November 2021

Hydrolysis of rice straw for production of soluble sugars

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Hydrolysis of rice straw for production of soluble sugars

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A thesis submitted in partial fulfillment of the requirements for the degree of

Master of Science in Environmental Engineering

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Spring 2013

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To department of Environmental Engineering

April 2013

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Masters of Science in Environmental Engineering.

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بسم الله الرحمـٰن الرحيم

ACKNOWLEDGEMENTS:

I would like to thank my parents and my family for their support throughout my life and the past years. Special thanks to my teachers who guided me and enhanced my knowledge and skills. I am very blessed to have finished this work.

ABSTRACT

Hydrolysis is an effective way of rice straw management and effective treatment to produce soluble sugars. Due to the refractory nature of biodegradation of rice straw in the fields, new higher rate conversion techniques such as hydrolysis are favorable to quickly treat large quantities of rice straw. The hydrolysis process involves chemical pretreatment and thermal treatment of rice straw. Hydrolysis process is capable of reducing rice straw bulk volume by 75% and reducing rice straw mass by 42% using single stage hydrolysis technique. The hydrolysis process is capable of adding value to the raw material rice straw by producing sugars, and producing degraded biomass, and significantly reducing volume. The hydrolysis process was capable of producing at highest 167g of sugars out of 400g of rice straw (41.7%) while consuming 9 KwHr/Kg of energy, and acid consumption of 0.78Kg acid/Kg sugar produced. The degraded biomass is no longer a refractory material to biodegradation due to the thermal treatment and chemical pretreatment. The degraded biomass can be recycled into the hydrolysis process, or used as a raw material to another process.

The hydrolysis process was utilized to test the effect of rice straw pretreatment with 0.5%, 1.0%, 2.0% and 4.25% sulfuric acid and treatment with 3 bar, 4 bar, and 5 bar pressures at different retention times of 30min to 120min to produce soluble sugars. Sugar production was found to be very costly using low concentrations of sulfuric acid due to the high energy consumption during treatment. Sugar production was also found to be costly also with high concentrations of acid due to the high cost of acid per amount of sugars produced. Cost effective sugar production was obtained with 1.0% acid concentration at retention times not exceeding 60min at 5bar and 3bar.

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CHAPTER (1)
INTRODUCTION

Rice straw is one of the abundant lignocellulosic waste materials in the world. In terms of total production, rice is the third most important grain crop in the world behind wheat and corn. As per FAO statistics, world annual rice production in 2007 was about 650 million tons and grew to 696 million tons in 2010 as per latest available statistics. Egypt's share of global rice production is 4.3 million tons in 2010 while the consumption was 4.6 million tons for the same year. Egypt still has the potential to grow production to meet domestic market demand. Every kilogram of grain harvested is accompanied by production of 1 to 1.5 kg of the straw (Maiorella, 1985). It gives an estimation of about 696 to 1044 million tons of rice straw produced globally and a large part of this is going as cattle feed and rest as waste. The options for the disposition of rice straw are limited by the low bulk density, slow degradation in the soil, harboring of rice stem diseases, and high mineral content. Nowadays, field burning is the major practice for removing rice straw, but it increases the air pollution and consequently affects public health (Mussatto and Roberto, 2003). As climate change is extensively recognized as a threat to development, there is growing interest in alternative uses of agro-industrial residues for energy applications. In this context, rice straw would be a potential candidate for our future energy needs. This review aims to give an overview of the available technologies for treatment of rice straw to produce soluble sugars and degraded biomass. The soluble sugars can be utilized for bioethanol production. The degraded biomass can be used in other applications.

Today's agricultural demands will only increase in the future. With this in mind, it is important to recognize that more agricultural products must be made available in the future to accommodate the growing populations of the world.

Egypt's agricultural wastes were reported to be 30 million tons in 2008 growing to be 30.4 million tons in 2010 according to EEAA 2010 annual report. With growing population, this number is expected to grow.

Cellulosic and lignocellulosic materials such as crop residues are the main bulk of agricultural wastes. Natural degradation of these materials takes time and requires a large space and extended periods of time. Driven by the market needs, investments were made by many private and governmental organizations to increase the yield of production from the fields. On the other hand little investments were made to study the effect on

downstream problems of waste generation. Based on environmental needs, investments need to be made to process the huge amounts of wastes into useful products.

One of the most challenging lignocellulosic materials is rice straw due to its slow degradation naturally and high mineral content. Rice stem diseases can spread if natural decomposition in the fields is chosen over the conventional open field burning. The main challenge with natural degradation comes from its high silica and lignin content and its resistance to biological degradation. Dealing with rice straw in the conventional way (burning) has a much larger impact on air quality than any other agricultural waste.

The aim of this study is to produce soluble sugars from rice straw using the hydrolysis process at different conditions.

1.1 Objective Scope of work:

There is a need to stop rice straw open fields burning while providing an alternative to the slow biodegradation. Biodegradation can take many months and pose a health and fire risk. The target is to process rice straw quickly and effectively to produce more useful products. The aim is to process rice straw to produce

1- Soluble sugars (namely a. Glucose, b. Xylose and c. Fructose) and

2- Degraded biomass, through the hydrolysis process.

The process needs to be capable of high rate conversion of rice straw into its processed form of soluble sugars and degraded biomass in the most simple and low tech method. Large quantities are to be converted into useful products with less or no negative environmental impact instead of being burned in the fields and wasted. Such simple low tech process can be of benefit to urban communities, generating job opportunities for farmers and peasants and transportation personnel upstream as well as industrial, mechanical, electrical, chemical and biochemical technicians and engineers downstream.

The financial benefit (resulting from the main two output byproducts 1- Soluble Sugars and 2- degraded Rice straw biomass) is intended to support the activities related to the process under study. The environmental benefits are a bonus to the activities related to the process under study.

The process has input raw materials as

1- Rice Straw,
2- Water and Chemicals in the form of catalysts.

The Process has output products as

1- Soluble fermentable sugars,

2- Degraded Rice Straw Biomass,

3- Water and Chemicals for reuse.

The scope is to use a low technology hydrolysis system and devices to utilize the agricultural waste rice straw as raw material for sugar production. The system should be low tech to suite the local market and socio economic conditions of low budget, low level of education of workers, low periodic maintenance and other local chronic problems. This sugar can then be separated and used to produce other useful products such as ethanol.

CHAPTER (2)
LITERATURE REVIEW

Most literatures discussed the processing of Rice Straw and other agricultural wastes as a source of sustainable biomass for sugar production. Subsequent to sugar production, the most promising product in downstream projects is the production of ethanol as an alternative sustainable fuel to fossil fuels. The focus of this study remains on the production of soluble sugars from rice straw. While literature discussed sugar production as a step in the process, the focus of this study is to produce soluble sugars. This study focuses on pretreatment with dilute sulfuric acid at different concentrations of pretreatment (0.5%, 1.0%, 2.0% and 4.25%) at different treatment pressures of 3bar, 4bar and 5bar at retention times of 30, 60, 90 and 120 minutes.

2.1 Rice Straw structure and breakdown

Rice Straw composition is predominantly cellulose with 32 to 47 %, hemicellulose ranging between 19 to 27% and lignin ranging between 5 to 24%. Pentose sugars (five carbon sugars) are the dominant figure in hemicellulose out of which xylose is the most important sugar as seen in table 2.1. The amount of carbohydrates in rice straw range from 41 to 43.4 % Glucose and 14.8 to 20.2% xylose and 2.7 to 4.5% arabinose and 1.8% manose and 0.4% galactose. (Roberto et al. 2003)

Material	Cellulose	Hemicellulose	Lignin	Glucose	Xylose	Arabinose	Manose	Glactose
% composition	32-47	19-27	5-24	41-43	15-20	3-5	2	0.4

Glucose is most easily converted into ethanol by the action of yeast. Xylose and other sugars are also convertible into ethanol, but with genetic engineering microorganisms to be used for its fermentation process. If rice straw can be utilized to produce soluble sugars then there is potential to produce ethanol from obtained solutions.

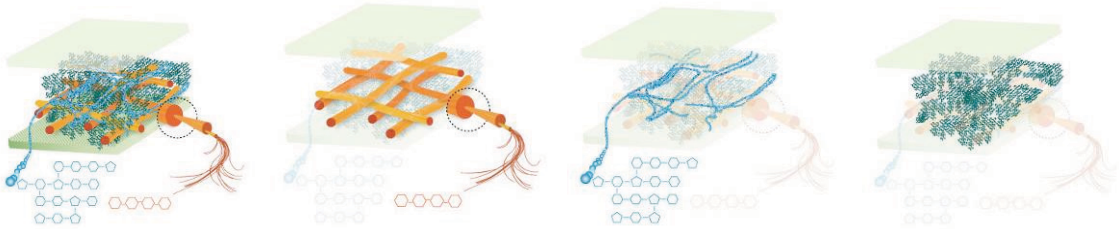
2.1.1. Plant cell wall

The plant cell wall of Rice straw is composed of Cellulose, Hemicelluose, Ligning, and Membrane. The building blocks of cellulose are only hexoses namely glucose. The building blocks for hemicellulose are hexoses and pentoses namely glucose, xylose, mannose, galactose rhamnose and arabinose. The building blocks for lignin are

Figure 2.2 Cellulose in Cell Wall (http://www.ceres.net/images)

Figure 2.1 Plant cell wall and its components, Cellulose, Hemicellulose and Lignin. http://www.ceres.net/images)

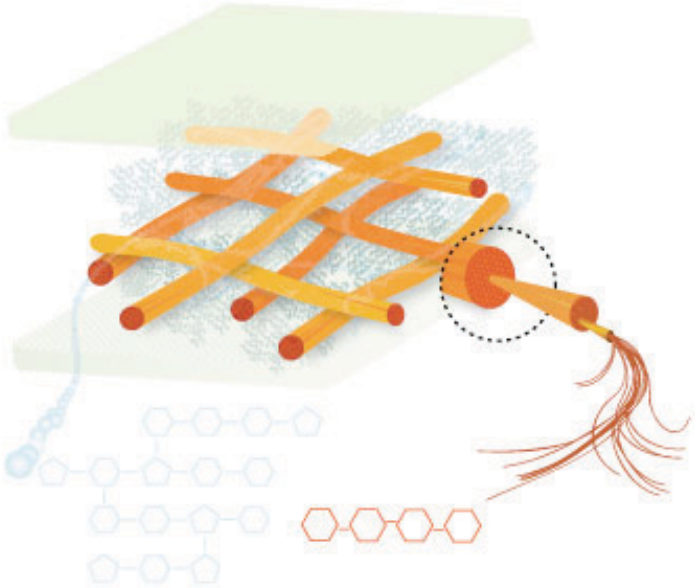
monolignols methoxylate in their three main forms p-coumaryl alcohol, coniferyl alcohol, and sinapyl alcohol (Freudenberg et al 1968).



Complete cell wall Cellulose Hemicellulose Lignin

As shown in Figure 2.1 cellulose is very densely packed and forms well-structured linear fibers while hemicellulose is more random in nature. Lignin on the other hand is a planar sheet like structure. Each morphology is the result of the building blocks and affects the reaction to pretreatment and treatment parameters in later phases.

2.1.2. Cellulose $(C_6H_{10}O_5)_n$

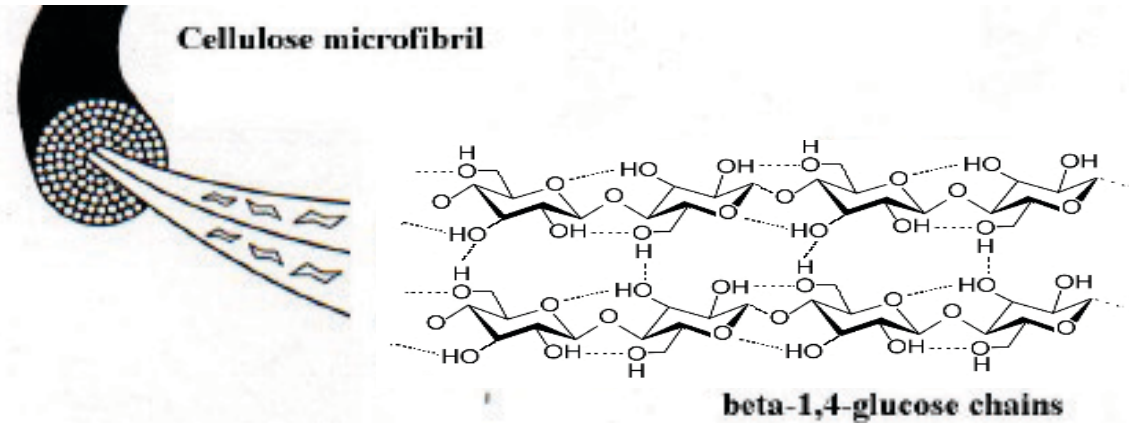


Cellulose is an organic compound with the formula $(C_6H_{10}O_5)_n$ as seen highlighted in orange in Figure 2.2 cellulose is well packed within the cell wall. It is a linear chain polysaccharide. Cellulose is very abundant material, it constitutes about 33% of all

Figure 2.3 Cellulose microfibril, composed of chains of glucose building blocks in β-1-4 glucose chains, with cross linking hydrogen bonds between chains.
(Sun, 2002)

cultivated and wild plant matter put together. For some plant components the cellulose content could be as high as 90% such as cotton fibers. For other hardwoods the cellulose content is 50%. Cellulose is the structural component of the primary cell wall of green plants. (Domalski 1987)

Cellulose is tasteless odorless and hydrophilic. It is not soluble in water and in most organic solvents and it is biodegradable. Cellulose has as its building blocks Glucose units which come together though β(1→4)-glycosidic bonds. This linkage type is what segregates cellulose from starch or glycogen which are α(1→4)-glycosidic bonds. Cellulose's structure is a straight chain polymeric material and no coiling takes place such as that occurring in starch. The many Hydroxyl groups on the six carbon backbone form the main chain undergo hydrogen bonding with oxygen molecules from the nearby chains. This inter-chain hydrogen bonding holds the adjacent chains together more firmly building microfibrils, as seen in figure 2.3, with high tensile strength and promotes crystallinity. Hydrogen bonding and high density microfibril packing requires high temperatures and high pressure (320°C, 25 bar) to reach the amorphous phase in water. Glucose as the main building block of cellulose, it is not easily broken down from the stiff matrix. This is due to the strong bonds between glucose molecules forming long chains, and also to the multiple hydrogen bonds cross linking these strong chains (Deguchi et al. 2006)



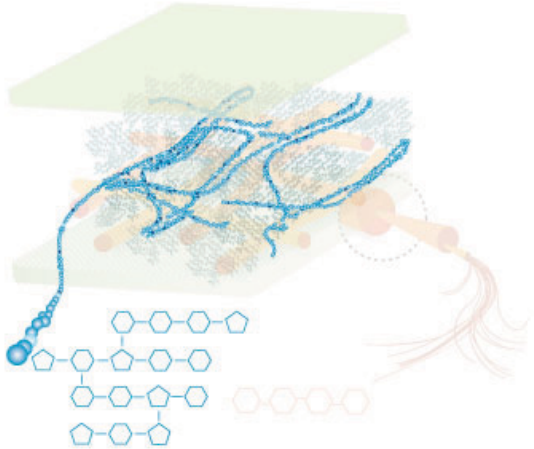
The dependent properties of cellulose are caused by the degree of polymerization due to chain length. This is summed up by the number of glucose building blocks that make up one polymer molecule. Wood pulp cellulose has chain lengths between 300 to 1700

glucose units. Cotton and other plant fibers have chain lengths between 800 and 104 glucose units. Molecules of very small chain length of cellulose are also known as cellodextrins which in contrast with long chains, are soluble in water and organic solvents. There are many solvents that dissolve cellulose and generally can degrade it in the process. (Klemm et al. 1998).

The degradation of cellulose does not yield glucose directly. Cellulose is broken down to glucan and oligosccahrides prior to glucose as follows: Cellulose → Glucan → Oligosaccharides → Glucose → HMF → Levulinic acid. The formation of HMF and Levulinic acid is not favorable because they act as an inhibitor to fermentation in later stages. (Karimi et al. 2006)

Figure 2.4 Hemicellulose in Cell Wall (http://www.ceres.net/images)

2.1.3. Hemicellulose:



Hemicellulose is composed of several heteropolymer as a matrix of polysaccharides. It is present in the cell walls of almost all plant cell walls alongside cellulose. Unlike cellulose's crystallinity and strength and resistance to hydrolysis, hemicellulose has a random and amorphous structure, as seen in blue in figure 2.4, which is one of the reasons why it is found lacking in strength comparing to cellulose. Hemicellulose is more readily hydrolyzed by acid or base or enzymes compared to cellulose. The constituents of hemicellusoe are mostly pentoses such as xylose, mannose, galactose rhamnose and arabinose. Xylose is the sugar peresent in largest amounts.

2.1.4. Lignin:

Lignin is what fills the spaces between the components of the plant structure such as Cellulose, Hemicellulose and Pectin. Lignin is covalently bonded to Hemicellulose and cross linking occurs with other polysaccharides. This cross linking transfers mechanical strength to cell walls and the plant as a whole. (Chabannes et al 2001)

Lignin is a cross-linked macromolecule spreading as a sheet within the cell wall as

igure 2.5 Lignin in Cell Wall
ttp://www.cerespot/images) 1.5 with molecular mass exceeding 104u. It is hydrophobic to an extent and aromatic in nature. The degree to which polymerization occurs in natural fibers is difficult to measure due to fragmentation during extraction processes and the presence of various types of substructures repeating randomly as shown in figure 2.6 with at least three types of cross linked monomer building blocks.

Figure 2.6 Lignin structure and its building blocks.

There are three known monomers known as monolignols methoxylated as seen in figure 2.6, p-coumaryl alcohol, coniferyl alcohol, and sinapyl alcohol (Freudenberg et al 1968)

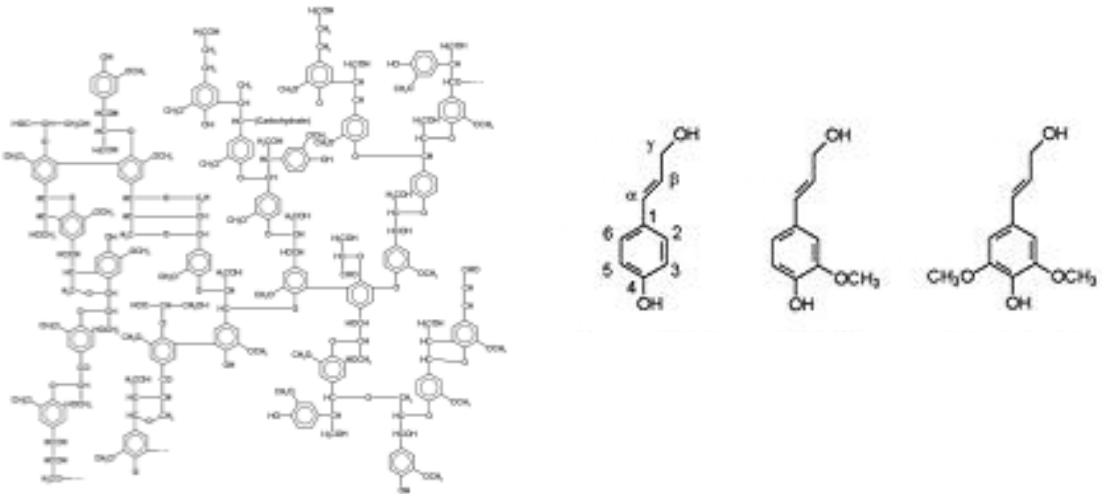
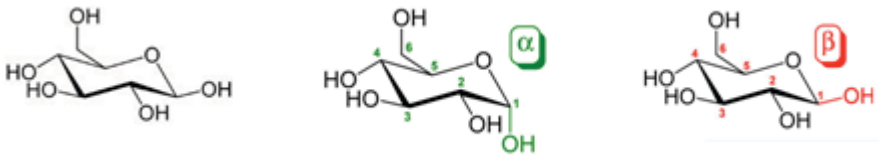


Figure 2.7 Cyclic glucose in α and β forms

These lignols are incorporated into lignin in the form of the phenylpropanoids p-hydroxyphenyl, guaiacyl, and syringal respectively (Boerjan et al. 2003)

Adding confusion and uncertainty regarding Lignin, it was found that all lignins contain little amounts of incomplete monolignols. Other monomers are also present in non-woody plants such as rice straw. More studies are needed in this area. (Ralph et al. 2001)

2.1.5. Glucose ($C_6H_{12}O_6$)



There are two forms that glucose can exist as in solution, namely cyclic (ring) and acyclic (open chain). The cyclic form is favored in equilibrium conditions. It is favored in cellulose as its building blocks. The cyclic form can exist as α or β forms as shown in figure 2.7. The α form (position of the OH group) can be processed by human enzymes breaking down the bonds between glucose molecules. The β form cannot be processed by human enzymes, they require specialized bacteria to process these bonds such as cattle's intestinal cultures.

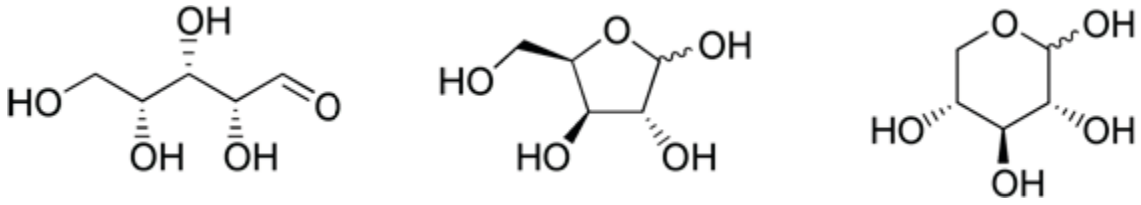
The α and β forms interchange with time in aqueous solutions to reach equilibrium conditions of α to β 36 to 64 %. This process called mutarotation. (McMurry 1998)

Yeast such as *Saccharomyces cerevisiae* can metabolize glucose in the absence of oxygen to produce ethanol and carbon dioxide. In the presence of oxygen the same yeast will produce carbon dioxide and water. The biochemical reaction is as follows converting glucose to ethanol. $C_6H_{12}O_6$ + yeast → 2 CH_3CH_2OH + 2 CO_2. It is important to note that the yeast is sensitive to fermentation by-product ethanol. Even the most resistant strains of yeast will not survive in ethanol concentrations more than 15% by volume (Morais et al. 1996).

Figure 2.9 Fructose molecules, Cyclic and open chain

2.1.6 Xylose ($C_5H_{10}O_5$)

Figure 2.8 Xylose molecules, cyclic and open chain



Xylose is a monosaccharide pentose usually called wood sugar. It is better known for its alcohol form (xyilito $C_5H_{12}O_5$). shown in figure 2.8 Xylose is one of the building blocks of hemicellulose. "The hydrolysis of hemicellulose may lead first to the monomeric sugar:

Hemicellulose → Oligosaccharides → Sugars

(xylose; arabinose; glucose; mannose; galactose)

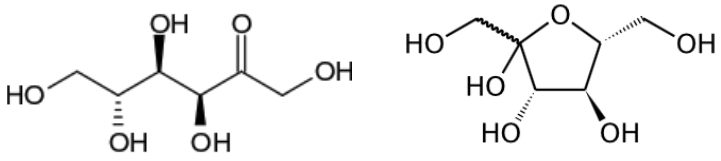
These reactions may further continue to some other by products:

Pentoses → Furfural → Furfural resinification and condensation products;

Hexoses → HMF →Levulinic acid."(Karimi et al. 2006)

Xylose can also be converted into ethanol by yeast fermentation. Yeasts such as *Pichia stipitis* produces ethanol as a by product of xylose catabolism. *Pichia stipitis* is very sensitive to ethanol, other types have been genetically altered to produce ethanol. This way the yeast remains unaffected by ethanol's presence in the solution. One such example is Saccharomyces cerevisiae which successfully expresses the XYL1 and XYL2 genes needed for the breakdown of xylose sugar. (Eliasson et al., 2000).

2.1.7. Fructose ($C_6H_{12}O_6$)

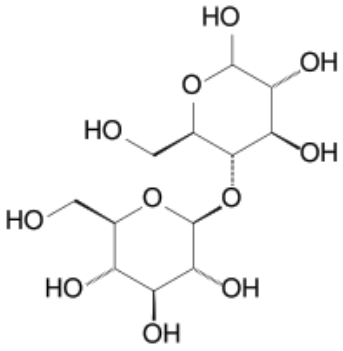


Fructose is a monosaccharide hexose and is the most water soluble of all the sugars (Hyvonen et al. 1982). There are many forms of Fructose that can exist in a solution which are cyclic and open chain shown in figure 2.9, such as D Fructose and L Fructose.

Figure 2.10 Cellobiose molecule

It is one of the three monosaccharaides that are absorbed directly into the bloodstream during the digestion process. In plants it is found as part of the disaccharide sucrose which is made up of one Fructose molecule and one Glucose molecule. Most importantly, Fructose is fermentable by the action of yeast and bacterial to produce ethanol.

2.1.8. Cellobiose ($C_{12}H_{22}O_{11}$)



Cellobiose is a disaccharide with the chemical formula $[HOCH_2CHO(CHOH)_3]_2O$ which comes as the result of the condensation of two glucose molecules joint in a $\beta(1\rightarrow4)$ bond (shown in figure 2.10) in the presence of acidic media. Cellobiose can be hydrolyzed into two glucose molecules by the action of bacteria or the action of cationic ion exchange resins

2.1.9 Hydrolysis

Hydrolysis is the method understudy to produce sugars from Rice straw. Other biological and enzymatic techniques are available but Hydrolysis is a quicker process. Hydrolysis involves blasting the Rice straw with high pressure, high temperature saturated steam. The Rice straw needs to be pretreated with acidic or basic media to facilitate the degradation process. The result from hydrolysis is reduction of the rice straw bulk by 42% by weight and 75% by volume. The recovery of glucose and xylose is achievable via downstream separation techniques. The production of high quality compost is more readily achievable with the remaining 58% of the rice straw as a result of the chemical and thermal treatments.

The aim of this study is to replicate the process of hydrolysis using local rice straw in order to study the amount of soluble sugars generated by the process.

When hydrolysis is performed on Rice Straw the material's chemical structure and chemical bonds are attacked with the acid and the steam breaking them up into their building blocks glucose, xylose, arabinose, manose, glactose, etc. The main interest is in glucose and xylose.

Glucose could originate from either the Hemicellulose or cellulose fractions of Lignocellulose. The glucose liberated at mild hydrolysis conditions most likely originated from Hemicellulose

Hemicellulose, being more readily susceptible to hydrolysis due to its structure and weaker bonds, breaks down in serial:

Hemicellulose → Oligosaccharides → Sugars (xylose; arabinose; glucose; mannose; galactose

Cellulose on the other hand is well packed and stronger bonds hold the cellulose chains together. This requires secondary treatment because single stage treatment with acid and steam is not sufficient to break down the bonds. Nevertheless if too much steam and acid are used then the cellulose will be broken down and also the building blocks themselves (in this case glucose only) will be broken down as well resulting in HMF and Levulinic acid by products which are not favorable products.

Cellulose (Glucan(→ Oligosaccharides → Glucose → HMF → Levulinic acid.
(Karimi et al. 2000)

In general, both Pentoses and Hexoses are susceptible to breakdown at high pressure and temperature.
Pentoses → Furfural → Furfural resinification condensation and other products
Hexoses → HMF → Levulinic acid.

Avoiding monosaccharides' degradation is key to improve the yield of hydrolysis, and to avoid the problems with the inhibition of fermentation of sugars to, ethanol and / or xylitol in downstream processes. (Sanchez G. et.al 2004, Converti A. et.al 2000)
In general, mild pressure and temperature are more favorable for downstream processes because the yields of Furfural from Pentoses and Levulinic acid from Hexoses are

significantly less at mild hydrolysis conditions. The results of Keikhosro Karimi show that the yield of HMF gradually increases when the hydrolysis pressure is increased, regardless of the type of hydrolysis and stage number. (Karimi et al. 2006)

2.1.10 Caramelization:

Sugars react to temperature same as all other matter. With sugars the case if slightly different due to the caramelization phenomenon. Sugars caramelize when they are slowly heated above their melting temperatures. The sugar molecules break down and reform into products that are similar to caramel color (light brown) and smell (full of aroma). Glucose's melting temperature is between 146 and 150 degrees Celsius. Xylose's melting temperature is 144 and 145 degrees Celsius and their caramelization temperature is arround 160 degrees Celsius. Caramelization reactions are also very sensitive in highly reactive chemical environment where the pH is too aggressive, best conditions is neutral pH to avoid excessive caramelization (Vilamiel et al. 2006).

According to steam tables the corresponding steam pressures in the range from 144 to 150 degrees Celsius is 4.1 to 4.8 bar. It is recommended not to exceed 5bar during the Hydrolysis treatment period to avoid such unfavorable by-products which will break down and reform into more complex compounds or residues. It is more suitable to control temperature than pressure in this case because pressure inside a constant volume vessel will increase while injecting steam. As steam is cooled down it condenses into hot water which will increase the pressure inside the vessel without direct contribution to temperature.

2.1.11. Fermentation:

Fermentation is a naturally occurring metabolic process capable of converting sugars to alcohol and carbon dioxide. This process has long been used to produce wine, champagne, beer and other alcoholic drinks. The fermentation process occurs as a byproduct to the action of yeast or bacteria growing and living in the sugar medium. As with all by products of living microorganisms, if the by product increases to levels that are toxic, the organisms die and the process stops.

There are many types of microorganisms that can ferment different types of sugars. For example: Xylose is a five-carbon sugar that can be metabolized into ethanol by a yeast called *Pichia stipitis*. This yeast can metabolize Xylose to produce ethanol due to

Table 2.2 Rice Straw composition and theoretical ethanol yield from available carbohydrate. (Binod et al.2010)

the presence of the XYL1 and XYL2 genes in its DNA that are necessary for enzyme production to breakdown the xylose sugar. Drawbacks are related to *Pichia Stipitis* being sensitive to ethanol and the yeast cells die if accumulation occurs. Therefore a process is needed to remove the by product ethanol at the same rate of its production to save the yeast.

Genetic engineering of yeast provided another strain that is resistant to ethanol. This yeast is called *Saccharomyces cerevisiae* also expresses the XYL1 and XYL2 genes. This strain can allow for effective production of ethanol with significantly reducing the risk of the culture being affected. (Eliasson et al. 2000).

2.2. Sugar production from rice straw instead of food crops

The production of soluble sugars from food crops such as grains has resulted in an undesirable direct competition with food supply. Soluble sugar production from rice straw biomass has become an increasingly more economical alternative to soluble sugar production from food crops. The use of the produced soluble sugars from food crops was first generation biofuels production as gasoline additives or substitute. A switch to a more abundant inedible plant material such as rice straw will help to reduce pressure on the food crops. Large parts of rice straw plant materials are made up of complex carbohydrates such as cellulose and hemicelluloses which can be converted to soluble sugars. Ethanol fermenting microorganisms can utilize these sugars and convert into ethanol. Rice straw has several characteristics that make it a potential feedstock for fuel ethanol production. It has high cellulose and hemicelluloses content that can be readily hydrolyzed into fermentable sugars. In terms of chemical composition, the straw predominantly contains cellulose (32–47%), hemicellulose (19–27%) and lignin (5–24%) (Garrote et al., 2002; Maiorella, 1983; Saha, 2003; Zamora and Crispin, 1995). The pentose sugars are dominant in hemicellulose, in which xylose is the most important sugar (14.8–20.2%) (Maiorella, 1983; Roberto et al., 2003). The carbohydrate composition and theoretical ethanol yields of rice straw is shown in Table 2.2, theoretical ethanol yield is 0.42L of ethanol from 1Kg of dry rice straw.

Cellulose	38.60%
Hemicellulose	19.70%
Theoretical ethanol yield (L/Kg dry)	0.42

Table 2.3 Proximate composition and selected major elements of ash of rice straw, rice husk and wheat straw. (Jenkins et al. 1998)

Table 2.4 Worldwide quantities of rice straw available and theoretical ethanol yield. (Kim and Dale 2004).

The chemical composition of feedstock has a major influence on the efficiency of bioenergy generation. Table 2.3 lists the chemical properties of rice straw, rice husk, and wheat straw to highlight the particular differences in feedstock. The low feedstock quality of rice straw is primarily determined by a high ash content (10– 17%) as compared to wheat straw (around 3%) and also high silica content in ash (SiO_2 is 75% in rice and 55% in wheat). On the other hand, rice straw as feedstock has the advantage of having a relatively low total alkali content (Na_2O and K_2O typically comprise <15% of total ash), whereas wheat straw can typically have >25% alkali content in ash.

	Rice Straw	Rice Husk	Wheat Straw
Proximate analysis (%dry fuel)			
Fixed carbon	15.86	16.22	17.71
Volatile matter	65.47	63.52	75.27
Ash	18.67	20.26	7.02
Elemental Composistion of ash (%)			
SiO_2	74.67	91.42	55.32
CaO	3.01	3.21	6.14
MgO	1.75	0.01	1.06
Na_2O	0.96	0.21	1.71
K_2O	12.3	3.71	25.6

Straw quality varies substantially within seasons as well as within regions. If straw is exposed to precipitation in the field, alkali and alkaline compounds are leached, improving the feedstock quality. Thus, the preferred use of this material for bioethanol production is related to both quality and availability.

2.3. Availability of Rice Straw

Rice straw is one of the highly abundant lignocellulosic crop residues in the world. Its annual production is about 730 million tons (696–1044 range) distributed in Africa, Asia, Europe and America as shown in Table 2.4.

	Rice Straw Availability (Million MT)	Theoretical ethanol yield (Billion Liters)
Africa	20.93	8.83
Asia	667.59	281.72
Europe	3.92	1.65
North America	10.95	4.62
Central America	2.77	1.17
South America	23.51	9.92

This amount of rice straw can potentially produce 308 billion liters bioethanol per year. In Asia it is a major field-based residue that is produced in large amounts (667.59 million tons).

The total amount equaling 730 million MT could produce theoretically 308 billion liters of ethanol if the technology were available. However, an increasing proportion of this rice straw undergoes field burning. This waste of energy seems inapt, given the high fuel prices and the great demand for reducing greenhouse gas emissions as well as air pollution (Kim and Dale, 2004). There are primarily two types of residues such as straw and husk from rice cultivation that have potential in terms of sugar production for potential energy production. Although the technology of using rice husk is well established in many Asian countries, rice straw is rarely used as a source of renewable energy. One of the principal reasons for the preferred use of husk is its easy procurement as it is available at the rice mill. But the collection of rice straw is laborious and its availability is limited to harvest time. The logistics of collection could be improved through baling, but the high cost of equipment makes it uneconomical for most of the rice farmers. Thus, the technologies to use rice straw for the energy purpose must be especially efficient to compensate for the high costs involved in straw collection.

2.4 Production of Sugars From Rice Straw

2.4.1. Basic concept

Rice straw consists of three main components, cellulose, hemicellulose and lignin. Technologies for conversion of this feedstock to potential ethanol production have been developed on two platforms, which can be referred to as the sugar platform and the synthesis gas (or syngas) platform. The aim of this study is to focus on the sugar platform to produce soluble sugars that can be used by a downstream project. The basic steps of these platforms are shown in figure 2.11. In sugar platform, cellulose and hemicellulose are first converted to soluble sugars. These soluble sugars can then be fermented in a later process to produce ethanol. The focus of this study remains on the sugar production portion of the sugars platform.

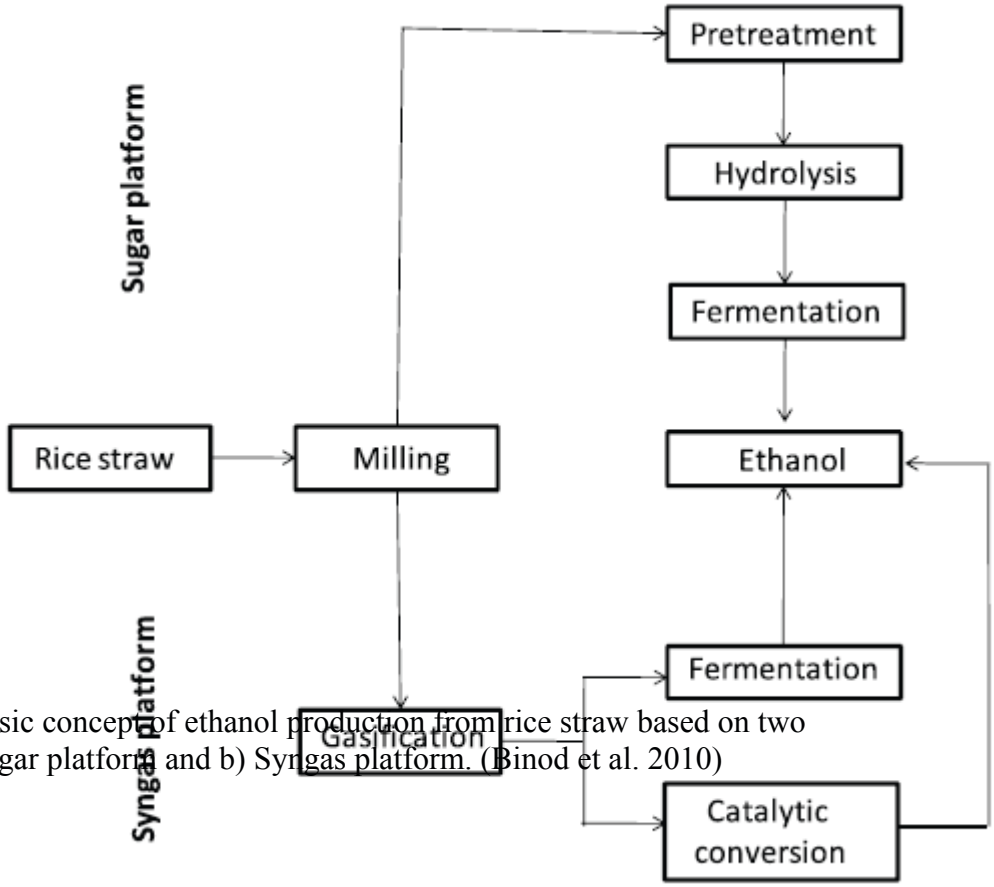


Figure 2.11 Basic concept of ethanol production from rice straw based on two platforms a) Sugar platform and b) Syngas platform. (Binod et al. 2010)

The fermentable sugars include glucose, xylose, arabinose, galactose, and mannose. Hydrolysis of cellulose and hemicellulose to generate these sugars can be carried out by using either acids or enzymes (Drapcho et al., 2008). In the syngas platform, the biomass is subjected through a process called gasification. In this process, the biomass is heated with no oxygen or only about one-third the oxygen normally required for complete combustion. It subsequently converts to a gaseous product, which contains mostly carbon monoxide and hydrogen. The gas, which is called synthesis gas or syngas, can be fermented by specific microorganisms or converted catalytically to ethanol. In the sugar platform, only the carbohydrate fractions are utilized for ethanol production, whereas in the syngas platform, all three components of the biomass are converted to ethanol (Drapcho et al., 2008).

2.4.2. Importance of pretreatment

Rice straw is composed of heterogeneous complex of carbohydrate polymers. Cellulose and hemicellulose are densely packed by layers of lignin, which protect them

against enzymatic hydrolysis. So it is necessary to have a pretreatment step to break lignin seal to expose cellulose and hemicellulose for enzymatic action. Pretreatment aims to decrease crystallinity of cellulose, increase biomass surface area, remove hemicellulose, and break lignin seal. It makes cellulose more accessible to enzymes so that conversion of carbohydrate polymers into fermentable sugars can be achieved more rapidly and with more yields. Pretreatment includes physical, chemical and biological methods and their combinations. It has been viewed as one of the most expensive processing steps in cellulosic biomass-to-fermentable sugars conversion (Mosier et al., 2005).

2.4.3. Types of pretreatment

There are many types of pretreatment such as physical pretreatment, chemical pretreatment, biological pretreatment and combinations of pretreatment processes. The goal behind pretreatment is to minimize the use of energy, chemicals and low value byproducts.

2.4.3.1. Physical pretreatment

Increasing the accessible surface area and size of pores is achievable effectively with Physical pretreatment. It decreases the crystalline and degrees of polymerization of cellulose. Commonly used physical pretreatments of lignocellulosic residues include, grinding and milling, irradiation and microwave pretreatment.

2.4.3.1.1. Grinding and milling.

Usually grinding and milling are the initial steps of pretreatment of any biomass which reduces particle size, though the combination of grinding with other pretreatment method has been tried. Grinding and milling reduce the crystallinity of the biomass. Superfine grinding of steam exploded biomass has been tried and proved better than ground residue when hydrolyzed (Jin and Chen, 2006) though energy required for the process also has to be considered while going for commercial applications. For grinding rice straw wet disk milling proved better than ball milling both in terms of glucose recovery as well as energy saving (Hideno et al., 2009). Developments in this field provide a number of pretreatment which permits enzymatic saccharification, e.g. ball milling, roll milling, wet disk milling, and several type of grinding has been tried based on the biomass, though there are no reports particularly on rice straw as such.

2.4.3.1.2. Electron beam irradiation.

The cellulosic fraction of the lignocellulosic materials can be degraded by irradiation to fragile fibers, low molecular weight oligosaccharides and cellobiose (Kumakura and Kaetsu, 1983). It could be due to preferential dissociation of the glucosidal bonds of the cellulose molecular chains by irradiation in the presence of lignin. Irradiation methods are expensive, high energy demanding and have difficulties in industrial application. Jin et al. (2009) carried out physical pretreatment of milled dry rice straw using electron beam irradiation with accelerated electrons by a linear electron accelerator that had the capacity to produce electron beams. Enzymatic hydrolysis of electron beam irradiated and untreated rice straw were carried out and the result indicate that the untreated rice straw produced a glucose yield of 22.6% and the electron beam irradiated sample produced a glucose yield of 52.1% after hydrolysis for 132 h. SEM and X-ray diffraction analysis for the treated rice straw shows physical changes after electron beam irradiation. Because these methods do not involve the use of extreme temperatures, the generation of inhibitory substances produced during acid or alkali pretreatment can be either avoided or minimized.

2.4.3.1.3. Microwave pretreatment.

Microwave irradiation has been widely used in many areas because of its high heating efficiency and easy operation. Microwave irradiation could change the ultra structure of cellulose (Xiong et al., 2000) degrade lignin and hemicelluloses in lignocellulosic materials, and increase the enzymatic susceptibility of lignocellulosic materials (Azuma et al., 1984). Enzymatic hydrolysis of rice straw could be enhanced by microwave pretreatment in presence of water (Azuma et al., 1984; Ooshima et al., 1984) and also in glycerine medium with lesser amount of water (Kitchaiya et al., 2003). Rice straw treated by microwave irradiation alone had almost the same hydrolysis rate and reducing sugar yield compared to the raw straw (Zhu et al., 2005).

2.4.3.2. Chemical pretreatment

Chemical pretreatment of rice straw works on breaking the molecular bonds between sugar molecules and building blocks of the fibers. Chemical pretreatment is expensive and can pose a hazard to the environment and health risk if not well controlled.

Chemical waste treatment can also be an issue in developing countries where regulations are not strict, or laws are not implemented to force proper treatment of waste.

There are many chemical pretreatments available, and the most promising chemicals for pretreatment of rice straw include alkali and ammonia pretreatments. Chemical pretreatment is very important because it was found that later phases of treatment could not effectively convert lignocelluloses to soluble sugars without effective chemical pretreatment.

