Biomass Conversion and Biorefinery
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Lecture 11
Dilute Acid, Alkali, Ozone

Good morning students. Today is lecture 2 under module 4 and as you know that this module is dedicated to our Biomass pretreatment and we have discussed basics in the last class. Today we will discuss some of the most important pre-treatment processes such as dilute acid, alkali based pre-treatment, auto hydrolysis, Ozone based pre-treatment and few others.

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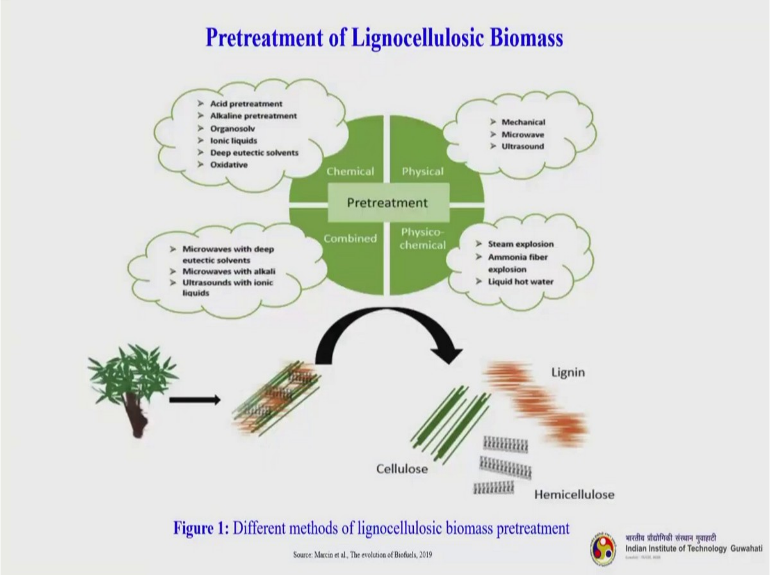

Pretreatment of Lignocellulosic Biomass

- Acid pretreatment
- Alkaline pretreatment
- Organosolv
- Ionic liquids
- Deep eutectic solvents
- Oxidative

- Mechanical
- Microwave
- Ultrasound

- Microwaves with deep eutectic solvents
- Microwaves with alkali
- Ultrasounds with ionic liquids

- Steam explosion
- Ammonia fiber explosion
- Liquid hot water

Figure 1: Different methods of lignocellulosic biomass pretreatment

Source: Marcin et al., The evolution of Biofuels, 2019

Let us start. As you know that pre-treatment of lignocellulosic biomasses can be carried out by various methods: chemical, physical, combined and physico-chemical. So, some of these we have discussed; some we're going to discuss today. So, under chemical: it is acid pre-treatment, alkaline, organosolv, ionic liquids, deep eutectic solvents. All these things we will be discussing today; physical we have already discussed.

So, in the combined pre-treatment it is actually with Microwave based pre-treatment, then Microwave with the alkali, Microwave with deep eutectic solvents, then ultrasound with ionic liquids, like this, basically a combination of two pre-treatment technologies. And then physicochemical: steam explosion, Ammonia fibre explosion, liquid hot water these things also we're going to discuss today.

So, the goal is again the same as we have already discussed; it is to release or defragment cellulose, Lignin and hemicellulose. So, remove Lignin and purify cellulose and hemicellulose so that they can be converted into various value-added products.

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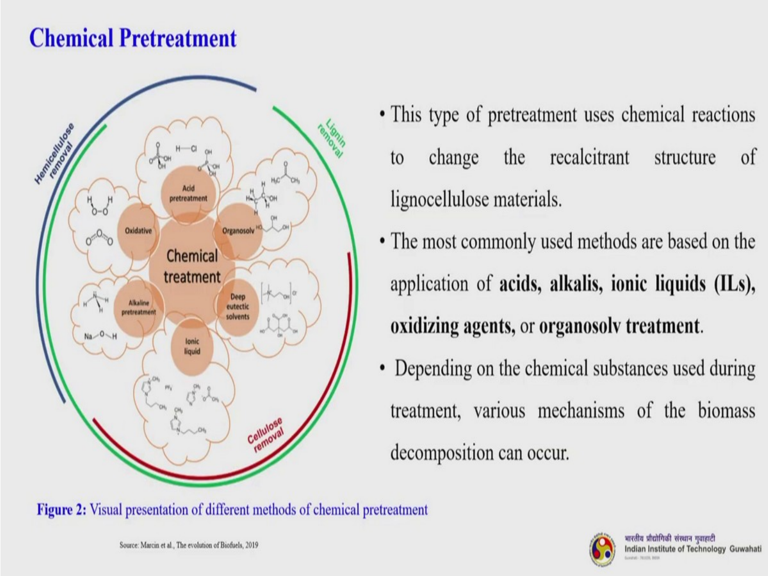

Chemical Pretreatment

- This type of pretreatment uses chemical reactions to change the recalcitrant structure of lignocellulose materials.
- The most commonly used methods are based on the application of **acids, alkalis, ionic liquids (ILs), oxidizing agents,** or **organosolv treatment**.
- Depending on the chemical substances used during treatment, various mechanisms of the biomass decomposition can occur.

Figure 2: Visual presentation of different methods of chemical pretreatment

Source: Marcin et al., The evolution of Biofuels, 2019

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Let us talk about chemical pre-treatment. This particular image is a beautiful image from the perspective that it can tell us how different types of chemical pre-treatment are there and how they overlap each other in a doing a particular job. Now, let us look at this particular green one. The green boundary here. You can see, that is almost taking into everything except little part of the acid pre-treatment.

With this green boundary pertains to the lignin removal. All these processes under this green they will do Lignin removal more efficiently than other processes. Similarly, if you see this red one, so that one is for the cellulose removal, whether ionic liquid or deep eutectic solvents, alkaline pretreatment, even part of little organosolv pretreatment also. Similarly, oxidative, acid pretreatment and alkaline. This takes care of the hemicellulose removal.

This type of pretreatment uses chemical reactions to change the recalcitrant structure of lignocellulosic materials. Most commonly used are acid, alkaline, ionic liquids, oxidizing agents and organosolv pretreatment. Now depending upon the chemical substances used during pretreatment various mechanisms of the Biomass decomposition can occur.

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Acidic Pretreatment

- Pretreatment of lignocelluloses with acids is one of the most effective methods of solubilizing the hemicelluloses, making cellulose more accessible.
- Acid pretreatment involves the use of **concentrated** and **diluted acids** to break the rigid structure of the lignocellulosic material.
- The main reactions during acid treatment are the hydrolysis of hemicellulose and condensation and precipitation of solubilized lignin.
- The most commonly used acid is ***dilute sulphuric acid*** (H_2SO_4), which has been commercially used to pre-treat a wide variety of biomass types - switchgrass, corn stover, spruce (softwood), and poplar.

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Let us understand acidic pretreatment: So, pretreatment of lignocelluloses with acids is one of the most effective method of solubilizing the hemicellulose making cellulose more accessible. Now acid pretreatment involves the use of concentrated and dilute acid both, to break the rigid structure of the lignocellulosic materials. The main reactions during acid pretreatment at the hydrolysis of hemicellulose and condensation and precipitation of the solubilized Lignin.

The most commonly used acid is the dilute sulphuric acid which has been commercially used to pre-treat a wide variety of biomass types whether it is switchgrass, cronstover, Spruce and poplar. So, this list is basically endless. If you see just type dilute acid pretreatment of lignocellulosic biomass there are hundreds and hundreds of excellent research papers available.

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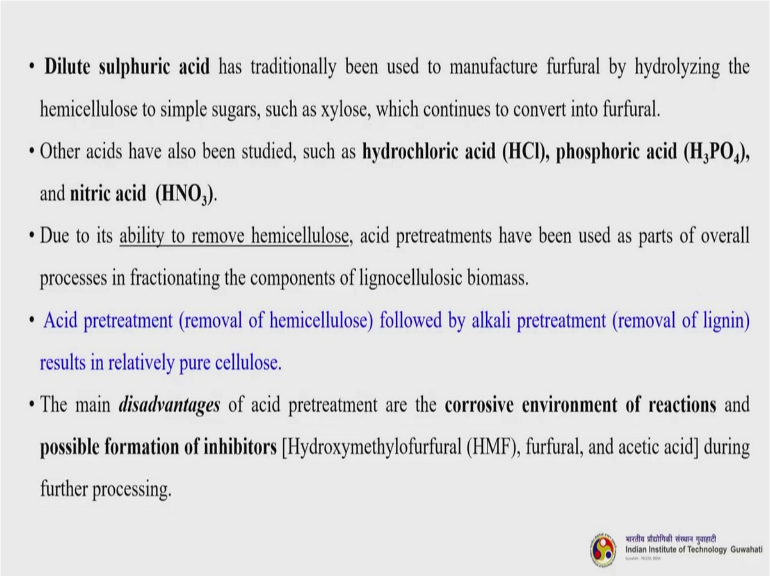

- **Dilute sulphuric acid** has traditionally been used to manufacture furfural by hydrolyzing the hemicellulose to simple sugars, such as xylose, which continues to convert into furfural.
- Other acids have also been studied, such as **hydrochloric acid (HCl), phosphoric acid (H_3PO_4)**, and **nitric acid (HNO_3)**.
- Due to its ability to remove hemicellulose, acid pretreatments have been used as parts of overall processes in fractionating the components of lignocellulosic biomass.
- Acid pretreatment (removal of hemicellulose) followed by alkali pretreatment (removal of lignin) results in relatively pure cellulose.
- The main ***disadvantages*** of acid pretreatment are the **corrosive environment of reactions** and **possible formation of inhibitors** [Hydroxymethylofurfural (HMF), furfural, and acetic acid] during further processing.

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Dilute sulphuric acid has traditionally been used to manufacture furfurals by hydrolysing the hemicellulose to simple sugars such as xylose which continues to convert into furfurals. Other acids have also been studied such as hydrochloric acid, then Phosphoric acid and nitric acid. Due to its ability to remove hemicellulose acid pretreatment has been used as part of overall processes in fractionating the components of lignocellulosic biomass.

Acid pretreatment followed by alkali pretreatment results in relativity pure cellulose. Now, please give little more emphasis on this particular sentence, which I have highlighted in blue colour. So, acid pretreatment which basically talking about the removal of hemicellulose in a more efficient manner than that of cellulose and Lignin followed by alkalis. Alkali is more predominantly will be doing the role to remove lignin.

So, if you combine this it becomes a hybrid process. Dilute acid followed by Alkali so both hemicellulose and Lignin will be removed and whatever left out is cellulose. So, the disadvantages of acid pretreatment are corrosive environment of reaction and possible formation of inhibitors like HMF (hydroxymethylfurfural) and acetic acid during further processing. Now when you talk about corrosive environment of the reaction that means you have to use a very sophisticated reactor of a particular material so that it can deal with the corrosive environment. So, glass is better. But you know that glass you cannot makeup in very big size reactors. It is very difficult to do that. This basically adds on to extra additional cost to the entire process. HMF is a very high value material or solvent. So, it is a fuel additive also.

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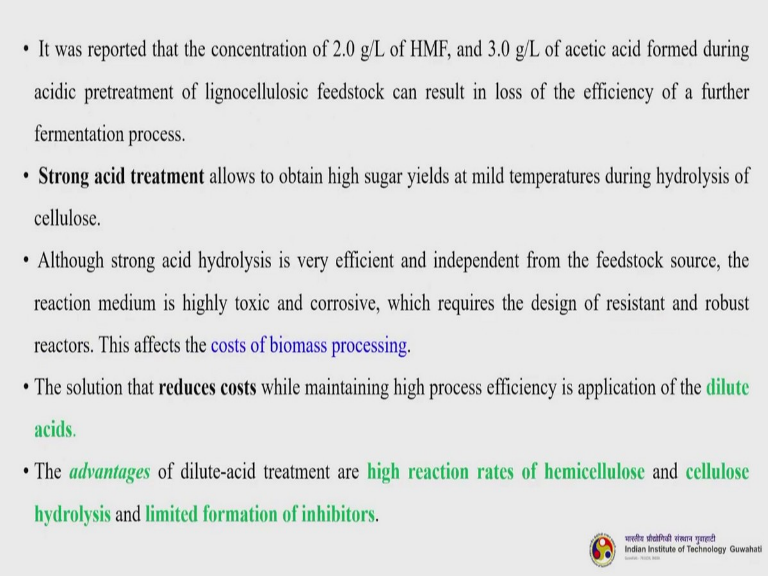

- It was reported that the concentration of 2.0 g/L of HMF, and 3.0 g/L of acetic acid formed during acidic pretreatment of lignocellulosic feedstock can result in loss of the efficiency of a further fermentation process.
- **Strong acid treatment** allows to obtain high sugar yields at mild temperatures during hydrolysis of cellulose.
- Although strong acid hydrolysis is very efficient and independent from the feedstock source, the reaction medium is highly toxic and corrosive, which requires the design of resistant and robust reactors. This affects the costs of biomass processing.
- The solution that **reduces costs** while maintaining high process efficiency is application of the **dilute acids.**
- The ***advantages*** of dilute-acid treatment are **high reaction rates of hemicellulose** and **cellulose hydrolysis** and **limited formation of inhibitors**.

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So, it was reported that the concentration of 2 grams per litre of HMF and 3 grams per litre of Acetic Acid formed during acidic pretreatment of lignocellulosic feedstock can result in the loss of the efficiency of further fermentation process. The meaning of this particular sentence is that for certain cases as for example dilute acid pretreatment of few biomasses, it has been noticed that beyond 2 grams of HMF and 3 grams per litre of Acetic Acid the process and the fermentation is not proceeding in a proper direction because this HMF and acetic acid is becoming toxic for the fermentation to proceed.

Strong acid treatment allows to obtain high sugar yield at mild temperatures during hydrolysis of cellulose. Although strong acid hydrolysis is very efficient and independent from the feedstock source the reaction medium is highly toxic and corrosive which requires the design of resistant and robust reactors; that affects the cost of biomass processing. So that means adding additional cost.

So, the solution that reduces cost while maintaining the high process efficiency is the application of the dilute acid. If you talk about dilute acid also, the problem of corrosiveness also comes down to a lesser extent. So, the advantages of dilute acid treatment are: high reaction rates of hemicellulose and cellulose hydrolysis and limited formation of inhibitors.

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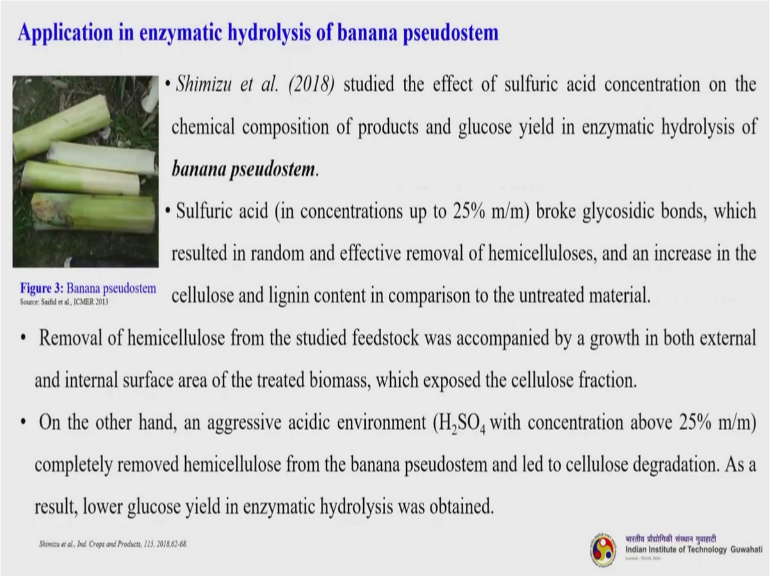

Application in enzymatic hydrolysis of banana pseudostem

- *Shimizu et al. (2018)* studied the effect of sulfuric acid concentration on the chemical composition of products and glucose yield in enzymatic hydrolysis of ***banana pseudostem***.
- Sulfuric acid (in concentrations up to 25% m/m) broke glycosidic bonds, which resulted in random and effective removal of hemicelluloses, and an increase in the cellulose and lignin content in comparison to the untreated material.

Figure 3: Banana pseudostem
Source: Saiful et al., ICMER 2013

- Removal of hemicellulose from the studied feedstock was accompanied by a growth in both external and internal surface area of the treated biomass, which exposed the cellulose fraction.
- On the other hand, an aggressive acidic environment (H_2SO_4 with concentration above 25% m/m) completely removed hemicellulose from the banana pseudostem and led to cellulose degradation. As a result, lower glucose yield in enzymatic hydrolysis was obtained.

Shimizu et al., Ind. Crops and Products, 115, 2018,62-68.

See few examples. The first one is application of dilute acid pretreatment in enzymatic hydrolysis of Bananas pseudostem. Shimizu et al studied the effect of sulfuric acid concentration on the chemical composition of products and glucose yield in enzymatic hydrolysis of Banana pseudostem so this the image. So, the sulphuric acid in concentration up to 25% broke glycosidic bond which resulted in random and effective removal of hemicellulose and an increase in the cellulose and Lignin content in comparison to the untreated material.

Removal of hemicellulose from the studied feedstock was accompanied by growth in both external and internal surface area of the treated biomass which exposes the cellulose fraction. On the other hand, aggressive acidic environment that means sulphuric acid with the concentration of above 25% completely remove hemicellulose from the banana pseudostem and lead to cellulose degradation.

And that is what also we do not want. So as a result, lower glucose yield in enzymatic hydrolysis was obtained; because cellulose was degraded. Now, for all such a processes whether it is dilute acid pretreatment, then enzymatic hydrolysis, fermentation you need to optimise the process parameters so that you get a proper yield of the cellulose and then it gets converted to glucose.

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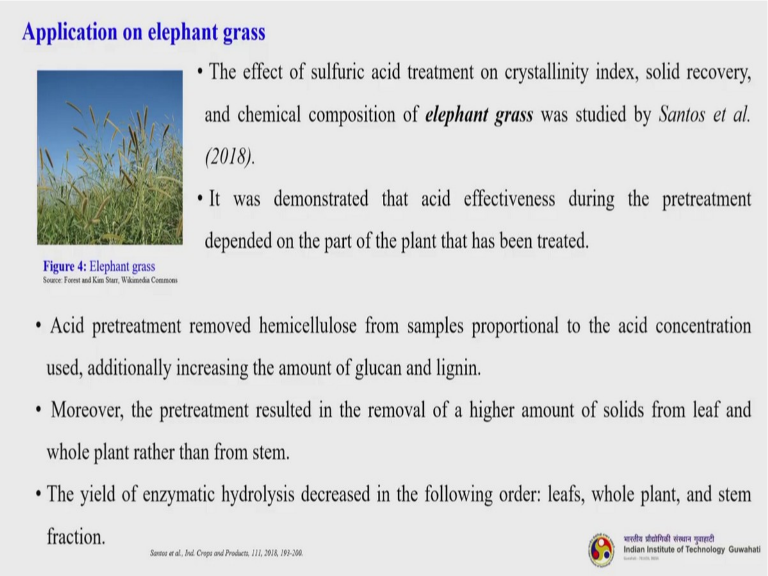

Application on elephant grass

Figure 4: Elephant grass
Source: Forest and Kim Starr, Wikimedia Commons

- The effect of sulfuric acid treatment on crystallinity index, solid recovery, and chemical composition of ***elephant grass*** was studied by *Santos et al. (2018).*
- It was demonstrated that acid effectiveness during the pretreatment depended on the part of the plant that has been treated.
- Acid pretreatment removed hemicellulose from samples proportional to the acid concentration used, additionally increasing the amount of glucan and lignin.
- Moreover, the pretreatment resulted in the removal of a higher amount of solids from leaf and whole plant rather than from stem.
- The yield of enzymatic hydrolysis decreased in the following order: leafs, whole plant, and stem fraction.

Santos et al., Ind. Crops and Products, 111, 2018, 193-200.

