

# **SOME STUDIES ON ANTIMICROBIAL REGENERATED CELLULOSE MEMBRANES USING LIGNOCELLULOSIC WASTE**

*A Major Project Dissertation  
submitted in partial fulfilment of the requirement for degree of*

**Master of Technology  
In  
Polymer Technology**

*Submitted by*

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### **CERTIFICATE**

This is to certify that this is a bonafide record of project work based on topic **“Some Studies on Antimicrobial Regenerated Cellulose Membranes using Lignocellulosic waste”** is submitted by **Anuja Agrawal** (2K13/PTE/02). This project was carried under my supervision in year 2014-15 and being submitted towards partial fulfilment of the requirement for the award of Master of Technology in Polymer Technology in Delhi Technological University.

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## **ABSTRACT**

This study focuses on the fabrication and characterization of antibacterial regenerated cellulose membranes for water treatment using lignocellulosic waste. Coir pith and sugarcane bagasse are chosen for this purpose. The extracted cellulose is characterized. As the amount of extracted cellulose is greater for sugarcane bagasse, so we fabricated our membranes using sugarcane bagasse. 7% NaOH/12% urea is used as solvent and silver modified montmorillonite as an antibacterial agent. Methanol, calcium chloride and gelatin are used as coagulant. Membranes coagulated with methanol show maximum strength while with gelatine we get good porosity and comparable strength. Membranes obtained through calcium chloride coagulant have good strength and moderate porosity. Antibacterial membranes are fabricated using calcium chloride as a coagulant and as a pore forming agent. Membranes are characterized by DTA, TGA, SEM techniques. Pore size of membranes is in the range of 10-30  $\mu\text{m}$ . Water content and pure water flux measurement are carried out. Pure water flux of the membranes decreases with the increase in concentration of silver modified montmorillonite upto 1.5%. Antibacterial testing is carried out for both Gram positive (*S.aureus*) and Gram negative (*E. Coli*) bacteria. Membranes show good antibacterial properties for both the tested bacteria. So, these membranes may be a good alternative for pretreatment of RO, UF or NF processes or may be used as cell strainers.

# **TABLE OF CONTENTS**

	<b>PAGE NO.</b>
Certificate	i
Acknowledgement	ii
Abstract	iii
Table of Contents	iv
List of Figures	vii
List of Tables	ix
<b>Chapter 1 Introduction and objectives</b>	<b>1</b>
<b>1.1</b> Introduction	<b>1</b>
<b>1.2</b> objectives	<b>3</b>
<b>Chapter 2 Literature review</b>	<b>5</b>
<b>2.1.</b> Cellulose, a renewable biomaterial	<b>5</b>
<b>2.1.1</b> Molecular structure of cellulose	<b>5</b>
<b>2.1.2</b> Supramolecular structure of cellulose	<b>6</b>
<b>2.2</b> Solvent systems for cellulose	<b>8</b>
<b>2.3</b> Dissolution of cellulose in NaOH-Urea system	<b>9</b>
<b>2.4</b> Regeneration of cellulose	<b>10</b>
<b>2.5.</b> Methods used for waste water treatment	<b>10</b>
<b>2.6</b> Coconut coir pith	<b>11</b>
<b>2.6.1</b> Properties of coir pith	<b>11</b>
<b>2.6.2</b> Production of coir pith	<b>12</b>
<b>2.7</b> Sugarcane bagasse	<b>12</b>

	<b>2.8</b>	Membranes for waste water treatment	13
	<b>2.9</b>	Regenerated cellulose membranes for wastewater treatment	14
	<b>2.10</b>	Antibacterial membranes for wastewater treatment	14
<b>Chapter 3</b>	<b>Experimental</b>		17
	<b>3.1.</b>	Materials	17
	<b>3.2.</b>	Methods	17
	<b>3.2.1</b>	Extraction of cellulosic material from coir pith	17
	<b>3.2.2</b>	Extraction of cellulosic material from sugarcane bagasse	18
	<b>3.2.3.</b>	Dissolution of cellulose	18
	<b>3.2.4</b>	Fabrication of membranes	18
	<b>3.2.4.1</b>	Fabrication of membranes using different coagulants	18
	<b>3.2.4.2</b>	Fabrication of antimicrobial membranes	19
	<b>3.2.5</b>	Determination of $\alpha$ -cellulose content of autoclaved material	19
	<b>3.2.6</b>	Determination of molecular weight of regenerated cellulose	20
	<b>3.2.7</b>	Characterization of raw material	21
	<b>3.2.7.1</b>	Fourier Transform Infrared Spectroscopy (FTIR)	21
	<b>3.2.8</b>	Characterization of regenerated membranes prepared using different coagulants	21
	<b>3.2.9</b>	Characterization of regenerated cellulose membranes with silver modified montmorillonite	21
	<b>3.2.9.1</b>	Water content	21
	<b>3.2.9.2</b>	Thermo Gravimetric Analysis (TGA)	22

	3.2.9.3 Differential Thermal Analysis (DTA)	22
	3.2.9.4 Flux measurement	22
	3.2.9.5 Antimicrobial testing	23
<b>Chapter 4</b>	<b>Results and Discussion</b>	24
	4.1 extraction of cellulose from coir pith	24
	4.2 Extraction of cellulose from sugarcane bagasse	25
	4.3 $\alpha$ - cellulose content of autoclaved material	25
	4.4 Determination of viscosity average molecular weight	26
	4.5 Characterization of raw materials	27
	4.5.1 Fourier Transform Infrared Spectroscopy (FTIR)	27
	4.6 Characterization of regenerated membranes prepared using different coagulants	29
	4.7 Characterization of regenerated cellulose membranes with silver modified montmorillonite	31
	4.7.1 Thermo Gravimetric Analysis (TGA)	31
	4.7.2 Differential Thermal Analysis (DTA)	35
	4.7.3 Water content	38
	4.7.4 Flux measurement	39
	4.7.5 Antimicrobial testing	40
<b>Chapter 5</b>	<b>Conclusions</b>	41
<b>Chapter 6</b>	<b>Future Plans</b>	42
<b>Chapter 7</b>	<b>References</b>	43

## LIST OF FIGURES

<b>Figures</b>	<b>Page No.</b>
Figure 1—Molecular structure of cellulose	6
Figure 2—Interconversion of the cellulose polymorphs.	7
Figure 3—[a] coir husk [b] wet coir pith [c] dried coir pith [d] coir fibre [e] autoclaved coir pith.	24
Figure 4—[a] raw sugarcane bagasse and [b] autoclaved sugarcane bagasse	25
Figure 5—Plots of inherent /reduced viscosity vs. concentration	26
Figure 6—FTIR spectra of [a]raw coir pith, [b]raw sugarcane bagasse,	28
Figure 7—SEM micrograph of cellulose membranes by using [a], [c] 25% Cacl <sub>2</sub> and [b], [d] 20% Cacl <sub>2</sub> .	29
Figure 8—SEM micrograph of cellulose membranes by using [a], [b] 20% gelatin and [c], [d] 25% gelatine	30
Figure 9—SEM micrograph of cellulose membranes by using methanol as coagulant	31
Figure 10—TGA thermogram of regenerated cellulose membranes with [a] 0%clay, [b] 0.5%clay, [c] 1.0%clay, [d] 1.5%clay and [e] 2.0%clay (silver modified montmorillonite)	34
Figure 11—DTA plots for regenerated cellulose membranes with [a]0%clay, [b] 0.5%clay, [c] 1.0%clay, [d] 1.5%clay and [e] 2.0%clay (montmorillonite)	37



<p>Figure 12—Zone of inhibition for E.coli treated [a] 0% clay [b] 0.5%clay [c] 1.0%clay [d] 1.5%clay and [e] 2.0%clay (silver modified montmorillonite) loaded membranes</p>	<p>40</p>
<p>Figure 13—Zone of inhibition for S.aureus treated [a] 0%clay [b] 0.5%clay [c] 1.0%clay [d] 1.5%clay and [e] 2.0%clay (silver modified montmorillonite) loaded membranes</p>	<p>41</p>

## LIST OF TABLES

<b>Tables</b>	<b>Page No.</b>
Table 1—Data for viscosity measurement	26
Table 2— Data for water content measurement	38
Table 3—Data for rate of filtration through the membranes	39
Table 4—Zone of inhibition for membranes treated with E. coli	42
Table 5—Zone of inhibition for membranes treated with S. aureus	42

# CHAPTER 1: INTRODUCTION AND OBJECTIVES

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## 1.1 Introduction

Membrane separation is gradually emerging as a powerful separation tool for purification, fractionation, separation and concentration of products in wastewater treatment. It is the most effective technique for water filtration. Membrane processes are generally classified into different categories such as reverse osmosis, nanofiltration, ultrafiltration and microfiltration. Biofouling is a major problem in membrane desalination processes as it generates the requirement for higher operating pressures and backwashing or chemical cleanings. The main reasons for the membrane fouling may be contaminants adsorption, pore clogging, microbial activity, formation of the gel layer etc [1]. Different pretreatment processes such as activated carbon adsorption, coagulation, microfiltration, ozone oxidation etc. are used to prevent the membrane fouling in RO, UF or NF processes and to increase the life of these membranes. Pretreatment plays a pivotal role in reducing the deposition of pollutants on the membrane surface and as a result increasing the life span of membranes.

Polymer-matrix nanocomposite membranes are advanced membranes with nanomaterials dispersed in their polymer matrices. The nanocomposite membrane in which the nanofillers have been incorporated within the polymer matrix have improved the separation performance of the membrane and increased the permeability, selectivity and stability of the membrane which are the key factors for water purification applications [2, 3]. The incorporation of nanomaterials into polymers could not only tune structure and physicochemical properties (hydrophilicity, porosity, charge density, chemical, thermal and mechanical stability) of membranes, but also introduce unique functionalities such as antibacterial characteristics into

the membranes. Developing antimicrobial membranes increase membrane efficiency and application duration significantly. In addition, use of antimicrobial membrane helps to provide pathogen-free clean water.

Montmorillonite is the most widely used clay for different purposes and treatment of many skin problems. It is also effective in the adsorption of heavy metals. It is clear from many clinical studies that clay may be an effective remedy for many medical issues. Montmorillonite clay has antibacterial properties and also effective for the topical treatments. It has been used to treat contact dermatitis. Montmorillonite clay may also be used to treat some internal medical problems in humans as well as in animals. Silver (Ag) is the most widely explored antimicrobial agent in nanocomposite membrane due to its excellent biocidal properties and successful applications in many areas such as antimicrobial plastics, coatings and wound and burn dressing [4]. The properties of different clay polymer nanocomposites and their use as adsorbents in the removal of micropollutants (inorganic, organic and biological) from aqueous solutions have been studied [5]. The fabrication of Flat sheet nanocomposite membranes of PSf/clay with varying clay concentration has been analysed [6]. Nanocomposite membranes have been fabricated for water treatment from propionated lignin and cellulose triacetate [7].

The most widely used polymer for membrane fabrication is cellulose and its derivatives. Agricultural waste contains different type of lignocellulosic materials. Lignocellulosic wastes contain a fair amount of cellulose and lignin. So we can use these lignocellulosic materials for the fabrication of membranes used for water filtration. Due to the presence of lignocellulose their biodegradability is low and their proper disposal is a problem. Coir pith and sugarcane bagasse are some of them. If we are able to recover the cellulose present in these agricultural wastes, we can use it in many places such as preparation of cellulose

membranes for waste water treatment or bio-ethanol etc [8-11]. Agricultural solid wastes have been used as an adsorbent for dye removal from wastewater [4, 12-14].

Coir pith is generally dumped as an agro waste. When we extract one ton of fiber, approximately two tons of pith is produced. It is found that presently there is a stock of  $10 \times 10^6$  metric tons of coir pith in the coconut producing states of India and annual production of coir pith is about  $7.5 \times 10^5$  tons in India [9, 15]. Pollutant removal by adsorption using different form of coir pith has been studied [11,14].

A sugar factory produces approximately 3 tonnes of waste sugarcane bagasse, if 10 tonnes of sugarcane is crushed. Sugarcane bagasse contains a high moisture content of about 40 to 50%, which is a problem in its use as a fuel. Cellulose acetate membranes have been prepared using sugarcane bagasse [15-17]. Cellulose acetate membrane from biomass of ramie fibers [18] and from biomass: newspaper and mango seed [19] has been fabricated for water treatment purposes. But no study has been conducted on regeneration of cellulose and fabrication of antibacterial membranes using regenerated cellulose for water treatment.

The percentage of the cellulose is around 35% in coir pith and above 45% in sugarcane bagasse. The reuse of coir pith and sugarcane bagasse can also reduce the environmental pollution. Raw material used can be considered as a low cost, abundant and inexpensive in India. Besides that, the production of cellulose has a potential in a future because from the cellulose, many valuable product can be produce such as bio-ethanol.

## **1.2 Objectives**

The broad objective of the project is to develop the antimicrobial membranes from cellulose, obtained from agricultural waste, using silver modified montmorillonite for water treatment.

The specific objectives are -

- Extraction of cellulose from coir pith waste and its characterization.

- Extraction of cellulose from sugarcane bagasse and its characterization.
- Fabrication of regenerated membranes from extracted cellulose of sugarcane bagasse using calcium chloride, gelatine and methanol as a coagulant and their characterization.
- Fabrication of regenerated membranes from extracted cellulose of sugarcane bagasse using silver modified montmorillonite and calcium chloride.
- Characterization of membranes for physical and thermal properties.
- Characterization of membranes for water filtration and antimicrobial activity.

## CHAPTER 2: LITERATURE REVIEW

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### 2.1 Cellulose: a renewable biomaterial

Cellulose is used either in its native or in the derivate form in a wide range of industries, out of which pharmaceuticals and apparel are two main industries. Cellulose molecule is a renewable, biodegradable and environment-friendly biomaterial, so other industries has also started to show an increased interest in its use. The fuel and energy sectors have more recently begun to utilize cellulose. Besides it, other wood components, such as hemicellulose and lignin, have also become an interesting source of raw material for the production of bioethanol and biodiesel [18, 20].

Cellulose is an organic polymer that exists in the cell wall of all green plants. It gives mechanical strength to them. So it is the most abundant organic compound on Earth. The present volume of cellulose is believed to be around 700 billion tons. It is a polysaccharide based on the monomer anhydroglucopyranose unit (AGU) with the molecular formula  $C_6H_{10}O_5$  [20].

