Chapter

Bioethanol Production: An Overview

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Abstract

Bioethanol is currently being considered as a potential replacement for the conventional gasoline, especially as it possesses similar and some superior qualities enabling reduction in GHG and increases fuel reserve. Bioethanol used for commercial purposes is usually produced from edible feedstocks such as corn and sugar cane which increases the production cost. The high cost of these feedstocks is the driving force behind the search for the second, and third generations (3G) bioethanol produced from cheaper and available feedstocks. The fourth-generation bioethanol is being developed to further advance the 3G bioethanol to enhance the potential of algae to capture CO₂ and to increase the production of specific compounds. Despite the efforts been made to reduce the cost of production through the use of diverse non-edible feedstocks, the cost of processing the feedstocks is still very high, thereby making bioethanol uncompetitive with the conventional gasoline. The life cycle assessment and techno-economic analyses are usually conducted to assess the economic feasibility and the environmental impact of the bioethanol production processes. This chapter thus, covers the State-of-the-art processes involved in bioethanol production including pretreatment, hydrolysis, fermentation processes, bioethanol recovery, integrated processes, Life cycle assessment, techno-economic analysis, exergy analysis and process simulation.

Keywords: bioethanol, lignocellulose, hydrolysis, pretreatment, fermentation, distillation, exergy, simulation, techno-economic, life cycle assessment

1. Introduction

The depletion of the fossil fuel and global warming caused by the emission of greenhouse gases from the combustion of fossil is currently driving researchers in the direction of finding alternative and environmentally friendly fuel. Biofuels are one of the numerous options being considered. Bioethanol is considered as the most promising biofuel to replace gasoline, especially due to its properties. This biofuel is a liquid oxygenated fuel containing 35% oxygen produced from the microbial fermentation of monomeric sugar obtained from carbohydrate sources such as corn, soybeans and sugar cane. The bioethanol produced globally in 2018 was 110 billion liters and is expected to increase to 140 billion liters in 2022 with compound annual growth rate (CAGR) of 7.6% due to anticipated economic feasibility of the process [1]. The US, Brazil, European Union, China and Canada respectively are the global powerhouses in bioethanol production. The US uses corn as the feedstock to produce bioethanol and obtained a production capacity of ~57.7 billion liters while
Brazil produces bioethanol from sugar cane and had a total production capacity of \(~27.6\) billion liters in 2016 respectively [2].

Bioethanol is considered a potential substitute for the conventional gasoline and can be used directly in vehicles or blended with the gasoline, thereby reducing greenhouse gas emissions and consumption of gasoline [3]. For direct application (E100), the timing (and electronic control system if in use) of the gasoline engine is adjusted, and larger gasoline tank is used. However, the use of bioethanol (E100) is usually characterized with difficulty in starting the engine at a low temperature or during the cold weather due to higher heat of vaporization. Required. The blending of bioethanol with gasoline might not require modifying the engine, rather it will help to enhance ignition or engine performance. The most commonly used blends are E85 and E10. Advantages of bioethanol include high-octane rating resulting to increased engine efficiency and performance, low boiling point, broad flammability, higher compression ratio and heat of vaporization, comparable energy content, reduced burning time and lean burn engine [4]. The disadvantages include high production cost resulting from high cost of feedstock, enzymes, detoxification and ethanol recovery, respectively. Bioethanol possesses a low volumetric energy density, meaning that more volume of bioethanol/km (up 50%) will be consume compared to the conventional gasoline [3]. The use of bioethanol in engines might require frequent replacing the engine parts as the bioethanol has the capacity to degrade some elastomers and cause corrosion of metals [5]. However, in attempt to reduce the cost of production, lignocellulosic biomass is being considered as feedstocks because of availability and low cost of acquisition. Unfortunately, the processing cost is still high, thereby, making the process unattractive economically [6].