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So, another application on Elephant grass. The effect of sulphuric acid treatment on crystallinity index, solid recovery and chemical composition of elephant grass was studied by Santos et al. It was demonstrated that acid effectiveness during the pretreatment depended on the part of the plant that has been treated. Acid pretreatment removed hemicellulose from samples proportional to the acid concentration used additional increasing the amount of glucan and Lignin.

Moreover, the pretreatment resulted in the removal of a higher amount of solid from leaf and whole plant rather than from the stem. The yield of enzymatic hydrolysis decreases in the following order: Leaves, whole plant and stem fraction.

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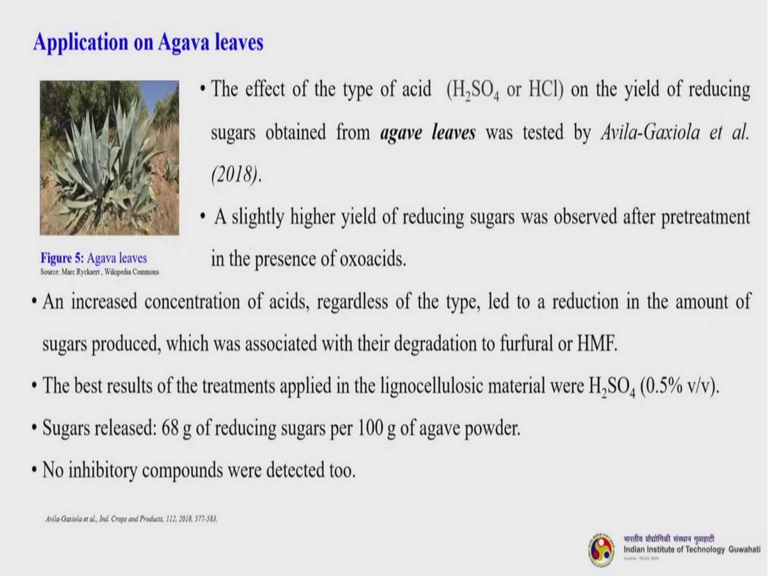

Application on Agava leaves

Figure 5: Agava leaves
Source: Marc Ryckaert , Wikipedia Commons

- The effect of the type of acid (H_2SO_4 or HCl) on the yield of reducing sugars obtained from ***agave leaves*** was tested by *Avila-Gaxiola et al. (2018).*
- A slightly higher yield of reducing sugars was observed after pretreatment in the presence of oxoacids.
- An increased concentration of acids, regardless of the type, led to a reduction in the amount of sugars produced, which was associated with their degradation to furfural or HMF.
- The best results of the treatments applied in the lignocellulosic material were H_2SO_4 (0.5% v/v).
- Sugars released: 68 g of reducing sugars per 100 g of agave powder.
- No inhibitory compounds were detected too.

Avila-Gaxiola et al., Ind. Crops and Products, 112, 2018, 577-583.

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So, another one application on the Agava leaves. Effect of the type of acid whether sulfuric acid or hydrochloric acid on the yield of reducing Sugars obtained from Agava leaves was tested by Avila-Gaxiola et al in 2018. So, you can refer that reference has been given. A slightly higher yield of reducing Sugars was observed after pretreatment in the presence of oxoacids. An increase concentration of acids regardless of the type led to a reduction in the amount of sugar produced which is associated with their degradation to furfurals or hydroxymethylfurfural.

The best results of the treatments applied in the lignocellulosic materials were sulphuric acid at only 0.5% volume by volume basis, very diluted concentration. Sugars released is 68 grams of reducing sugar per 100 grams of Agava powder, which is actually very good yield. No inhibitory compounds were detected; that’s because the concentration of the sulphuric acid is very low.

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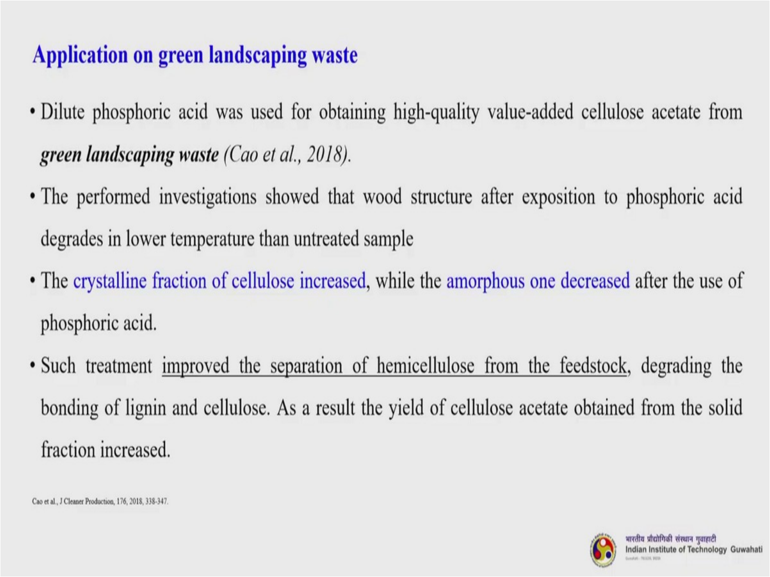

Application on green landscaping waste

- Dilute phosphoric acid was used for obtaining high-quality value-added cellulose acetate from ***green landscaping waste*** *(Cao et al., 2018)*.
- The performed investigations showed that wood structure after exposition to phosphoric acid degrades in lower temperature than untreated sample
- The crystalline fraction of cellulose increased, while the amorphous one decreased after the use of phosphoric acid.
- Such treatment improved the separation of hemicellulose from the feedstock, degrading the bonding of lignin and cellulose. As a result the yield of cellulose acetate obtained from the solid fraction increased.

Cao et al., J Cleaner Production, 176, 2018, 338-347.

So, another application on green landscaping waste. Dilute phosphoric acid was used for obtaining high quality value-added cellulose Acetate from Green landscaping waste. So, the performed investigation showed that wood structure after exposition to phosphoric acid degrades in lower temperature than untreated sample. And the crystalline fraction of the cellulose increased, while the amorphous one decreased after the use of the phosphoric acid.

Such treatment improves the separation of hemicellulose from the feedstock degrading the bonding of Lignin and cellulose. As a result, the yield of cellulose acetate obtained from the solid fraction increased.

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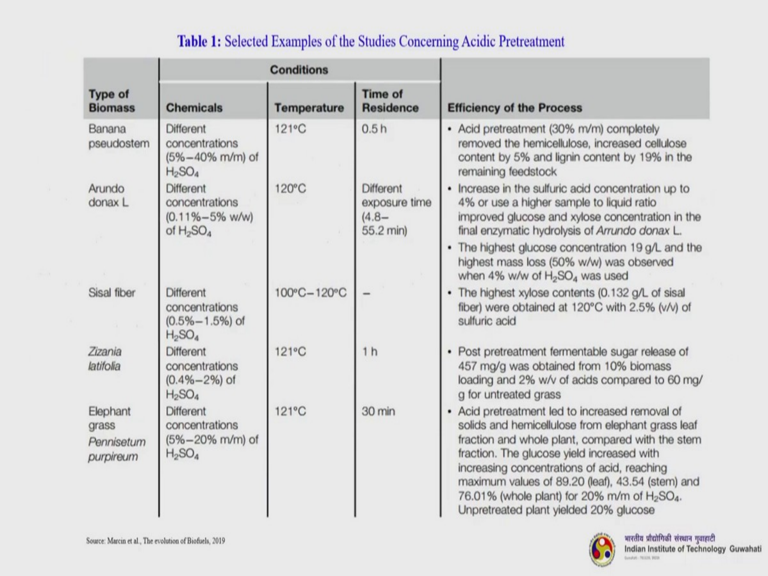

Table 1: Selected Examples of the Studies Concerning Acidic Pretreatment

Type of Biomass	Conditions			Efficiency of the Process
	Chemicals	Temperature	Time of Residence	
Banana pseudostem	Different concentrations (5%–40% m/m) of H_2SO_4	121°C	0.5 h	• Acid pretreatment (30% m/m) completely removed the hemicellulose, increased cellulose content by 5% and lignin content by 19% in the remaining feedstock
Arundo donax L	Different concentrations (0.11%–5% w/w) of H_2SO_4	120°C	Different exposure time (4.8–55.2 min)	• Increase in the sulfuric acid concentration up to 4% or use a higher sample to liquid ratio improved glucose and xylose concentration in the final enzymatic hydrolysis of *Arrundo donax* L. • The highest glucose concentration 19 g/L and the highest mass loss (50% w/w) was observed when 4% w/w of H_2SO_4 was used
Sisal fiber	Different concentrations (0.5%–1.5%) of H_2SO_4	100°C–120°C	–	• The highest xylose contents (0.132 g/L of sisal fiber) were obtained at 120°C with 2.5% (v/v) of sulfuric acid
Zizania latifolia	Different concentrations (0.4%–2%) of H_2SO_4	121°C	1 h	• Post pretreatment fermentable sugar release of 457 mg/g was obtained from 10% biomass loading and 2% w/v of acids compared to 60 mg/g for untreated grass
Elephant grass *Pennisetum purpireum*	Different concentrations (5%–20% m/m) of H_2SO_4	121°C	30 min	• Acid pretreatment led to increased removal of solids and hemicellulose from elephant grass leaf fraction and whole plant, compared with the stem fraction. The glucose yield increased with increasing concentrations of acid, reaching maximum values of 89.20 (leaf), 43.54 (stem) and 76.01% (whole plant) for 20% m/m of H_2SO_4. Unpretreated plant yielded 20% glucose

Source: Marcin et al., The evolution of Biofuels, 2019

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This particular table, please refer to this one. So here it is given that selected samples of the studies concerning acid pretreatment. This list is only a representative in nature. There are many. This has been taken from a particular reference which is listed here. You can refer to it later there are many. I just quickly explain to you. Banana pseudostem we have already seen. Let us see this Sisal fibre. So different concentration, say 0.5 to 1.5%, so different concentrations of sulphuric acid has been tested.

The condition is; temperature is around 100 degrees to 120 degrees' centigrade; time of residence is of course not mentioned and the efficiency is that the highest xylose concentration of 0.132 gram per litre of the fibre was obtained at 120 degrees centigrade with 2.5 % volume by volume of sulphuric acid. Like similarly, I do not want to read out all these things you can refer it later on and I deliberately added all these things so that those (of you) who will be working on Biomass related topics for their academic interest or otherwise for doing some research, it will be helpful for them to get some first-hand information about the different effects of the dilute acid pretreatment. Similarly, this is continuing, so you can see the Agava plant which we have discussed, this is cornstover dilute phosphoric acid 50 degree centigrade, 10 hours. So you can see that it substantially decreases the gaseous product formation, but increase the amount of liquid fraction in the pyrolysis process. This is particularly done, pre-treated cornstover with an aim to pyrolyze it.

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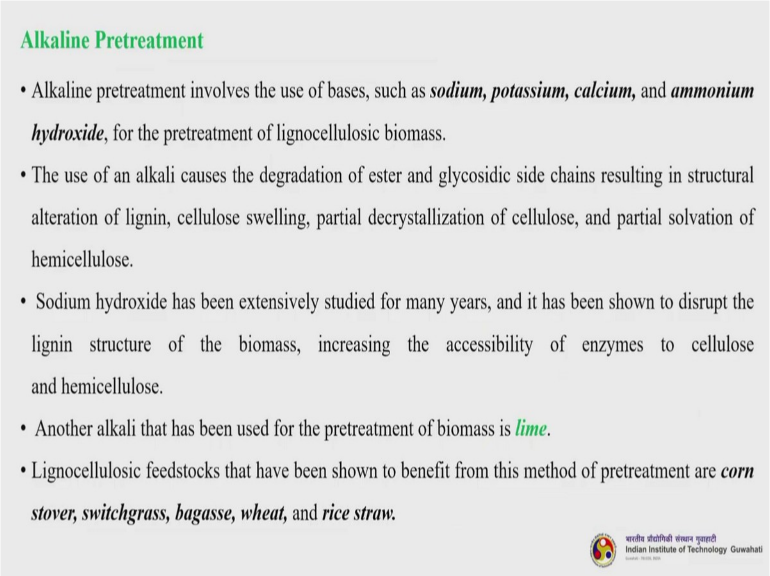

Alkaline Pretreatment

- Alkaline pretreatment involves the use of bases, such as ***sodium, potassium, calcium,*** and ***ammonium hydroxide***, for the pretreatment of lignocellulosic biomass.
- The use of an alkali causes the degradation of ester and glycosidic side chains resulting in structural alteration of lignin, cellulose swelling, partial decrystallization of cellulose, and partial solvation of hemicellulose.
- Sodium hydroxide has been extensively studied for many years, and it has been shown to disrupt the lignin structure of the biomass, increasing the accessibility of enzymes to cellulose and hemicellulose.
- Another alkali that has been used for the pretreatment of biomass is *lime*.
- Lignocellulosic feedstocks that have been shown to benefit from this method of pretreatment are ***corn stover, switchgrass, bagasse, wheat,*** and ***rice straw.***

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The next one is alkaline pretreatment: Alkaline pretreatment involves the use of bases such as Sodium, Potassium, calcium and ammonium Hydroxide for the pretreatment of lignocellulosic biomass. The use of an alkali causes the degradation of Ester and glycosidic side chains resulting in structural alteration of Lignin and cellulose swelling, partially de-crystallization of cellulose and partial solvation of hemicellulose.

Sodium hydroxide has been used extensively by researchers for many years and it has been shown to disrupt the lignin structure of the Biomass increasing the accessibility of enzyme to cellulose and hemicellulose. Another alkali that has been used for the pretreatment of biomass is the lime. Lignocellulosic feedstocks that have been shown to benefit from this method of pretreatment are cornstover, switchgrass, bagasse, wheat, rice straw and the list is actually end less there are many other. Just a few are listed here.

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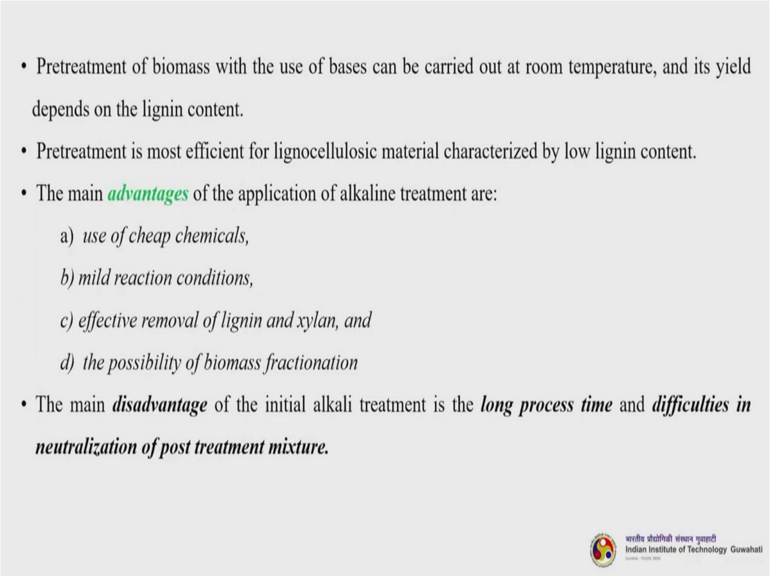

- Pretreatment of biomass with the use of bases can be carried out at room temperature, and its yield depends on the lignin content.
- Pretreatment is most efficient for lignocellulosic material characterized by low lignin content.
- The main ***advantages*** of the application of alkaline treatment are:
  - a) *use of cheap chemicals,*
  - *b) mild reaction conditions,*
  - *c) effective removal of lignin and xylan, and*
  - *d) the possibility of biomass fractionation*
- The main ***disadvantage*** of the initial alkali treatment is the ***long process time*** and ***difficulties in neutralization of post treatment mixture.***

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So, pretreatment of biomass with the use of bases can be carried out at room temperature and its yield depends on the lignin content. Pretreatment is most efficient for lignocellulosic materials characterized by low lignin content. The advantages of this particular alkaline treatment are: (a) use of cheap chemicals: actually that means the low-cost chemicals; (b) mild reaction conditions; (c) Effective removal of Lignin and xylan and (d) possibility of biomass fractionation. However, the biggest disadvantage of that particular process is the long process time it requires and difficulties in neutralization of the post treatment mixture. So that means you have to neutralize after the processes is over. There will be still so much of Alkaline content in the reaction mixture.

So, you need to take out your Lignin and cellulose, hemicellulose. So, you need to neutralize it. So that neutralization also adds some another cost. So, you have to add some acid to neutralize it. The most important thing is of course the long process time. So that is the disadvantage and in commercial or industrial applications we have to have the processes which are faster and as well as low cost.

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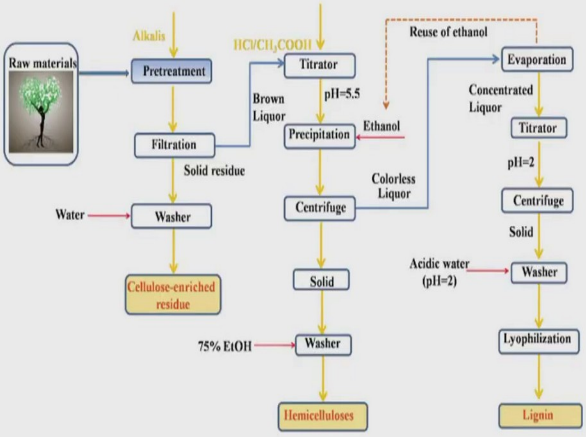

Figure 6: Flow chart of pretreatment using alkalis

Source: Xu et al., Biomass Fractionation Technologies for a Lignocellulosic Feedstock Based Biorefinery, 2016, Pages 431-459.

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This is an excellent slide. You see this is a flowchart of a pretreatment process using the alkalis. What it does. This is a representative one. It is not true for every biomass. So let us just understand. A biomass - so, you give alkalis to pre-treat it, so whatever you get is (subjected to) a filtration. So, it is a solid residue - you wash it - then whatever the solid residue that is obtained is nothing but the cellulose enriched residues.

So, you can further process it to get pure cellulose out of that. So, the brown liquor after the alkali pretreatment. So, alkali pretreatment will result into two things. One is this solid rich leftover mass. So that is basically Cellulose rich and another is a brown liquor liquid. So that brown liquor when you do the further processing by adjusting its pH and all these things it goes for precipitation – centrifugation - then again, you will get a solid part - wash it - you get hemicellulose. And from here this particular process whatever the solid part is come down as hemicellulose whatever liquid part remain it has become by the time colourless liquid. That goes to further processing of evaporation - centrifugation - washer - lyophilization and you get lignin. Here actually during this evaporation what is evaporated is this alcohol.

So this alcohol or in this case ethanol will be again recycled back to this particular space. Now you can see and understand that from this particular process that in three subsequent steps we are getting cellulose rich residue, then hemicelluloses and Lignin by alkali pretreatment.

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Application on bamboo

- *Yuan et al. (2018)* investigated in detail the effect of sodium hydroxide concentration on the chemical composition of **bamboo**, which can be used as a potential feedstock for the production of sugars and alcohols.
- Additionally, they showed that post-treatment liquor can be a source of *high-value lignin* and *silica*.
- It was demonstrated that severe conditions can lead to degradation of cellulose and hemicellulose monomers, hence decreasing the sugar yield.
- Effective removal of sugars from the solid fraction resulted in their larger share in the liquid fraction.
- Furfural and hydroxymethylfurfural, potential inhibitors of further processing, were not detected in liquid fraction which can be beneficial for the high yield of bio-ethanol formation.

Yuan et al., Bioresource Technology, 247, 2018, 242-249.

So, few applications we will see, application on bamboo. Yuan et al investigated in detail the effect of sodium hydroxide concentration on the chemical composition of bamboo which can be used as a potential feedstock for the production of Sugars and alcohol. Additionally, they showed that post treatment liquor can be a source of high-value Lignin and silica. It was demonstrated that severe conditions can lead to degradation of cellulose and hemicellulose monomer hence decreasing the sugar yield.

