

Biofuel: A hope for sustainable future

Himanshu Singh, Prabjot Kaur, Moumita Goswami, Animesh Mishra, and Purnima Tiwari

Department of Biotechnology, School of Bioengineering and Biosciences, Lovely Professional University, Phagwara, Punjab.

Abstract:

Large amount of food waste is generated by various industries, during agricultural processes, household waste, etc every year. Fruit waste contain high amount of carbohydrates which is highly recommended for the production of bioethanol. Fruit waste such as banana peel, citrus peel, orange peel, apple peel was taken from the local vendors outside Lovely Professional University. These waste generated contain high amount of lignocellulose that has the potential to produce bioethanol as a renewable form of energy. Our study consists of the size reduction followed by pre-treatment which includes chemical pre-treatment by the utilization of compound such as NaOH, HCL or alkali and utilization of microorganisms such as *Trichoderma resie* and *Sacchromyces Servesie* as well. The amount of glucose content is checked with the help of glucose standard curve. We utilized *S. cerevisiae* for sacchrification, ie. help in production of cellulase enzyme that will break the cellulose into glucose and galactose.

Keywords: Biofuel, Pretreatment, Bioethanol, Lignocellulosic Biomass, Saccharification

Introduction:

Bioethanol is an liquor made by aging. Bioethanol is created by the sugar or from cellulose by the method of aging and can moreover be gotten from the chemical response of ethylene with steam. Basically sugar is utilized as source for generation of ethanol. Source comes from vitality crops. The rule fuel utilized as petroleum interchange is bioethanol. The sources for sugar required to deliver ethanol comes from fuel or vitality crops .The basically utilized crops are too developed for particular vitality utilize these crops are maize, corn, sugarcane, rice and wheat straw crops, squander straw, sorghum plants, sawdust , ruddy canary grass.

Ethanol with the molecular formula $\text{CH}_3\text{CH}_2\text{OH}$ also called ethyl alcohol, also known as $\text{C}_2\text{H}_5\text{OH}$ or $\text{C}_2\text{H}_6\text{O}$. Ethanol can cause alcohol intoxication when consumed. It is best known as the type of alcohol used in alcohol-containing drinks, and is also used as a solvent and as a fuel in thermometers. Commonly, it is commonly used as spirits or alcohol.

The utilization of energy is definitely expanding day by day. Primary source of energy is fossil fuel. Bioethanol, an ethanol fluid that's known to be a clean fuel for combustion engines, may be a promptly accessible substitute since it can be inferred from plant based materials. The full utilization of bioethanol was more than 65,000 million liters and the it is developing rapidly [Kim, Taylor at.al 2008].The per capita squander era in India is around 0.37 kg/person/day.[RK Annepu. Sustainable Solid Waste Management in India, 2012.]. Lignocellulose biomass is conveyed by its carbohydrate substance for generation ethanol. The biomass is composed of cellulose (40–50%), hemicellulose (25–35%) and lignin (15–20%)[Kay, Zaho et, al.2006]. Citrus peel waste(CPW) which incorporates orange, lemon, kinnow etc. can moreover be use for the generation of bioethanol because it comprises of tall sum of lignin, cellulose, hemicellulose, pectin which is required for the generation [Siles, Thompson et, al.2008]. Cellulose is a structural polymer of glucose joined together by β -1, 4 glyosidic linkages. Hemicellulose can be hydrolysed effortlessly by acidic treatment or enzymatic activity.Hemicellulose can be hydrolysed effortlessly by acidic treatment or enzymatic movement.

CPW requires extra preparing some time recently bioethanol generation. Usually since in spite of the fact that CPW is wealthy in different dissolvable and insoluble sugars, making it an perfect feedstock, it too contains a solid microbial inhibitor alluded to as D-limonene. The generation of D-limonene from citrus peel is financially reasonable, as this by-product has tall included esteem as a enhancing operator and for different applications within the chemical industry. In this way, evacuating and recouping D-limonene earlier to the yeast maturation prepare serves two purposes: high-value utilization and upgraded aging of CPW-derived sugars. The productive expulsion of D-limonene from CPW requires a pre-treatment step. Most pre-treatment strategies are based on thermochemical or thermophysical forms such as

processing or steam explosion [Boluda-Aguilar M, López-Gómez 2013]. Substitution of gasoline by liquid fills made from renewable sources may be a high-priority objective in various countries around the world. It is driven by the focuses of a secure and viable essentialness supply and a need to diminish the nursery impact.