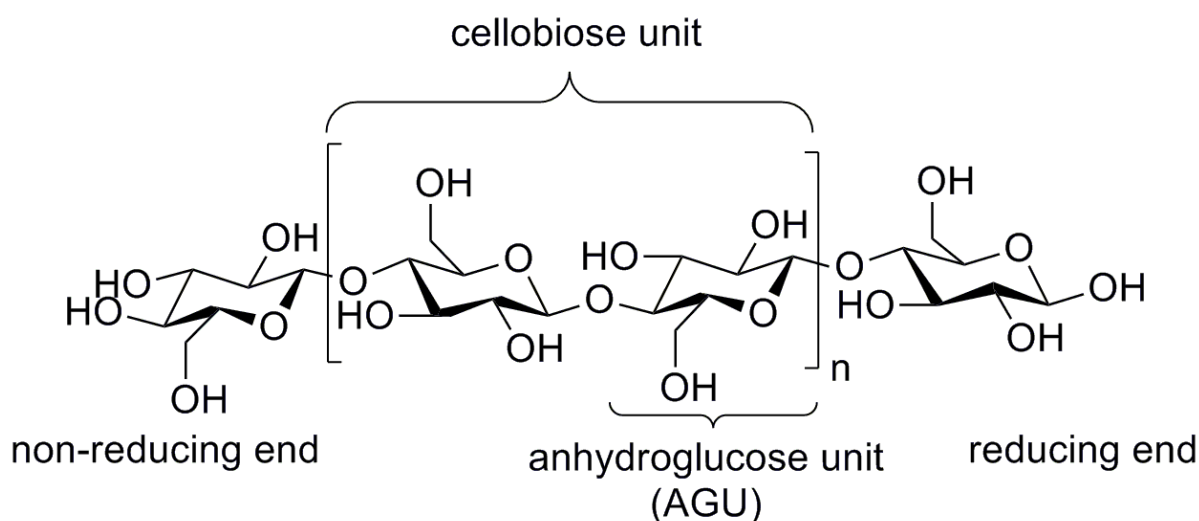
#### 2.1.1 Molecular structure of cellulose

The AGU monomers, which compose the cellulose molecule, are linked together by  $\beta$ -glycosidic linkages that are formed between the carbon atom C-1 and the C-4 of an adjacent unit of cellulose polymer. A dimer of AGUs (cellobiose) can therefore be considered as the basic unit of the cellulose molecule. It also shows that the cellulose molecule may be considered as a linear, unbranched polymer with repeating units of cellobiose.

The chain length of the cellulose molecule is expressed as the constituent of AGUs, which is commonly known as the degree of polymerization (DP). The DP of cellulose depends on the origin of cellulose molecule. Untreated cellulose, isolated from cotton, wood etc., has a DP  $\geq$  1000 while regenerated cellulose fibres and powder have a DP of 250-600 and  $\leq$  200, respectively. The DP is always an average value because the cellulose or other polymer substrates are a polydisperse mixture of polymers with different chain lengths. The correlation between the DP and the weight-average molecular mass ( $M_w$ ) of the cellulose molecule may be expressed by the following Equation -

$$DP = M_w/162$$

Each AGU contains a hydroxyl group on the carbon atoms C-2, C-3 and C-6. By using these hydroxyl groups we get the opportunity of modifying the cellulose into its derivatives, in different manners, with reactions that are commonly used in primary and secondary alcohols, such as esterification and etherification. Due to the presence of hydroxyl groups on each ends of the molecule, a chemical polarity may also be generated.



**Figure 1—Molecular structure of cellulose [20]**



### 2.1.2 Supramolecular structure of cellulose

The cellulose molecule has not only the ability of generating intramolecular hydrogen bonds (bonds within a molecule) but also intermolecular bonds (bonds between molecules) from carbon C-6 in one chain to carbon C-3 on the neighbouring molecule. The cellulose molecules have the tendency to aggregate into highly well-defined ordered regions due to their chemical structure and ability to generate intra and intermolecular hydrogen bonds, hydrophobic interactions and attractive vander Waals forces. These are normally known as “crystalline regions” and can be arranged in different polymorphs named as cellulose I ( $I\alpha$  and  $I\beta$ ), II, III ( $III_I$  and  $III_{II}$ ) and IV ( $IV_I$  and  $IV_{II}$ ) [18, 20, 21]. The native cellulose, cellulose I, has a parallel crystalline structure composed of the phases  $I\alpha$  and  $I\beta$  where the latter is thermodynamically more stable than the former. The composition of  $I\alpha$  and  $I\beta$  depends on the origin of the raw material: cellulose extracted from higher plants, such as wood, consists mainly of  $I\beta$  whereas that from more primitive organisms, such as bacteria, is enriched with  $I\alpha$ . [18, 20].

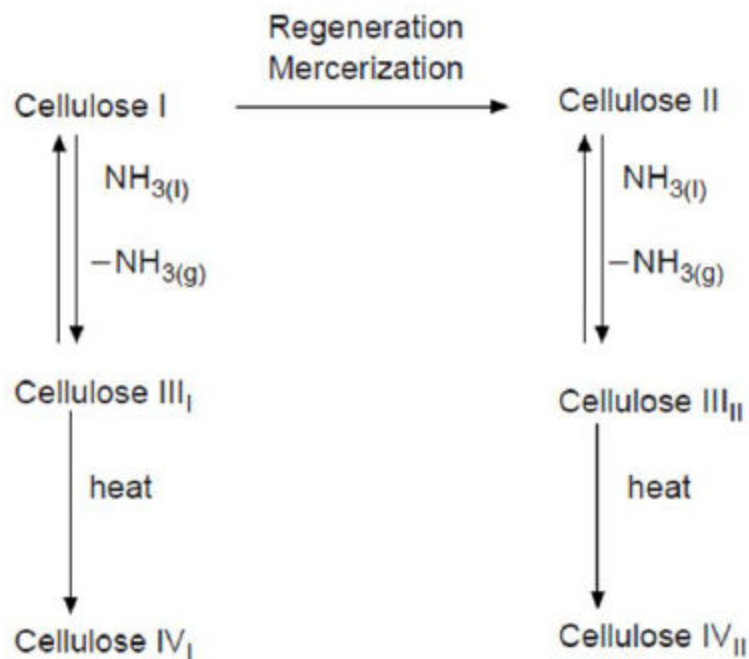


Figure 2—Interconversion of the cellulose polymorphs [20]

## 2.2 Solvent systems for cellulose

Cellulose is not a thermoplastic polymer and degrades before it melted, thus melting is not an option to fabricate a membrane. Dissolution is a process step that is therefore necessary in order to transform the solid cellulose into a liquid phase. However, due to its well defined structure, cellulose is barely accessible to any organic and inorganic solvents. The main problem is its low solubility in common solvents. Using organic solvents and ionic liquid may also dissolve cellulose, but the high cost and problem associated with the recovery of organic solvent hinder their further applications on a wide scale.

The solvent systems available for cellulose can be classified into two groups, derivatizing and non-derivatizing solvents, latter of which can be divided into two subgroups, namely aqueous and non-aqueous media [21].

Derivatizing solvents, as the name reveals, modify the cellulose prior to its dissolution. The viscose process utilizes carbon disulfide to form a soluble cellulose derivatized intermediate prior to the dissolution step. These solvents react with one, or several, of the three reactive hydroxyl groups in the cellulose, so that cellulose may be more soluble in common solvents. The non-derivatizing solvents, on the other hand, do not modify the cellulose prior to dissolution step but dissolve it instead, by disturbing the forces keeping it together without any chemical modification [20].

Sulphuric acid can be used to dissolve and then regenerate the cellulose fibres. Using optimal conditions of the acidic treatment (57–60 wt.% Sulphuric acid, Temperature = 45°C; acid to cellulose ratio 8-10, time = 1 h) in combination with the high-power disintegration permitted obtaining the nanocrystalline cellulose particles (NCP) having dimensions 150–200 × 10–20nm with the good yield (65–70%). At sulphuric acid concentration 65% and temperature 25°C cellulose can be regenerated by dilution with water [24].

Ionic liquids may be used as a green solvent for dissolution of cellulose [25].

NaOH-based aqueous systems provide an environment friendly, economically feasible and simple process for cellulose dissolution. Intermolecular hydrogen bonding of polysaccharides can be broken by using different additives with sodium hydroxide [26]. It is evident from the researches that NaOH and urea break intermolecular hydrogen bonding of different polysaccharides which enhances its solubility in water. By adding some organic or inorganic compounds (urea, thiourea etc) to NaOH solution, solubility of cellulose may be increased [27-33].

### **2.3 Dissolution of cellulose in NaOH-Urea solution**

The molecular weight of polymer is a key parameter in dissolution process; the higher the molecular weight, the weaker is the contribution of entropic force for dissolution. Under these conditions, the enthalpy term becomes crucial in calculating the changes in Gibbs free energy.

In the case of the cold alkali, one of the leading opinions is that NaOH forms hydrates with water capable to break the intermolecular and intramolecular hydrogen bonds. These hydrates have the capacity to bind with one or two hydroxyl groups of each AGU. The reason for the enhanced solubility of cellulose is the contribution of dissociated counterions to the entropy of mixing [30, 33].

The role of urea and thiourea is to prevent the reassociation of cellulose molecules, therefore providing stability to the solution. They are also supposed to prevent cellulose from regenerating its intermolecular hydrogen bonding again by forming a stable “hydrate coat” on their surface. Urea has no direct interaction with cellulose but it helps the alkali to penetrate into cellulose crystalline regions, so stabilizing the swollen cellulose molecules. Urea hinders the hydrophobic association of cellulose molecules.

Another important variable in NaOH-based solvents is the relatively low temperature needed to efficiently dissolve cellulose in opposition to the general picture of solubility, which implies that the entropic driving force is strengthened with increasing temperature. Typically, sub-zero temperatures are required for dissolution in sodium hydroxide. It is suggested that the process of cellulose dissolution in water or sodium hydroxide is exothermic and thus takes place at a lower temperature [34].

#### **2.4 Regeneration of cellulose**

The regeneration mechanism for alkali systems suggests that the complex formed with cellulose, NaOH (with or without urea or thiourea) hydrates is broken by adding a non-solvent such as water, calcium chloride etc. which leads to the self-association of cellulose molecules. The regenerated cellulose film is considered to be formed by the rearrangement of the hydrogen bonds. Moreover, in acidic non-solvents, it is assumed that the H<sup>+</sup> ions play a key role to trigger cellulose regeneration by neutralizing the alkaline content [34].

#### **2.5. Methods used for waste water treatment**

Adsorption is the most used method in wastewater treatment, which can mix the wastewater and the porous material powder or granules, such as activated carbon and clay, or let the wastewater through its filter bed composed of granular materials. Through this method, pollutants in the wastewater are adsorbed and removed on the surface of the porous material or filter [35].

The technologies for wastewater treatment may be divided into three main categories:

- (i) Conventional methods
- (ii) Established recovery processes
- (iii) Emerging removal methods

The expectations for developing an effective method for the treatment of these wastes are quite promising, but require continuous optimization and knowledge of new aspects [36].

It is well known that dye removal by using adsorbents is a cost effective method and efficient way. Many studies has been conducted recently on various biosorbent materials such as fungal or bacterial biomass and biopolymers that may be obtained in large quantities and that are harmless to nature. One of them is coir pith. It is produced after the removal of fibres from coconut husk. It is an agricultural waste product [37].

Agricultural wastes are renewable materials, available in large quantity and less expensive in comparison to other materials used as adsorbents. There are specific alternative agricultural by-products which are being used intensely as dye adsorbents such as peanut hull, coir pith obtained from coconut and rice husk.

## **2.6 Coconut coir pith**

Coir pith is a by-product of coir fibre production which is an important industry in most countries where coconust are grown in large amount. The extraction of the coconut fibre from husks gives us this by-product called coir pith. Coir pith is a 100% natural growing medium. This coir pith dried in the natural sun, are processed to produce different items namely coir pith block, coir pith briquettes, coir pith tablets etc. [9, 11].

### **2.6.1 Properties of coir pith**

Coir pith is the spongy, peat like residue from the processing of coconut husks (mesocarp) for coir fibre. Also known as cocopeat, it consists of short fibres (<2cm) around 2% - 13% of the total and cork like particles ranging in size from granules to fine dust. Coir pith strongly absorbs liquids and gases. Coir pith has been used as a casing layer in mushroom production

and as a biological filter for odour control. It is estimated that the production of coir pith in India is about 7.5 million tonnes per year [14].

### **Chemical composition of coir pith**

Moisture	11.9%
Ash	8.7%
Fat and resin	1.8%
Lignin	25.2%
Cellulose	35%
Pentosan	7.45%

### **2.6.2 Production of coir pith**

After the husk has been separated from the inner hard shelled nut, it is soaked in water to soften the pith and loosen the fibres. This is usually done by floating the husks in a lagoon for several months. The waste coir fibre (coir dust) was until recently, the only part of the coconut tree that had no real value. Even the roots have a use as they release a potent narcotic when chewed. Coir pith is a poor fuel because it tends to smoulder and give off more smoke than heat [9].

### **2.7 Sugarcane bagasse**

Sugarcane bagasse is the fibrous residue that remains after sugarcanes are crushed to extract their juice. It may be used as a biofuel, for the production of electricity, in the manufacture of paper and pulp and building materials. A sugar factory produces approximately 3 tonnes of waste sugarcane bagasse, if 10 tonnes of sugarcane is crushed. Sugarcane bagasse contains a high moisture content of about 40 to 50%, which is a problem in its use as a fuel [38].

Approximate composition of sugarcane bagasse is -

Cellulose 40–50%

Hemicellulose 20–30%

Lignin 18–24%

Ash 1–6%

Waxes <2%

G. R. Filho et al. analyzed the properties of methyl cellulose produced from sugarcane bagasse[3].

J.X. Sun et al. isolated the cellulose from sugarcane bagasse and studied its properties [39]. They also studied the properties of cellulose obtained from barley straw [40].

## **2.8 Membranes for waste water treatment**

Electro-spinning process is a very effective method to make porous hydrophobic membranes. Furthermore, electrospun nanofibrous membranes possess several attractive qualities, such as high porosity, pore sizes ranging from tens of nanometer to several micrometers, interconnected open pore structure and high permeability of gases. As well, the higher porosity of the nanofiber membranes enables them to have a large surface area per unit volume of membrane accessible to the liquid filtration applications, e.g. membrane distillation. So far, nature and synthetic polymers such as cellulose, polysulphone, polyacrylonitrile and chitosan/nylon-6 were electrospun into nanofibers to prepare the desired affinity membranes [40,41].

Recently, affinity membrane chromatography, as a promising alternative technology to the traditional packed-bed column chromatography, has attracted much attention. This is attributed to the advantages of the affinity membranes, such as low pressure drops, high flow

rates and no intraparticle diffusion, overcoming the basic limitations of traditional column methods. Moreover, the surface immobilized affinity ligands allowed the membranes to become more efficient and specific in capturing target biomolecules. Solvent casting is also a widely used method for membrane fabrication [42].

## **2.9 Regenerated cellulose membranes for wastewater treatment**

Cellulose membranes have been prepared for different purposes using different lignocellulosic materials [4, 18, 25].

Cellulose fibres have also been prepared for different purposes using different solvents [24,31]. Y. Mao et al. prepared regenerated cellulose membranes using NaOH-urea system as solvent [43]. R. Li et al. prepared regenerated cellulose using cellulose from different sources [30]. X. Xiong et al. prepared hybrid regenerated cellulose membranes using chitosan [6].

Sodium hydroxide pretreatment can enhance lignocelluloses digestibility [34]. Fan et al. fabricated cellulose acetate membrane from biomass of ramie fibers for water treatment purposes [44]. Carla da Silva Meireles et al. prepared asymmetric membranes using cellulose acetate (CA) obtained from biomass (newspaper and mango seed) [19].