And effective removal of sugar from the solid fraction resulted in their larger share in the liquid fraction. Furfural and hydroxymethylfurfural potential inhibiter for further processing were not detected in liquid fraction. So that is a good thing actually. So we will get more bioethanol.

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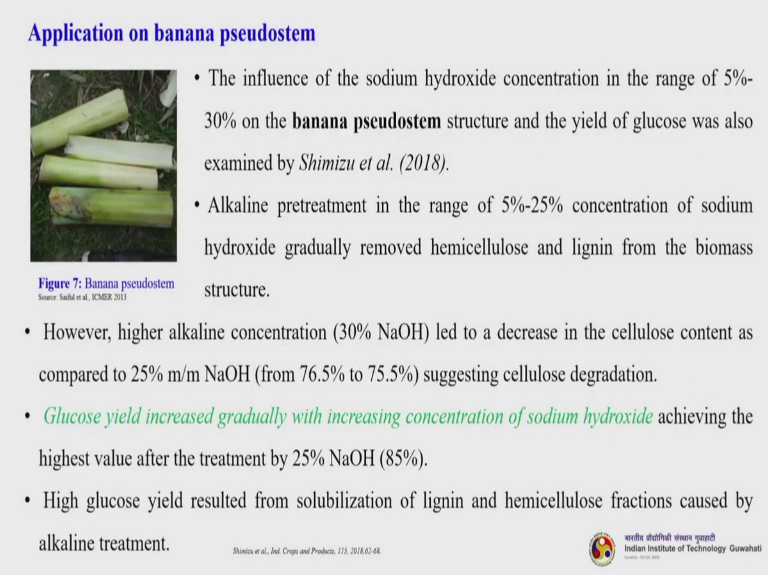

Application on banana pseudostem

- The influence of the sodium hydroxide concentration in the range of 5%-30% on the **banana pseudostem** structure and the yield of glucose was also examined by *Shimizu et al. (2018)*.
- Alkaline pretreatment in the range of 5%-25% concentration of sodium hydroxide gradually removed hemicellulose and lignin from the biomass structure.

Figure 7: Banana pseudostem
Source: Saiful et al., ICMER 2013

- However, higher alkaline concentration (30% NaOH) led to a decrease in the cellulose content as compared to 25% m/m NaOH (from 76.5% to 75.5%) suggesting cellulose degradation.
- *Glucose yield increased gradually with increasing concentration of sodium hydroxide* achieving the highest value after the treatment by 25% NaOH (85%).
- High glucose yield resulted from solubilization of lignin and hemicellulose fractions caused by alkaline treatment.

Shimizu et al., Ind. Crops and Products, 115, 2018,62-68.

Application on banana pseudostem. So, by again Shimizu et al. Influence of sodium hydroxide in the range of 5 to 30% was studied for the yield of glucose. Alkaline pretreatment in the range of 5 to 25% concentration of sodium hydroxide gradually removed hemicellulose and Lignin from the Biomass structure. However higher alkaline concentration, that is 30% and above led to a decrease in the cellulose content as compared to 25% Sodium Hydroxide suggesting cellulose degradation.

So, this is what is I was talking about optimisation. So, you have to take different concentrations and to see at what concentration of sodium hydroxide or alkali, you are getting the best result, the higher yield of reducing sugar. So, glucose yield is increased gradually with increasing concentration of sodium hydroxide achieving the highest value after treatment by 25% sodium hydroxide.

So, high glucose yield resulted from solubilization of Lignin and hemicellulose fractions caused by the alkali pretreatment; this is the reason for high yield.

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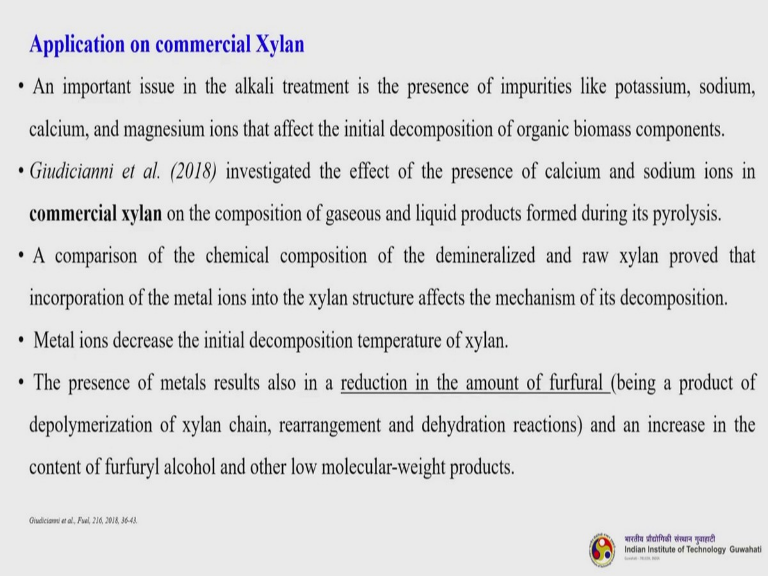

Application on commercial Xylan

- An important issue in the alkali treatment is the presence of impurities like potassium, sodium, calcium, and magnesium ions that affect the initial decomposition of organic biomass components.
- *Giudicianni et al. (2018)* investigated the effect of the presence of calcium and sodium ions in **commercial xylan** on the composition of gaseous and liquid products formed during its pyrolysis.
- A comparison of the chemical composition of the demineralized and raw xylan proved that incorporation of the metal ions into the xylan structure affects the mechanism of its decomposition.
- Metal ions decrease the initial decomposition temperature of xylan.
- The presence of metals results also in a reduction in the amount of furfural (being a product of depolymerization of xylan chain, rearrangement and dehydration reactions) and an increase in the content of furfuryl alcohol and other low molecular-weight products.

Giudicianni et al., Fuel, 216, 2018, 36-43.

So then application on commercials Xylan. An important issue in the alkali pretreatment is the presence of impurities like potassium, sodium, calcium and magnesium ions that affect the initial decomposition of the organic Biomass components. Giudicianni et al in 2018 investigated the effect of the presence of calcium and sodium ions in commercials Xylan on the composition of gaseous and liquid products formed during its pyrolysis.

So, a comparison of the chemical composition of the demineralized and raw Xylan prove that incorporation of the metal ions into the Xylan structure affect the mechanism of its decomposition. Metal ions decrease the initial decomposition temperature of Xylan. The presence of metals results also in the reduction in the amount of furfural (being a product of depolymerization of Xylan chain, rearrangement and dehydration reactions) and an increase in the content of furfuryl alcohol and other low molecular weight products.

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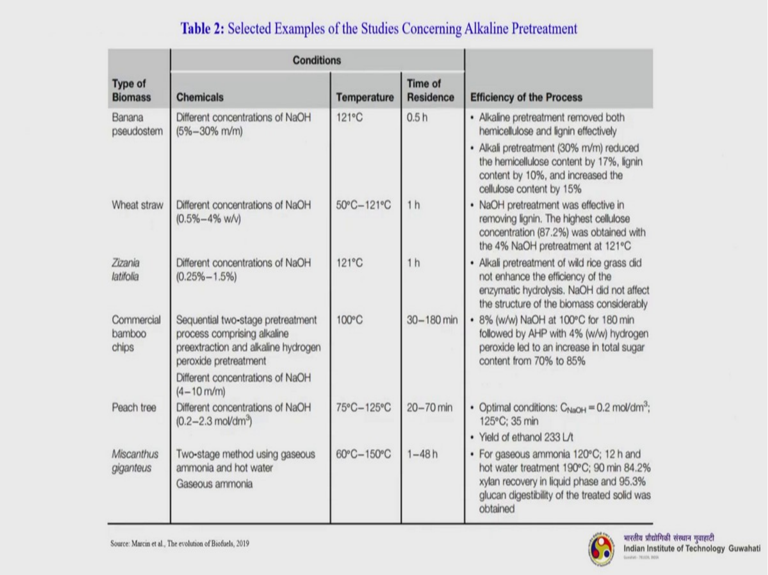

Table 2: Selected Examples of the Studies Concerning Alkaline Pretreatment

Type of Biomass	Conditions			Efficiency of the Process
	Chemicals	Temperature	Time of Residence	
Banana pseudostem	Different concentrations of NaOH (5%–30% m/m)	121°C	0.5 h	• Alkaline pretreatment removed both hemicellulose and lignin effectively • Alkali pretreatment (30% m/m) reduced the hemicellulose content by 17%, lignin content by 10%, and increased the cellulose content by 15%
Wheat straw	Different concentrations of NaOH (0.5%–4% w/v)	50°C–121°C	1 h	• NaOH pretreatment was effective in removing lignin. The highest cellulose concentration (87.2%) was obtained with the 4% NaOH pretreatment at 121°C
Zizania latifolia	Different concentrations of NaOH (0.25%–1.5%)	121°C	1 h	• Alkali pretreatment of wild rice grass did not enhance the efficiency of the enzymatic hydrolysis. NaOH did not affect the structure of the biomass considerably
Commercial bamboo chips	Sequential two-stage pretreatment process comprising alkaline preextraction and alkaline hydrogen peroxide pretreatment Different concentrations of NaOH (4–10 m/m)	100°C	30–180 min	• 8% (w/w) NaOH at 100°C for 180 min followed by AHP with 4% (w/w) hydrogen peroxide led to an increase in total sugar content from 70% to 85%
Peach tree	Different concentrations of NaOH (0.2–2.3 mol/dm^3)	75°C–125°C	20–70 min	• Optimal conditions: C_{NaOH} = 0.2 mol/dm^3; 125°C; 35 min • Yield of ethanol 233 L/t
Miscanthus giganteus	Two-stage method using gaseous ammonia and hot water Gaseous ammonia	60°C–150°C	1–48 h	• For gaseous ammonia 120°C; 12 h and hot water treatment 190°C; 90 min 84.2% xylan recovery in liquid phase and 95.3% glucan digestibility of the treated solid was obtained

Source: Marcin et al., The evolution of Biofuels, 2019

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So, this is again another table which gives selected examples of studies concerning alkaline pretreatment. So, you can see so many are mentioned here actually if you look at literature, there are hundreds of papers on alkali pretreatment of different Biomass. So here we have listed Banana pseudostem, wheat straw, *Zizania latifolia*, commercial Bamboo chips, peach tree, miscanthus and giganteus and then many. So, you please refer to it later on, I am not going to read this table.

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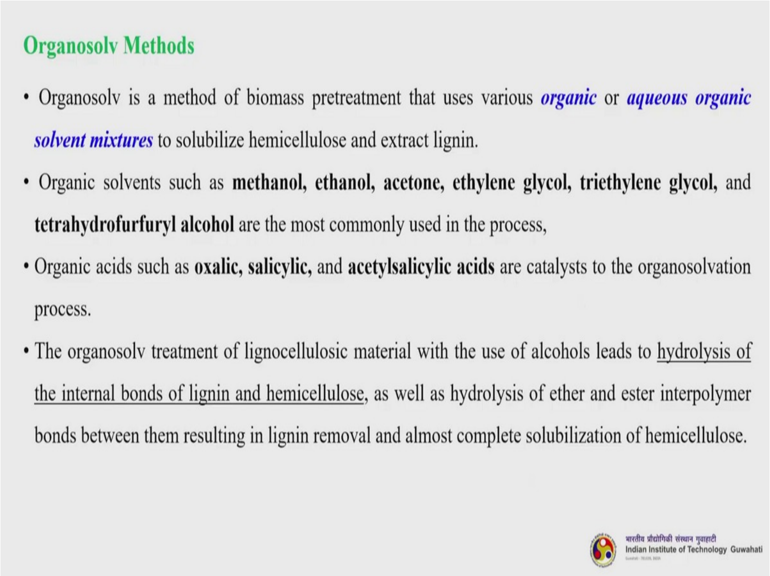

Organosolv Methods

- Organosolv is a method of biomass pretreatment that uses various ***organic*** or ***aqueous organic solvent mixtures*** to solubilize hemicellulose and extract lignin.
- Organic solvents such as **methanol, ethanol, acetone, ethylene glycol, triethylene glycol,** and **tetrahydrofurfuryl alcohol** are the most commonly used in the process,
- Organic acids such as **oxalic, salicylic,** and **acetylsalicylic acids** are catalysts to the organosolvation process.
- The organosolv treatment of lignocellulosic material with the use of alcohols leads to hydrolysis of the internal bonds of lignin and hemicellulose, as well as hydrolysis of ether and ester interpolymer bonds between them resulting in lignin removal and almost complete solubilization of hemicellulose.

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And we will move into the next pretreatment method which is a very important class of pretreatment methods known as organosolv method. The organosolv is a method of biomass pretreatment that uses various organic and aqueous organic solvent mixture to solubilize hemicellulose and extract Lignin. Please understand acids are out of this; no acids and alkalis are covered under this.

So organic solvent such as methanol, ethanol and Acetone, ethylene glycol, Triethylene glycol and tetrahedrofurfuryl alcohol are the most commonly used solvent for the organosolv pretreatment process. Organic acids such as Oxalic, salicylic and acetylsalicylic acid are catalyst to organosolv solvation process. So, they act as catalyst, they are not the main solvents. So the organosolv pretreatment of lignocellulosic materials with the use of alcohol leads to the hydrolysis of the internal bonds of Lignin and hemicellulose as well as hydrolysis of ether and Ester interpolymer bond between them resulting in lignin removal and almost complete solubilisation of the hemicellulose.

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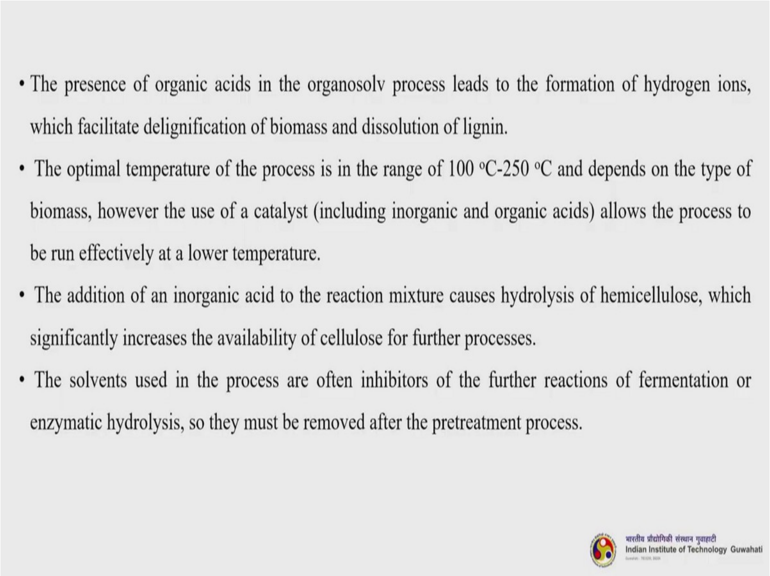

- The presence of organic acids in the organosolv process leads to the formation of hydrogen ions, which facilitate delignification of biomass and dissolution of lignin.
- The optimal temperature of the process is in the range of 100 °C-250 °C and depends on the type of biomass, however the use of a catalyst (including inorganic and organic acids) allows the process to be run effectively at a lower temperature.
- The addition of an inorganic acid to the reaction mixture causes hydrolysis of hemicellulose, which significantly increases the availability of cellulose for further processes.
- The solvents used in the process are often inhibitors of the further reactions of fermentation or enzymatic hydrolysis, so they must be removed after the pretreatment process.

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The presence of organic acids in the organosolv process leads to the formation of hydrogen ions, which facilitate delignification of biomass and dissolution of Lignin. So, this the mechanism. So, the optimal temperature of the process is in the range 100 degree to 250 degree centigrade and depends on the type of biomass. However, the use of a catalyst (so that includes any organic or inorganic acids) allows the process to be run effectively at lower temperature.

So, you use catalyst the temperature will come down. Then addition of an inorganic acid to the reaction mixture causes hydrolysis of hemicellulose which significantly increases the availability of cellulose for further process. The solvents used in the process are open inhibitors for further reactions of fermentation or enzymatic hydrolysis, so they must be removed after the pretreatment process.

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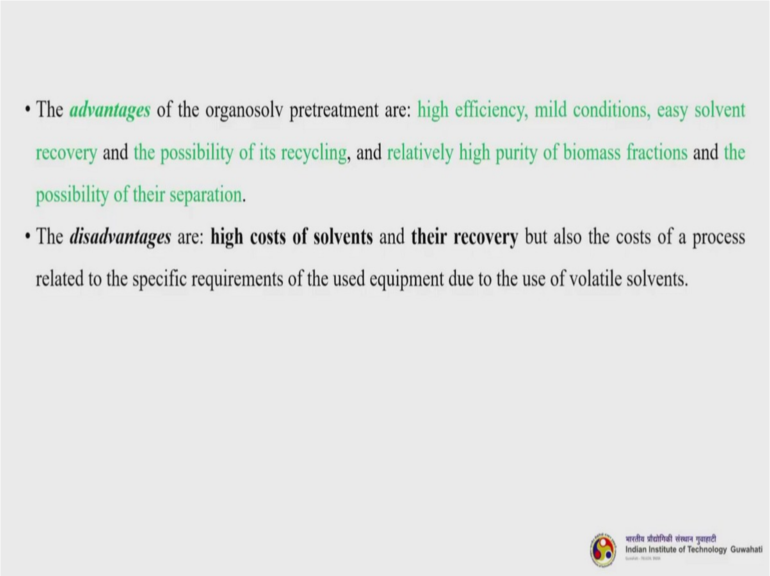

- The ***advantages*** of the organosolv pretreatment are: high efficiency, mild conditions, easy solvent recovery and the possibility of its recycling, and relatively high purity of biomass fractions and the possibility of their separation.
- The ***disadvantages*** are: **high costs of solvents** and **their recovery** but also the costs of a process related to the specific requirements of the used equipment due to the use of volatile solvents.

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The advantages of the organosolv pretreatment are: high efficiency, mild conditions, easy solvent recovery, and the possibility of its recycling and relatively high purity of biomass fractions and the possibility of their separation. The disadvantages are: high cost of solvent and their recovery but also, the cost of a process related to the specific requirement of the used equipment due to the use of volatile solvents.

So again, you need specific type of sophisticated equipment to carry out this particular pretreatment.

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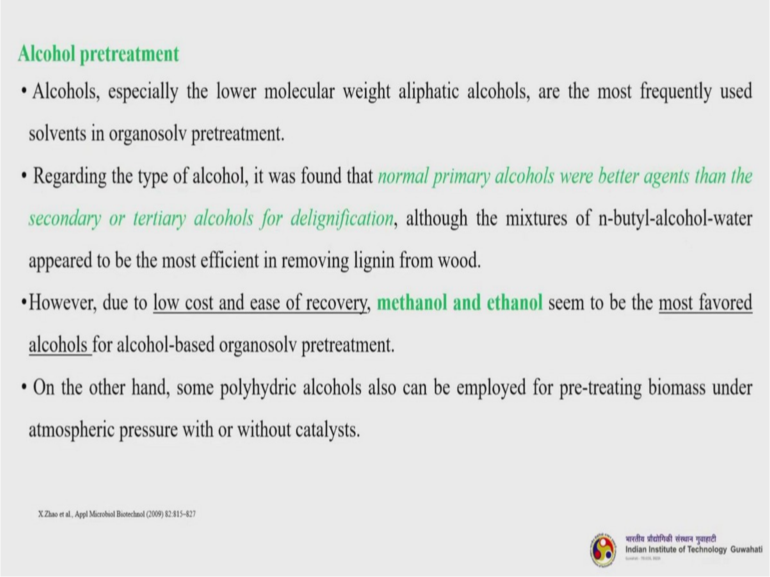

Alcohol pretreatment

- Alcohols, especially the lower molecular weight aliphatic alcohols, are the most frequently used solvents in organosolv pretreatment.
- Regarding the type of alcohol, it was found that *normal primary alcohols were better agents than the secondary or tertiary alcohols for delignification*, although the mixtures of n-butyl-alcohol-water appeared to be the most efficient in removing lignin from wood.
- However, due to low cost and ease of recovery, **methanol and ethanol** seem to be the most favored alcohols for alcohol-based organosolv pretreatment.
- On the other hand, some polyhydric alcohols also can be employed for pre-treating biomass under atmospheric pressure with or without catalysts.

