MEMBRANE SEPARATION OF ACID FROM LIGNOCELLULOSIC HYDROLYSATE

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in

Bioprocess Engineering

by

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(Enrollment no. 16556011)

under the supervision of

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DEPARTMENT OF BIOTECHNOLOGY

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May 9th, 2018

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I, after checking the dissertation mentioned above, hereby states my approval of the dissertation submitted in partial fulfillment of the requirements for the degree of Master of Technology in Bioprocess Engineering at Indian Institute of Technology Roorkee. I am satisfied with the volume, quality, correctness, and originality of the work.

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This is to certify that the project dissertation entitled "**MEMBRANE SEPARATION OF ACID FROM LIGNOCELLULOSIC HYDROLYSATE**" by **Shristi Baxi** (Enrollment No: **16556011**) in partial fulfillment of the requirements for the award of Degree of Master of Technology in Bioprocess Engineering at Indian Institute of Technology Roorkee is an authentic work carried out by her during a period from July 2017 to May 2018 under my supervision and guidance. To my best of knowledge, neither this dissertation nor any part of it has been submitted to any university or institute in India or abroad for the award of any degree.

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I, **Shristi baxi** (Enrollment No. **16556011**), hereby declare that this dissertation entitled **MEMBRANE SEPARATION OF ACID FROM LIGNOCELLULOSIC HYDROLYSATE** represents my original work carried out as a postgraduate student of IIT Roorkee and, to the best of my knowledge, it contains no material previously published or written by another person, nor any material presented for the award of any other degree of IIT Roorkee or any other institution. Any contribution made to this research by others, with whom I have worked at IIT Roorkee or elsewhere, is explicitly acknowledged in the dissertation. Works of other authors cited in this dissertation have been duly acknowledged under the section "Bibliography". I have also submitted my original research records to the scrutiny committee for evaluation of my dissertation.

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Shristi Baxi

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May 9th, 2018

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ABSTRACT

Conventional bioethanol production process consists of four major steps: pretreatment, hydrolysis, detoxification and fermentation. Acid hydrolysis of lignocellulosic biomass is a crucial technique to break the rigid crystalline structure of lignocellulosic biomass into carbohydrate monomers. However, acid present in the hydrolysate inhibits the fermentation of the cellulosic liquor into bioethanol. Currently, over liming followed by precipitation is used in the pilot scale facilities. In the present work, the recovery of sulfuric acid from lignocellulosic hydrolysate by means of single cell electrodialysis is proposed for bioethanol production

Henceforth, to increase the performance and decrease the cost of the process, effective electrochemical separation using ion exchange membranes was evaluated. The limiting current was determined by operating the electrodialysis unit in potentiostatic mode as with the increase in current, recovery of acid decreases. This is attributed to the enhancement of water dissociation as the limiting current density exceeds and equilibrium shift at the solution/ membrane boundary as more sulfate ions cross the anion exchange membrane. The effects of flow rates on the current efficiency were also investigated as increased flow rate increases the amount of ion removal while increasing the mean current. The concentration of the reducing sugars, i.e., glucose and xylose in the was examined before and after the run and after the dialysis run. It was reported that there was a minimal change in the concentration of both glucose and xylose. We have reported approximately 70% recovery of the sulfuric acid from lignocellulosic liquor at an optimal flow rate of 18ml/min at an advantageous limiting current of 1.5 mA. We also to check the total reducing sugar concentration and check the separation efficiency. The separation of sulfuric acid from lignocellulosic hydrolysate increased indicating the importance of number of cell pair as a vital parameter.

Keywords: Lignocellulosic hydrolysate, Electrodialysis, Ion exchange membranes, Limiting current density.

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ABBREVIATIONS AND SYMBOLS	
NH_4OH	Ammonium hydroxide
N	Amount of ions removed during time t
A	Ampere
AEM	Anion Exchange Membrane
A	Anode
~	Approximately
$Ca(OH)_2$	Calcium hydroxide
CaO	Calcium oxide
$CaSO_4$	Calcium sulfate
C	Cathode
CEM	Cation Exchange Membrane
cm	Centimeter
Ct	Concentration at time t
CC	Concentrate Chamber
I	Current
CE	Current (coulombic) efficiency
°C	Degree Celsius
DC	Diluate Chamber
ED	Electrodialyser
F	Faraday's constant
1G	First Generation
g/l	Gram per liter
>	Greater than
h	Hour
HCL	Hydrochloric acid

H^+	Hydrogen ion
OH^-	Hydroxyl ion
C_o	Initial concentration
I_{lim}	Limiting Current
L	Liter
mg	Milligram
mL	Milliliter
mm	Millimeter
mL/min	Milliliter per minute
mV	Millivolt
MT	Million tones
min	Minute
M	Molarity
N	Number of membrane pairs
Δn	Number of moles removed in time t
%	Percentage
KOH	Potassium hydroxide
pH	Potential of Hydrogen
RE	Removal Efficiency
s	Second
2G	Second Generation
NaOH	Sodium hydroxide
S	Spacer
SEEC	specific electrical energy consumption
SO_4^{2-}	Sulfate ion
H_2SO_4	Sulfuric Acid

z	Valency of ion
V	Voltage
w/w	Weight by weight
wt %	Weight percentage

CHAPTER 1

INTRODUCTION

Over the years, the depletion of petroleum resources has created environmental and political concerns paving ways for increased demand to produce renewable fuels. 5% biofuel blending has been set mandatory by Government of India in both petrol and diesel. Presently, diesel biofuel blending is insignificant and petrol blending is only around 3% from molasses-based ethanol. In India, the annual requirement of ethanol is about 5×10^{10} L but total installed capacity is just about half. Therefore, the target of 20% blending by 2020 which is set by biofuel policy of India looks too far unless bioethanol production technologies are successfully demonstrated commercially (Mishra et al., 2018).

Bioethanol is produced by sugar fermentation and the origin of sugar determines its generation. Starch as the source of fermentable sugars produces first generation or 1G ethanol, which is found in the feedstocks like sugarcane and cereals. However, 1G ethanol leads to food-fuel conflicts. Hence, focus has shifted to second generation or 2G ethanol, which is produced from cellulosic and hemi cellulosic plant biomass like agricultural residues, municipal wastes and corn stover and wheat straw. With annual production estimated 1×10^{10} MT worldwide lignocellulose forms the most abundant renewable biomass reducing high energy cost and shortages in the upcoming days (Sánchez and Cardona, 2008). *Saccharum spontaneum* (Kansas grass) is a tall perennial grass, with high level of carbohydrate in its cell wall (~68% on a dry solid basis), making it a suitable feedstock for ethanol production (Chandel et al. 2011).

A new technique has been developed in our lab which can convert lignocellulosic biomass to fuel ethanol in just two process steps with high conversion efficiency and thus hope to bring down the cost effectively. This technique gives pentose and hexose sugars as separate fractions, direct from lignocellulosic biomass with negligible toxics. Fig. 1 represents a schematic diagram of bioethanol production from lignocellulosic biomass using fractional hydrolysis process. Fractional hydrolysis process is tested with various inorganic acids; highest scarification was achieved using sulfuric acid (~85%). Hemi cellulosic part of lignocellulose degrades to produce xylose, mannose, acetic acid, galactose and glucose, while cellulosic part

produces glucose (Eva et al., 2000). However, the presence of acid in the hydrolysate liquor is detrimental for the fermentation.

Conditioning of the hydrolysate for acid removal and exposing the sugars for fermentation is generally categorized into biological, physical and chemical methods. Biological and chemical methods focus on the conversion of more toxic compounds to less toxic compounds while physical method focuses on removal of toxic compounds form hydrolysate (Pienkos, 2009). Currently, sulfuric acid is being removed by adding lime (CaO) to the hydrolysate liquor and precipitating the sulfate ions as gypsum ($CaSO_4$). Conditioning with lime is extensively used as it is relatively cheap and effective. However, over liming increases the cost, time, use of additional chemicals and equipment. Furthermore, formation of insoluble gypsum hinders the scale up of the ethanol production process (Datta et al., 2013). To overcome all these prevailing challenges, in the present work, an efficient separation process- ion exchange membrane electrodialysis is used for removal of sulfuric acid from hydrolysate obtained from Kansas grass biomass after fractional hydrolysis process. This method provides a cost-effective pathway with fewer unit operations, lower operation time, and reduced use of chemicals and water.

As the current passes through ion selective membranes, ions are removed from the solution near the diluate surface of the membrane and concentrates at the concentrate section by convection and diffusion forming a stationary layer which increases current density. Further, leading to the disassociation of the water molecules, if the ion concentration reaches zero forming H^+ and OH^- ions. The electric current from this point is termed as "limiting current". It is directly proportional to the bulk concentration of the ions and inversely proportional to the thickness of the stationary layer and the ion selectivity of the membrane. (Ibanez et. al., 2004).

Flow rate is another essential parameter which affects removal efficiency of ions decreasing liquid film formed between at the interface of membrane surface and the liquid adjacent to the membrane thus causing a decrease in concentration polarization. Conversely, increase in flow rate decreases the current efficiency which leads to higher energy consumption as the amount of the material removed remains consistent even though mean current passing through the electrodialysis stack increases.

In this study, electrodialysis was employed to remove sulfuric acid from dilute acid pre-treated Kansas grass hydrolysate liquor. This strategy also paves way of sulfuric acid recovery into nearly pure acid streams and reuse while parallelly proving the feasibility of the process

at a larger scale. This selective separation process would enable reuse of sulfuric acid used in the acid pre-treatment step. Glucose and xylose were also estimated at both beginning and end of the process to know the process efficiency.

