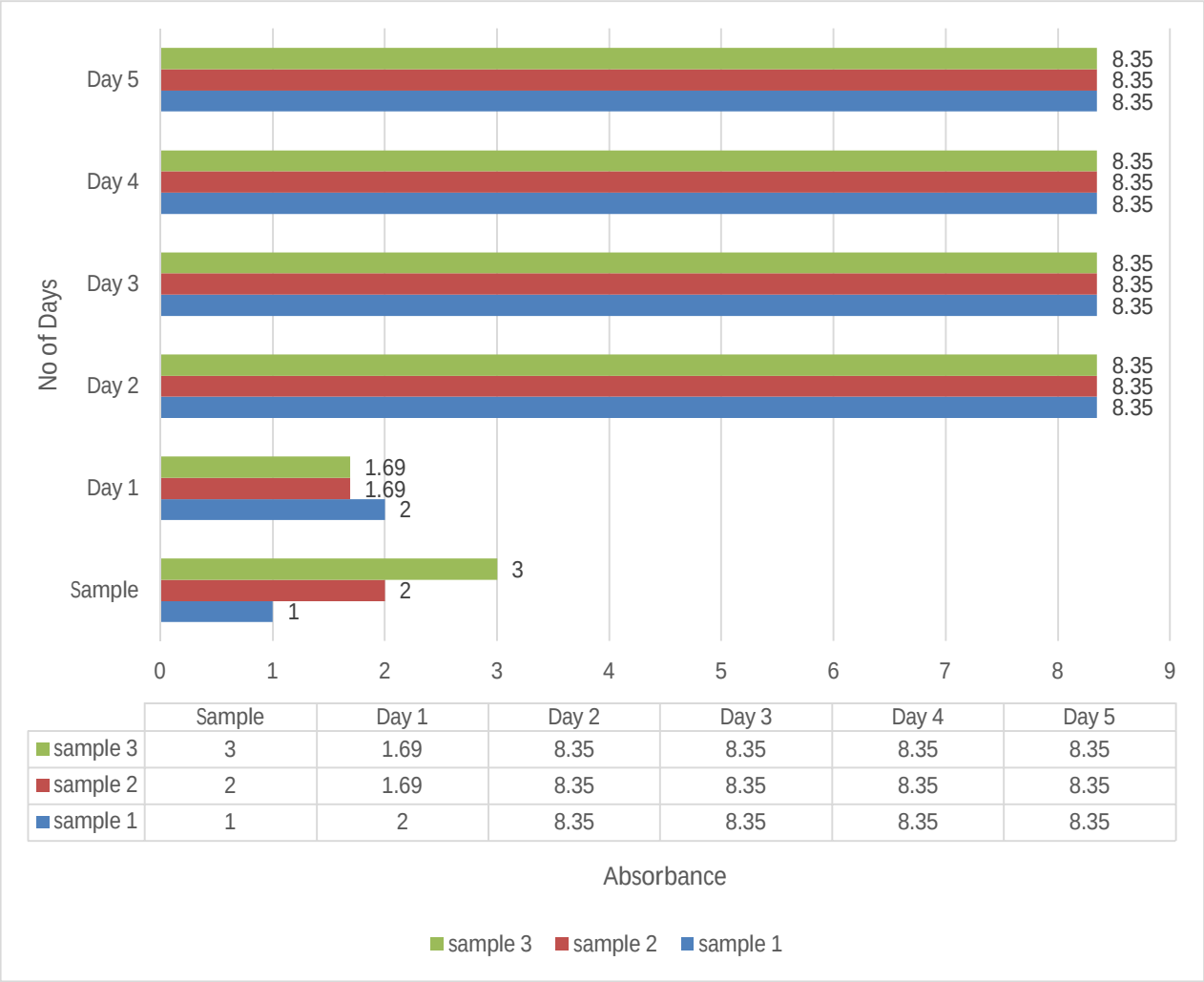
3,5-dinitrosaalicylic acid is used extensive In biochemistry for the estimation. of reducing sugar, it detect the presence of free carbonyl group of reducing sugars. This involves the oxidation of aldehyde functional group and the ketone functional group.