The transportation fragment inside the European Union (EU) is completely subordinate on imported fossil fills, and in this way enormously powerless to advertise unsettling influences. It to boot the division tried and true for the foremost parcel of the increment in CO₂ surges. The utilize of biofuels inside the EU is energized by a Mandate that set a target of 2% substitution of gasoline and diesel with biofuels in 2005 on an essentialness preface, which got to have extended to 5.75% by 2010. Bioethanol is expected to be one of the overpowering renewable biofuels within the transportation division interior the coming 20 a long time, and has as of presently been presented on a colossal scale in Brazil, the USA and a couple of European nations. The central focuses of bioethanol are that it can be made from a collection of crude materials, it is non-toxic and is successfully displayed into the existing establishment. In any case, about all bioethanol these days is made from sugar or starch-based agrarian crops, utilizing so-called first-generation progresses. In show disdain toward of the reality that this ethanol is made at a competitive brought, the unrefined fabric supply will not be satisfactory to meet the growing ask for fuel ethanol, and as well the diminishment of nursery gasses coming almost from the utilize of sugar or starch-based ethanol isn't as tall as desirable.

One of the preeminent promising choices to meet this challenge is the generation of bioethanol from lignocellulose feedstocks, such as agrarian buildups (e.g. wheat straw, sugar cane bagasse, corn) and timberland build-ups (e.g. sawdust, decreasing rests), as well as from committed crops (Salix, switch grass) utilizing second-generation developments. These rough materials are adequately plenteous and make especially net nursery gas radiations, decreasing normal impacts. Be that as it may, to compete with gasoline the era gotten must be impressively brought down. These days, rough texture and protein era are two of the most supporters to the for the most part era taken a toll. Profitable utilize of the total alter is required, i.e. tall in common resin of ethanol conveyed by hydrolysis and maturing of the carbohydrate division (hemicellulose and cellulose), as well as a tall abandon of the foremost co-product (lignin). The least expensive and effectively accessible crude fabric for the generation of bioethanol is fruit waste. It could be a potential source, from which ethanol can be delivered. Fruit waste which is disposed of has incredible antimicrobial and cell support potential. In this project, looking at the ethanol effectiveness delivered by maturation process from blend source of differing natural products, apples, papaya, grapes, and bananas, we consider the impact of different parameters like; pH, temperature, particular gravity and concentration [M.s. et. al, 2009]. The yield of ethanol included up to more vital than 80% of the fermentable sugar expended. *Saccharomyces cerevisiae* is utilized as a portion of the maturation prepare since it changes over sugar with oxygen to grant carbon dioxide.

As demonstrated by the Worldwide Energy Organization, cellulosic ethanol may allow ethanol powers to accept a much more noteworthy portion afterward on than already suspected [Inderwildi et, al. 2009]. Ethanol an imperative biofuel, having tall calorific esteem has the included advantage of being less contaminating than most sources of energy that are in utilize nowadays. Reports accessible propose that past characteristic substrates for ethanol generation through saccharification have included sugarcane bagasse, wheat straw, corn, softwood etc [Galbe et. al, 2012]. Fruit rinds are great sources of cellulose which can be utilized for the generation of ethanol by means of saccharification taken after by fermentation

Organic matter counting natural product skins may be a major portion of wastes created every day by families, agrarian segment and nourishment handling businesses. Natural products are utilized on a little and huge scale for family utilization and by nourishment preparing businesses like mash and stick producers. In urban zones, a significant portion of solid waste incorporates natural product squander created by natural product juice merchants and eateries. These businesses and foundations more often than not dispose of the unpalatable parts of the natural products which incorporate the exocarp commonly alluded to as 'rind' or 'peel'. In most cases, these squander materials are dumped in landfills which lead unhygienic conditions. Be that as it may, utilization of these squander materials in generation of bio-fuels would be of incredible natural and financial advantage because it may diminish the burden on conventional sources of vitality conjointly get freed of the wastes.