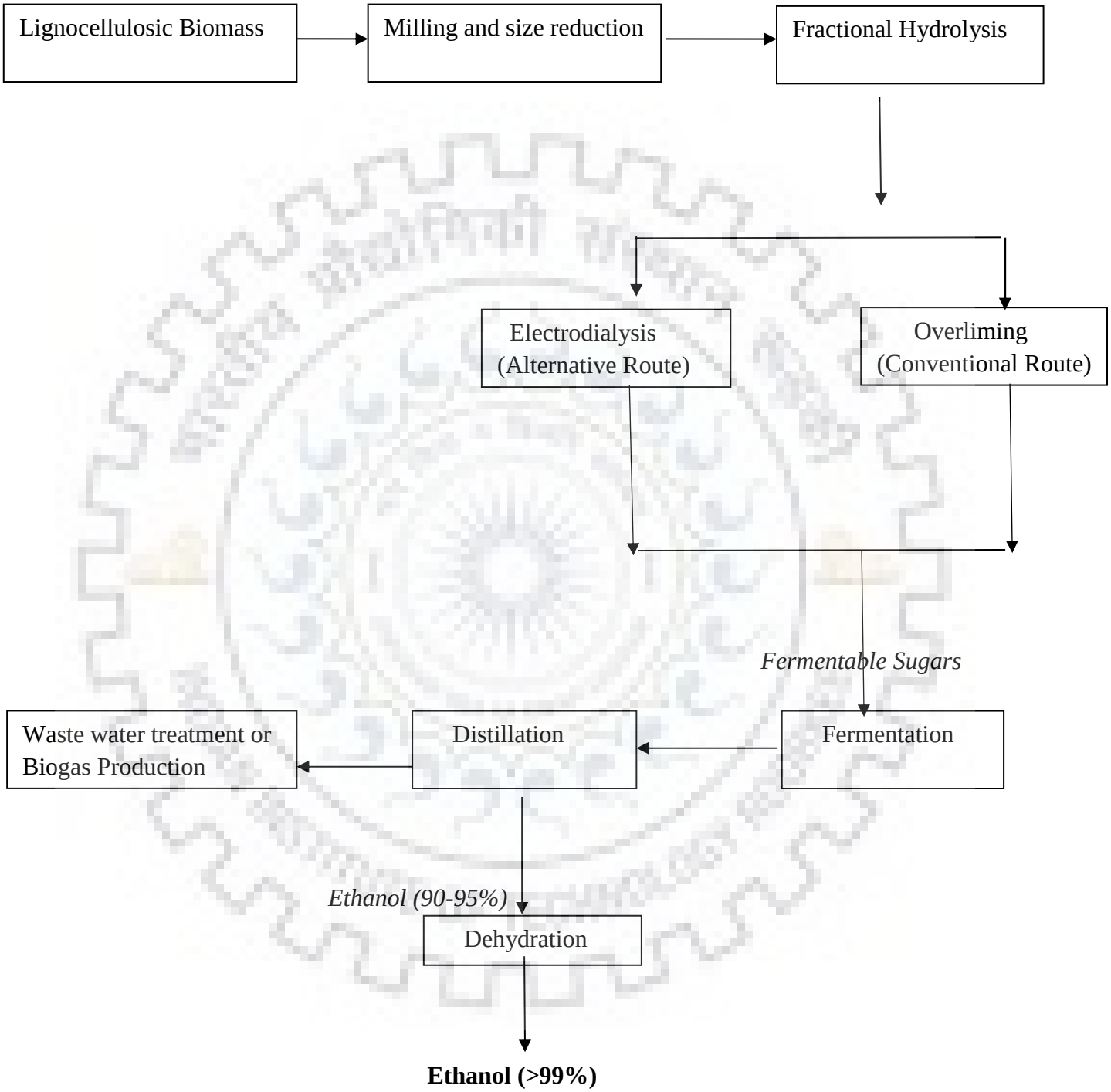


Figure 1.1: Schematic of the lignocellulosic biomass derived biofuel production pathway and the role of intermediate separation steps. Electrodialysis could be used as an alternative option

for removing sulfuric acid instead of over liming. ED enables a pathway with fewer unit operations, reduced use of chemicals (lime and others) and water, and the potential of recycling the acid.

1.1 Objective

The operation of the ED unit at constant voltage in potentiostatic mode will be taken to demonstrate the ability and efficiency of this novel operation.

The key objectives for this study are as follows:

- Fabrication of membrane-based dialysis chamber by coupling with flowmeters and power supply.
- Evaluation of limiting current for synthetic hydrolysate liquor and confirm whether the same limiting current value applies to the Kansas grass hydrolysate liquor.
- Analysis of the effect of flow rate on current for Kansas grass hydrolysate liquor.
- Comparative analysis of separation performance of synthetic hydrolysate liquor and Kansas grass hydrolysate liquor.
- Investigation of change in the concentration of reducing sugars in the acid hydrolysate.
- Reusability study of the strength of sulfuric acid obtained.

1.2 Significance of the present study

- A novel electrodialyser was fabricated using ion exchange membranes for the separation of sulfuric acid from the lignocellulosic hydrolysate.
- Optimum value for limiting current was unveiled for the measure synthetic hydrolysate liquor followed by Kansas grass hydrolysate liquor.
- Separation performance was analyzed of synthetic hydrolysate liquor and Kansas grass hydrolysate liquor as a function of time.
- The concentration of reducing sugars in the acid hydrolysate was analyzed to investigate the loss of reducing sugars during the experiment.
- Reusability studies were performed to evaluate the strength of sulfuric acid obtained from the lignocellulosic hydrolysate.
- The number of cell pairs were increased to check importance of number of cell pairs as an operational parameter.

CHAPTER 2

REVIEW OF LITERATURE

2.1 Status of the problem

Due to the high cost required to run Electrodialysis in parallel to the other process needed instead makes Electrodialysis a confined field of usage. Henceforth, the existing prior art is more found in the field of dealing with maximization in ion removal such as purification of amino acids, water purification or microelectronics. The work presented here deals with the separation of sulfuric acid from lignocellulosic acid hydrolysate by using electrodialysis is still at a budding stage and is not being widely used for industrial applications.

Maigrot and Sabates (1890) insinuated the idea of combining electrolysis and dialysis as an original concept. However, the foremost manuscript was published in a scientific periodical using was by Morse and Pierce (1903). Unfortunately, while remembering the 100th issue of the Journal of Membrane Science in 1995 electrodialysis went unmentioned though standard membrane separation works were published.

Goldstein et. a1. 1989 produced an inclusive study on the recovery of sulfuric acid and hydrochloric acid by electrodialysis and its fiscal allegations for reduced wood acid hydrolysis. The invention discloses a method for separating acid from an acid-sugar hydrolysate in an electrodialysis apparatus comprising the steps of continuously passing the hydrolysate through a diluate compartment of the electrodialysis apparatus, continuously passing a carrier fluid through a concentrate compartment of the electro dialysis apparatus, and maintaining a current between the anode-cathode pair of the electrodialysis apparatus that forces the anions and cations of the acid to migrate from the diluate compartment into a concentrate compartment..

Several works have been done on lignocellulosic hydrolysate conditioning obtained from acid-based processes in the past. Dilute acid hydrolysate of spruce and an inhibitor cocktail was treated with different bases and reduced to 5.5 with HCl/H_2SO_4 (Persson et al., 2002). The treatments didn't have much effect on glucose and mannose concentration or on acetic or formic acid. Treatment at pH 5.5 didn't have much effect on the toxic concentration while major changes were observed with ammonia treatment to pH 10. Recovery of sulfuric

acid was studied from acid mine drainage by means of 3-compartment electrodialysis (Calatayud et al., 2014), in which abrupt increase in the cell voltage was seen, resulted from the formation of precipitates at surface of the cation-exchange membrane; thereby limiting the process. These reports offer some insights in the use of electrodialysis and the removal of sulfuric acid; though, increases production costs and lack of efficient pathway. Henceforth, preventing scale up of the process and deployment at an industrial level.

2.2 Fabrication of Electrodialyser stack

A classical electrodialysis stack represents a sandwich model of stacks consisting of

- *Ion exchange Membranes*

Ion exchange membranes represents the core of the electrodialysis stack which is used to separate the ions depending on the charge. Cation exchange membranes allow the passage of cations while rejecting anions due to the presence of negatively charged groups, such as $–SO^{3-}$-, $-COO^{-}$-, $–PO_3^{2-}$-, $–PO_3H–$, $–C_6H_4O–$, etc. Whereas, anion exchange membranes allow the passage of anions but reject cations due to the presence of positively charged groups, such as $–NH^{3+}$, $–NRH^{2+}$, $–NR_2H^{+}$, $–NR^{3+}$, $–PR^{3+}$, etc. (Hideo et. al., 1991). Figure 2.1 represents a typical membrane used in the electrodialysis stack.