In amber colour bottle 1 gm of 3,5-dinitro Salicylic acid slowly add is gm of Sodium potassium tartarate phenol, Then add 10 gm of NAOH. Slowly added and continuous shaking in 100 ml of Water store in Dark place Prepare test tube for test sample and one control. Take 1 mL of dinitro solution and odd 4 mL of reducing sugar Sample in test tube. Boil in water both for 15 min at 70℃ temp. Get reading for 5 days to cheek the Stability of reducing sugar of it.



Graph by colorimetric analysis and Observation Table

The given spectra of the graph indicate that the esterification of the three sample is done for this we did this same procedure of each three sample by conjugative 5 days.




| | Sample | Day 1 | Day 2 | Day 3 | Day 4 | Day 5 |
|---|---|---|---|---|---|---|
| sample 3 | 3 | 1.69 | 8.35 | 8.35 | 8.35 | 8.35 |
| sample 2 | 2 | 1.69 | 8.35 | 8.35 | 8.35 | 8.35 |
| sample 1 | 1 | 2 | 8.35 | 8.35 | 8.35 | 8.35 |

*F. Phytochemical Test*

The confirmatory qualitative phytochemical screening of plant extract was performing to identify The main classes of compound ( Tannins ,Saponins , glycosides ,flavonoids , alkaloids ).

| SR NO | CHEMICAL TEST | OBSERVATION | INTERFACE | RESULT |
|---|---|---|---|---|
| I) | Detection of carbohydrates<br>1) Molisch's Test<br>2 ml offiltrate,2 drops of alcoholic solution of <- naphthol<br>Is added . shake well . add 1 ml of H2So 4 along the side ,cool<br>Violet ring at the junction of | Violet ring are Junction of two liquids | Presence of carbohydrates | The test is Positive. Shown in fig no-25 |

| | | | | |
|---|---|---|---|---|
| | two liquid .B<br><br>2)Fehling's Test<br>1 ml of filtrate is boiled ,<br>1 ml etching solution A&B<br>Formation off red precipitate.<br><br>3)Barfoed's Test<br>Sample + 2 ml Barford's reagent boil water bath reddish brown colour obtain | Red precipitate<br><br>Radish brown colour Obtain | Presence of carbohydrates<br><br>Presence of carbohydrates | The test is Positive. Shown in fig no-26<br><br>The test is Positive. Shown in fig no-27 |
| II) | Detection of flavonoid<br>1)Alkaline reagent Test<br>Extract is treated with 10% NAOH solution formation of intense yellow colour indicate presence of flavonoid.<br><br>2)NH4OH Test<br>3 ml of extra is 10% NH4OH solution development of yellow fluorescence indicate a positive test<br><br>3)Mg Turning Test<br>Extract treated with mg turning & add CONC. HCL to this solution add 5 ml of 95% ethanol . formation of crimson red colour indicate flavonoid . | Instance yellow colour<br><br>Yellow fluorescence<br><br>Crimson red colour | Presence of flavonoid<br><br>Presence of flavonoid<br><br>Presence of flavonoid | The test is Positive. Shown in fig no-28<br><br>The test is Positive. Shown in fig no-29<br><br>The test is Positive. Shown in fig no-30 |
| | 4)Zn Test<br>2 ml extract were treated with 2n dust and conc. HCL. Development of red | | | |