## **2.10 Antibacterial membranes for wastewater treatment**

Biofouling of membranes is a major problem in membrane separation processes because due to biofouling membranes require higher operating pressures and more frequent chemical cleaning. In addition membrane life is decreased as well as product water quality is compromised. Low-biofouling composite membranes may be fabricated using interfacial polymerization, surface grafting, coating of a protective layer and surface modification with nanoparticles [45]. Y. Ma et al. analysed the fabrication of flat sheet nanocomposite membranes of PSf/clay with different clay concentration [43].



L. M. Nevárez et al. used propionated lignin and cellulose triacetate to fabricate nanocomposite membranes for water treatment [7].

It has been demonstrated that incorporation of nanomaterials into the polymer matrix could increase the water permeability and water flux without reducing the rejection of salts. Based on the literature study, polymer based nanocomposites have been proposed as membrane materials for water desalination.

Polymer-matrix nanocomposite membranes are advanced membranes with nanomaterials dispersed in their polymer matrices. They could be used for gas–gas, liquid–liquid, and liquid–solid separations. The incorporation of nanomaterials into polymers could not only tune structure and physicochemical properties (hydrophilicity, porosity, charge density, chemical, thermal and mechanical stability) of membranes, but also introduce unique functionalities such as antibacterial and photocatalytic characteristics into the membranes. Incorporation of nanomaterials in the membranes can change their water permeability and solute rejection [42,46].

Membrane biofouling decreases membrane permeability, reduces permeate quality and increases energy costs of the separation process. Developing antimicrobial membranes will likely increase membrane efficiency and application duration significantly. In addition, use of antimicrobial membrane helps to provide pathogen-free clean water. Silver (Ag) is the most widely explored antimicrobial agent in nanocomposite membrane due to its excellent biocidal properties and successful applications in many areas such as antimicrobial plastics, coatings and wound and burn dressing [4].

E.I. Unuabonah et al. reviewed the properties of different clay polymer nanocomposites and their use as adsorbents in the removal of micropollutants (inorganic, organic and biological) from aqueous solutions [5].

Anti-bacterial nanocomposites (NC1–NC4) based on cellulose acetate were prepared by dispersing ZnO nanofillers in the cellulose acetate matrix [46] but till date no study is conducted on fabrication of antibacterial membranes using regenerated cellulose. So in this study we will prepare antibacterial membranes using regenerated cellulose obtained from lignocellulosic waste.

## CHAPTER 3: EXPERIMENTAL

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### 3.1. Materials

Coconut husk and sugarcane bagasse is purchased from nearby market of Delhi, India. All of the chemical reagents (sodium hydroxide, urea, calcium chloride, gelatine, methanol, sulphuric acid etc) used in this work are of analytical grade and are purchased from commercial sources in India. Silver modified montmorillonite is taken from our lab which was prepared and characterized by Mr. Priyadarshan Sahoo for his M.Tech. project on “Preparation and Characterization of Antimicrobial Absorbent Layer for Baby Diaper”. Distilled water is used for all the experimental work.

### 3.2. Methods

#### 3.2.1. Extraction of cellulosic material from coir pith

Coir pith is extracted from the coir husk of coconut. The coir waste is kept soaked in water for 7 days then it is rinsed to remove the coir pith from the fibre. The recovered coir pith is washed with water till colourless drained water is obtained. Coir pith is dried under sun for 2 days and oven dried at 80°C upto constant weight. It is kept in dessicator till use. Coir pith is autoclaved 2 times in 8% NaOH solution at 120°C for 20 minutes. Excess water is drained and recovered solid material is thoroughly washed with water and oven dried at 80°C.

### **3.2.2 Extraction of cellulosic material from sugarcane bagasse**

Sugarcane bagasse is washed with water thoroughly to remove sugar content. Sugarcane bagasse is dried under sun for 2 days and oven dried at 80°C upto constant weight. Bagasse is autoclaved 2 times in 8% NaOH solution at 120°C for 20 minutes. Excess water is drained and recovered solid material is thoroughly washed with water and oven dried at 80°C.

### **3.2.3. Dissolution of cellulose**

5 grams of cellulose is dispersed in 100 grams of 7% NaOH /12% urea solution and stirred for 5 minutes to obtain slurry. This dispersion is kept in freezer (-5°C) for about 24 hours. The obtained solid mass is thawed at room temperature and stirred vigorously. The resulting solution is subjected to centrifugation at 2000 rpm for 30 minutes to remove the excess gel and to carry out degasification. After it we got a clear cellulose solution of approximately 4% concentration.

### **3.2.4 Fabrication of membranes**

#### **3.2.4.1 Fabrication of membranes using different coagulants**

The prepared solution of cellulose is casted on a glass plate to give the thickness of solution 0.5 mm and cellulose is regenerated using different coagulants calcium chloride, gelatine and methanol.

For calcium chloride, different concentrations (10%,15%,20% and 25%) of calcium chloride is mixed with thawed cellulose solution and immediately casted on the glass plate. After drying, membrane is removed carefully from the glass plate and treated with 3% HCl for 2 minutes.

For gelatine, different concentrations (10%, 15%, 20% and 25%) of gelatine are mixed with thawed cellulose solution and immediately casted on the glass plate. After drying, membrane

is removed from the glass plate carefully and kept in distilled water at 40°C for 24 hours to remove gelatine.

For methanol, thawed cellulose solution is casted on the glass plate and immediately immersed in a bath containing methanol for 5 minutes. Membrane is washed with distilled water, dried and removed.

#### **3.2.4.2 Fabrication of antimicrobial membranes**

Antimicrobial membranes are fabricated using silver modified montmorillonite as an antimicrobial agent in different concentrations (0.5%, 1.0%, 1.5% and 2.0%) and calcium chloride as coagulant using the same method described earlier.

#### **3.2.5 Determination of $\alpha$ -cellulose content of autoclaved material**

$\alpha$ -cellulose content of the cellulosic material obtained after autoclaving is determined to find out the amount of accessible cellulose. Chemical composition of sugarcane bagasse is estimated according to the following procedures:

1.4 g of sample is immersed in 75 mL 17.5% NaOH and stirred at 25°C for 30 minutes. Add 25.0 mL NaOH (17.5%) to the beaker by rinsing the stirrer, so that the volume of reagent added to the sample becomes exactly 100 mL and wait for 30 minutes. The suspension is filtered after 60 minutes. Filtrate is used to determine the chemical composition of the sample. To determine the chemical composition, 25 mL of the prepared filtrate is taken and 10 mL of 0.5 N potassium dichromate and 50 mL of conc. sulphuric acid is added to it with stirring. In the last add 50 mL of water. Titrate it with 0.1N ferrous ammonium sulphate(FAS) solution by using Ferroin as an indicator. 12.5 ml NaOH(17.5%) and 12.5 ml of water is mixed and this solution is used as blank reading.

$\alpha$ -cellulose content is determined using the following equation[42]:

$$\alpha - cellulose\% = 100 - \frac{6.85(V_b - V_t) \times N_f \times 20}{S \times W_s} \quad (1)$$

where:

$V_t$  = titration of the pulp filtrate, mL

$V_b$  = blank titration, mL

$N_f$  = exact normality of the ferrous ammonium sulphate solution

$S$  = volume of the sample filtrate used in the oxidation, mL

$W_s$  = oven-dry weight of sample, g

24.52 g of  $K_2Cr_2O_7$  is dissolved in 1000 mL of water to prepare 0.5 N Potassium Dichromate solution and 40.5 g of  $Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$  is dissolved in 1000 mL of water and add 10 mL of concentrated  $H_2SO_4$  to prepare 0.1N Ferrous Ammonium Sulphate solution (FAS).

### 3.2.6 Determination of molecular weight of regenerated cellulose

Viscosity-average molecular weights ( $M_v$ ) of the original cellulose material is determined in 6% NaOH/4%urea at 25°C by using a capillary viscometer and is calculated from the following equation :

$$[\eta]_{int} = K_p M_v^\alpha \quad (2)$$

Where  $[\eta]_{int}$  is the intrinsic viscosity,  $M_v$  is Molecular weight,  $K_p$  and  $\alpha$  are constants for different polymer- solvent systems [47].

If we know the  $K_p$  and  $\alpha$  values for a given polymer solution the intrinsic viscosity and molecular weight can be calculate using the above equation.

Values of  $K_p$  and  $\alpha$  for cellulose solution in 6% NaOH/4% urea are  $2.45 \times 10^{-2}$  and 0.815 respectively.

Intrinsic viscosity of polymer samples is measured by preparing different solutions of known concentrations and the flow times of solvent and the solutions of different concentrations are measured using capillary viscometer. Reduced viscosity and inherent viscosity is calculated and double extrapolation plots of reduced viscosity vs. concentration and inherent viscosity

vs. concentration is plotted. The intrinsic viscosity is calculated by the common ordinate intercept of these two graphs.

### **3.2.7 Characterization of raw material**

#### **3.2.7.1 Fourier Transform Infrared Spectroscopy (FTIR) analysis**

Fourier Transform Infrared Spectroscopy (FTIR) is an analytical tool which is used to get information about the presence of some specific chemical groups in the sample. The experiments are carried out in an FTIR using transmittance Mode on Thermo Scientific Nicolet 380 Spectrophotometer (Nicolet) USA for raw coir pith. Thirty-two scans are measured for each spectrum with a step size of  $4\text{ cm}^{-1}$  and the range is  $4000 - 400\text{ cm}^{-1}$ . The powdered material (Raw coir pith) is prepared as pellets, mixing them with KBr for the analysis.

FTIR spectra of sugarcane bagasse is measured by Thermo Fisher FTIR (NICOLET 8700) analyser and the range is  $4000 - 600\text{ cm}^{-1}$ .

### **3.2.8 Characterization of regenerated membranes prepared using different coagulants**

The surface morphology of membranes is examined by using Hitachi S-3700N scanning electron microscope (Germany). Prior to examination, samples are kept in liquid nitrogen for 10 minutes and then freeze dried, after it samples are gold-sputter coated to render them electrically conductive and then scanned at an accelerating voltage of 15 kV.

### **3.2.9 Characterization of regenerated cellulose membranes with silver modified montmorillonite**

#### **3.2.9.1 Water content**

The ability of membranes to absorb water is determined by weighing membranes after soaking in water for 24 h and then dried them upto constant weight in the oven and the

weights of the dry membranes are recorded. The water content, absorbed by the membranes, is determined by the following equation:

$$\text{Water content} = \frac{W_a - W_b}{W_b} \times 100 \quad (3)$$

Where,  $W_a$  - weights of membranes in wet conditions

$W_b$  - weights of membranes in dry conditions

The readings for water content are measured three times for each sample and the results are given as average of these readings.

### **3.2.9.2 Thermo Gravimetric Analysis (TGA)**

TGA experiments are carried out by using Perkin Elmer (Pyris-6) TG/DTA equipment using alumina pans under nitrogen atmosphere. The samples are first heated from room temperature to 120°C and kept for 10 minutes. After then samples are heated upto 500°C and the scanning rate is kept 10°C/min.

### **3.2.9.3 Differential Thermal Analysis (DTA) measurement**

Differential Thermal Analysis (DTA) measurement experiments are conducted by using Perkin Elmer (Pyris-6) TG/DTA equipment. The tests performed (5 mg of sample) under nitrogen flow in the temperature range of 30°C- 650°C and the heating rate is kept 10°C/min.

### **3.2.9.4 Flux measurement analysis**

Permeation flux measurement analysis is carried out by using gravity filtration process.

$$\text{Flux} = \frac{Q}{A.t} \quad (4)$$

Where,

Q is the quantity of permeate in Litre, A is the effective membrane area (25cm<sup>2</sup>) and t is the time in minutes.



### 3.2.9.5 Antimicrobial testing

The Antibacterial activity of regenerated cellulose films is evaluated using disc diffusion method and the pathogen considered are Gram positive bacteria species (*Staphylococcus aureus*) and Gram negative bacteria species (*Escherichia coli*). The bacteria were grown in Luria Agar medium. In this test, 15 mm diameter discs of the films, prepared by incorporating different concentration of silver modified montmorillonite and without silver modified montmorillonite, are gently pressed on to the surface of the plate and kept in incubator at 37°C for 24 h. The diameter of the zone of inhibition demonstrates the antibacterial activity of membranes and is compared with the zone of inhibition of the membrane without silver modified montmorillonite. Zone of inhibition is calculated by using the following equation:

$$C = \frac{W-T}{2} \quad (5)$$

Where C is the width of clear zone of inhibition (mm), W is the width of test specimen and clear zone (mm) and T is the width of test specimen (mm).

## CHAPTER 4: RESULTS AND DISCUSSION

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### 4.1 Extraction of cellulose from coir pith

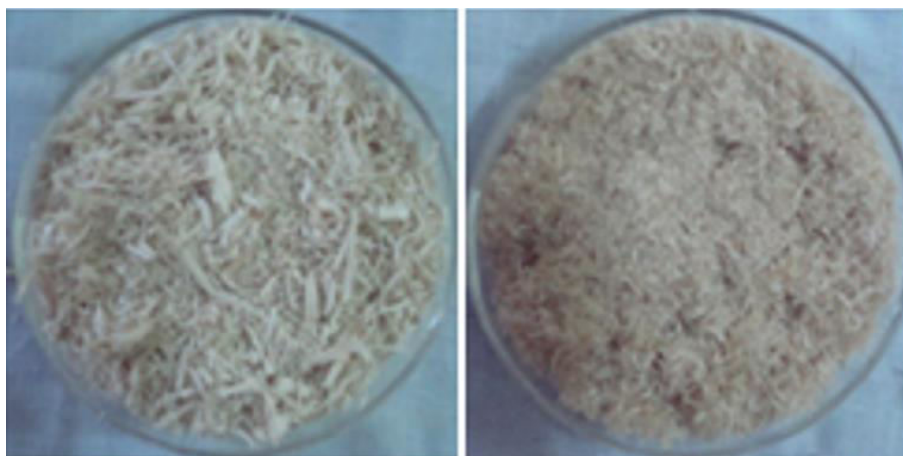
Coir pith is extracted from coir husk after soaking for 7 days in water and then rinsing them. The obtained coir pith is dried and autoclaved 2 times with 2N NaOH solution at 120°C for 20 minutes. Figure 3 shows the images of coir husk and coir pith before and after autoclaving. The yield of cellulosic material from coir pith after autoclaving is around 30%.



**Figure 3—[a] coir husk [b] wet coir pith [c] dried coir pith [d] coir fibre [e] autoclaved coir pith**

## 4.2 Extraction of cellulose from sugarcane bagasse

Cellulosic material from sugarcane bagasse is extracted by using the same method used for the extraction of coir pith.



**Figure 4—[a] raw sugarcane bagasse and [b] autoclaved sugarcane bagasse**

The images of sugarcane bagasse before and after autoclaving are shown in figure 4. The yield of cellulose from sugarcane bagasse is more than 45%. So we choose sugarcane bagasse for our further experiments.