2.4.3.2.1. Alkali pretreatment.

It involves the application of alkaline solutions like NaOH or KOH to remove lignin and a part of the hemicelluloses, and efficiently increase the accessibility of enzymes to the cellulose in later treatment phases. The alkali pretreatment can result in a sharp increase in fiber breakdown into its sugar molecules building blocks, resulting in higher saccharification yields. Pretreatment can be performed at low temperatures but with a relatively long time and high concentration of the Alkali solution. Compared with acid or oxidative reagents, alkali treatment appears to be the most effective method in breaking the ester bonds between lignin, hemicellulose and cellulose, and avoiding fragmentation of the hemicellulose polymers (Gaspar et al., 2007).

Alkaline pretreatment of chopped rice straw with 2% NaOH with 20% solid loading at 85°C for 1 h decreased the lignin by 36% (Zhang and Cai, 2008). The separated and fully exposed micro fibrils increased the external surface area and the porosity of the rice straw, thus facilitating enzymatic hydrolysis. The main effect of sodium hydroxide pretreatment on lignocellulosic biomass is delignification by breaking the ester bonds cross-linking lignin and xylan, thus increasing the porosity of biomass (Tarkov and Feist, 1969).

2.4.3.2.2. Ammonia pretreatment.

As a pretreatment reagent ammonia has number of desirable characteristics. It is an effective swelling reagent for lignocellulosic materials. It has high selectivity for reactions with lignin over those with carbohydrates. Its high volatility makes it easy to recover and reuse. It is a non-polluting and noncorrosive chemical. One of the known reactions of aqueous ammonia with lignin is the cleavage of C–O–C bonds in lignin as well as ether and ester bonds in the lignin–carbohydrate complex (Kim and Lee, 2007).

A flow-through process called Ammonia Recycle Percolation (ARP) was developed for pretreatment. In this process, ammonia is pumped through a bed of biomass maintained at 170 °C. By this process up to 85% delignification and almost theoretical yield of glucose in enzyme hydrolysis can be achieved (Drapcho et al., 2008). Soaking in Aqueous Ammonia (SAA) pretreatment at mild temperatures ranging from 40 to 90 °C for longer reaction times has been used to preserve most of the glucan and xylan in the samples, which is subsequently fermented using the simultaneous saccharification and co-fermentation (SSCF) process (Kim and Lee, 2007; Kim et al., 2008). SAA is still a new method and its effectiveness has not yet been tested for many lignocellulosic feedstock including rice straw. Comparing to other alkalis such as sodium hydroxide or lime, ammonia is highly selective for lignin removal and shows significant swelling effect on lignocellulose. Also, it is easily recoverable due to its high volatility (Wyman et al., 2005).

The effectiveness of the SAA process is strongly dependent on the pretreatment temperature. The ammonia fiber/freeze explosion/expansion (AFEX) process uses anhydrous ammonia instead of aqueous ammonia. Similar to the ARP and SAA process, the ammonia used in the AFEX process can be recovered and recycled due to its high volatility. After treatment the only exit stream is a gas mix containing ammonia and water vapor. All biomass components remain with the treated solids. Thus, there is no loss of any carbohydrate fraction. Since all of the ammonia will quickly evaporate, there is no need for pH adjustment of the treated material over a wide range before it can be used in subsequent enzyme hydrolysis and soluble sugars production.

Enzyme hydrolysis of AFEX-treated biomass can produce glucose with greater than 90% theoretical yield and xylose with up to 80% theoretical yield. There is no formation of inhibitory compounds (Drapcho et al., 2008). AFEX is reported as an effective pretreatment process for rice straw as it resulted 3% sugar loss during pretreatment (Zhong et al., 2009).

Ferrer et al. (1997) carried out pretreatment of rice straw by a process called Ammonia Pressurization and Depressurization (PDA) using a laboratory-scale ammonia reactor unit consisting of a 4-L reactor with appropriate support equipment. Pretreatment followed by enzymatic hydrolysis resulted significant increase in sugar yield. Ko et al. (2009) carried out aqueous ammonia pretreatment and the optimum conditions were 21% ammonia concentration at 69 °C for 10 h. When AFEX was used in conjunction with 60 FPU of

cellulase/g-glucan and b-glucosidase, xylanase and other supplements, the typical glucose yields after 72–168 h of hydrolysis were 60–100% of the theoretical maximum.

2.4.3.2.3. Acid pretreatment.

Pretreatment of lignocellulose with acids at ambient temperature enhance the anaerobic digestibility. Dilute acid pretreatment predominantly affect hemicellulose with little impact on lignin degradation. Pretreatment for prolonged periods of time proves effective in impregnation of acid into densely packed fibers. Acid pretreatment will attack the hemicellulose, and by this, making the cellulose better accessible to enzymes. Acid pretreatment is usually carried out using mineral acids like HCl and H_2SO_4. Following dilute acid treatment, the enzyme cellulase can be utilized for hydrolysis of the remaining carbohydrates in the treated biomass. Dilute acid pretreatment can be a simple single-stage process in which biomass is treated with dilute sulfuric acid at suitable acid concentrations and temperatures for a period of time. To reduce enzyme requirements, a two-stage process was developed at the National Renewable Energy Laboratory (NREL) in Golden, Colorado. A schematic diagram of this process is shown in figure 2.12. Literatures regarding dilute acid hydrolysis of rice straw is limited because of the inability of the process to remove lignin and low sugar yield (Sumphanwanich et al., 2008).

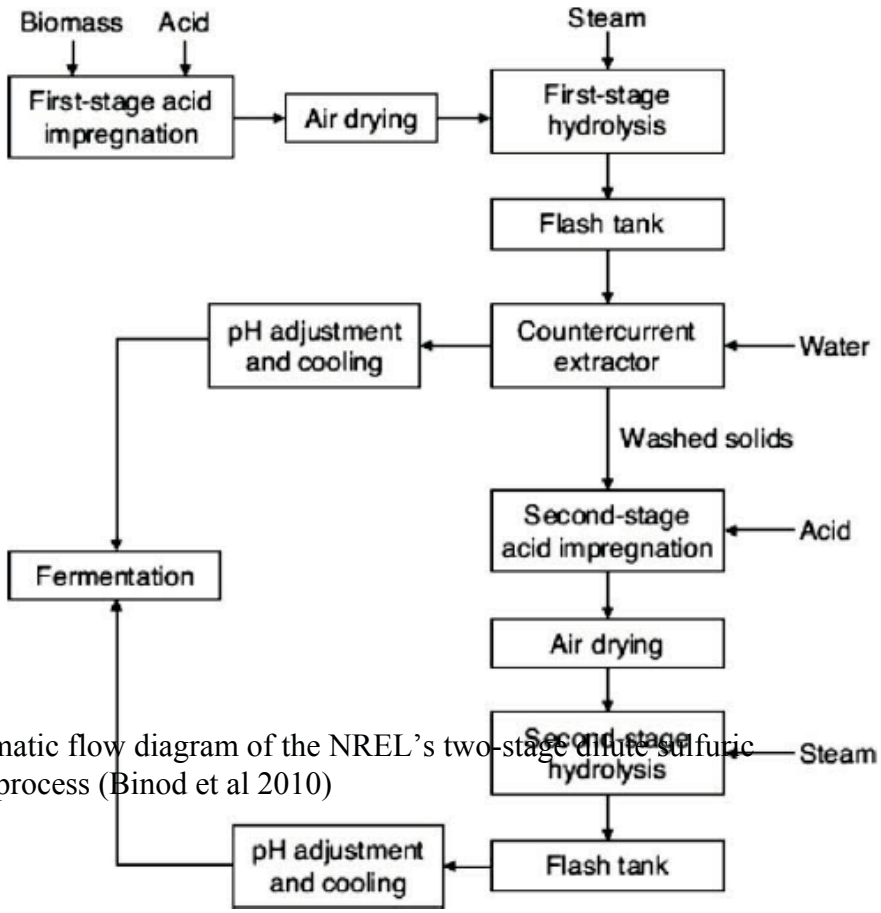


Figure 2.12 Schematic flow diagram of the NREL's two-stage dilute sulfuric acid pretreatment process (Binod et al 2010)

2.4.3.2.4. Pretreatment with oxidizing agent.

Oxidative pretreatment involves the addition of an oxidizing compound, like hydrogen peroxide or peracetic acid, to the biomass, which is suspended in water. This pretreatment remove the hemicellulose and lignin to increase the accessibility of the cellulose. During oxidative pretreatment several reactions can take place, like electrophilic substitution, displacement of side chains, cleavage of alkyl aryl ether linkages or the oxidative cleavage of aromatic nuclei (Hon and Shiraishi, 2001). Hydrogen peroxide pretreatment utilizes oxidative delignification to detach and solubilize the lignin and loosen the lignocellulosic matrix thus improving enzyme digestibility (Martel and Gould, 1990).

Wei and Cheng (1985) evaluated the effect of hydrogen peroxide pretreatment on the change of the structural features and the enzymatic hydrolysis of rice straw. Changes in the lignin content, weight loss, accessibility for Cadoxen, water-holding capacity, and crystallinity of straw were measured during pretreatment to express the modification of

the lignocellulosic structure of straw. The rates and the extents of enzymatic hydrolysis, cellulase adsorption, and cellobiose accumulation in the initial stage of hydrolysis were determined to study the pretreatment effect on hydrolysis. Pretreatment at 60 °C for 5 h in a solution with 1% (w/w) H_2O_2 and NaOH resulted in 60% delignification, 40% weight loss, a fivefold increase in the accessibility for Cadoxen, one times increase in the water-holding capacity and only a slight decrease in crystallinity as compared with that of the untreated straw. Improvement on the pretreatment effect could be made by increasing the initial alkalinity and the pretreatment temperature of hydrogen peroxide solution.

A saturated improvement on the structural features was found when the weight ratio of hydrogen peroxide to straw was above 0.25 g H_2O_2/g straw in an alkaline H_2O_2 solution with 1% (w/w) NaOH at 32 °C. The initial rates and extents of hydrolysis, cellulase adsorption, and cellobiose accumulation in hydrolysis were enhanced in accordance with the improved structural features of straw pre-treated. A four times increase in the extent of the enzymatic hydrolysis of straw for 24 h was attributed to the alkaline hydrogen peroxide pretreatment.

Reports are there for employing per acetic acid for the pretreatment of rice straw (Taniguchi et al., 1982; Toyama and Ogawa, 1975). Quantitative changes in the composition of the treated straw, crystallinity of the treated straw and extracted cellulose, and susceptibility of the treated straw with per acetic acid resulted in a slight loss in hemicellulose and cellulose in the straw. The per acetic acid treatments caused little or no breakdown of the crystalline structure of cellulose in the straw. The degree of enzymatic solubilization relative to the amount of residual straw was 42% after treatment with 20% per acetic acid.

2.4.3.2.5. Organosolv pretreatment.

Organosolv pretreatment enhances the enzymatic digestibility mainly by delignification and hemicellulose removal leaving a cellulose-rich residue, which can be hydrolyzed with enzymes at high rates and to almost theoretical glucose yield. Hemicellulose and lignin can be recovered for production of high-value co-products. The change of cellulose crystallinity during organosolv pretreatment is not clear yet, but it has been found that the swelling of cellulose in organic solvent strongly depends on the species of organic solvents, solvent concentration and temperature (Mantanis et al., 1994,

1995). The organosolv process uses hot organic solvents such as ethanol at acidic pH to fractionate biomass components. It was first considered for paper making, but recently it has also been considered for pretreatment of lignocellulosic feedstock for ethanol production. There are some inherent drawbacks to the organosolvent pretreatment.

Organosolvent pretreatment is expensive at present than the leading pretreatment processes but the separation and recycling of the applied solvent could reduce the operational costs of the process. It also requires strict controlled conditions due to the volatility of organic solvents. Removal of solvents from the pre-treated cellulose is usually necessary because the solvents might inhibit enzymatic hydrolysis and fermentation or digestion of hydrolysate (Xuebing et al., 2009). The commonly used organic solvents for pretreatment are solvents with low boiling points like ethanol and methanol and alcohols with high boiling points like ethylene glycol, glycerol, tetrahydrofurfuryl alcohol and other organic compounds like dimethylsulfoxide, ethers, ketone, and phenols (Thring et al., 1990). Organosolv processes, if the pretreatment is conducted at high temperatures (185–210 °C), there is no need for acid addition but at lower temperature requires addition of catalysts.

Jamshid et al. (2005) reported rice straw pulping using diethylene glycol, mixture of diethylene glycol and ethylene glycol at atmospheric pressure. Pretreatment with high boiling point solvents enhance delignification. The most important advantage for high boiling point alcohol pretreatment is that the process can be performed under atmospheric pressure. Jahan (2006) reported acetic acid or formic acid pretreatment of rice straw with the variation of reaction variables. Maximum pentosan dissolution was observed in 80% acetic acid with 0.6% H_2SO_4 catalyst at 80 °C for 120 min. Acetic acid dissolved pentosan more slowly than formic acid.

2.4.3.3. Biological pretreatment

Biological pretreatment offers some conceptually important advantages such as low chemical and energy use, but a controllable and sufficiently rapid system has not yet been found. Biological pretreatment is less hazardous than chemical pretreatments and produces less unfavorable byproducts. Chemical pretreatments have serious disadvantages in terms of the requirement for specialized corrosion resistant equipment, extensive washing, and proper disposal of chemical wastes. Biological pretreatment is a

safe and environmentally-friendly method for lignin removal from lignocellulose. The most promising microorganisms for biological pretreatment are white-rot fungi that belong to class Basidiomycetes.

The effects of biological pretreatment of rice straw using four white-rot fungi *(Phanerochaete chrysosporium, Trametes versicolor, Ceriporiopsis subvermispora, and Pleurotus ostreatus)* were evaluated on the basis of quantitative and structural changes in the components of the pre-treated rice straw as well as susceptibility to enzymatic hydrolysis (Taniguchi et al., 2005). Of these white- rot fungi, P. ostreatus selectively degraded the lignin fraction of rice straw rather than the hemicellulose component. When rice straw was pre-treated with P. ostreatus for 60 d, the total weight loss and the degree of Klason lignin degraded were 25% and 41%, respectively.

After the pretreatment, the residual amounts of cellulose and hemicellulose were 83% and 52% of those in untreated rice straw, respectively. By enzymatic hydrolysis with a commercial cellulase preparation for 48 h, 52% hemicellulose and 44% cellulose in the pre-treated rice straw were solubilized. The net sugar yields based on the amounts of hemicellulose and cellulose of untreated rice straw were 33% for total soluble sugar from hemicellulose and 32% for glucose from cellulose (Taniguchi et al., 2005). The biological pretreatment induces structural loosening of cells with a simultaneous increase in porosity. The Scanning Electron Microscopic (SEM) observations show that the pretreatment with P. ostreatus resulted in an increase in susceptibility of rice straw to enzymatic hydrolysis due to partial degradation of the lignin that is responsible for preventing penetration of cellulase in the rice straw as described above.

Patel et al. (2007) did a preliminary study on the microbial pretreatment and fermentation of the agricultural residues like rice straw. A combination of five different fungi viz. Aspergillus niger, Aspergillus awamori, Trichoderma reesei, Phenerochaete chrysosporium, Pleurotus sajor-caju, obtained from screening were used for pretreatment and Saccharomyces cereviseae (NCIM 3095) was used for carrying out fermentation. Pretreatment with A. niger and A. awamori and later fermentation yielded highest amount of ethanol (2.2 g L^{-1}).

2.4.4. Combined pretreatment

Kun et al. (2009) reported pretreatment of rice straw with alkali assisted by photocatalysis which efficiently changed the physical properties and microstructure of rice straw also resulted in decrease in lignin content and thereby increasing the enzymatic hydrolysis rate of the pre-treated rice straw had. Alkali treatment of rice straw in the absence of H_2O_2 favored solubilization of the small molecular size of hemicelluloses, which are rich in glucose, probably originating from a-glucan, while the second stage treatment by alkaline peroxide enhanced dissolution of larger molecular size hemicelluloses, which were rich in xylose.

Microwave is emerging as an important and efficient pretreatment method when applied in combination with other methods. Zhu et al. (2006) reported several combinations of microwave pretreatment of rice straw along with acid and alkali which removes hemicellulose and lignin, respectively, and microwave removes more lignin compared to pretreatment with alkali alone. The results show that higher microwave power with shorter pretreatment time and the lower microwave power with longer pretreatment time had almost the same effect on the weight loss and composition at the same energy consumption. Microwave enhances some reactions in the pretreatment, but the detailed mechanism is still unclear.

Lu and Minoru (1993) reported radiation pretreatment of rice straw in the presence of NaOH solutions using an electron beam accelerator. Electron beam irradiation alter lignocellulosic structure so that NaOH solution could enter easily into the lignocellulosic complex and increase the rate of reaction so the lignin will be eliminated more easily and cellulose or hemicellulose scissored by irradiation was degraded slightly by NaOH which in turn increase the enzyme accessibility. Jin and Chen (2006) studied a combination of steam explosion and superfine grinding of rice straw and its enzymatic hydrolysis. Superfine grinding were combined with low severity steam explosion for treating rice straw to shorten the grinding time, save the energy cost, avoid the inhibitors, and obtain high enzymatic hydrolysis.

Superfine grinding was conducted after rice straw was steam exploded at low Ro (steam explosion severity factor) to avoid excessive decomposition of hemicellulose and side products generation from sugars and lignin. It shows difference in enzymatic

hydrolysis, chemical compositions, fiber characteristics and composed cells contents of the superfine ground steam exploded rice straw product and the ground steam exploded rice straw residue. Enzymatic hydrolysis of the superfine ground product gained the highest hydrolytic rate and yielded more reducing sugar, while the reducing sugar yield generated from the superfine ground residue was even lower than that from the untreated rice straw. Steam explosion and super fine grinding decrease particle size and improve reactive surface to the largest content, and it had been considered to be no more energy consuming than traditional mechanical grinding with respect to the increase of surface area.

2.5 Enzymatic hydrolysis

Enzymatic hydrolysis involves breaking down the bonds between the sugar building blocks of cellulose fibers, as well as hemicellulose by using enzymes. The cellulose usually contains only glucans, whereas hemicellulose contains polymers of several sugars such as mannan, xylan, glucan, galactan, and arabinan. Consequently, the main hydrolysis product of cellulose is glucose, whereas the hemicellulose gives rise to several pentoses and hexoses (Taherzadeh and Niklasson, 2004).

High lignin content blocks enzyme accessibility, causes end-product inhibition, and reduces the rate and yield of hydrolysis. In addition to lignin, cellobiose and glucose also act as strong inhibitors of cellulose enzymes. It is recommended to continuously separate the produced soluble sugars such as cellobiose and glucose in order to stop inhibition of the enzymes action (Knauf and Moniruzzaman, 2004). Various factors influencing the yields of the lignocellulose to the monomeric sugars and the by-products are, e.g., particle size, liquid to solid ratio, type and concentration of acid used, temperature, and reaction time, as well as the length of the macromolecules, degree of polymerization of cellulose, configuration of the cellulose chain, association of cellulose with other protective polymeric structures within the plant cell wall such as lignin, pectin, hemicellulose, proteins, and mineral elements.

Recent advances in enzyme technology for the conversion of cellulosic biomass to sugars have brought significant progress in lignocellulosic ethanol research. Enzymatic hydrolysis is usually carried out under mild conditions, i.e., low pressure and long retention time in connection to the hydrolysis of hemicellulose. Valdes and Planes (1983) studied the hydrolysis of rice straw using 5– 10% H_2SO_4 at 80–100 °C. They reported the best sugar yield at 100 °C with 10% H_2SO_4 for 240 min. Yin et al. (1982) studied the

hydrolysis of hemicellulose fraction of rice straw with 2% H_2SO_4 at 110–120 °C, where they succeeded to hydrolyze more than 70% of pentoses. Valkanas et al. (1998) carried out hydrolysis of rice straw with different acids with varying concentrations (0.5– 1% H_2SO_4, 2–3% HCl and 0.5–1% H_3PO_4) and they found that after 3 h retention time, rice straw pentosans converted to a solution of monosaccharides, suitable for fermentation.

Roberto et al. (2003) studied the effects of H_2SO_4 concentration and retention time on the production of sugars and the by-products from rice straw at relatively low temperature (121 °C) and long time (10– 30 min) in a 350-L batch reactor. The optimum acid concentration of 1% and retention time of 27 min was found to attain high yield of xylose (77%). The pretreatment of the straw with dilute sulfuric acid resulted in 0.72 g g1 sugar yield during 48 h enzymatic hydrolysis, which was higher than steam-pretreated (0.60 g g1) and untreated straw (0.46 g g1) (Abedinifar et al., 2009). When they increased the concentration of substrate from 20 to 50 and 100 g L1 sugar yield lowered to 13% and 16%, respectively.

The kinetics of glucose production from rice straw by Aspergillus niger was studied by Aderemi et al. (2008). Glucose yield was found to increase from 43 to 87% as the rice straw particle size decreased from 425 to 75 lm, while the optimal temperature and pH were found within the range of 45–50°C and 4.5–5, respectively. The study shows that the concentration and rate of glucose production is depend on pretreatment of rice straw, substrate concentration and cell loading. Enzymatic hydrolysis of alkali assisted photocatalysis of rice straw resulted 2.56 times higher hydrolysis rate than that of alkali process (Kun et al., 2009) whereas, ammonia treated rice straw resulted an increase of monomeric sugars from 11% in the untreated to 61% (Sulbaran-de-Ferrer et al., 2003). Hydrolysis efficiency of lignocellulosic biomass increases when combination of enzymes such as cellulase, xylanases and pectinases are employed rather than only cellulase (Zhong et al., 2009) but the cost of the process increases drastically even though from ecological point of view it is highly desirable.

2.6 Fermentation

Fermentation of rice straw can be utilized to process the large quantities of straw available. Making soluble sugars available for the microorganisms to feed on is the target. Two processes can be followed:

a) Soluble sugars can be generated from rice straw into a solution and then can be separated for introduction to the microorganisms to feed on. This process is called Separate Hydrolysis and Fermentation (SHF)

b) Soluble sugars can be generated and made available for the microorganisms to feed on simultaneously in the same solution as the fermentation solution. This process is called Simultaneous Saccharification and Fermentation (SSF)

The cellulose and hemicellulose fraction of rice straw can be converted to ethanol by either SSF or SHF processes. SSF is more favored because of its low potenpotential costs (Wyman, 1994). It results in higher yield of ethanol compared to SHF by minimizing product inhibition. One of the drawbacks of SHF is the difference in optimum temperature of the hydrolyzing enzymes and fermenting microorganisms which can result in microorganisms death.

Most of the reports states that the optimum temperature for enzymatic hydrolysis is at 40–50 °C, while the microorganisms with good ethanol productivity and yield do not usually tolerate this high temperature. This problem can be avoided by applying thermo-tolerant microorganisms such as Kluyveromyces marxianus, Candida lusitaniae, and Zymomonas mobilis or mixed culture of some microorganisms like Brettanomyces clausenii and Saccharomyces cerevisiae (Golias et al., 2002; Spindler et al., 1988). Punnapayak and Emert (1986) studied SSF of alkali-pre-treated rice straw with Pachysolen tannophilus and Candida brassicae, where P. tannophilus resulted in higher ethanol yields than C. brassicae in all the experiment.

However, they achieved only less than 30% of theoretical ethanol yield. SSF of acid-pre-treated rice straw with Mucor indicus, Rhizopus oryzae, and S. cerevisiae resulted an overall yield of 40–74% of the maximum theoretical ethanol yield (Karimi et al., 2006). The SSF of alkali and microwave/alkali pretreated rice straws to ethanol using cellulase from T. reesei and S. cerevisiae were studied by Zhu et al. (2006). Under the optimum conditions ethanol concentration reached 29.1 g L1 and ethanol yield was 61.3%. The study shows that production of ethanol from microwave/alkali pre-treated rice straw had lower enzyme loading, shorter reaction time, and achieved higher ethanol concentration and yield than rice straw pre-treated by alkali alone. There are many reports

stating that the simultaneous saccharification and fermentation (SSF) is superior to the traditional saccharification and subsequent fermentation in the production of ethanol from rice straw because the SSF process can improve ethanol yields by removing end-product inhibition of saccharification process and eliminate the need for separate reactors for saccharification and fermentation (Chadha et al., 1995).

Separate enzymatic hydrolysis and fermentation of rice straw by M. indicus, R. oryzae, and S. cerevisiae were studied by Abedinifar et al. (2009). Their study concludes that M. indicus is able to produce ethanol from pentoses. This species seems to be a good strain for production of ethanol from lignocelluloses, particularly for rice straw.

In addition to SSF and SHF, there is another process called consolidated bioprocessing (CBP). In this process, cellulase production, biomass hydrolysis, and ethanol fermentation are carried out together in a single reactor. A microorganism that can efficiently ferment cellulose directly to ethanol, such as Clostridium phytofermentans, will be most suitable for this process. Glucose and xylose are two dominating sugars in the lignocellulosic hydrolysates. The main difficulty of using two microorganisms for the co-fermentation of these two sugars is the inability to provide optimal environmental conditions for the two strains simultaneously (Chandrakant and Bisaria, 1998). A majority of previous studies on strain co-cultures reported that, while the fermentation of glucose in the sugar mixture proceeded efficiently with a traditional glucose-fermenting strain, the fermentation of xylose was often slow and of low efficiency due to the conflicting oxygen requirements between the two strains and/or the catabolite repression on the xylose assimilation caused by the glucose.

Approaches in both process engineering and strain engineering have been carried out to circumvent these difficulties and to improve the system efficiency. Examples of process engineering include continuous culture (Grootjen et al., 1991; Laplace et al., 1993; Delgenes et al., 1996), the immobilization of one of the strains (Grootjen et al., 1991), co-immobilization of two strains (Grootjen et al., 1991; deBari et al., 2004), two stage fermentation in one bioreactor (i.e. sequential culture) (Fu and Peiris, 2008), and separate fermentation in two bioreactors.

2.7 Combined processes:

Haagensen et.al. (2002) used alkaline wet oxidation as pretreatment method of Sugarcane Bagasse (SB) and Rice Straw (RS) prior to enzymatic hydrolysis and glucose fermentations with *Saccharomyces cerevisiae.* At high enzyme loadings, the enzymatic hydrolysis of Wet Oxidized Sugarcane Bagasse (SBWO) resulted in the highest degree of saccharification compared to Wet Oxidized Rice Straw (RSWO). However, at enzyme concentrations below 10 FPU/g-cellulose, wet oxidized rice straw showed faster hydrolysis and higher levels of saccharification. Incomplete hydrolysis was found for both biomass suspensions with maximum yields of 73% and 62% (of theoretical) for SBWO and RSWO, respectively. Ethanol yields from simultaneous saccharification and fermentation (SSF) were similar to what would be expected from the enzymatic hydrolysis. Based on this, it was concluded that the results of enzymatic hydrolysis was not affected by feedback inhibition of the enzymes. The maximum ethanol yields from SSF of SBWO and RSWO were 0.39 g-ethanol/g-glucose and 0.31 g-ethanol/g-glucose, respectively.

Similar ethanol yields of SBWO and RSWO was seen at enzyme loadings of 25 FPU/g-cellulose when separate hydrolysis and fermentation (SHF) was applied. However, SHF of SBWO resulted in a specific ethanol yield (222 1-ethanol/t-SB) that was 19% higher than that of RSWO (186 1-ethanol/t-RS). The specific ethanol yields obtained correspond to 89% and 87% of the theoretical yield based on the cellulose content of SB an RS, respectively. The results indicate that alkaline wet oxidation is a promising technology for pretreatment of sugarcane bagasse and rice straw in bioethanol production.

Sun (2002) studied lignocellulosic biomass to produce ethanol as a promising alternative source of energy instead of crude oil. In the study, two main processes were involved in the conversion: Hydrolysis of cellulose in the biomass lignocellulosic material to produce reduced sugars as well as fermentation of produced sugars into ethanol. In his study it was found that the cost of ethanol production from lignocellulosic materials was very high with the use of today's technologies. The most significant challenges were the low yields of ethanol and high cost of the hydrolysis process. Considerable efforts had been made to improve yields from lignocellulosic materials. Pretreatment to remove lignin and hemicelluloses significantly enhanced the hydrolysis process of cellulose. The use of enzymes can also affect the hydrolysis process

positively. Most importantly glucose presence in the hydrolysate is an essential key factor to cellulase inhibition. It was recommended to conduct simultaneous saccharification and fermentation to remove the glucose and achieve higher cellulose hydrolysis and more glucose yields.

Roberto et al (2003) investigated the effects of H_2SO_4 concentration and reaction time on the production of sugars (xylose, glucose and arabinose) and on the reaction byproducts (furfural, hydroxymethylfurfural (HMF) and acetic acid). Dilute sulfuric acid was used as a catalyst for the hydrolysis of rice straw at 121°C in a 350-l batch hydrolysis reactor. Rationale for conducting this study was determined based on a central composite statistical design. Response surface methodology (RSM) was adopted to optimize the hydrolysis conditions aiming to attain high xylose selectivity. The optimum H_2SO_4 concentration of 1% and reaction time of 27 min was found. Under these conditions, 77% of xylose yield and 5.0 g g^{-1} of selectivity were attained.

Yukihiko Matsumura et.al (2005) discussed the use of agricultural residue in Japan as an energy resource, based on the amounts produced and availability. The main agricultural residues in Japan were rice straw and rice husk. Based on a scenario wherein these residues were collected as was the rice product, we evaluate the size, cost, and CO_2 emission for power generation. Rice residue has a production potential of 12 Mt-dry yearly, and 1.7 kt of rice straw was collected for each storage location. As this is too small an amount even for the smallest scale of power plant available, 2-month operation per year is assumed. Assuming a steam boiler and turbine with an efficiency of 7%, power generation from rice straw biomass can supply 3.8 billion kW h of electricity per year, or 0.47% of the total electricity demand in Japan. The electricity generated from this source costs as much as 25 JPY/kW h about 0.21 USD/kW h (1 USD = 120 JPY), more than double the current price of electricity. With heat recovery at 80% efficiency, the simultaneous heat supplied via cogeneration reaches 10% of that supplied by heavy oil in Japan. Further cost incentives will be required if the rice residue utilization is to be introduced. It will also be important to develop effective technologies to achieve high efficiency even in small-scale processes. If Japanese technologies enable the effective use of agricultural residue abroad as a result of Japanese effort from the years after 2010, the resulting reduction of greenhouse gas emission can be counted under the framework of the Kyoto Protocol.

Karimi et al (2006) investigated hydrolysis of rice straw by dilute sulfuric acid at high temperature and pressure in one and two stages. The hydrolyses were carried out in a 10-l reactor, where the hydrolysis retention time (3–10 min), pressure (10–35 bar) and acid concentration (0–1%) were examined. Optimization of first stage hydrolysis is desirable to achieve the highest yield of the sugars from hemicellulose and also as a pretreatment for enzymatic hydrolysis. The results show the ability of first stage hydrolysis to depolymerize xylan to xylose with a maximum yield of 80.8% at hydrolysis pressure of 15 bar, 10 min retention time and 0.5% acid concentration. However, the yield of glucose from glucan was relatively low in first stage hydrolysis at a maximum of 25.8%. The solid residuals were subjected to further dilute-acid hydrolysis in this study. This second-stage hydrolysis without addition of the acid could not increase the yield of glucose from glucan beyond 26.6%. On the other hand, the best results of the hydrolysis were achieved, when 0.5% sulfuric acid was added prior to each stage in two-stage hydrolysis. The best results of the second stage of the hydrolysis were achieved at the hydrolysis pressure and the retention time of 30 bar and 3 min in the second stage hydrolysis, where a total of 78.9% of xylan and 46.6% of glucan were converted to xylose and glucose, respectively in the two stages. Formation of furfural and HMF were functions of the hydrolysis pressure, acid concentration, and retention time, whereas the concentration of acetic acid were almost constant at pressure of higher than 10 bar and a total retention time of 10 min.

Abou Zeid et al (2008) From a previous research done by Abou Zeid, a local yeast isolate Candida tropicalis gave the highest yield production of xylitol in fermentation medium in which xylose was used as the sole carbon source compared with Candida guilliermondii NRRLY-488. The produced amounts of xylose reductase (XR) enzyme and xylitol sugar as well as the biomass of both tested yeast isolates inoculated in fermentation media containing hot water hydrolysate of rice straw without additional carbon source, were very low. Meanwhile, treated rice straw with Na OH or H SO_4 increased the xylose and total sugars several times in the hydrolysate but toxic compounds were obtained, namely acetic acid, furfural, 5- hydroxy methyl furfural (HMF) and phenolic compounds. Thus, activated charcoal was used to eliminate the toxic compounds produced in the treated rice straw hydrolysate (acid + heat) inoculated with C. tropicalis and C. guilliermondii. Xylitol yield produced as a result to charcoal treatment reached 36.63 and 41.50g/l out of 60g/l xylose compared with 15.0and 28.0g/l

out of 65g/l xylose without charcoal treatment for both isolates, respectively. Meanwhile inoculating the previous hydrolysate medium with the adapted cells of the two tested isolates produced the highest xylitol yield (45.2 and 47.35g/l) for both strains, respectively. The purification procedure resulted in 2.774 and 14.917 folds of xylose reductase (XR) from ammonium sulphate and Sephadex-G200 purification steps with a recovery of 69.806 and 27.139% and specific activity of 1.990 and 19.514 U/mg proteins, respectively. The molecular masses of purified xylose reductase (XR) and xylitol dehydrogenase (XD) were found to be 36.48 and 89.5 k Daltons, respectively. The amino acids analysis of xylose reductase (XR) showed that glutamic and aspartic acids are present in high percentages while tyrosine and methionine were in low values and cysteine was not detected.

Taherzadeh et.al. (2008) dedicated to reviewing the methods that have been studied for pretreatment of lignocellulosic wastes for conversion to ethanol or biogas. Effective parameters in pretreatment of lignocelluloses, such as crystallinity, accessible surface area, and protection by lignin and hemicellulose are described first. Then, several pretreatment methods were discussed and their effects on improvement in ethanol and/or biogas production were described. They include milling, irradiation, microwave, steam explosion, ammonia fiber explosion (AFEX), supercritical CO_2 and its explosion, alkaline hydrolysis, liquid hot-water pretreatment, organosolv processes, wet oxidation, ozonolysis, dilute- and concentrated-acid hydrolyses, and biological pretreatments. All these methods should make the lignocelluloses available to the enzymatic attack, where crystallinity of cellulose, its accessible surface area and protection by lignin and hemicellulose were the main factors in order to obtain an efficient hydrolysis. In addition, the efficient utilization of the hemicelluloses was an opportunity to reduce the cost of ethanol or biogas production.

Diverse advantages had been reported for most of the pretreatment methods, which make them interesting for industrial applications. While methods such as dilute acid, hot water, AFEX, ammonia recycle percolation, and lime were capital-intensive, some other methods such as biological pretreatment were extremely slow. Furthermore, some technological factors such as energy balance, solvent recycling and corrosion, as well as environmental factors such as wastewater treatment, should be carefully considered for the selected method.

Ramos (2009) indicated that pretreatment of lignocellulosic materials was an essential step for bioconversion because of the various chemical and physical barriers that can greatly inhibit their susceptibility to bioprocess namely hydrolysis and fermentation. His aim was to review some of the important pretreatment methods developed to enhance the conversion of lignocellulosics. Steam explosion precluded the treatment of biomass with high pressure steam as method of choice. The optimal pretreatment condition for different plant biomass was different. The resulting best substrate for hydrolysis was obtained with minimal losses of soluble sugars to side reactions. Pretreatment optimization was the result of a compromise between opposing constraints. The main reason was inherent to acid hydrolysates which can only be maximized by lowering pretreatment severities while developing substrate requires more severe pretreatment conditions. In either case, severe conditions upstream or downstream will result in sugar losses. For this reason, it is best to use a weak acid at low concentration while increasing the reaction time and lowering the pressure and lowering the temperature below the degradation limits for sugars.

Sorahi et al (2009) were successfully converted rice straw to ethanol by separate enzymatic hydrolysis and fermentation by Mucor indicus, Rhizopus oryzae, and Saccharomyces cerevisiae. The hydrolysis temperature and pH of commercial cellulase and b-glucosidase enzymes were first investigated and their best performance obtained at 45°C and pH 5.0. The pretreatment of the straw with dilute-acid hydrolysis resulted in 0.72 g g-1 sugar yield during 48 h enzymatic hydrolysis, which was higher than steam-pretreated (0.60 g g^{-1}) and untreated straw (0.46 g g^{-1}). Furthermore, increasing the concentration of the dilute-acid pretreated straw from 20 to 50 and 100 g L^{-1} resulted in 13% and 16% lower sugar yield, respectively. Anaerobic cultivation of the hydrolyzates with M. indicus resulted in 0.36–0.43 g g-1 ethanol, 0.11–0.17 g g^{-1} biomass, and 0.04–0.06 g g-1 glycerol, which is comparable with the corresponding yields by S. cerevisiae (0.37–0.45 g g^{-1} ethanol, 0.04–0.10 g g^{-1} biomass and 0.05– 0.07 glycerol).

These two fungi produced no other major metabolite from the straw and completed the cultivation in less than 25 h. However, R. oryzae produced lactic acid as the major by-product with yield of 0.05–0.09 g g^{-1}. This fungus had ethanol, biomass and glycerol yields of 0.33–0.41, 0.06–0.12, and 0.03–0.04 g g^{-1}, respectively. The results of this work showed that the dilute-acid pretreatment is more efficient in improving enzymatic hydrolysis than just steaming. It is probably due to better removal of hemicellulose and lignin by dilute-acid pretreatment. The optimum conditions for enzymatic hydrolysis with

respect to pH, temperature and substrate concentration were investigated and chosen for this work. Generally, the optimum conditions depend on the properties of the applied enzyme. Enzyme inactivation and inhibition by hydrolysis products could be factors accounting for the low degree of carbohydrate conversion at higher substrate concentrations. These could be the reason for higher sugar yield in lower substrate concentration in the current work.

Teng-Chieh et al (2010) aimed in their study to propose operational conditions for the dilute acid pretreatment of rice straw and to explore the effect of the structural properties of the solid residues on the enzymatic hydrolysis. A maximal sugar yield of 83% was achieved when the rice straw was pretreated with 1% (w/w) sulfuric acid with a reaction time of 1–5 min at 160°C or 180°C, followed by enzymatic hydrolysis. The completely release of sugar (xylose and glucose) increased the pore volume of the pretreated solid residues resulted in an efficiency of 70% for the enzymatic hydrolysis. The extra pore volume was generated by the release of acid-soluble lignin and this resulted in the enzymatic hydrolysis being enhanced by nearly 10%. The increase in the crystallinity index of the pretreated rice straw was limited. These results were consistent with those from the Fourier transformer infrared (FTIR) analysis.

Park et al. (2011) determined the major carbohydrates of rice straw samples in order to evaluate the potential of using rice straw as a feedstock for ethanol production in Japan. Straw samples were harvested by cutting the plants at ground level when the grains were mature and immediately heating or chilling the samples. In all cases, significant amounts (62-303 g kg-1) of soft carbohydrates defined as consisting of glucose, fructose, sucrose, starch and b-1,3-1,4- glucan were detected, in addition to structural carbohydrates (cellulose and xylan). These results indicate that rice straw is a rich source of fermentable sugars from both soft carbohydrates and lignocellulosic portions of the cell wall.

Cai et al (2012) studied the central composite design of response surface method to optimize dilute H_2SO_4 pretreatment of corncob, in respect to acid concentration (0.16e1.84%), treatment time (0.16e1.84 h) and temperature (105-130°C) for xylose production. Enzymatic hydrolysis of the remaining solid was carried out further to evaluate the acid pretreatment conditions for maximizing glucose production. The results showed that pretreatment conditions for the highest xylose production was 1% sulfuric acid for 1.5 h at 123°C, corresponding to 87.2% total xylan converted to xylose and that for the highest glucose + cellobiose recovery was 0.5% sulfuric acid for 30 min at 125 C,

corresponding to 78.1% total glucan converted to glucose + cellobiose. In the subsequent simultaneous saccharification and fermentation (SSF) experiments using 14% glucan substrates pretreated under above two kinds of conditions, 47 g $^{-1}$ ethanol with a 65.8% theoretical yield and 50.2 g l $^{-1}$ ethanol with a 70.4% theoretical yield were obtained, respectively. This study had demonstrated that xylan in the corncob can be removed efficiently by dilute H_2SO_4 pretreatment. The optimal combination of pretreatment conditions was found to be 1% sulfuric acid and treatment time of 1.5 h at 123°C, corresponding to 87.2% total xylan converted to xylose. Conditions for the highest xylose yield in acid hydrolysis stage did not give the best glucose yields in enzymatic hydrolysis stage. The highest glucose + cellobiose recovery was 0.5% sulfuric acid for 30 min at 125°C, corresponding to 78.1% total glucan converted to glucose + cellobiose. Pretreatment conditions required for best sugar yields depended on which sugars and which products were targeted.

In the subsequent simultaneous saccharification and fermentation (SSF) experiments using 14% glucan substrates pretreated under above two kinds of conditions, 47 g l $^{-1}$ ethanol with a 65.8% theoretical yield and 50.2 g l^{-1} ethanol with a 70.4% theoretical yield were obtained. This study showed some xylose release during enzymatic hydrolysis or SSF process. If the xylose and arabinose presented in the broth at the end of the SSF could be fermented to ethanol, another 8.0 g l^{-1} ethanol could theoretically be produced (0.51 g ethanol/g pentose).

Guerra et al (2012) studied the xylose production from wheat straw by sulphuric acid hydrolysis at 130 °C. Several mass fraction of acid (1, 2, 3, 4 or 5%) were evaluated. Kinetic models were developed to explain the variation with time of xylose, glucose, arabinose, furfural, 5-(hydroxymethyl)-2- furaldehyde and acetic acid in the hydrolysates. Optimal conditions found were a H_2SO_4 mass fraction of 2% at 130°C for 29 min, which yielded a solution with xylose, 18.9 kg m^{-3}; glucose, 3.5 kg m^{-3}; arabinose, 3.1 kg m^{-3}; furfural, 0.6 kg m^{-3}; 5 (hydroxymethyl)-2-furaldehyde, 0.3 kg m^{-3}and acetic acid, 2.3 kg m^{-3}.

In these conditions, 99% of the hemicelluloses and 11% of the glucan were hydrolysed. The operational conditions 2% H_2SO_4 at 130°C for 29.3 min were selected because it resulted in solutions with high concentration of fermentable sugars (25.5 kg m^{-3}) and low concentrations of growth inhibitors (less than 0.9 kg m^{-3} for furfural-HMF and 2.3 kg m^{-3}for acetic acid). In these conditions, approximately 99% of the hemicellulosic

sugars were hydrolysed with a small concentration of by-products and only 12% degradation of the glucan fraction.

Kittamas Sirichai et al (2010) utilized pretreatment of lignocellulosic waste for rice straw, cassava pulp and cassava peels. Acidic and alkaline solutions were used in combination with heat, either individually or in combination, to establish a feasible pretreatment method prior to enzymatic hydrolysis. Individual substrates were also used for further bioethanol production. Pretreatment of rice straw using 2% NaOH at 85°C for 1 hour prior to enzymatic hydrolysis yielded glucose and xylose as 430.0 ± 0.5 and 162.0 ± 0.2 mg per g dried substrate, respectively. The cassava pulp pretreatment by 1N HCl prior to enzymatic hydrolysis revealed the amount of glucose and xylose as 410.3± 0.5 and 31.2 ± 0.1 mg per g dried substrate, respectively. Similar acid pretreatment scheme was also found to be feasible for cassava peel as a result of 414.1 ± 0.5 mg glucose and 24.3 ± 0.1 mg xylose obtained per gram of dried substrate.