| | | | | |
|---|---|---|---|---|
| | colour indicate presence of flavonoids . | | | The test is Negative |
| III) | Detection of Tannins<br>1)Ferric Chloride Test<br>About 50mg extract Is dissolved in distilled water few of nature 5% ferric chloride solution is added<br><br>2)Gelatin Test<br>A little quantity of extract is dissolved in distilled water and 2 ml of 1% solution of gelatin containing 10% sodium chloride is added to it . development of white precipitate.<br><br>3)Lead Acetate Test<br>A small quantity extract dissolved in dist. H2O. 3 ml of 10% lead acetate solution is added . A bulky white precipitate indicate<br>4)Shinoda Test<br>A little quantity of extract dissolve in alcohol , few fragment of , mg . turning and CONC . HCL added . if any pink or crimson added colour develops | Formation of blue , green , violet colour | Presence of tannins | The test is Positive. Shown in fig no-31<br><br>The test is Negative<br><br>The test is Positive. Shown in fig no-32<br><br>The test is Negative |
| iv) | Detection of glycosides .<br>1)Borntrager's Test<br>2 ml of filtrate Hydrolysate 3 ml of ethyl acetate is added and shaken , ethylate layer is separated and 10% , ammonia solution's added to it . formation of pink colour .<br><br>2)Legal's Test<br>20 mg of extract is dissolved in pyridine sodium nitro preside solution is added & mad alkaline using 10% sodium Hydroxide solution . | Pink colour | Presence of glycosides | The test is Negative<br><br>The test is Positive. Shown in fig no-33 |

| | | | | |
|---|---|---|---|---|
| v) | Detection of Amino Acid<br>1)Ninhydrin Test<br>To 1 ml of sample add 5 drops of ninhydrin reagent heated in boiling water bath for 2 min A purple colour indicate the presence of amino acid's<br><br>2)Million's Test<br>1 ml of sample , add 1ml of million's reagent and heated for 3 minute then 1% sodium nitrate is added . Red colour formed indicate the presence of tyrosine | | | The test is Negative<br><br>The test is Negative |
| xi) | Detection of Alkaloids<br>1)Wagner's Test<br>Few ml of filtrate few drops of Wagner's reagent were added along with the sides of the test tube . formation of reddish brown precipitate indicate test .<br><br>2)Hager's Test<br>To few ml of filtrate<br>1 or 2 ml of Hager's<br>Regent is added .<br><br>3)Dragendorff's reagent<br>A few ml of filtrate 1 or 2 ml of Dragendorff's reagent is added a<br><br>4)Mayer's Test<br>To a few drop's of<br>Filtrate , two drops of Mayer's reagent is added along with side test tube if the test is positive it gives white or creamy precipitate. | Reddish brown precipitate<br><br>Aprominent reddish brown precipitate | Alkaloids is present<br><br>Alkaloids is present | The test is Positive. Shown in fig no-34<br><br>The test is Negative<br><br>The test is Positive. Shown in fig no-35<br><br>The test is Negative |

*G. Clarification*

The process is one of the Important steps to produce biofuel with certain specification. This method used certain adsorbents to absorb Impurities contain crude biodiesel as a product of transesterification reaction. An adsorbent material that has a high Silica content such as bentonite has The potential to be used, in clarification process. Firstly, filter the sample by using Whatman filter paper. Bentonite is used as clarify agent. The dry washing process of biofuel was carried out in conical flask. sample added 5 gm of bentonite, Placed in at room temp without disturbed. up to sample get transparent and pipette out. clarification was done. We get 147 mL of sample after pipette out.

*H. Potassium Dichromate Test*

Take potassium dichromate in test tube ,add few drops of alcohol (test sample). " acidify with dilute sulfuric and warm The test tube in hot water bath. orange solution turns into green. (Primary or secondary alcohol).

*I. Flammability Test*

*a) Objective*

The objective of this test is to determine whether the alcohol being tested is flammable or not. Flammability refers to the ability of a substance to ignite and sustain combustion in the presence of an ignition sources.

On a separate non-flammable Surface place a few drops of the alcohol sample using the ignition. Source, ignite the pooled alcohol on the surface observe if the alcohol on the surface ignites and burns. with a visible flame. Take note Some factors. (Colour size, and intensity).

*b) Conclusion*

Based on the observation from the Surface Ignition test analyze the result to determine the alcohol being tested is flammable (Booster Esterifies).