When bioethanol is produced from edible feedstocks such as corn and sugar cane, it is called first generation (1G) bioethanol and 2G second-generation (2G) bioethanol if the feedstock is a lignocellulose. Examples of these lignocellulose biomass is switch grass, cornstalks, wood, herbaceous crops, waste paper and paper products, agricultural and forestry residues, pulp and paper mill waste, municipal solid waste and food industry waste. Lignocellulosic biomass is made up of cellulose, hemicellulose, lignin, protein, ash, and minor extractives [7]. Lignocellulosic biomass is being considered as feedstocks for bioethanol production due to relatively low cost of acquisition, availability and sustainability of supply. This biomass has the capacity to increase the current production rate of bioethanol and is being speculated to produce approximately 442 billion liters per year of bioethanol globally. The 2G-bioethanol has a greater potential to reduce the greenhouse gases emission compared to 1G -bioethanol. The third generation (3G) bioethanol is obtained when algae are used as the feedstock. Algae bioethanol is gaining traction possibly due to high carbohydrate content and absence of lignin in most available algae. With this kind of feedstock, the cost of pretreatment is expected to reduce as the complex

| Algae                        | Bioethanol yield (%) | Ref. |
|------------------------------|----------------------|------|
| Nannochloropsis Oculata      | 3.68                 | [9]  |
| Tetraselmis suecica          | 7.26                 | [9]  |
| Scenedesmus dimorphus        | 49.7                 | [10] |
| Porphyrudium cruentum (seawater) | 65.4                 | [11] |
| Porphyrudium cruentum (fresh water) | 70.3                 | [12] |
| Padina Tetrastomatica        | 16.1                 | [12] |

Table 1. Yield of different species of algae.
lignin removal process is eliminated [8]. Numerous researchers have investigated the use of algae as feedstock for bioethanol production. Based on the results obtained, the species of algae with high productivity are presented in Table 1.

The fourth-generation (4G) bioethanol is obtained from the modification of E. coli gene altercations through the application of metabolic engineering or systems biology strategies [13].

2. Bioethanol production process

The processes involved in the production of bioethanol from different feedstocks include pretreatment, hydrolysis, fermentation and ethanol recovery. These processes are explained below:

2.1 Pretreatment

Pretreatment is one of the costliest steps in the production of bioethanol from lignocellulosic biomass accounting to approximately $0.30/gallon of ethanol produced. There exist different pretreatment methods aimed at increasing the reactivity of cellulose and the potential yield of the fermentable sugars. These may be either traditional or advanced pretreatments. Traditional pretreatments are classified into four categories which include chemical, physical, physicochemical, and biological methods while advanced pretreatment method may be either acid-based fractionation or ionic liquid-based fractionation (ILF) [14]. Amongst the traditional pretreatment methods, chemical categories are the most efficient and hence predominantly used [15].

The pretreatment of lignocellulosic biomass through various methods helps to release cellulose usually embedded in a matrix of polymers consisting of lignin and hemicellulose by disrupting the original structure (Figure 1). With this, cellulose is separated from the polymer matrix and is more accessible for enzymatic hydrolysis, thereby resulting to increased sugar yields greater than 90% (theoretical yield) using feedstocks such as grasses, corn and wood [16]. This means that cellulose is more susceptible to enzymatic hydrolysis when its crystalline structure is disrupted. Without the disruption, enzymes bind on the surface of the lignin and not the cellulose chains impeding enzymatic hydrolysis.

Other advantages of pretreatment include helping to prevent the degradation of sugars (pentoses); ensuring viability of the bioethanol production processes by using moderate size reactors and minimizing heat and power requirements, and minimizing the formation of inhibitors which reduces the yield of the hydrolysis and hence the fermentation of sugar to ethanol [16].

2.1.1 Traditional pretreatments

These pretreatments method have been discussed extensively in the literature. As mentioned earlier, the method is categorized as: (1) physical pretreatment- this involves the breaking down of the size of the lignocellulosic biomass and crystallinity by methods such as milling, grinding, irradiation and extrusion. The resultant effect of which are increased surfaced area and pore size of the biomass enabling increase in the enzymatic hydrolysis efficiency. Physical pretreatment may need combining with chemical pretreatment to enhance the efficiency of deconstruction of the lignocellulose [17]. (2) Chemical pretreatment: these include acid, alkali, oxidative delignification, and organic acid (organosolvation) methods. They are highly selective for specific type of feedstocks, and are used to deconstruct and remove lignin and/or hemicellulose from the polymer matrix. Chemical pretreatments
are undoubtedly effective but require harsh operating conditions which may have adverse effect on the downstream processing and the by-products may need special disposal procedures [17]. (3) Physicochemical: this combines the features of both physical and chemical pretreatments. Examples are steam explosion, liquid hot water, microwave irradiation and CO\textsubscript{2} explosion [18]. (4) Biological pretreatment: This involves the use of microorganisms to breakdown lignocellulosic biomass for further enzymatic hydrolysis. These organisms include white-rot, brown-rot and soft-rot fungi, and bacteria [19].