Yamaguchi et.al. (2010) investigated the hydrolysis of lignocellulosic materials, specifically rice traw, bagasse and Japanese cedar, with a highly active solid acid catalyst, a carbon material bearing SO_3H, COOH and OH groups at 373 K through an artificial neural network (ANN) and a response surface methodology (RSM). The ANN models developed for experimental design accurately reflect the novel solid-solid interface catalysis. The ANN models revealed that the amount of water dominates the hydrolysis reaction. The correlations between the reaction properties and the properties of these lignocellulosic materials are discussed on the basis of the reaction mechanism. The catalytic hydrolysis of lignocellulosic materials (rice-straw, bagasse, and Japanese cedar) into glucose using the carbonbased solid acid catalyst proceeds as well as with sulfuric acid, even though lignocellulosic material has a very complex structure, the cell wall. The formation rates of glucose in the hydrolysis of lignocellulosic materials were lower than those in the hydrolysis of pure crystalline cellulose. This is attributed to the complex structure of the cell wall of lignocellulosic material, especially the presence of lignin. It is expected that the catalytic activity of the carbon material would be improved by pretreatment to remove lignin.

CHAPTER (3)
EXPERIMENTAL WORK

Introduction:

Specialized equipment is required to carry out the experimental work. Requirements are steam and pressure and control media to safely administer the thermal treatment at different conditions.

Parameters to consider:

- Reactor volume of 9L to 10L to be able to treat batches of 400g rice straw after soaking in Acid of concentrations 0.5%, 1.0%, 2.0% and 4.25%
- Boiler volume of 3L to 4L to produce sufficient amount of steam sufficient to carry out the longest experiments retention times of 120 minutes.
- The apparatus can be operated as a batch reactor for experimental amounts of RS.
- The materials used to build the apparatus needs to be selected to resist chemicals and temperatures shock for safe operation such as stainless steel 316.
- The operating process needs to be made low tech / simple in order to be viable.

The necessary specialized equipment was not available on shelf or for direct usage or purchase. Equipment components were purchased from local market and assembled in house to fulfill the operating parameter requirements.

3.1 Experimental Apparatus:

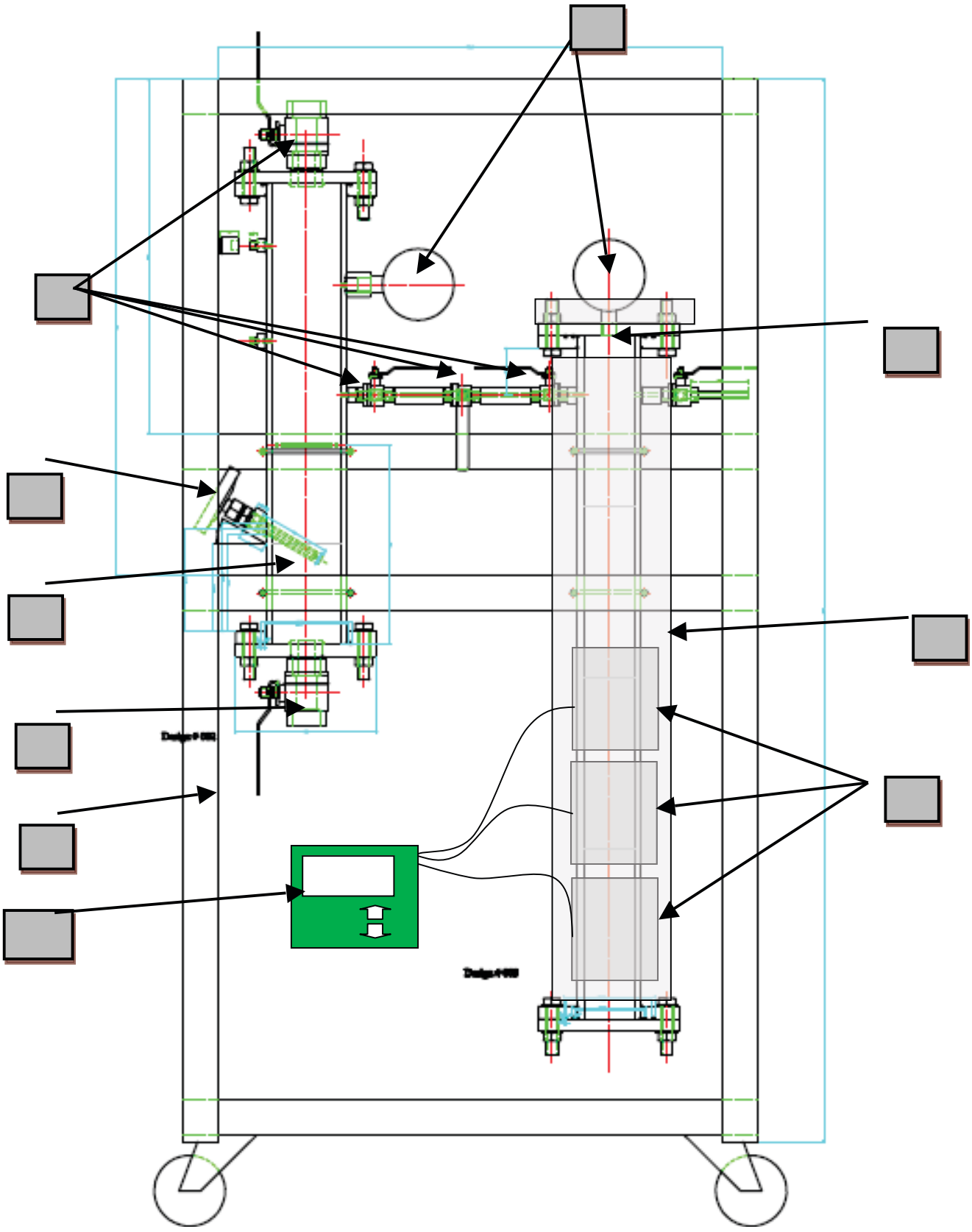
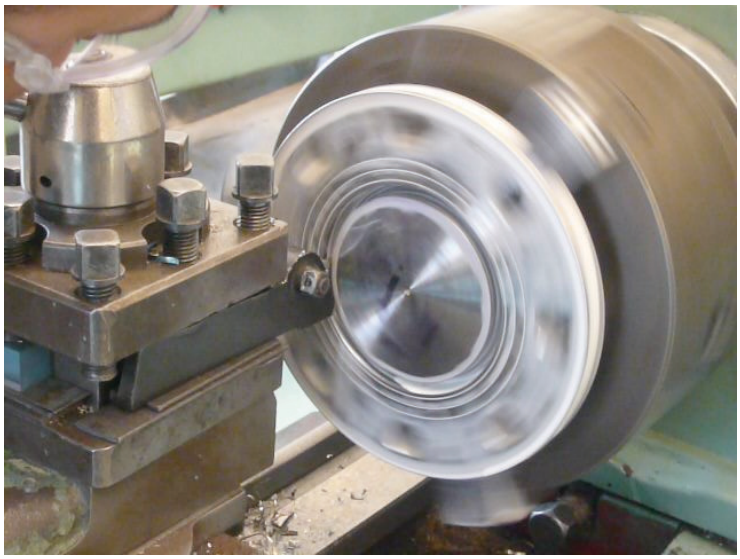


Figure 3.3 Machining of Experimental Apparatus.
Flanges machining and sealing grooves preparation.

The apparatus in figure 3.1 was designed and built locally for the purpose of processing rice straw at different retention times and pressures for different pretreatment acid percent concentrations for soluble sugars production. The reactor body is made of stainless steel 316 t=8mm which is capable of withstanding 200 bar at 400^0C. The main body is made of a standard seamless pipe section cut to length to achieve the desired volume inside the reactor as seen in figure 3.2.

Figure 3.2 Machining of Experimental Apparatus.
Main reactor body and boiler body machining from standard seamless pipe.

Four stainless steel flanges were also machined as seen in figure 3.3, from a 20mm thick plate. The material was also selected to be stainless steel 316. Standard size grooves were made to incorporate standard high pressure sealing rings that are to be fitted in each end.



All ball valves used are stainless steel chemical resistant ball valves. The Pressure rating for all used ball valves is 70 bar each. Double blocking is used for safety where two consecutive ball valves in case one fails or leaks as outlined in figure 3.4. Water is introduced into the boiler from valve set #3. Valve set #1 allows flow of steam from the Boiler to the reactor. Valve set #5 is used to collect the effluent. Valve set #4 is used to release the steam pressure. Valve set #2 is used to inject cooling air into the reactor body after each run.

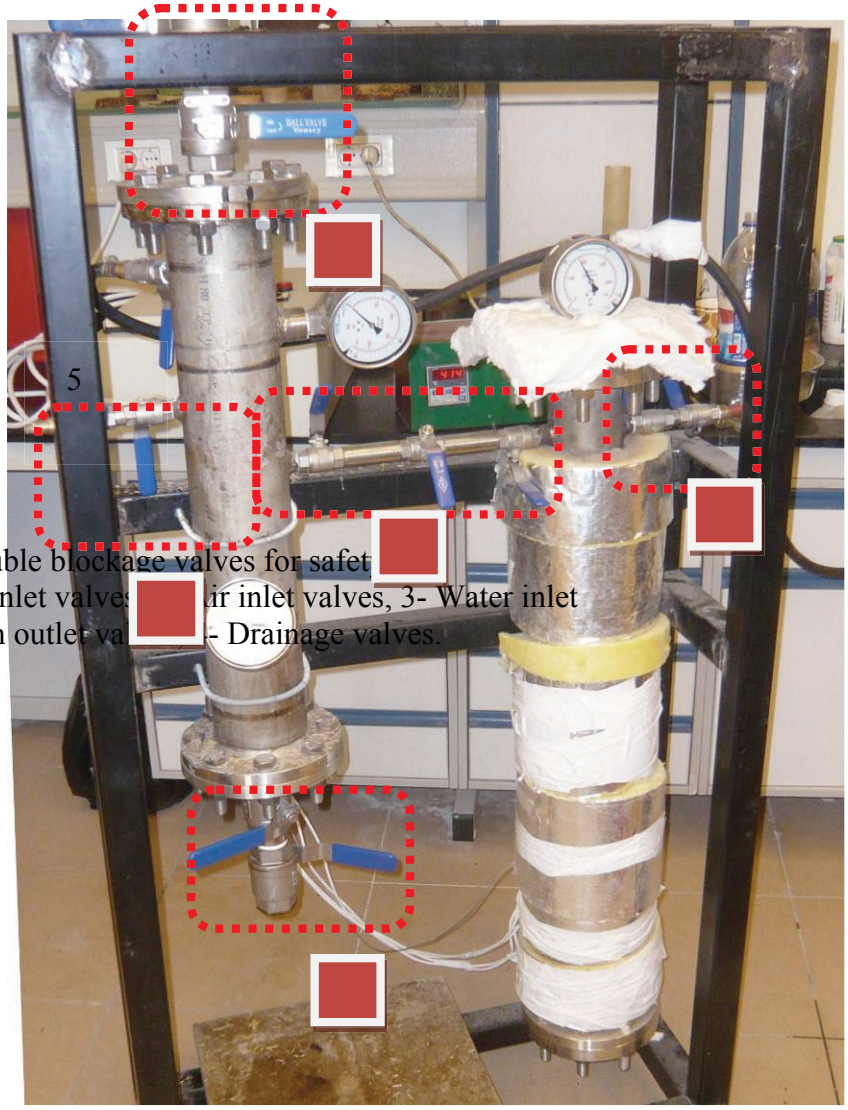
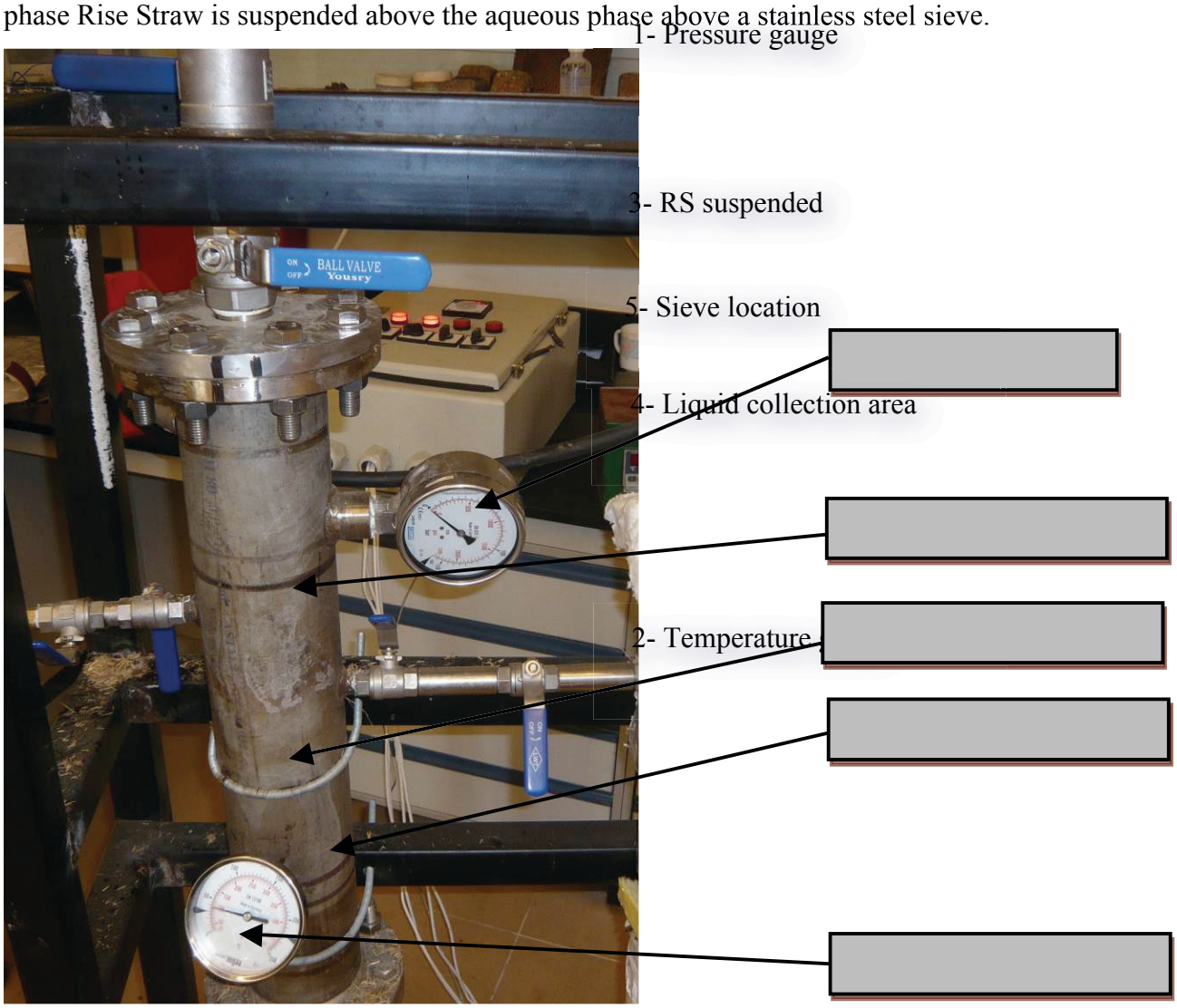


Figure 3.4 Double blockage valves for safet
Set#: 1- Steam inlet valves, 2- Air inlet valves, 3- Water inlet valves, 4- Steam outlet valves, 5- Drainage valves.

The design incorporates a pressure release valve which is connected to the main body of the reactor and connected to a high pressure hose leading to a remote drain for cooling and collection. Sudden pressure release is achievable away form the device in case of any emergency.

igure 3.5 Reactor.
-Pressure gauge, 2- Temperature gauge. 3- Rice straw suspension area,
- liquid collection area, 5- Sieve location

The reactor is equipped with a pressure gauge attached in the top side in the gaseous phase, and a thermometer attached in the bottom in the aqueous phase. The solid phase Rise Straw is suspended above the aqueous phase above a stainless steel sieve.



The design allows for pre-soaked Rice Straw charges to be loaded from the top of the reactor. The loaded rice straw remains suspended above the sieve. Figure 3.5 shows the sieve from the top of the reactor. The sieve is capable of holding the small chopped straw pieces even after being milled to small segments.

Further improvements can be made to add insulation to reduce the rate of energy consumption while still allowing for condensation to occur in order to wash away produced sugars from the rice straw to avoid scarification inhibition.

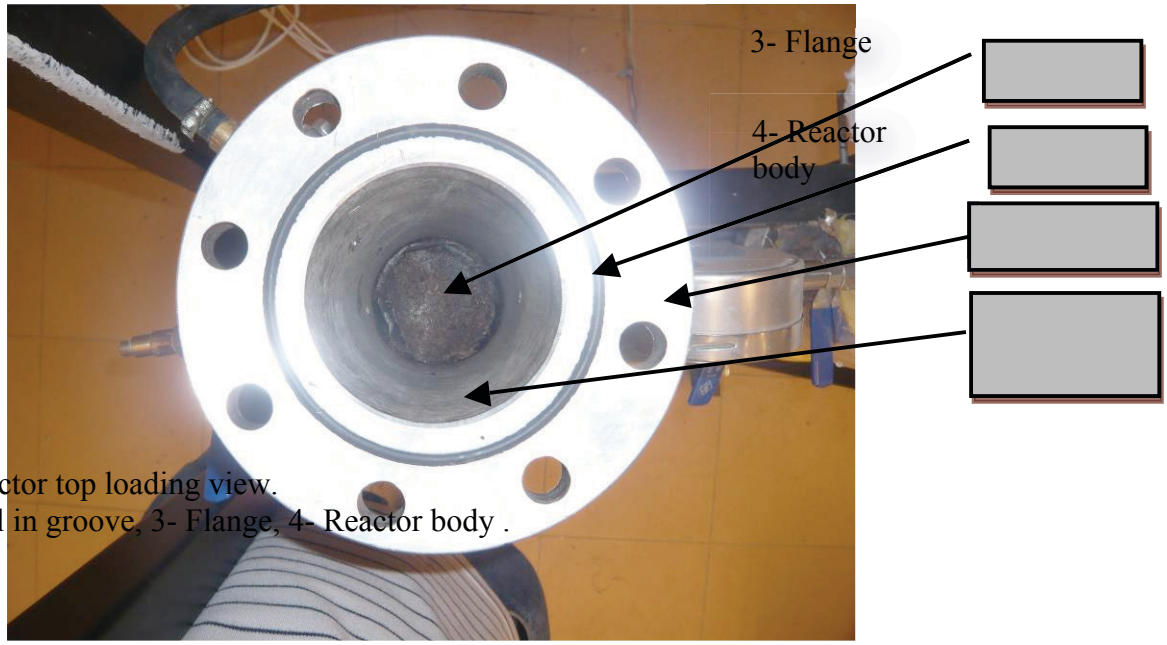


Figure 3.6 Reactor top loading view.
1- Sieve, 2- Seal in groove, 3- Flange, 4- Reactor body .

The reactor is designed to treat 400g of dry rice straw with a volume of about four liters. Due to the fact that pretreatment of the rice straw increases its weight more than 4 times after soaking in acid (up to 1700g) while the volume increase is negligible, it is possible to compact the Rice Straw into the reactor. Compaction inside the reactor does not result in fluid loss from the pretreated rice straw as it is pressed to remove excess fluids prior to introduction into the apparatus from the top till the sieve seen in figure3.6.

The apparatus design allows steam to be introduced into the reactor from the top side to allow good distribution of steam into the presoaked rice straw. At operating conditions a temperature differential develops due to condensation from top to bottom of the reactor. The apparatus design allows for condensates to develop as the main reactor body is not insulated which allows for cooling. As steam is consumed and condensates accumulate they trickle down through the rice straw and reach the sieve and subsequently reach the collection area in the bottom of the apparatus reactor body. The design allows for produced sugars to become soluble in the condensates, and quickly separate from the rice straw. The design is useful in this regards as it limits scarification inhibition to sugar production as it removes the produced sugars regularly. After treatment with steam, the design allows for the effluent to be collected from the bottom. The design accommodates the recommendation of previous works to separate the produced sugars from the rice straw to avoid scarification inhibition.

The boiler is designed with a capacity of 4L and is capable of producing steam pressure up to 70 bar. Electric heaters (3x500Watt) are used to heat up and maintain the water and steam temperature in the boiler. Figure 3.7 shows the temperature is regulated automatically via a controller connected to thermocouples and pressure is regulated manually via ball valve throttling.

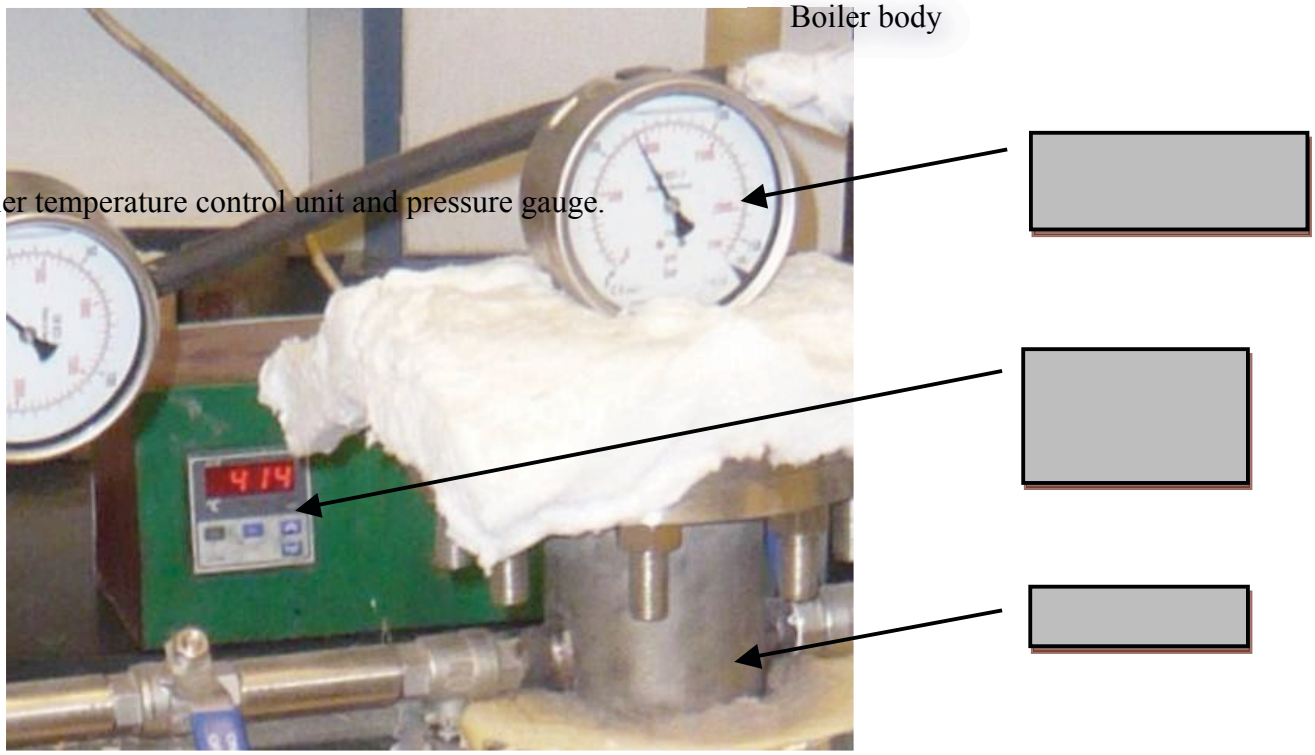
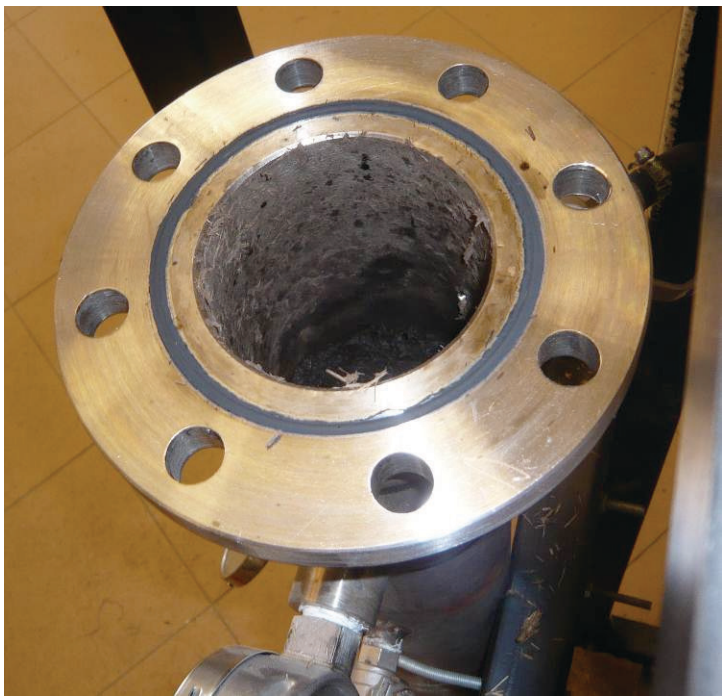


Figure 3.7 Boiler temperature control unit and pressure gauge.

A stainless steel pressure gauge is attached to the boiler to monitor the pressure. The temperature is monitored via a digital gauge. Once the pressure in the boiler reaches 65bar, the heaters are switched off and the injection process into the reactor begins. It is not recommended to exceed 65 bar pressure in the boiler as the valves can only support 70 bar safely. Even with double blockage valves it is best to operate based on the nominal operating condition for one ball valve.

3.2 Experimental Procedure:

Sample preparation starts with milled Rice Straw batches weighed to 400g. Each batch is placed in a container in preparation for acid pretreatment at different acid concentrations. The pretreatment is done for a number of hours soaking in the dilute acid concentration with duration of 24+/-2hrs. Rice straw is pre-treated with dilute acid to attack the bonds between the sugar building blocks on the molecular level. This step is important to increase the yield of sugars after steam blasting. Sulfuric acid 0.5%, 1.0%, 2.0% and 4.25% concentration were used as acid pretreatment media. The amount of dilute acid at different concentrations is 4L. After the soaking period, the excess solution is drained and the remaining acid collected. The acid from this stage was on average 2.1L. The wet remaining mixture of rice straw is then introduced into the reactor where the final stage of hydrolysis is to take place with steam blasting.



igure 3.8 Reactor Top flange unbolted to load Rice Straw Batches for treatment.

In preparation for treatment, the top flange of the reactor is unbolted and the cover is removed and a batch load of presoaked rice straw is loaded as seen in figure 3.8. The seals surfaces are checked to make sure no rice straw fibers are interfering with the sealing area and the flange is bolted tightly. Steam is suddenly introduced from the boiler until the reactor reaches the desired operating pressure.

The hydrolysis process is replicated at pressures of 3, 4, and 5 bar. The holding time in the reactor is from 30 minutes to 120 minutes. To support these experimental conditions, high temperature, high pressure steam is prepared in the 1.5kW 4 L electric boiler filled with 3-4L of water. The boiler is super heated to 400^0C and pressure is allowed to rise to 65 bar as seen in figure 3.9.

Figure 3.9 Boiler pressure after heating.

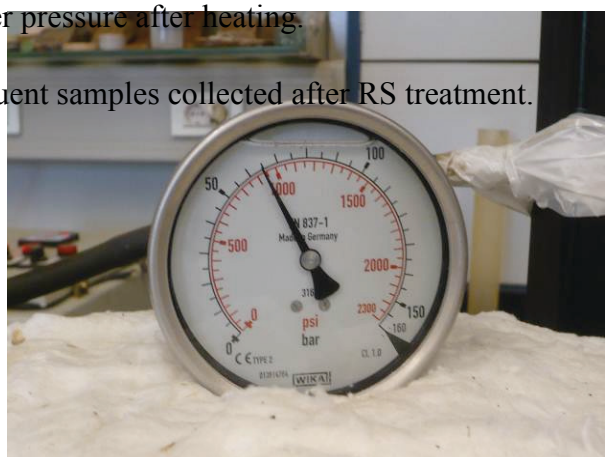


Figure 3.10 Effluent samples collected after RS treatment.

By opening a series of three ¼ inch ball valves, steam is introduced into the reactor. The pressure in the reactor is then maintained at 3, 4, and 5 bar for the rest of the reaction time. After the reaction holding time, at the respective pressures (in the reactor) the pressure is suddenly dropped from the reaction pressure to atmospheric pressure via a drainage hose. The hose is cooled to maintain its physical integrity at high temperature. After reaching atmospheric pressure in the reactor and sufficient reactor body cooling, the main flange is unbolted to release the remaining wet rice straw. The solution mixture's volume is recorded and then placed in a sealed glass bottle and allowed to cool to room temperature in a water bath as seen in figure 3.10.

Figure 3.12 Treated Rice straw - degradation and volume reduction observed

After the effluent is allowed to cool to room temperature, 40 ml samples are taken to be sent for testing as seen in figure 3.11.

The top flange is unbolted, and the solid RS remains are collected from above the sieve as seen in Figure 3.12. It is important to note that the Rice straw volume is significantly reduced after steam treatment of 30 min at 3 bar, which corresponds to the lowest testing condition.

The solid Degraded RS remains are collected and dried in an oven at 90^0C. The degraded RS solid remains are weighed periodically until no significant change in weight is obtained. It is important not to overheat the Degraded RS solid remains as some components can volatilize or catch on fire. Special care is needed in handling the dry

matter to avoid losses of fine particles that can become air bourn dust. Figure 3.13 shows Degraded RS solid remains versus dry rice straw bulk before soaking.

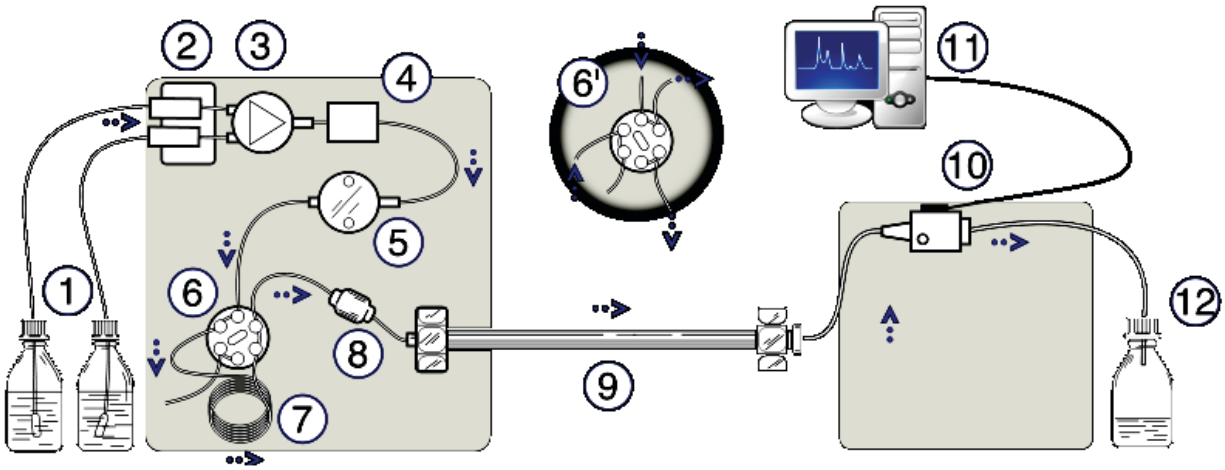
Figure 3.13 - Left Dry Rice straw before treatment.
- Right Degraded Rice Straw after treatment.

After drying, the Degraded Rice Straw solid remains samples are allowed to normalize to room temperature for 24 hrs (same condition as milled rice straw before treatment). The weights are then measured one final time and recorded.

Figure 3.14 High pressure Liquid Chromatograph (HPLC) Apparatus

3.3. Experimental Testing:

Samples testing using High Pressure Liquid Chromatography (HPLC) is the chosen method for testing for soluble sugars. The 40 ml sample solutions were prepared by precipitating the suspended solids using centrifuge at 4000rpm for 10 minutes. The soluble portion is collected and injected into the HPLC devise.



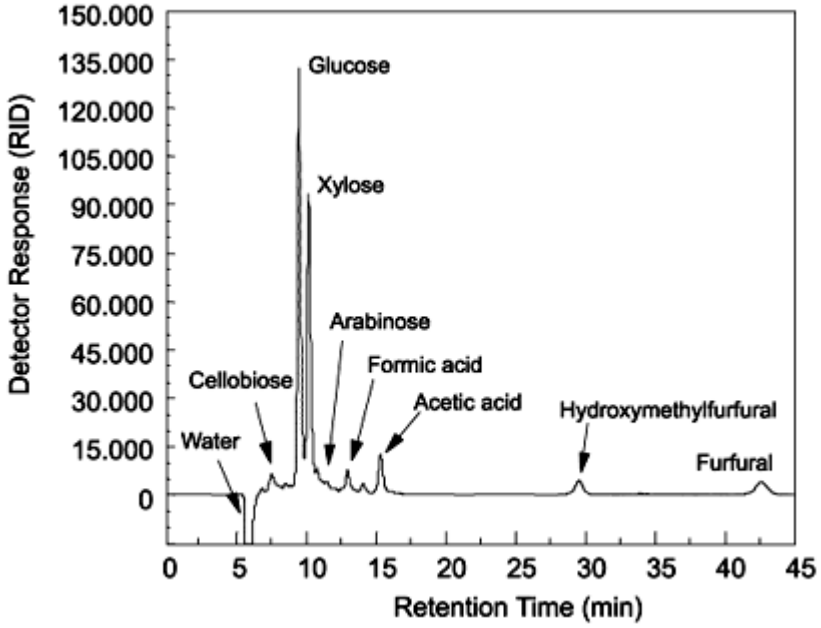
Schematic representation of an HPLC unit:
(1) Solvent reservoirs, (2) Solvent degasser, (3) Gradient valve, (4) Mixing vessel for delivery of the mobile phase, (5) High-pressure pump, (6) Switching valve in "inject position", (6') Switching valve in "load position", (7) Sample injection loop, (8) Pre-column (guard column), (9) Analytical column, (10) Detector (i.e. IR, UV), (11) Data acquisition, (12) Waste or fraction collector.

Meyer, Veronika. Practical High-performance Liquid Chromatography, 4th edition, John Wiley and Sons, 2004, ISBN 0470093781, p. 7.

HPLC utilizes a solvent (eluent) as the carrier of the sample solution (analyte). The mixture of eluent and analyte solution is heated and pressurized and forced into a long thin pipe (coiled into a loop) as seen in figure 3.14 item #7. The mixture is then introduced into the analytical column for fractionation. The eluate emerges after fractionation after the column. The eluate is passed onto a detector which reports the magnitude of the signal resulting from the amounts of material in the eluate. The signal is proportional with the concentration of the analyte components separated. The detector produces a chromatogram which is a series of peaks at different retention times depending on the size and speed of fractionation. Each standard material has a

Figure 3.15 High pressure Liquid Chromatograph (HPLC)
Chromatograph of HPLC analysis of wood hydrolysate by acid catalysed steam explosion. (Ramos, 2003)

"fingerprint" chromatogram which allows for material identification and concentration calculation. An example of detector data acquisition and representation is the detector response vs. time curve in figure 3.15. Different components of the solution travel at different rates through the long sample injection loop and the analytical column. The result is different time of appearance at the detector to provide a "response" signal vs. time. In the case of severe hydrolysis more byproducts appear such as Hydroxymethylfurfural and furfural.



CHAPTER (4)

RESULTS AND DISCUSSION:

4.1 Standard Solutions:

Samples of standard solutions of known concentrations were used to obtain the signature areas corresponding to each standard solution that is unique to that given standard solution. Standard solution signature areas are later compared to test solution areas to obtain actual concentrations in test solutions. Standard solutions were provided by the National Center for Research المركز القومي للبحوث.

Standard solutions are Glucose, Xylose, Fructose, Cellobiose and Glucuronic.

Standard Solutions, Glucose,

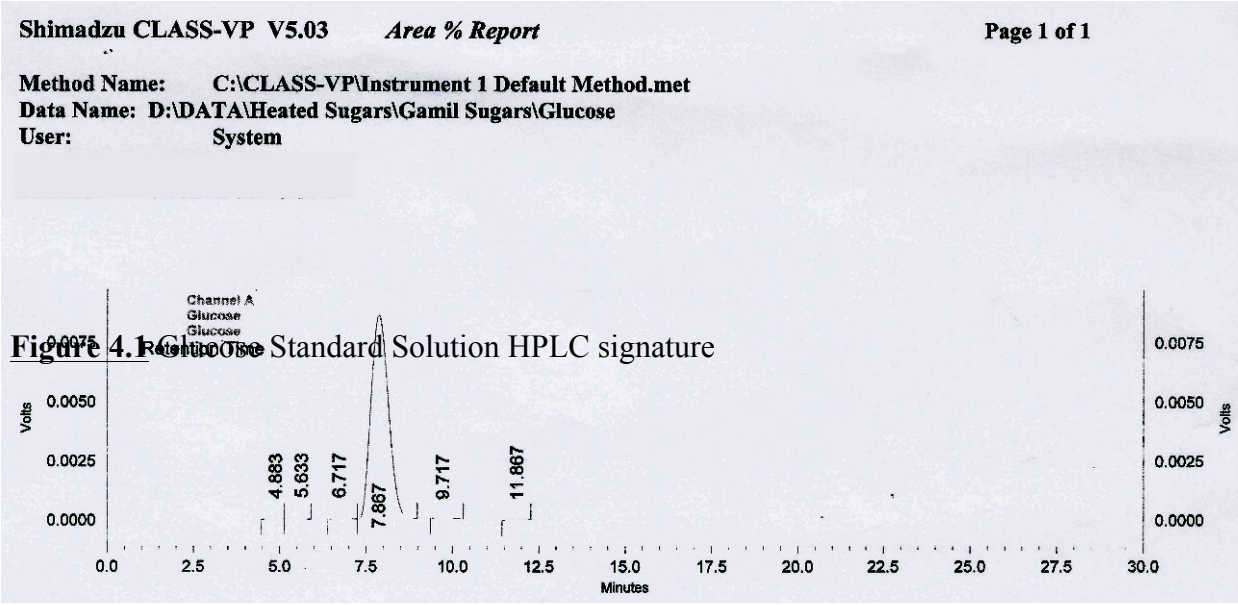


Figure 4.1 Glucose Standard Solution HPLC signature

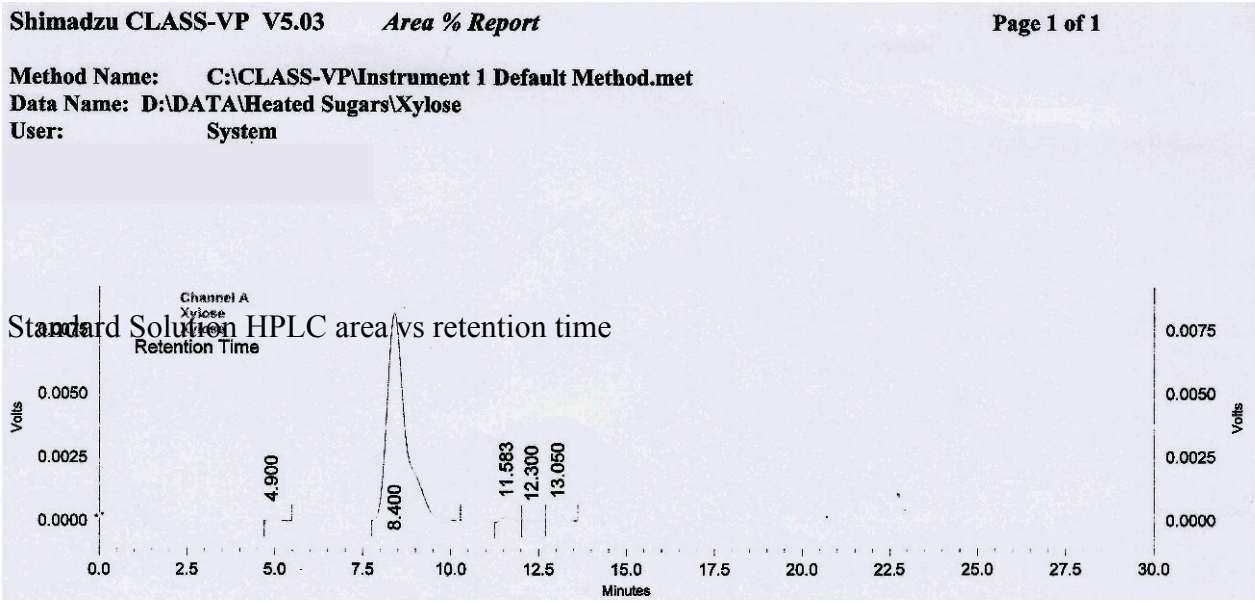
Table 4.1 Glucose Standard Solution HPLC area vs retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	4.883	2536	0.812	116	1.272
2	5.633	3235	1.035	140	1.536
3	6.717	3181	1.018	147	1.613
4	7.867	300098	96.030	8600	94.340
5	9.717	1129	0.361	38	0.417
6	11.867	2327	0.745	75	0.823
Totals		312506	100.000	9116	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.1 are for Glucose with concentration of 1 mg/mL

Standard Solution, Xylose,



able 4.2 Xylose Standard Solution HPLC area vs retention time

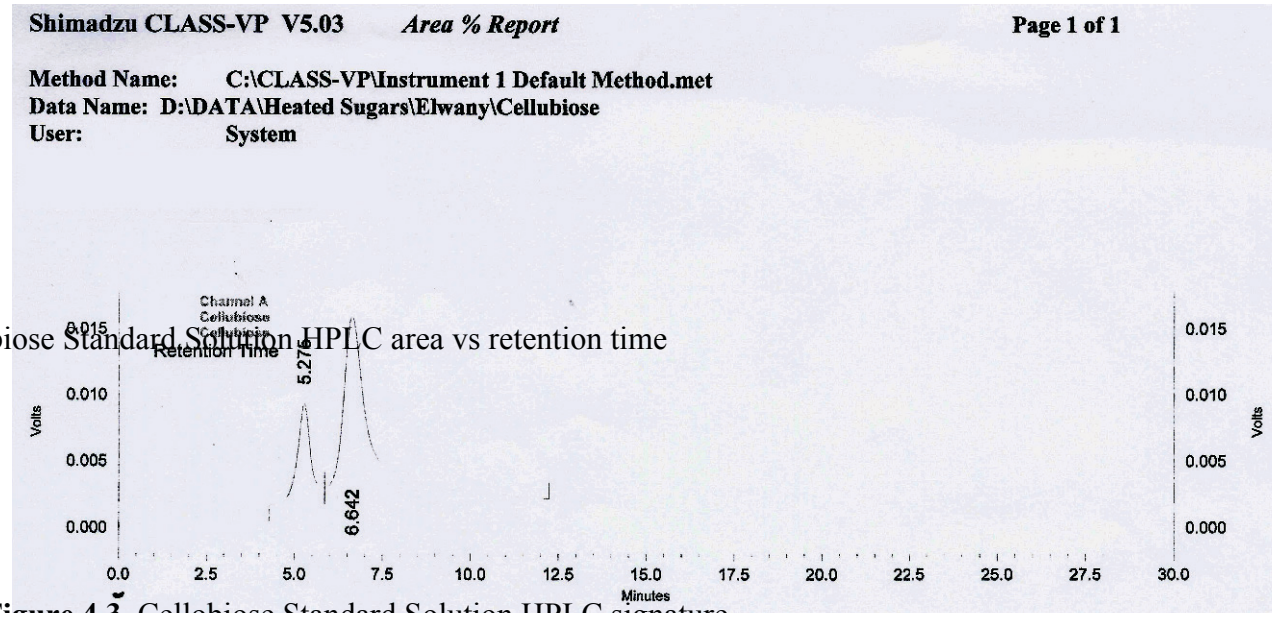
Figure 4.2 Xylose Standard Solution HPLC signature

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	4.900	964	0.306	50	0.574
2	8.400	301965	95.940	8166	93.668
3	11.583	5475	1.740	246	2.822
4	12.300	2241	0.712	82	0.941
5	13.050	4097	1.302	174	1.996
Totals		314742	100.000	8718	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.2 are for Xylose with concentration of 1 mg/mL

Standard Solutions, Cellobiose,



able 4.3 Cellobiose Standard Solution HPLC area vs retention time

Figure 4.3 Cellobiose Standard Solution HPLC signature

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.275	211383	19.390	7603	35.048
2	6.642	878789	80.610	14090	64.952
Totals		1090172	100.000	21693	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.3 are for Cellobiose with concentration of 2 mg/mL

Standard solutions,Glucoronic

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**

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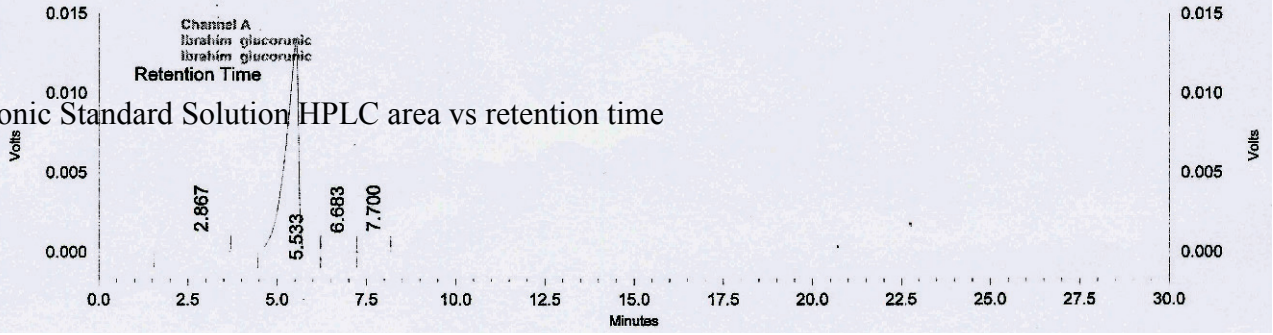


Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	2.867	6124	1.800	82	0.603
2	5.533	329169	96.770	13355	98.220
3	6.683	1701	0.500	47	0.346
4	7.700	3163	0.930	113	0.831
Totals		340157	100.000	13597	100.000

Table 4.4 Glucoronic Standard Solution HPLC area vs retention time

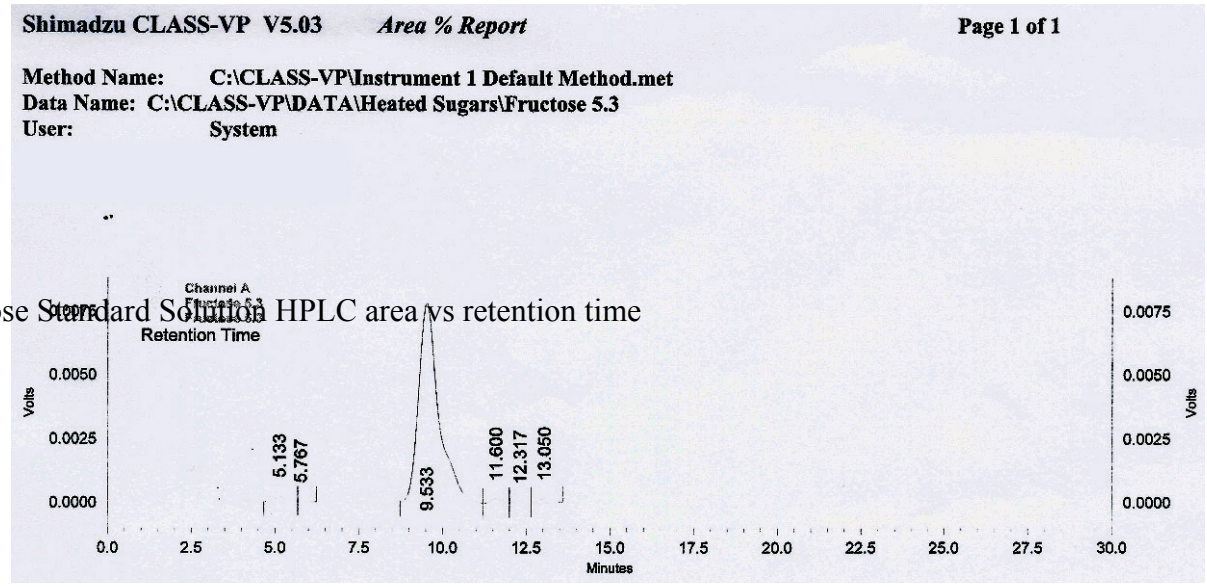
Figure 4.4 Glucuronic Standard Solution HPLC signature

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.4 are for Glucuronic acid with concentration of 1 mg/mL.