X.Zhao et al., Appl Microbiol Biotechnol (2009) 82:815–827

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So, we will see alcohol pretreatment under organosolv. Alcohol, especially the lower molecular weight aliphatic alcohols are the most frequently used solvents in the organosolv pretreatment. Regarding the type of alcohol, it was found that normal primary alcohols were better agent than the secondary or tertiary alcohols for delignification. Although the mixtures of n-butyl alcohol water appeared to be the most efficient in removing Lignin from the wood. However, due to the low cost and ease recovery methanol and ethanol seems to be the most favoured alcohol for alcohol based organosolv pretreatment.

On the other hand, some polyhydric alcohol also can be employed for the pre-treating Biomass under atmospheric pressure with or without catalyst.

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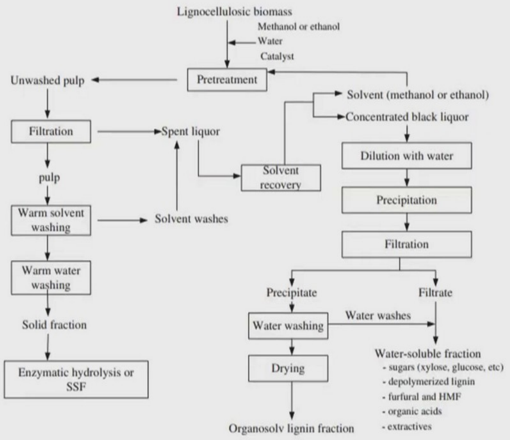

Figure 8: Process flowchart of methanol and ethanol pretreatment

Source: X.Zhao et al., Appl Microbiol Biotechnol (2009) 82:815–827

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So, let us see this process flowchart for methanol and ethanol pretreatment. So the lignocellulosic Biomass, you use either ethanol or methanol water. You pre-treat it, so whatever you get is basically the unwashed pulp. So that you filter it then again go for warm solvent washing so you get the solvent washes, again it goes to the solvent recovery process. You need to recover the solvent because solvents are costly.

Then again, followed by a warm water washing, solid fraction, you will be getting and this goes to the enzymatic hydrolysis or SSF. So, whatever spent liquor you are getting here that goes to the solvent recovery process and the solvent is again feed back into the pretreatment process. So, this is from here again, you can see that. From the solvent recovery you will get something a concentrated Black Liquor.

So that is rich in lignin fraction. So, you need to recover Lignin. So, you go for the different processes such as dilution with water followed by precipitation filtration. So then again, you see water washing, drying and you get organosolv lignin fraction. And then here again the water washing and filter again recovery and the reuse of this process. This is one simple system where you can see that we are getting the solid fraction which can be processed for the enzymatic hydrolysis or SSF. And then we get a lignin there apart from that some other solvents.

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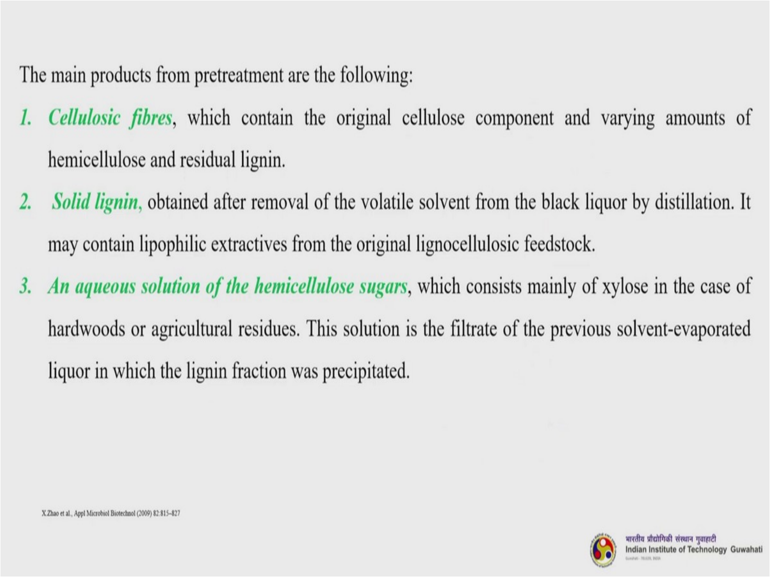

The main products from pretreatment are the following:

1. *Cellulosic fibres*, which contain the original cellulose component and varying amounts of hemicellulose and residual lignin.
2. *Solid lignin*, obtained after removal of the volatile solvent from the black liquor by distillation. It may contain lipophilic extractives from the original lignocellulosic feedstock.
3. *An aqueous solution of the hemicellulose sugars*, which consists mainly of xylose in the case of hardwoods or agricultural residues. This solution is the filtrate of the previous solvent-evaporated liquor in which the lignin fraction was precipitated.

X.Zhao et al., Appl Microbiol Biotechnol (2009) 82:815–827

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So, the main products from pretreatment are the following. First one is the cellulosic fibres. So which contain the original cellulose component and varying amounts of hemicellulose and little residual Lignin which could not be taken out. The second part, solid Lignin obtained after removal of the volatile solvent from the black liquor by distillation. It may contain lipophilic extractives from the original lignocellulosic feedstock. And the third one is an aqueous solution of the hemicellulose sugars, which consists mainly of xylose in the case of hard wood or agricultural residues. You do not get xylose every time. It depends which Biomass you are using. This solution is the filtrate of the previous solvent-evaporated liquor in which the lignin fraction was precipitated.

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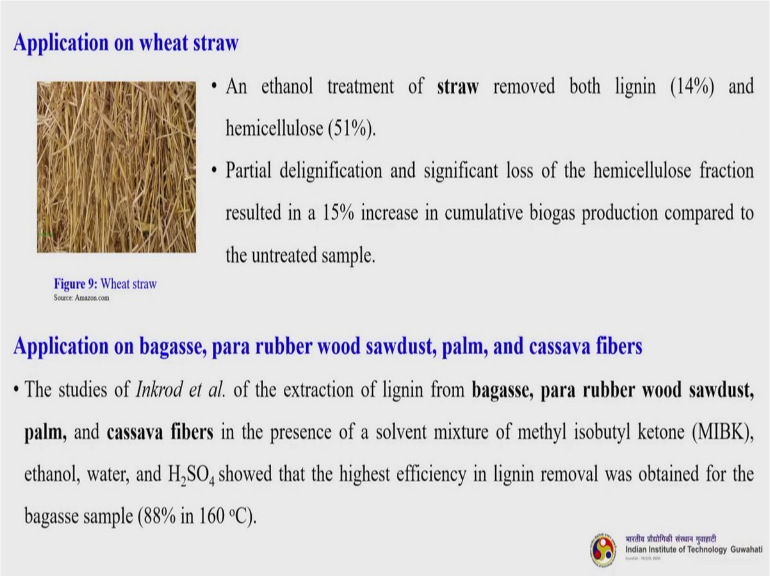

Application on wheat straw

- An ethanol treatment of **straw** removed both lignin (14%) and hemicellulose (51%).
- Partial delignification and significant loss of the hemicellulose fraction resulted in a 15% increase in cumulative biogas production compared to the untreated sample.

Figure 9: Wheat straw
Source: Amazon.com

Application on bagasse, para rubber wood sawdust, palm, and cassava fibers

- The studies of *Inkrod et al.* of the extraction of lignin from **bagasse, para rubber wood sawdust, palm,** and **cassava fibers** in the presence of a solvent mixture of methyl isobutyl ketone (MIBK), ethanol, water, and H_2SO_4 showed that the highest efficiency in lignin removal was obtained for the bagasse sample (88% in 160 °C).

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So, we will see 1 or 2 applications. So, application on wheat straw. An ethanol treatment of straw remove both lignin 14% and hemicellulose 51%. Partial delignification and significant loss of hemicellulose fraction resulted in a 15% increase in cumulative biogas production compared to the untreated sample.

Another application on bagasse, para rubber wood sawdust, Palm and cassava fibre. So, the studies of Inkrod et al of the extraction of Lignin from Bagasse, para rubber wood sawdust, palm and cassava fibres in the presence of a solvent mixture of methyl isobutyl ketone, ethanol, water, and sulfuric acid showed that the highest efficiency in Lignin removal was obtained for the bagasse sample that is 88% in 160 degree centigrade.

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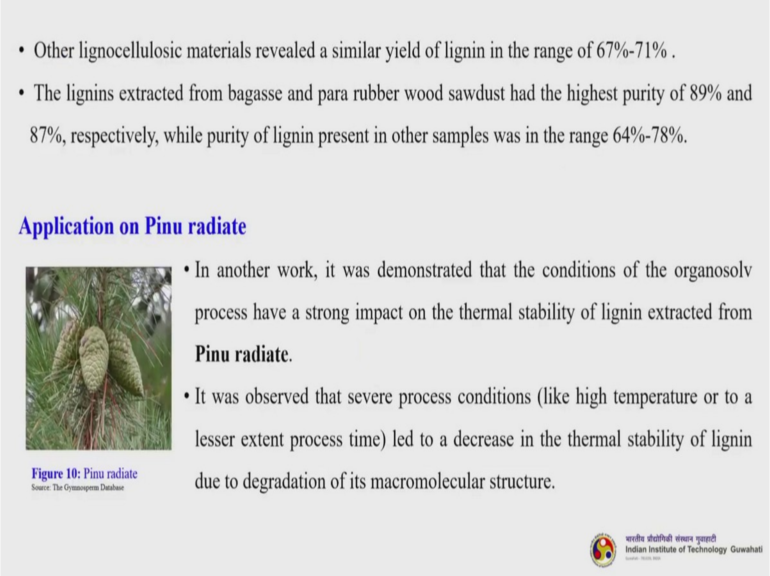

- Other lignocellulosic materials revealed a similar yield of lignin in the range of 67%-71% .
- The lignins extracted from bagasse and para rubber wood sawdust had the highest purity of 89% and 87%, respectively, while purity of lignin present in other samples was in the range 64%-78%.

Application on Pinu radiate

- In another work, it was demonstrated that the conditions of the organosolv process have a strong impact on the thermal stability of lignin extracted from **Pinu radiate**.
- It was observed that severe process conditions (like high temperature or to a lesser extent process time) led to a decrease in the thermal stability of lignin due to degradation of its macromolecular structure.

Figure 10: Pinu radiate
Source: The Gymnosperm Database

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Other lignocellulosic materials revealed a similar yield of Lignin in the range of 67 to 71%. The Lignins extracted from bagasse and para rubber wood sawdust had the highest purity of 89% and 87% respectively while purity of Lignin present in other samples was in the range of 64 to 78%. And it is a good process and quite efficient.

Another application of this process on Pinu radiate. So, in another work it was demonstrated that the conditions of the organosolv process have a strong impact on the thermal stability of Lignin extracted from Pinu radiate. It was observed that severe process conditions like high temperature or to a lesser extent process time lead to a decrease in the thermal stability of Lignin due to the degradation of its macromolecular structure. Lignin is a very complex molecule. So, we have seen the structure. Though it is a very rigid structure, however too harsh treatment will result in the lignin degradation.

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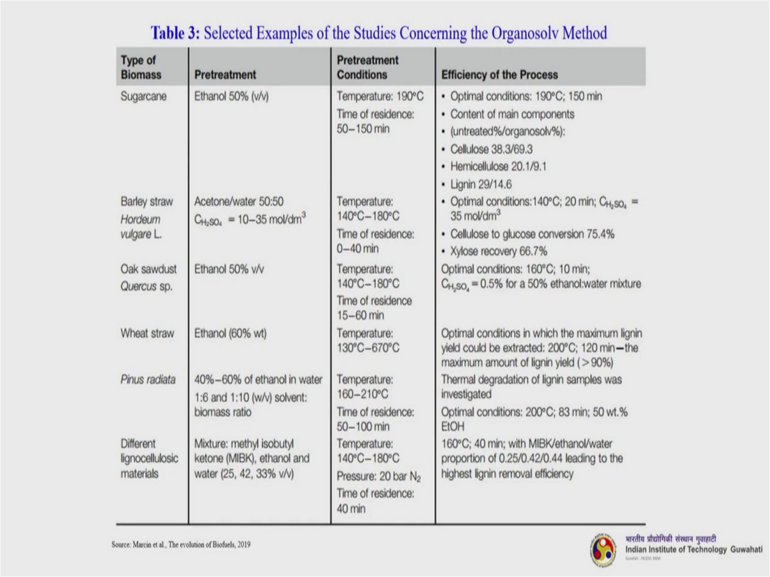

Table 3: Selected Examples of the Studies Concerning the Organosolv Method

Type of Biomass	Pretreatment	Pretreatment Conditions	Efficiency of the Process
Sugarcane	Ethanol 50% (v/v)	Temperature: 190°C Time of residence: 50–150 min	• Optimal conditions: 190°C; 150 min • Content of main components • (untreated%/organosolv%): • Cellulose 38.3/69.3 • Hemicellulose 20.1/9.1 • Lignin 29/14.6
Barley straw *Hordeum vulgare* L.	Acetone/water 50:50 $C_{H_2SO_4}$ = 10–35 mol/dm^3	Temperature: 140°C–180°C Time of residence: 0–40 min	• Optimal conditions:140°C; 20 min; $C_{H_2SO_4}$ = 35 mol/dm^3 • Cellulose to glucose conversion 75.4% • Xylose recovery 66.7%
Oak sawdust *Quercus* sp.	Ethanol 50% v/v	Temperature: 140°C–180°C Time of residence 15–60 min	Optimal conditions: 160°C; 10 min; $C_{H_2SO_4}$ = 0.5% for a 50% ethanol:water mixture
Wheat straw	Ethanol (60% wt)	Temperature: 130°C–670°C	Optimal conditions in which the maximum lignin yield could be extracted: 200°C; 120 min—the maximum amount of lignin yield (>90%)
Pinus radiata	40%–60% of ethanol in water 1:6 and 1:10 (w/v) solvent: biomass ratio	Temperature: 160–210°C Time of residence: 50–100 min	Thermal degradation of lignin samples was investigated Optimal conditions: 200°C; 83 min; 50 wt.% EtOH
Different lignocellulosic materials	Mixture: methyl isobutyl ketone (MIBK), ethanol and water (25, 42, 33% v/v)	Temperature: 140°C–180°C Pressure: 20 bar N_2 Time of residence: 40 min	160°C; 40 min; with MIBK/ethanol/water proportion of 0.25/0.42/0.44 leading to the highest lignin removal efficiency

Source: Marcin et al., The evolution of Biofuels, 2019

So, this is selected examples of some of the studies concerning the organosolv method. You can see that some of the Biomasses like sugarcane, barley, straw, oak sawdust, wheat, pinus all these things are there and different pretreatment processes. Ethanol 50% volume by volume here acetone and water is mixed in 50-50 ratio, and sulfuric acid concentration of 10 to 35 moles per dm cube.

That is very small amount. It acts as catalyst. As I mentioned that in the alcohol adding acid, acid is actually working as a catalyst. So then ethanol 50% and then again ethanol 60% by weight 40 to 60% of Ethanol in water. There are so many different types of pretreatment processes and then followed by their conditions. Again, I am telling you for your understanding whatever you are seeing here; let us say the time of residence to 50 to 60 minutes, temperature 130 to 670 degree, these are the range and they have varied, like 15, 20, 30, 40, 50, 60 like that they might have varied. I am just telling you for your understanding. That you will understand how to optimise the process. Temperature again, 130, 140, 150, 200, 300, 500 up to 670. So this is the range they have varied and they might have got an optimised condition. And let us see the optimised condition he has given here for one particular optimal condition; 160 degrees centigrade, 10 minutes and 0.5% concentration of sulphuric acid for 50% ethanol water mixture.

This is what is called the optimised process condition for the most efficient or the highest reducing sugar yield you can say.

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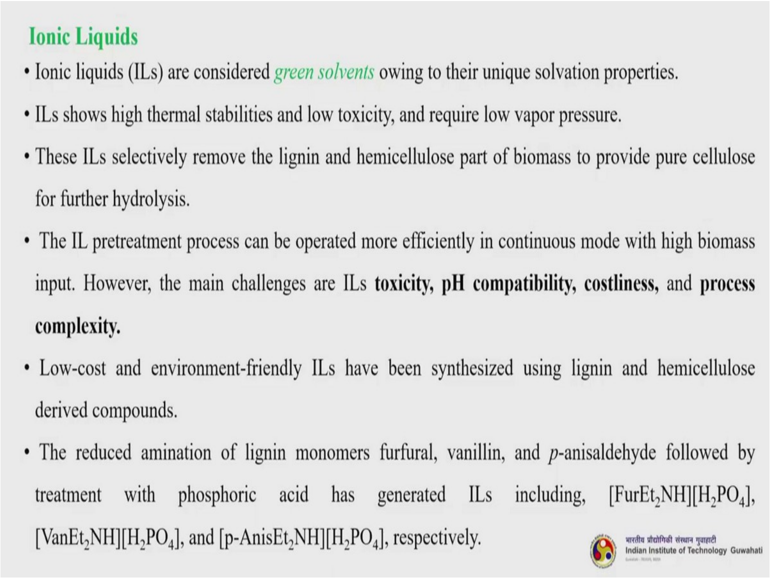

Ionic Liquids

- Ionic liquids (ILs) are considered *green solvents* owing to their unique solvation properties.
- ILs shows high thermal stabilities and low toxicity, and require low vapor pressure.
- These ILs selectively remove the lignin and hemicellulose part of biomass to provide pure cellulose for further hydrolysis.
- The IL pretreatment process can be operated more efficiently in continuous mode with high biomass input. However, the main challenges are ILs **toxicity, pH compatibility, costliness,** and **process complexity.**
- Low-cost and environment-friendly ILs have been synthesized using lignin and hemicellulose derived compounds.
- The reduced amination of lignin monomers furfural, vanillin, and *p*-anisaldehyde followed by treatment with phosphoric acid has generated ILs including, $[FurEt_2NH][H_2PO_4]$, $[VanEt_2NH][H_2PO_4]$, and $[p\text{-}AnisEt_2NH][H_2PO_4]$, respectively.

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So, the next class of solvents are ionic liquids. Ionic liquids are considered as the green solvents owing to their unique solvation properties. So Ionic liquids shows high thermal stability and low toxicity and required low vapour pressure. This ionic liquid selectively remove the Lignin and hemicellulose part of biomass to provide pure cellulose for further hydrolysis. The IL pretreatment process can be operated more efficiently in continuous mode with high biomass input.

However, the main challenges are IL toxicity, pH compatibility, costliness - Ionic liquid still whatever it is commercially available most of them are very costly - and process complexity. Low cost and environmentally friendly IL’s have been synthesized using Lignin and hemicellulose derived compounds. The reduced amination of Lignin monomers, furfural, vanillin, and P-anisaldehyde followed by treatment with phosphoric acid has generated ionic liquids including this.

So, these are some of the ionic liquids which are actually being generated from Lignin and lignin derived products.