*c) Result*



Fig no-1. Starch extraction



fig no-2. Sterilization



fig no-3. Autoclave

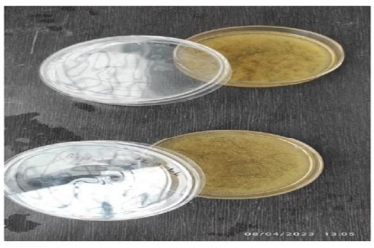


fig no-4. Solidification of PDA




fig no-5. Cultivation of saccharomyces cerevisiae




Fig No.6 Incubation



fig no.7 saccharomyces cerevisiae



fig no.8 Innoculation

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Fig no.9 Mold formation




Fig no.10 Gas formation

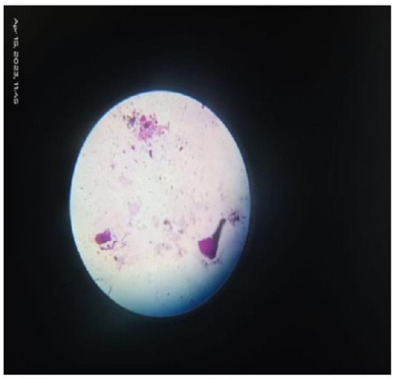


Fig no. 11 Acid fast bacteria




Fig no.12 Straw of wheet and Gram



Fig no 13. Materials



fig no.14 Addition of materials



Fig no.15 Addition of water



Fig no-16. Innoculation



fig no-17.fermantation tank



Fig no.18 Chemical for DNSA



Fig no. 19 DNSA



Fig No.20 DNSA Requirments

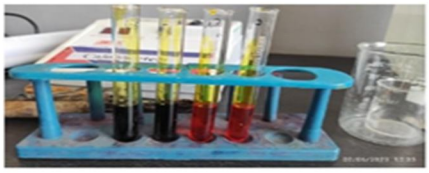


Fig No. 21 Preparation



Fig No. 22 Heating

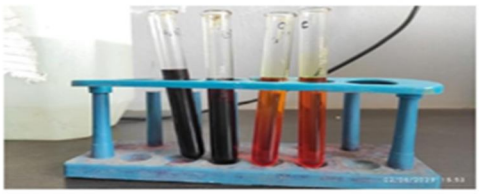


Fig no. 23 Result of DNSA



Fig No. 25 Molisch's Test



Fig no 26 Fehling's Test



Fig no. 27 Barfoed's test



Fig no. 28 Alkaline reagent test



Fig no 29 $NH_4OH$ Test



**Fig no.30 Mg turning test**



**Fig no.31 Ferric chloride test**



**Fig no.32 Lead Acetate**

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Fig no.33 Legal's test



Fig no.34 Wagner's test



Fig no.35 Dragendorff's test



Fig no.36 pH meter



Fig no.37 pH meter



Fig no.38 Clarification



Fig no.39 Filtration

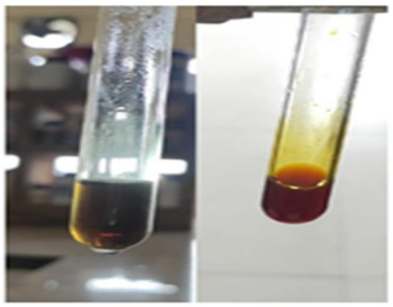


Fig no.40 Potassium Dichromate test

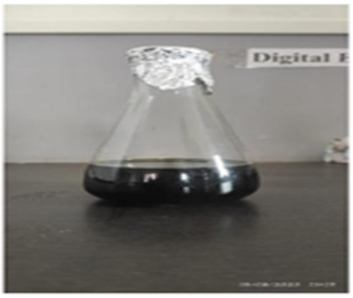


Fig no.41 Sample

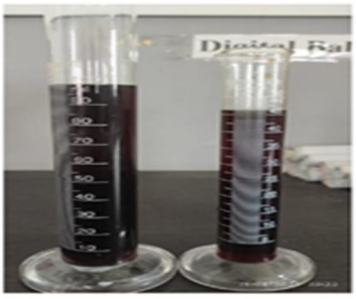


Fig no.42 Quantity of sample

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| Sr. no | Test | Result |
|---|---|---|
| 1 | Appearance | Liquid |
| 2 | Colour | Dark reddish brown |
| 3 | Odor | Pleasant |
| 4 | Viscosity | Low viscosity |
| 5 | Phase | Single phase |
| 6 | PH | 4.0 |