## 4.3 $\alpha$ - cellulose content of autoclaved material

Volume of the FAS used for titration of bagasse filtrate ( $V_t$ ) = 22 mL

Blank titration ( $V_b$ ) = 52 mL

Normality of the FAS solution ( $N_f$ ) = 0.1N

Volume of the filtrate (bagasse) used for titration ( $S$ ) = 25 mL

Oven-dry weight of sample (bagasse) ( $W_s$ ) = 1.4 g

$$\alpha - cellulose\% = 100 - \frac{6.85(V_b - V_t) \times N_f \times 20}{S \times W_s}$$

So,  $\alpha - cellulose = 88.25\%$

#### 4.4 Determination of viscosity average molecular weight

6% NaOH/4% Urea is used as solvent and different concentration (0.025%, 0.05%, 0.075%, 0.1%, 0.25% and 0.5%) solutions are prepared.

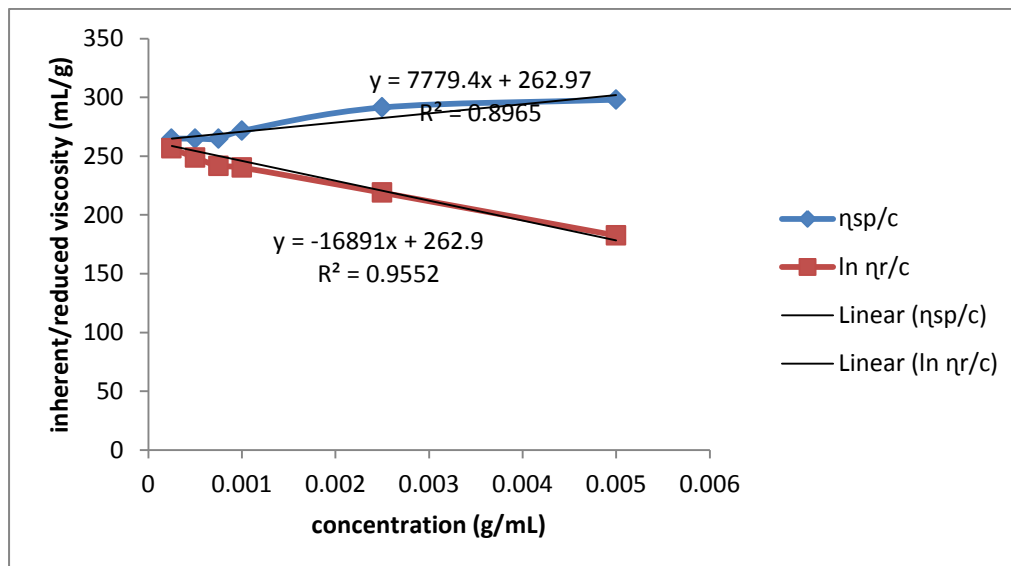


Figure 5—Plots of inherent /reduced viscosity vs. concentration

Table 1—Data for viscosity measurement

S. no.	Conc. (g/mL)	Flow time of pure solvent ( $t_0$ ) sec	Flow time of solution (t) sec	Relative viscosity ( $\eta_r$ )	Specific viscosity ( $\eta_{sp}$ )	Reduced viscosity ( $\eta_{sp}/C$ ) mL/g	Inherent viscosity ( $\ln \eta_r/C$ ) mL/g
1.	0.00025	1.51	1.61	1.066225	0.066225	264.9007	256.4981
2.	0.0005		1.71	1.13245	0.13245	264.9007	248.7674
3.	0.00075		1.81	1.198675	0.198675	264.9007	241.6229
4.	0.001		1.92	1.271523	0.271523	271.5232	240.2155
5.	0.0025		2.61	1.728477	0.728477	291.3907	218.8962
6.	0.005		3.76	2.490066	1.490066	298.0132	182.4619

Reduced viscosity and inherent viscosity is calculated and double extrapolation plots of reduced viscosity vs. concentration and inherent viscosity vs. concentration is plotted.

Figure 5 shows the plots of inherent viscosity vs. concentration and reduced viscosity vs. concentration. By plotting graphs inherent /reduced viscosity vs. concentration, we get the value of common ordinate intercept. It gives the value of intrinsic viscosity, which is 262.9 mL/g. By putting this value in Mark- Houwink equation, we can determine the value of viscosity average molecular weight of obtained cellulose.

$$[\eta] = K_p M_v^\alpha$$

$$\text{Or, } 262.9 = 2.45 \times 10^{-2} (M)^{0.815}$$

$$\text{Or, } M_v = 7.68 \times 10^4 \text{ g/mol}$$

## **4.5 Characterization of raw materials**

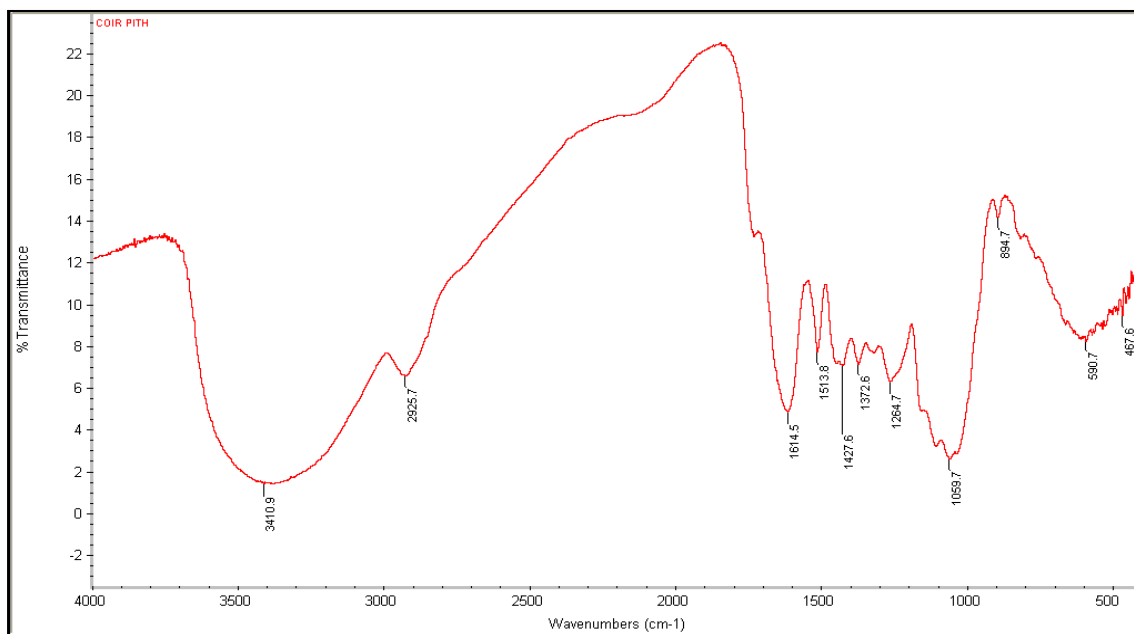
### **4.5.1 Fourier Transform Infrared Spectroscopy (FTIR)**

FTIR is an analytical tool used to identify the presence of some certain functional groups in a sample molecule. We can confirm the identity of a compound in the sample by analyzing the collection of absorption bands in FTIR analysis. Figure 6 shows the FTIR data of raw coir pith, raw sugarcane bagasse.

The peak at  $1614.5 \text{ cm}^{-1}$  in raw coir pith is due to the presence of acetyl ester or uronic ester linkage of hemicelluloses. It may be attributed to the ester linkage of carboxylic group present in p-coumeric acids or ferulic acid of lignin or hemicelluloses. Two peaks at  $1513.8 \text{ cm}^{-1}$  and  $1427.6 \text{ cm}^{-1}$  are due to the C=C stretch aromatic peak from aromatic ring of lignin. Hemicelluloses give the peak at  $1264.7 \text{ cm}^{-1}$  in raw coir pith. The –CH symmetric stretching of aromatic lignin gives the peak at  $1372.6 \text{ cm}^{-1}$  in raw coir pith. Raw coir pith and sugarcane bagasse both show a peak at  $3410.9 \text{ cm}^{-1}$  corresponding to cellulose–OH. The peak at  $1059.7$

cm<sup>-1</sup> in raw coir pith and sugarcane bagasse is due to β-glycosidic linkage at 1–4 position of glucose ring in cellulose.

[a]



[b]

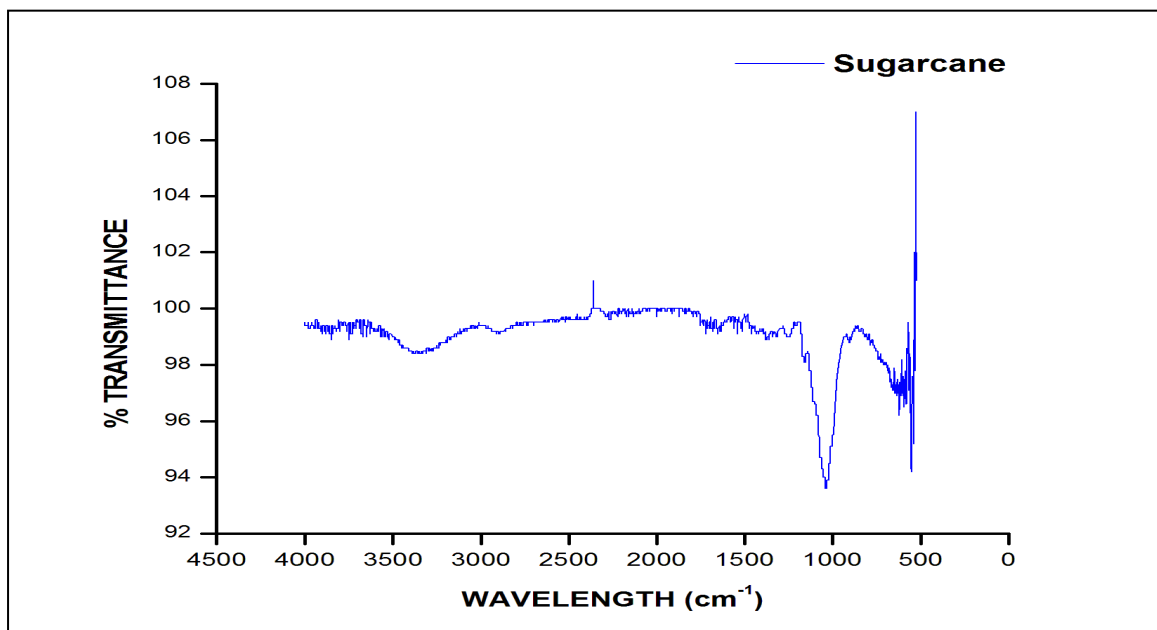
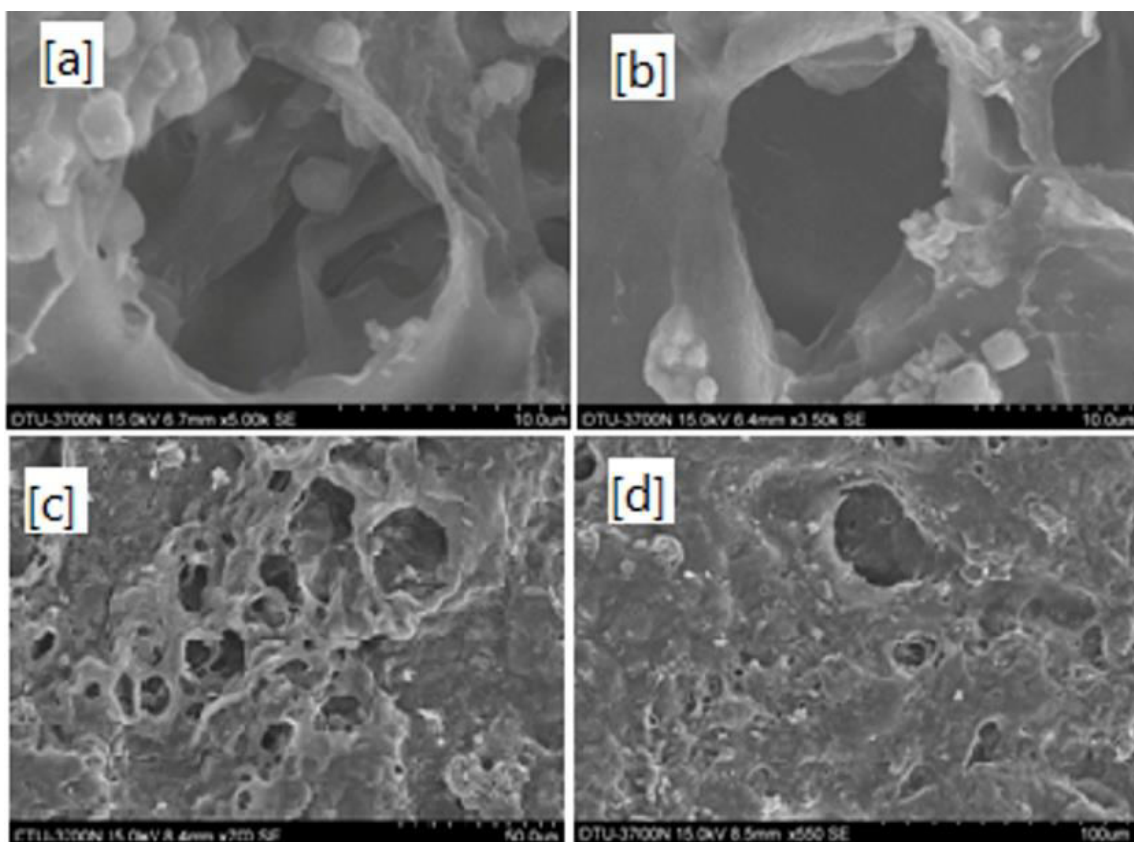


Figure 6—FTIR spectra of [a]raw coir pith, [b]raw sugarcane bagasse

#### 4.6 Characterization of regenerated membranes prepared using different coagulants

Three different coagulants namely calcium chloride, gelatine and methanol have been used to prepare the regenerated cellulose membranes. The surface morphology and pore size of the prepared membranes were analyzed using SEM.

Figure 7-9 show the SEM micrograph of different membranes prepared by using different coagulants.