Leftover biomass incorporate bagasse sugar cane, corn straw and fiber, wheat and rice straw, eucalyptus wood and edit squanders from commercial development of natural products such as bananas, grapes and apples [Rivas-Cantu et al.,

2013]. Innovations for the transformation of biomass to ethanol are moreover beneath different stages of advancement. The utilize of these lignocellulosic buildups requires a few partition of cellulose and hemicellulose from lignin, taken after by hydrolysis of sugars, and this bioconversion has been broadly considered utilizing the distinctive sorts of squanders. The potential yield of ethanol from lignocellulosics changes altogether between feedstocks, so numerous applications in alcoholic maturation are detailed within the writing with different squanders. Particularly within the case of ethanol from bananas, the few considers that have been distributed include the utilize of the natural product, clears out and other squander such as the pseudostem. [Tewari et al. 1986] reported the reasonableness of banana peel for liquor aging. Hammond et al. (1996) displayed ethanol abdicate (on a dry weight premise) from ready bananas as higher than from most other agrarian commodities. [Velásquez-Arredondo et al. 2010] explored the corrosive hydrolysis of banana mash and fruit and the enzymatic hydrolysis of flower stalk and banana skin, and the comes about gotten illustrated a positive vitality adjust for the four generation courses assessed. The study by Graefe et al. (2011) presents comes about of a case consider in Costa Rica and Ecuador which found that significant sums of ethanol may be created from banana bunches that don't meet quality standards, as well as from which are incompletely cleared out to decay within the areas. [Oberoi et al. 2011] moreover illustrated that banana peel might serve as an perfect substrate for the generation of ethanol through concurrent saccharification and aging. [Hossain et al. 2011] assessed bioethanol from spoiled banana and concluded that this will be utilized in engine vehicle motors, creating moo outflows, and hence it can be utilized as an natural reusing handle for squander administration. [Arumugam and Manikandan 2011] investigate the potential application of mash and banana peel squanders in bioethanol generation utilizing weaken corrosive pretreatment taken after by enzymatic hydrolysis. [Gonçalves Filho et al. 2013] evaluate the same techniques with banana tree pseudostem.

Biofuel can be classified into three generations:

First generation biofuel:

To begin with era biofuel feedstocks begin from cultivating grain and sugar crops that are also source of human (and creature) food. The biofuel made by aging of sugar for example- sugarcane (Macedo et al., 2008), whey and molasses are routinely known as to begin with first generation biofuel. Sugar surrender require fair a preparing strategy for the extraction of sugars to aging (not requiring any movement of hydrolysis), turning into a by and large fundamental strategy of sugar alter into ethanol (Icoz et al., 2009). In this method, ethanol can be matured straightforwardly from cane juice or beet juice or from molasses by and huge got as a result after the extraction of sugar. In shapes that utilization starch from grains like corn, saccharification is fundamental some time recently maturation. In this movement, starch is gelatinized by cooking and submitted to enzymatic hydrolysis to glucose monomers, which can be aged by microorganisms.

Second generation:

Second generation biofuel alludes to fuel liquor conveyed from non-nourishment biomass source for case- lignocellulose. Lignocellulosic biomass as wood and horticulture stores is for all bury and purposes boundless, since their generation depends on the photosynthetic method which is almost 60% of the total biomass conveyed (Kuhad et al., 1997). Second generation biofuels decrease net carbon outflow, increment vitality efficiency and diminish vitality reliance, overcoming the impediment of clench hand era biofuels (Silva et al., 2010). Polysaccharides show in lignocellulosic materials counting cellulose and hemicellulose are of awesome intrigued as feedstocks for moment era ethanol production.

Third generation:

Third-generation biofuels are created from algal biomass, which incorporates a exceptionally unmistakable development abdicate as compared with classical lignocellulosic biomass. Third era biofuels inferred from microalgae are considered to be a reasonable elective vitality asset that's void of the major disadvantages related with firstand moment era biofuels such as microalgae are able of all year circular generation, they can be developed in brackish water on non-arable arrive and so may not bring about land-use alter, limiting related natural impacts (Searchinger et al., 2008).