## III. CONCLUSION

In conclusion, the recent increase in demand for biofuels, coupled with the necessity to reduce production costs and improve environmental sustainability in biomass production, highlights the significance of optimizing the processes involved. This study specifically focused on the kinetic and optical properties of biofuel and biomass production, with a particular emphasis on esterification reactions as a means to efficiently convert waste biomass. The development of an efficient acidic catalyst for pretreatment was identified as a key step in this process.

Esterification of acetic acid, a common component in biomass, was investigated as a model reaction. This method offers the potential to remove organic acids from biomass by reacting them with alcohol present in biomass, producing water as a byproduct. Additionally, the quality of slow pyrolysis-produced biomass was compared with biomass collected by condensing pyrolysis vapor using atomized ethanol (EtOH).

The findings of this study underscore the importance of carefully considering the techno-economic feasibility of various processes when selecting feedstock for industrialization. Furthermore, the development of efficient catalysts and optimization of esterification reactions hold promise for enhancing the overall efficiency and sustainability of biofuel and biomass production. These efforts contribute positively to the reduction of fossil fuel dependency and the promotion of a more environmentally friendly energy future.

## IV. DISCUSSION

Certainly, let's discuss the various aspects of the subject you've presented:

[1] *Importance of Biofuels*: The increasing demand for biofuels is driven by the need to reduce our reliance on fossil fuels and their associated environmental impact. Biofuels, derived from renewable feedstocks, offer a more sustainable energy source.

[2] *Cost Reduction and Environmental Sustainability*: Reducing production costs is essential for the widespread adoption of biofuels. Simultaneously, improving the environmental sustainability of biomass production and biofuel processing is crucial to mitigate their environmental impact.

[3] *Kinetic vs. Optical Properties*: Your text mentions focusing on the kinetic properties or optical properties of biofuel and biomass production. It's important to clarify the specific properties you're referring to, as this can influence the efficiency and quality of production processes.

[4] *Process Optimization*: Process optimization plays a pivotal role in enhancing the efficiency of biofuel production. Optimization includes factors such as reaction kinetics, temperature, pressure, and catalysts.

[5] *Esterification and Transesterification*: These are key reactions in biofuel production, particularly in the synthesis of biodiesel. Esterification reactions, often acid-catalyzed, are used to pretreat biomass and remove organic acids, making the feedstock more suitable for biofuel production.

[6] *Pyrolysis and Condensation*: Slow pyrolysis and condensation of pyrolysis vapor using atomized ethanol are techniques used to produce biomass. The quality and composition of the resulting biomass can vary based on these processes.

[7] *Types of Biofuels*: Biofuels can be categorized into different generations. Second-generation bioethanol processes aim to complement traditional fuels. Third-generation biofuels, often referred to as advanced biofuels, are needed for applications like aviation and are less reliant on food crops.

[8] *Environmental Impact*: While biofuels are considered greener alternatives, their production processes can have a significant environmental impact. Finding ways to reduce this impact is crucial for their sustainability.

[9] *Lignocellulosic Biomass*: This type of biomass, derived from sources like maize, straw, and sugarcane, is abundant and suitable for biofuel production, particularly biodiesel.

[10] *Yeast Saccharomyces cerevisiae*: This yeast is widely used in the biofuel industry, especially for esterification processes. It's a workhorse for converting feedstock into biofuels.

[11] *CO2 Reduction*: Biofuels offer a closed-loop system for CO2 emissions, as they absorb CO2 during growth and release it when burned. This makes them a valuable tool in reducing greenhouse gas emissions.

[12] *Challenges and Future Research*: Discuss the challenges in scaling up biofuel production and the need for ongoing research to improve efficiency, reduce costs, and minimize environmental impacts.