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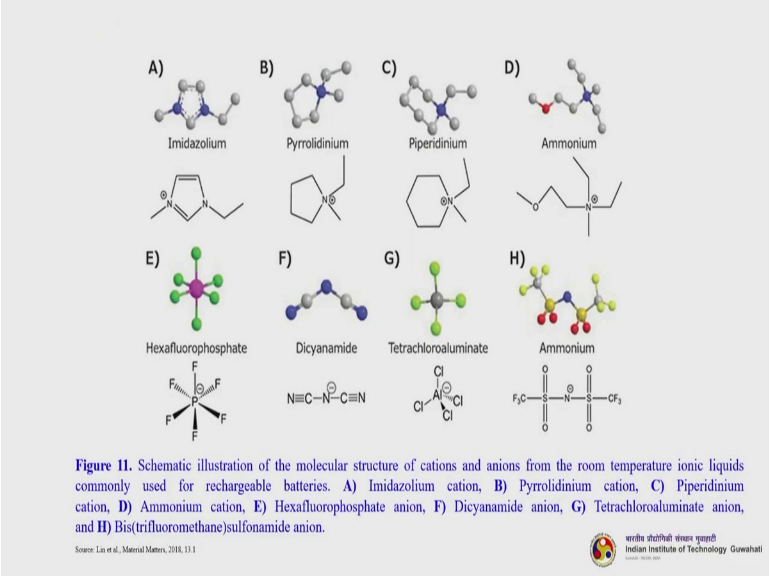

Figure 11. Schematic illustration of the molecular structure of cations and anions from the room temperature ionic liquids commonly used for rechargeable batteries. **A)** Imidazolium cation, **B)** Pyrrolidinium cation, **C)** Piperidinium cation, **D)** Ammonium cation, **E)** Hexafluorophosphate anion, **F)** Dicyanamide anion, **G)** Tetrachloroaluminate anion, and **H)** Bis(trifluoromethane)sulfonamide anion.

Source: Lin et al., Material Matters, 2018, 13.1

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How do you prepare ionic liquids? It is very simple, mix this and this; representative am telling you. what is that? These 4 on the top are all cations. These 4 on the bottom are all anions. So, any cation and any anion you fuse them together you get an ionic liquid. It is not so easy to do that. It can be done in the lab scale but you need further processing. Fusing together at certain conditions is fine.

But then the purity comes into picture. So, it may be 50%, 60%, 70% pure but then you need to purify to 80% 85% 90% it is again a challenge. So, these are different types of ionic liquids cations and anions; imidazolium based, pyrrolidinium based, Piperidinium based and based on ammonium. And these are some of the anions; hexafluorophosphate, dicyanamide, tetra chloroaluminate and ammonium.

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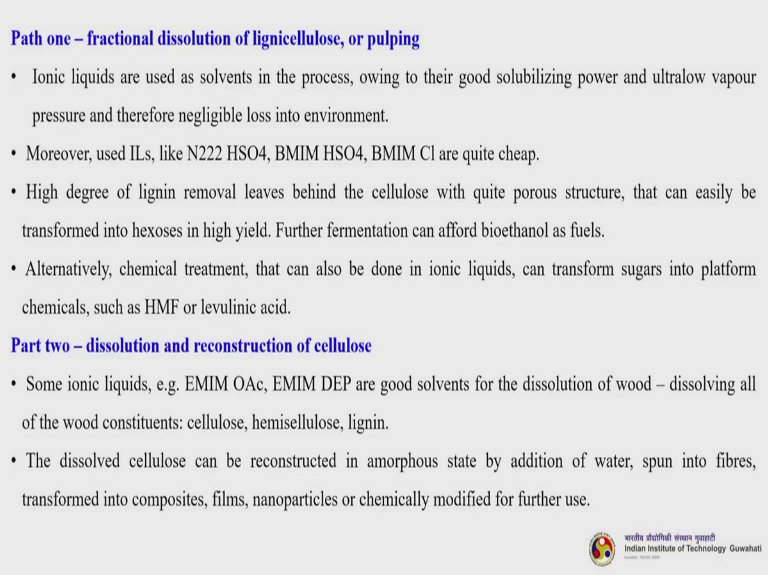

Path one – fractional dissolution of lignicellulose, or pulping

- Ionic liquids are used as solvents in the process, owing to their good solubilizing power and ultralow vapour pressure and therefore negligible loss into environment.
- Moreover, used ILs, like N222 HSO4, BMIM HSO4, BMIM Cl are quite cheap.
- High degree of lignin removal leaves behind the cellulose with quite porous structure, that can easily be transformed into hexoses in high yield. Further fermentation can afford bioethanol as fuels.
- Alternatively, chemical treatment, that can also be done in ionic liquids, can transform sugars into platform chemicals, such as HMF or levulinic acid.

Part two – dissolution and reconstruction of cellulose

- Some ionic liquids, e.g. EMIM OAc, EMIM DEP are good solvents for the dissolution of wood – dissolving all of the wood constituents: cellulose, hemisellulose, lignin.
- The dissolved cellulose can be reconstructed in amorphous state by addition of water, spun into fibres, transformed into composites, films, nanoparticles or chemically modified for further use.

So, there are 2 ways you can do. The path one is fractional dissolution of lignocellulose or pulping and path 2 is dissolution and reconstruction of cellulose.

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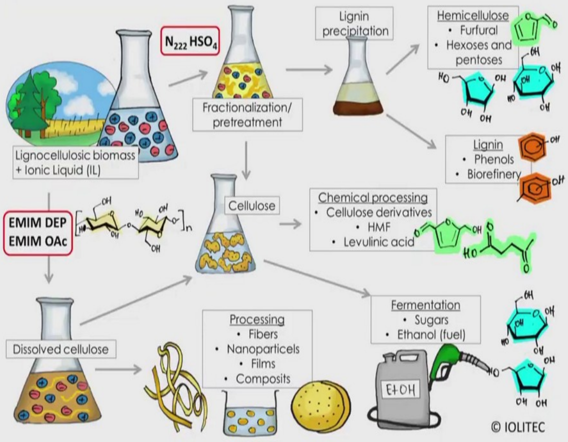

Figure 12: Pretreatment of lignocellulosic biomass using ionic liquid

Source: IOLITEC (Ionic liquid technologies)

Which is being explained in more detail in such a nice schematic representation, but let us first understand what is Path 1. So, ionic liquids are used as solvents in the process owing to their good solubilizing power and Ultra low Vapour pressure and therefore negligible loss into the environment. Moreover, used IL's like N222 HSO4, BMIM HSO4, BMIM Cl are quite cheap. I can tell you, see let us look at this, BMIM is your cation and HSO4 is anion, like that you can understand.

So that BMIM is the cation and Cl is the anion. So high degree of Lignin removal leaves behind the cellulose with quite porous structure that can easily be transformed into hexoses in high yield. Further fermentation can afford bioethanol is fuels. Alternatively, chemical pretreatment that can also be done in ionic liquid can transform sugar into platform chemical such as a HMF or Levulinic acid.

Part 2 is dissolution and reconstruction of cellulose. So, some ionic liquids like EMIM OAc, EMIM DEP are good solvent for the dissolution of wood - hardwoods basically - dissolving all of the wood constituents whether it is cellulose, hemicellulose and Lignin. Now, this dissolved cellulose can be reconstructed in amorphous state by addition of water, spun into fibres, transformed into composites, films, nanoparticles or chemically modified for further use.

So how these two things have been done here, we will try to understand. So, this is the lignocellulosic Biomass. So, you add ionic liquid to it. Then follow the proper protocol how to do it. There are steps and process conditions. Now the first one use EMIM DEP or EMIM OAc, you got, this is the part 2 basically which we have discussed, dissolved cellulose. Now this dissolved cellulose can be purified into cellulose.

It goes for further processing like cellulose derivatives, HMF, Levulinic acid and fermentation to Sugars, ethanol, this is alcohol or biofuel platform. So, then this dissolved cellulose can be further spun into fibres, nanoparticles, films and composite and you can use them. Now in the part 1 which discussed if you use N222 HSO4 and similar other ionic liquids so you are getting for a fractionalization.

So, you pre-treat and pretreatment process, then you get cellulose and further processing and whatever precipitation is remaining that is rich in Lignin basically, deep brown in colour. You can see the colour is also been shown here. Again, you further process it so you get 2 things hemicellulose rich fraction to be purified to get hemicellulose and lignin rich fraction you get lignin. So, you are getting cellulose, hemicellulose and Lignin in ionic liquid pretreatment process and all are in a better yield.

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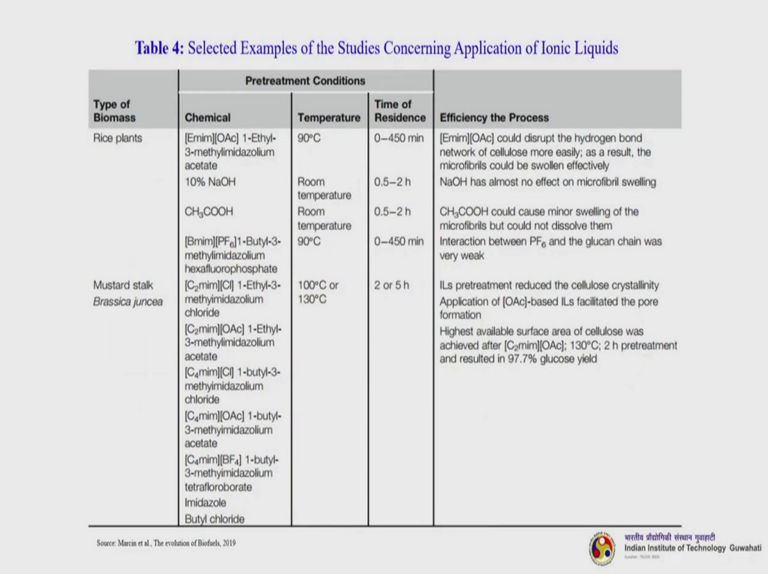

Table 4: Selected Examples of the Studies Concerning Application of Ionic Liquids

Type of Biomass	Pretreatment Conditions			Efficiency the Process
	Chemical	Temperature	Time of Residence	
Rice plants	[Emim][OAc] 1-Ethyl-3-methylimidazolium acetate	90°C	0–450 min	[Emim][OAc] could disrupt the hydrogen bond network of cellulose more easily; as a result, the microfibrils could be swollen effectively
	10% NaOH	Room temperature	0.5–2 h	NaOH has almost no effect on microfibril swelling
	CH_3COOH	Room temperature	0.5–2 h	CH_3COOH could cause minor swelling of the microfibrils but could not dissolve them
	[Bmim][PF_6]1-Butyl-3-methylimidazolium hexafluorophosphate	90°C	0–450 min	Interaction between PF_6 and the glucan chain was very weak
Mustard stalk *Brassica juncea*	[C_2mim][Cl] 1-Ethyl-3-methyimidazolium chloride [C_2mim][OAc] 1-Ethyl-3-methylimidazolium acetate [C_4mim][Cl] 1-butyl-3-methyimidazolium chloride [C_4mim][OAc] 1-butyl-3-methyimidazolium acetate [C_4mim][BF_4] 1-butyl-3-methyimidazolium tetrafloroborate Imidazole Butyl chloride	100°C or 130°C	2 or 5 h	ILs pretreatment reduced the cellulose crystallinity Application of [OAc]-based ILs facilitated the pore formation Highest available surface area of cellulose was achieved after [C_2mim][OAc]; 130°C; 2 h pretreatment and resulted in 97.7% glucose yield

Source: Marcin et al., The evolution of Biofuels, 2019

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So, this table is showing you selected examples of the studies concerning application of the ionic liquids. So again, I am not going to read out this. So please refer to it later. So, the temperature, residence time, efficiency of the process, what are the Chemicals, what are the different types of ionic liquids being used and these are the different types of biomass.

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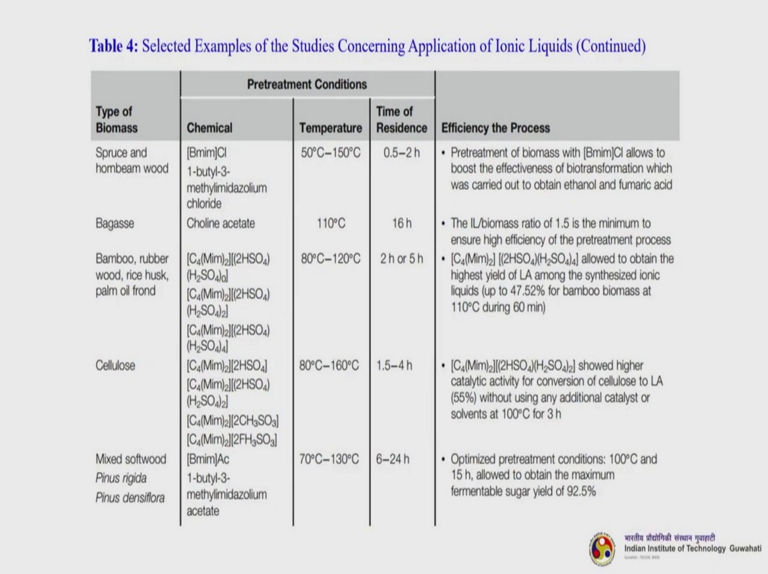

Table 4: Selected Examples of the Studies Concerning Application of Ionic Liquids (Continued)

Type of Biomass	Pretreatment Conditions			Efficiency the Process
	Chemical	Temperature	Time of Residence	
Spruce and hornbeam wood	[Bmim]Cl 1-butyl-3-methylimidazolium chloride	50°C–150°C	0.5–2 h	• Pretreatment of biomass with [Bmim]Cl allows to boost the effectiveness of biotransformation which was carried out to obtain ethanol and fumaric acid
Bagasse	Choline acetate	110°C	16 h	• The IL/biomass ratio of 1.5 is the minimum to ensure high efficiency of the pretreatment process
Bamboo, rubber wood, rice husk, palm oil frond	[C_4(Mim)$_2$][(2HSO_4)(H_2SO_4)$_0$] [C_4(Mim)$_2$][(2HSO_4)(H_2SO_4)$_2$] [C_4(Mim)$_2$][(2HSO_4)(H_2SO_4)$_4$]	80°C–120°C	2 h or 5 h	• [C_4(Mim)$_2$] [(2HSO_4)(H_2SO_4)$_4$] allowed to obtain the highest yield of LA among the synthesized ionic liquids (up to 47.52% for bamboo biomass at 110°C during 60 min)
Cellulose	[C_4(Mim)$_2$][2HSO_4] [C_4(Mim)$_2$][(2HSO_4)(H_2SO_4)$_2$] [C_4(Mim)$_2$][2CH_3SO_3] [C_4(Mim)$_2$][2FH_3SO_3]	80°C–160°C	1.5–4 h	• [C_4(Mim)$_2$][(2HSO_4)(H_2SO_4)$_2$] showed higher catalytic activity for conversion of cellulose to LA (55%) without using any additional catalyst or solvents at 100°C for 3 h
Mixed softwood *Pinus rigida* *Pinus densiflora*	[Bmim]Ac 1-butyl-3-methylimidazolium acetate	70°C–130°C	6–24 h	• Optimized pretreatment conditions: 100°C and 15 h, allowed to obtain the maximum fermentable sugar yield of 92.5%

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Similar table with different Biomass again.

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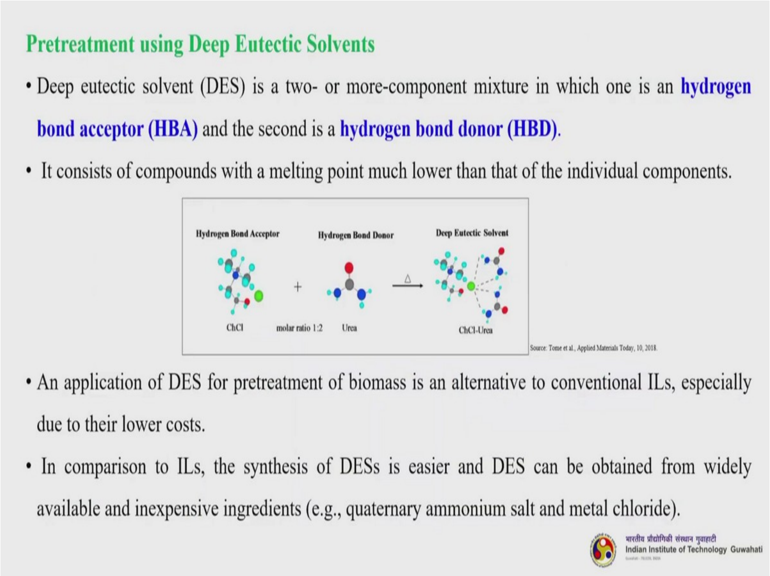

Pretreatment using Deep Eutectic Solvents

- Deep eutectic solvent (DES) is a two- or more-component mixture in which one is an **hydrogen bond acceptor (HBA)** and the second is a **hydrogen bond donor (HBD)**.
- It consists of compounds with a melting point much lower than that of the individual components.

Source: Tome et al., Applied Materials Today, 10, 2018.

- An application of DES for pretreatment of biomass is an alternative to conventional ILs, especially due to their lower costs.
- In comparison to ILs, the synthesis of DESs is easier and DES can be obtained from widely available and inexpensive ingredients (e.g., quaternary ammonium salt and metal chloride).

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So next class of solvent is Deep Eutectic Solvents. So deep eutectic solvents come in the picture as a result of certain disadvantages of the ionic liquids. So, DES, these are two or more components mixture in which one is a hydrogen bond acceptor, which is known as HBA, another is a hydrogen bond donor (HBD). So, you fuse together a hydrogen bond acceptor like ChCl or hydrogen bond donor like urea in a molar ratio that you decide.

So, you will get ChCl urea, which is the deep eutectic solvent. It is just like ionic liquid you are fusing one cation and anion. Here we are talking in terms of hydrogen bond acceptor and hydrogen donor. So, an application of the DES for pretreatment of biomass is an alternative to conventional IL's especially due to their lower cost. In comparison to the IL's the synthesis of DES is easier and DES can be obtained from widely available and inexpensive ingredients.

So, for example your quaternary ammonium salt and metal chloride; these are less costly than the ionic liquids.

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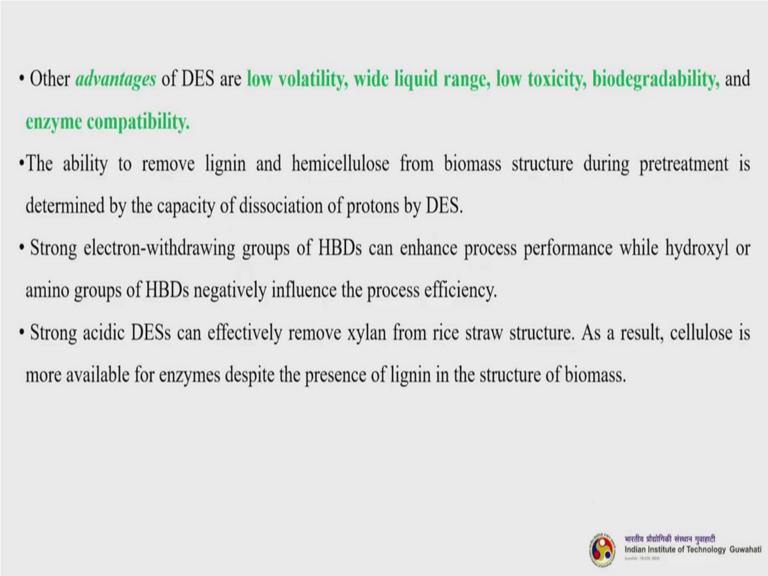

- Other *advantages* of DES are **low volatility, wide liquid range, low toxicity, biodegradability,** and **enzyme compatibility.**
- The ability to remove lignin and hemicellulose from biomass structure during pretreatment is determined by the capacity of dissociation of protons by DES.
- Strong electron-withdrawing groups of HBDs can enhance process performance while hydroxyl or amino groups of HBDs negatively influence the process efficiency.
- Strong acidic DESs can effectively remove xylan from rice straw structure. As a result, cellulose is more available for enzymes despite the presence of lignin in the structure of biomass.

The other advantages of DES are low-volatility, wide liquid range, low toxicity biodegradability and enzyme compatibility. Biodegradability is very important feature for a solvent to be extraordinarily green solvent. So, the ability to remove Lignin and hemicellulose from Biomass structure during pretreatment is determined by the capacity of the dissociation of protons by this DES.