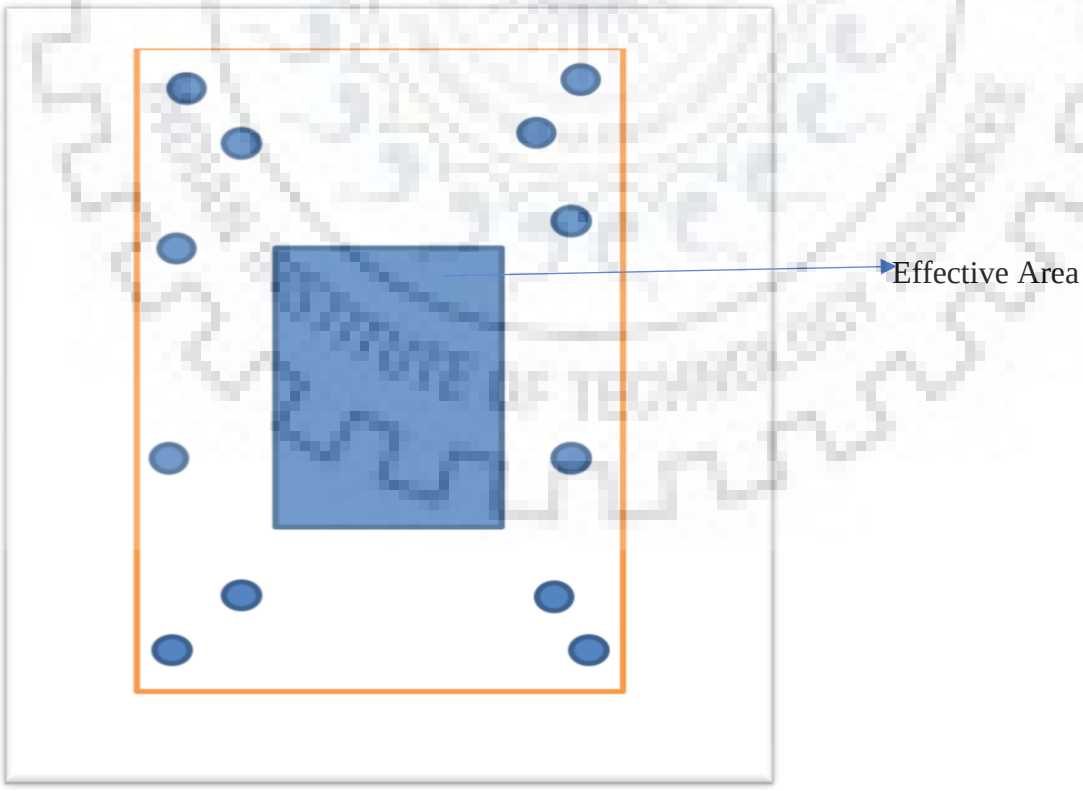


Figure 2.1: The figure repersents a typical membrane in the electrodialysis stack.

❖ *Flow plates*

Figure 2.2 represents a typical flow plate used in the electrodialysis stack. The flow paltes determine the flow pattern of the solution in a zig-zag motion decreaing the size of polarization layer formed at the membrane surface hindering the removal of ions due to incresed coefficients of mass transfer.

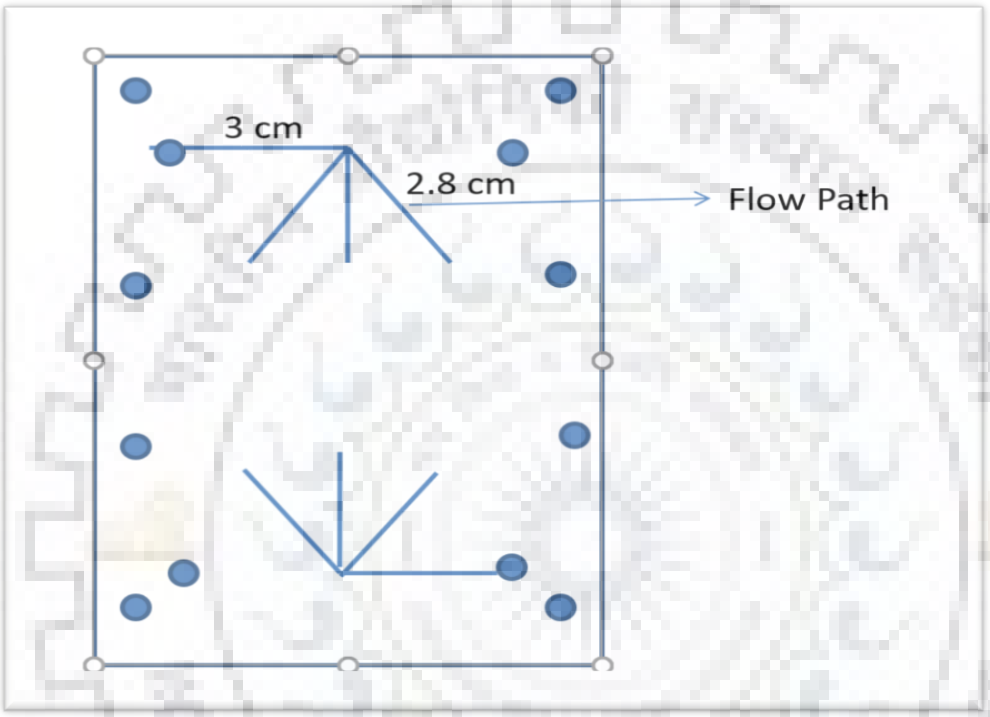


Figure 2.2: A typical flow plate used in the electrodialysis stack.

❖ *Spacer*

Inert spacers separate all the other stacks present in the electrodialysis stack from each other They are used to provide:

- Mechanical support
- Turbulence
- Geometry of flow channel

❖ *Electrodes*

Electrodes are used to provide contact to the solution with the power supply and helps in the passage of electrical current into the system. The mesh type electrodes are most common as they maximize the area from which the electric current passes. The material chosen is mostly inert in relation to the solution to be separated so that it doesn't degrade when in contact with

the solution. Titanium is one of the most preferred material for electrode construction due to its inert nature. Electrode is kept in an inert chamber which contain the ports through which all the solutions enter the electrodialysis chamber for separation further flowing into flow plates. Figure 2.3 represents a typical electrode used in the electrodialysis stack

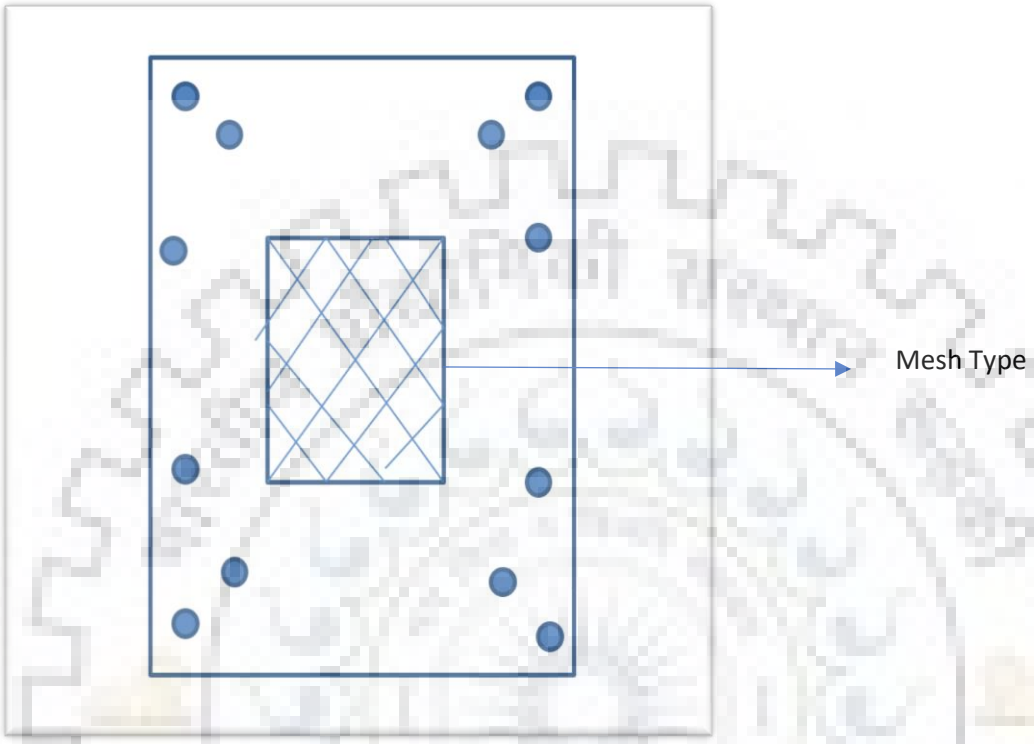


Figure 2.3: The figure represents the typical electrode used in the electrodialysis stack

❖ *D.C Power Supply*

The DC power supply is used to provide current for the ions to break in the electrodialysis chamber as they do not have cycles as AC current have and easy handling.

One anion exchange membrane(AEM) and two cation exchange membranes(CEM) form one membrane pair along with flow plates and spacers stacked in between the electrodes. The electrolyte rinsing solution passes through electrodes in a recirculating manner. DC power supply passes the current through the electrodialysis stack as a driving force to break the ions of the solution starting the process. The passage of solutions in the electrodialysis stack results in two chambers:

- Concentrate compartment (C)
- Diluate compartment (D)

The hydrogen ions move towards the cathode while sulfate ions anions move towards the anode through the membranes.