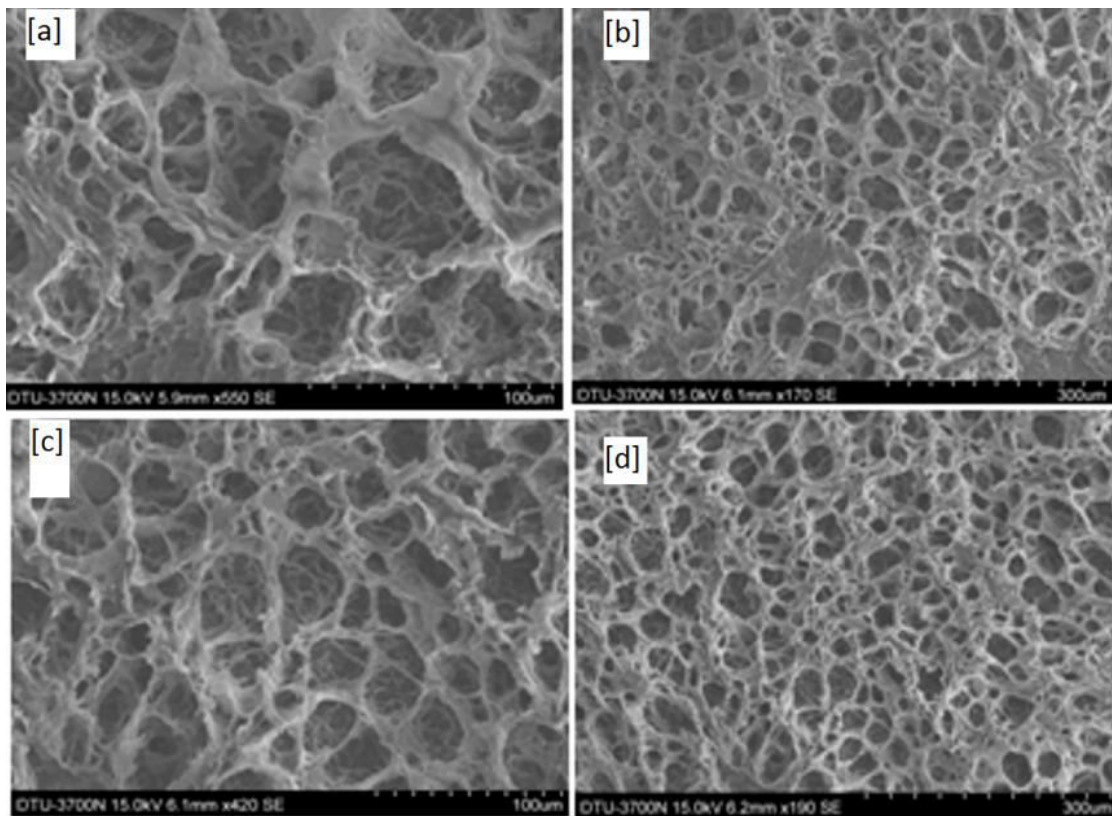


**Figure 7—SEM micrograph of cellulose membranes by using [a], [c] 25% CaCl<sub>2</sub> and [b], [d] 20% CaCl<sub>2</sub>**

Figure 7 shows SEM images of membranes after coagulating with 20% and 25% calcium chloride. We were not able to fabricate proper membranes after coagulating with 5% and 10% calcium chloride. The reason behind it may be that, by using low concentration of

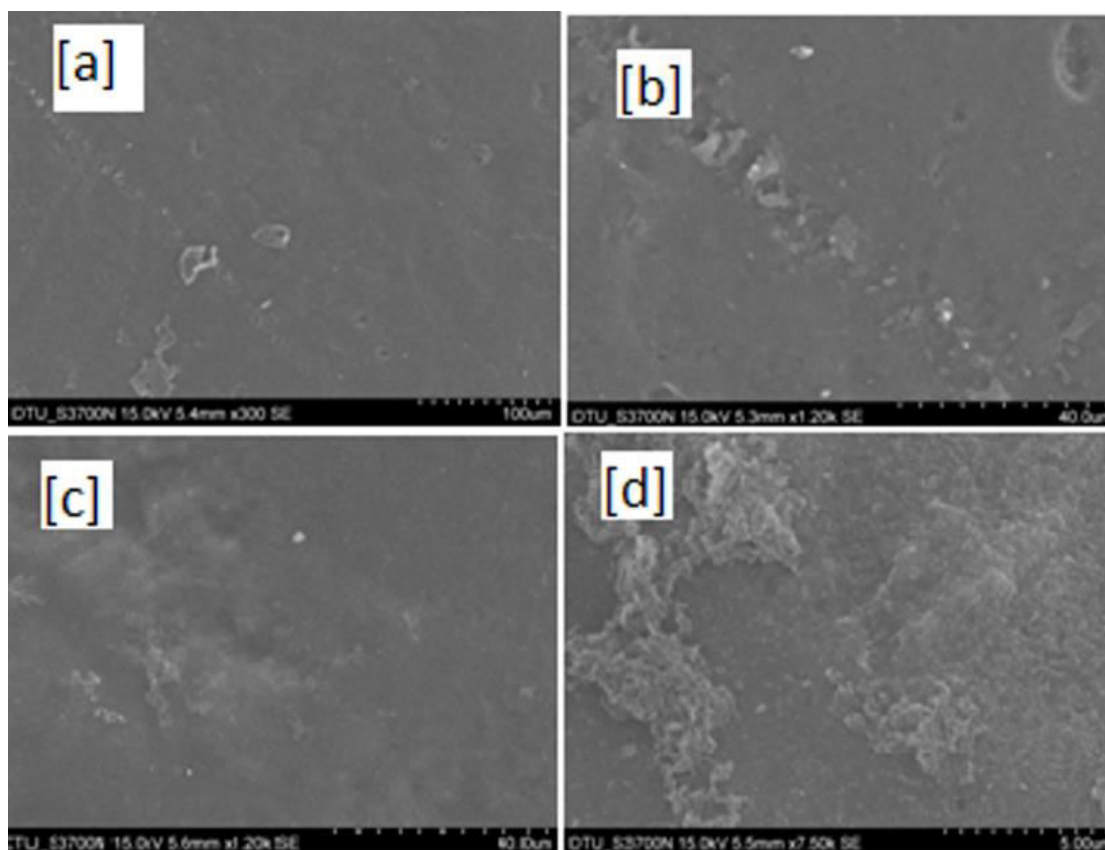
calcium chloride we are not able to regenerate the whole amount of cellulose so the concentration of cellulose is less, which gives membrane with cracks due to low concentration of cellulose. Pore size in membranes fabricated using 25% calcium chloride is around 10-20  $\mu\text{m}$ .

Figure 8 shows SEM images of cellulose membranes after using gelatine as a pore forming agent and as a coagulant. At low concentration of gelatin, membranes were very weak. It is due to the low concentration of regenerated cellulose as described earlier for calcium chloride. Membranes formed by 25% gelatin shows lower pore size (10-30 $\mu\text{m}$ ) but more porosity as compared to 20% gelatine (20-50 $\mu\text{m}$ ).



**Figure 8—SEM micrograph of cellulose membranes by using [a], [b] 20% gelatin and [c], [d] 25% gelatine**





**Figure 9—SEM micrograph of cellulose membranes by using methanol as coagulant**

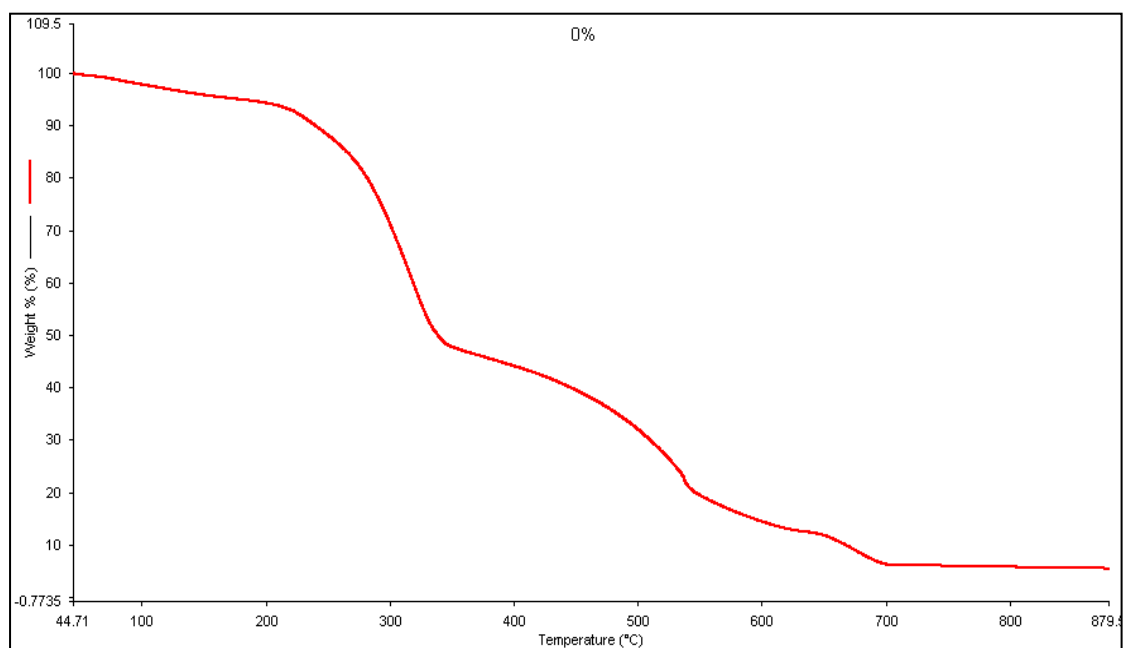
Figure 9 shows the SEM images of cellulose membranes fabricated by coagulating with methanol. No pores can be visible even at 5 µm resolution but the strength is greater than membranes fabricated by using calcium chloride and gelatin. So we may get non-porous membranes by using methanol as a coagulant. Since we obtained good strength and moderate porosity in calcium chloride coagulated regenerated membranes, the further studies are carried out using calcium chloride as a coagulant.

#### **4.7 Characterization of regenerated cellulose membranes with silver modified montmorillonite**

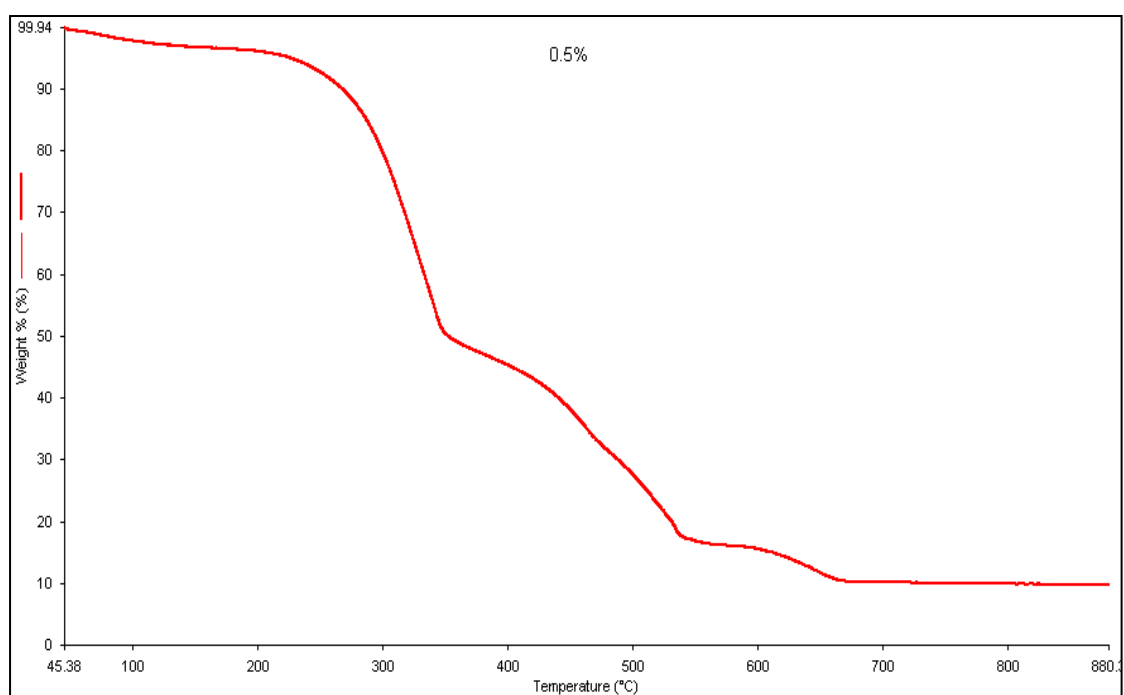
##### **4.7.1 Thermo Gravimetric Analysis (TGA)**

TGA measures the mass change of a sample as a function of time (or temperature). It is used to measure the composition and the thermal stability of the sample materials.