Strong electron withdrawing groups of HBD's - hydrogen bond donors - can enhance process performance while hydroxyl or amino groups of the hydrogen bond donors negatively influence the process efficiency. So, strong acidic DES can effectively remove Xylan from rice straw structure. As a result, cellulose is more available for enzyme despite the presence of Lignin in the structure of biomass.

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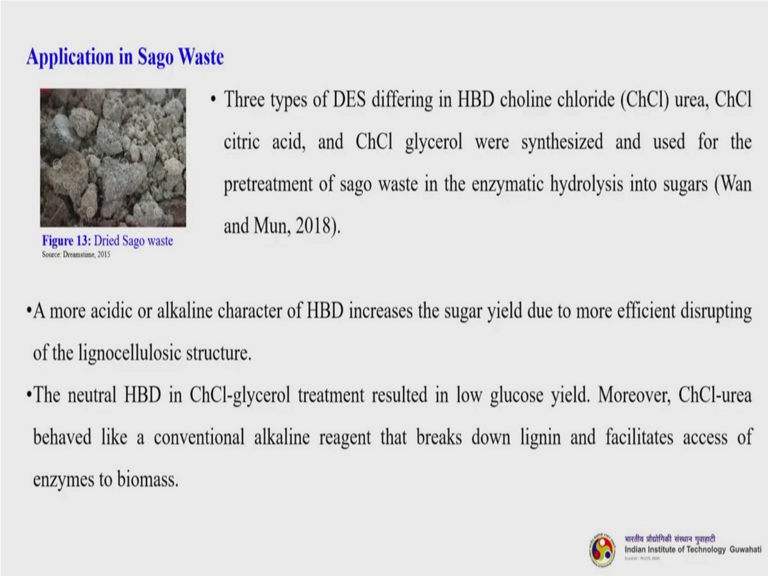

Application in Sago Waste

Figure 13: Dried Sago waste

Source: Dreamstime, 2015

- Three types of DES differing in HBD choline chloride (ChCl) urea, ChCl citric acid, and ChCl glycerol were synthesized and used for the pretreatment of sago waste in the enzymatic hydrolysis into sugars (Wan and Mun, 2018).
- A more acidic or alkaline character of HBD increases the sugar yield due to more efficient disrupting of the lignocellulosic structure.
- The neutral HBD in ChCl-glycerol treatment resulted in low glucose yield. Moreover, ChCl-urea behaved like a conventional alkaline reagent that breaks down lignin and facilitates access of enzymes to biomass.

So, we will see one application in Sago waste. So 3 types of DES differing in HBD choline chloride that is ChCl urea and ChCl citric acid and ChCl glycerol was synthesized and used for the pretreatment of sago waste in the enzymatic hydrolysis into sugar. So, basically three different types of Deep eutectic solvents are prepared. A more acidic or alkaline character of the hydrogen bond donor increases sugar yield due to more efficient disrupting of the lignocellulosic structure.

The neutral HBD in ChCl glycerol pretreatment resulted in low glucose yield. Moreover, ChCl urea DES deep eutectic solvent behaves like a conventional alkaline reagent that breaks down the Lignin and facilitate excess of enzymes to the Biomass.

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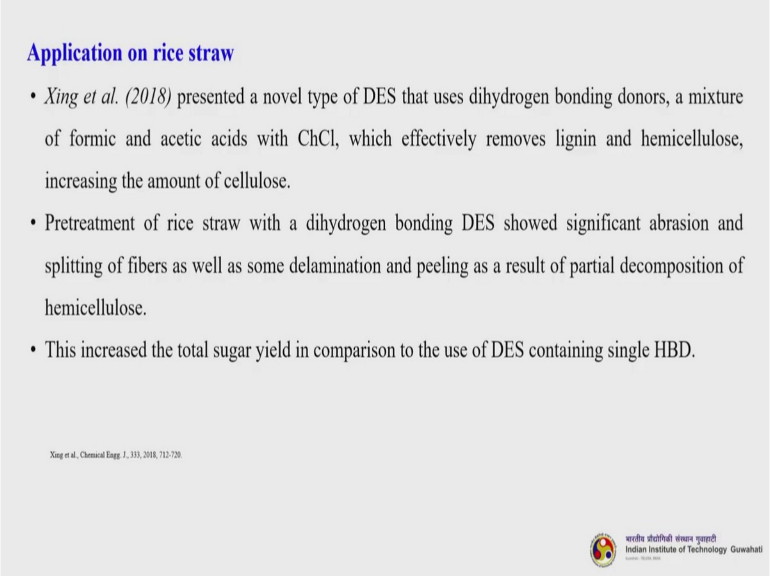

Application on rice straw

- *Xing et al. (2018)* presented a novel type of DES that uses dihydrogen bonding donors, a mixture of formic and acetic acids with ChCl, which effectively removes lignin and hemicellulose, increasing the amount of cellulose.
- Pretreatment of rice straw with a dihydrogen bonding DES showed significant abrasion and splitting of fibers as well as some delamination and peeling as a result of partial decomposition of hemicellulose.
- This increased the total sugar yield in comparison to the use of DES containing single HBD.

Xing et al., Chemical Engg. J., 333, 2018, 712-720.

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So, another application on rice straw. Xing et al presented a novel type of DES that uses the dihydrogen bonding donors, a mixture of formic acid and acetic acid with ChCl, which effectively removes Lignin and hemicellulose increasing the amount of cellulose. Pretreatment of rice straw with a dihydrogen bonding DES showed significant abrasion and splitting of fibres as well as some delamination and peeling as a result of partial decomposition of hemicellulose.

This increased the total sugar yield in comparison to the use of DES containing single hydrogen bond donor. This is a very good work. I have given you the reference. It is a published work in chemical engineering journal. Those who are interested please refer to this.

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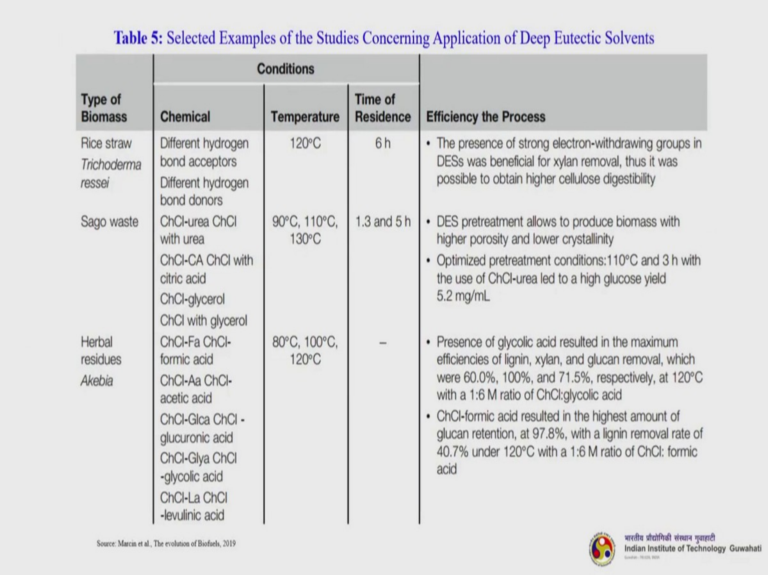

Table 5: Selected Examples of the Studies Concerning Application of Deep Eutectic Solvents

Type of Biomass	Conditions			Efficiency the Process
	Chemical	Temperature	Time of Residence	
Rice straw *Trichoderma ressei*	Different hydrogen bond acceptors Different hydrogen bond donors	120°C	6 h	• The presence of strong electron-withdrawing groups in DESs was beneficial for xylan removal, thus it was possible to obtain higher cellulose digestibility
Sago waste	ChCl-urea ChCl with urea ChCl-CA ChCl with citric acid ChCl-glycerol ChCl with glycerol	90°C, 110°C, 130°C	1.3 and 5 h	• DES pretreatment allows to produce biomass with higher porosity and lower crystallinity • Optimized pretreatment conditions:110°C and 3 h with the use of ChCl-urea led to a high glucose yield 5.2 mg/mL
Herbal residues *Akebia*	ChCl-Fa ChCl-formic acid ChCl-Aa ChCl-acetic acid ChCl-Glca ChCl -glucuronic acid ChCl-Glya ChCl -glycolic acid ChCl-La ChCl -levulinic acid	80°C, 100°C, 120°C	–	• Presence of glycolic acid resulted in the maximum efficiencies of lignin, xylan, and glucan removal, which were 60.0%, 100%, and 71.5%, respectively, at 120°C with a 1:6 M ratio of ChCl:glycolic acid • ChCl-formic acid resulted in the highest amount of glucan retention, at 97.8%, with a lignin removal rate of 40.7% under 120°C with a 1:6 M ratio of ChCl: formic acid

Source: Marcin et al., The evolution of Biofuels, 2019

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And again, one more table in which listed are the studies concerning application of the different Deep eutectic solvents with respect to different Biomass like say rice straw, Sago waste, herbal residues, what are the Chemicals and HDB's all that is being used, what is the temperature, time of residence and what is the efficiency. So please refer to this later on.

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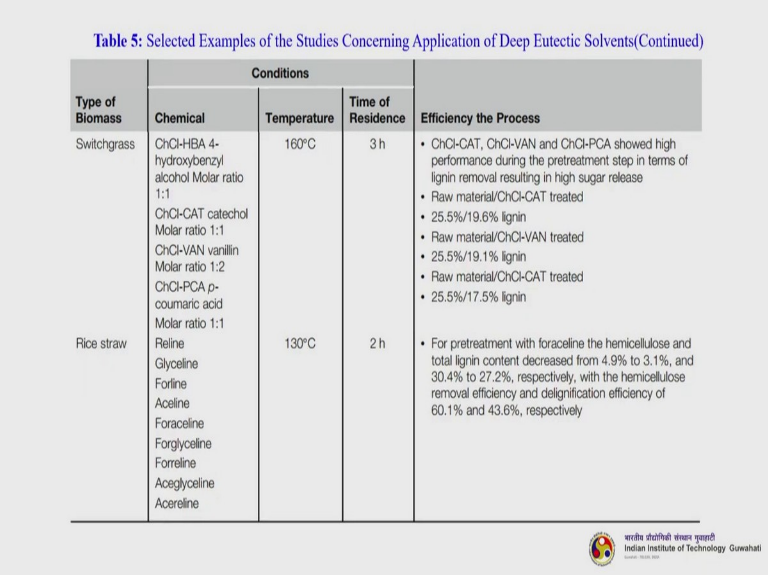

Table 5: Selected Examples of the Studies Concerning Application of Deep Eutectic Solvents(Continued)

Type of Biomass	Conditions			Efficiency the Process
	Chemical	Temperature	Time of Residence	
Switchgrass	ChCl-HBA 4-hydroxybenzyl alcohol Molar ratio 1:1 ChCl-CAT catechol Molar ratio 1:1 ChCl-VAN vanillin Molar ratio 1:2 ChCl-PCA *p*-coumaric acid Molar ratio 1:1	160°C	3 h	• ChCl-CAT, ChCl-VAN and ChCl-PCA showed high performance during the pretreatment step in terms of lignin removal resulting in high sugar release • Raw material/ChCl-CAT treated • 25.5%/19.6% lignin • Raw material/ChCl-VAN treated • 25.5%/19.1% lignin • Raw material/ChCl-CAT treated • 25.5%/17.5% lignin
Rice straw	Reline Glyceline Forline Aceline Foraceline Forglyceline Forreline Aceglyceline Acereline	130°C	2 h	• For pretreatment with foraceline the hemicellulose and total lignin content decreased from 4.9% to 3.1%, and 30.4% to 27.2%, respectively, with the hemicellulose removal efficiency and delignification efficiency of 60.1% and 43.6%, respectively

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Again, this is continuing with switchgrass and rice straw.

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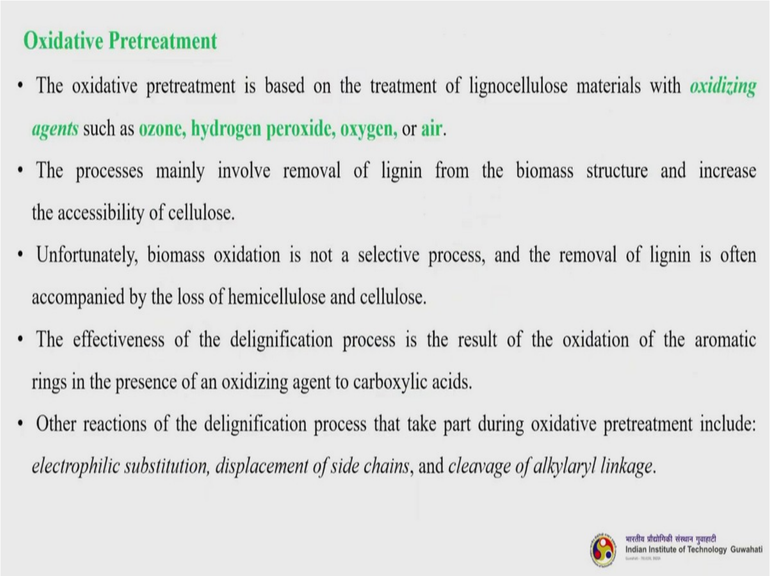

Oxidative Pretreatment

- The oxidative pretreatment is based on the treatment of lignocellulose materials with *oxidizing agents* such as ozone, hydrogen peroxide, oxygen, or air.
- The processes mainly involve removal of lignin from the biomass structure and increase the accessibility of cellulose.
- Unfortunately, biomass oxidation is not a selective process, and the removal of lignin is often accompanied by the loss of hemicellulose and cellulose.
- The effectiveness of the delignification process is the result of the oxidation of the aromatic rings in the presence of an oxidizing agent to carboxylic acids.
- Other reactions of the delignification process that take part during oxidative pretreatment include: *electrophilic substitution, displacement of side chains*, and *cleavage of alkylaryl linkage*.

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So will move to the next class of treatment which is called oxidative pretreatment. The oxidative pretreatment is based on the treatment of lignocellulosic materials with oxidizing agent. So, what are those? Those can be Ozone, hydrogen peroxide, oxygen or even air. The processes mainly involve removal of Lignin from the Biomass structure and increase the accessibility of cellulose. Unfortunately, Biomass oxidation is not a selective process and the removal of Lignin is often accompanied by the loss of hemicellulose and Cellulose.

The effectiveness of the delignification process is the result of the oxidation of the aromatic rings in the presence of an oxidizing agent to Carboxylic acids. Other reactions of the delignification process that take part during oxidative pretreatment include electrophilic substitution, displacement of sides chains, and cleavage of alkylaryl linkage. So, these are the other type of reactions.

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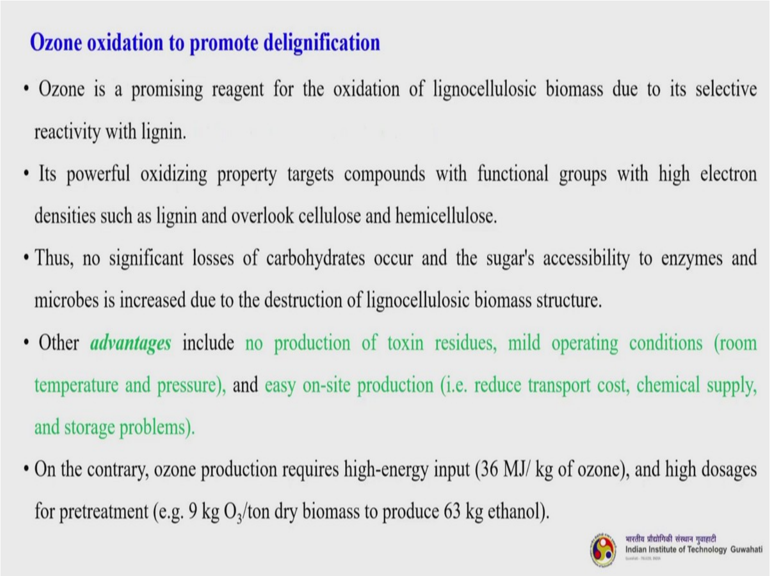

Ozone oxidation to promote delignification

- Ozone is a promising reagent for the oxidation of lignocellulosic biomass due to its selective reactivity with lignin.
- Its powerful oxidizing property targets compounds with functional groups with high electron densities such as lignin and overlook cellulose and hemicellulose.
- Thus, no significant losses of carbohydrates occur and the sugar's accessibility to enzymes and microbes is increased due to the destruction of lignocellulosic biomass structure.
- Other ***advantages*** include no production of toxin residues, mild operating conditions (room temperature and pressure), and easy on-site production (i.e. reduce transport cost, chemical supply, and storage problems).
- On the contrary, ozone production requires high-energy input (36 MJ/ kg of ozone), and high dosages for pretreatment (e.g. 9 kg O_3/ton dry biomass to produce 63 kg ethanol).

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Let us understand the Ozone oxidation to promote delignification. Ozone is a promising reagent for the oxidation of lignocellulosic biomass due to its selective reactivity lignin. Its powerful oxidising property target compounds with functional groups with high electron density such as Lignin and overlook cellulose and hemicellulose. Thus, no significant losses of Carbohydrates occur and the Sugars' accessibility to enzymes and microbes is increased due to the destruction of lignocellulosic biomass structure.

Advantages include: no production of toxic residues, mild operating conditions - basically room temperature and pressure, easy onsite production - that is reduced transport cost, chemical supply and storage problems. On the contrary Ozone production requires high energy inputs (36 megajoule per kg of Ozone) and high doses for pretreatment (example, 9kg Ozone per tonne of dry biomass to produce 63 kg of ethanol).

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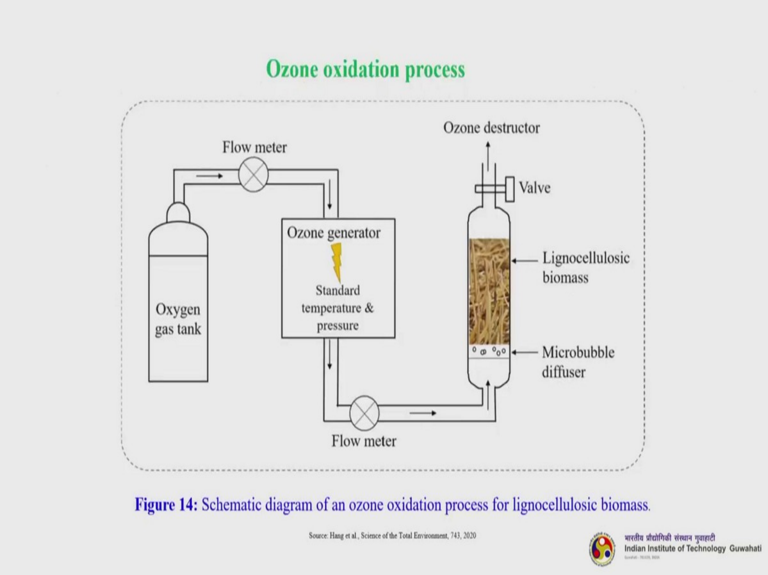

Ozone oxidation process

Figure 14: Schematic diagram of an ozone oxidation process for lignocellulosic biomass.

Source: Hang et al., Science of the Total Environment, 743, 2020

This is a schematic of the Ozone oxidation process. So, basically you have an Oxygen gas tank here - then by flowmeter it goes to a unit, which is basically Ozone generator. So, you maintain standard temperature and pressure. Now this ozone will be fed to a packed bed type of column which is having this lignocellulosic Biomass.

And you have a micro bubble diffuser - so the Ozone will be diffused and it will come in contact with the lignocellulosic Biomass, then whatever the Ozone is getting or moving out of this column has to be passed through an Ozone destructor. So again, convert Ozone to oxygen. So, this is very simplest way - this can be done in a large scale very easily. No harsh pretreatment condition and no requirement of sophisticated instrumentation only you need a Ozone generator and oxygen cylinder.