Note:

Glucuronic acid is a carboxylic acid resulting from the oxidization of glucose. Its structure is similar to glucose with a sixth carbon attached to a carboxylic group (COOH) instead of an (OH) group. Glucuronic acid is soluble in water.

Standard solutions, Fructose,



'able 4.5 Fructose Standard Solution HPLC area vs retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.133	5553	1.609	259	3.032
2	5.767	632	0.183	44	0.515
3	9.533	329682	95.498	7832	91.677
4	11.600	4194	1.215	190	2.224
5	12.317	1394	0.404	56	0.656
6	13.050	3769	1.092	162	1.896
Totals		345224	100.000	8543	100.000

Figure 4.5 Fructose Standard Solution HPLC signature

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.5 are for Fructose with concentration of 2 mg/mL.

Figure 4.6 The effect of: 3bar, 30min, 0.5% H2SO4 HPLC

4.2. Different parameters to be investigated.

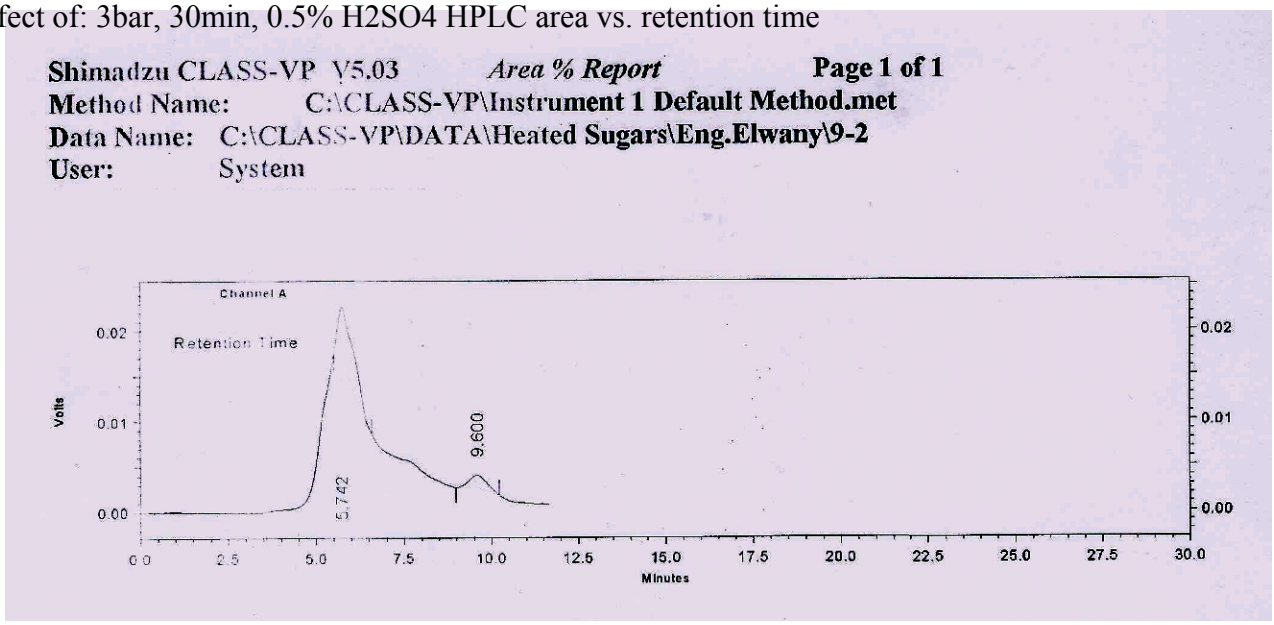
The hydrolysis process has different a) pretreatment and b) treatment parameters to investigate their effect on sugars production:

1- Pretreatment acid concentrations of 0.5%, 1.0%, 2.0% and 4.25% sulfuric acid.
2- Treatment pressures of 3bar, 4bar, and 5bar.
3- Treatment retention times of 30 minutes to 120 minutes.

4.2.1. The effect of pretreatment with 0.5% Sulfuric at 3bar.

3 bar 30 min, 0.5% Sulfuric

Table 4.6 The effect of: 3bar, 30min, 0.5% H2SO4 HPLC area vs. retention time



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.742	235698	78.534	7187	80.383
2	9.600	64424	21.466	1754	19.617
Totals		300122	100.000	8941	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.6 are shown in table 4.6 which corresponds to:

0.6g glucuronic acid

0.4g fructose

The resulting total soluble sugar equivalent is 1.0g of sugars produced.

3 bar 90 min, 0.5% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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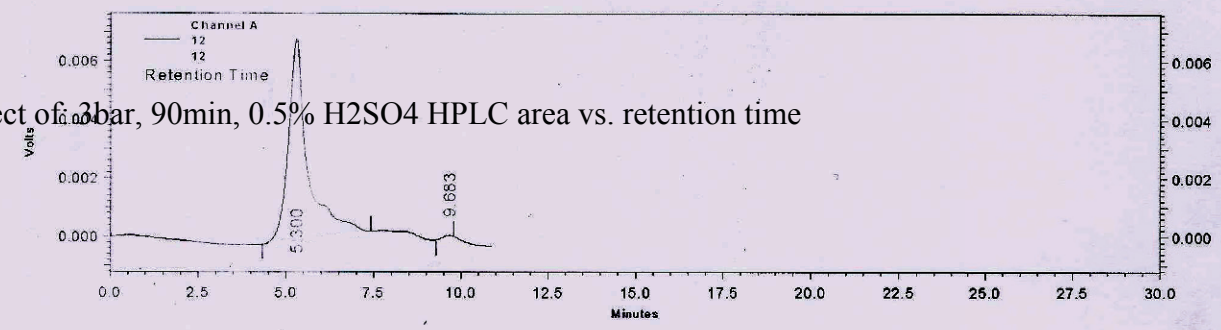

Table 4.7 The effect of 3bar, 90min, 0.5% H2SO4 HPLC area vs. retention time

Figure 4.7 The effect of: 3bar, 90min, 0.5% H2SO4 HPLC

Detection A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.300	266978	99.568	6876	99.135
2	9.683	1158	0.432	60	0.865
Totals		**268136**	**100.000**	**6936**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.7 are shown in table 4.7 which corresponds to:

1.1g glucuronic acid

0.01g fructose

The resulting total soluble sugar equivalent is 1.11g of sugars produced.

3 bar 120 min, 0.5% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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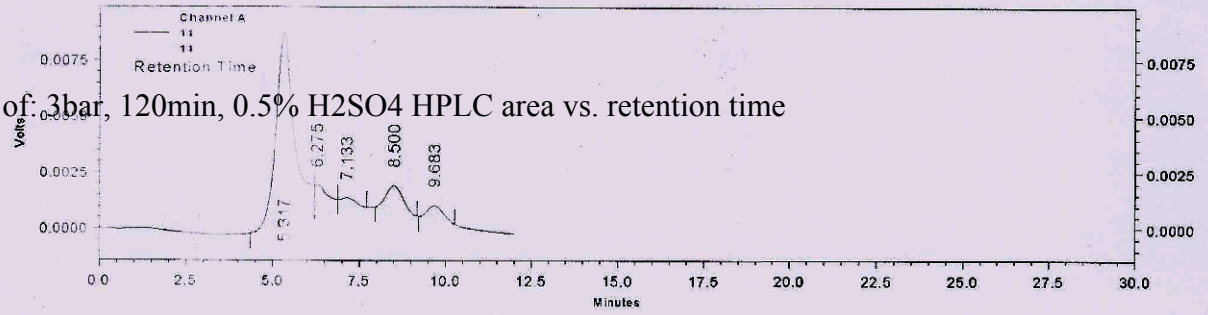

Figure 4.8 The effect of: 3bar, 120min, 0.5% H2SO4 HPLC

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.317	324478	73.403	8645	69.343
2	6.275	41850	9.467	1473	11.815
3	7.133	18975	4.293	593	4.757
4	8.500	37429	8.467	1125	9.024
5	9.683	19317	4.370	631	5.061
Totals		442049	100.000	12467	100.000

Table 4.8 The effect of: 3bar, 120min, 0.5% H2SO4 HPLC area vs. retention time

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.8 are shown in table 4.8 which corresponds to:

1.5g glucuronic acid

0.6g cellobiose

0.1g glucose

0.2g xylose

0.2g fructose

The resulting total soluble sugar equivalent is 2.6g of sugars produced.

4.2.2. The effect of pretreatment with 0.5% Sulfuric at 4bar.

4 bar 30 min, 0.5% Sulfuric

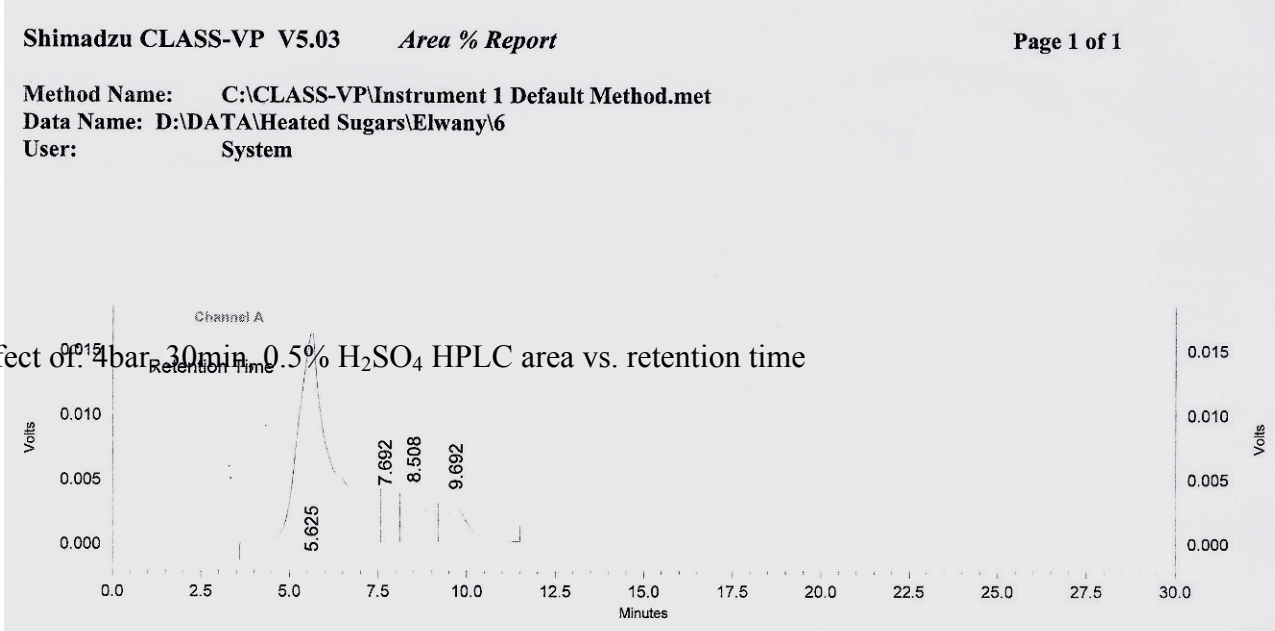

Figure 4.9 The effect of: 4bar, 30min, 0.5% H_2SO_4 HPLC

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.625	1107638	73.700	16335	65.051
2	7.692	90793	6.041	2959	11.784
3	8.508	171443	11.407	3192	12.712
4	9.692	133024	8.851	2625	10.454
Totals		1502898	100.000	25111	100.000

able 4.9 The effect of: 4bar, 30min, 0.5% H_2SO_4 HPLC area vs. retention time

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.9 are shown in table 4.9 which corresponds to:

4.0g glucuronic acid

0.0g cellobiose

0.4g glucose

0.7g xylose

1.0g fructose

The resulting total soluble sugar equivalent is 6.0g of sugars produced.

4 bar 60 min, 0.5% Sulfuric

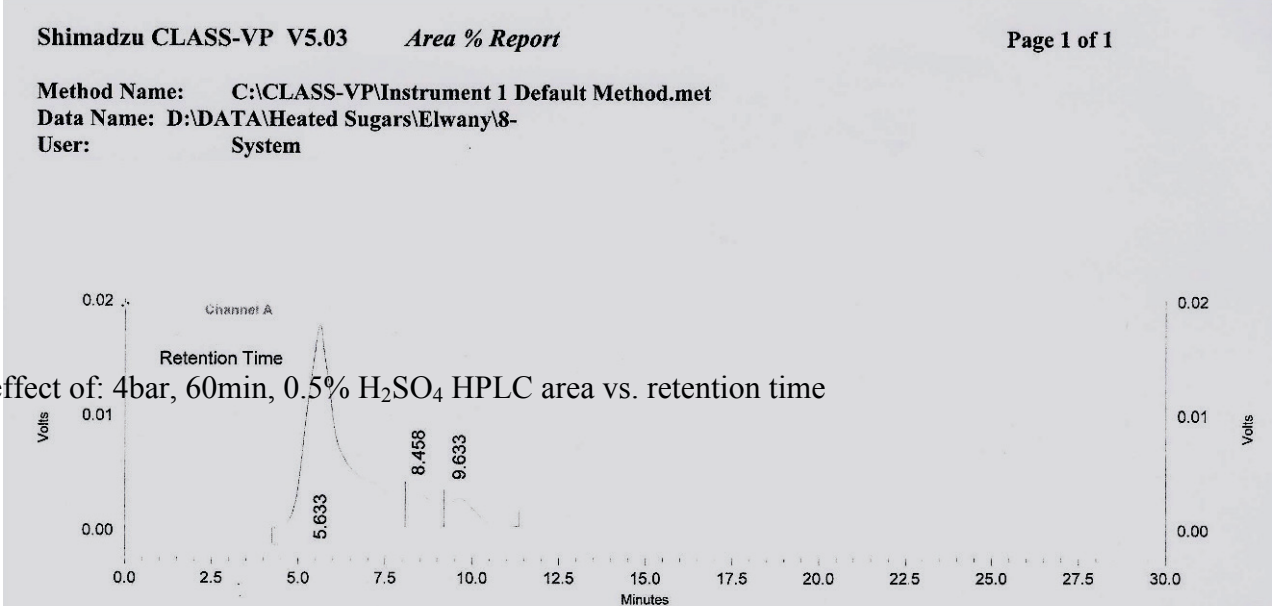

Figure 4.10 The effect of: 4bar, 60min, 0.5% H_2SO_4 HPLC

Table 4.10 The effect of: 4bar, 60min, 0.5% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.633	1307527	80.722	17708	76.552
2	8.458	164208	10.135	2918	12.615
3	9.633	138405	8.543	2506	10.833
Totals		1620140	100.000	23132	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.10 are shown in table 4.10 which corresponds to:

7.0g glucuronic acid

0.0g cellobiose

0.0g glucose

0.9g xylose

1.5g fructose

The resulting total soluble sugar equivalent is 9.4g of sugars produced.

4 bar 90 min, 0.5% Sulfuric

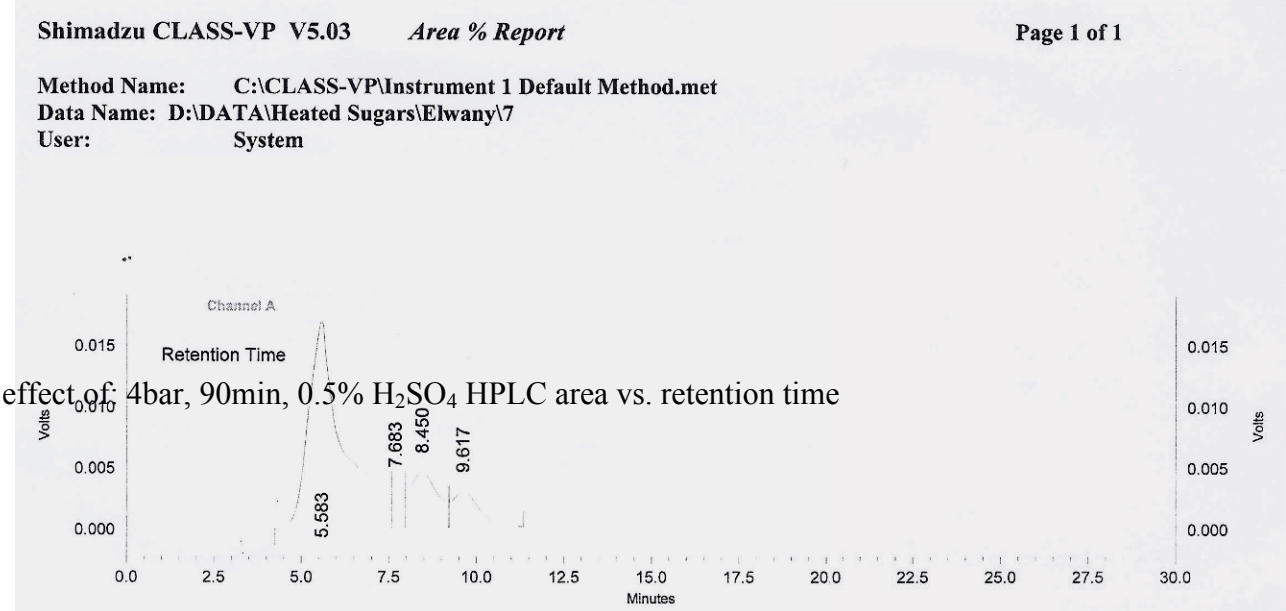

Table 4.11 The effect of: 4bar, 90min, 0.5% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.583	1193539	71.096	16998	60.955
2	7.683	78315	4.665	3409	12.225
3	8.450	254267	15.146	4546	16.302
4	9.617	152651	9.093	2933	10.518
Totals		1678772	100.000	27886	100.000

Figure 4.11 The effect of: 4bar, 90min, 0.5% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.11 are shown in table 4.11 which corresponds to:

6.3g glucuronic acid

0.0g cellobiose

0.5g glucose

1.5g xylose

1.6g fructose

The resulting total soluble sugar equivalent is 9.8g of sugars produced.

Figure 4.12 The effect of: 4bar, 120min, 0.5% H_2SO_4 HPLC

4 bar 120 min, 0.5% Sulfuric

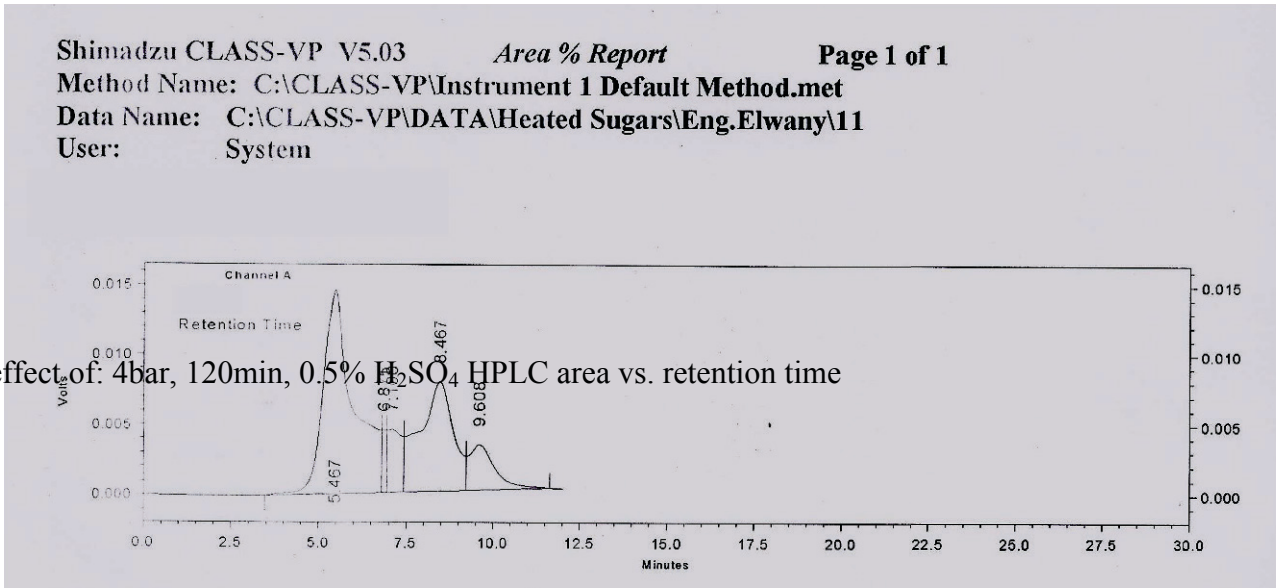

Table 4.12 The effect of: 4bar, 120min, 0.5% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.467	884852	50.788	14589	42.192
2	6.875	40069	2.300	4463	12.907
3	7.100	124255	7.132	4508	13.037
4	8.467	527003	30.249	7787	22.520
5	9.608	166059	9.531	3231	9.344
Totals		**1742238**	**100.000**	**34578**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.12 are shown in table 4.12 which corresponds to:

4.5g glucuronic acid

0.6g cellobiose

0.7g glucose

2.9g xylose

1.7g fructose

The resulting total soluble sugar equivalent is 10.5g of sugars produced.

4.2.3. The effect of pretreatment with 0.5% Sulfuric at 5bar.

5 bar 45 min, 0.5% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* Page 1 of 1

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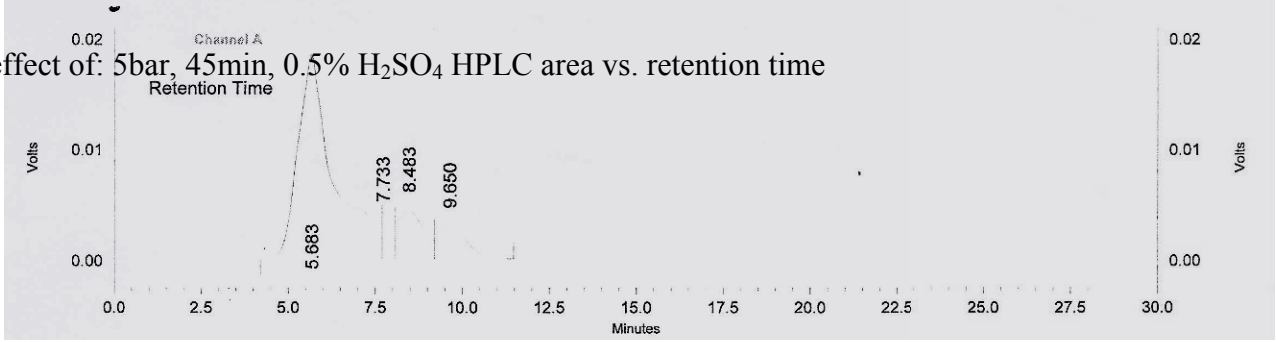

Figure 4.13 The effect of: 5bar, 45min, 0.5% H_2SO_4 HPLC

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.683	1343874	73.865	18572	63.129
2	7.733	75431	4.146	3518	11.958
3	8.483	231328	12.715	4380	14.888
4	9.650	168737	9.274	2949	10.024
Totals		1819370	100.000	29419	100.000

Table 4.13 The effect of: 5bar, 45min, 0.5% H_2SO_4 HPLC area vs. retention time

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.13 are shown in table 4.13 which corresponds to:

6.0g glucuronic acid

0.0g cellobiose

0.4g glucose

1.1g xylose

1.5g fructose

The resulting total soluble sugar equivalent is 9.1 g of sugars produced.

5 bar 60 min, 0.5% Sulfuric

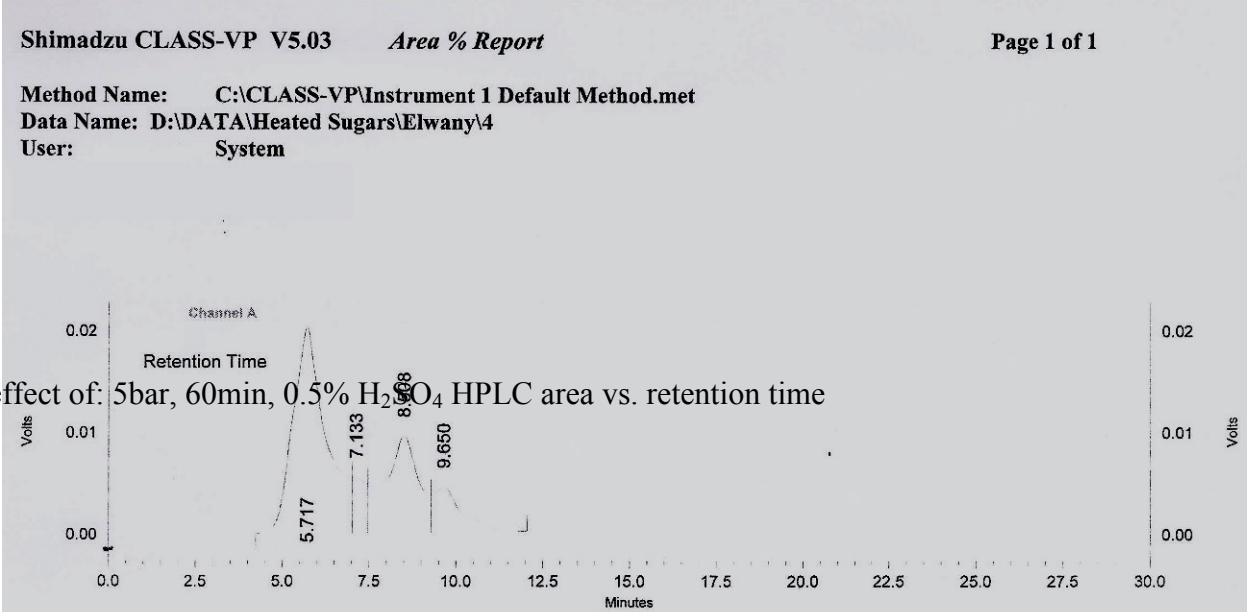

Table 4.14 The effect of: 5bar, 60min, 0.5% H_2SO_4 HPLC area vs. retention time

Figure 4.14 The effect of: 5bar, 60min, 0.5% H_2SO_4 HPLC

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.717	1342268	56.043	20182	50.907
2	7.133	142937	5.968	5528	13.944
3	8.508	657472	27.451	9488	23.932
4	9.650	252393	10.538	4447	11.217
Totals		2395070	100.000	39645	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.14 are shown in table 4.14 which corresponds to:

6.7g glucuronic acid

0.0g cellobiose

0.8g glucose

3.6g xylose

2.5g fructose

The resulting total soluble sugar equivalent is 13.6 g of sugars produced.

Figure 4.15 The effect of: 5bar, 90min, 0.5% H_2SO_4 HPLC

5 bar 90 min, 0.5% Sulfuric

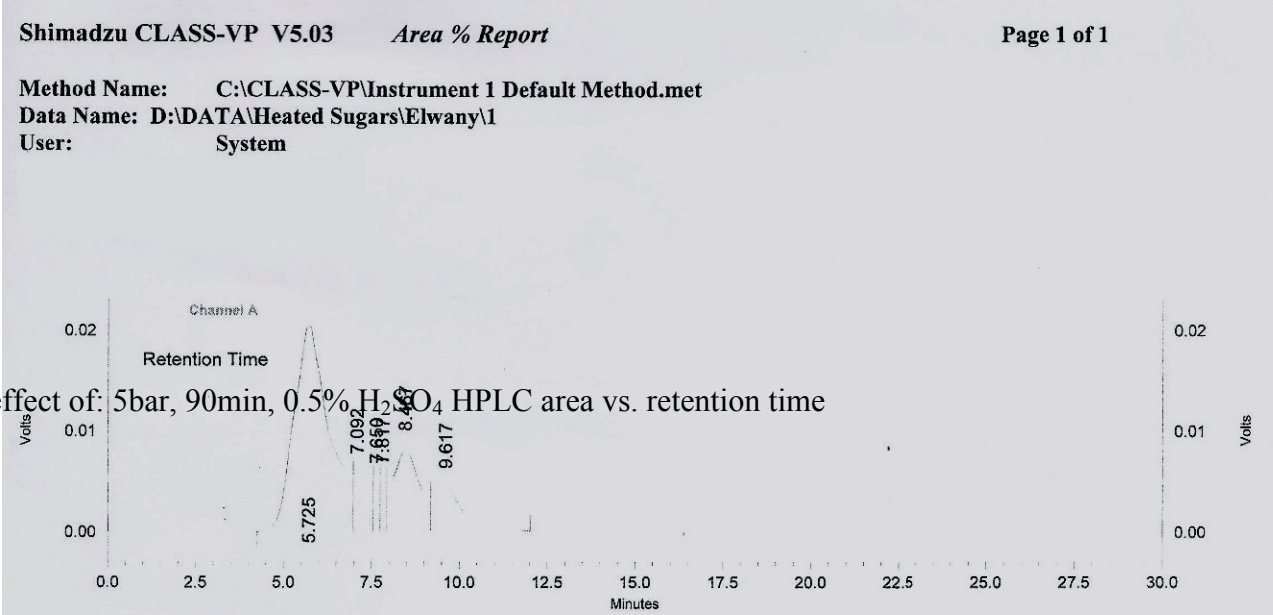

Table 4.15 The effect of: 5bar, 90min, 0.5% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.725	1423182	58.903	20483	42.185
2	7.092	182264	7.544	5822	11.991
3	7.650	56220	2.327	4902	10.096
4	7.817	56075	2.321	4894	10.079
5	8.467	434690	17.991	8092	16.666
6	9.617	263729	10.915	4362	8.984
Totals		2416160	100.000	48555	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.15 are shown in table 4.15 which corresponds to:

9.1g glucuronic acid

3.6g cellobiose

0.8g glucose

3.0g xylose

3.4g fructose

The resulting total soluble sugar equivalent is 19.9 g of sugars produced.

5 bar 120 min, 0.5% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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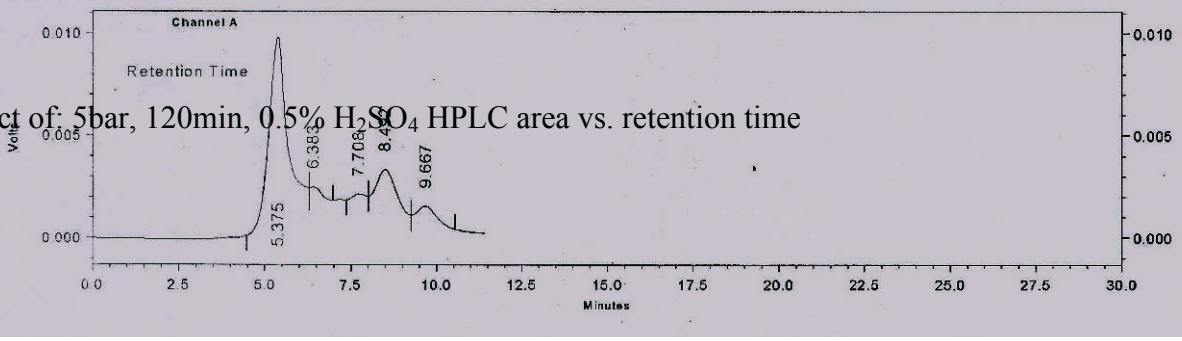

Figure 4.16 The effect of: 5bar, 120min, 0.5% H_2SO_4 HPLC

Table 4.16 The effect of: 5bar, 120min, 0.5% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.375	337898	70.565	9110	68.332
2	6.383	23555	4.927	1049	7.868
3	7.708	12907	2.700	449	3.368
4	8.492	77678	16.249	1984	14.881
5	9.667	26096	5.459	740	5.551
Totals		478054	100.000	13332	100.000

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.16 are shown in table 4.16 which corresponds to:

2.2g glucuronic acid

0.5g cellobiose

0.1g glucose

0.6g xylose

0.3g fructose

The resulting total soluble sugar equivalent is 3.7g of sugars produced.

4.2.4. The effect of pretreatment with 1.0% Sulfuric at 3bar.

3 bar 30 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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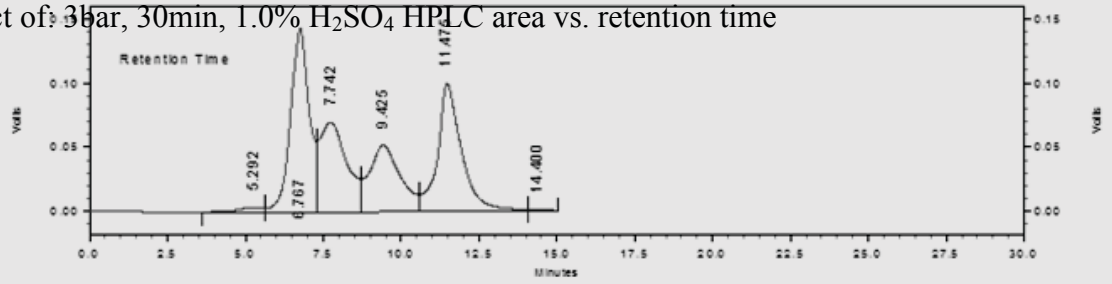
User: System



able 4.17 The effect of: 3bar, 30min, 1.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.292	188776	0.981	3687	1.003
2	6.767	5953941	30.942	142091	38.672
3	7.742	4185220	21.750	69535	18.925
4	9.425	3575214	18.580	51622	14.050
5	11.475	5290183	27.493	99266	27.017
6	14.400	48715	0.253	1225	0.333
Totals		**19242049**	**100.000**	**367426**	**100.000**

Figure 4.17 The effect of: 3bar, 30min, 1.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.17 are shown in table 4.17 which corresponds to:

0.7g glucuronic acid

73.2g cellobiose

18.1g glucose

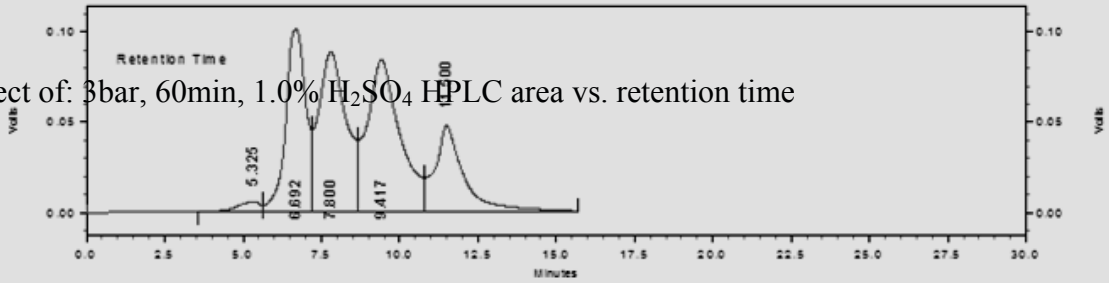
0.0g xylose

28.2g fructose

The resulting total soluble sugar equivalent is 120.3g of sugars produced.

3 bar 60 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
Method Name: C:\CLASS-VP\Instrument 1 Default Method.met
Data Name: D:\DATA\Heated Sugars\EL-11-11
User: System



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.325	291573	1.497	5581	1.708
2	6.692	4527588	23.251	101204	30.973
3	7.800	5445564	27.965	88374	27.047
4	9.417	6175853	31.716	84080	25.732
5	11.500	3031885	15.570	47508	14.540
Totals		**19472463**	**100.000**	**326747**	**100.000**

Table 4.18 The effect of: 3bar, 60min, 1.0% H_2SO_4 HPLC area vs. retention time

Figure 4.18 The effect of: 3bar, 60min, 1.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.18 are shown in table 4.18 which corresponds to:

1.4g glucuronic acid

67.7g cellobiose

28.7g glucose

0.0g xylose

59.2g fructose

The resulting total soluble sugar equivalent is 156.9g of sugars produced.

3 bar 90 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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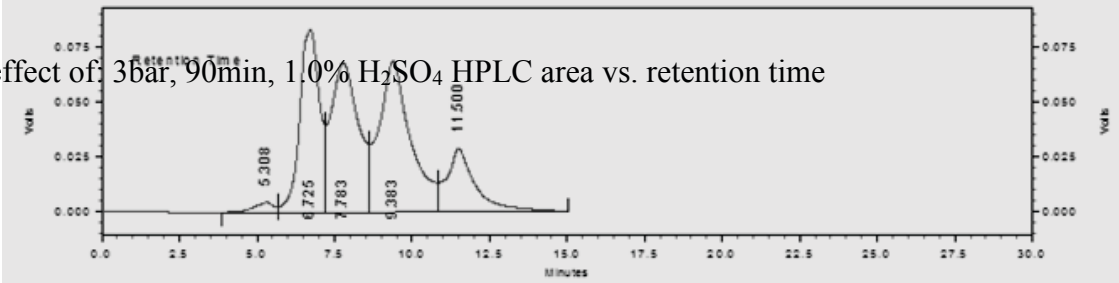
Data Name: D:\DATA\Heated Sugars\EL-9-11
User: System



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.308	202137	1.357	4411	1.746
2	6.725	3743486	25.122	82952	32.830
3	7.783	4210841	28.258	67946	26.891
4	9.383	4871833	32.694	68652	27.170
5	11.500	1872919	12.569	28713	11.364
Totals		**14901216**	**100.000**	**252674**	**100.000**

Table 4.19 The effect of: 3bar, 90min, 1.0% H_2SO_4 HPLC area vs. retention time

Figure 4.19 The effect of: 3bar, 90min, 1.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.19 are shown in table 4.19 which corresponds to:

0.7g glucuronic acid

50.6g cellobiose

15.8g glucose

0.0g xylose

30.4g fructose

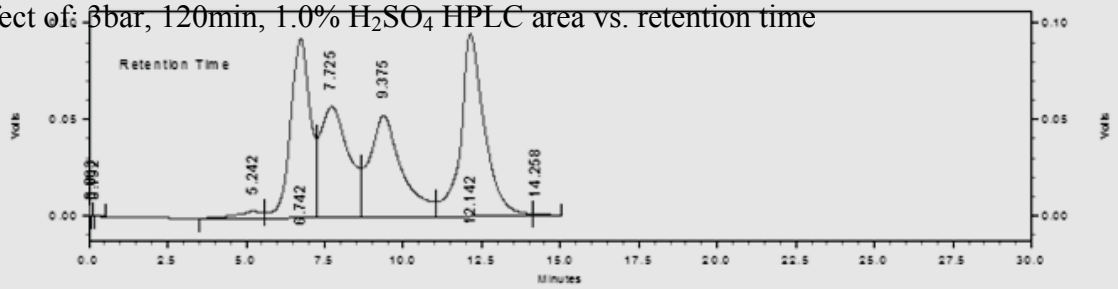
The resulting total soluble sugar equivalent is 97.6g of sugars produced.