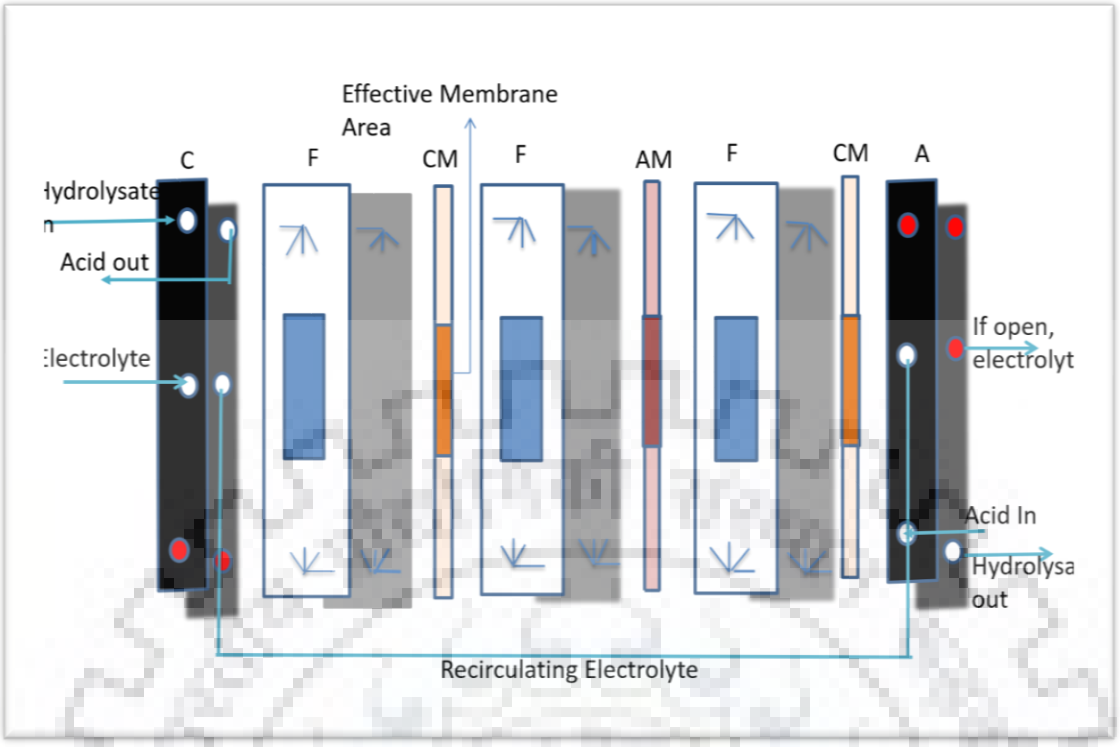


Figure 2.4: The figure represents the schematic diagram of electrodialysis stack.

2.3 Electrodialysis Operation Modes

2.3.1 Potentiostatic vs. galvanostatic mode

There are two prior modes defined for the operation of electrodialysis operation:

- Potentiostatic mode (constant voltage)
- Galvanostatic mode (constant current)

The constant voltage mode is rather chosen above the constant current mode as in constant current mode unrestrained increase in voltage occurs causing breakdown in the current source or even Joule heating causing of the crumbling of electrodialysis stack. While in constant voltage electric conductivity of solution limits increasing the electrical resistance.

2.3.2 Process solutions flow regime

The detrimental formation of gas bubbles occurs due to flow of solution from the electrodialysis stack which either comes out of electrodes or membrane stack resulting in:

(a) electric resistance increments the of the intermembrane space and

(b) reduction of effective surface area of membrane

thus, reduce the performance of the unit. Note, that bubbles can enter unit with process solutions or gas may desorb from the solutions directly in CC or DC.

2.3.3 Batch and Continuous Flow

There are two operating regimes for the flow of solution in the electrodialysis stack:

- batch flow
- continuous flow

The operating mode to be chosen depends on:

- Volume of solution to be filtered
- Application of the Electrodialysis

During the batch flow, the constant amount of solution to be filtered is added and it is recycled to achieve the desired concentration in the final output tank. However, in the concentration procedure follows until the desired concentration is achieved in a single flow

CHAPTER 3

MATERIALS AND METHODS

3.1 Equipment and Reagents

The experiments were conducted using homogenous anion exchange membranes (AEM) and cation exchange membranes (CEM) with polyethylene backbone were procured from Permionics Membranes Pvt. Ltd. (Vadodara, India). The membrane size was 200 mm × 120 mm. The laboratory ED unit was fabricated using transparent acrylic plate purchased from Sushil enterprises (Saharanpur, India). The titanium electrodes (anode and cathode) were procured from Titanium Tantlum Pvt. Ltd., Chennai. The direct current was supplied by a power supply operating at 0-16 volt. Sulfuric acid, glucose, xylose and phenolphthalein were obtained from HiMedia Laboratories (Mumbai, India) and used without any further purification. Ultrapure Type 1 was used throughout the experiments.

3.2 Lignocellulosic Hydrolysate Preparation

The Kansas grass hydrolysate liquor was prepared in the Biochemical Engineering Laboratory, Indian Institute of Technology Roorkee, Roorkee in a laboratory scale fractional hydrolysis column operated at conditions of 100°C, 30 minutes residence time, 10 % (w/w) total solid loading. The synthetic hydrolysate solutions were prepared constituting different concentration of sulfuric acid to conduct the initial set of experiments for optimization.

3.3 Experimental Procedure

3.3.1 Fabrication of Setup

All the experiments were conducted in a laboratory scale parallel planar electrodialyser at room temperature using one cell pair of electrodialysis stack as shown in figure 2.1. Figure 3.2 depicts the conceptual diagram of principle of process proposed through the single cell electrodialyser stack. The cell unit is composed of one anode and one cathode as well as one anion exchange membrane (A) and two cation exchange membrane (C) along with spacers (S). The effective area of electrodialyser cell is represented by the opening at the center of the cell which is 36 cm^2. The membranes were placed at distance of 7 cm. When the hydrolysate liquor

passes through the diluate chamber, sulfuric acid breaks down into H^+ and SO_4^{2-} and ions are transported from the diluate compartment to the concentrate compartment across the ion exchange membranes. Henceforth, sulfuric acid is transported out of the diluate chamber from the hydrolysate liquor and accumulates in the recovery solution. A rectifier is provided to supply a DC power at constant voltage (17 V maximum) or constant current (max. 4 A). As the concentration decreases, electrical conductivity decreases causing an increase in electrical resistance making potentiostatic mode (constant voltage) preferable. Three pumps have capacities of max. 19 L/min to avoid radical concentration changes during the procedure. Three solution tanks (each 2 L) are used for holding the diluted, the concentrated, and the electrode rinse solutions. The filtered hydrolysate and filtered acid samples were taken from separate compartments. Initially, a set of experiments was conducted to determine the limiting current followed by the study of effect of flow rate on the removal of sulfuric acid from the lignocellulosic liquor.

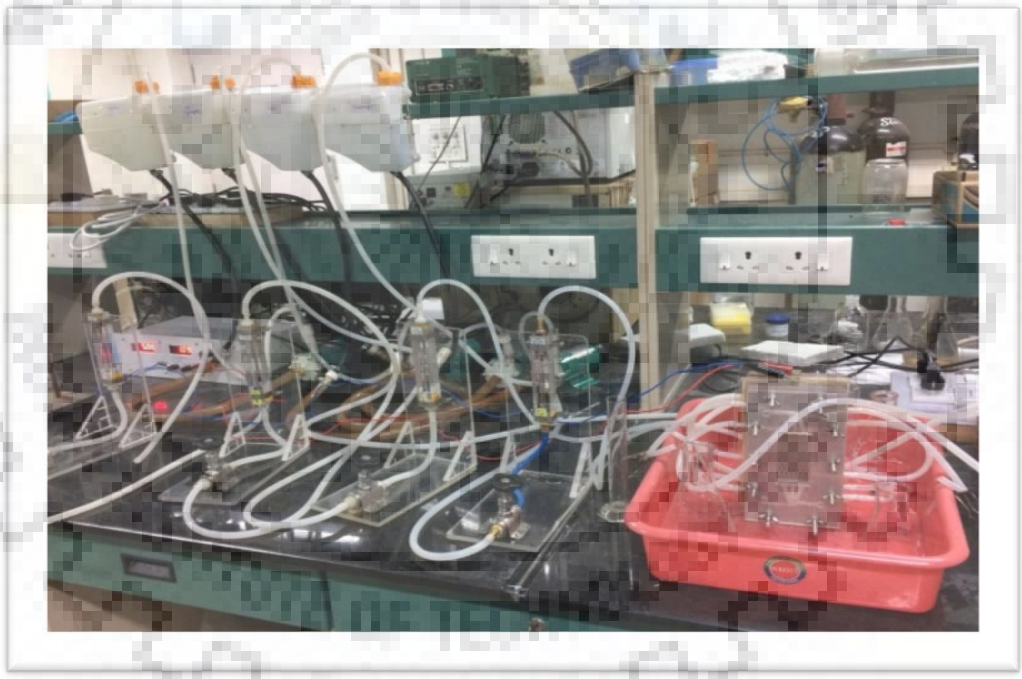


Figure 3.1: Experimental setup depicting the various components of a laboratory scale electrodialyser. Hydrolysate is transferred from the overhead tanks to the ED stack through pump and flowmeter.