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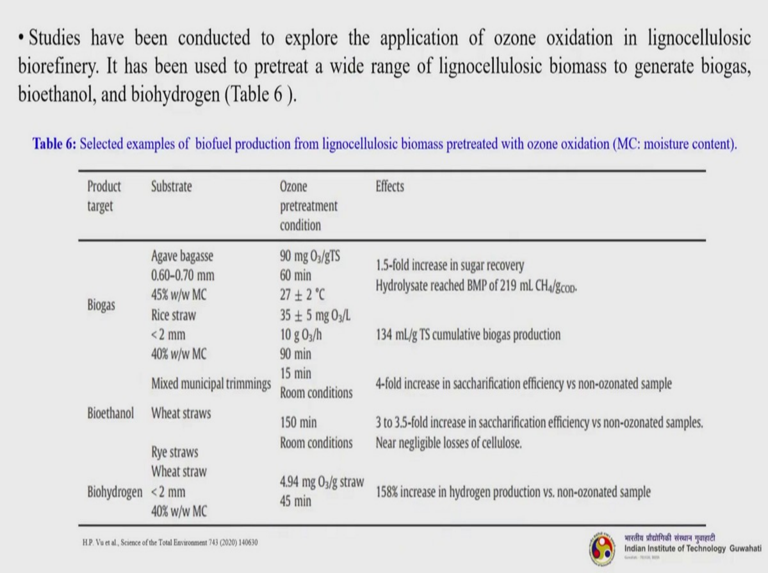

- Studies have been conducted to explore the application of ozone oxidation in lignocellulosic biorefinery. It has been used to pretreat a wide range of lignocellulosic biomass to generate biogas, bioethanol, and biohydrogen (Table 6).

Table 6: Selected examples of biofuel production from lignocellulosic biomass pretreated with ozone oxidation (MC: moisture content).

Product target	Substrate	Ozone pretreatment condition	Effects
Biogas	Agave bagasse 0.60–0.70 mm 45% w/w MC	90 mg O_3/gTS 60 min 27 ± 2 °C	1.5-fold increase in sugar recovery Hydrolysate reached BMP of 219 mL CH_4/g_{COD}.
	Rice straw < 2 mm 40% w/w MC	35 ± 5 mg O_3/L 10 g O_3/h 90 min	134 mL/g TS cumulative biogas production
Bioethanol	Mixed municipal trimmings	15 min Room conditions	4-fold increase in saccharification efficiency vs non-ozonated sample
	Wheat straws Rye straws	150 min Room conditions	3 to 3.5-fold increase in saccharification efficiency vs non-ozonated samples. Near negligible losses of cellulose.
Biohydrogen	Wheat straw < 2 mm 40% w/w MC	4.94 mg O_3/g straw 45 min	158% increase in hydrogen production vs. non-ozonated sample

H.P. Vu et al., Science of the Total Environment 743 (2020) 140630

So, studies have been conducted to explore the application of Ozone oxidation in lignocellulosic bio-refinery. It has been used to pretreat a wide range of lignocellulosic biomass to generate biogas, bioethanol and biohydrogen. This table again list out selected examples of biofuel production from lignocellulosic Biomass pre-treated with Ozone oxidation. So, MC is moisture content. So you can see it is written 45% w/w MC that is moisture content.

So, it is biogas. The target is biogas production, bioethanol production, biohydrogen production. So, please refer to the slide later on.

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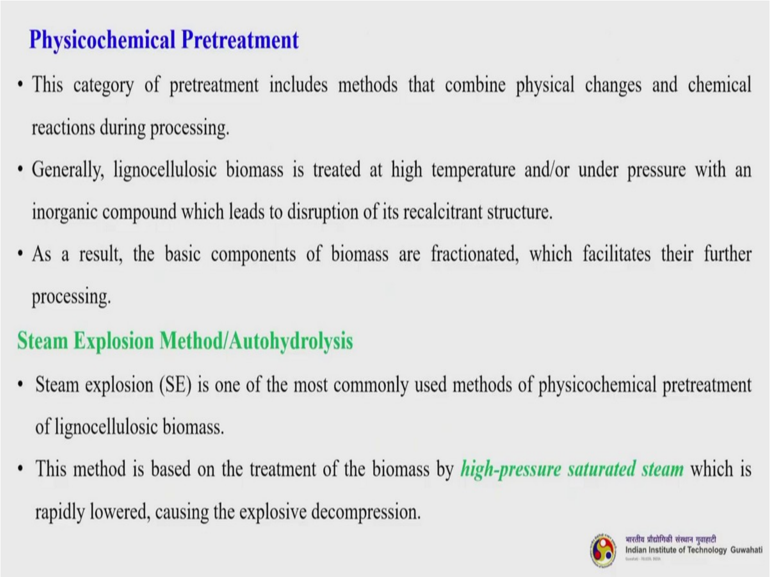

Physicochemical Pretreatment

- This category of pretreatment includes methods that combine physical changes and chemical reactions during processing.
- Generally, lignocellulosic biomass is treated at high temperature and/or under pressure with an inorganic compound which leads to disruption of its recalcitrant structure.
- As a result, the basic components of biomass are fractionated, which facilitates their further processing.

Steam Explosion Method/Autohydrolysis

- Steam explosion (SE) is one of the most commonly used methods of physicochemical pretreatment of lignocellulosic biomass.
- This method is based on the treatment of the biomass by ***high-pressure saturated steam*** which is rapidly lowered, causing the explosive decompression.

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So, we will talk about the physicochemical pretreatment methods. This category of pretreatment includes methods that combine physical changes and chemical reactions during the processing hence the name physicochemical. Generally, lignocellulosic Biomass is treated at high temperature and under pressure with an inorganic compound which leads to disruption of its recalcitrant structure. As a result the basic components of biomass are fractionated, which facilitates further processing.

We will see one example, one such pre-treatment method which is steam explosion method or auto hydrolysis. So, Steam explosion is one of the most commonly used methods of physicochemical pretreatment of lignocellulosic biomass. So, this method is based on the treatment of the biomass by high pressure saturated steam, which is rapidly lowered. That is why it is causing explosion. Causing the explosive decompression.

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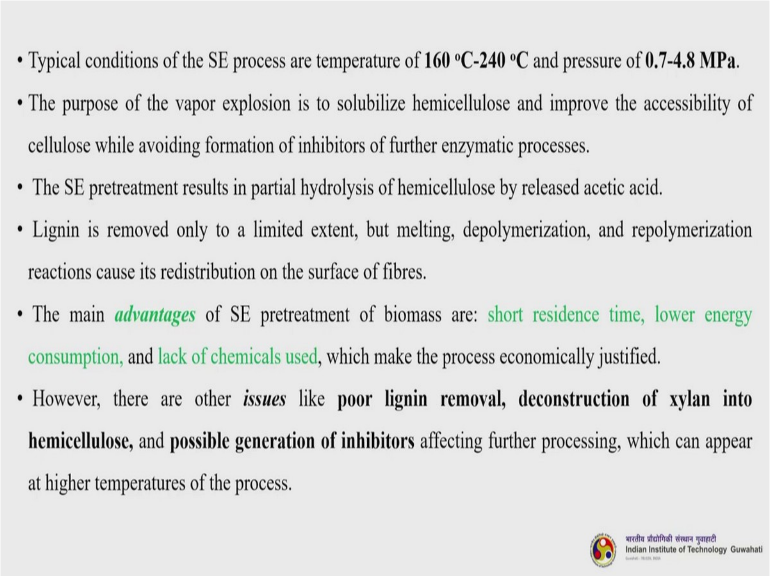

- Typical conditions of the SE process are temperature of **160 °C-240 °C** and pressure of **0.7-4.8 MPa**.
- The purpose of the vapor explosion is to solubilize hemicellulose and improve the accessibility of cellulose while avoiding formation of inhibitors of further enzymatic processes.
- The SE pretreatment results in partial hydrolysis of hemicellulose by released acetic acid.
- Lignin is removed only to a limited extent, but melting, depolymerization, and repolymerization reactions cause its redistribution on the surface of fibres.
- The main ***advantages*** of SE pretreatment of biomass are: short residence time, lower energy consumption, and lack of chemicals used, which make the process economically justified.
- However, there are other ***issues*** like **poor lignin removal, deconstruction of xylan into hemicellulose,** and **possible generation of inhibitors** affecting further processing, which can appear at higher temperatures of the process.

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So typical conditions of the steam explosion process are temperature 160 to 240 degree and a pressure of 0.7 to 4.8 Mega Pascal. The purpose of the vapour explosion is to solubilize hemicellulose and improve the accessibility of cellulose while avoiding formation of inhibitors for further enzymatic processes. The steam explosion pretreatment results in partial hydrolysis of hemicellulose by released acetic acid.

Lignin is removed only to a limited extent, but melting, depolymerization and repolymerization reaction causes its redistribution on the surface of the fibre. The main advantages of the steam explosive pretreatment are residence time is very short - so, it is good for commercial application, low energy consumption and lack of chemicals used. So the entire process is very economically justified.

However, there are certain issues, like poor lignin removal, deconstruction of Xylan into hemicellulose and possible generation of inhibitors affecting further processing. Again this HMF and acetic acid and all this. So, if you go for a high temperature application.

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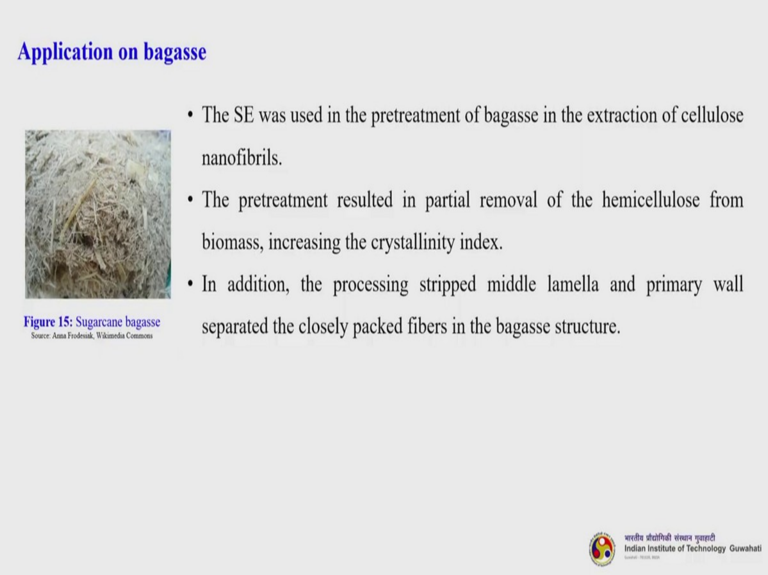

Application on bagasse

- The SE was used in the pretreatment of bagasse in the extraction of cellulose nanofibrils.
- The pretreatment resulted in partial removal of the hemicellulose from biomass, increasing the crystallinity index.
- In addition, the processing stripped middle lamella and primary wall separated the closely packed fibers in the bagasse structure.

Figure 15: Sugarcane bagasse
Source: Anna Frodesiak, Wikimedia Commons

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So, we will see one application on bagasse. So, steam explosion was used in the pretreatment of bagasse in the extraction of cellulose nanofibrils. The pretreatment result in partial removal of hemicellulose from Biomass increasing the crystallinity index. In addition, the processing stripped middle lamella and primary wall separated the closely packed fibres in the bagasse structure.

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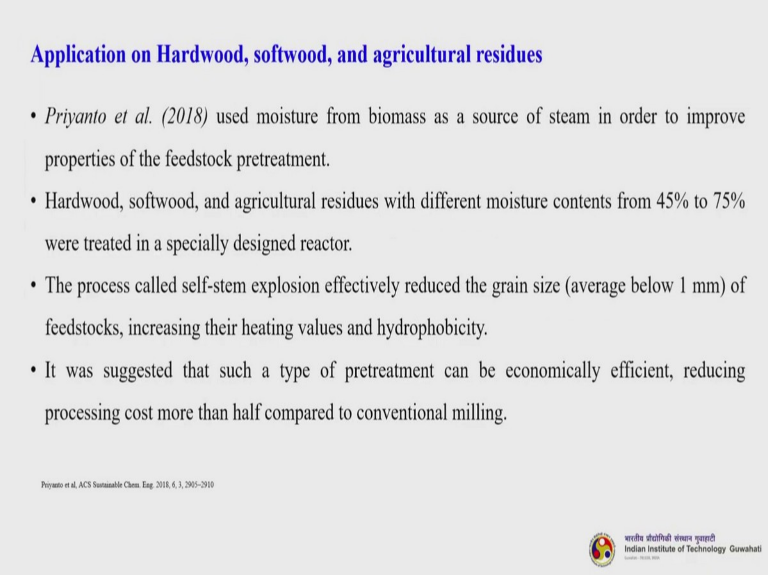

Application on Hardwood, softwood, and agricultural residues

- *Priyanto et al. (2018)* used moisture from biomass as a source of steam in order to improve properties of the feedstock pretreatment.
- Hardwood, softwood, and agricultural residues with different moisture contents from 45% to 75% were treated in a specially designed reactor.
- The process called self-stem explosion effectively reduced the grain size (average below 1 mm) of feedstocks, increasing their heating values and hydrophobicity.
- It was suggested that such a type of pretreatment can be economically efficient, reducing processing cost more than half compared to conventional milling.

Priyanto et al, ACS Sustainable Chem. Eng. 2018, 6, 3, 2905−2910

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Another application on hardwood, soft wood and agricultural residues. Priyanto et al, this is a very good study published in ACS Sustainable Chemistry engineering. So, what they did actually they used moisture from Biomass as a source of steam in order to improve properties of feedstock pretreatment. This is very interesting study. Hardwood, softwood and agricultural residues with different moisture content from 45% to 75% were treated in a specially designed reactor.

The process called self-steam explosion effectively reduced the grain size (almost average below 1 mm) of feedstock increasing the heating values and hydrophobicity. It was suggested that such a type of pretreatment can be economically efficient reducing processing cost more than half compared to the conventional milling.

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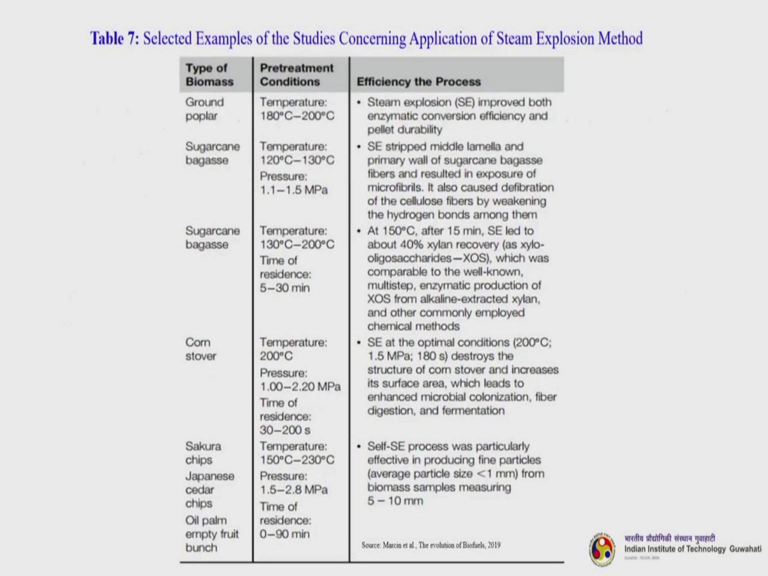

Table 7: Selected Examples of the Studies Concerning Application of Steam Explosion Method

Type of Biomass	Pretreatment Conditions	Efficiency the Process
Ground poplar	Temperature: 180°C–200°C	• Steam explosion (SE) improved both enzymatic conversion efficiency and pellet durability
Sugarcane bagasse	Temperature: 120°C–130°C Pressure: 1.1–1.5 MPa	• SE stripped middle lamella and primary wall of sugarcane bagasse fibers and resulted in exposure of microfibrils. It also caused defibration of the cellulose fibers by weakening the hydrogen bonds among them
Sugarcane bagasse	Temperature: 130°C–200°C Time of residence: 5–30 min	• At 150°C, after 15 min, SE led to about 40% xylan recovery (as xylo-oligosaccharides—XOS), which was comparable to the well-known, multistep, enzymatic production of XOS from alkaline-extracted xylan, and other commonly employed chemical methods
Corn stover	Temperature: 200°C Pressure: 1.00–2.20 MPa Time of residence: 30–200 s	• SE at the optimal conditions (200°C; 1.5 MPa; 180 s) destroys the structure of corn stover and increases its surface area, which leads to enhanced microbial colonization, fiber digestion, and fermentation
Sakura chips Japanese cedar chips Oil palm empty fruit bunch	Temperature: 150°C–230°C Pressure: 1.5–2.8 MPa Time of residence: 0–90 min	• Self-SE process was particularly effective in producing fine particles (average particle size <1 mm) from biomass samples measuring 5 – 10 mm

Source: Marcin et al., The evolution of Biofuels, 2019

This table is giving you selected examples of some of the studies concerning application of the steam explosion method. Various types of biomass, their pretreatment conditions and the efficiency has been listed, please refer to it later on.

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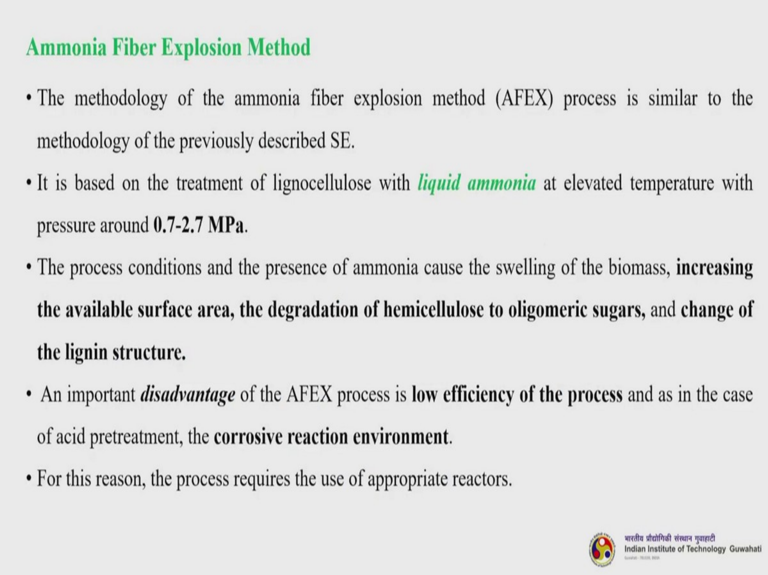

Ammonia Fiber Explosion Method

- The methodology of the ammonia fiber explosion method (AFEX) process is similar to the methodology of the previously described SE.
- It is based on the treatment of lignocellulose with ***liquid ammonia*** at elevated temperature with pressure around **0.7-2.7 MPa**.
- The process conditions and the presence of ammonia cause the swelling of the biomass, **increasing the available surface area, the degradation of hemicellulose to oligomeric sugars,** and **change of the lignin structure.**
- An important ***disadvantage*** of the AFEX process is **low efficiency of the process** and as in the case of acid pretreatment, the **corrosive reaction environment**.
- For this reason, the process requires the use of appropriate reactors.

So, we move ahead to one of the again more studied method which is called AFEX that is Ammonia fibre explosion method. So here, it is similar to the steam explosion. So here it is based in the pretreatment of lignocellulose with liquid Ammonia at elevated temperature. With the pressure around 0.7 to 2.7 mega pascal. The process conditions and the presence of ammonia causes the swelling of the biomass, increasing the available surface area, the degradation of hemicellulose to oligomeric sugars and the change of the lignin structure. Basically, the mechanism is that, the ammonia is swelling the Biomass making it more porous and they will be more accessible to the enzyme. So an important disadvantage of this AFEX process is the low efficiency of the process as in the case of acid pretreatment, the corrosive reaction and environment also.