Lignocellulose bioethanol

Ethanol is gasoline fortified with bio-based oxygen. The main requirement for the development of bioethanol as a biofuel is the development of lignocellulose bio mass which serves as low cost and is enriched with biomass from carbon and energy feed. Lignocellulose means drying plant matter, so that there is an excess of raw material on earth. Lignocellulose composed of two polymers of carbohydrate protein-cellulose and hemicellulose. Carbohydrate polymer consists of various sugar monomers and tightly bound to lignin..(Saha *et al.* 2005). There are numerous and many advanced

technologies for the bioconversion of lignocellulose into biofuel, and the choice of technologies is based on economic and environmental factors (Carroll *et al.*, 2009).

Lignocellulose is very recalcitrant, so to simplify recalcitrant structures pretreatment is done. Hydrolysis is done to hydrolyze the sugars into polysaccharides, such as cellulose and hemicellulose. Fermentation is done to convert fermentable sugar to ethanol (Huang *et al.*, 2011).

Lignocellulose compounds are taken for agribusiness squander, ranger service, natural product and family squanders. Hydrolysis of cellulose causes generation of glucose and hemicellulose causes generation of both hexose and pentose sugar. Amid the method of hydrolysis production of acidic corrosive is additionally there that can moreover be calculate for restraint in ethanol maturation handle. Lignin is generally the by-product. Contrast between the handling steps of starch and lignocellulose feedstock but hydrolysis stages of lignocellulose is more complicated (Chandel *et al.*, 2007).

Lignocellulosic biomass (LCB) is the foremost luxuriously open bioresource. The hydrolysis of LCB brings around the discharge of distinctive diminishing sugars which are exceedingly esteemed within the era of biofuels for illustration- bioethanol and biogas, distinctive normal acids, phenols, and aldehydes. Most of LCB is made out of natural polymers, for illustration, cellulose, hemicellulose, and lignin, which are emphatically associated with one another by covalent and hydrogen bonds in this way forming a significantly adamant structure (Chen *et al.*, 2017). The nearness of lignin renders the bio-polymeric structure exceedingly safe to solubilization in this way quelling the hydrolysis of cellulose and hemicellulose which shows a critical test for the isolation of the person bio-polymeric parts (Behera *et al.*, 2014).

Structure of Lignocellulosic biomass and its components:

LCB is for the foremost portion made out of three polymers: cellulose, hemicellulose and lignin along side minor measures of diverse compounds for case- proteins, flotsam and jetsam, and gelatin (Akhtar *et al.*, 2016). In common the cellulose, hemicellulose and lignin substance in a normal LCB drop interior the scope of 30–60, 20–40 and 15–25%, separately (Dahadha *et al.*, 2017).

Cellulose could be a guideline basic and fundamental portion of LCB which could be a straight polysaccharide and comprises of D-glucose subunits associated by β -(1,4)- glycosidic securities (Kumar *et al.*, 2017). This polymer is insoluble in water unless at amazingly moo or tall pH levels.

Hemicellulose could be a moment critical section of LCB that comprises of brief chains of different polysaccharides for illustration- xylan, galactomannan, glucomannan and xyloglucan that are held together by β -(1,4)- and also β -(1,3)- glycosidic bonds (Zhou S. *et al.*, 2017).

Lignin is commonly the foremost complex and little division, talking to approximately 10% to 25% of the biomass. It features a long-chain, sweet-smelling polymer made to a incredible degree out of phenyl propane units. Lignin acts like a glue by filling the gap between and around the cellulose and hemicellulose composition with the polymers (Chen *et al.*, 2011).