In conclusion, the development and adoption of biofuels are critical steps toward a more sustainable energy future. Achieving these goals requires a multidisciplinary approach that combines chemistry, biology, engineering, and environmental science to optimize processes and reduce their environmental footprint.

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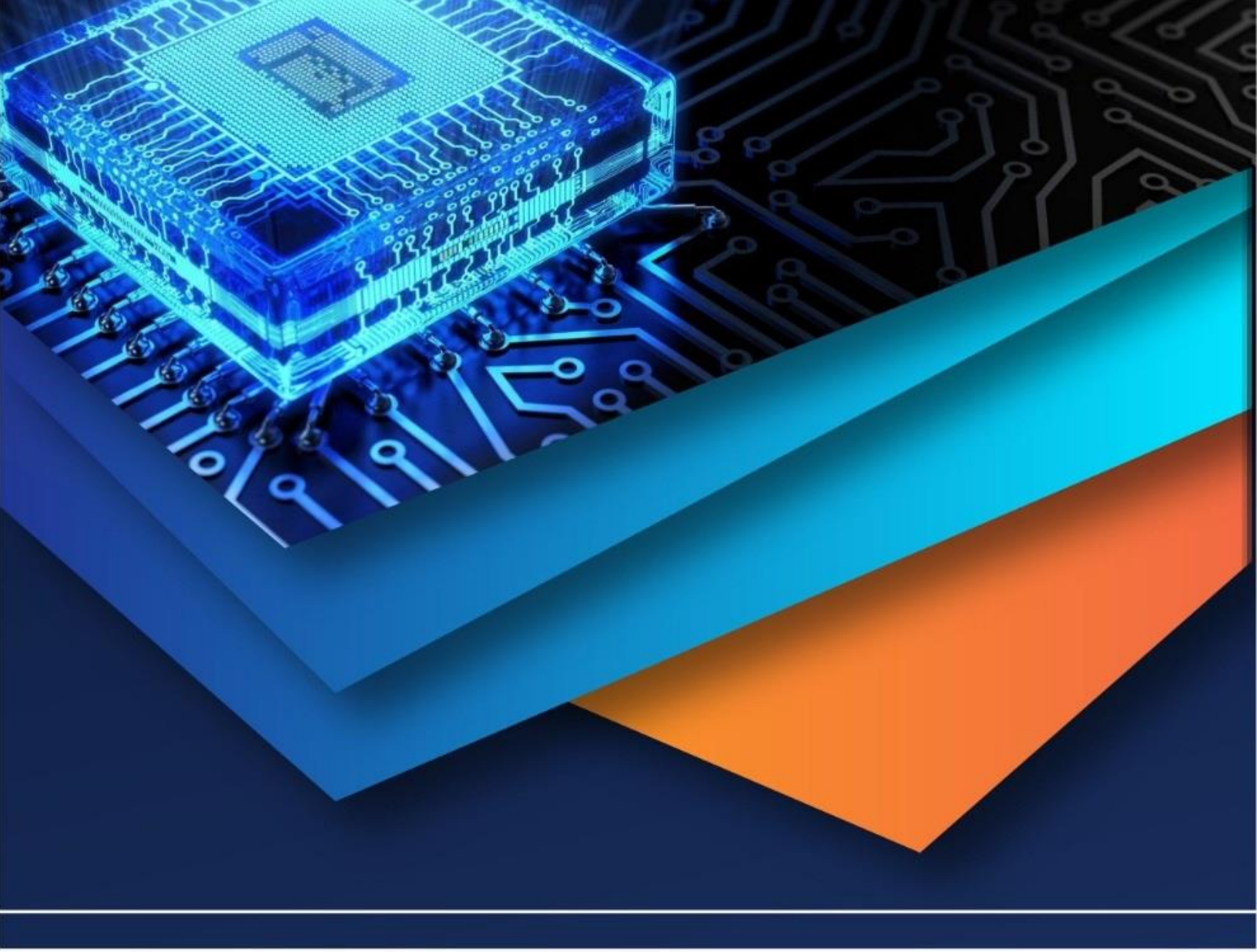
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