So, for this reason the process requires the use of appropriate reactors and hence again additional cost come into the picture.

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Application on corn stover, prairie cord grass, and switchgrass

Figure 16: Corn stover
Source: Jessica Eise, phys.org, 2015

Figure 17: Switch grass
Source: Dennis Pennington, Farm Energy, 2019

Figure 18: Prairie cord grass
Source: Minnesota Water Stewards

- The AFEX process was used as a pretreatment method of corn stover, prairie cord grass, and switch grass before the pyrolysis process.
- The impact of the treatment was negligible and did not significantly improve the properties of the tested materials.
- Parameters such as moisture content, volatile matter, ash content, or heating values were almost the same before and after treatment.
- However, the AFEX process increased pellet durability and decreased the temperature of sample degradation.
- During the pyrolysis process, the yield to bio-char and to bio-oil increased only from 22% to 25% and from 46% to 48%, respectively, after AFEX pretreatment.

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We will see some of these AFEX study. So the AFEX process was used as a pretreatment method of Corn stover, prairie cord grass and switchgrass before the pyrolysis process. All this has been used for pyrolysis but before that they are pre-treated with AFEX to increase the pyrolytic product yield. The impact of the treatment was negligible and did not significantly improved the properties of tested materials.

Parameters such as moisture content, volatile matter, ash content or heating values were almost the same before and after pretreatment. However, the AFEX process increased pellet durability and decrease the temperature of the sample degradation, not much effect has been actually reported. During the pyrolysis process the yield just increased for biochar from 22 to

25% and for the bio-oil it is 46 to 48% after the AFEX pretreatment which is very, very marginal actually not significant.

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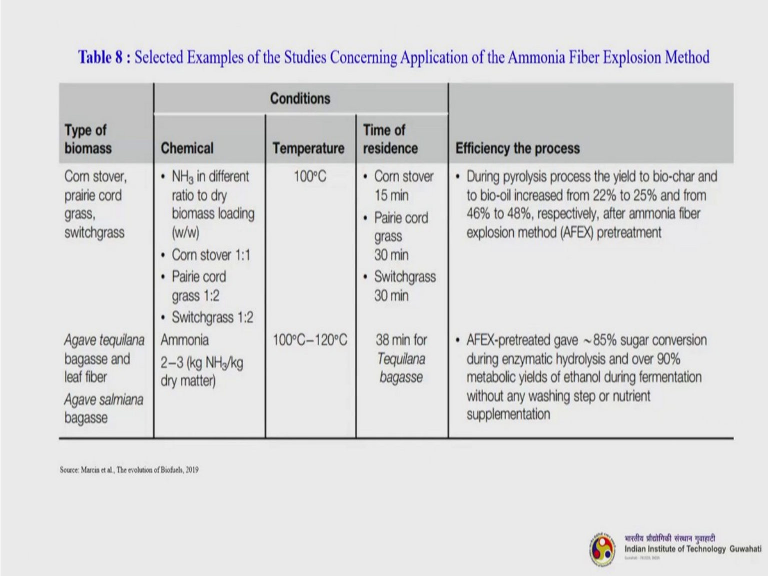

Table 8 : Selected Examples of the Studies Concerning Application of the Ammonia Fiber Explosion Method

Type of biomass	Conditions			Efficiency the process
	Chemical	Temperature	Time of residence	
Corn stover, prairie cord grass, switchgrass	• NH_3 in different ratio to dry biomass loading (w/w) • Corn stover 1:1 • Pairie cord grass 1:2 • Switchgrass 1:2	100°C	• Corn stover 15 min • Pairie cord grass 30 min • Switchgrass 30 min	• During pyrolysis process the yield to bio-char and to bio-oil increased from 22% to 25% and from 46% to 48%, respectively, after ammonia fiber explosion method (AFEX) pretreatment
Agave tequilana bagasse and leaf fiber *Agave salmiana* bagasse	Ammonia 2–3 (kg NH_3/kg dry matter)	100°C–120°C	38 min for *Tequilana bagasse*	• AFEX-pretreated gave ~85% sugar conversion during enzymatic hydrolysis and over 90% metabolic yields of ethanol during fermentation without any washing step or nutrient supplementation

Source: Marcin et al., The evolution of Biofuels, 2019

This is again selected examples of the studies concerning application of the AFEX method. Various biomass is like cornstover, grass, switchgrass, which we just saw. Agava, bagasse and leaf, *Agava salmiana* bagasse. So please see the different chemical that is listed. The conditions, time of residence and the efficiency of the process, please refer to it later.

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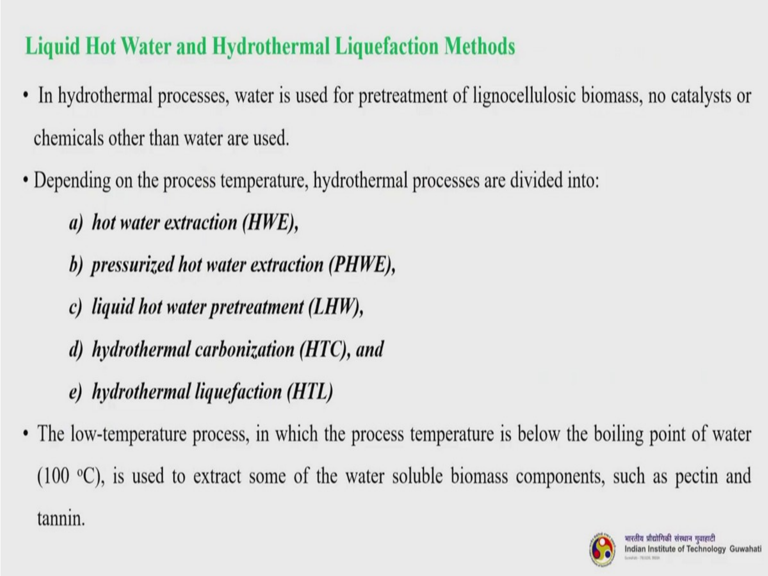

Liquid Hot Water and Hydrothermal Liquefaction Methods

- In hydrothermal processes, water is used for pretreatment of lignocellulosic biomass, no catalysts or chemicals other than water are used.
- Depending on the process temperature, hydrothermal processes are divided into:
  - *a) hot water extraction (HWE),*
  - *b) pressurized hot water extraction (PHWE),*
  - *c) liquid hot water pretreatment (LHW),*
  - *d) hydrothermal carbonization (HTC), and*
  - *e) hydrothermal liquefaction (HTL)*
- The low-temperature process, in which the process temperature is below the boiling point of water (100 °C), is used to extract some of the water soluble biomass components, such as pectin and tannin.

And we will see one of the most important class of this pretreatment process, which is called liquid hot water and hydrothermal liquefaction methods. In hydrothermal process water is used for the pretreatment of lignocellulosic biomass. No catalyst or chemicals other than water are used. So that it is called hydrothermal method. So, depending on the process temperature hydrothermal processes are divided into 5 types.

(a) The first one is the hot water extraction; (b) second one is pressurised hot water extraction; (c) Third one is the liquid hot water pretreatment; (d) forth one is hydrothermal carbonization or HTC; and, (e) hydrothermal liquefaction or HTL. HTC and HCL are widely studied since last decade and have lot of applications and excellent studies have been reported. So, the low-temperature process in which the process temperature is below the boiling point of the water (less than hundred degree centigrade) is used to extract some of the water-soluble Biomass components such as pectin and tannin.

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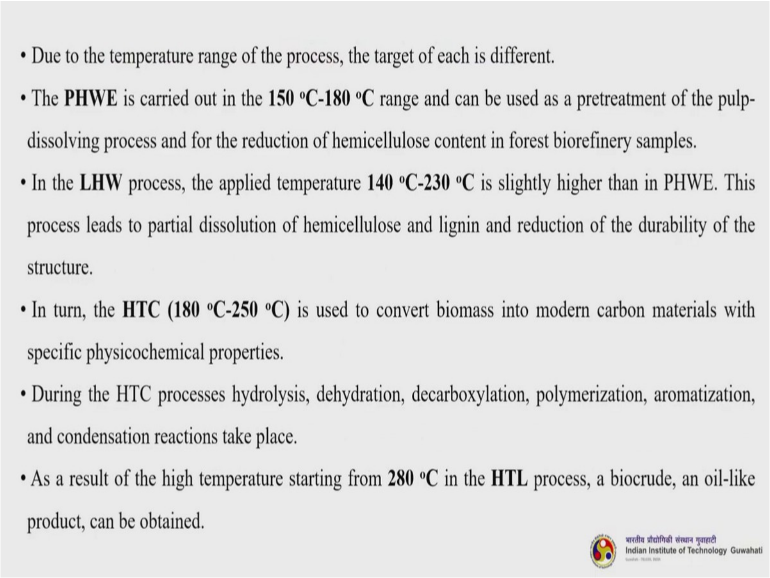

- Due to the temperature range of the process, the target of each is different.
- The **PHWE** is carried out in the **150 °C-180 °C** range and can be used as a pretreatment of the pulp-dissolving process and for the reduction of hemicellulose content in forest biorefinery samples.
- In the **LHW** process, the applied temperature **140 °C-230 °C** is slightly higher than in PHWE. This process leads to partial dissolution of hemicellulose and lignin and reduction of the durability of the structure.
- In turn, the **HTC (180 °C-250 °C)** is used to convert biomass into modern carbon materials with specific physicochemical properties.
- During the HTC processes hydrolysis, dehydration, decarboxylation, polymerization, aromatization, and condensation reactions take place.
- As a result of the high temperature starting from **280 °C** in the **HTL** process, a biocrude, an oil-like product, can be obtained.

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Due to the temperature range of the process, the target of each is different. So that we need to understand. Here whatever is being listed you’re seeing, everything has a certain different temperature. The temperature, temperature range depending upon their product whatever coming out will also be different. We will see. So in the pressurized hot water extraction, it is carried out in the 150 to 180 degree centigrade in that range and can be used as a pretreatment of the pulp dissolving process and for the reduction of hemicellulose content in the forest bio refineries samples.

In the liquid hot water process the applied temperature is 140 to 230 degree centigrade is little or a we can say slightly higher than the PHWE. So this process leads to partial dissolution of hemicellulose and Lignin and reduction of the durability of the structure. In turn in the HTC or the hydrothermal carbonization, which is carried out at 180 to 250 degree centigrade is used to convert Biomass into modern carbon materials with specific physicochemical properties.

High quality carbon can be prepared that is why the name is actually hydrothermal carbonization. Then during the HTC process hydrolysis, dehydration, decarboxylation, polymerization, aromatization and condensation reaction takes place. As a result of the high-temperature starting from 280 degree centigrade in the HTL process, a biocrude that is oil like product can be obtained. We will discuss HTL later on, its a very good process actually.

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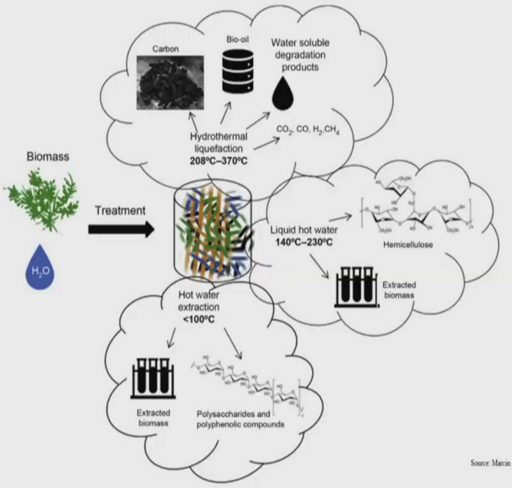

Source: Marcin et al., The evolution of Biofuels, 2019

Figure 19: Visual presentation of hydrothermal methods

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Whatever we have discussed this has been given in this particular some sort of schematic representation of graphical abstract type you can see. Biomass, the treatment method. You can see this. The first one is the hydrothermal liquefaction HTL which was discussed at the end. So that converts, you will get Carbon, you get bio-oil and you get water soluble degradation products.

Apart from that you get carbon dioxide, carbon monoxide, hydrogen and methane. When you go for the liquid hot water pretreatment, you get hemicellulose and extracted biomass here. The solid biomass can be fractionated again into valuable products. For hot water extraction, which is below 100 degree centigrade we get extracted biomass which can be for the processed to again value-added products and we get the polysaccharides and polyphenolic compounds.

So, this is the lowest in the lowest range of the hydrothermal processes and HTL is the highest range in terms of the temperature and the uses.

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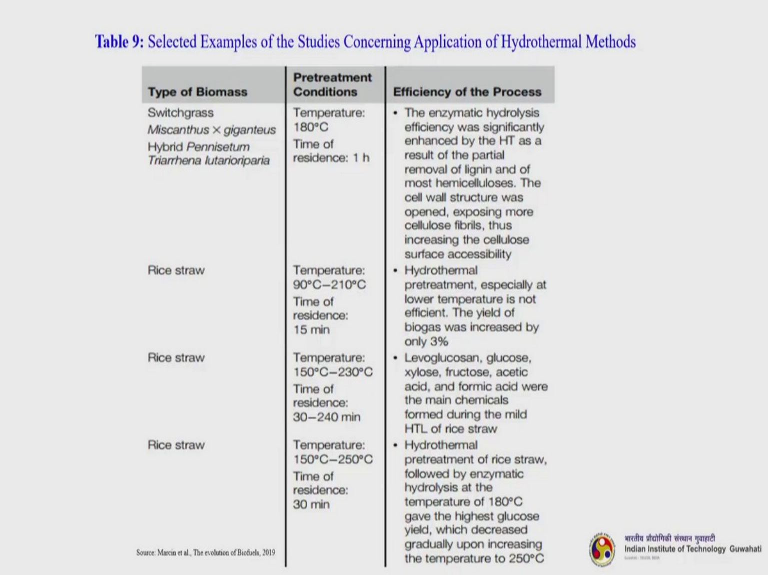

Table 9: Selected Examples of the Studies Concerning Application of Hydrothermal Methods

Type of Biomass	Pretreatment Conditions	Efficiency of the Process
Switchgrass *Miscanthus × giganteus* Hybrid *Pennisetum* *Triarrhena lutarioriparia*	Temperature: 180°C Time of residence: 1 h	• The enzymatic hydrolysis efficiency was significantly enhanced by the HT as a result of the partial removal of lignin and of most hemicelluloses. The cell wall structure was opened, exposing more cellulose fibrils, thus increasing the cellulose surface accessibility
Rice straw	Temperature: 90°C–210°C Time of residence: 15 min	• Hydrothermal pretreatment, especially at lower temperature is not efficient. The yield of biogas was increased by only 3%
Rice straw	Temperature: 150°C–230°C Time of residence: 30–240 min	• Levoglucosan, glucose, xylose, fructose, acetic acid, and formic acid were the main chemicals formed during the mild HTL of rice straw
Rice straw	Temperature: 150°C–250°C Time of residence: 30 min	• Hydrothermal pretreatment of rice straw, followed by enzymatic hydrolysis at the temperature of 180°C gave the highest glucose yield, which decreased gradually upon increasing the temperature to 250°C

Source: Marcin et al., The evolution of Biofuels, 2019

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So, a table line gives you the studies concerning application of the hydrothermal method. So again, I am telling you that you please refer to it later on. So different Biomass are listed, different pretreatment conditions and their efficiency of the process has been listed. So, this will help you if at all going to do some work on Biomass fractionation or biomass pretreatment.

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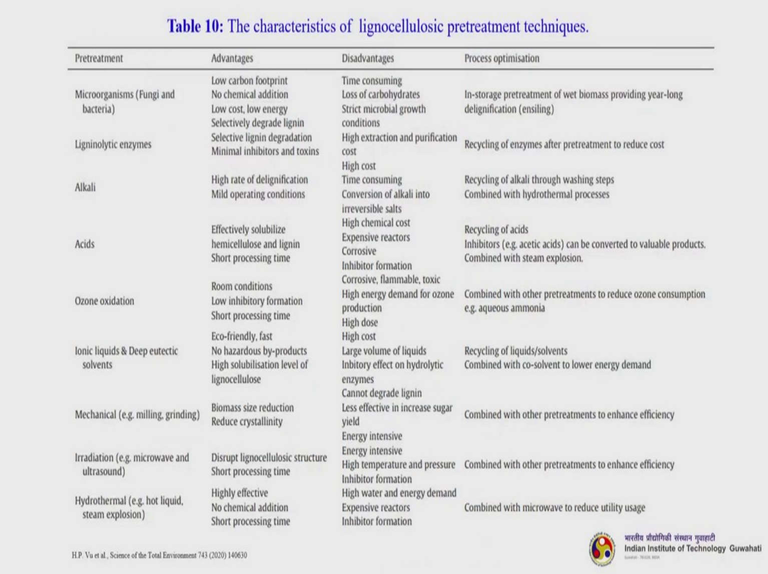

Table 10: The characteristics of lignocellulosic pretreatment techniques.

Pretreatment	Advantages	Disadvantages	Process optimisation
Microorganisms (Fungi and bacteria)	Low carbon footprint No chemical addition Low cost, low energy Selectively degrade lignin	Time consuming Loss of carbohydrates Strict microbial growth conditions	In-storage pretreatment of wet biomass providing year-long delignification (ensiling)
Ligninolytic enzymes	Selective lignin degradation Minimal inhibitors and toxins	High extraction and purification cost	Recycling of enzymes after pretreatment to reduce cost
Alkali	High rate of delignification Mild operating conditions	High cost Time consuming Conversion of alkali into irreversible salts	Recycling of alkali through washing steps Combined with hydrothermal processes
Acids	Effectively solubilize hemicellulose and lignin Short processing time	High chemical cost Expensive reactors Corrosive Inhibitor formation	Recycling of acids Inhibitors (e.g. acetic acids) can be converted to valuable products. Combined with steam explosion.
Ozone oxidation	Room conditions Low inhibitory formation Short processing time	Corrosive, flammable, toxic High energy demand for ozone production High dose	Combined with other pretreatments to reduce ozone consumption e.g. aqueous ammonia
Ionic liquids & Deep eutectic solvents	Eco-friendly, fast No hazardous by-products High solubilisation level of lignocellulose	High cost Large volume of liquids Inbitory effect on hydrolytic enzymes	Recycling of liquids/solvents Combined with co-solvent to lower energy demand
Mechanical (e.g. milling, grinding)	Biomass size reduction Reduce crystallinity	Cannot degrade lignin Less effective in increase sugar yield Energy intensive	Combined with other pretreatments to enhance efficiency
Irradiation (e.g. microwave and ultrasound)	Disrupt lignocellulosic structure Short processing time	Energy intensive High temperature and pressure Inhibitor formation	Combined with other pretreatments to enhance efficiency
Hydrothermal (e.g. hot liquid, steam explosion)	Highly effective No chemical addition Short processing time	High water and energy demand Expensive reactors Inhibitor formation	Combined with microwave to reduce utility usage

H.P. Vu et al., Science of the Total Environment 743 (2020) 140630

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We will see this the characteristics of the lignocellulosic pretreatment techniques. This is an overall idea. So, you can see this, different pre-treatment methods like microorganisms - the biological one, followed by ligninolytic enzymes, alkali method, acids or dilute acids method, Ozone oxidation, ionic liquids and deep eutectic solvents, you have mechanical - this is a mechanical and physical method milling and grinding, then irradiation using using microwave, ultrasound, hydrothermal - like hot liquid and steam explosion. So, I will leave it to you for read and referred to it later on. There is not much to again describe or there is no much point in reading. Whatever being listed here we have already discussed throughout our today's lecture. This in a single table is this has been compared. So, you can understand what are the advantages, disadvantages of a particular process for a particular biomass.

So, with this I conclude my today's lecture. So please feel free to post your query in the Swayam portal as well as drop a mail to me at kmohanty@iitg.ac.in. I shall be happy to answer and in the next class that is the third of the module 4 we will be discussing about hybrid methods and the role of pretreatment in a biorefinery concept. Thank you very much.