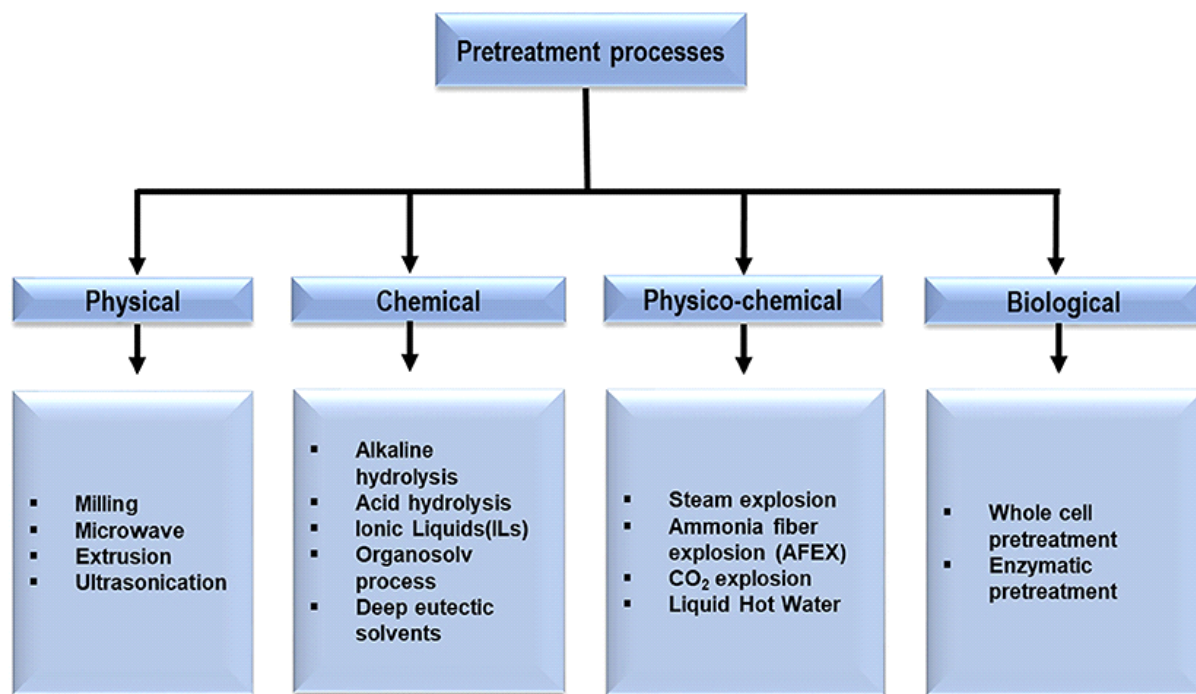


Figure 1. Pretreatment technique used in lignocellulosic biomass (Luo et al., 2014).

Physical Pretreatment

It is basically carried out to decrease the atom estimate that results within the increment in surface zone, and reducing in level of polymerization and crystallinity (Rajendran et al., 2017). The frequently common physical pretreatment strategies incorporates processing, expulsion, microwave treatment and ultrasonication.

Diverse strategies of pretreatment are physical, physico-chemical, chemical, organic prepare utilized agreeing to the taken a toll compelling. The most reason of pretreat is to clear absent lignin and hemicellulose, subside cellulose crystallinity and thus the porosity of the feedstock wheat straw. Pretreatment is pointed for no arrangement of byproducts those act as inhibitor to assist handle that's hydrolysis, no misfortune of carbohydrates, and more arrangement of sugar (sun et al., 2010).

• **Milling**

Processing is utilized to diminish the crystallinity and atom estimate of LCB. Processing can diminish the particle estimate upto 0.2 mm. The diminish in atom estimate and crystallinity is decided by the sort of processing technique gotten, planning time and moreover the kind of biomass utilized (Kumar et al., 2017). Preparing pretreatment of unrefined LCB for solid state anaerobic digestion prepare illustrated that diminish in particle measure essentially grows the substrate dissolvability in this way growing the reaction vitality (Motte et al., 2014). Processing pretreatment doesn't bring around any poisonous or inhibitory compounds it could be a favored preparatory pretreatment strategy for a wide assortment of lignocellulosic feedstocks.

• **Ultrasonification**

Ultrasonication pretreatment depends on the guideline of cavitation through the work of ultrasonic radiation. The cavitation produces shear strengths that isolates the complex organize structure of LCB and advances the extraction of wanted compounds for case cellulose, hemicellulose, and lignin (Ravindran et al., 2016). A few elements influencing the sonication treatment incorporates ultrasound frequency, sonication span, sonication power and temperature (Liyakathali et al., 2016) have discovered that the enzymatic digestibility of energy cane bagasse increases with increase in the sonication time and temperature while ultrasonic frequencies had no impact on enzymatic digestibility.