Figure 4.20 The effect of: 3bar, 120min, 1.0% H_2SO_4 HPLC

3 bar 120 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
Method Name: C:\CLASS-VP\Instrument 1 Default Method.met
Data Name: D:\DATA\Heated Sugars\EL-10-11
User: System

Table 420 The effect of: 3bar, 120min, 1.0% H_2SO_4 HPLC area vs. retention time



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	0.083	47	0.000	18	0.006
2	0.192	270	0.002	29	0.010
3	5.242	180463	1.078	3517	1.161
4	6.742	4054482	24.231	92722	30.618
5	7.725	3583776	21.418	57510	18.990
6	9.375	3801108	22.716	52884	17.463
7	12.142	5064421	30.266	94723	31.279
8	14.258	48319	0.289	1434	0.474
Totals		**16732886**	**100.000**	**302837**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.20 are shown in table 4.20 which corresponds to:

1.0g glucuronic acid

60.2g cellobiose

23.9g glucose

0.0g xylose

50.2g fructose

The resulting total soluble sugar equivalent is 135.4g of sugars produced.

Figure 4.21 The effect of: 4bar, 30min, 1.0% H_2SO_4 HPLC

4.2.5. The effect of pretreatment with 1.0% Sulfuric at 4bar.

4 bar 30 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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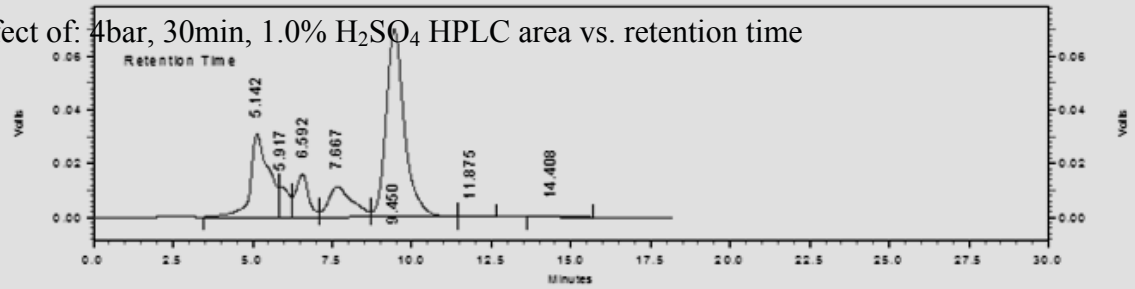
Data Name: D:\CLASS-VP\DATA\Heated Sugars\EL-8-11
User: System



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.142	1243744	23.068	30850	22.066
2	5.917	253463	4.701	11293	8.077
3	6.592	444063	8.236	15691	11.223
4	7.667	585474	10.859	11382	8.141
5	9.450	2829925	52.488	69831	49.947
6	11.875	8269	0.153	216	0.154
7	14.408	26642	0.494	547	0.391
Totals		**5391580**	**100.000**	**139810**	**100.000**

Table 4.21 The effect of: 4bar, 30min, 1.0% H_2SO_4 HPLC area vs. retention time

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.21 are shown in table 4.21 which corresponds to:

5.5g glucuronic acid

5.2g cellobiose

2.4g glucose

0.0g xylose

21.1g fructose

The resulting total soluble sugar equivalent is 34.4g of sugars produced.

Figure 4.22 The effect of: 4bar, 60min, 1.0% H_2SO_4 HPLC

4 bar 60 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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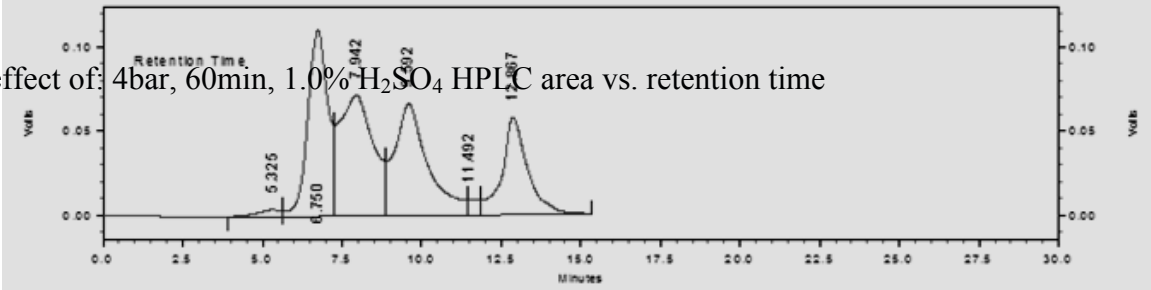
Data Name: D:\DATA\Heated Sugars\EL-7-11
User: System



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.325	191556	1.021	4007	1.252
2	6.750	4890927	26.058	110597	34.564
3	7.942	5282881	28.146	71730	22.417
4	9.592	4885906	26.031	66578	20.807
5	11.492	207915	1.108	9262	2.895
6	12.867	3310444	17.637	57801	18.064
Totals		**18769629**	**100.000**	**319975**	**100.000**

Table 4.22 The effect of: 4bar, 60min, 1.0% H_2SO_4 HPLC area vs. retention time

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.22 are shown in table 4.22 which corresponds to:

0.9g glucuronic acid

68.0g cellobiose

25.9g glucose

0.0g xylose

43.6g fructose

The resulting total soluble sugar equivalent is 138.3g of sugars produced.

Figure 4.23 The effect of: 4bar, 90min, 1.0% H_2SO_4 HPLC

4 bar 90 min, 1.0% Sulfuric

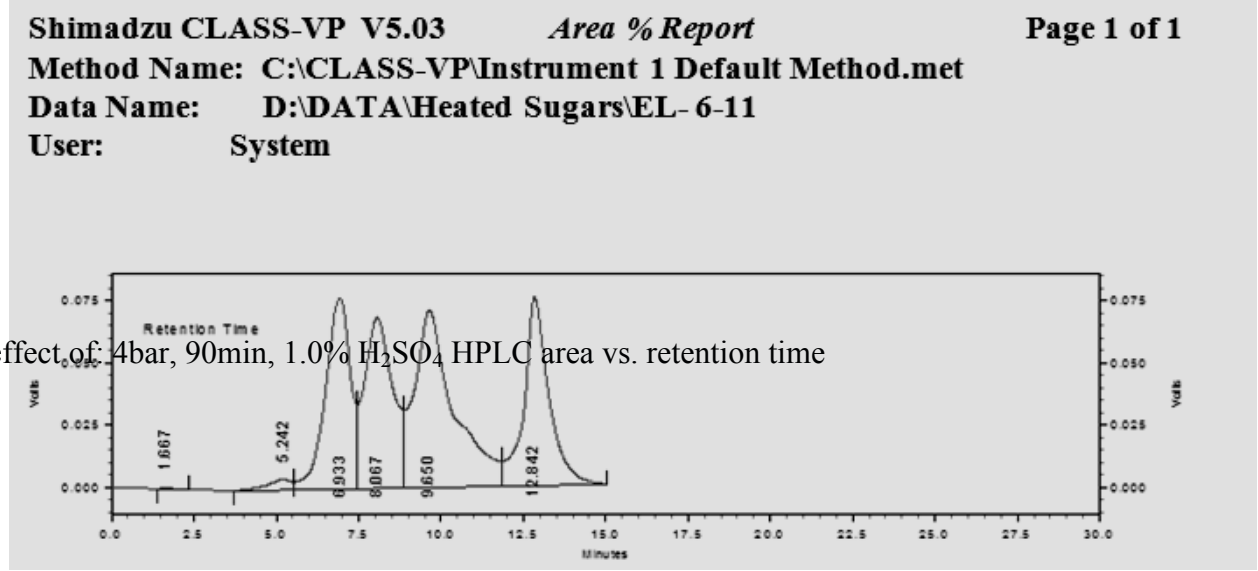


Table 4.23 The effect of: 4bar, 90min, 1.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	1.667	9181	0.050	340	0.115
2	5.242	198012	1.080	4024	1.359
3	6.933	4066559	22.189	76463	25.815
4	8.067	4103111	22.388	68578	23.153
5	9.650	5919757	32.301	71305	24.073
6	12.842	4030358	21.991	75488	25.486
Totals		**18326978**	**100.000**	**296198**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.23 are shown in table 4.23 which corresponds to:

1.0g glucuronic acid

65.4g cellobiose

23.2g glucose

0.0g xylose

61.1g fructose

The resulting total soluble sugar equivalent is 150.7g of sugars produced.

Figure 4.24 The effect of: 4bar, 120min, 1.0% H_2SO_4 HPLC

4 bar 120 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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User: System

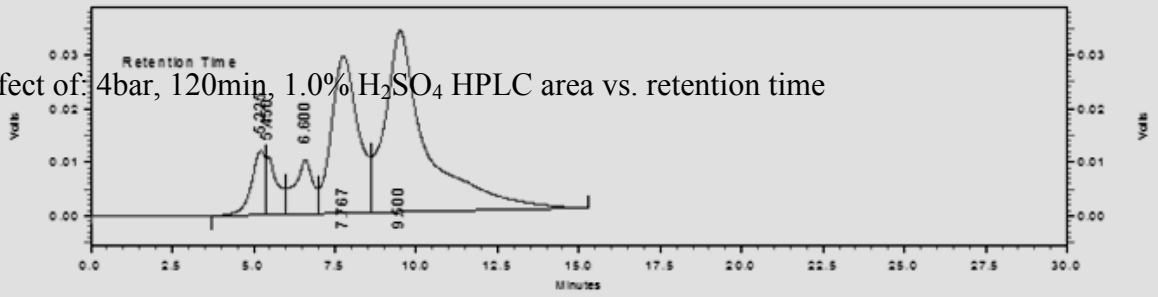

Table 4.24 The effect of: 4bar, 120min, 1.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.225	348497	6.058	11946	12.490
2	5.450	260812	4.534	10717	11.205
3	6.600	398174	6.922	10006	10.462
4	7.767	1669289	29.020	29218	30.550
5	9.500	3075511	53.466	33754	35.292
Totals		**5752283**	**100.000**	**95641**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.24 are shown in table 4.24 which corresponds to:

3.7g glucuronic acid

7.4g cellobiose

11.0g glucose

0.0g xylose

36.8g fructose

The resulting total soluble sugar equivalent is 58.8g of sugars produced.

Figure 4.25 The effect of: 5bar, 30min, 1.0% H_2SO_4 HPLC

4.2.6. The effect of pretreatment with 1.0% Sulfuric at 5bar.

5 bar 30 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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User: System

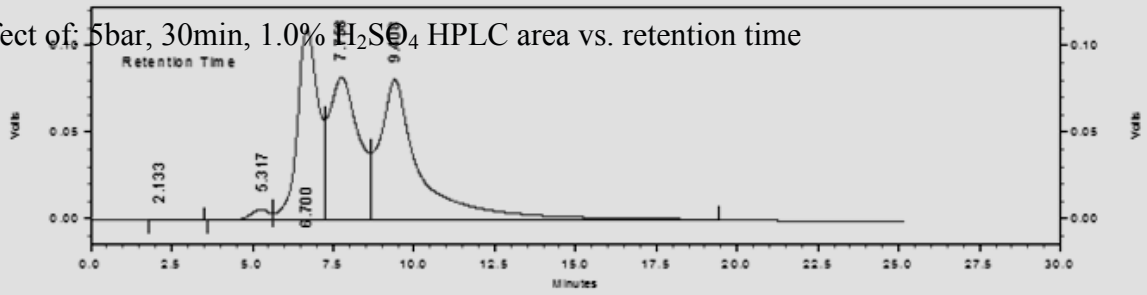


Table 4.25 The effect of: 5bar, 30min, 1.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	2.133	8460	0.047	166	0.059
2	5.317	257023	1.432	6414	2.291
3	6.700	5006055	27.891	109482	39.107
4	7.758	5286408	29.453	82706	29.542
5	9.408	7390969	41.178	81188	29.000
Totals		**17948915**	**100.000**	**279956**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.25 are shown in table 4.25 which corresponds to:

1.0g glucuronic acid

62.5g cellobiose

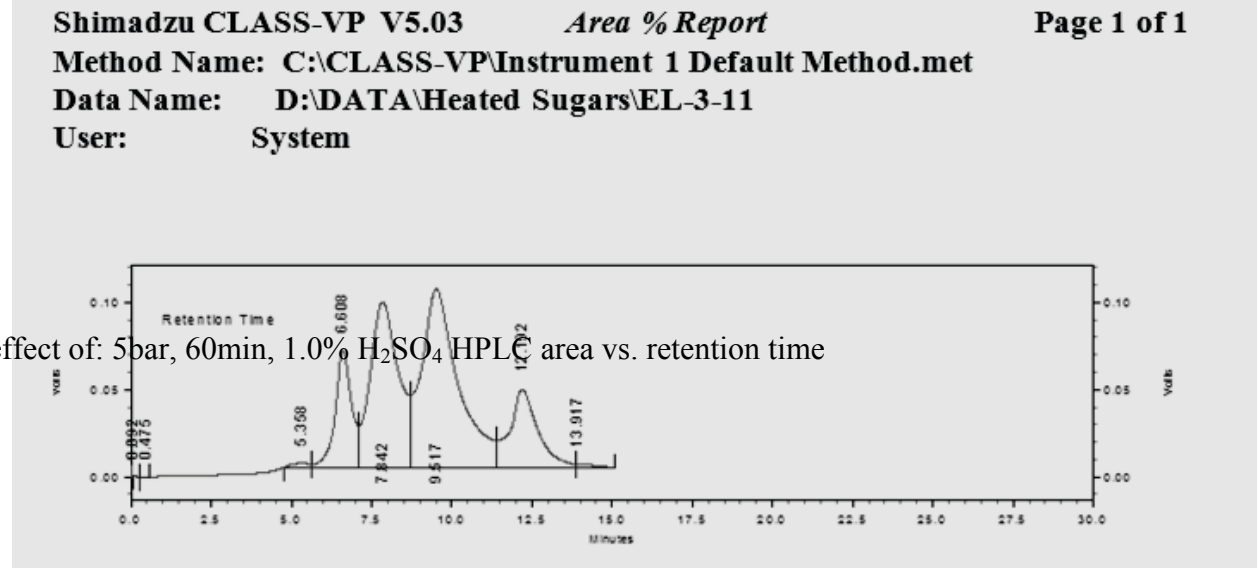
23.3g glucose

0.0g xylose

59.2g fructose

The resulting total soluble sugar equivalent is 146.0g of sugars produced.

5 bar 60 min, 1.0% Sulfuric



able 4.26 The effect of: 5bar, 60min, 1.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	0.092	897	0.005	173	0.055
2	0.475	982	0.005	112	0.036
3	5.358	90279	0.465	2692	0.856
4	6.608	2408114	12.401	67126	21.343
5	7.842	5839244	30.069	95014	30.210
6	9.517	8270167	42.588	102695	32.652
7	12.192	2731580	14.066	44785	14.239
8	13.917	77942	0.401	1917	0.610
Totals		**19419205**	**100.000**	**314514**	**100.000**

Figure 4.26 The effect of: 5bar, 60min, 1.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.26 are shown in table 4.26 which corresponds to:

0.4g glucuronic acid

34.3g cellobiose

29.3g glucose

0.0g xylose

75.5g fructose

The resulting total soluble sugar equivalent is 139.5g of sugars produced.

5 bar 90 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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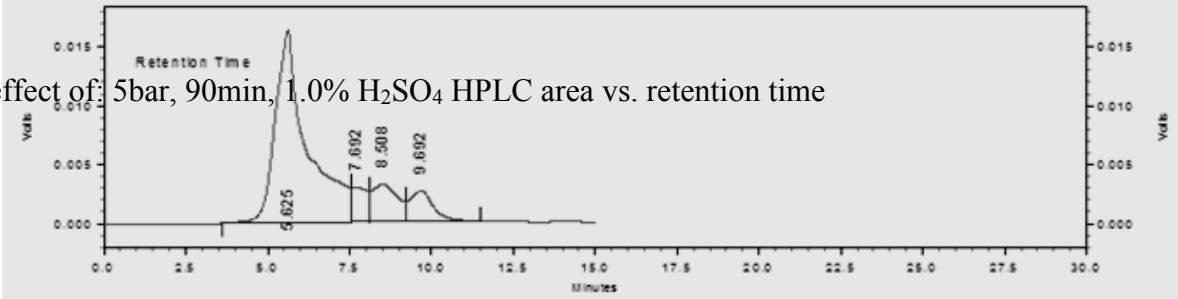
Data Name: D:\DATA\Heated Sugars\EL-2-11
User: System



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.625	1107638	73.700	16335	65.051
2	7.692	90793	6.041	2959	11.784
3	8.508	171443	11.407	3192	12.712
4	9.692	133024	8.851	2625	10.454
Totals		**1502898**	**100.000**	**25111**	**100.000**

Table 4.27 The effect of: 5bar, 90min, 1.0% H_2SO_4 HPLC area vs. retention time

Figure 4.27 The effect of: 5bar, 90min, 1.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.27 are shown in table 4.27 which corresponds to:

5.3g glucuronic acid

0.0g cellobiose

0.5g glucose

0.9g xylose

1.3g fructose

The resulting total soluble sugar equivalent is 7.9g of sugars produced.

Figure 4.28 The effect of: 5bar, 120min, 1.0% H_2SO_4 HPLC

5 bar 120 min, 1.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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Data Name: D:\CLASS-VP\DATA\Heated Sugars\EL-8-11
User: System

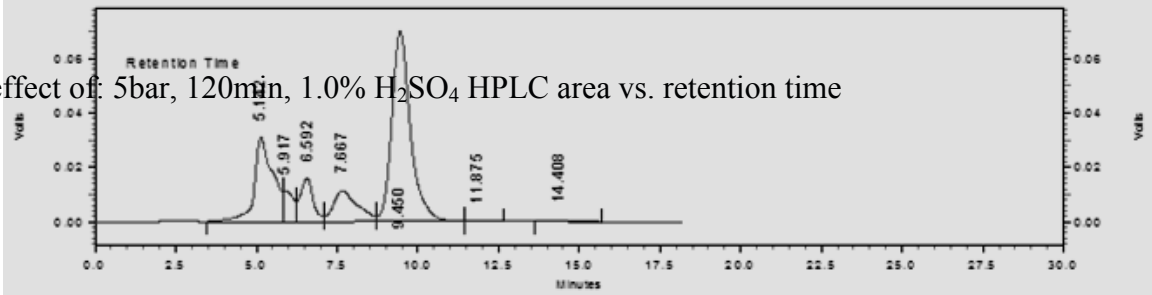

Table 4.28 The effect of: 5bar, 120min, 1.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.142	1243744	23.068	30850	22.066
2	5.917	253463	4.701	11293	8.077
3	6.592	444063	8.236	15691	11.223
4	7.667	585474	10.859	11382	8.141
5	9.450	2829925	52.488	69831	49.947
6	11.875	8269	0.153	216	0.154
7	14.408	26642	0.494	547	0.391
Totals		**5391580**	**100.000**	**139810**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.28 are shown in table 4.28 which corresponds to:

3.9g glucuronic acid

2.9g cellobiose

3.7g glucose

0.0g xylose

22.4g fructose

The resulting total soluble sugar equivalent is 32.9g of sugars produced.

Figure 4.29 The effect of: 3bar, 30min, 2.0% H_2SO_4 HPLC

4.2.7. The effect of pretreatment with 2.0% Sulfuric at 3bar.

3 bar 30 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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Data Name: C:\DATA\Heated Sugars\EL-new\11-24
User: System

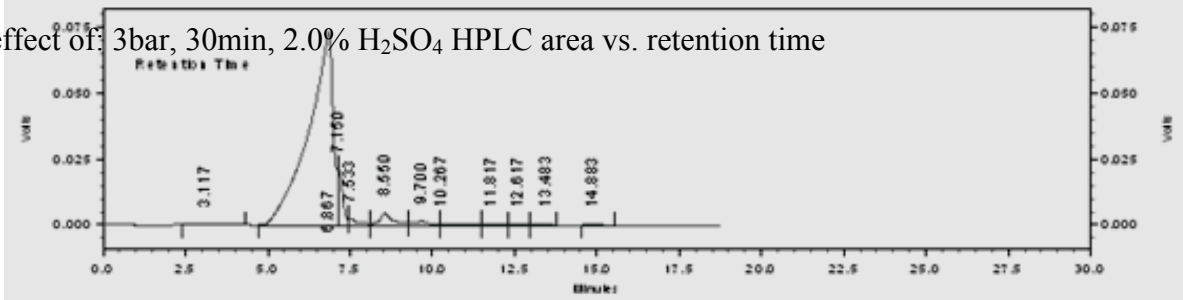

Table 4.29 The effect of: 3bar, 30min, 2.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	3.117	5512	0.130	93	0.090
2	6.867	3781571	89.153	73176	70.750
3	7.150	211230	4.980	20948	20.254
4	7.533	46822	1.104	2335	2.258
5	8.550	116728	2.752	4468	4.320
6	9.700	47870	1.129	1331	1.287
7	10.267	18517	0.437	516	0.499
8	11.817	5280	0.124	161	0.156
9	12.617	1777	0.042	59	0.057
10	13.483	4983	0.117	292	0.282
11	14.883	1368	0.032	50	0.048
Totals		**4241658**	**100.000**	**103429**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.29 are shown in table 4.29 which corresponds to:

0.0g glucuronic acid

48.0g cellobiose

0.2g glucose

0.5g xylose

0.4g fructose

The resulting total soluble sugar equivalent is 49.0g of sugars produced.

3 bar 60 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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Data Name: C:\DATA\Heated Sugars\EL-new\11-23
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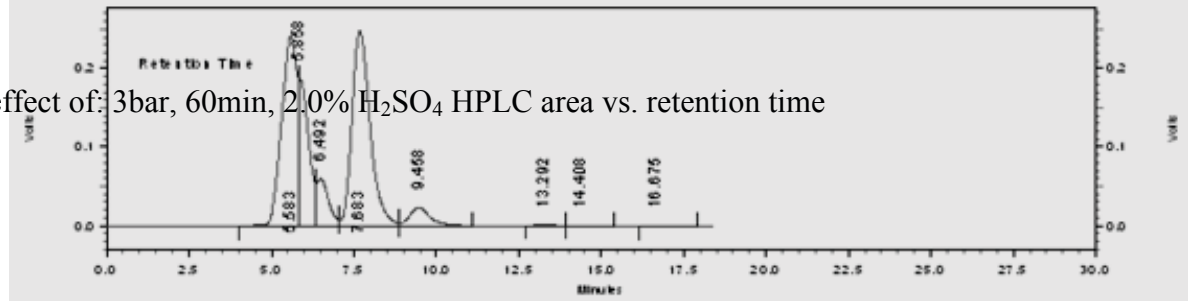

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.583	8023444	34.040	240839	31.735
2	5.858	3597251	15.262	186799	24.614
3	6.492	1486466	6.306	60114	7.921
4	7.683	9438215	40.043	246845	32.526
5	9.458	985430	4.181	23358	3.078
6	13.292	19570	0.083	538	0.071
7	14.408	9652	0.041	200	0.026
8	16.675	10444	0.044	221	0.029
Totals		**23570472**	**100.000**	**758914**	**100.000**

able 4.30 The effect of: 3bar, 60min, 2.0% H_2SO_4 HPLC area vs. retention time

Figure 4.30 The effect of: 3bar, 60min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.30 are shown in table 4.30 which corresponds to:

47.7g glucuronic acid

19.0g cellobiose

42.5g glucose

0.0g xylose

8.1g fructose

The resulting total soluble sugar equivalent is 117.2g of sugars produced.

3 bar 90 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
Method Name: C:\CLASS-VP\methods\Multilevel Calibration.met
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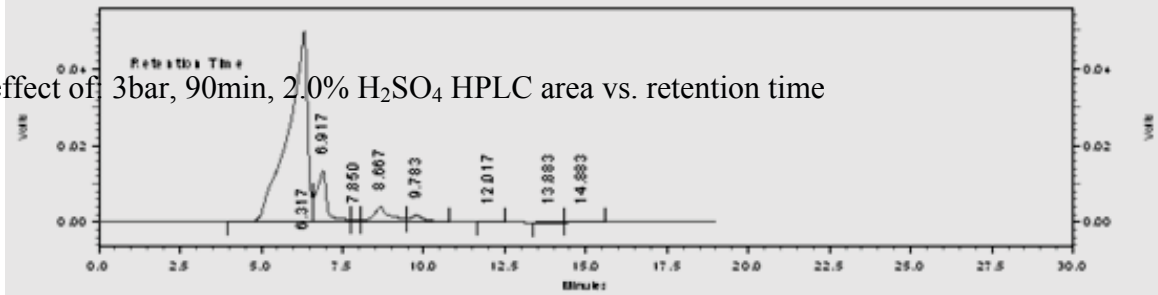
User: System



Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	6.317	2017916	80.632	49526	72.247
2	6.917	300206	11.996	13095	19.103
3	7.850	4657	0.186	308	0.449
4	8.667	121400	4.851	3711	5.413
5	9.783	39006	1.559	1435	2.093
6	12.017	799	0.032	28	0.041
7	13.883	9278	0.371	242	0.353
8	14.883	9363	0.374	206	0.301
Totals		**2502625**	**100.000**	**68551**	**100.000**

Table 4.31 The effect of: 3bar, 90min, 2.0% H_2SO_4 HPLC area vs. retention time

Figure 4.31 The effect of: 3bar, 90min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.31 are shown in table 3.31 which corresponds to:

0.0g glucuronic acid

40.4g cellobiose

0.0g glucose

0.7g xylose

0.4g fructose

The resulting total soluble sugar equivalent is 41.6g of sugars produced.

3 bar 120 min, 2.0% Sulfuric

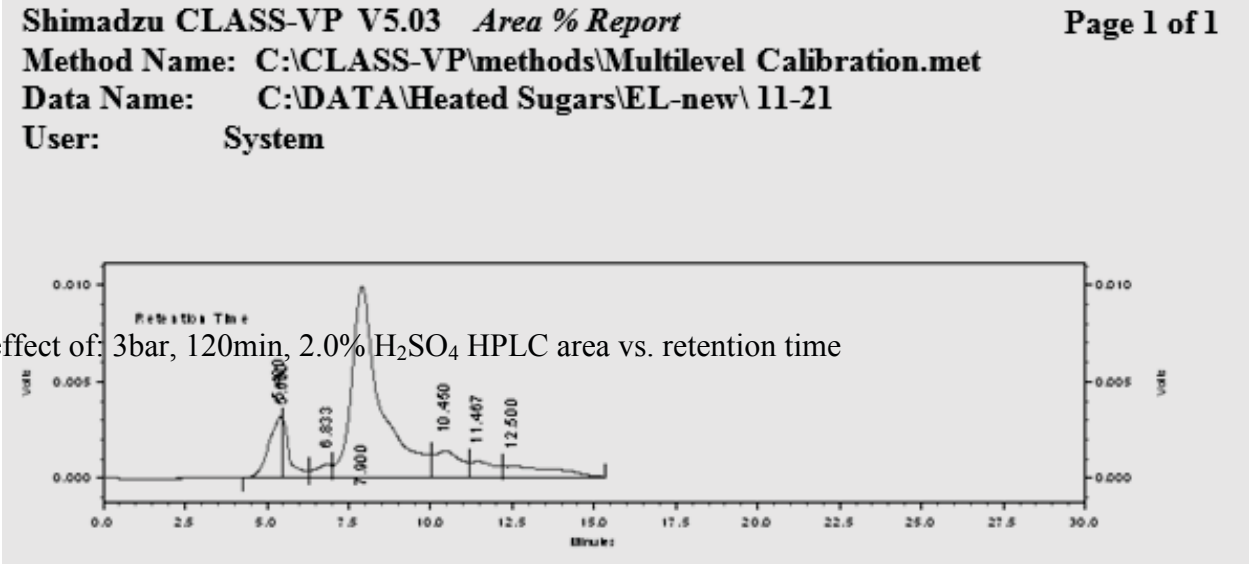


Table 4.32 The effect of: 3bar, 120min, 2.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.400	90693	9.453	3180	16.323
2	5.500	43439	4.527	2935	15.065
3	6.833	23779	2.478	681	3.496
4	7.900	616991	64.306	9884	50.734
5	10.450	78052	8.135	1400	7.186
6	11.467	41331	4.308	817	4.194
7	12.500	65175	6.793	585	3.003
Totals		**959460**	**100.000**	**19482**	**100.000**

Figure 4.32 The effect of: 3bar, 120min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.32 are shown in table 4.32 which corresponds to:

0.9g glucuronic acid

0.5g cellobiose

4.3g glucose

0.0g xylose

0.0g fructose

The resulting total soluble sugar equivalent is 5.6g of sugars produced.

Figure 4.33 The effect of: 4bar, 30min, 2.0% H_2SO_4 HPLC

4.2.8. The effect of pretreatment with 2.0% Sulfuric at 4bar.

4 bar 30 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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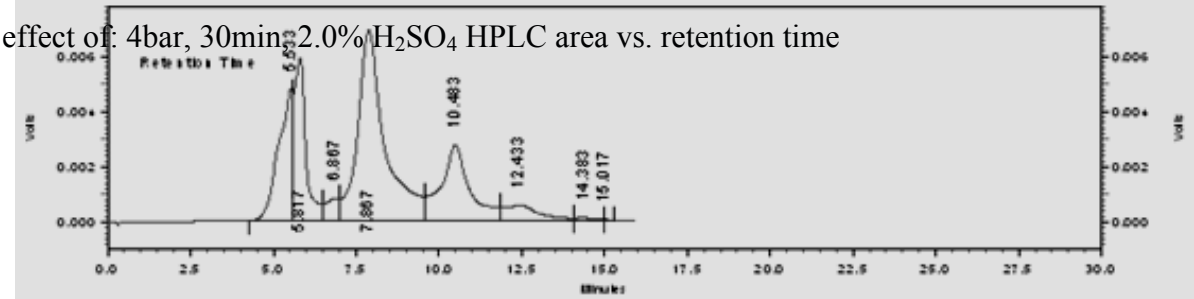

Table 4.33 The effect of: 4bar, 30min, 2.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.533	140197	15.453	4777	21.983
2	5.817	140979	15.539	5895	27.128
3	6.867	22760	2.509	814	3.746
4	7.867	395055	43.544	6883	31.675
5	10.483	164079	18.085	2735	12.586
6	12.433	40592	4.474	527	2.425
7	14.383	3416	0.377	82	0.377
8	15.017	173	0.019	17	0.078
Totals		**907251**	**100.000**	**21730**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.33 are shown in table 4.33 which corresponds to:

1.0g glucuronic acid

0.3g cellobiose

1.6g glucose

0.0g xylose

0.0g fructose

The resulting total soluble sugar equivalent is 3.0g of sugars produced.

Figure 4.34 The effect of: 4bar, 60min, 2.0% H_2SO_4 HPLC

4 bar 60 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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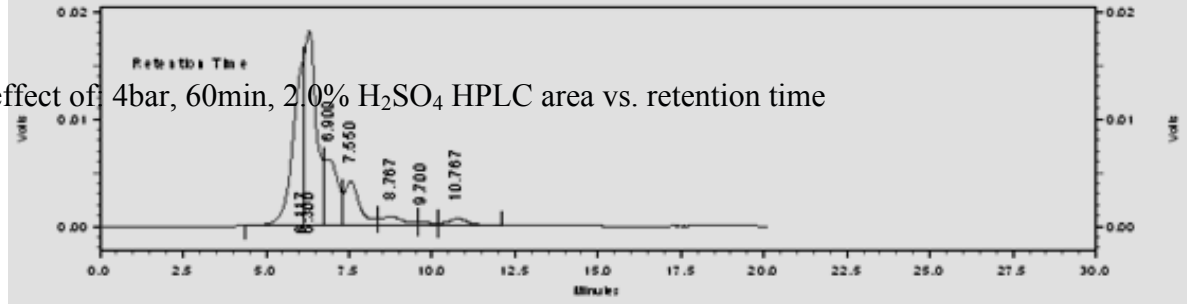

Table 4.34 The effect of: 4bar, 60min, 2.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	6.117	321403	27.326	15420	33.787
2	6.300	474710	40.361	18132	39.729
3	6.900	173209	14.727	6209	13.605
4	7.550	130435	11.090	4176	9.150
5	8.767	40298	3.426	821	1.799
6	9.700	9702	0.825	273	0.598
7	10.767	26409	2.245	608	1.332
Totals		**1176166**	**100.000**	**45639**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.34 are shown in table 4.34 which corresponds to:

0.0g glucuronic acid

14.6g cellobiose

0.7g glucose

0.2g xylose

0.1g fructose

The resulting total soluble sugar equivalent is 15.6g of sugars produced.

Figure 4.35 The effect of: 4bar, 90min, 2.0% H_2SO_4 HPLC

4 bar 90 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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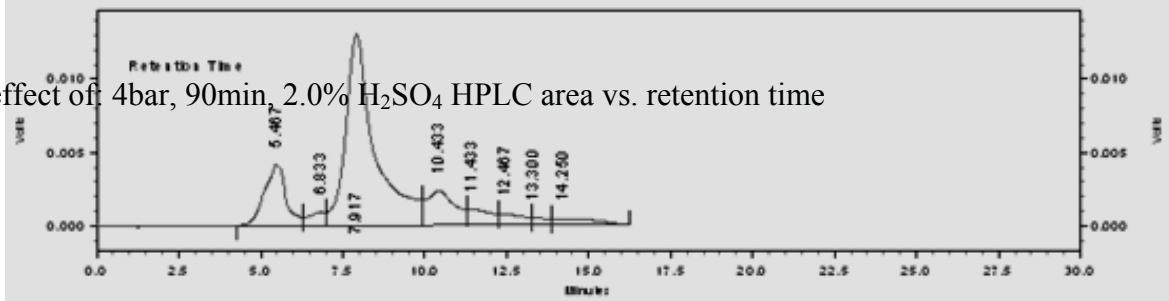

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.467	190644	14.333	4150	17.826
2	6.833	32371	2.434	941	4.042
3	7.917	816230	61.366	13023	55.938
4	10.433	138938	10.446	2360	10.137
5	11.433	55648	4.184	1109	4.764
6	12.467	38990	2.931	773	3.320
7	13.300	16243	1.221	486	2.088
8	14.250	41029	3.085	439	1.886
Totals		**1330093**	**100.000**	**23281**	**100.000**

Table 4.35 The effect of: 4bar, 90min, 2.0% H_2SO_4 HPLC area vs. retention time

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.35 are shown in table 4.35 which corresponds to:

1.0g glucuronic acid

0.5g cellobiose

4.8g glucose

0.0g xylose

0.0g fructose

The resulting total soluble sugar equivalent is 6.3g of sugars produced.

4 bar 120 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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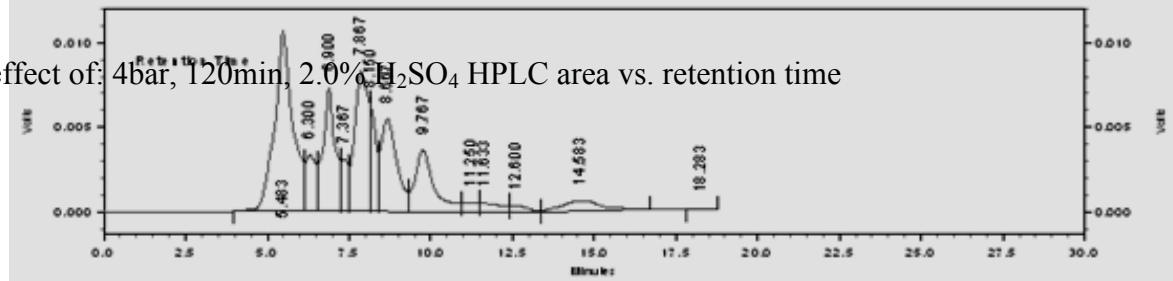

able 4.36 The effect of: 4bar, 120min, 2.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.483	412359	28.065	10664	21.294
2	6.300	70799	4.819	3341	6.671
3	6.900	191210	13.014	7328	14.633
4	7.367	41544	2.828	3091	6.172
5	7.867	241838	16.460	8276	16.526
6	8.150	78557	5.347	6381	12.742
7	8.667	182552	12.425	5480	10.942
8	9.767	150259	10.227	3607	7.202
9	11.250	17311	1.178	514	1.026
10	11.633	21034	1.432	481	0.960
11	12.600	11835	0.805	312	0.623
12	14.583	48437	3.297	560	1.118
13	18.283	1540	0.105	45	0.090
Totals		**1469275**	**100.000**	**50080**	**100.000**

Figure 4.36 The effect of: 4bar, 120min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.36 are shown in table 4.36 which corresponds to:

3.0g glucuronic acid

5.9g cellobiose

2.2g glucose

2.0g xylose

2.2g fructose

The resulting total soluble sugar equivalent is 15.3g of sugars produced.

4.2.9. The effect of pretreatment with 2.0% Sulfuric at 5bar.

5 bar 30 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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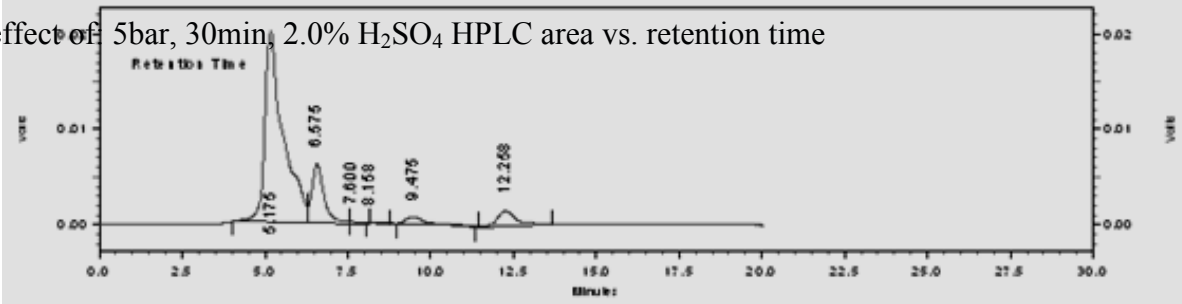

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.175	775637	74.768	19964	69.799
2	6.575	159346	15.360	6129	21.429
3	7.600	5286	0.510	190	0.664
4	8.158	1463	0.141	5	0.017
5	9.475	32877	3.169	764	2.671
6	12.258	62787	6.052	1550	5.419
Totals		**1037396**	**100.000**	**28602**	**100.000**

Table 4.37 The effect of: 5bar, 30min, 2.0% H_2SO_4 HPLC area vs. retention time

Figure 4.37 The effect of: 5bar, 30min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.37 are shown in table 4.37 which corresponds to:

3.2g glucuronic acid

2.0g cellobiose

0.0g glucose

0.0g xylose

0.3g fructose

The resulting total soluble sugar equivalent is 5.5g of sugars produced.

5 bar 60 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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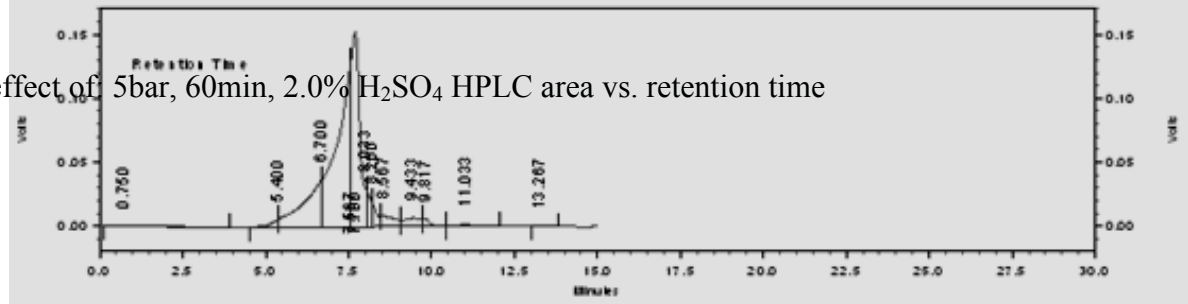

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	0.750	36538	0.415	478	0.121
2	5.400	85141	0.968	6060	1.538
3	6.700	1405829	15.977	36594	9.285
4	7.567	3589505	40.795	128682	32.650
5	7.700	2604511	29.600	151723	38.497
6	8.033	242868	2.760	28760	7.297
7	8.200	185858	2.112	19743	5.009
8	8.567	256318	2.913	7996	2.029
9	9.433	230328	2.618	6594	1.673
10	9.817	95481	1.085	5946	1.509
11	11.033	65218	0.741	1492	0.379
12	13.267	1336	0.015	52	0.013
Totals		**8798931**	**100.000**	**394120**	**100.000**

able 4.38 The effect of: 5bar, 60min, 2.0% H_2SO_4 HPLC area vs. retention time

Figure 4.38 The effect of: 5bar, 60min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.38 are shown in table 4.38 which corresponds to:

0.0g glucuronic acid

0.0g cellobiose

46.1g glucose

0.0g xylose

0.0g fructose

The resulting total soluble sugar equivalent is 46.1g of sugars produced.

Figure 4.39 The effect of: 5bar, 90min, 2.0% H_2SO_4 HPLC

5 bar 90 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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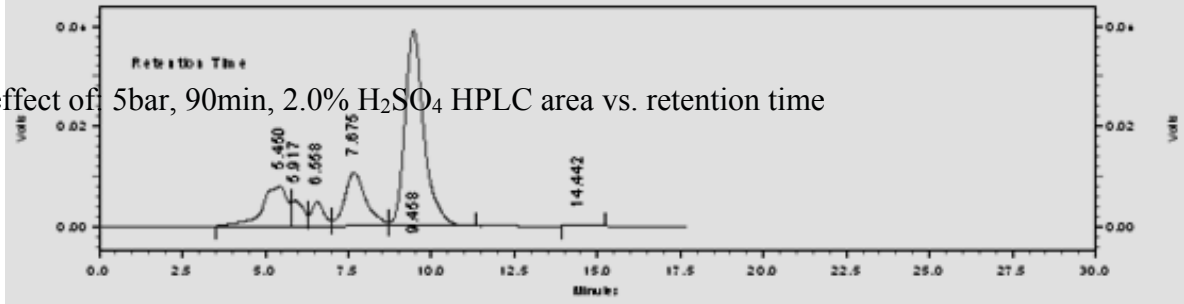

Table 4.39 The effect of: 5bar, 90min, 2.0% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.450	412670	15.190	8017	11.800
2	5.917	131018	4.822	5142	7.569
3	6.558	128759	4.739	4873	7.173
4	7.675	476312	17.532	10676	15.714
5	9.458	1561449	57.474	39060	57.494
6	14.442	6602	0.243	170	0.250
Totals		**2716810**	**100.000**	**67938**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.39 are shown in table 4.39 which corresponds to:

3.1g glucuronic acid

2.3g cellobiose

3.0g glucose

0.0g xylose

18.0g fructose

The resulting total soluble sugar equivalent is 26.5g of sugars produced.