2.1.2 Advanced pretreatment methods for lignocellulose

These methods are also called lignocellulose fractionation pretreatment and are targeted at reducing the cost of cellulosic ethanol production by fractionating the lignocellulose in such a way to generate value-added co-products under a mild operating condition like 50\textdegree{}C and atmospheric pressure [20]. This gain is achieved by using cellulose solvents which enhances the cellulose accessibility and separation of cellulose, hemicellulose, and lignin to produce value-added co-products [21]. The method is also known as Cellulose solvent-based lignocellulose fractionation (CSLF). The operation helps to reduce the quantities of enzymes required for the subsequent enzymatic hydrolysis and could be used for varieties of feedstocks [21].

There are two general techniques used in CSLF which include (1) acid-mediated fractionation and (2). Ionic liquid-based fractionation (ILF). These are discussed below:
2.1.2.1 Acid-mediated fractionation

The cellulose solvents such as phosphoric acid and organic solvents like acetone or ethanol are usually used at mild operating conditions of 1 atm and 50°C to separate lignocellulosic biomass. The effectiveness of the separation is dependent on the solubility properties of the cellulose, hemicellulose, and lignin in the cellulose solvent, organic solvent and water, respectively [20]. Separating lignin and hemicellulose from the cellulose fraction helps to reduce substrate recalcitrant and competitive binding sites, unwanted sugar degradation, cost and production of the inhibitors [20]. This method has been used efficiently to pretreat varieties of lignocellulose such as bamboo, corn stover, sugarcane, switchgrass and elephant grass [22].

2.1.2.2 Ionic liquid-based fractionation

Ionic liquids (ILs) are salt solutions consisting of significant quantity of organic cations and small/inorganic anions that exists as liquid at relatively low temperatures like room temperature. They are used to fractionate lignocellulose to obtain specific, purified and polymeric raw materials which are intact and are easily separated and used as value-added co-products. In comparison of the conventional lignocellulosic biomass, ILs pretreatment methods show some advantages such as less energy intensive, simplicity of operation and capacity to separate specific components [20]. The properties such as low vapor pressure and high thermal stability suggest that ILs are environmentally friendly and as such are considered as green solvents. ILs are also considered to be tunable due to such properties as hydrophobicity, polarity, and solvent power which can be adjusted to achieve specific desirable results. These properties of ILs with those of antisolvent and lignocellulose (type, moisture content, partial size, and load) with temperature, pretreatment time can be used to determine the overall efficiency of the ionic liquid pretreatment method [23].

However, the most frequently used pretreatment method is steam explosion. This patronage could be due to its low capital investment, high energy efficiency, less environmental impact, less hazardous process chemicals and conditions, and complete sugar recovery [24].

2.2 Hydrolysis

Following the pretreatment of the lignocellulosic biomass is the hydrolysis of polymeric carbohydrate (cellulose and hemicellulose) to produce sugar monomers. This stage is required since enzymes needed in the succeeding stage (fermentation) can only digest sugar monomers. The process can be catalyzed either by acid or enzymes. Acid-catalyzed hydrolysis is the most commonly used method and it involves either the use of concentrated or dilute acid (see Eq. (1)). Example of such acids are H$_2$SO$_4$ and HCl. The concentrated acid-catalyzed hydrolysis is used at lower temperature and high acid concentration, resulting to 90% sugar recovery at a short period of time [25]. The disadvantage of this method is the high cost of production due to difficulty in acid recovery, disposal, concentration control and recycling [26]. Another problem with the concentrated acid-catalyzed hydrolysis treatment is its capability to degrade sugar monomers due to the prevailing acidic environment. The dilute acid-catalyzed hydrolysis requires high temperature and low acid concentration. The most predominantly used acid is dilute acid. The
problem with this method of hydrolysis is that the process results to the formation of inhibitors compared to the acid-catalyzed hydrolysis.

\[
(G_6H_{10}O_5)_{n} + H_3O^+ \rightarrow H_2O + nC_6H_{12}O_6
\]  

(1)

Acid hydrolysis of the lignocellulose is carried out in two stages. Stage one is where the hemicellulose is hydrolyzed with the help of dilute acid and in the second stage, cellulose is hydrolyzed using concentrated acid [25].