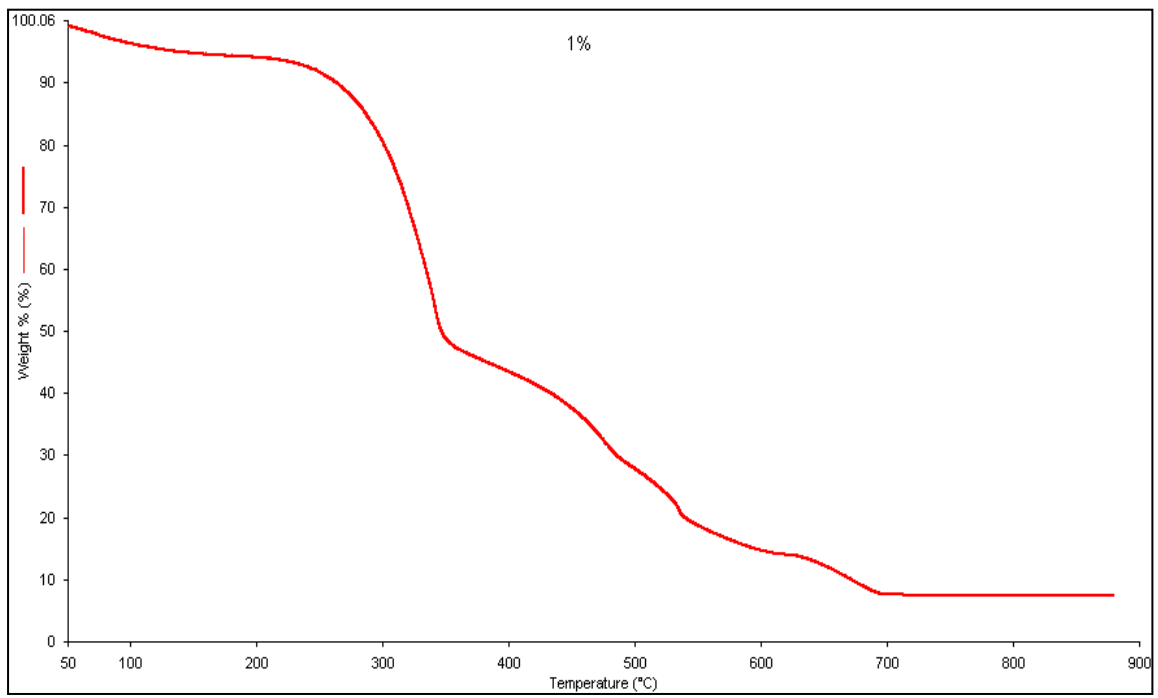
**[a]**



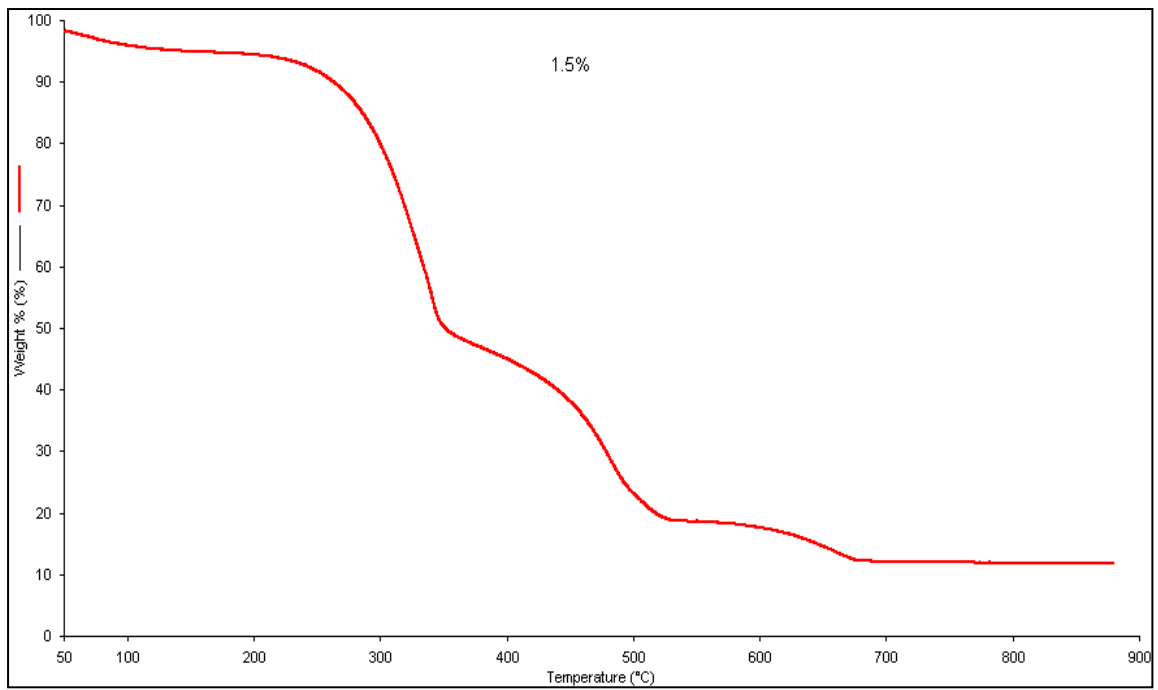
**[b]**



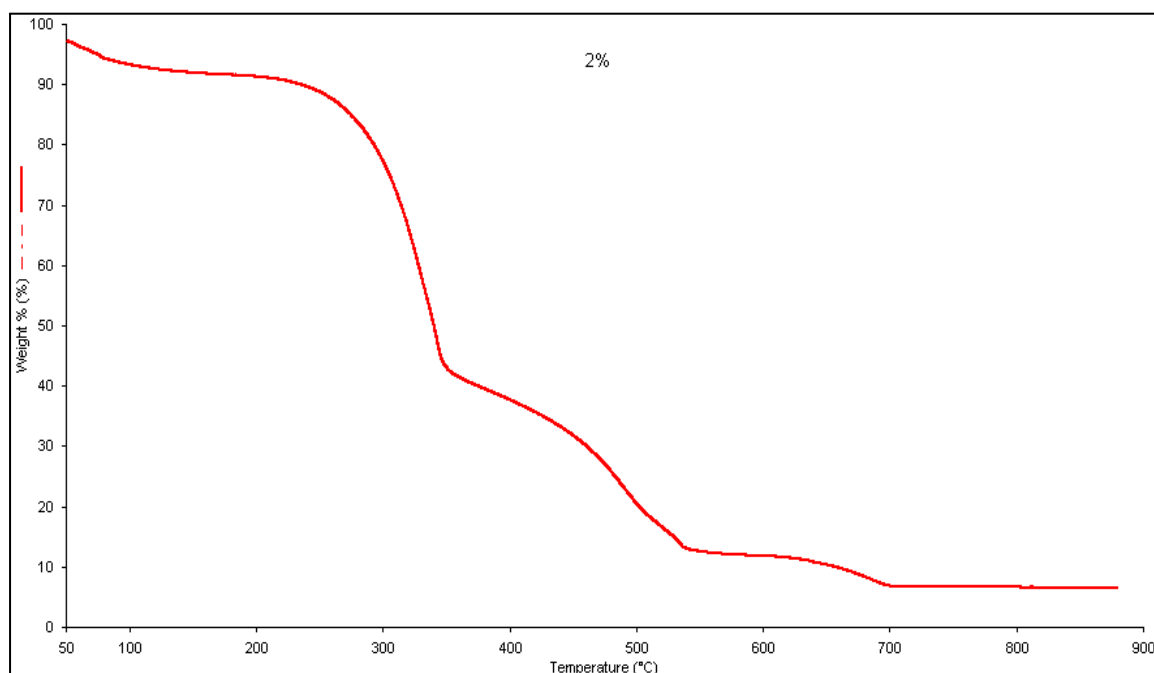
[c]



[d]



[e]



**Figure 10—TGA thermogram of regenerated cellulose membranes with [a] 0% clay, [b] 0.5% clay, [c] 1.0% clay, [d] 1.5% clay and [e] 2.0% clay (silver modified montmorillonite)**

By using TGA as an analytical tool, we can get an idea about the temperature range at which membranes can be used. It also shows the effect of incorporation of clay on the thermal properties of membranes.

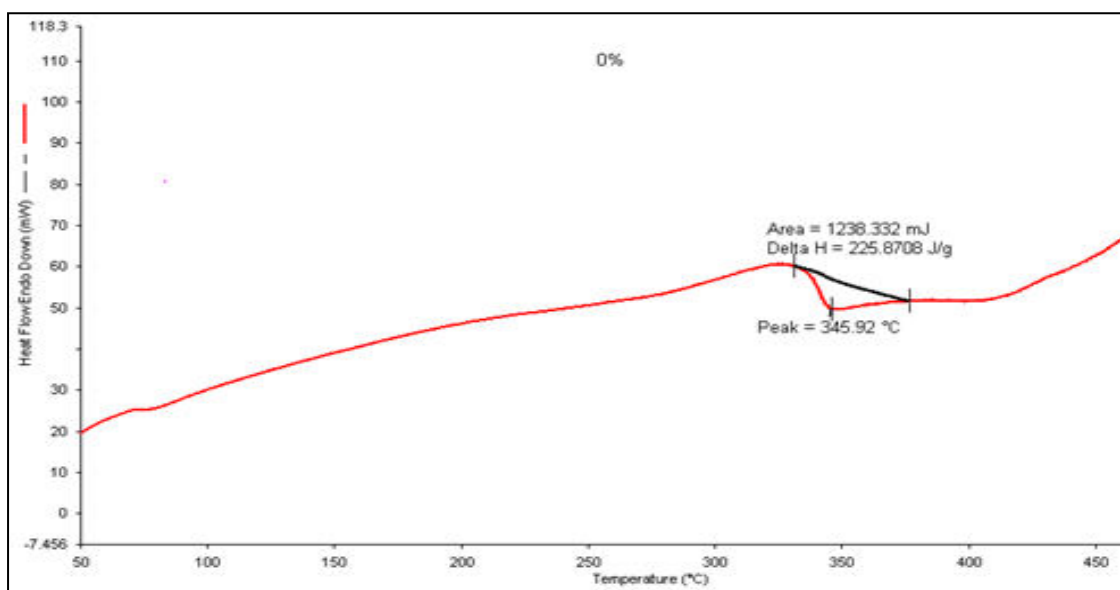
Figure 10 shows the TGA thermograms of regenerated cellulose membranes loaded with different concentration of silver modified montmorillonite. Thermal behaviour and thermal stability of the membranes is carried out by TGA in a nitrogen atmosphere. We can see that maximum weight loss temperature is around 320°C for all the membranes and weight loss at 320°C decreases by incorporating clay upto 1.5% concentration but again increases on increasing the concentration of montmorillonite to 2.0%. It shows the agglomeration of clay on increasing the concentration of clay above 1.5%. For the membranes without clay, a constant weight loss of 10% is reported upto the temperature of 200°C, then a rapid weight

loss is reported. But for the membranes with clay content, there is a weight loss of 2-5% upto the temperature of 100°C, then there is no apparent weight loss upto the temperature of 250°C. A rapid weight loss is start after 250°C. So we can say that membranes with 1.5% clay show better thermal properties as compared to other membranes and working temperature of membranes is increased from 200°C to 250°C by the incorporation of silver modified montmorillonite.

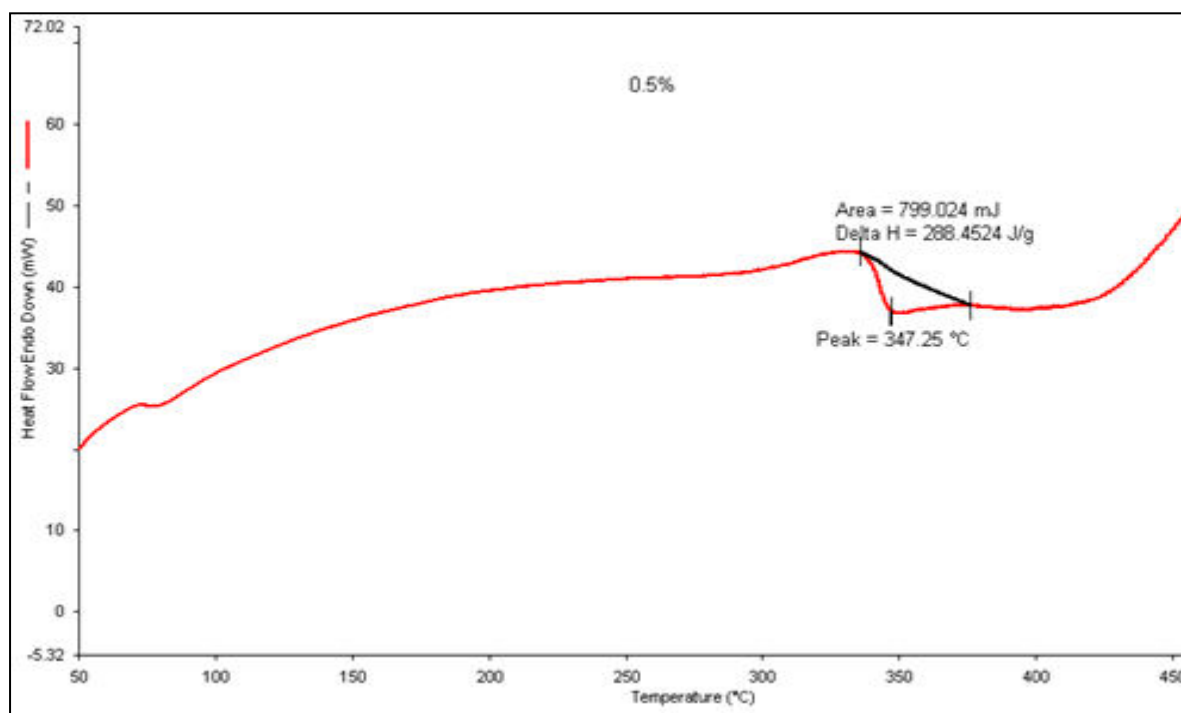
#### 4.7.2 Differential Thermal Analysis (DTA)

Differential Thermal Analysis (DTA), is a thermal analysis technique that gives an idea about the change of a material's heat capacity ( $C_p$ ) with temperature. For DTA measurement, a specimen of known mass is either heated or cooled and the heat capacity changes are recorded as changes in the heat flow. It also gives an idea about the degradation temperature and the crystallinity of the material. TG/DTA instrument may also be used to calculate heat flow by the right calibrations and is also be used for the DSC analysis. This is conducted by measuring temperature differences and heat changes between a specimen and a reference or the heat flux.

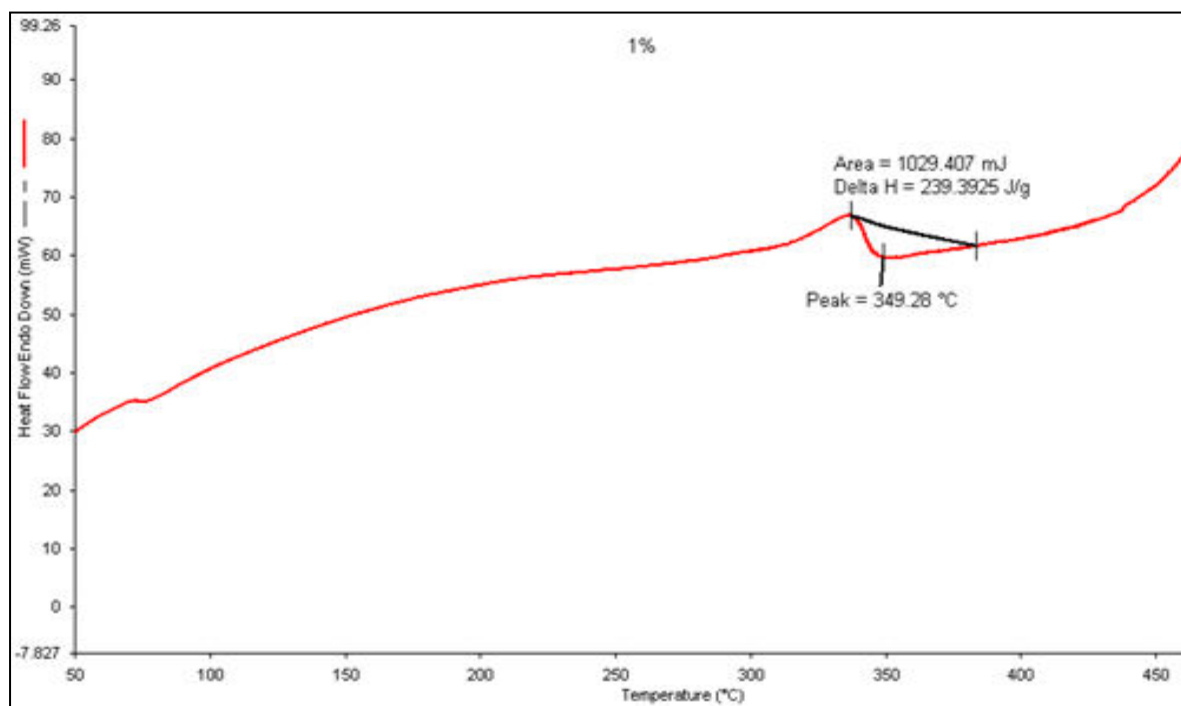
[a]