5 bar 120 min, 2.0% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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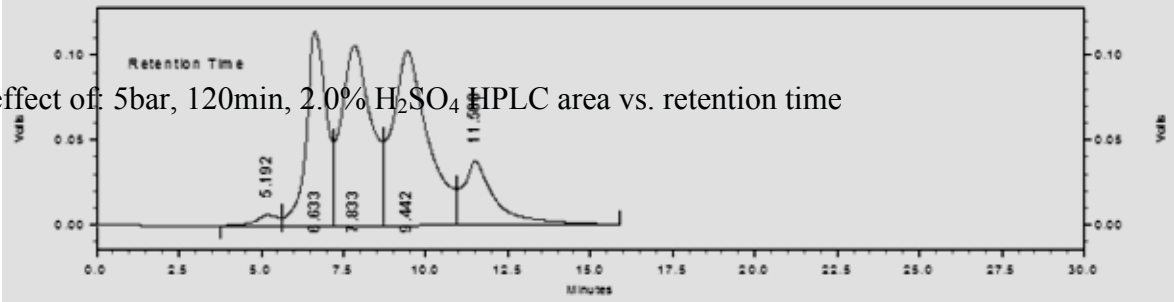

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.192	285320	1.286	6109	1.666
2	6.633	4878862	21.997	114111	31.115
3	7.833	6659171	30.024	105983	28.899
4	9.442	7772252	35.042	102743	28.015
5	11.500	2584105	11.651	37795	10.306
Totals		**22179710**	**100.000**	**366741**	**100.000**

Table 4.40 The effect of: 5bar, 120min, 2.0% H_2SO_4 HPLC area vs. retention time

Figure 4.40 The effect of: 5bar, 120min, 2.0% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.40 are shown in table 4.40 which corresponds to:

0.2g glucuronic acid

10.3g cellobiose

5.0g glucose

0.0g xylose

10.6g fructose

The resulting total soluble sugar equivalent is 26.1g of sugars produced.

Figure 4.41 The effect of: 5bar, 30min, 4.25% H_2SO_4 HPLC

4.2.10. The effect of pretreatment with 4.25% Sulfuric at 5bar.

5 bar 30 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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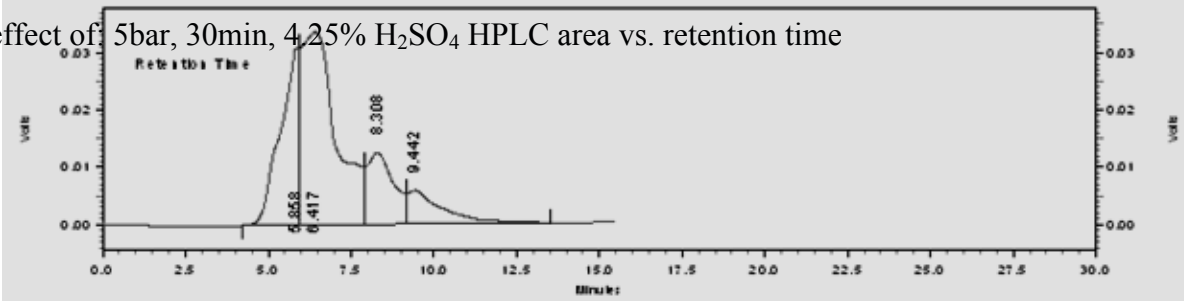

Table 4.41 The effect of: 5bar, 30min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.858	1178688	24.617	30990	37.348
2	6.417	2472844	51.646	33651	40.555
3	8.308	708273	14.793	12505	15.071
4	9.442	428230	8.944	5830	7.026
Totals		**4788035**	**100.000**	**82976**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.41 are shown in table 4.41 which corresponds to:

5.8g glucuronic acid

37.9g cellobiose

0.0g glucose

3.8g xylose

4.2g fructose

The resulting total soluble sugar equivalent is 51.7g of sugars produced.

Figure 4.42 The effect of: 5bar, 60min, 4.25% H_2SO_4 HPLC

5 bar 60 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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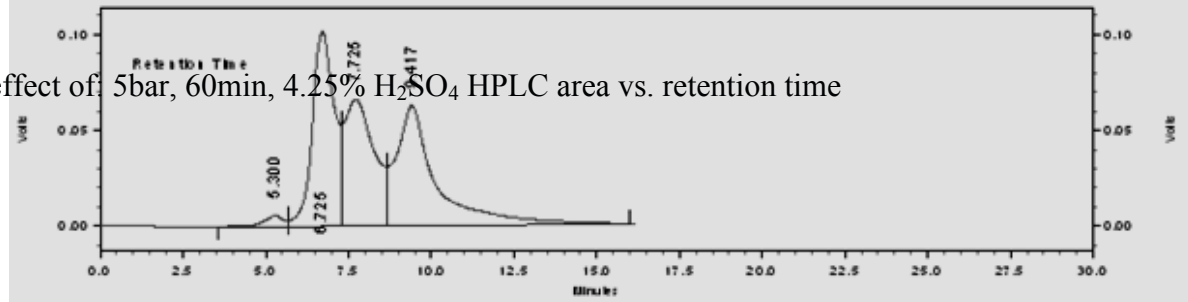

able 4.42 The effect of: 5bar, 60min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.300	213816	1.496	5550	2.362
2	6.725	4692930	32.842	101156	43.051
3	7.725	4084153	28.582	65852	28.026
4	9.417	5298458	37.080	62408	26.560
Totals		**14289357**	**100.000**	**234966**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.42 are shown in table 4.42 which corresponds to:

1.2g glucuronic acid

81.5g cellobiose

25.0g glucose

0.0g xylose

59.0g fructose

The resulting total soluble sugar equivalent is 166.6g of sugars produced.

Figure 4.43 The effect of: 5bar, 90min, 4.25% H_2SO_4 HPLC

5 bar 90 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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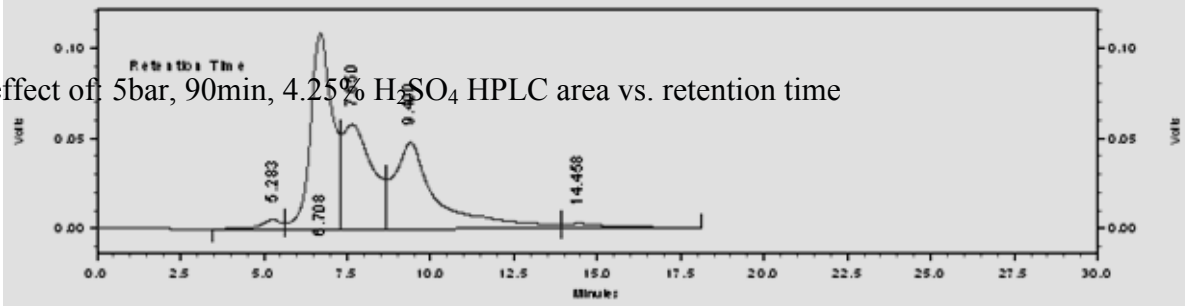

Table 4.43 The effect of: 5bar, 90min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.283	214370	1.602	5404	2.436
2	6.708	4988695	37.274	108111	48.732
3	7.650	3572092	26.690	57720	26.018
4	9.400	4308761	32.194	47835	21.562
5	14.458	299770	2.240	2780	1.253
Totals		**13383688**	**100.000**	**221850**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.43 are shown in table 4.43 which corresponds to:

1.1g glucuronic acid

82.8g cellobiose

20.9g glucose

0.0g xylose

45.9g fructose

The resulting total soluble sugar equivalent is 150.7g of sugars produced.

Figure 4.44 The effect of: 5bar, 120min, 4.25% H_2SO_4 HPLC

5 bar 120 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**

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User: System

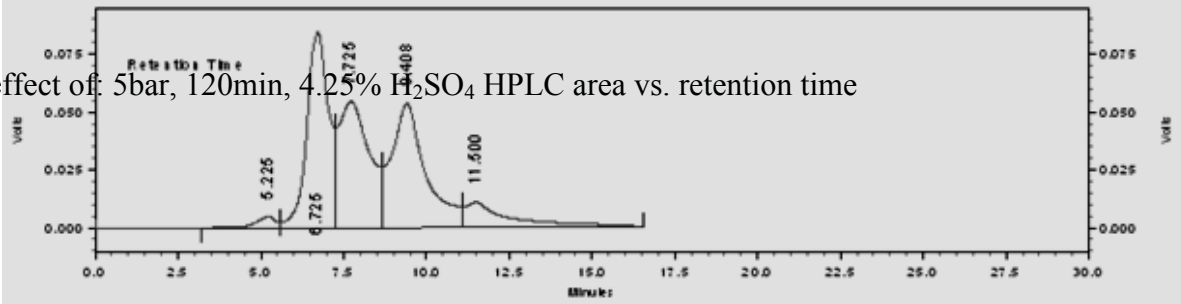

Table 4.44 The effect of: 5bar, 120min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.225	197834	1.596	4851	2.338
2	6.725	3887946	31.367	84169	40.564
3	7.725	3430377	27.675	54338	26.187
4	9.408	3883405	31.330	53461	25.764
5	11.500	995511	8.032	10680	5.147
Totals		**12395073**	**100.000**	**207499**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.44 are shown in table 4.44 which corresponds to:

1.2g glucuronic acid

71.7g cellobiose

22.3g glucose

0.0g xylose

45.9g fructose

The resulting total soluble sugar equivalent is 141.1g of sugars produced.

4.2.11. The effect of pretreatment with 4.25% Sulfuric at 4bar.

4 bar 30 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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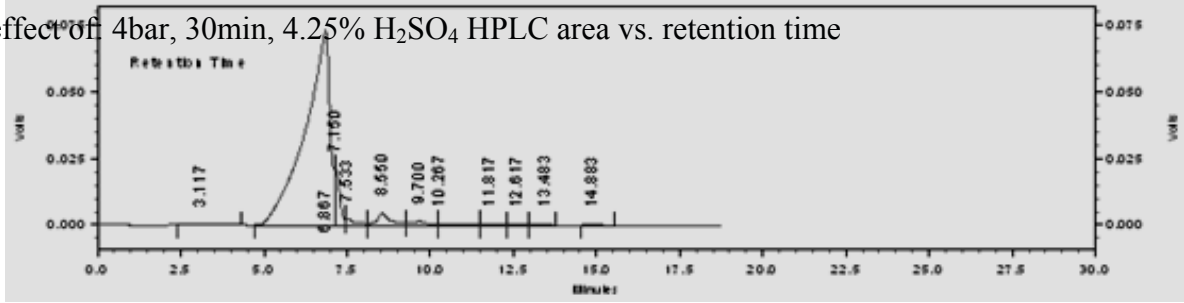

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	3.117	5512	0.130	93	0.090
2	6.867	3781571	89.153	73176	70.750
3	7.150	211230	4.980	20948	20.254
4	7.533	46822	1.104	2335	2.258
5	8.550	116728	2.752	4468	4.320
6	9.700	47870	1.129	1331	1.287
7	10.267	18517	0.437	516	0.499
8	11.817	5280	0.124	161	0.156
9	12.617	1777	0.042	59	0.057
10	13.483	4983	0.117	292	0.282
11	14.883	1368	0.032	50	0.048
Totals		**4241658**	**100.000**	**103429**	**100.000**

'**able 4.45** The effect of: 4bar, 30min, 4.25% H_2SO_4 HPLC area vs. retention time

Figure 4.45 The effect of: 4bar, 30min, 4.25% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.45 are shown in table 4.45 which corresponds to:

1.0g glucuronic acid

0.0g cellobiose

27.8g glucose

2.3g xylose

3.1g fructose

The resulting total soluble sugar equivalent is 34.2g of sugars produced.

4bar 60 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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Data Name: C:\DATA\Heated Sugars\EL-new\12-29
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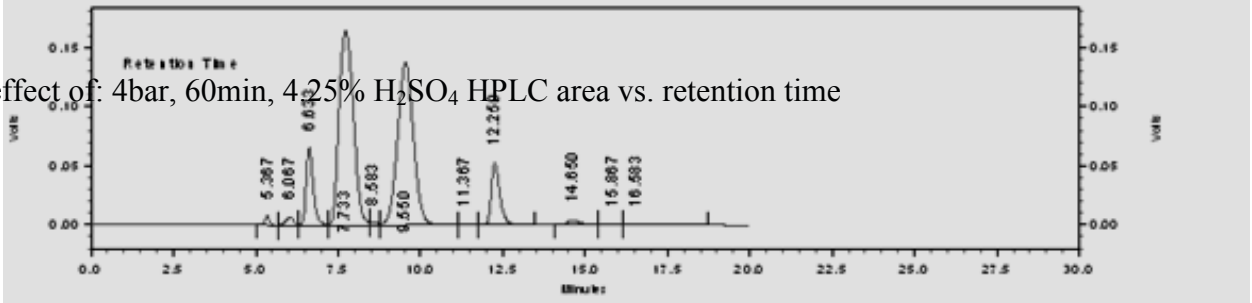

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.367	75451	0.628	7902	1.788
2	6.067	110074	0.917	7232	1.636
3	6.633	1007091	8.388	65185	14.750
4	7.733	4962534	41.332	163620	37.024
5	8.583	36986	0.308	2543	0.575
6	9.550	4674340	38.932	136863	30.969
7	11.367	4283	0.036	206	0.047
8	12.250	937392	7.809	52606	11.904
9	14.650	109080	0.909	4076	0.922
10	15.867	27690	0.231	850	0.192
11	16.583	61259	0.510	846	0.191
Totals		**12006380**	**100.000**	**441929**	**100.000**

Table 4.46 The effect of: 4bar, 60min, 4.25% H_2SO_4 HPLC area vs. retention time

Figure 4.46 The effect of: 4bar, 60min, 4.25% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.46 are shown in table 4.46 which corresponds to:

0.3g glucuronic acid

14.0g cellobiose

24.3g glucose

0.2g xylose

41.7g fructose

The resulting total soluble sugar equivalent is 80.5g of sugars produced.

4 bar 90 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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Data Name: C:\DATA\Heated Sugars\EL-new\12-28
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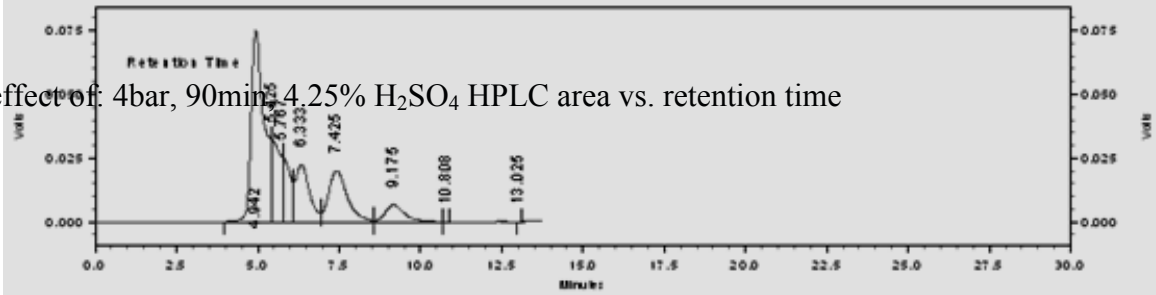

Table 4.47 The effect of: 4bar, 90min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	4.942	2099009	43.261	74813	41.263
2	5.425	589064	12.141	32181	17.750
3	5.767	423206	8.722	25291	13.949
4	6.333	666691	13.741	22499	12.409
5	7.425	802432	16.538	19915	10.984
6	9.175	271339	5.592	6569	3.623
7	10.808	123	0.003	17	0.009
8	13.025	54	0.001	21	0.012
Totals		**4851918**	**100.000**	**181306**	**100.000**

Figure 4.47 The effect of: 4bar, 90min, 4.25% H_2SO_4 HPLC

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.47 are shown in table 4.47 which corresponds to:

14.3g glucuronic acid

9.5g cellobiose

4.0g glucose

0.0g xylose

2.5g fructose

The resulting total soluble sugar equivalent is 30.3g of sugars produced.

Figure 4.48 The effect of: 4bar, 120min, 4.25% H_2SO_4 HPLC

4 bar 120 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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Data Name: C:\DATA\Heated Sugars\EL-new\12-27
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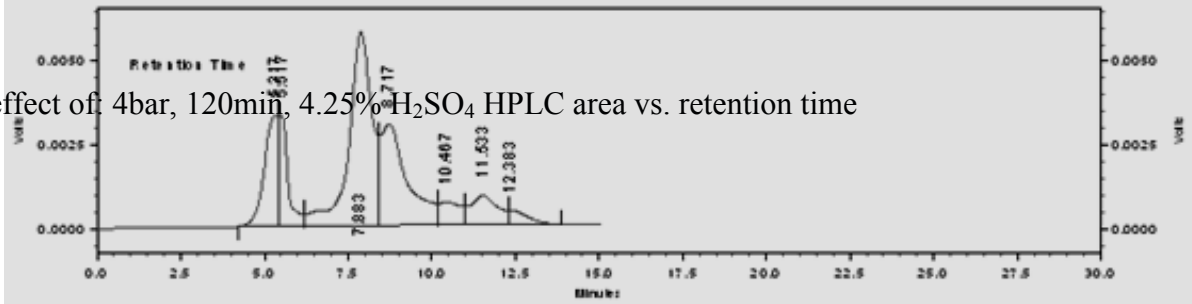

Table 4.48 The effect of: 4bar, 120min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.317	92512	13.115	3311	19.015
2	5.517	64353	9.123	3315	19.038
3	7.883	282868	40.100	5778	33.182
4	8.717	168878	23.940	3028	17.389
5	10.467	30337	4.301	687	3.945
6	11.533	50426	7.148	887	5.094
7	12.383	16040	2.274	407	2.337
Totals		**705414**	**100.000**	**17413**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.48 are shown in table 4.48 which corresponds to:

0.7g glucuronic acid

0.0g cellobiose

1.5g glucose

0.9g xylose

0.0g fructose

The resulting total soluble sugar equivalent is 3.0g of sugars produced.

Figure 4.49 The effect of: 3bar, 30min, 4.25% H_2SO_4 HPLC

4.2.12. The effect of pretreatment with 4.25% Sulfuric at 3bar.

3 bar 30 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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Data Name: C:\DATA\Heated Sugars\EL-new\ 12-34
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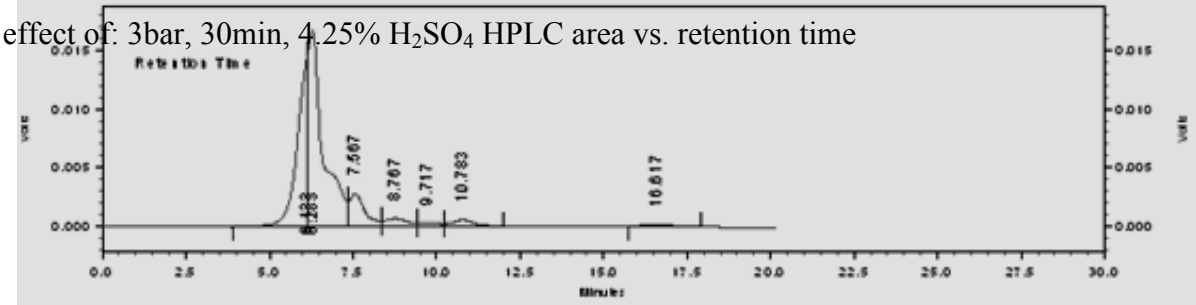

Table 4.49 The effect of: 3bar, 30min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	6.133	293863	29.783	14517	40.783
2	6.283	527186	53.431	16677	46.851
3	7.567	91586	9.282	2806	7.883
4	8.767	30707	3.112	650	1.826
5	9.717	11681	1.184	274	0.770
6	10.783	21938	2.223	504	1.416
7	16.617	9705	0.984	168	0.472
Totals		**986666**	**100.000**	**35596**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.49 are shown in table 4.49 which corresponds to:

0.0g glucuronic acid

11.3g cellobiose

0.4g glucose

0.1g xylose

0.1g fructose

The resulting total soluble sugar equivalent is 12.0g of sugars produced.

Figure 4.50 The effect of: 3bar, 60min, 4.25% H_2SO_4 HPLC

3 bar 60 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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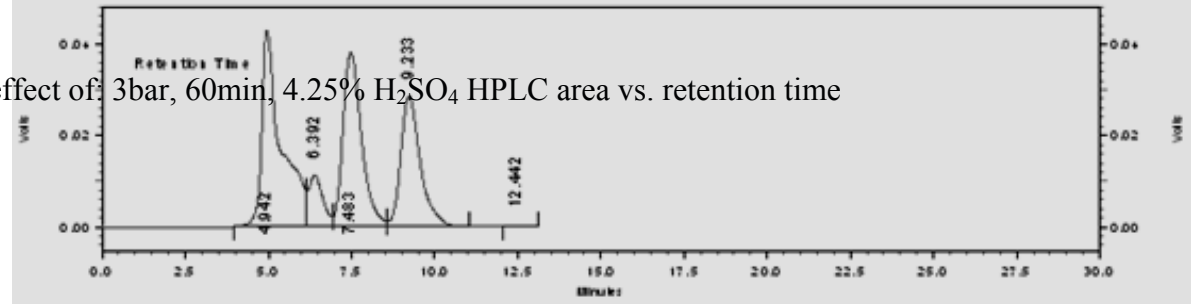

able 4.50 The effect of: 3bar, 60min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	4.942	1765515	37.179	42679	35.405
2	6.392	333300	7.019	11287	9.363
3	7.483	1481150	31.191	37913	31.451
4	9.233	1163355	24.498	28480	23.626
5	12.442	5369	0.113	186	0.154
Totals		**4748689**	**100.000**	**120545**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.50 are shown in table 4.50 which corresponds to:

0.0g glucuronic acid

13.5g cellobiose

0.6g glucose

0.2g xylose

0.1g fructose

The resulting total soluble sugar equivalent is 14.4g of sugars produced.

Figure 4.51 The effect of: 3bar, 90min, 4.25% H_2SO_4 HPLC

3 bar 90 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 ***Area % Report*** **Page 1 of 1**
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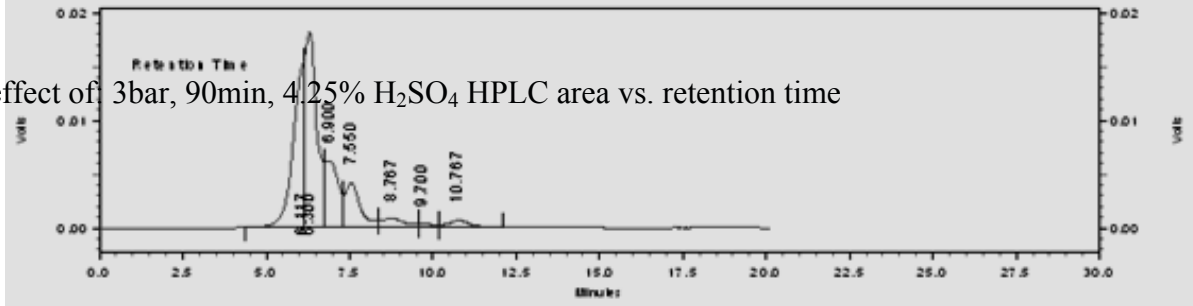

Table 4.51 The effect of: 3bar, 90min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	6.117	321403	27.326	15420	33.787
2	6.300	474710	40.361	18132	39.729
3	6.900	173209	14.727	6209	13.605
4	7.550	130435	11.090	4176	9.150
5	8.767	40298	3.426	821	1.799
6	9.700	9702	0.825	273	0.598
7	10.767	26409	2.245	608	1.332
Totals		**1176166**	**100.000**	**45639**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.51 are shown in table 4.51 which corresponds to:

9.3g glucuronic acid

5.5g cellobiose

8.5g glucose

0.0g xylose

12.2g fructose

The resulting total soluble sugar equivalent is 35.5g of sugars produced.

Figure 4.52 The effect of: 3bar, 120min, 4.25% H_2SO_4 HPLC

3 bar 120 min, 4.25% Sulfuric

Shimadzu CLASS-VP V5.03 *Area % Report* **Page 1 of 1**
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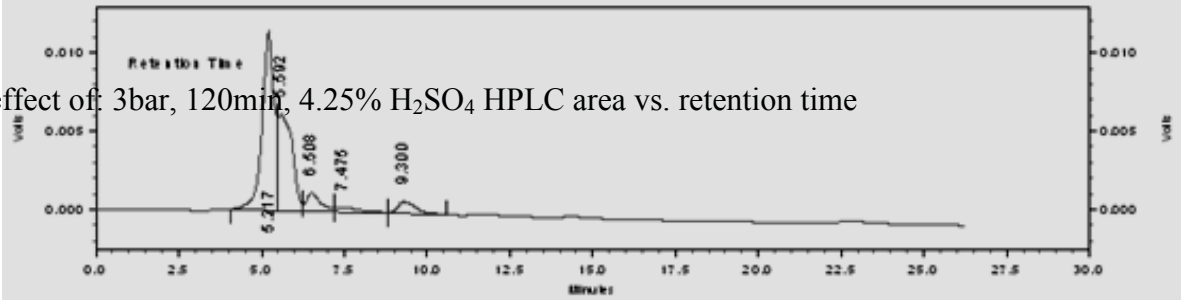


Table 4.52 The effect of: 3bar, 120min, 4.25% H_2SO_4 HPLC area vs. retention time

Detector A

Pk #	Retention Time	Area	Area %	Height	Height %
1	5.217	291822	53.198	11411	57.675
2	5.592	177820	32.416	6154	31.104
3	6.508	35402	6.454	1200	6.065
4	7.475	15330	2.795	281	1.420
5	9.300	28188	5.139	739	3.735
Totals		**548562**	**100.000**	**19785**	**100.000**

Resulting chromatograph of HPLC analysis for the areas under the curve for Figure 4.52 are shown in table 4.52 which corresponds to:

2.6g glucuronic acid

0.6g cellobiose

0.1g glucose

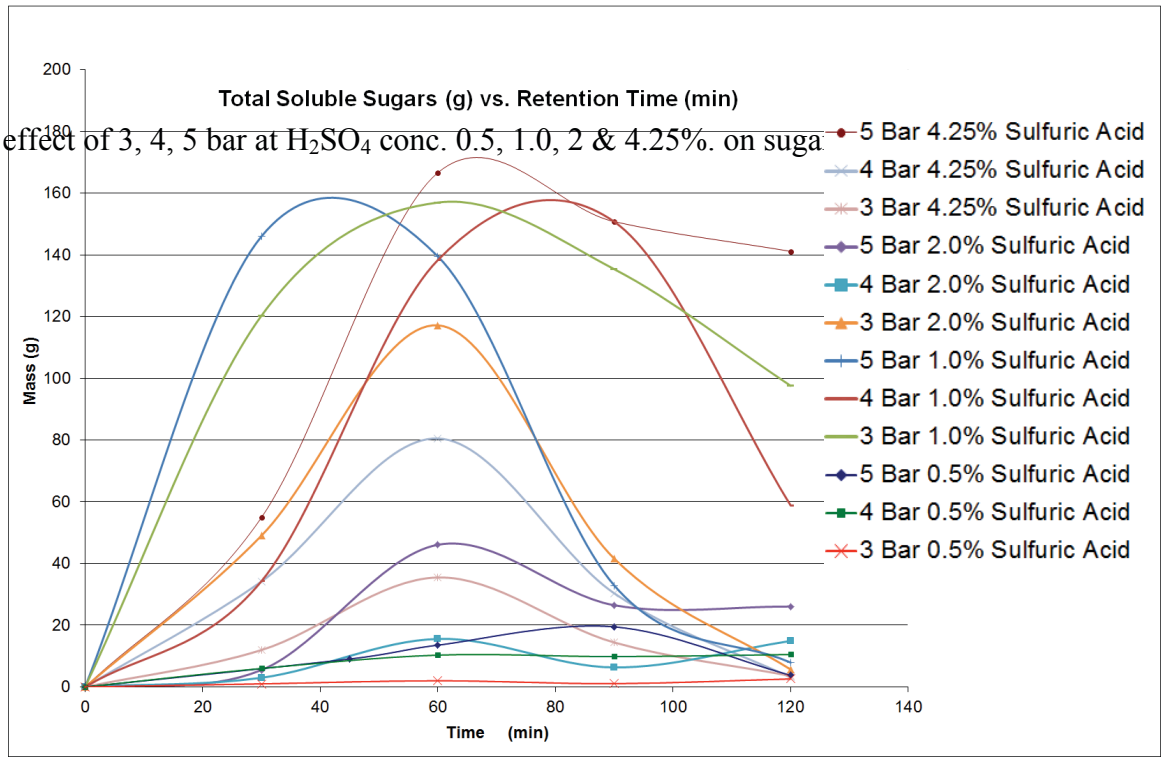
0.0g xylose

0.3g fructose

The resulting total soluble sugar equivalent is 3.6g of sugars produced.

4.3 DISCUSSION:

The overall results show general trends of low sugars production at low acid concentrations and low retention times. Generally speaking high retention times of 120 minutes produce less soluble sugar amounts than medium retention times of 90 minutes. Similarly retention times of 90 minutes produce less soluble sugar amounts than lesser retention times of 60 minutes. On the other hand, retention times of 60 minutes produce more soluble sugars than 30 minutes retention time with the exception of 1% sulfuric acid pretreatment condition at different reaction pressures of 3, 4 and 5 bar.



igure 4.53 The effect of 3, 4, 5 bar at H_2SO_4 conc. 0.5, 1.0, 2 & 4.25%. on suga

The highest yield of sugars achieved is at the condition of 4.25% Sulfuric Acid pretreatment and 5 bar pressure at 60 minutes retention time as seen in Figure 4.53. The yield of sugars at this condition is 166.63g from 400g rice straw.

The second highest yield of sugars achieved is at the condition of 1.0% Sulfuric Acid pretreatment and 3 bar pressure at 60 minutes retention time. The yield of sugars at this condition is 156.95g from 400 g rice straw.

Due to the high energy consumption to generate steam pressure at 5 bar vs. 3 bar, calculations indicate that treatment with 3bar steam for 60 minutes consumes less energy than treatment at a higher pressure of 5 bar for the same duration.

4.3.1. The effect of 3bar, 0.5% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 3 bar, 0.5% Sulfuric acid at different retention times

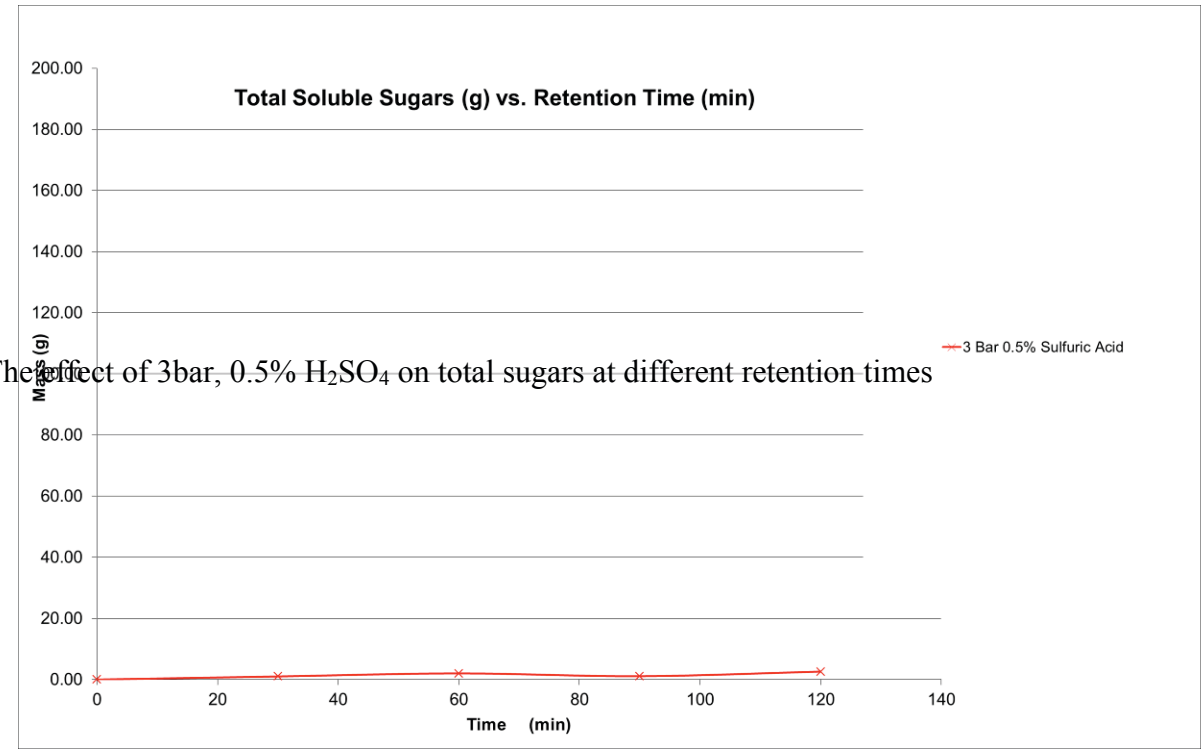


Figure 4.54 The effect of 3bar, 0.5% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 2.6 g at around 120 minutes retention time at 3 bar with 0.5% sulfuric acid pretreatment as seen in figure 4.54.

The results indicate low yield of sugars with the treatment at the pressure of 3 bar and 0.5% Sulfuric acid pretreatment.

4.3.2. The effect of 4bar, 0.5% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 4 bar, 0.5% Sulfuric acid at different retention times

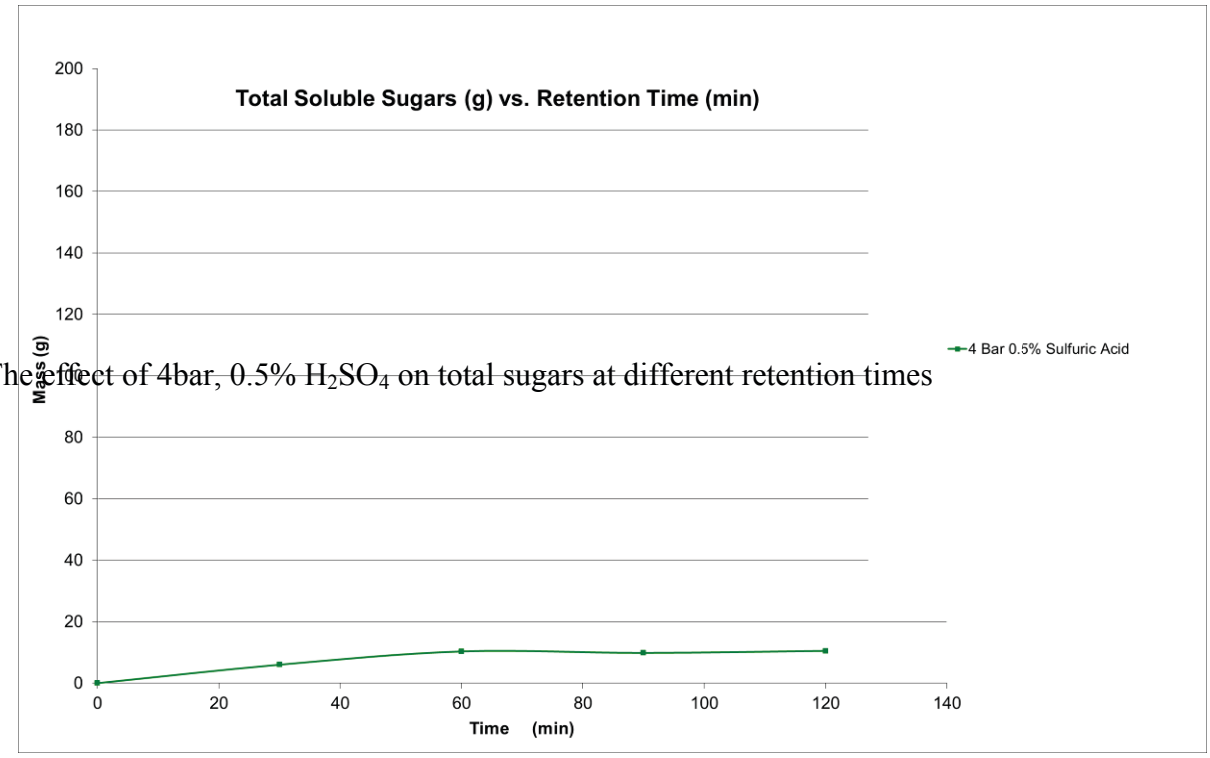


Figure 4.55 The effect of 4bar, 0.5% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 10.5 g at around 120 minutes retention time at 4 bar with 0.5% sulfuric acid pretreatment as seen in firure 4.55.

The graph indicates the peak value will be within 60 to 70 minutes of retention time.

The results also indicate low yield of sugars with the treatment at the pressure of 4 bar and 0.5% Sulfuric acid pretreatment.

4.3.3. The effect of 5bar, 0.5% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 5 bar, 0.5% Sulfuric acid at different retention times

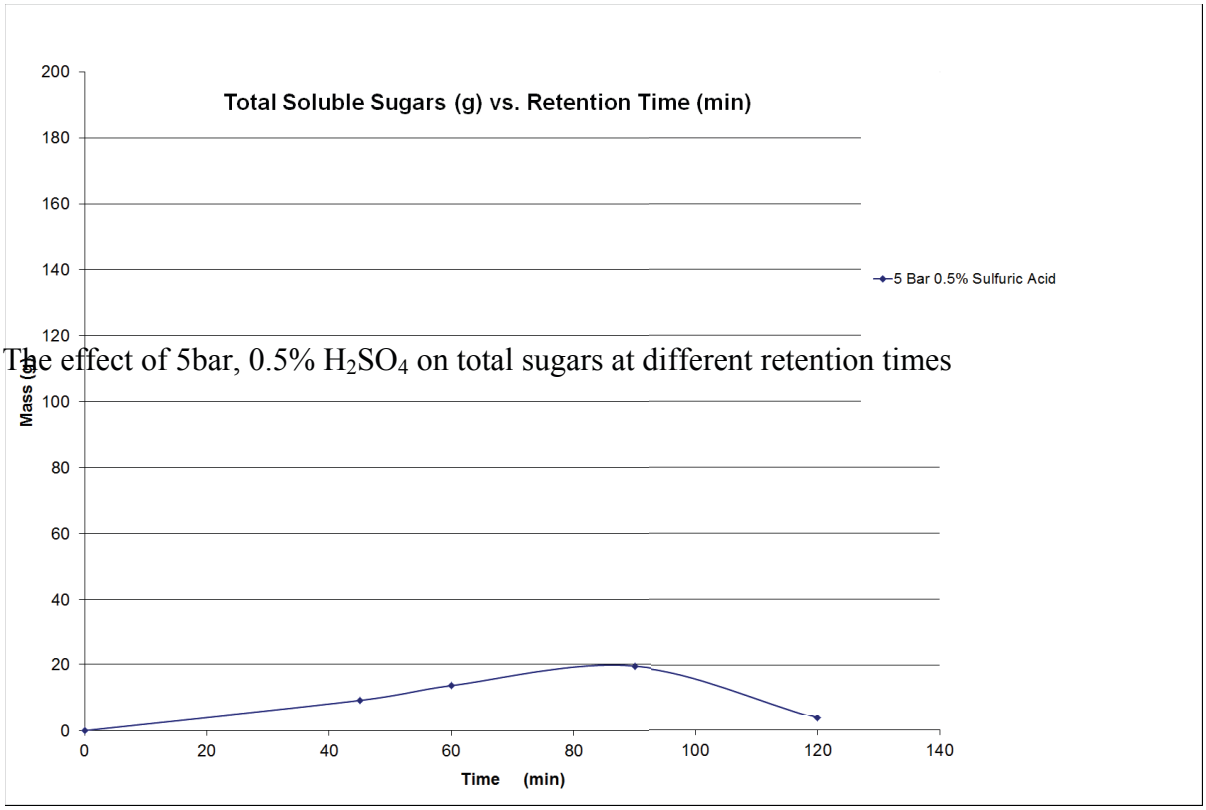


Figure 4.56 The effect of 5bar, 0.5% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 19.5 g at around 90 minutes retention time at 5 bar with 0.5% sulfuric acid pretreatment as seen in figure 4.56.

The graph indicates the peak value will be within 80 to 100 minutes of retention time.

The results also indicate low yield of sugars with the treatment at the pressure of 5 bar and 0.5% Sulfuric acid pretreatment.

4.3.4. The effect of 3bar, 1.0% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 3 bar, 1.0% Sulfuric acid at different retention times

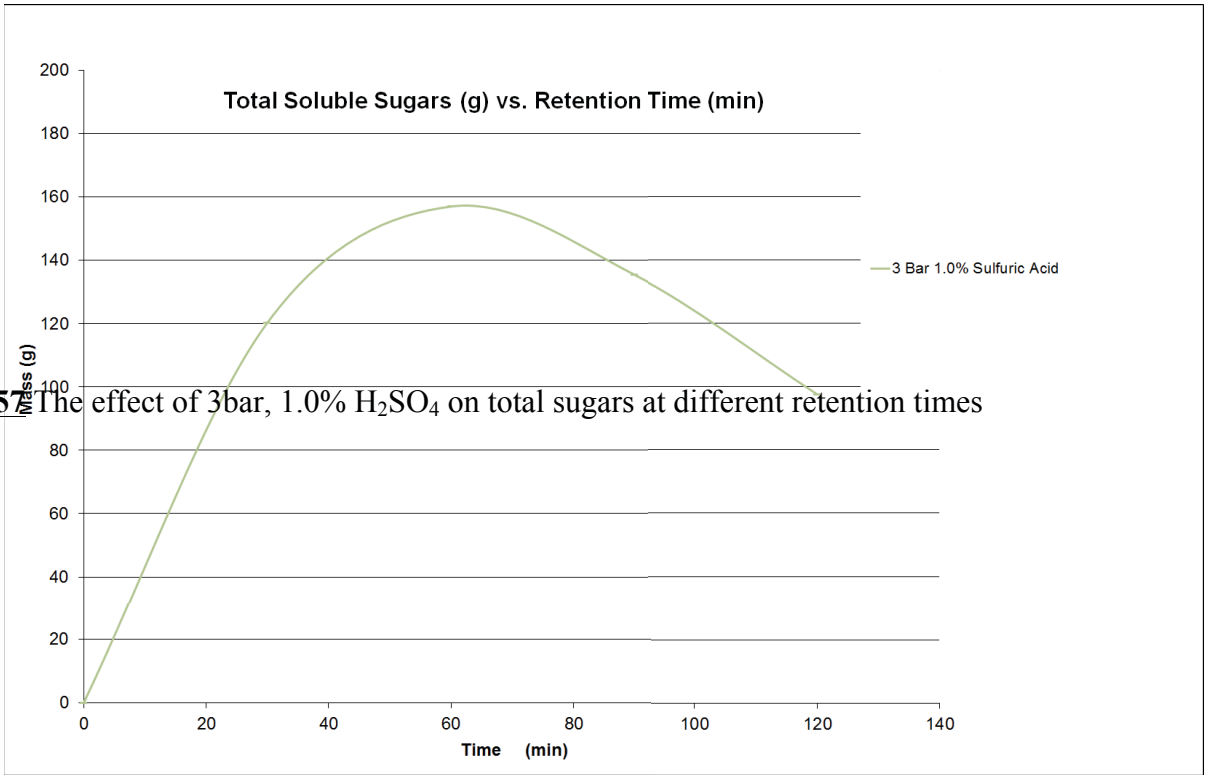


Figure 4.57 The effect of 3bar, 1.0% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 157g at around 60 minutes retention time at 3 bar with 1.0% sulfuric acid pretreatment as seen in figure 4.57.

The graph indicates the peak value will be within 50 to 70 minutes of retention time.

The results also indicate very high yield of sugars with the treatment at the pressure of 3 bar and 1.0% Sulfuric acid pretreatment.