Enzyme-catalyzed hydrolysis uses enzymes to hydrolyze polymeric carbohydrate to sugar monomers under mild operating conditions of temperature 45–50°C and pH 4.8–5.0. This method is efficient and results to high sugar recovery without inhibitor formation and tendency to cause corrosion. The efficacy of the enzyme-catalyzed hydrolysis is affected by factors such as pH, enzyme loading, time, temperature and substrate concentration. The hydrolytic process can be catalyzed by three kinds of cellulase enzymes, name endo-1,4-β-glucanases, cellobiohydrolases and β-glucosidases. These enzymes are usually very expensive due to high demand from various industries such as paper, textile and food processing industries [1]. The high cost of these enzymes also impacts on the overall cost of production especially as large quantities of enzymes are required. Based on the cost, microorganisms with the potential of secreting cellulosytic enzymes are broadly used in the contemporary times. These include Clostridium, cellulomonas, Erwinia, Thermoospora, Bacteroides, Bacillus, Ruminococcus, Acetivibrio, and Streptomyces. Others include fungi such as Trichoderma, Penicillium, Fusarum, Phanerochaete, Humicola, and Schizophillum sp. The most commonly used microbial enzymes amongst these microorganisms is Trichoderma species [27]. The problems with the microbial enzymes are stability, substrate or product inhibition and catalytic efficiency. Although, with advances in genetic modifications, recombinant DNA techniques and application of various strategies to improve the strains help to increase the quantity of enzymes produced, make them more robust and economically feasible. The efficiency of the cellulose hydrolysis can also be improved by the addition of Polyethylene glycol (PEG) or Tween 20 resulting to increased enzymatic saccharification and reduction in the adsorption of cellulose on lignin [25].

The mechanism of the hydrolysis of lignocellulosic biomass to glucose occurs in three steps and are presented in Figures 2–6. The first step is the linking of the β-1,4 bond of the cellulose with water molecule catalyzed by endoglucanase (1,4-β-D-glucanohydrolase) resulting to the formation of celloextrin with a shorter chain, and free-chains ends (reducing and non-reducing ends) (Figure 2) [28]. The second step is the degrading of celloextrin to a two-unit glucoses (cellobiose) with the help of exoglucanase (1,4-β-D-glucan cellobiohydrolase) by adjusting the reducing and non-reducing chains (Figure 3) [29]. The third step is the formation of glucose obtained when the β-glucosidases strikes the cellobiose (Figure 3) [30]. The production of glucose is necessary because, the subsequent process which is fermentation requires the use of the simplest monomer as feedstock.

The hydrolysis of hemicellulose is easier compared to cellulose due to its possession of more amorphous property. The hemicellulose contains 10–15% and 10–35% of xylan in soft and hard woods, respectively. Xylan has both main and outer chains. The former can be degraded using endo-β-1,4-xylanase (EC 3.2.1.8) and β-xylosidase (EC 3.2.1.37). The main chain of xylan is hydrolyzed to a short chain xylan oligosaccharide through the help of endo-β-1,4-xylanase (Figure 5). The oligosaccharide is further degraded to a pyranose form of xylan known as xyropyranose by β-xylosidase (Figure 6) [32]. On the contrary, the outer chains of the xylan
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can be degraded by enzymes known as accessory xylanolytic enzymes such as feruloyl esterase (EC 3.1.1.73), α-L-arabinofuranosidase (EC 3.2.1.55), α-glucuronidase (EC 3.2.1.139), and acetylxylan esterase (EC 3.1.1.72).

Figure 2.
Hydrolysis of long chain cellulose to a shorter chain cellulose (cellodextrin) [28].

Figure 3.
Hydrolysis of cellodextrin to cellobiose catalyzed by exoglucanase (1,4-β-D-glucan cellobiohydrolase) [29].
2.3 Fermentation processes

This is a biological process that involves the conversion of the monomeric units of sugars obtained from the hydrolysis step into ethanol, acids and gases using microorganisms such as yeast, fungi or bacteria (see Eq. (2)) [1, 33]. The most commonly used microorganism is yeast especially \textit{Saccharomyces cerevisiae} due to high yield of ethanol and high tolerance limits [34]. \textit{Saccharomyces cerevisiae} converts glucose,

\[
C_6H_{12}O_6 + \text{yeast} \rightarrow 2C_2H_5OH + 2CO_2
\]  

mannose or fructose which can be obtained from the hydrolysis of cellulose to ethanol while xylan from the hydrolysis of hemicellulose can be converted to xylose. Some examples of different microorganisms used in fermentation of simple sugars and their respective ethanol yields at varying operating conditions are presented in Table 2. As can be seen, \textit{Saccharomyces Cerevisiae 3013} followed by \textit{Zymomonas Mobilis ZMA7-2} gave the maximum ethanol yield [33].