Chemical Pretreatment

- **Alkali Pretreatment**

Alkali pretreatment may be a considered chemical pretreatment procedure which depends on the solubilization of lignin within the soluble base treatment. The diverse essential reagents utilized for the most part for soluble base pretreatment are the hydroxides of sodium, potassium, calcium and ammonium. This results in solubilisation of lignin and hemicellulose parts within the dissolvable base course of action and gets the cellulose the collaboration of chemicals (Sun S. et al., 2016). Furthermore, dissolvable base pretreatment changes the lignocellulosic structure through cellulose extending that prompts diminish in crystallinity and level of polymerization in this way extending interior surface region (Behera et al., 2014).

- **Acid Pretreatment**

Acid pretreatment of LCBs depends on the feebleness of the glucosidic bonds among hemicellulose and cellulose to corrosive. Hydronium particles which starts from the corrosive catalyst cause breakdown of the long cellulose and hemicellulose chains into sugar monomers (Lloyd et al., 2005).

References:

- 1.) Asli, M.S., "A study on some efficient parameters in batch fermentation of ethanol using *Saccharomyces cerevisiae* extract from fermented siahe sardasht pomace", *African Journal of Biotechnology*, vol.9 (20), pp.2906- 2912, 2010.
- 2.) O. R. Inderwildi, D. A. King (2009). *Quo Vadis Biofuel*. *Energy & Environmental Science* 2: 343.
- 3.) Anderson WF, Akin DE (2008). Structural and chemical properties of grass lignocelluloses related to conversion for biofuels. *J Ind Microbiol Biotechnol*. 35:355–366.
- 4.) Galbe M, Zacchi G (2012). A review of the production of ethanol from softwood. *Appl Microbiol Biotechnol* 59:618–628.
- 5.) Bigelow M, Wyman CE (2002). Cellulase production on bagasse pretreated with hot water. *Appl Biochem Biotechnol*. 98–100:921–934.
- 6.) Omojasola, P Folakemi, Jilani, Omowumi Priscilla, Ibiyemi SA (2008). Cellulase production by some fungi cultured on pineapple waste. *Nature and Science*. 6(2):1545-0740
- 7.) Oberoi, H.S., Vadlani, P.V., Saida, L., Bansal, S., Hughes, J.D., 2011. Ethanol production from banana peels using statistically optimized simultaneous saccharification and fermentation process. *Waste Manage*. 31, 1576–1584.
- 8.) O'Brien, D.J., Senske, G.E., Kurantz, M.J., Craig Jr., J.C., 2004. Ethanol recovery from corn fiber hydrolysate fermentations by pervaporation. *Bioresour. Technol*. 92, 15–19.
- 9.) Tewari, H.K., Marwaha, S.S., Rupal, K., 1986. Ethanol from banana peels. *Agric. Wastes* 16, 135–146
- 10.) Graefe, S., Dufour, D., Giraldo, A., Muñoz, L.A., Mora, P., Solís, H., Garcés, H., Gonzalez, A., 2011. Energy and carbon footprints of ethanol production using banana and cooking banana discard: a case study from Costa Rica and Ecuador. *Biomass Bioenergy* 35, 2640–2649.
- 11.) Hammond, J.B., Egg, R., Diggins, D., Coble, C.G., 1996. Alcohol from bananas. *Bioresour. Technol*. 56, 125–130.
- 12.) Hossain, A.B.M.S., Ahmed, S.A., Ahmed, M.A., Faris, M.A.A., Annuar, M.S.M., Hadeel, M., Norah, H., 2011.
- 12.) Bioethanol fuel production from rotten banana as an environmental waste management and sustainable energy. *Afr. J. Microbiol. Res*. 5, 586–598.
- 13.) Mojovic L, Nikolic S, Rakin M and Vukasinovi c M, Production of bioethanol from corn meal hydrolyzates. *Fuel* 85:1750–1755 (2006).
- 14.) Park, Se-Joon, et al. "A Study on bio-ethanol production from fruit wastes." *Transactions of the Korean hydrogen and new energy society* 20.2 (2009): 142-150.
- 15.) Choi, In Seong, et al. "A low-energy, cost-effective approach to fruit and citrus peel waste processing for bioethanol production." *Applied Energy* 140 (2015): 65-74.