4.3.5. The effect of 4bar, 1.0% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 4 bar, 1.0% Sulfuric acid at different retention times

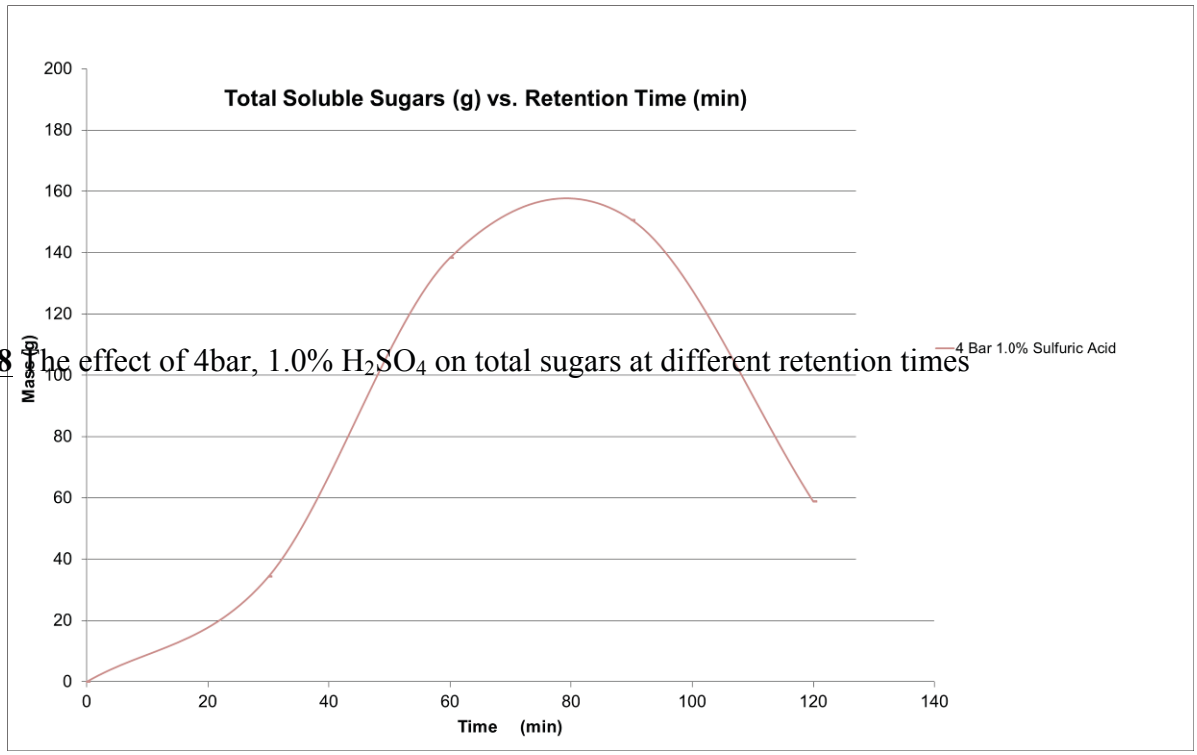


Figure 4.58 The effect of 4bar, 1.0% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 151 g at around 90 minutes retention time at 4 bar with 1.0% sulfuric acid pretreatment as seen in figure 4.58.

The graph indicates the peak value will be within 80 to 100 minutes of retention time.

The results also indicate very high yield of sugars with the treatment at the pressure of 4 bar and 1.0% Sulfuric acid pretreatment.

4.3.6. The effect of 5bar, 1.0% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 5 bar, 1.0% Sulfuric acid at different retention times

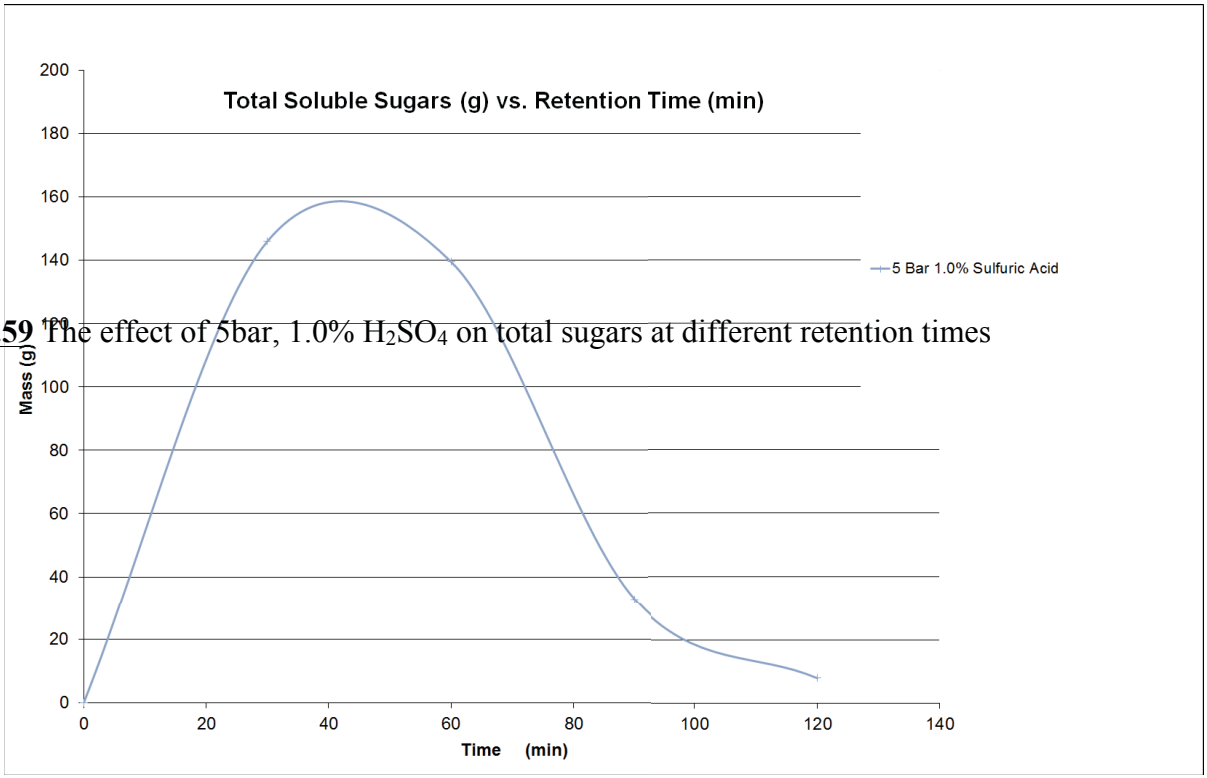


Figure 4.59 The effect of 5bar, 1.0% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 146g at around 30 minutes retention time at 5 bar with 1.0% sulfuric acid pretreatment as seen in figure 4.59.

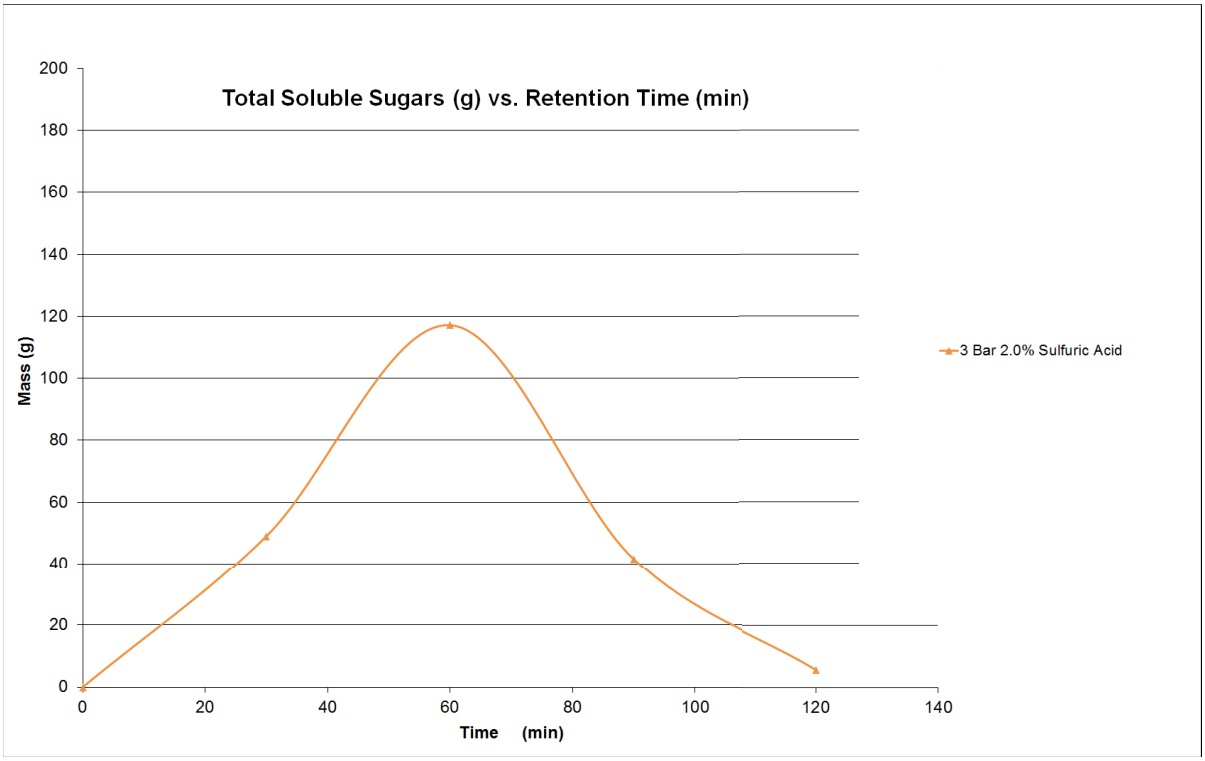
The graph indicates the peak value will be within 20 to 50 minutes of retention time.

The results also indicate very high yield of sugars with the treatment at the pressure of 5 bar and 1.0% Sulfuric acid pretreatment.

Figure 4.60 The effect of 3bar, 2.0% H_2SO_4 on total sugars at different retention times

4.3.7. The effect of 3bar, 2.0% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 3 bar, 2.0% Sulfuric acid at different retention times



Total Soluble Sugars achieved highest value of 117.2g at around 60 minutes retention time at 3 bar with 2.0% sulfuric acid pretreatment as seen in figure 4.60.

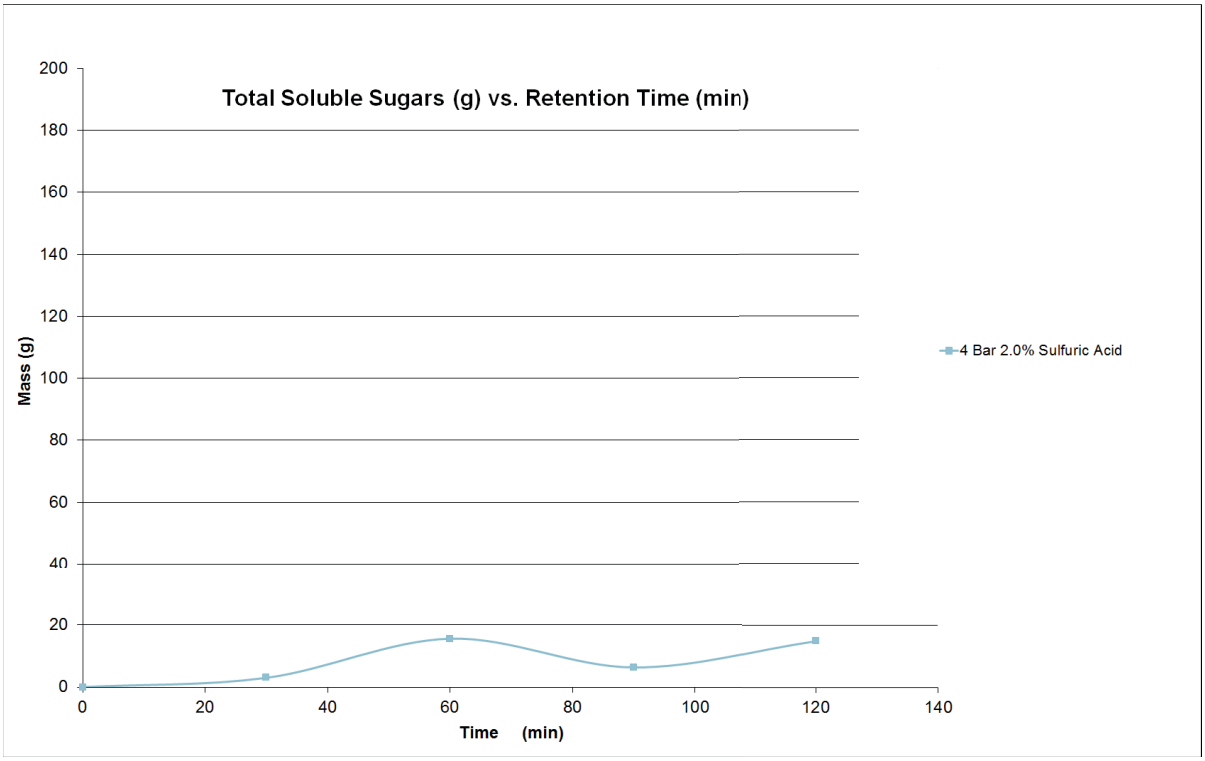
The graph indicates the peak value will be within 50 to 70 minutes of retention time.

The results also indicate high to medium yield of sugars with the treatment at the pressure of 3 bar and 2.0% Sulfuric acid pretreatment.

Figure 4.61 The effect of 2.0% H_2SO_4 on total sugars at different retention times

4.3.8. The effect of 4bar, 2.0% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 4 bar, 2.0% Sulfuric acid at different retention times



Total Soluble Sugars achieved highest value of 15.6g at around 60 minutes retention time at 4 bar with 2.0% sulfuric acid pretreatment as seen in figure 4.61.

The graph indicates the peak value will be within 50 to 70 minutes of retention time.

The results also indicate low yield of sugars with the treatment at the pressure of 4 bar and 2.0% Sulfuric acid pretreatment.

4.3.9. The effect of 5bar, 2.0% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 5 bar, 2.0% Sulfuric acid at different retention times

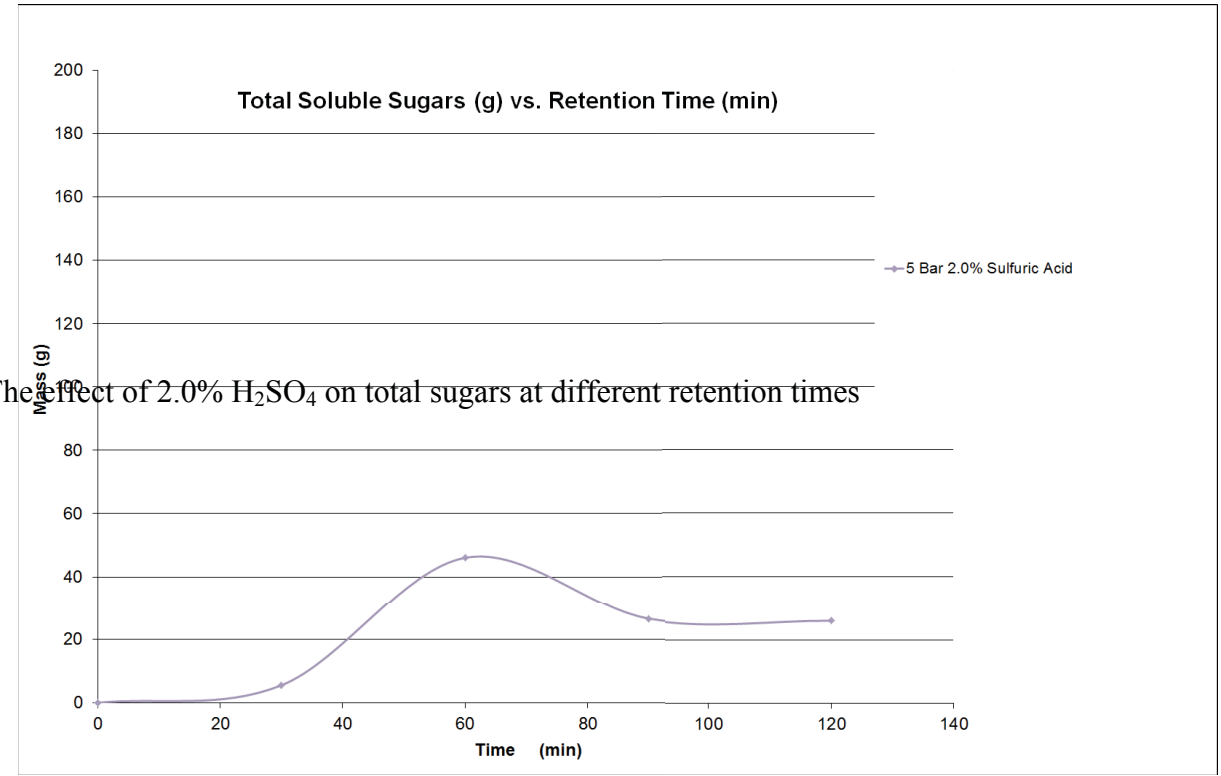


Figure 4.62 The effect of 2.0% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 46.1g at around 60 minutes retention time at 5 bar with 2.0% sulfuric acid pretreatment as seen in figure 4.62.

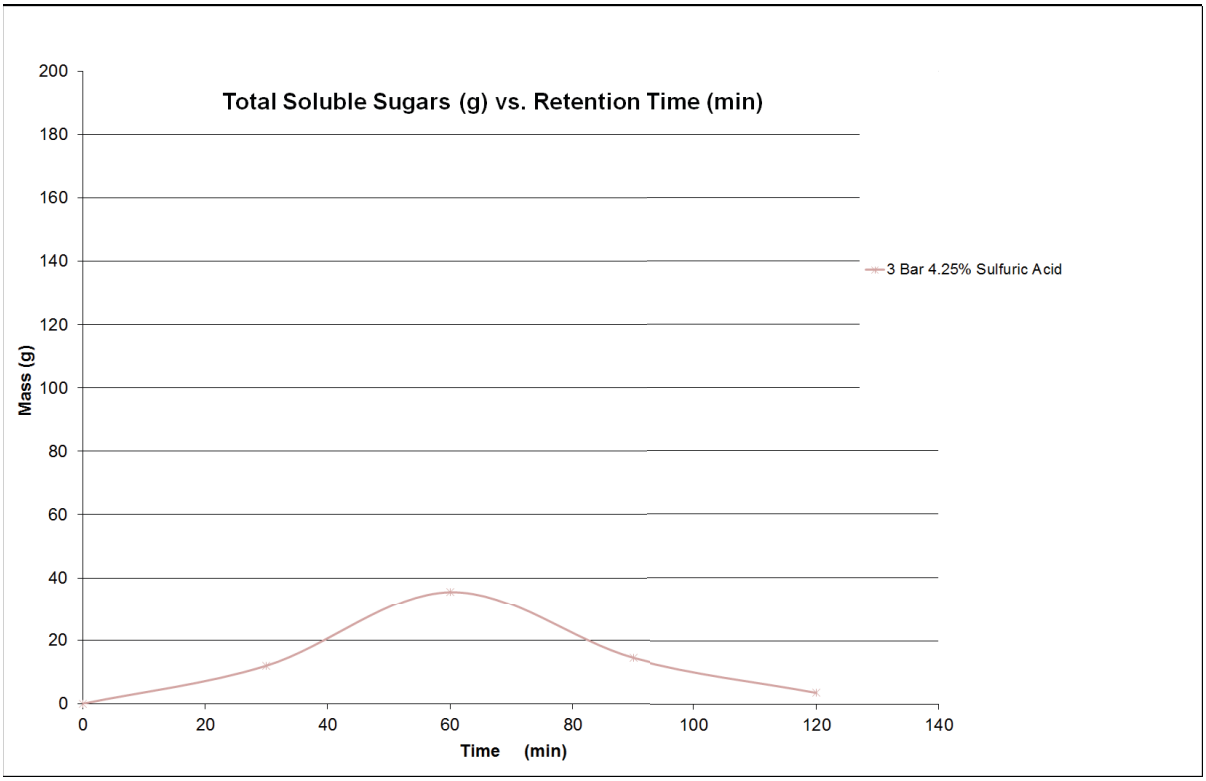
The graph indicates the peak value will be within 50 to 70 minutes of retention time.

The results also indicate low to moderate yield of sugars with the treatment at the pressure of 5 bar and 2.0% Sulfuric acid pretreatment.

Figure 4.63 The effect of 3bar, 4.25% H_2SO_4 on total sugars at different retention times

4.3.10. The effect of 3bar, 4.25% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 3 bar, 4.25% Sulfuric acid at different retention times



Total Soluble Sugars achieved highest value of 35.5g at around 60 minutes retention time at 3 bar with 4.25% sulfuric acid pretreatment as seen in figure 4.63.

The graph indicates the peak value will be within 50 to 80 minutes of retention time.

The results also indicate low to moderate yield of sugars with the treatment at the pressure of 3 bar and 4.25% Sulfuric acid pretreatment.

4.3.11. The effect of 4bar, 4.25% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 4 bar, 4.25% Sulfuric acid at different retention times

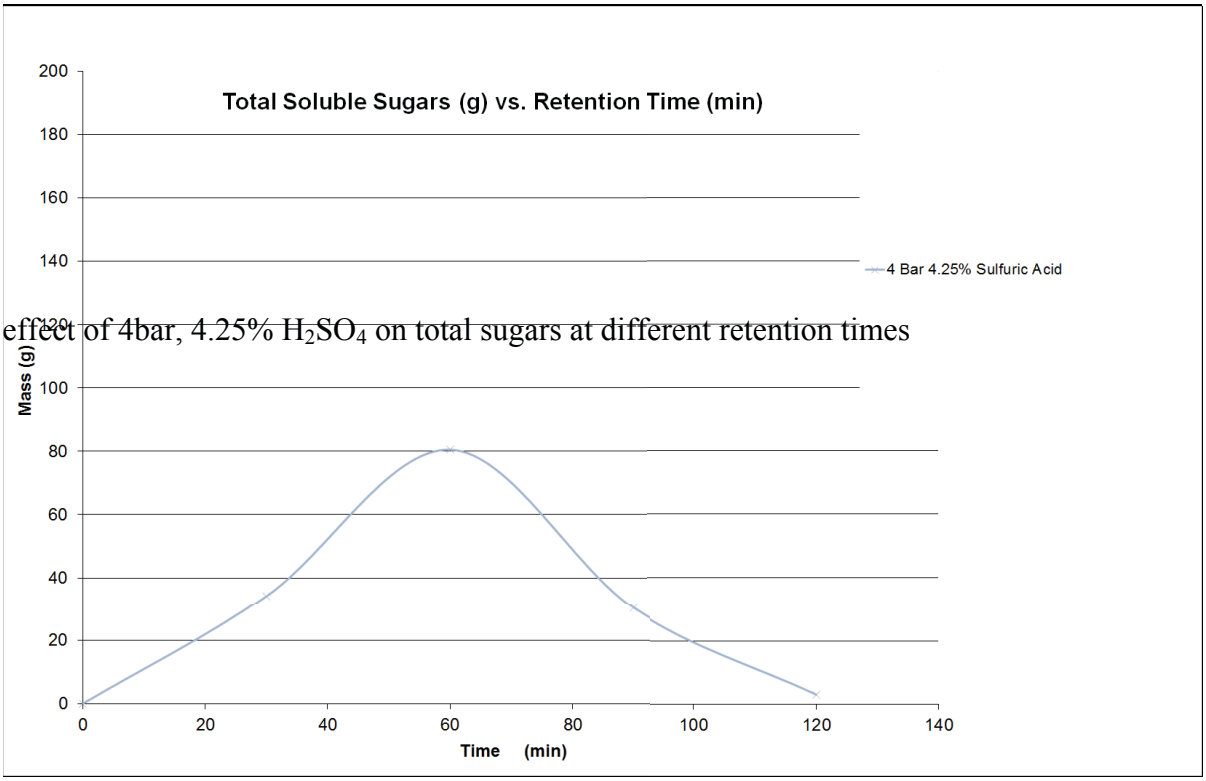


Figure 4.64 The effect of 4bar, 4.25% H_2SO_4 on total sugars at different retention times

Total Soluble Sugars achieved highest value of 80.5g at around 60 minutes retention time at 4 bar with 4.25% sulfuric acid pretreatment as seen in figure 4.64.

The graph indicates the peak value will be within 50 to 70 minutes of retention time.

The results also indicate medium to moderate yield of sugars with the treatment at the pressure of 4 bar and 4.25% Sulfuric acid pretreatment.

4.3.12. The effect of 5bar, 4.25% H_2SO_4 on total sugars vs. retention time.

Resulting total soluble sugars for input parameters of 5 bar, 4.25% Sulfuric acid at different retention times

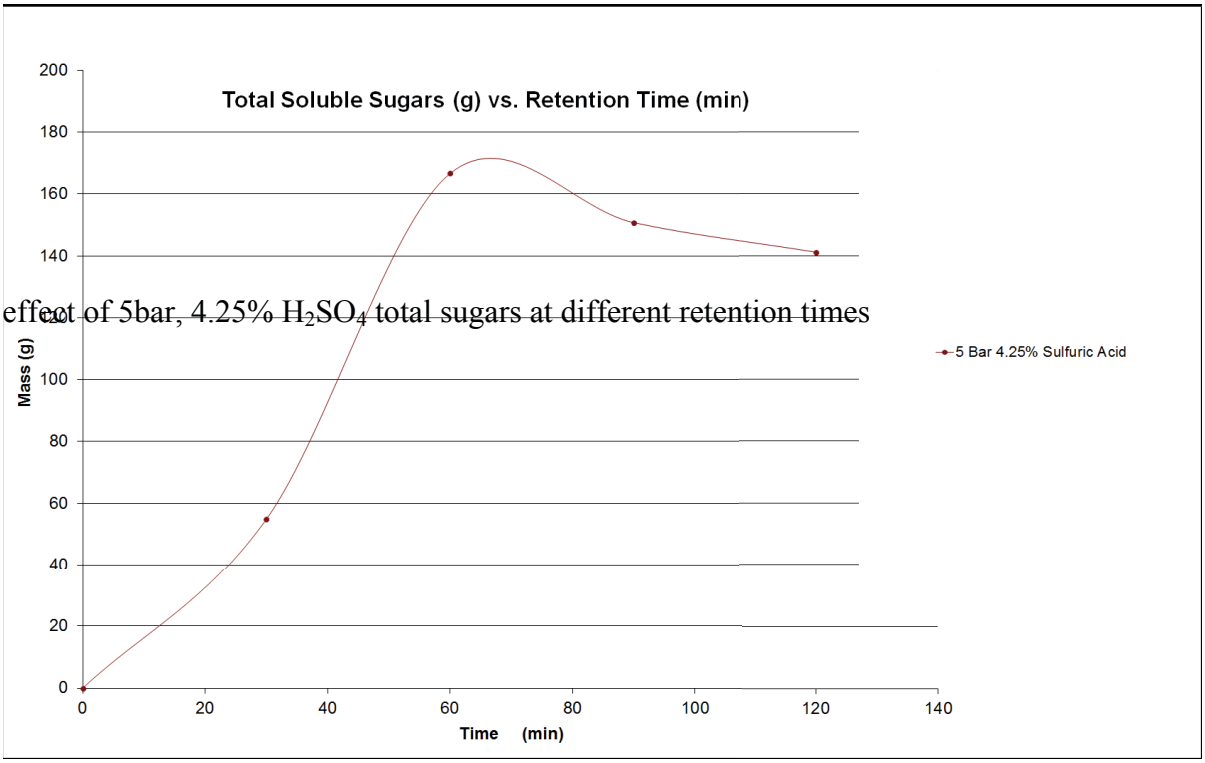


Figure 4.65 The effect of 5bar, 4.25% H_2SO_4 total sugars at different retention times

Total Soluble Sugars achieved highest value of 166.6g at around 60 minutes retention time at 5 bar with 4.25% sulfuric acid pretreatment as seen in figure 4.65.

The graph indicates the peak value will be within 50 to 80 minutes of retention time.

The results also indicate high yield of sugars with the treatment at the pressure of 5 bar and 4.25% Sulfuric acid pretreatment.

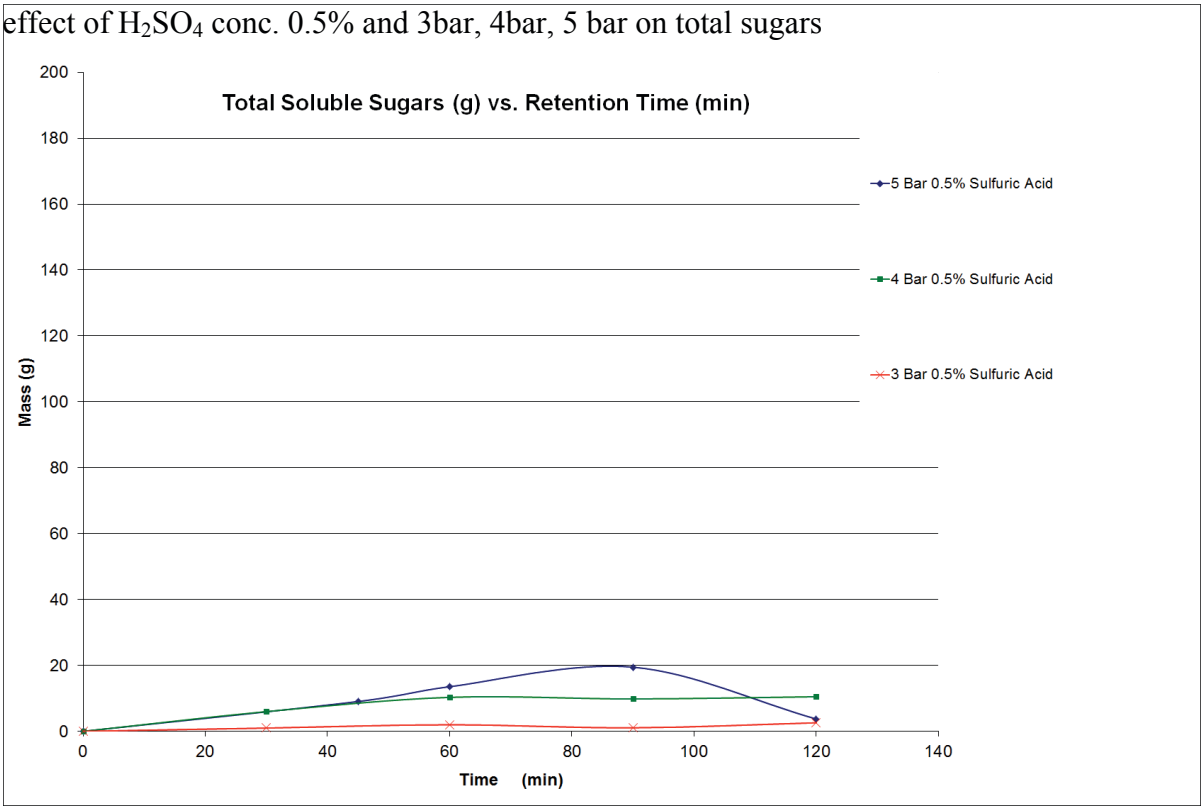
4.4 The effect of pretreatment acid % at different pressures

It is noticeable that pretreatment with different acid concentrations resulted in different soluble sugars production after treatment. In general low concentrations of acid resulted in low sugar yields at different pressures while higher concentrations of acid resulted in higher soluble sugars production except for 1.0% acid concentration.

4.4.1. The effect of 0.5% H_2SO_4 at 3,4 & 5bar on total sugars vs. retention time.

For the condition of 0.5% Sulfuric Acid pretreatment at retention times of 30, 60, 90 and 120 minutes results are shown at different treatment pressures of 3, 4 and 5 bar.

Figure 4.66 The effect of H_2SO_4 conc. 0.5% and 3bar, 4bar, 5 bar on total sugars



At 0.5% Sulfuric Acid, the highest yield is at around 90 minutes with 19.5g of sugars

The highest yield achieved is 4.87% produced sugars from 400g of rice straw as seen in figure 4.66.

Lower yield of 0.93% is the result of treatment at the same pressure of 5 bar but at a higher retention time of 120 minutes. This indicates that the rate of sugar production is lower than the rate of sugar loss from 90 to 120 minutes retention time.

4.4.2. The effect of 1.0% H_2SO_4 at 3,4 & 5bar on total sugars vs. retention time.

For the condition of 1.0% Sulfuric Acid pretreatment at retention times of 30, 60, 90 and 120 minutes results are shown at different treatment pressures of 3, 4 and 5 bar.

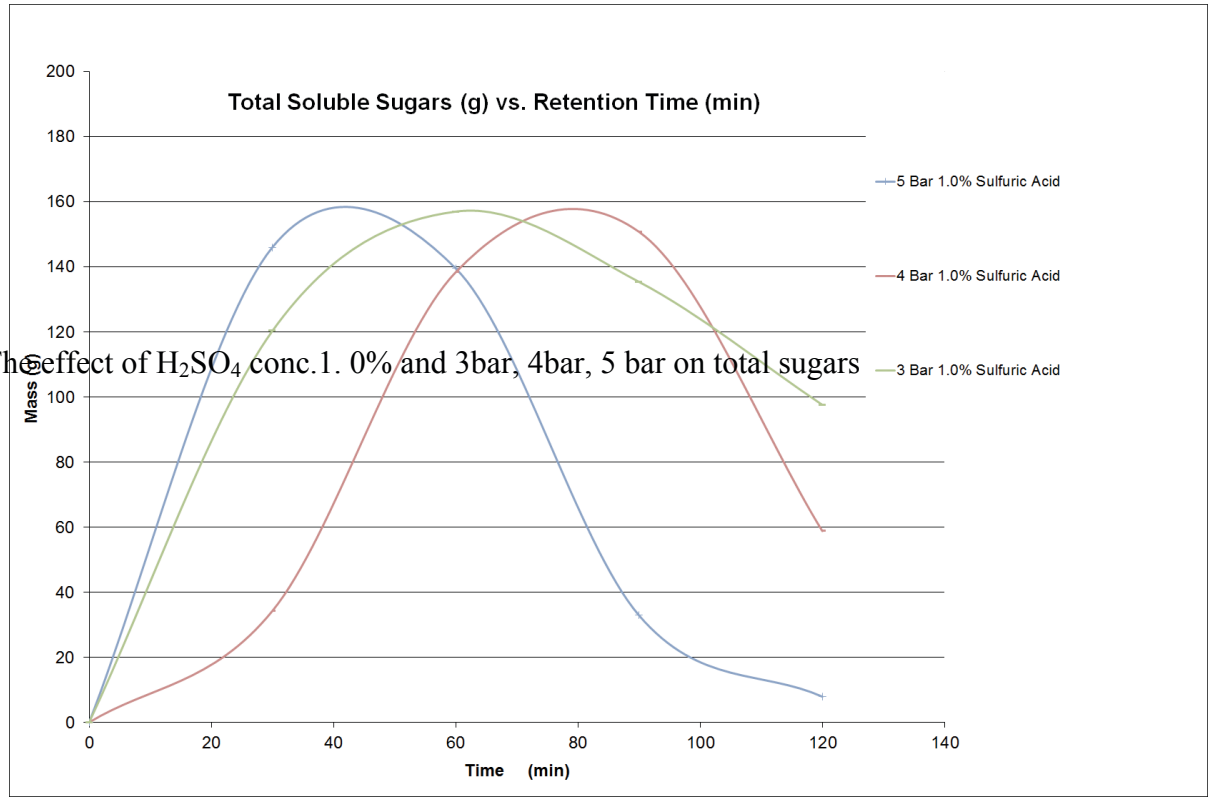


Figure 4.67 The effect of H_2SO_4 conc.1. 0% and 3bar, 4bar, 5 bar on total sugars

At 1.0% Sulfuric Acid, the highest yield is at around 60 minutes with 156.95g of sugars. The highest yield achieved is 39.24% produced sugars from 400g of rice straw as seen in figure 4.67.

Lower yield of 33.84% is the result of treatment at 1.0% and 5 bar but at a higher retention time of 90 minutes. This indicates that the rate of sugar production is lower than the rate of sugar loss from 60 to 90 minutes retention time resulting in 5.4% sugar loss at 1% and 5 bar. Further sugar loss of 9.45% is the result of higher retention time from 90 to 120 minutes.

4.4.3. The effect of 2.0% H_2SO_4 at 3,4 & 5bar on total sugars vs. retention time.

For the condition of 2.0% Sulfuric Acid pretreatment at retention times of 30, 60, 90 and 120 minutes results are shown at different treatment pressures of 3, 4 and 5 bar.

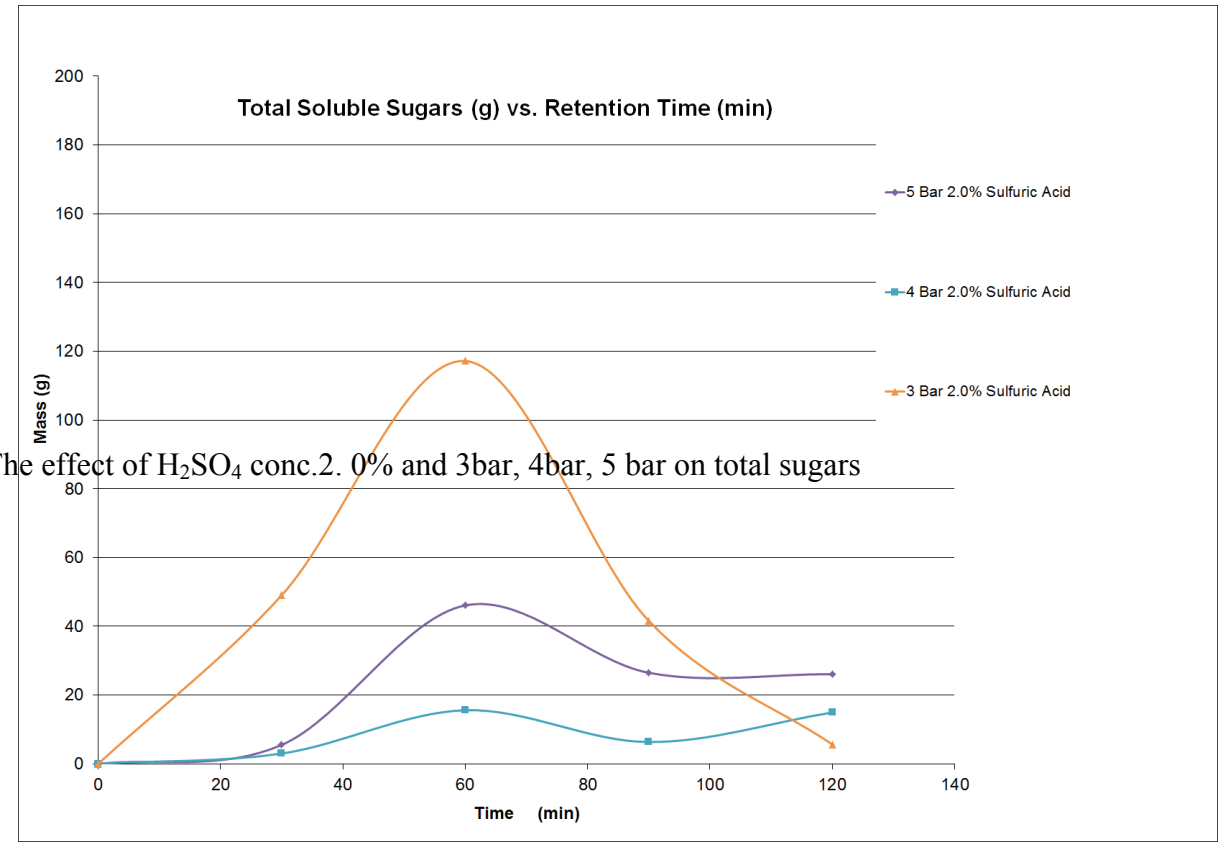


Figure 4.68 The effect of H_2SO_4 conc.2. 0% and 3bar, 4bar, 5 bar on total sugars

At 2.0% Sulfuric Acid, the highest yield is at around 60 minutes with 117.17g of sugars. The highest yield achieved is 29.29% produced sugars from 400g of rice straw as seen in figure 4.68.

Lower yield of 10.39% is the result of treatment at 2.0% and 3 bar but at a higher retention time of 90 minutes. This indicates that the rate of sugar production is lower than the rate of sugar loss from 60 to 90 minutes retention time resulting in 18.9% sugar loss at 2% and 3 bar. Further sugar loss of 9.98% is the result of higher retention time from 90 to 120 minutes.

4.4.4. The effect of 4.25% H_2SO_4 at 3,4 & 5bar on total sugars vs. retention time.

For the condition of 4.25% Sulfuric Acid pretreatment at retention times of 30, 60, 90 and 120 minutes results are shown at different treatment pressures of 3, 4 and 5 bar.

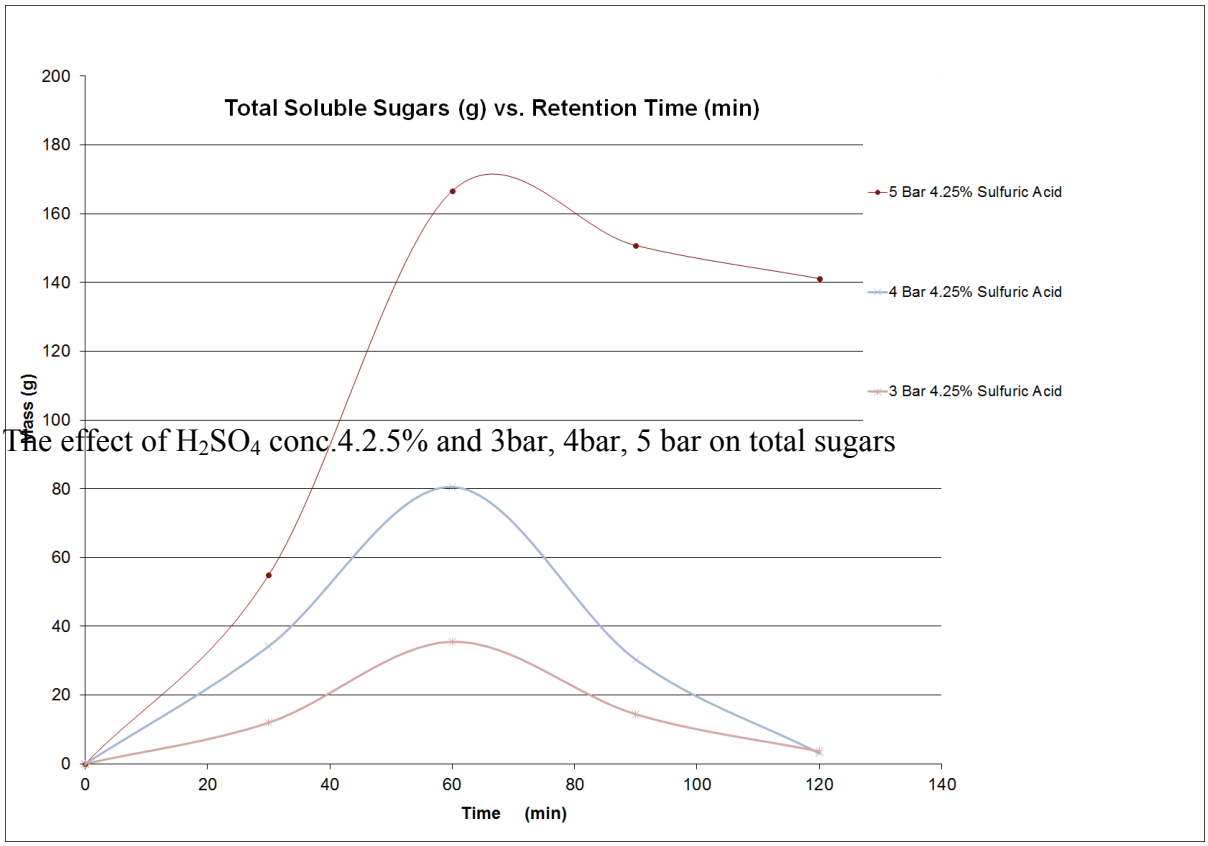


Figure 4.69 The effect of H_2SO_4 conc.4.2.5% and 3bar, 4bar, 5 bar on total sugars

At 4.25% Sulfuric Acid, 5 Bar, the highest yield is at 60 minutes with 166.63g of sugars. The highest yield achieved is 41.66% produced sugars from 400g of rice straw as seen in figure 4.69.