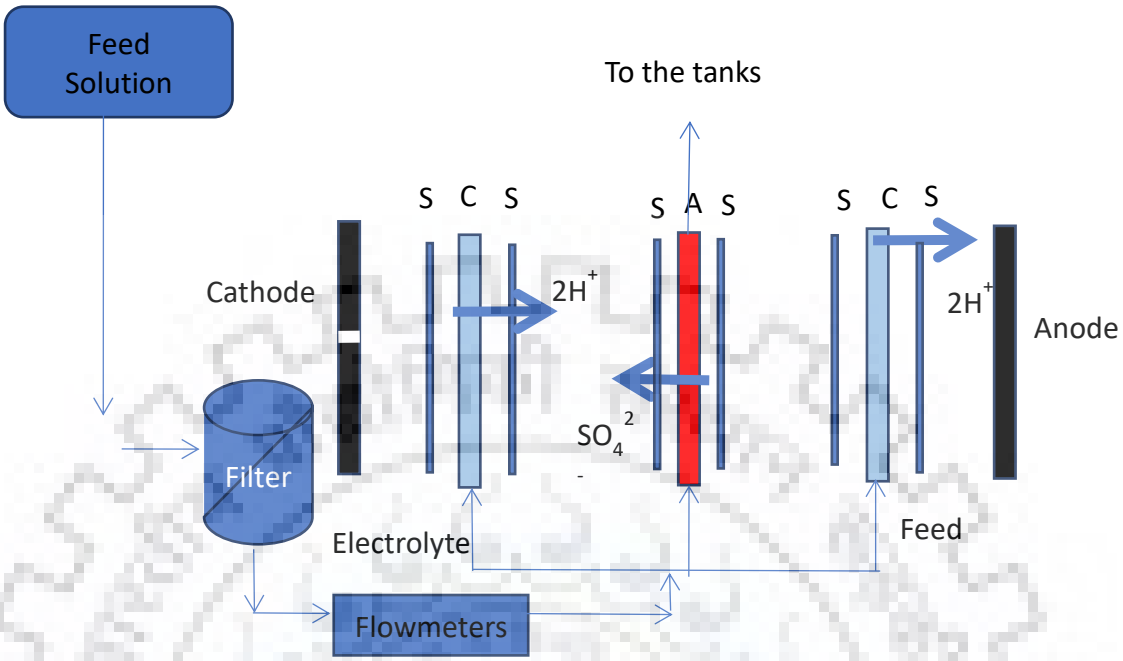


Figure 3.2: Schematic of different components inside a single cell electrodialyser stack. Under the applied electric field, the acid is transferred to the concentrate compartment from the diluate compartment. C, A, S represents cation, anion and spacer respectively.

3.3.2 Limiting current measurement.

Determination of limiting current is highly necessary as it reduces current efficiency of ED as well as efficiency of sulfuric acid separation (J C. Forgacs et. al., 1972). In general, limiting current density could be determined from the following three different combinations of the experimental data (Cowan and Brown, 1959; Rosenberg and Tirrell, 1957):

(1) current and pH value,

(2) resistance and reciprocal of current density, and

(3) current and voltage.

The last type of measurement procedure was employed in this work. Different concentration of sulfuric acid solution was prepared, and voltage was varied keeping the flow rate constant. Finally, the limiting current was found using the method of V-I curve.

3.3.3 Current efficiency calculation.

The current efficiency (CE%) calculation was done from (Yu, Gao, Hao& Jiang, 2000):

3.5 (a) $$CE = \frac{zF\Delta n}{NI\Delta t}$$

Where,

F is Faraday constant,

Δn is the number of moles removed in time t (mol),

N is the number of compartment in ED stack,

I is the electrical current applied (A) and

Δt is the time interval (s).

3.3.5 Effect of flow rate and Separation performance.

The effect of flow rate was first studied in a synthetic hydrolysate mixture (M) with same concentration as in hydrolysate stream. The synthetic hydrolysate liquor contained sulfuric acid (of same molarity as of hydrolysate liquor) and 0.6 g/l of glucose. The membranes were dipped in water followed by dilute acid concentration and used to assemble the electrodialysis chamber. All the operations were conducted in the batch mode by filling 1.5 L of overhead tanks with hydrolysate, reference solution and electrode rinsing solution. The solutions were passed through the pumps and entered the electrodialysis chamber at their respective ports. Flow rate was varied between 3 and 20 mL/min, whereas, the applied electric potential was kept constant at 10 mV. This value of applied current was found using the limiting current principle. The results of the effect of feed flow rate on sulfate ions removal were utilized to optimize separation of sulfuric from the lignocellulosic hydrolysate liquor and can be used for further scaling up. The ED stack was further disassembled and washed by water.

Further, experiments with Kansas grass hydrolysate liquor were conducted in a way like that of the synthetic hydrolysate liquor except the Kansas grass hydrolysate liquor was filtered with muslin cloth followed by vacuum-based membrane filter prior to passing it through electrodialyser. Similarly, the separation performance of sulfuric acid in the Kansas grass hydrolysate liquor was analyzed for the performance evaluation of the process. These experiments were conducted using the Kansas grass hydrolysate liquor prepared in our laboratory

Removal efficiency (RE %) can be calculated using the following equation (Marder et. al., 2016);

3.7 (a). $$RE\% = \frac{C_0 - C_t}{C_0} \times 100$$

Where,

C_o is the initial concentration of sulfate ion expressed in molarity and

C_t is the concentration of sulfate ions at time t.

3.3.6 Sugar Analysis

The amount of glucose and xylose that passes the membrane was analyzed to check the efficiency of the ion exchange membranes. Total reducing sugar in the hydrolysate sample was estimated using DNS method. Further, phloroglucinol test was used to test the xylose and finally the amount of glucose is also determined of the samples both before and after the electrodialysis.

3.3.6 Analytical Methods.

Sulfuric acid was analyzed through titration using 0.1M NaOH solution using phenolphthalein ($C_{20}H_{14}O_4$) as indicator. It turns colorless in acidic solutions and pink in basic solutions. The change in pH values was also measured using pH meter (B. R Biochem Lifesciences, India). Dinitro salicylic (DNS) method was applied to estimate the concentration of reducing sugars in a sample (Miller, 1959). Reducing sugars contain free carbonyl group, have the property to reduce many of the reagents. When alkaline solution of 3,5-dinitrosalicylic acid reacts with reducing sugars it is converted into 3-amino-5-nitrosalicylic acid forming orange color. Further, xylose was estimated colorimetric phloroglucinol test. The pentoses form, by reaction with phloroglucinol in an acid medium, forms a complex that can be determined by spectrophotometry.

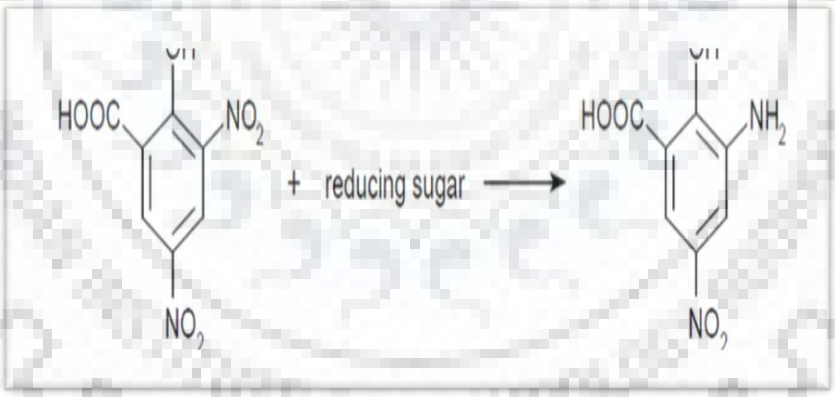


Figure 3.3: The change in 3,5-dinitrosalicylic acid to 3-amino-5-nitrosalicylic after reacting with reducing sugar.

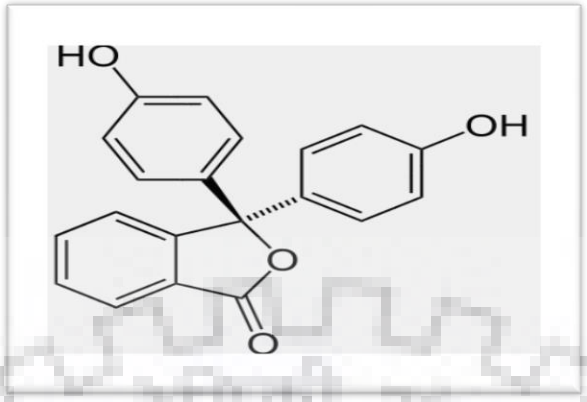


Figure 3.4: The molecular structure of Phenolphthalein (3,3-Bis(4-hydroxyphenyl)-2-benzofuran-1(3*H*)-one)

CHAPTER 4

RESULTS AND DISCUSSIONS

4.1 Preparation of Kansas Grass Hydrolysate

The Kansas grass hydrolysate was prepared in the laboratory in nine different steps with different normality. Table 4.1 indicates the acid concentration used to prepare the Kansas Grass Hydrolysate and the normality at which it is obtained.

Sl. No.	Acid Concentration (%)	Hydrolysate Obtained (N)
1.	1	0.0799
2.	3	0.2167
3.	5	0.4421
4.	10	0.8478
5.	12	1.0307
6.	15	1.4507
7.	20	2.0000
8.	25	2.2823
9.	30	3.1343

Table 4.1: The table indicates the acid concentration used to prepare the Kansas Grass Hydrolysate.

4.2 Effect of limiting current.

The effects of limiting current were studied to access the effectiveness of the process at different sulfuric acid concentration. The limiting current for the synthetic hydrolysate liquor

was first studied and then verified for the Kansas grass hydrolysate liquor, the results are shown in Figure 4.1. It can be seen from the curve that current obviously increases with the increase in the applied voltage in a linear pattern. However, as the voltage reaches around 10 mV, the current becomes stable indicating the limiting current for the sulfuric acid concentration. Also, from Figure 4.2, it can be noted that current increases with the increasing sulfuric acid concentration from 0.02 M to 0.2 M. All experiments were carried out for 30 min. From the above analysis it can be concluded that for the corresponding voltage of 10 V, the limiting current is 1.5 mA at the sulfuric acid concentration of 0.2 M in the Kansas grass hydrolysate liquor.