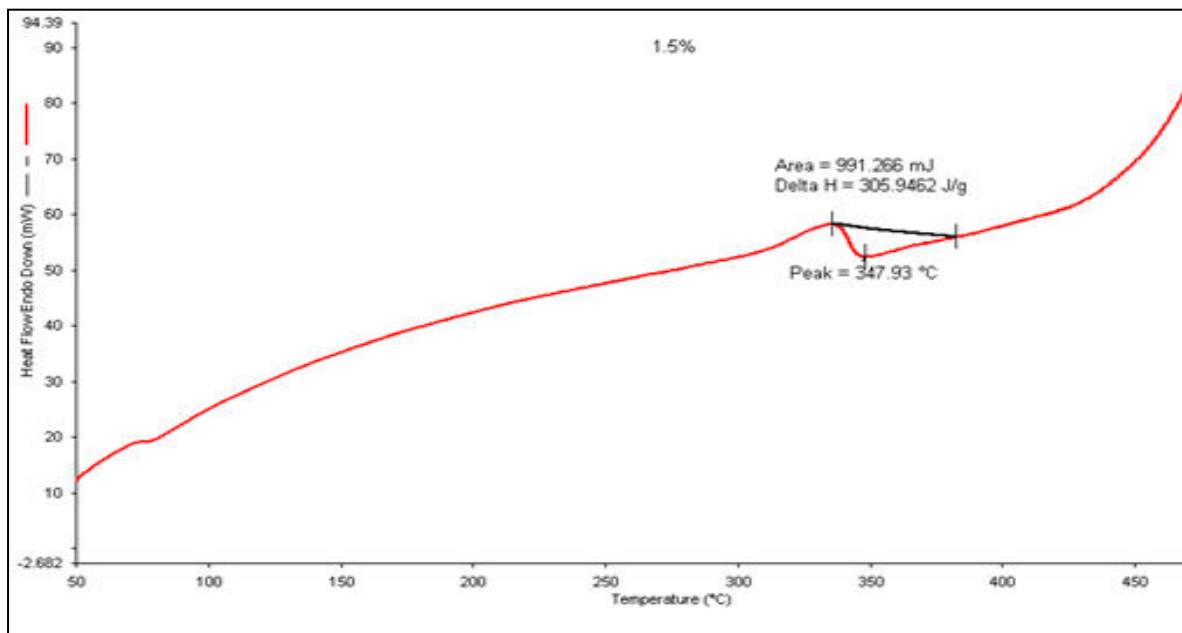
[b]



[c]



[d]



[e]

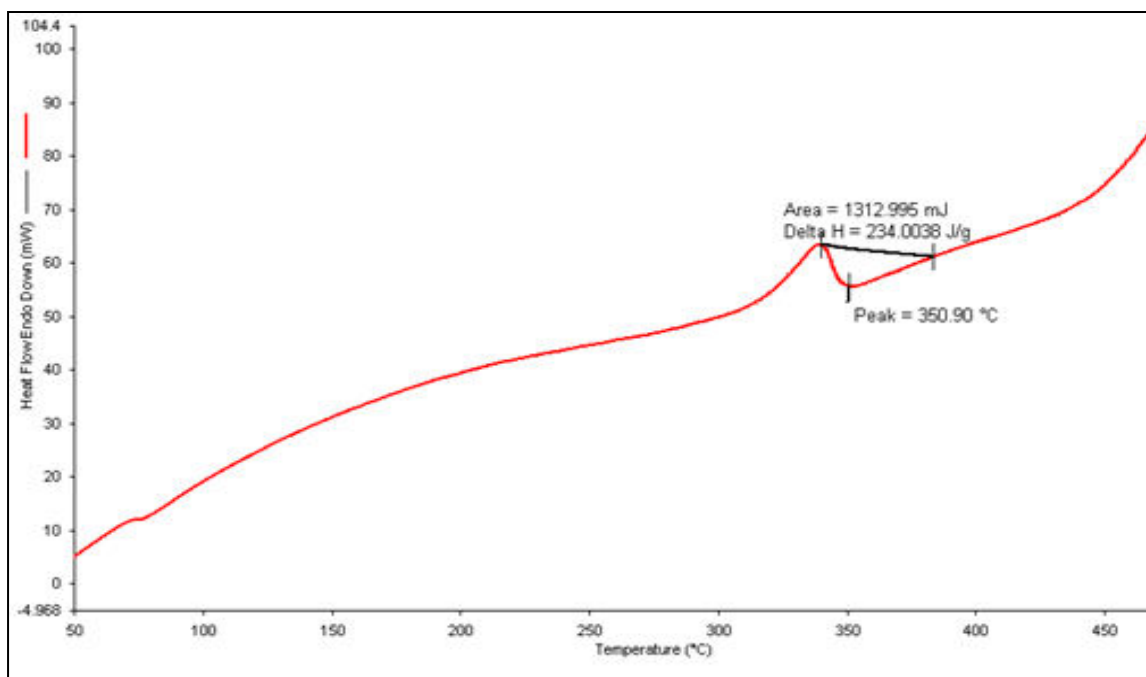


Figure 11—DTA plots for regenerated cellulose membranes with [a]0% clay, [b] 0.5% clay, [c] 1.0% clay, [d] 1.5% clay and [e] 2.0% clay(montmorillonite)

Like all DTA measurements, the TG/DTA instrument actually measures the temperature difference between specimen and reference and calculates heat flow from the calibrated data.

DTA curves of regenerated cellulose membranes are shown in figure 11. All the membranes show a major peak at temperature around 345°C-350°C, which is associated with the degradation or thermal depolymerisation of cellulose. Another peak around 470°C is attributed to the oxidation of the char. As the percentage of silver modified montmorillonite increases the degradation temperature also increases.

#### 4.7.3 Water content

Water content of the membranes is measured to get the information about the minimum amount of water required to wet the membranes. Water content of wet cellulose membrane is measured by using the following equation:

$$\text{Water content} = \frac{W_a - W_b}{W_b} \times 100$$

**Table 2— Data for water content measurement**

S.no.	Clay concentration(%)	Weight of wet membrane, $W_a$ (g)	Weight of dry membrane, $W_b$ (g)	Water content (%)
1.	0	0.0321	0.0104	208.6538
2.	0.5	0.0312	0.0098	218.3673
3.	1.0	0.0314	0.0093	237.6344
4.	1.5	0.0301	0.0087	245.977
5.	2.0	0.0308	0.0086	258.1395

Table 2 shows the data for water content measurement. It is clear from the data shown in table 2 that as the concentration of clay increases the water content of membranes also increases. It is due to the fact that the silver modified montmorillonite being a clay increases the absorption of water by the membranes.



#### 4.7.4 Flux measurement

The permeability of ultrapure water through the regenerated cellulose membranes is measured by using gravity filtration method. As the average pore size of these membranes is 10-20  $\mu\text{m}$ , they may be used as cell strainers or for pretreatment of RO, UF or NF membranes. For every membrane three reading are taken and the average is reported as time of filtration. Table 3 shows the data for rate of filtration and flux using gravity filtration method.

**Table 3—Data for rate of filtration through the membranes**

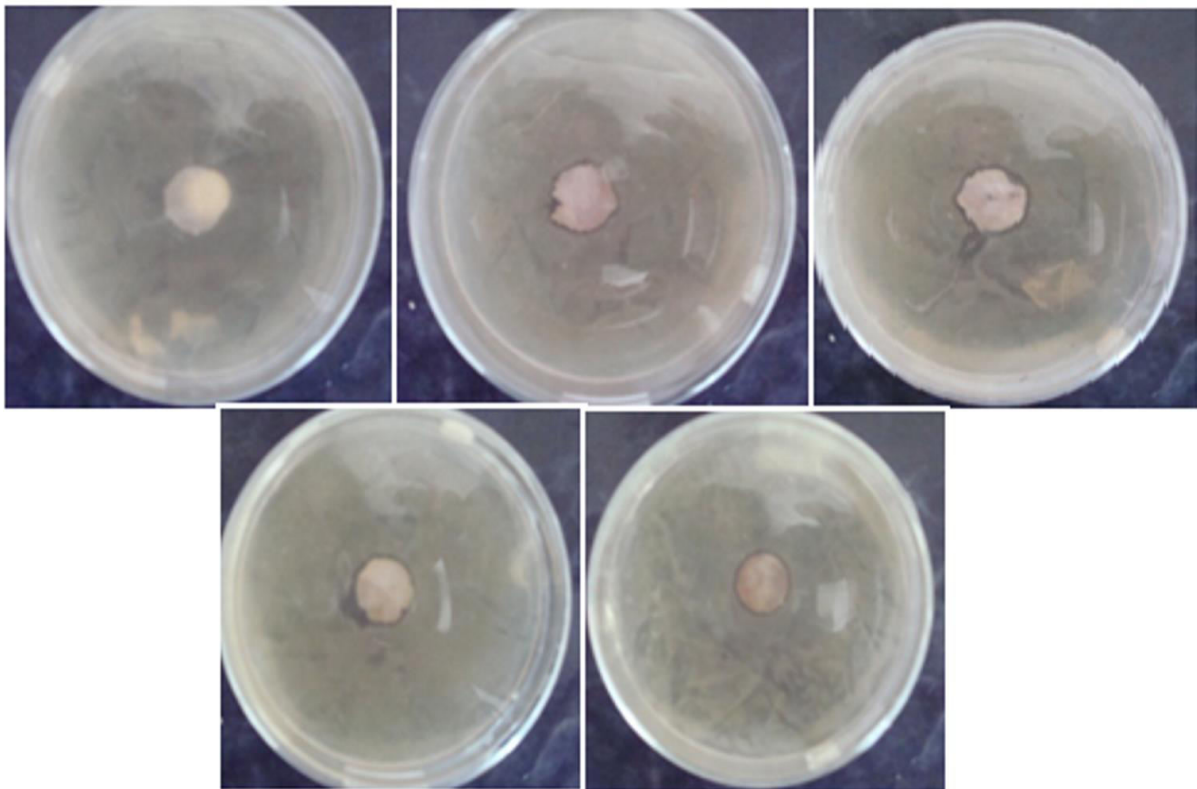
S.No.	Concentration of clay(%)	Time of filtration(250 mL of water)		Rate of filtration(mL/minute)	Flux (mL/min/cm <sup>2</sup> )
1.	0	1 <sup>M</sup> 2.16 <sup>S</sup>	1 <sup>M</sup> 2.16 <sup>S</sup>	241.3127	3.074
		1 <sup>M</sup> 2.18 <sup>S</sup>			
		1 <sup>M</sup> 2.14 <sup>S</sup>			
2.	0.5	1 <sup>M</sup> 10.76 <sup>S</sup>	1 <sup>M</sup> 10.71 <sup>S</sup>	212.1341	2.702
		1 <sup>M</sup> 10.81 <sup>S</sup>			
		1 <sup>M</sup> 10.56 <sup>S</sup>			
3.	1.0	1 <sup>M</sup> 12.32 <sup>S</sup>	1 <sup>M</sup> 12.37 <sup>S</sup>	207.2682	2.640
		1 <sup>M</sup> 12.42 <sup>S</sup>			
		1 <sup>M</sup> 12.36 <sup>S</sup>			
4.	1.5	1 <sup>M</sup> 16.31 <sup>S</sup>	1 <sup>M</sup> 16.48 <sup>S</sup>	196.1297	2.498
		1 <sup>M</sup> 16.61 <sup>S</sup>			
		1 <sup>M</sup> 16.51 <sup>S</sup>			
5.	2.0	1 <sup>M</sup> 7.16 <sup>S</sup>	1 <sup>M</sup> 6.36 <sup>S</sup>	226.0398	2.879
		1 <sup>M</sup> 6.75 <sup>S</sup>			
		1 <sup>M</sup> 5.16 <sup>S</sup>			

As the flux of water through these membranes first decreases on increase of the clay content upto 1.5%, then again increases on increasing the clay content further. It is attributed to the fact that montmorillonite remains in platelets form while finely dispersed in a solution. When the water passes through these platelets, it forms a torturous path for the water and the water

flux decreases. But after increasing the concentration of clay above 1.5%, clay starts to agglomerate giving a less torturous path for the passage of water.

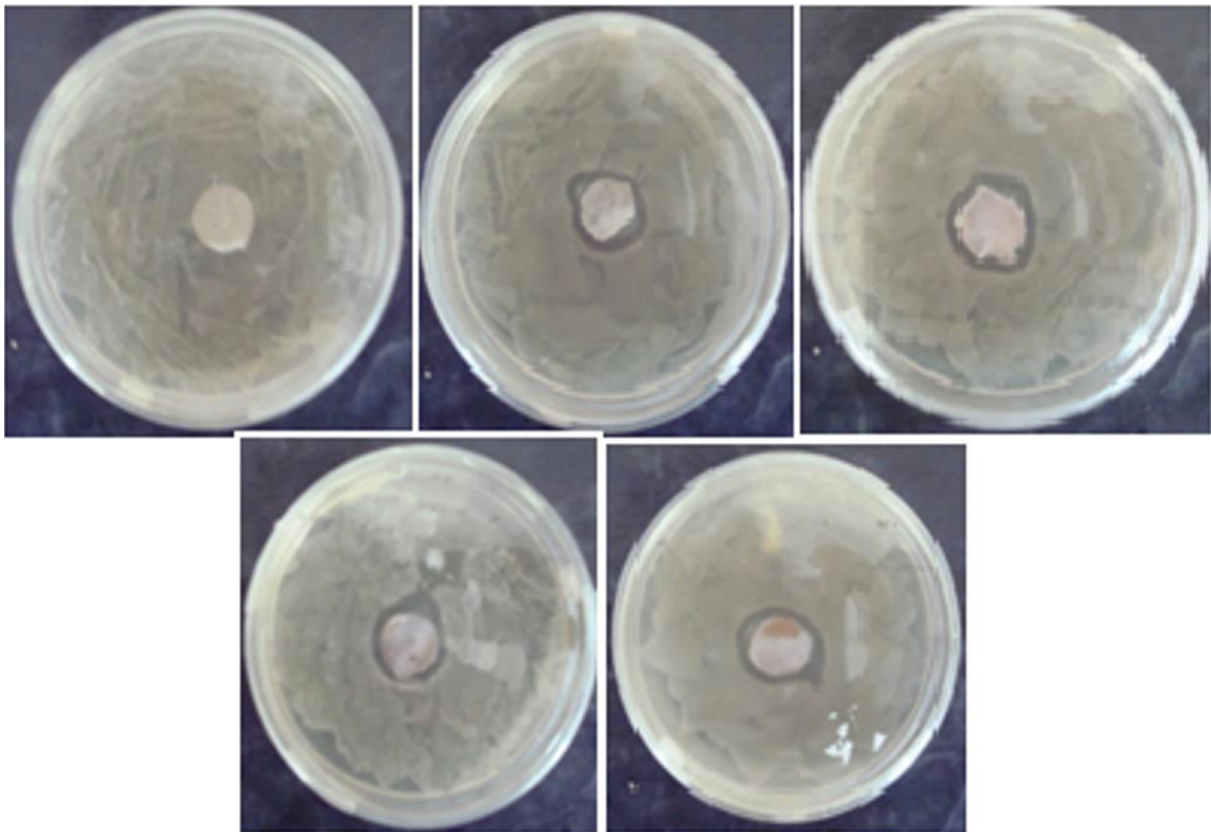
#### 4.7.5 Antimicrobial testing

As the fouling of membranes due to the bacterial activity is a major area of concern, we conducted antimicrobial testing on these membranes. Figures 10 and 11 shows the results of the disc susceptibility tests and the data of the results are given in Tables 4 and 5. Membranes loaded with silver modified clay show good antibacterial activity for both Gram positive bacteria species *S. aureus* and Gram negative bacteria species *E. Coli*. Zone of inhibition for the species *S. aureus* is greater than that of the species *E.coli*.



**Figure 12—Zone of inhibition for the species *E.coli* treated [a] 0% clay [b] 0.5% clay [c] 1.0% clay [d] 1.5% clay and [e] 2.0% clay silver modified montmorillonite loaded membranes**

The antibacterial properties of these regenerated cellulose membranes loaded with silver modified clay has been attributed to the fact that due to the electrostatic forces, of the negatively charged membrane of the bacteria to the surface of the clay, where the positive charged silver ions kills the bacteria or renders them unable to replicate.