Lower yield of 37.69% is the result of treatment at 4.25% and 5 bar but at a higher retention time of 90 minutes. This indicates that the rate of sugar production is lower than the rate of sugar loss from 60 to 90 minutes retention time resulting in 3.97% sugar loss at4. 25% and 5 bar. Further sugar loss of 2.41% is the result of higher retention time from 90 to 120 minutes.

Table 4.53 Cost calculation at 0.5% H_2SO_4 at 3bar, 4 bar, and 5 bar at different reaction times

4.5 Financial calculation (LE per Kg Sugars produced):

It is important to relate the results not only the maximum amount of sugars produced, but also the amount of energy consumed. Equally important to include the amount of acid consumed to produce the sugars.

4.5.1 Financial calculation using 0.5% H2SO4 acid.

Calculations show that the cost per Kg of sugars produced using 0.5% Sulfuric Acid concentration is very high due to the low yield of sugars as shown in table 4.53. Both the cost of energy and the cost of the acid are very high compared to the low yield of sugars. It is shown that the lowest cost per Kg of produced sugars is 37.92LE/Kg at 0.5% Sulfuric Acid, 90 min retention time, and 5 Bar Pressure as seen in table 4.53.

Reaction Pressure (P)	Reaction Time Retention (min)	Acid %	Total Soluble sugars (g)	Energy Consumption per Sugars produced (Kw.hr/Kg)	Energy Cost (LE/Kw.hr)	Energy Cost per unit Sugar (LE/Kg)	Acid per sample (ml)	Acid Cost LE/ unit Sugar Produced (LE/Kg)	Cost per unit Sugar (Energy + Acid) (LE/Kg)
5	45	0.5%	9.06	124.130	0.30	37.24	16.00	19.42	56.66
5	60	0.5%	13.55	110.697	0.30	33.21	16.00	12.99	46.20
5	90	0.5%	19.48	96.267	0.30	28.88	16.00	9.04	37.92
5	120	0.5%	3.74	601.626	0.30	180.49	16.00	47.06	227.55
4	30	0.5%	6.00	124.995	0.30	37.50	16.00	29.33	66.83
4	60	0.5%	10.32	145.379	0.30	43.61	16.00	17.06	60.67
4	90	0.5%	9.84	190.556	0.30	57.17	16.00	17.89	75.05
4	120	0.5%	10.50	214.200	0.30	64.26	16.00	16.76	81.02
3	30	0.5%	1.00	752.877	0.30	225.86	16.00	176.68	402.54
3	60	0.5%	2.08	721.154	0.30	216.35	16.00	84.62	300.96
3	90	0.5%	1.08	1742.904	0.30	522.87	16.00	163.60	686.47
3	120	0.5%	2.60	866.530	0.30	259.96	16.00	67.78	327.74

Excessively high cost per unit sugar in LE/Kg is at 90 minutes retention time and 3 bar pressure at 0.5% Sulfuric Acid pretreatment. The cost is 686LE/Kg mainly driven by the low sugar production and the long retention time resulting in high energy consumption. It is therefore not recommended to produce sugars under these conditions due to negative energy balance resulting in negative financials while the intention is to produce a higher value product financially and a potential sustainable fuel / energy substitute for downstream processes.

Cost of acid consumption per unit sugar produced can be improved if the remaining acid is recycled. However this amount is neglected at this phase to show the full cost per unit. Acid recycling and other process enhancements are excluded from this study.

Table 4.54 Cost calculation at 1.0% H_2SO_4 at 3bar, 4 bar, and 5 bar at different reaction times

4.5.2 Financial calculation using 1.0% H2SO4 acid.

The cost per Kg of sugars produced using 1.0% Sulfuric Acid concentration is low due to the high yield of sugars.

Both the cost of energy and the cost of the acid are very low due to the high yield of sugars.

The lowest cost per Kg of produced sugars is 3.9LE/Kg at 1.0% Sulfuric Acid, 30 min retention time, and 5 Bar Pressure as seen in table 4.54.

Reaction Pressure (P)	Reaction Time Retention (min)	Acid %	Total Soluble sugars (g)	Energy Consumption per Sugars produced (Kw.hr/Kg)	Energy Cost (LE/Kw.hr)	Energy Cost per unit Sugar (LE/Kg)	Acid per sample (ml)	Acid Cost LE/ unit Sugar Produced (LE/Kg)	Cost per unit Sugar (Energy + Acid) (LE/Kg)
5	30	1.0%	145.99	5.137	0.30	1.54	31.00	2.34	3.88
5	60	1.0%	139.49	10.753	0.30	3.23	31.00	2.44	5.67
5	90	1.0%	32.87	57.036	0.30	17.11	31.00	10.37	27.48
5	120	1.0%	7.92	284.223	0.30	85.27	31.00	43.08	128.34
4	30	1.0%	34.28	21.880	0.30	6.56	31.00	9.95	16.51
4	60	1.0%	138.33	10.844	0.30	3.25	31.00	2.47	5.72
4	90	1.0%	150.73	12.440	0.30	3.73	31.00	2.26	5.99
4	120	1.0%	58.78	38.277	0.30	11.48	31.00	5.80	17.28
3	30	1.0%	120.30	6.234	0.30	1.87	31.00	2.83	4.70
3	60	1.0%	156.95	9.557	0.30	2.87	31.00	2.17	5.04
3	90	1.0%	135.35	13.853	0.30	4.16	31.00	2.52	6.68
3	120	1.0%	97.56	23.062	0.30	6.92	31.00	3.50	10.41

Excessively high cost per unit sugar in LE/Kg is at 120 minutes retention time and 5 bar pressure at 1.0% Sulfuric Acid pretreatment. The cost is 128LE/Kg mainly driven by the low sugar production and the long retention time resulting in high energy consumption.

It is therefore not recommended to produce sugars under these conditions.

It is also possible to consider acid recycling to reduce cost in later phases.

Table 4.55 Cost calculation at 2.0% H_2SO_4 at 3bar, 4 bar, and 5 bar at different reaction times

4.5.3 Financial calculation using 2.0% H2SO4 acid.

The cost per Kg of sugars produced using 2.0% Sulfuric Acid concentration is high due to the medium yield of sugars.

Both the cost of energy and the cost of the acid are high due to the medium yield of sugars.

The lowest cost per Kg of produced sugars is 9.7LE/Kg at 2.0% Sulfuric Acid, 60 min retention time, and 5 Bar Pressure as seen in table 4.55.

Reaction Pressure (P)	Reaction Time Retention (min)	Acid %	Total Soluble sugars (g)	Energy Consumption per Sugars produced (Kw.hr/Kg)	Energy Cost (LE/Kw.hr)	Energy Cost per unit Sugar (LE/Kg)	Acid per sample (ml)	Acid Cost LE/ unit Sugar Produced (LE/Kg)	Cost per unit Sugar (Energy + Acid) (LE/Kg)
5	30	2.0%	5.50	136.474	0.30	40.94	62.00	124.10	165.04
5	60	2.0%	46.07	32.557	0.30	9.77	62.00	14.80	24.57
5	90	2.0%	26.47	70.845	0.30	21.25	62.00	25.77	47.02
5	120	2.0%	26.07	86.318	0.30	25.90	62.00	26.16	52.06
4	30	2.0%	2.98	251.471	0.30	75.44	62.00	228.67	304.11
4	60	2.0%	15.58	96.283	0.30	28.88	62.00	43.78	72.66
4	90	2.0%	6.33	355.599	0.30	106.68	62.00	107.79	214.47
4	120	2.0%	14.93	150.681	0.30	45.20	62.00	45.67	90.88
	0								
3	30	2.0%	49.04	15.295	0.30	4.59	62.00	13.91	18.50
3	60	2.0%	117.17	12.801	0.30	3.84	62.00	5.82	9.66
3	90	2.0%	41.56	45.115	0.30	13.53	62.00	16.41	29.94
3	120	2.0%	5.65	398.532	0.30	119.56	62.00	120.80	240.36

Excessively high cost per unit sugar in LE/Kg is at 30 minutes retention time and 4 bar pressure at 2.0% Sulfuric Acid pretreatment. The cost is 304LE/Kg mainly driven by the low sugar production and the long retention time resulting in high energy consumption.

It is therefore not recommended to produce sugars under these conditions.

It is also possible to consider acid recycling to reduce cost in later phases.

Table 4.56 Cost calculation at 4.25% H_2SO_4 at 3bar, 4 bar, and 5 bar at different reaction times

4.5.4 Financial calculation using 4.25% H2SO4 acid.

The cost per Kg of sugars produced using 4.25% Sulfuric Acid concentration is high due to the high consumption of acid and the high cost of the acid.

Both the cost of energy and the cost of the acid are high even though there are samples with high yield of sugars.

The lowest cost per Kg of produced sugars is 11.2LE/Kg at 4.25% Sulfuric Acid, 60 min retention time, and 5 Bar Pressure as seen in table 4.56.

Reaction Pressure (P)	Reaction Time Retention (min)	Acid %	Total Soluble sugars (g)	Energy Consumption per Sugars produced (Kw.hr/Kg)	Energy Cost (LE/Kw.hr)	Energy Cost per unit Sugar (LE/Kg)	Acid per sample (ml)	Acid Cost LE/ unit Sugar Produced (LE/Kg)	Cost per unit Sugar (Energy + Acid) (LE/Kg)
5	30	4.25%	54.90	13.66	0.30	4.10	130.00	26.05	30.15
5	60	4.25%	166.63	9.00	0.30	2.70	130.00	8.58	11.28
5	90	4.25%	150.74	12.44	0.30	3.73	130.00	9.49	13.22
5	120	4.25%	141.13	15.94	0.30	4.78	130.00	10.13	14.91
4	30	4.25%	34.18	21.941	0.30	6.58	130.00	41.83	48.42
4	60	4.25%	80.52	18.630	0.30	5.59	130.00	17.76	23.35
4	90	4.25%	30.32	61.839	0.30	18.55	130.00	47.16	65.71
4	120	4.25%	3.05	738.496	0.30	221.55	130.00	469.36	690.90
3	30	4.25%	11.96	62.726	0.30	18.82	130.00	119.60	138.41
3	60	4.25%	35.48	42.274	0.30	12.68	130.00	40.30	52.98
3	90	4.25%	14.40	130.178	0.30	39.05	130.00	99.28	138.34
3	120	4.25%	3.59	626.626	0.30	187.99	130.00	398.26	586.24

Excessively high cost per unit sugar in LE/Kg is at 120minutes retention time and 4 bar pressure at 4.25% Sulfuric Acid pretreatment. The cost is 691LE/Kg mainly driven by the low sugar production and the long retention time resulting in high energy consumption.

It is therefore not recommended to produce sugars under these conditions.

It is also possible to consider acid recycling to reduce cost in later phases.

CHAPTER (5)
CONCLUSION

The hydrolysis process can be made economically sustainable by operating at the most economical conditions for Pretreatment with H_2SO_4 acid % concentration, treatment pressure, retention time for reaction, and resulting Cost. The condition of 1.0% H_2SO_4, 5 bar, 30 min provided cost per unit sugars produced of 3.88 LE/Kg (no cost reduction methods implemented namely acid recovery/reuse and insulation/heat loss reduction). While the market price is on average 6.25LE/Kg (market price is for reference only, processing and bleaching included). Other operating conditions also provided economical results as per the following ranking:

1- 1.0% H_2SO_4, 5 bar, 30 min, 3.88LE/Kg
2- 1.0% H_2SO_4, 3 bar, 30 min, 4.70LE/Kg
3- 1.0% H_2SO_4, 3 bar, 60 min, 5.04LE/Kg
4- 1.0% H_2SO_4, 5 bar, 60 min, 5.67LE/Kg
5- 1.0% H_2SO_4, 4 bar, 60 min, 5.72LE/Kg
6- 1.0% H_2SO_4, 4 bar, 90 min, 5.99LE/Kg

Treatment conditions of 0.5% H_2SO_4 provided non economical results due to the low yields of sugars and high energy consumption per unit sugar produced.

Treatment conditions of 2.0% H_2SO_4 provided non economical results due to the high acid consumption cost per unit sugar produced.

Treatment conditions of 4.25% H_2SO_4 also provided non economical results due to the high acid consumption cost per unit sugar produced.

ernorate	Feddan Cultivated RS	% Area
hlia	437,539	30.73%
ıia	271,237	19.05%
El Sheikh	255,098	17.91%
ira	195,758	13.75%
ɔia	161,731	11.36%
etta	64,777	4.55%
um	20,241	1.42%
ubia	17,566	1.23%

le 6.1 Rice straw cultivation area per governorate

CHAPTER (6)

RECOMENDATIONS

6.1 Soluble sugars production from Rice straw:

It is recommended to study treatment of the resulting soluble sugars to be utilized in a downstream process to be converted into a biofuel through genetically engineered microorganisms.

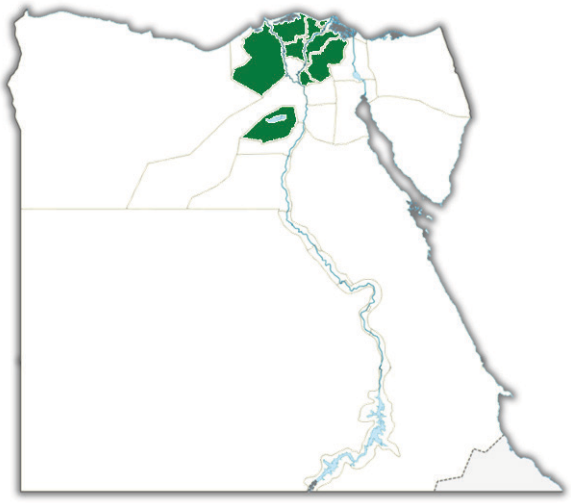
It is also recommended to study further treatment of the degraded rice straw after steam blasting to achieve higher yields of sugars production. There is great potential to achieve more sugar yields as a result of the chemical and thermal treatment.

Sugar production from rice straw proves to be a potential method to obtain low cost soluble sugars in a way that does not compete with food crops. Being a by- product of rice production, rice straw is already available in large quantities that are already available in many governorates in Egypt.

6.2 Potential useful application- National Project:

A national project needs to be put into place with segmented, decentralized layout to allow for processing Rice Straw in each governorate while minimizing transportation cost.

An estimate of a startup national project.



8 governorates will require a matrix of treatment sites as seen in table 1. The maximum distance is recommended to be 50Km maximum from the field to the treatment

site. "As a rule of thumb, transportation distances beyond a 25 to 50Km radius (depending on local infrastructure) are uneconomical." (Butchaiah 2011)

From the calculations, 42% sugars by weight can be recovered from the treated rice straw. It is important to note that degraded (treated) rice straw is a more valuable material than untreated rice straw because it is more readily composted, or used in mushroom production due to chemical and thermal treatment process etc.

Egypt's outlook for 2012 is 9 to 12 million tones of rice straw production based on calculations according to FAO forecasts. On average 10.5million tones of rice straw will be available. The processing capacity needed per working day is 43.75tons daily assuming 5 working days per week, and 2x8hr shifts and 21days holidays annually. The number of treatment vessels required to process this amount of rice straw on a daily basis is 81 vessels provided the 3m radius and 6m height. The highest cost effective yield obtained was at 60min treatment time and 5bar pressure with 1% Sulfuric Acid concentration pretreatment. Using the result of 39% Sugars recovery The yield is 4.1million tones soluble sugars with a potential of 25.6 billion Egyptian pounds in revenues per year.

Treatment of Rice Straw for sugars production will not be economical if significant transportation is employed to reach a centralized processing facility. Treatment facilities need to be spread out in a matrix layout around the producing governorates (reactors, soaking tanks, storage area, boilers)

Each reactor will require a number of soaking tanks to prepare the rice straw 24hr pretreatment soaking in acid solution. For the purpose of this study the number of saoking tanks needed is 40 due to retention time difference and number of shifts per day vs the 1 day capacity for the soaking tank. Each soaking tank needs 3 workers for preparation, loading and followup. Two eight hour shifts are required to cover daily production resulting in direct labor of 9671 personell. Also two eight hour shifts are required to operate the reacors with 4 presonell resulting in direct labor of 322 personell. Finally the buffer area, each worker can cover 200m2 area per day, resulting in potential direct labor of 65 personell.

Table 6.2 summarizes the inputs and calculations for a nationwide Processing Facilites to be located in each governorate. The facilitie will require a spread out network rather than a centralized headquater in order to decrease bulky transportation costs from the fields to the processing sides.

Rice Straw availability		
From	9,000,000,000	Kg
To	12,000,000,000	Kg
# of Facilities Calculation:		
Total Facility capacity /year	10,500,000,000	Kg / year
Processing days (365-104-21)	240	days
Facility capacity / day	43,750,000	Kg / day
Treatment Vessle Dimentions:		
R =	3	m
H =	6	m
t=	0.008	m
Treatment Vessle Costs:		
$V_{Stainless\ Steel}$ =	0.905	m^3
m_{vessle} =	6333	Kg
m_{other} =	1900	Kg
$Cost\ Material_{Stainless\ Steel}$ =	200	LE/kg
$Cost_{Vessle\ StSteel}$ =	1,646,697	LE
Treatment Vessle Volume:		
V_{Vessle} =	170	m^3
Extraction time =	0.5	hr
Volume Capacity $_{Vessle}$=	339	m^3/hr
Number of shifts per day =	2	shift /day
Shift Duration=	8	hr / shift
$\rho_{Rice\ Straw}$=	100	Kg/m^3
Mass Capacity $_{Vessle}$=	542,867	Kg /day
Country wide treatment capacity		
# of Vessles needed =	81	vessles
1 Vessle footprint area=	64	m^2
$Area_{Total\ Vessles}$ =	5,158	m^2
Total RS Volume =	437,500	m^3/day
Rice Straw Storage - Buffer Area		
h =	4	m
Stock in days =	120	days
Area $_{RS}$ =	13,125,000	m^2
Area $_{RS\ storage\ +\ Vessles}$=	13,130,158	m^2

Table 6.3 Facility calculation, Land costs, equipment costs, and labor costs.

Based on the previous assumptions and calculations, the 81 operational vessels can process the generated rice straw waste volume over the mentioned 8 governorates as follows in Table 6.3

Location	Feddan	% of Total	Treatment site area in Feddans	Facility Cost (Land+Site)	Equippement Cost	Land Price per Feddan	Land Cost	# of Reactors	Soaking tanks per reactor	# of soaking tanks	Vessle area per site (m^2)	Soaking tank area (m^2)	RS Buffer Area Per Site (m^2)
Qualubia	**17,566**	1.23%	39.2	35,313,908	19,645,292	400,000	15,668,616	**1**	**40**	**40**	**64**	**2,545**	**161,912**
Fayyium	**20,241**	1.42%	45.1	40,691,610	22,636,933	400,000	18,054,677	**1**	**40**	**46**	**73**	**2,933**	**186,568**
Damietta	**64,777**	4.55%	144.5	130,224,812	72,444,671	400,000	57,780,140	**4**	**40**	**147**	**235**	**9,385**	**597,071**
Gharbia	**161,731**	11.36%	360.7	325,136,839	180,875,143	400,000	144,261,696	**9**	**40**	**366**	**586**	**23,433**	**1,490,729**
Beheira	**195,758**	13.75%	436.5	393,543,212	218,929,929	400,000	174,613,284	**11**	**40**	**443**	**709**	**28,363**	**1,804,368**
Kafr El Sheikh	**255,098**	17.91%	568.9	512,837,720	285,294,021	400,000	227,543,699	**14**	**40**	**578**	**924**	**36,960**	**2,351,324**
Sharqia	**271,237**	19.05%	604.8	545,282,851	303,343,399	400,000	241,939,452	**15**	**40**	**614**	**982**	**39,299**	**2,500,083**
Daqahlia	**437,539**	30.73%	975.7	879,609,026	489,330,612	400,000	390,278,413	**25**	**40**	**991**	**1,585**	**63,394**	**4,032,945**
Total=	**1,423,947**		**3,175**	**2,862,639,977**	**1,592,500,000**		**1,270,139,977**	**81**		**3,224**	**5,158**	**206,312**	**13,125,000**

Labor=	322	9,671	65,625
			75,618
			Total

The result from rice straw processing activities is significant benefits to the local communities by employing 75,618 personnell and generating a substantial amount of industrial sugars that are a potential source raw material for biofuels.

For the process to be economical it is important to sell the produced sugars and sell the degraded rice straw at a competitive market price.

Assumptions: (market prices)

Rice straw from fields: 100LE/ton

Treated rice straw: 250LE/ton

Soluble Sugars 6,000LE/ton

Table 6.3 Facility calculation, revenues from produced sugars and soil conditioner and costs.

Table 6.4 Facility calculation, yearly direct labor costs, Transportation costs and treated RS.

6.2.1. National project benefits to rural economy:

As shown in table 6.4

1- Selling the rice straw waste, will be added income to the pesants.

100LE/ton x 10.5million tons = 1.05 billion LE/year

2- Tranporting the straw will be added income for truck drivers

60LE/ton x 10.5million tons = 630 million LE/year.

3- Job generation for processing labor at treatment sites is 1.13 billion LE/year in wages

4- 1100 vehicles will be needed on a daily basis for transportation securing an additional 3,300 jobs.

Location	Labor (Direct personell)	Labor cost / year	RS cost per ton (LE)	Treated tonns yearly (Ton)	Yearly RS cost (Treatment) (LE)	Transportation cost (LE/Ton)	Yearly Transportation cost (LE)	# of moves per vehicle daily	# of Vehicles needed daily (4 ton Veh.)
Qualubia	933	-13,992,550	-100	129,529	-12,952,940	-60	-7,771,764	10	13
Fayyium	1,075	-16,123,374	-100	149,255	-14,925,450	-60	-8,955,270	10	16
Damietta	3,440	-51,599,418	-100	477,657	-47,765,717	-60	-28,659,430	10	50
Gharbia	8,589	-128,830,071	-100	1,192,583	-119,258,336	-60	-71,555,002	10	124
Beheira	10,396	-155,934,960	-100	1,443,494	-144,349,403	-60	-86,609,642	10	150
Kafr El Sheikh	13,547	-203,203,427	-100	1,881,059	-188,105,948	-60	-112,863,569	10	196
Sharqia	14,404	-216,059,271	-100	2,000,066	-200,006,636	-60	-120,003,982	10	208
Daqahlia	23,235	-348,530,464	-100	3,226,356	-322,635,569	-60	-193,581,341	10	336
Total=	75,618	-1,134,273,536		10,500,000	-1,050,000,000		-630,000,000		1,094

On the other hand, as shown in table 6.5, enough soluble sugars and degraded rice straw will be produced to cover the costs of project startup and payback in a short period of time ranging from 5 months to 1 year.

Other costs include water and chemicals required for treatment predominantly Sulfuric acid. It is important to note that acid recovery and reuse is not considered in this calculation for simplicity. Acid recovery and reuse is a huge potential area of improvement that can significantly reduce the cost of producing sugars.

Location	Soluble sugar yearly output per	Soluble sugar yearly output per site (ton)	Prices sugar per ton (LE)	Soil Conditioner yearly output (Ton)	Soil Conditioner price (LE/Ton)	water consumption yearly (m^3)	water cost (LE/m^3)	water cost yearly (LE)	Acid consumption yearly (m^3)	Acid cost (LE/m^3)	Acid cost yearly (LE)
Qualubia	47,552	47,275	6000	82251.16876	250	971,470	-1.50	-1,457,206	9,838	-12,000	-118,053,095
Fayyium	47,552	54,474	6000	94776.60861	250	1,119,409	-1.50	-1,679,113	11,336	-12,000	-136,030,553
Damietta	47,552	174,333	6000	303312.3055	250	3,582,429	-1.50	-5,373,643	36,278	-12,000	-435,336,748
Gharbia	47,552	435,263	6000	757290.4346	250	8,944,375	-1.50	-13,416,563	90,577	-12,000	-1,086,920,476
Beheira	47,552	526,839	6000	916618.7119	250	10,826,205	-1.50	-16,239,308	109,633	-12,000	-1,315,600,463
Kafr El Sheikh	47,552	686,540	6000	1194472.768	250	14,107,946	-1.50	-21,161,919	142,866	-12,000	-1,714,397,608
Sharqia	47,552	729,974	6000	1270042.142	250	15,000,498	-1.50	-22,500,747	151,905	-12,000	-1,822,860,485
Daqahlia	47,552	1,177,539	6000	2048735.861	250	24,197,668	-1.50	-36,296,501	245,042	-12,000	-2,940,500,572
Total=		3,832,238		6,667,500		78,750,000		-118,125,000	797,475		-9,569,700,000

Table 6.6 Project baseline balance sheet, costs revenues and assumption values

Baseline calculation is needed to conduct sensitivity analysis should market prices fluctuate. The payback period of the project will vary according to the affected assumptions and fluctuations.

Market Price		Costs: EGP		
1.50	LE/m3	Water	-118,125,000	1%
1,250	LE/month	Labor	-1,134,273,536	6%
100	LE/m3	RS For treatment	-1,050,000,000	6%
1,541	LE/Ton	Electricity	-5,906,266,771	32%
60	LE/Ton	Transportation	-630,000,000	3%
12,000	LE/m3	Chemicals	-9,569,700,000	52%
		Total=	**-18,408,365,307**	

Market Price		Revenues: EGP		
6,000	LE/Ton	Soluble sugars	22,993,425,000	93%
250	LE/Ton	Degraded RS	1,666,875,000	7%
		Total=	**24,660,300,000**	

Net Operating Profit	6,251,934,693	**25%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**5 Months**	

Given current market prices on cost and revenue items, the national project can generate 25% annual net operating profit as seen in table 6.6.

The cost of investment (facility and land) can be recovered in 5 months.

Costs:

The highest cost was found to be related to the chemicals used, primarily Sulfuric Acid amounting to 52% of total annual costs. The cost is calculated based on one time use of the acid without recycling or reuse.

The second highest cost is related to energy consumption amounting to 32% of total annual costs. The cost is calculated based on the highest yield of sugars at lowest energy and acid consumption.

The Third highest cost is related to Labor amounting to 6% of annual costs.

The fourth highest cost is related to Rice straw amounting to 6% of annual costs.

Revenues:

Soluble sugars account to 93% of revenues and degraded Rice Straw accounts for 7% of revenues. There is a potential to add ferrous sulfate as another salable byproduct in case an acid recovery / reuse unit is put into consideration for each site.

Table 6.7 Sensitivity Analysis, Project balance sheet, +10% on costs

6.2.2. Sensitivity analysis:

Using the baseline calculation in Table 6.6, sensitivity analysis is conducted to study the effect of increasing/decreasing costs vs. increasing/decreasing revenues on 1) annual profitability and 2) project payback period. The change percent used is -10%, -20%, +10% and -20%.

6.2.2.1. Costs Sensitivity:

Sensitivity: +10% on costs

Market Price		Costs: EGP		
1.65	LE/m3	Water	-129,937,500	1%
1,375	LE/month	Labor	-1,247,700,889	6%
110	LE/m3	RS For treatment	-1,155,000,000	6%
1,695	LE/Ton	Electricity	-6,496,893,448	32%
66	LE/Ton	Transportation	-693,000,000	3%
13,200	LE/m3	Chemicals	-10,526,670,000	52%
		Total=	**-20,249,201,838**	

Market Price		Revenues: EGP		
6,000	LE/Ton	Soluble sugars	22,993,425,000	93%
250	LE/Ton	Degraded RS	1,666,875,000	7%
		Total=	**24,660,300,000**	

Net Operating Profit Annual	4,411,098,162	**18%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**8 Months**	

Given 10% increase in current market prices on cost only, the national project can generate 18% annual net operating profit instead of 25% as seen in table 6.7.

The cost of investment (facility and land) can therefore be recovered in 8 months instead of 5 months.

Table 6.8 Sensitivity Analysis, Project balance sheet, +20% on costs

Sensitivity: +20% on costs

Market Price		Costs: EGP		
1.80	LE/m3	Water	-141,750,000	1%
1,500	LE/month	Labor	-1,361,128,243	6%
120	LE/m3	RS For treatment	-1,260,000,000	6%
1,849	LE/Ton	Electricity	-7,087,520,126	32%
72	LE/Ton	Transportation	-756,000,000	3%
14,400	LE/m3	Chemicals	-11,483,640,000	52%
		Total=	**-22,090,038,369**	

Market Price		Revenues: EGP		
6,000	LE/Ton	Soluble sugars	22,993,425,000	93%
250	LE/Ton	Degraded RS	1,666,875,000	7%
		Total=	**24,660,300,000**	

Net Operating Profit	2,570,261,631	**10%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**13 Months**	

Given 20% increase in current market prices on cost only, the national project can generate 10% annual net operating profit instead of 25% as seen in table 6.8.

The cost of investment (facility and land) can therefore be recovered in 13 months instead of 5 months.

Table 6.9 Sensitivity Analysis, Project balance sheet, -20% on costs

Sensitivity: -20% on costs

Market Price		Costs: EGP		
1.20	LE/m3	Water	-94,500,000	1%
1,000	LE/month	Labor	-907,418,829	6%
80	LE/m3	RS For treatment	-840,000,000	6%
1,233	LE/Ton	Electricity	-4,725,013,417	32%
48	LE/Ton	Transportation	-504,000,000	3%
9,600	LE/m3	Chemicals	-7,655,760,000	52%
		Total=	**-14,726,692,246**	

Market Price		Revenues: EGP		
6,000	LE/Ton	Soluble sugars	22,993,425,000	93%
250	LE/Ton	Degraded RS	1,666,875,000	7%
		Total=	**24,660,300,000**	

Net Operating Profit Annual	9,933,607,754	**40%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**3 Months**	

Given 20% decrease in current market prices on cost only, the national project can generate 40% annual net operating profit instead of 25% as seen in table 6.9.

The cost of investment (facility and land) can therefore be recovered in 3 months instead of 5 months.

Table 6.10 Sensitivity Analysis, Project balance sheet, -10% on costs

Sensitivity: -10% on costs

Market Price		Costs: EGP		
1.35	LE/m3	Water	-106,312,500	1%
1,125	LE/month	Labor	-1,020,846,182	6%
90	LE/m3	RS For treatment	-945,000,000	6%
1,387	LE/Ton	Electricity	-5,315,640,094	32%
54	LE/Ton	Transportation	-567,000,000	3%
10,800	LE/m3	Chemicals	-8,612,730,000	52%
		Total=	**-16,567,528,776**	

Market Price		Revenues: EGP		
6,000	LE/Ton	Soluble sugars	22,993,425,000	93%
250	LE/Ton	Degraded RS	1,666,875,000	7%
		Total=	**24,660,300,000**	

Net Operating Profit Annual	8,092,771,224	**33%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**4 Months**	

Given 10% decrease in current market prices on cost only, the national project can generate 33% annual net operating profit instead of 25% as seen in table 6.10.

The cost of investment (facility and land) can therefore be recovered in 4 months instead of 5 months.

Table 6.11 Sensitivity Analysis, Project balance sheet, +10% Revenues

6.2.2.2. Revenues Sensitivity:

Sensitivity: +10% on Revenues

Market Price		Costs: EGP		
1.50	LE/m3	Water	-118,125,000	1%
1,250	LE/month	Labor	-1,134,273,536	6%
100	LE/m3	RS For treatment	-1,050,000,000	6%
1,541	LE/Ton	Electricity	-5,906,266,771	32%
60	LE/Ton	Transportation	-630,000,000	3%
12,000	LE/m3	Chemicals	-9,569,700,000	52%
		Total=	**-18,408,365,307**	

Market Price		Revenues: EGP		
6,600	LE/Ton	Soluble sugars	25,292,767,500	93%
275	LE/Ton	Degraded RS	1,833,562,500	7%
		Total=	**27,126,330,000**	

Net Operating Profit Annual	8,717,964,693	**32%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**4 Months**	

Given 20% increase in current market prices on revenues only, the national project can generate 32% annual net operating profit instead of 25% as seen in table 6.11.

The cost of investment (facility and land) can therefore be recovered in 4 months instead of 5 months.

Table 6.12 Sensitivity Analysis, Project balance sheet, +20% Revenues

Sensitivity: +20% on Revenues

Market Price		Costs: EGP		
1.50	LE/m3	Water	-118,125,000	1%
1,250	LE/month	Labor	-1,134,273,536	6%
100	LE/m3	RS For treatment	-1,050,000,000	6%
1,541	LE/Ton	Electricity	-5,906,266,771	32%
60	LE/Ton	Transportation	-630,000,000	3%
12,000	LE/m3	Chemicals	-9,569,700,000	52%
		Total=	**-18,408,365,307**	

Market Price		Revenues: EGP		
7,200	LE/Ton	Soluble sugars	27,592,110,000	93%
300	LE/Ton	Degraded RS	2,000,250,000	7%
		Total=	**29,592,360,000**	

Net Operating Profit Annual	11,183,994,693	**38%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**3 Months**	

Given 20% increase in current market prices on revenues only, the national project can generate 38% annual net operating profit instead of 25% as seen in table 6.12.

The cost of investment (facility and land) can therefore be recovered in 3 months instead of 5 months.

Table 6.13 Sensitivity Analysis, Project balance sheet, -20% Revenues

Sensitivity: -20% on Revenues

Market Price		Costs: EGP		
1.50	LE/m3	Water	-118,125,000	1%
1,250	LE/month	Labor	-1,134,273,536	6%
100	LE/m3	RS For treatment	-1,050,000,000	6%
1,541	LE/Ton	Electricity	-5,906,266,771	32%
60	LE/Ton	Transportation	-630,000,000	3%
12,000	LE/m3	Chemicals	-9,569,700,000	52%
		Total=	**-18,408,365,307**	

Market Price		Revenues: EGP		
4,800	LE/Ton	Soluble sugars	18,394,740,000	93%
200	LE/Ton	Degraded RS	1,333,500,000	7%
		Total=	**19,728,240,000**	

Net Operating Profit Annual	1,319,874,693	**7%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**26 Months**	

Given 20% decrease in current market prices on revenues only, the national project can generate 7% annual net operating profit instead of 25% as seen in table 6.13.

The cost of investment (facility and land) can therefore be recovered in 26 months instead of 5 months.

Table 6.14 Sensitivity Analysis, Project balance sheet, -10% Revenues

Sensitivity: -10% on Revenues

Market Price		Costs: EGP		
1.50	LE/m3	Water	-118,125,000	1%
1,250	LE/month	Labor	-1,134,273,536	6%
100	LE/m3	RS For treatment	-1,050,000,000	6%
1,541	LE/Ton	Electricity	-5,906,266,771	32%
60	LE/Ton	Transportation	-630,000,000	3%
12,000	LE/m3	Chemicals	-9,569,700,000	52%
		Total=	**-18,408,365,307**	

Market Price		Revenues: EGP		
5,400	LE/Ton	Soluble sugars	20,694,082,500	93%
225	LE/Ton	Degraded RS	1,500,187,500	7%
		Total=	**22,194,270,000**	

Net Operating Profit Annual	3,785,904,693	**17%**
Facility + Cost of Land=	2,862,639,977	
Facility payback period=	**9 Months**	

Given 10% decrease in current market prices on revenues only, the national project can generate 17% annual net operating profit instead of 25% as seen in table 6.14.

The cost of investment (facility and land) can therefore be recovered in 9 months instead of 5 months.

Costs: EGP	**Costs Sensitivity**				
	-20%	-10%	0%	10%	20%
Water =	-94,500,000	-106,312,500	-118,125,000	-129,937,500	-141,750,000
Labor =	-907,418,829	-1,020,846,182	-1,134,273,536	-1,247,700,889	-1,361,128,243
RS For treatment =	-840,000,000	-945,000,000	-1,050,000,000	-1,155,000,000	-1,260,000,000
Electricity =	-4,725,013,417	-5,315,640,094	-5,906,266,771	-6,496,893,448	-7,087,520,126
Transportation =	-504,000,000	-567,000,000	-630,000,000	-693,000,000	-756,000,000
Chemicals =	-7,655,760,000	-8,612,730,000	-9,569,700,000	-10,526,670,000	-11,483,640,000
Total=	-14,726,692,246	-16,567,528,776	-18,408,365,307	-20,249,201,838	-22,090,038,369
Revenues: EGP					
Soluble sugars	22,993,425,000	22,993,425,000	22,993,425,000	22,993,425,000	22,993,425,000
Degraded RS	1,666,875,000	1,666,875,000	1,666,875,000	1,666,875,000	1,666,875,000
Total=	24,660,300,000	24,660,300,000	24,660,300,000	24,660,300,000	24,660,300,000
Net Operating Profit Annual	9,933,607,754	8,092,771,224	6,251,934,693	4,411,098,162	2,570,261,631
Annual Profitability	**40%**	**33%**	**25%**	**18%**	**10%**
Facility + Cost of Land=	2,862,639,977	2,862,639,977	2,862,639,977	2,862,639,977	2,862,639,977
Facility payback period=	3	4	5	8	13

Costs: EGP	**Revenues Sensitivity**				
	20%	10%	0%	-10%	-20%
Water =	-118,125,000	-118,125,000	-118,125,000	-118,125,000	-118,125,000
Labor =	-1,134,273,536	-1,134,273,536	-1,134,273,536	-1,134,273,536	-1,134,273,536
RS For treatment =	-1,050,000,000	-1,050,000,000	-1,050,000,000	-1,050,000,000	-1,050,000,000
Electricity =	-5,906,266,771	-5,906,266,771	-5,906,266,771	-5,906,266,771	-5,906,266,771
Transportation =	-630,000,000	-630,000,000	-630,000,000	-630,000,000	-630,000,000
Chemicals =	-9,569,700,000	-9,569,700,000	-9,569,700,000	-9,569,700,000	-9,569,700,000
Total=	-18,408,365,307	-18,408,365,307	-18,408,365,307	-18,408,365,307	-18,408,365,307
Revenues: EGP					
Soluble sugars	27,592,110,000	25,292,767,500	22,993,425,000	20,694,082,500	18,394,740,000
Degraded RS	2,000,250,000	1,833,562,500	1,666,875,000	1,500,187,500	1,333,500,000
Total=	**29,592,360,000**	27,126,330,000	24,660,300,000	22,194,270,000	19,728,240,000
Net Operating Profit Annual	11,183,994,693	8,717,964,693	6,251,934,693	3,785,904,693	1,319,874,693
Annual Profitability	**38%**	**32%**	**25%**	**17%**	**7%**
Facility + Cost of Land=	2,862,639,977	2,862,639,977	2,862,639,977	2,862,639,977	2,862,639,977
Facility payback period=	**3**	**4**	5	**9**	26

able 6.17 Alternative agriculture residues as energy sources (Su Y. Matsumura et al. 2005)

It is important to note that payback period for the project is more sensitive to revenues reduction than any other factor. It is also important to note that revenues reduction is an unlikely scenario because the increasing cost and price of edible sugar for human consumption.

Excluding the 70 months payback period, the next longest payback period is just below two years. For a project with such large magnitude two years proves to be a very short period of time for project payback.

6.2.3. Cost Reduction potential

Energy consumption cost reduction:

Alternative sources of energy:

The remote nature of the treatment sites will face a challenge in energy availability for the facilities to be operational. This challenge is an opportunity to capitalize on other sources of energy such as other agricultural wastes in order to meet the demands steam generation required for hydrolysis process.

Agriculture residue	(KwHr/Ton)	Recovery Thermal Efficiency (80%) (KwHr/Ton)
Barley	3,989	3,191
Rice Straw	3,224	2,579
Corn	2,465	1,972
Soybean	1,768	1,414
Sorghum	1,731	1,385
Sugarcane	881	705
Potato	514	411
Sweet Potato	508	406
Wheat	233	186

Some assumptions are to be made in order to estimate the amount of biomass to burn to produce enough energy for treatment. Using energy values from table 6.17, Assuming

Biofuel: Rice straw

Energy: 3224KwHr/ton

Table 6.18 Calculated alternative biomass to produce 1 ton sugar from RS

Thermal recovery Efficiency: 80%

Net Energy: 2579 KwHr/ton

Result show that the least amount of energy consumed to produce 1 ton of sugars is 5137Kw. The amount of agriculture residue of different types is calculated accordingly. Barley is by far the highest in energy when burned as a biofuel relative to other agriculture residues.

Agriculture residue	Amount to burn per ton of Sugars produced (Ton)
Barley	1.6
Rice Straw	2.0
Corn	2.6
Soybean	3.6
Sorghum	3.7
Sugarcane	7.3
Potato	12.5
Sweet Potato	12.6
Wheat	27.6

As an example by using rice straw as a source of biofuel energy, from table 6.18, we get 2 ton of rice straw are to be burned to treat 2.74 tons of rice straw and to produce 1 ton of sugar (36.5% yield at 30 min, 5 bar and 1% Sulfuric Acid). For every 4.74tons (2.74ton +2.0ton) of rice straw entering the treatment sites, 2 tons will be burned as a source of energy, and 2.74 tons will be treated and the gain is 1ton of sugars.

Chemicals consumption cost reduction:

Acid recovery / reuse:

Sulfuric acid can be recovered by mixing scrap iron or ferrous oar with the waste acid. This process produces ferrous sulfate slurry that precipitates from solution. Ferrous sulfate can be sold commercially, or a process to recover Sulfuric acid can be put in place on site.

The recovery process involves heating the produced sulfates in an oven to 1000^{o}C and roasting to form SO2. Catalysts are then utilized to convert SO_2 to SO_3. The produced SO3 is then recovered by absorbing in water to produce concentrated acid. The process. (Klotz)

CHALLENGES:

Many challenges were encountered during this project. The challenges need to be mitigated in future potential studies. 48 samples were prepared for testing. Each sample took many hours to produce (25.5hrs to 27hrs) and had to be transported 25km in refrigerated condition to be tested at the National Center for Research " المركز القومي للبحوث". Each sample took 30 to 45 minutes in the HPLC unit at a cost of 200LE/sample. With high testing cost to conduct the sampling plan, it was difficult to increase the number of samples as the current cost reached 10,600LE.

It is recommended to maintain close proximity of HPLC analysis facility to the sample production facility to save transportation time, and to safeguard sample contamination / biodegradation etc. It is recommended that AUC acquires HPLC testing equipment needed to support further development of such projects and higher sampling plans.

Regarding highest sugar production at lowest cost per unit weight sugar produced, it was found The lowest cost per Kg of produced sugars is 3.9LE/Kg at 1.0% Sulfuric Acid, 30 min retention time, and 5 Bar Pressure. It is recommended to expand the study in future researches to find better ways to produce more sugars with less energy consumption.

Due to the complexity of biochemical, chemical and mechanical processes involved, the study of rice straw hydrolysis needs a team of researchers to optimize upstream and downstream activities related to the process. From the chemical perspective, minimal and optimal use of acid, water and energy is needed. From the biochemical perspective little or no inhibitor by products are needed to hinder the fermentation process in downstream processes.

From the mechanical perspective, structural integrity, material selection and fluid mechanics are needed to ensure the safe operation and smooth flow of materials from one stage to the next. Much work is needed in all related fields in order to bring the process to life with cost effective results that can make it compete with fossil fuels.

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