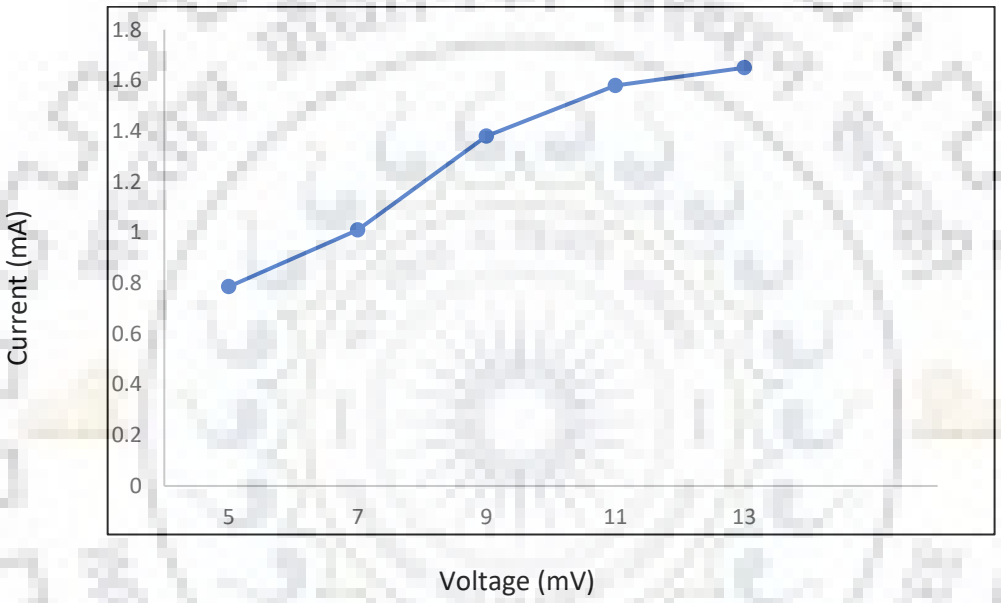


Figure 4.1: The limiting current curve for the Kansas grass hydrolysate liquor at concentration 0.2 M and flow rate 8 ml/min. Each data point indicates the current applied for sulfuric acid at varying voltage.

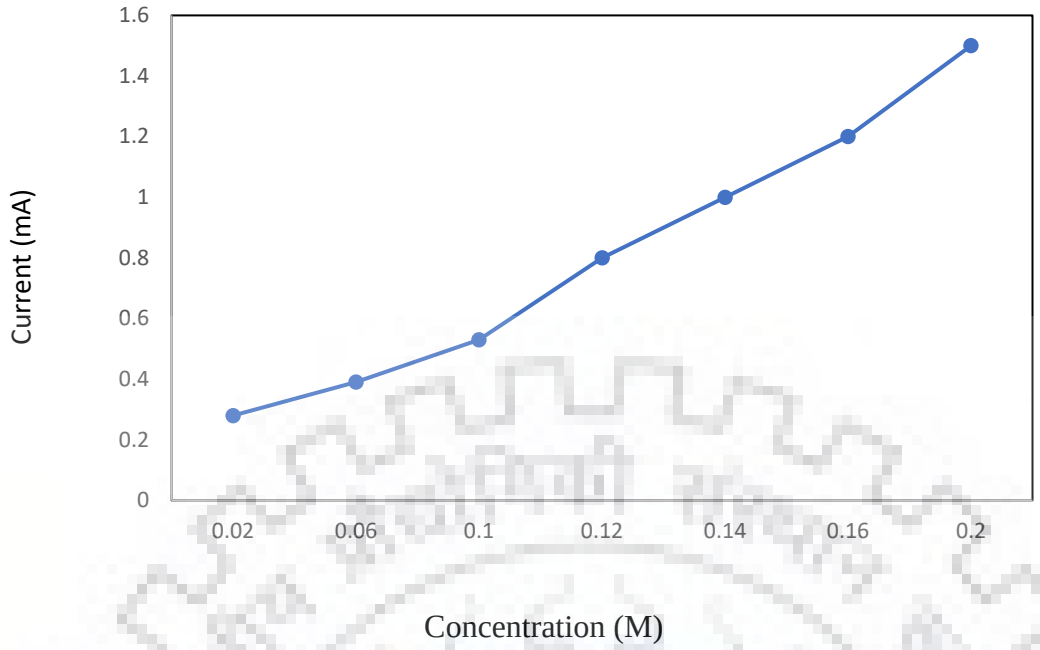


Figure 4.2: The limiting current curve for the synthetic hydrolysate liquor at increasing concentration and flow rate 8 ml/min. Each data point indicates the current applied for sulfuric acid at varying concentration.

4.2 Effect of Flow Rate.

Figure 4.3 illustrates the effect of feed flow rate on the removal of sulfuric acid from the lignocellulosic hydrolysate at applied electric potential of 10 mV and sulfuric acid concentration of 0.2 M across the electrodilayser stack. As the feed flow rate increases, there is a decrease in the residence time, leading to increase in sulfuric acid removal. The higher flow rate leads to better flow distribution increasing the mass transfer for the provided surface area throughout the electrodialyser stack. This advantage of better mass transfer is balanced by the disadvantage of the lower residence time. Henceforth, the total recovery decreases after it reaches the maximum value. Furthermore, Figure 4.4 indicates the effect of feed flow rate on the current for the removal of sulfuric acid from the lignocellulosic hydrolysate at applied electric potential of 10 mV and sulfuric acid concentration of 0.2 M in a single cell pair electrodialysis cell. The current increases from 0.42 mA to 1.6 mA with the increasing value of flowrate from 4ml/min to 16 ml/min due to increased ion transfer.

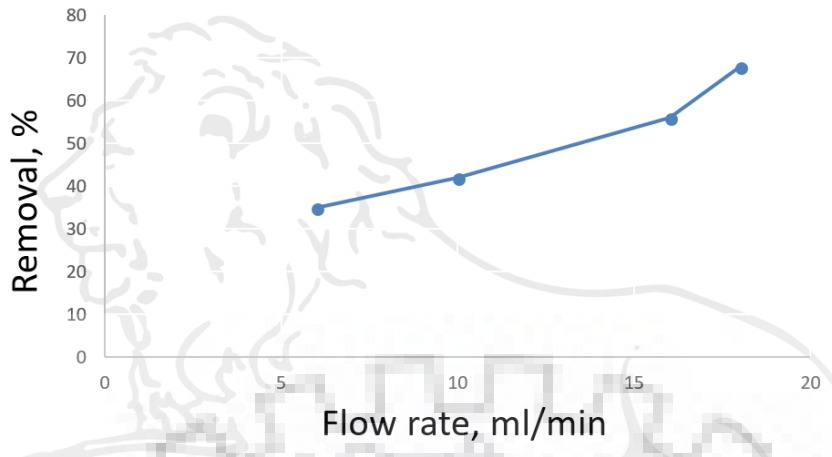


Figure 4.3: The effect of feed flow rate (residence time) on the removal % of sulfuric acid from the lignocellulosic hydrolysate at applied electric potential of 10 mV and sulfuric acid concentration of 0.4 M in a single cell pair electrodialysis cell. Each data point indicates the concentration for sulfuric acid at varying flow rate.

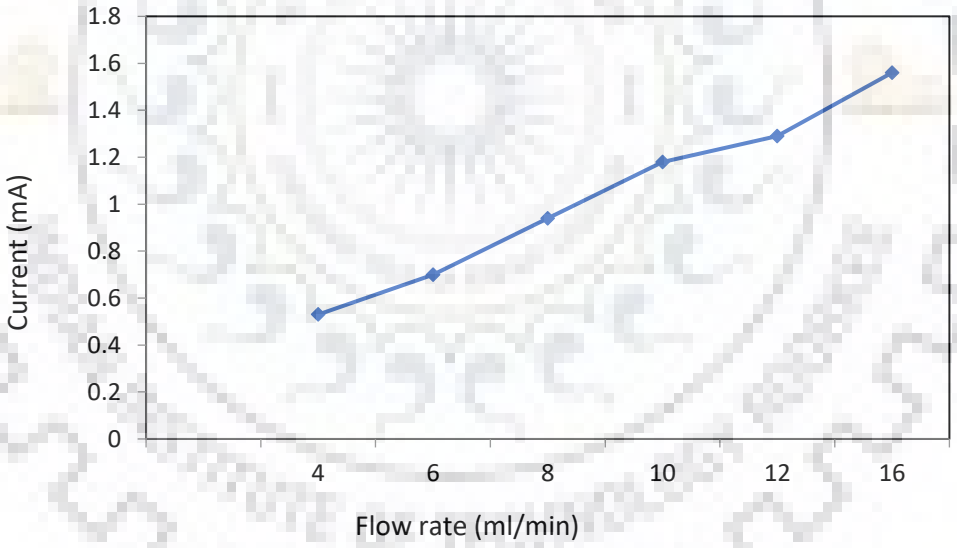


Figure 4.4: The effect of feed flow rate (residence time) on the current for the removal of sulfuric acid from the lignocellulosic hydrolysate at applied electric potential of 10 mV and sulfuric acid concentration of 0.2 M in a single cell pair electrodialysis cell. Each data point indicates the current applied for sulfuric acid at varying flow rate.

4.3 Separation Performance of Synthetic Hydrolysate Liquor

As depicted from the above experiments, the recovery of sulfuric acid from the synthetic hydrolysate liquor was hydrolysate was done at the voltage of 10 mV and flow rate

of 18 mL/min in a single cell electrodialyser stack. To evaluate the performance of the process, synthetic hydrolysate liquor was first used for the experiment. A typical ion removal profile for sulfuric acid is presented in Figure 4.4 that depicts the concentration of sulfuric acid in synthetic hydrolysate liquor as a function of operating time.