**Figure 13—Zone of inhibition for *S.aureus* treated [a] 0% clay [b] 0.5% clay [c] 1.0% clay [d] 1.5% clay and [e] 2.0% clay (silver modified montmorillonite) loaded membranes**

The antibacterial activity of films is demonstrated by the diameter of the zone of inhibition in comparison to the membranes with no montmorillonite and calculated using the following equation:

$$C = \frac{W - T}{2}$$

Where C is the width of clear zone of inhibition (mm), w is the width of test specimen and clear zone (mm) and T is the width of test specimen (mm).

**Table 4—Data for zone of inhibition for the specimens treated with *E. coli***

S. no.	sample	Width of Specimen (T, mm)	Width of specimen and clear zone (W, mm)	Clear Zone of Inhibition (C, mm)
1.	0% clay	15	15	No growth
2.	0.5% clay	15	16	0.5
3.	1.0% clay	15	20	2.5
4.	1.5% clay	15	18	1.5
5.	2.0% clay	15	17	1.0

**Table 5—Zone of inhibition for membranes treated with *S. aureus***

S. no.	sample	Width of Specimen (D, mm)	Width of specimen and clear zone (T, mm)	Clear Zone of Inhibition (W, mm)
1.	0% clay	15	15	No growth
2.	0.5% clay	15	21	3.0
3.	1.0% clay	15	23	4.0
4.	1.5% clay	15	25	5.0
5.	2.0% clay	15	24	4.5

## CHAPTER 5: CONCLUSIONS

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Cellulose is successfully extracted from sugarcane bagasse and coir pith by autoclaving. After autoclaving we get approximately 30% fibrous residue from coir pith and 45% residue from sugarcane bagasse. The percentage of  $\alpha$ -cellulose is 88.25% in autoclaved sugarcane bagasse and the viscosity average molecular weight is found to be  $7.68 \times 10^4$ . Antibacterial membranes are fabricated by using 7% NaOH/12% urea as a solvent, calcium chloride as a pore forming agent and silver modified montmorillonite as an antibacterial agent. Water content of membranes increases on the increase of clay content. Thermal properties of the membranes are also enhanced slightly by incorporation of clay. The pore size of the membranes is in the range of 10-20 $\mu$ m so these membranes may be use as cell strainers or for pretreatment of RO, UF or NF processes. These membranes show good antibacterial property for both Gram positive bacteria species (*S. Aureus*) and Gram negative bacteria species (*E. Coli*). The flux of water is around 2.5-3.1 mL/min/cm<sup>2</sup> for all the membranes. The flux of water first decreases on increasing the concentration of clay upto 1.5%, then again increases on increasing the concentration of silver modified montmorillonite.

## CHAPTER 6: FUTURE PLANS

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As the pore size of these membranes is in the range of 10-20  $\mu\text{m}$ , these membranes can be used for pretreatment of reverse osmosis, ultrafiltration or nanofiltration processes. These membranes may also be used as the cell strainers. Cell strainers may be of different pore size ranging from 10-70  $\mu\text{m}$  depending upon the size of the cell species to be strained. As the size of spores is 3-40  $\mu\text{m}$  and that of yeast cells is 1-50  $\mu\text{m}$ , so we can separate them from other cells or these cells may be separated from their suspension.

These membranes may be used in air filters to separate airborne particles (solids suspended in the air or Inhalable Dust)

There is a scope to improve the mechanical strength of these membranes, as the only demerit of these membranes is their low mechanical strength.

## CHAPTER 7: REFERENCES

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- [1] W. Sun, J. Liu, H. Chu , B. Dong, Pretreatment and Membrane Hydrophilic Modification to Reduce Membrane Fouling, *Membranes*, 3 (2013) 226-241
- [2] R. Munter, industrial waste water treatment, 195-210, [http://www.balticuniv.uu.se/index.php/component/docman/doc\\_download/286-water-use-and-management-19-industrial-wastewater-treatment](http://www.balticuniv.uu.se/index.php/component/docman/doc_download/286-water-use-and-management-19-industrial-wastewater-treatment)
- [3] S. Kamel, Nanotechnology and its applications in lignocellulosic composites, a mini review, *eXPRESS Polym. Lett.*, 1(9) (2007) 546–575
- [4] K. L. T. Carrillo, M. Tagaya, T. Kobayashi, Bamboo Fibers Elaborating Cellulose Hydrogel Films for Medical Applications, *J. Mater. Sci. Chem. Eng.* ,1 (2013) 7-12
- [5] E.I Unuabonah, A. Taubert, Clay–polymer nanocomposites (CPNs): Adsorbents of the future for water treatment, *Appl. Clay Sci.* (2014), <http://dx.doi.org/10.1016/j.clay.2014.06.016>
- [6] X. Xiong, J. Duan, W. Zou, X. He, W. Zheng, A pH-sensitive regenerated cellulose membrane, *J. Membrane Sci.*, 363 (2010) 96–102
- [7] L. M. Nevárez, L. B. Casarrubias, O. S. Canto, A. Celzard, V. Fierro, R. I. Gómez, G. G. Sánchez, Biopolymersbased nanocomposites: Membranes from propionated lignin and cellulose for water purification, *Carbohydr. Polym.*, 86 (2011) 732– 741
- [8] F. Cotana, G. Cavalaglio, A. Nicolini, M. Gelosia, V. Coccia, A. Petrozzi, L. Brinchi, Lignin as co-product of second generation bioethanol production from ligno-cellulosic biomass, *Energ. Procedia*, 45 ( 2014 ) 52 – 60

- [9] R. Narendar, K. Priya Dasan, Chemical treatments of coir pith: Morphology, chemical composition, thermal and water retention behaviour, *Composites: Part B*, 56 (2014) 770–779
- [10] S. Prabu, A .G Murugesan, Effective utilization and management of coir industrial waste for the production of poly- $\beta$  - hydroxybutyrate (PHB) using the bacterium *Azotobacter Beijerinckii*, *Int. J. Environ. Res.*, 4(3) (2010) 519-524
- [11] U.J. Etim, S.A. Umoren, U.M. Eduok, Coconut coir dust as a low cost adsorbent for the removal of cationic dye from aqueous solution, *J. Saudi Chemical Society*, <http://dx.doi.org/10.1016/j.jscs.2012.09.014>, (2012) 1-10
- [12] M. A. M. Salleh , D. K. Mahmoud , W. A. W. A. Karim , A. Idris, Cationic and anionic dye adsorption by agricultural solid wastes: A comprehensive review, *Desalination*, 280 (2011) 1–13
- [13] M. T. Yagub , T. K. Sen, S. Afroze , H.M. Ang, Dye and its removal from aqueous solution by adsorption: A review, *Adv. Colloid Interfac.*, 209 (2014) 172–184
- [14] M.V. Sureshkumar, C. Namasivayam, Adsorption behavior of Direct Red 12B and Rhodamine B from water onto surfactant-modified coconut coir pith, *Colloid. Surface. A*, 317 (2008) 277–283
- [15] G. R. Filho, D. S. Monteiro, C. S. Meireles, R. M. N. Assunc, D. A. Cerqueira , H. S. Barud, Sidney J.L. Ribeiro, Y. Messadeq, Synthesis and characterization of cellulose acetate produced from recycled newspaper, *Carbohydr. Polym.* , 73 (2008) 74–82
- [16] G. R. Filho, R.M.N. de Assuncao, J. G. Vieira, C. D. S. Meireles, D. A. Cerqueira, H. D. S. Barud, S.J.L. Ribeiro, Y. Messaddeq, Characterization of methylcellulose produced from sugar cane bagasse cellulose: Crystallinity and thermal properties, *Polym. Degrad. Stabil.*, 92 (2007) 205-210
- [17] G. R. Filho, S. D. Ribeiro, C. S. Meireles, L. G. Silva, R. Ruggiero, M. F. F. Junior, D. A. Cerqueira, R. M. N. Assunc, M. Zeni, P. Polleto, Release of doxycycline through cellulose



acetate symmetric and asymmetric membranes produced from recycled agroindustrial residue: Sugarcane bagasse, *Ind. Crop. Prod.*, 33 (2011) 566–571

[18] X. Qiu, S. Hu, “Smart” Materials Based on Cellulose: A Review of the Preparations, Properties, and Applications, *Materials*, 6(2013) 738-781

[19] C. D. S. Meireles, G. R. Filho, M. F. Ferreira Jr., D. A. Cerqueira, R. M. N. Assunção, E. A. M. Ribeiro, P. Poletto, M. Zeni, Characterization of asymmetric membranes of cellulose acetate from biomass: Newspaper and mango seed, *Carbohydr. Polym.*, 80 (2010) 954–961

[20] M. Kihlman, Dissolution of cellulose for textile fibre applications, (2012), <http://www.diva-portal.org/smash/get/diva2:513750/FULLTEXT03>

[21] T. Liebert ,Cellulose Solvents – Remarkable History, Bright Future, *American Chemical Society*,(2010) 1-54, <http://pubs.acs.org>

[22] I. Kilpelainen, H. Xie, A. King, M. Granstrom, S. Heikkinen, D. S. Argyropoulos, Dissolution of Wood in Ionic Liquids, *J. Agric. Food Chem.*, 55( 2007) 9142–9148

[23] S. Sen, J. D. Martin, D. S. Argyropoulos, Review of Cellulose Non-Derivatizing Solvent Interactions with Emphasis on Activity in Inorganic Molten Salt Hydrates, *ACS Sustainable Chemistry & Eng.*, (2013)1-13

[24] M. Ioelovich, Optimal Conditions for Isolation of Nanocrystalline Cellulose Particles, *Nanosci. Nanotechnol.*, 2(2) (2012) 9-13

[25] J. Pang, X. Liu, X. Zhang, Y. Wu, R. Sun, Fabrication of Cellulose Film with Enhanced Mechanical Properties in Ionic Liquid 1-Allyl-3-methylimidazolium Chloride (AmimCl),*Materials*, 6( 2013) 1270-1284

[26] W. Liu, T. Budtova, P. Navard, Influence of ZnO on the properties of dilute and semi-dilute cellulose-NaOH-water solutions, *Cellulose*, 18 (2011) 911–920

[27] D. Ruan, L. Zhang, Y. Mao, M. Zeng, X. Li, Microporous membranes prepared from cellulose in NaOH/thiourea aqueous solution, *J. Membrane Sci.*, 241 (2004) 265–274

- [28] J. Cai, L. Zhang, Rapid Dissolution of Cellulose in LiOH/Urea and NaOH/Urea Aqueous Solutions, *Macromol. Biosci.*, 5(2005) 539–548
- [29] L. Yan, Z. Gao, Dissolving of cellulose in PEG/NaOH aqueous solution, *Cellulose*, 15(2008)789–796
- [30] R. Li, S. Wang, A. Lu, L. Zhang, Dissolution of cellulose from different sources in an NaOH/urea aqueous system at low temperature, *Cellulose*, 22 (2015) 339–349
- [31] S. Zhang, F. Li, J. Yu, L. Gu, Novel Fibers Prepared from Cellulose in NaOH/Thiourea/Urea Aqueous Solution, *Fiber. Polym.*, 10(1) (2009) 34-39
- [32] S. Zhang, W. C. Wang, F. X. Li , J. Y. Yu, Swelling and dissolution of cellulose in NaOH aqueous solvent systems, *Cellulose Chem. Technol.*, 47 (2013) 671-679
- [33] Y. Mao, J. Zhou, J. Cai, L. Zhang , Effects of coagulants on porous structure of membranes prepared from cellulose in NaOH/urea aqueous solution, *J. Membrane Sci.*, 279 (2006) 246–255
- [34] B. Medronho, B. Lindman, Brief overview on cellulose dissolution/regeneration interactions and mechanisms, *Adv. Colloid Interfac.*, (2014) <http://dx.doi.org/10.1016/j.cis.2014.05.004>
- [35] Z. Wang, M. Xue, K. Huang, Z. Liu, Textile Dyeing Wastewater Treatment, Advances in Treating Textile Effluent, <http://www.intechopen.com/books/advances-in-treating-textile-effluent/textile-dyeing-wastewatertreatment>, (2011) 91-116
- [36] F. M. D. Chequer, G. A. R. de Oliveira, E. R. A. Ferraz, J. C. Cardoso, M. V. B. Zanoni, D. P. de Oliveira, Textile Dyes: Dyeing Process and Environmental Impact, Eco-Friendly Textile Dyeing and Finishing, (2013). <http://www.intechopen.com/books/eco-friendly-textile-dyeing-and-finishing/textile-dyes-dyeing-process-and-environmental-impact>

- [37] K. Singh and S. Arora, Removal of Synthetic Textile Dyes From Wastewaters: A Critical Review on Present Treatment Technologies, *Crit. Rev. Env. Sci. Tec.*, 41 (2011) 807–878
- [38] A. Pandey, R. S. Carlos, P. Nigam, T. S. Vanete, Biotechnological potential of agro-industrial residues. I: sugarcane bagasse, *Bioresource Technol.*, 74 (2000) 69–80
- [39] J.X. Sun, X.F. Sun, H. Zhao, R.C. Sun, Isolation and characterization of cellulose from sugarcane bagasse, *Polym. Degrad. Stabil.*, 84 (2004) 331-339
- [40] J.X. Sun, F. Xu, X.F. Sun, B. Xiao, R.C. Sun, Physico-chemical and thermal characterization of cellulose from barley straw, *Polym. Degrad. Stabil.*, 88 (2005) 521-531
- [41] C. Feng, K.C. Khulbe , T. Matsuura , S. Tabe , A.F. Ismail , Preparation and characterization of electro-spun nanofiber membranes and their possible applications in water treatment, *Sep. Purif. Technol.*, 102 (2013) 118–135
- [42] J. Yin, B. Deng, Polymer-matrix nanocomposite membranes for water treatment, *J. Membrane Sci.*, 479 (2015) 256–275
- [43] Y. Ma, F. Shi, Z. Wang, M. Wu, J. Ma, C. Gao, Preparation and characterization of PSf/clay nanocomposite membranes with PEG 400 as a pore forming additive, *Desalination*, 286 (2012) 131–137
- [44] X.S. Fan, Z.T. Liu, Z.W. Liu, J. Lu, Cellulose Acetate Membrane Synthesis from Biomass of Ramie, *J. Appl. Polym. Sci.*, 117 (2010) 588–595
- [45] V. Kochkodan, N. Hilal, A comprehensive review on surface modified polymer membranes for biofouling mitigation, *Desalination*, (2014), <http://dx.doi.org/10.1016/j.desal.2014.09.015>
- [46] S. B. Khan, A. A. Khalid, N. B. Elham, M. A. Abdullah, Y. Muhammad, G. Lassaad, R. Z. Ahmad, Assessment of antibacterial cellulose nanocomposites for water permeability and salt rejection, *J. Ind. Eng. Chem.*, (2014), <http://dx.doi.org/10.1016/j.jiec.2014.09.040>

[47] J. Zhou, L. Zhang and J. Cai, Behavior of cellulose in NaOH/Urea aqueous solution characterized by light scattering and viscometry, *J. Polym. Sci. Pol. Phys.*, 42 (2004) 347–

353