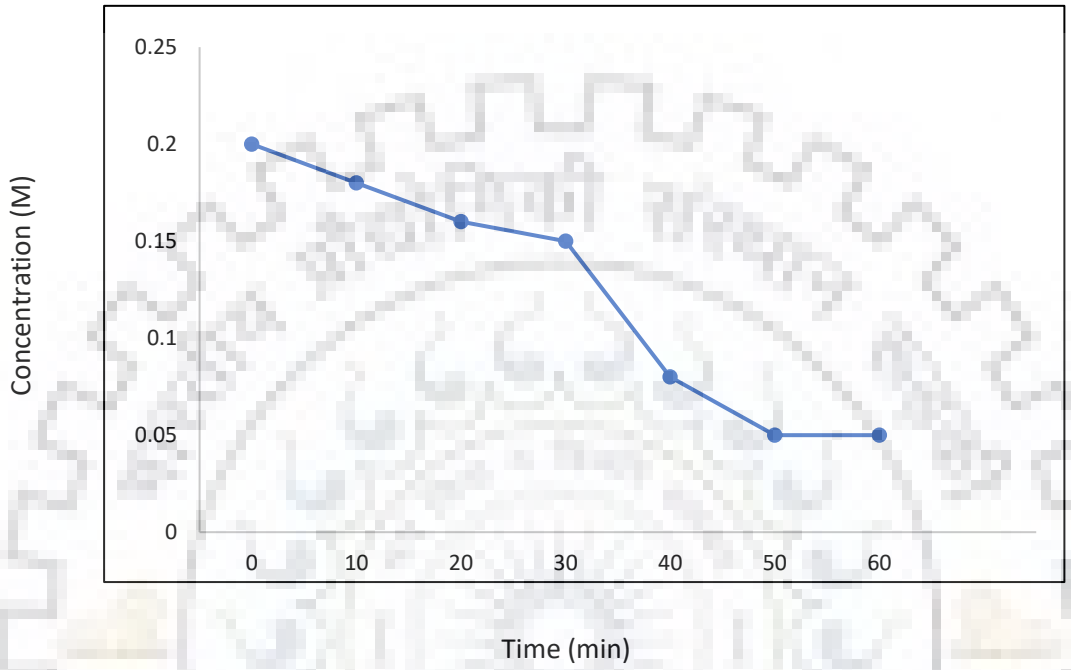


Figure 4.5: The removal of sulfuric acid from synthetic hydrolysate liquor using batch mode ED stack. The operating parameters are maintained to obtain remove of sulfuric acid 60 min of operation time.

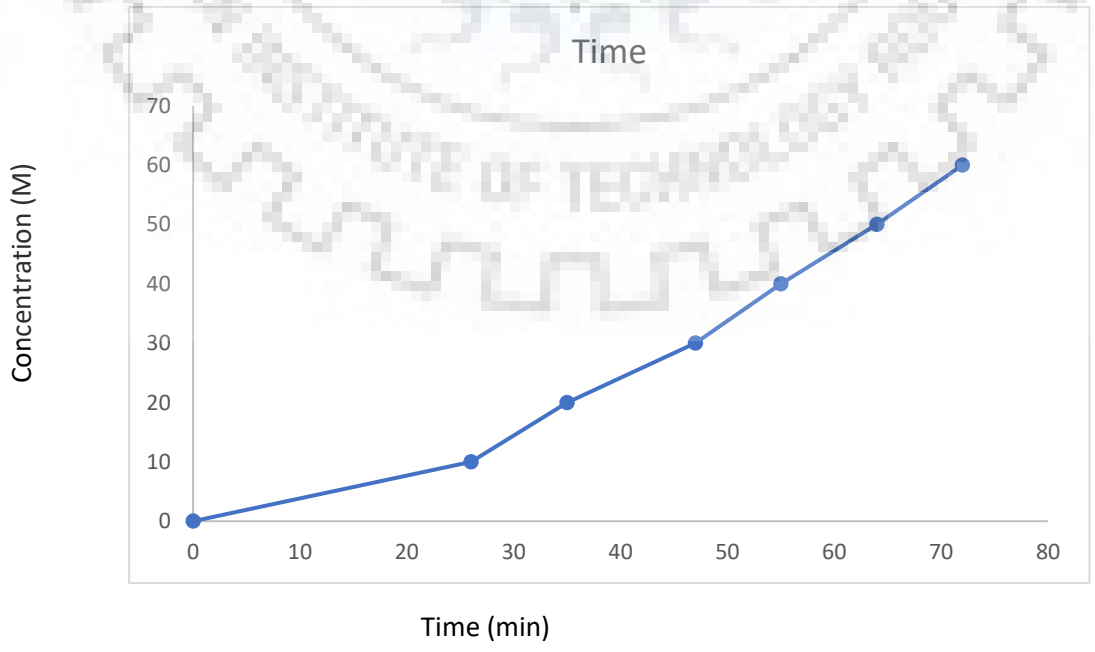


Figure 4.6: Concentration profile of the sulfuric acid in the recovery solution (concentrate solution) during the removal from synthetic hydrolysate liquor using batch mode ED.

4.4 Separation Performance of Kansas grass Hydrolysate Liquor

The same strategy was applied to lignocellulosic hydrolysate liquor. The hydrolysate liquor prepared in the laboratory was obtained with sulfuric acid concentration of 0.2 M and the electrolyte used had the concentration of 0.1 M. Figure 4.4 is a typical time profile of concentration of sulfuric acid in Kansas grass hydrolysate liquor, processed by one cell pair ED stack. After 60 min of operation 68 % of sulfate ions were removed from the hydrolysate. The experiment was paused after 60 min (with 68% sulfate) to replace the sulfuric acid enriched solution from the concentrate side. Three experiments have been conducted with Kansas grass hydrolysate liquor, and all of them demonstrated very similar separation performance. After each experiment, almost 70% of sulfuric acid. The process also demonstrates that more than 98% of total sugar was retained in the hydrolysate liquor at the end of the run. These results suggest that a very minimal amount of total sugars have been partially adsorbed on the diluate compartment. The time profile for sulfuric accumulation in the recovery solution (concentrate) is presented in Figure 4.5.

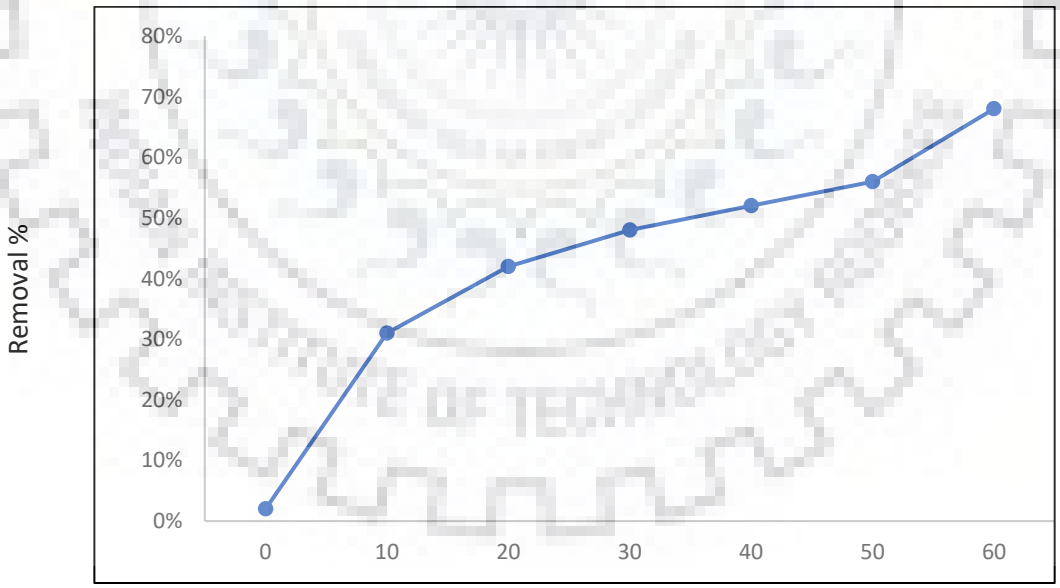
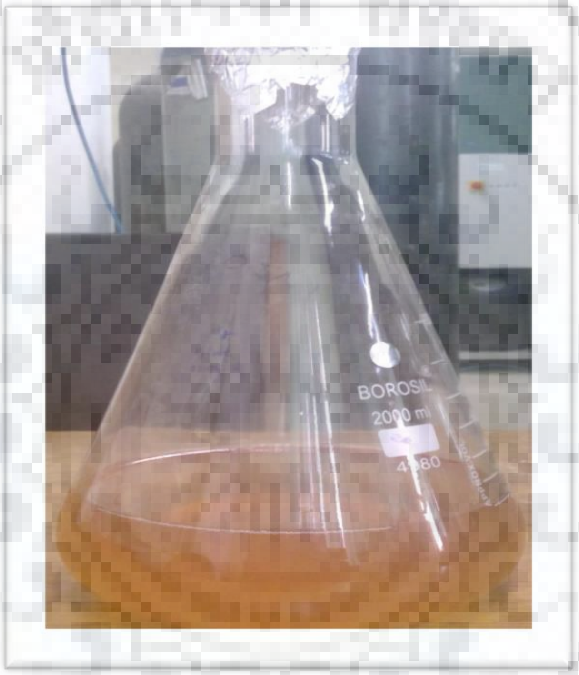


Figure 4.7: Concentration profile of the sulfuric acid in the recovery solution (concentrate solution) during the removal of sulfuric acid from Kansas grass hydrolysate liquor using batch mode electrodialysis.

Removal of sulfuric acid by electrodialysis eliminates the need for over liming, and therefore generation of low value solid waste gypsum. In the over liming process, the residence time for liming, settling of gypsum crystals, and pH adjustment is around 4.5 h (Aden et. al., 2002). Also, in the conventional conditioning process, about 20 wt % additional water is typically added while the liquor gets more concentrated by the process electrodialysis enabling the higher sugar titers. The recovered sulfuric acid enriched stream could be recycled back as-it-is for the dilute acid pre-treatment step, thereby further reducing water demand.



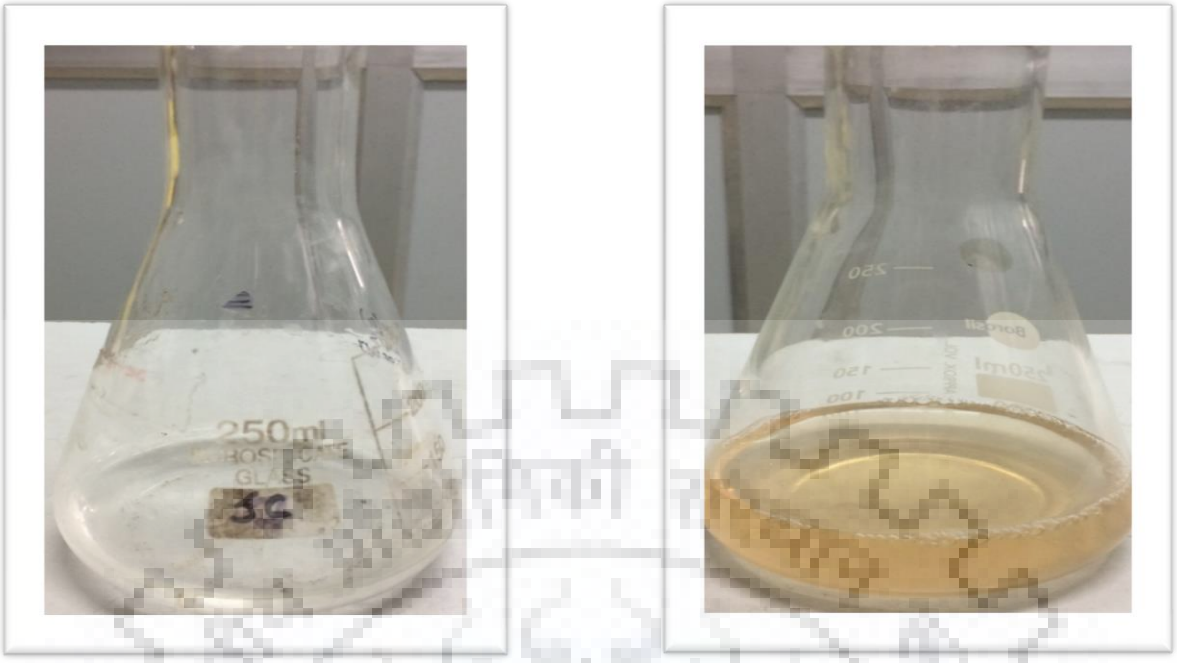


Figure 4.8: The conical flask containing (a) Lignocellulosic hydrolysate liquor, (b) Recovered Sulfuric acid obtained after Electrodialysis and (c) Concentrated Lignocellulosic Hydrolysate Liquor

4.5 Estimation of Reducing Sugars

The presence of reducing sugars (glucose and xylose) in the lignocellulosic hydrolysate liquor was evaluated to analyze the loss of sugars during the process. Glucose enriched solution is mainly obtained in the first four titer of the hydrolysate while xylose is obtained in the last five titers. Table 1 indicates the concentration of total reducing sugars, glucose and xylose in the before electrodialysis procedure while Table 2 contains the concentration of total reducing sugars, glucose and xylose the after the electrodialysis procedure at varying flow rate. This can be seen from the table 4.2 that there is very minimal change in the concentration of glucose and xylose after it passes the ion exchange membranes.

Absorbance

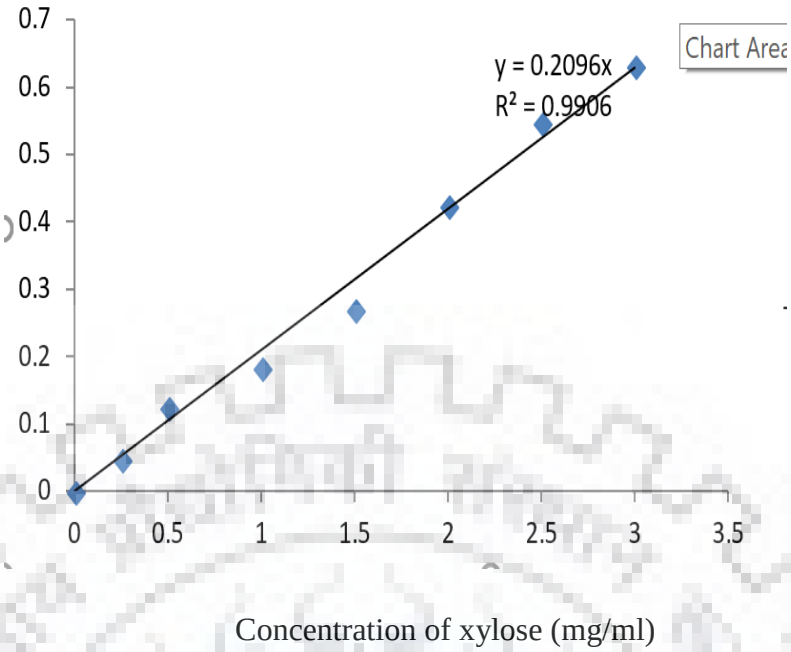


Figure 4.9: Calibration curve used to determine concentration of Glucose

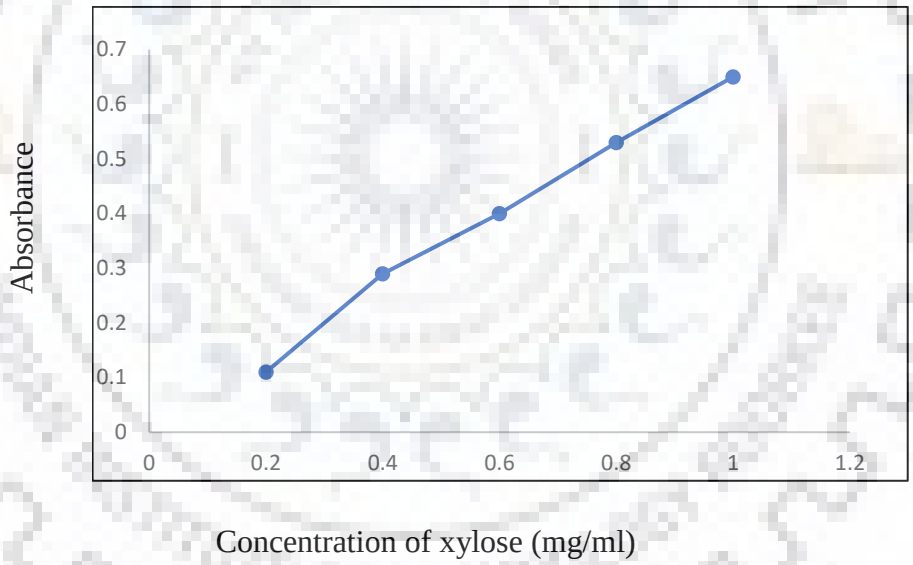


Figure 4.10: Calibration curve used to determine concentration of Xylose

Sl. No.		
1.	Concentration of Total Reducing Sugar (g/l)	10.4 g/l
2.	Concentration of glucose (g/l)	6.8 g/l
3.	Concentration of xylose (g/l)	3.6 g/l
4.	Molarity (M)	0.4 M

Table 4.2 : This table indicates the concentration of total reducing sugars, glucose and xylose before the electrodialysis procedure

SL. No.	Flow rate	Concentration of Total Reducing Sugar (mg/ml)	Concentration of glucose (mg/ml)	Concentration of xylose (g/l)
1	4	10	6.7	3.3
2	6	9.8	6.7	3.1
3	8	9.5	6.6	2.9
4	10	9.6	6.5	3.1
5	12	9.3	6.3	3.0

Table 4.2: This table indicates the concentration of total reducing sugars, glucose and xylose after the electrodialysis

CHAPTER 5

CONCLUDING REMARKS

5.1 Conclusion

Lignocellulosic hydrolysate liquor was deacidified using electrodialysis removing the sulfate and hydrogen ions. About 68 % of sulfuric acid was removed from the lignocellulosic hydrolysate liquor after the process. This proves electrodialysis as an effectual platform for removal of sulfuric acid from hydrolysate liquor and its reuse under the specified conditions. The operating parameters include flow rate and applied voltage, were considered due to major impacts on an ED system. A minimum electricity consumption of 10 mV was used to achieve the mentioned separation based on electrodialysis at 18 ml/min. The sulfuric acid enriched stream contained sulfuric acid 0.2 M. Finally, it can be concluded that these results depict the possibility usage of electrodialysis in removing the sulfuric acid from synthetic as well as lignocellulosic hydrolysate liquors. Electrodialysis if further employed can be to separate inhibitors such as HMF producing cost effective bioethanol.

5.2 Future Scope

The results depict the possibility of the continuous operation for the electrodialysis procedure. This could provide filtered hydrolysate and acid continuously with more efficiency. Even the number of chambers as well as the effective membrane size can be increased to increase the separation performance of the hydrolysate. This also indicate the reuse of sulfuric acid as an important product. This could result in the scaling up of the process.

CHAPTER 6

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