2.3.1 Fermentation technologies

The technologies used for the fermentation of monomeric units of sugar to ethanol include separate hydrolysis and fermentation, simultaneous
saccharification and fermentation (SSF), simultaneous saccharification and co-fermentation (SSCF), non-isothermal simultaneous saccharification and fermentation, simultaneous saccharification, filtration and fermentation, consolidated bioprocessing (CBP). The first three are commonly used [26]. Other types of fermentation include batch, fed-batch, continuous and solid-state fermentation. Some of these fermentation methods are discussed below.

2.3.1.1 Batch fermentation process

This is the simplest of the fermentation processes as it is flexible for a range of products, easy to control and has multi-vessel. The process involves adding the substrates, microorganism, culture medium and nutrients at the beginning of the operation in a closed system under favourable conditions at a predetermined time. The products are only withdrawn at the end of the fermentation time. The problems with this type of fermentation process are low yield, long fermentation time, and high labour cost making batch process unattractive for commercial production of bioethanol [26, 40]. Also, due to high sugar concentration in the fermentation medium, there could be substrate inhibition leading to inhibition of cell growth and ethanol production [41].

2.3.1.2 Continuous fermentation process

This process involves adding substrates, culture medium and nutrients into a fermentor containing active microorganisms and withdrawing the products continuously. The products obtained are usually ethanol, cells and residual sugar. The advantages of continuous fermentation process are high productivity, small fermenter volumes, and low investment and operational cost [42]. The disadvantages include possibility of product contamination, and potential decline in yeast capability to support ethanol production because of long cultivation time [43].
2.3.1.3 Fed-batch fermentation process

This is the combination of batch and continuous fermentation processes involving charging the substrate into the fermentor without removing the medium. Comparing with other fermentation processes, fed-batch process has higher productivity, more dissolved oxygen in medium, shorter fermentation time and lower toxic effect of the medium [43]. The disadvantage is that ethanol productivity is limited by cell mass concentration and feed rate [40].

2.3.1.4 Separate hydrolysis and fermentation (SHF)

The Enzymatic hydrolysis is separated from fermentation allowing enzymes to operate at high temperature and the fermentation microorganisms to function at moderate temperature for optimum performance [26]. Since the hydrolytic enzymes and the fermentation organisms operate at their optimum conditions, it is expected that the productivity of ethanol will be high. The disadvantages of SHF are high capital cost especially as two reactors are required, requirement of high reaction time, and possibility of limiting the cellulase activities by sugars released during the hydrolysis step [44].

2.3.1.5 Simultaneous saccharification and fermentation (SSF)

Here the saccharification of cellulose and the fermentation of monomeric sugars are carried out in the same reactor simultaneously [45]. Since the hydrolysate is simultaneously used for fermentation, the usual inhibition of the cellulase activities

| Microorganism | Temperature (°C) | pH | Fermentation time (h) | Sugar concentration (g/L) | Ethanol yield (g/L) | Ref. |
|---------------|-----------------|----|-----------------------|--------------------------|-------------------|-----|
| Yeasts        |                 |    |                       |                          |                   |     |
| Saccharomyces Cerevisiae 3013 | 30 | 5.5 | 65 | 280 | 130.12 | [35] |
| Saccharomyces Cerevisiae BY4742 | 35 | 5.0 | 96 | 80 | 39 | [36] |
| Bacteria      |                 |    |                       |                          |                   |     |
| Zymomonas Mobilis NRRL 806 | 30 | 6.5 | 18 | 117 | 30.4 | [37] |
| Zymomonas Mobilis ZMA7-2 | 30 | 4.0 | 44 | 200 | 99.78 | [38] |
| Fungi         |                 |    |                       |                          |                   |     |
| Aspergillus oryzae 694 | First aerobic step (30) | 5.0 | 24 | 50 | 24.4 | [39] |
| Second anaerobic step (30) | 5.0 | 144 | | | | |
| Rhizobium japonicus 2871 | First aerobic step (30) | 5.0 | 24 | 100 | 33 | [39] |
| Second anaerobic step (30) | 5.0 | 72 | | | | |

Table 2. Effects of microorganisms on the yield of ethanol under varying operating conditions [33].
can be avoided [46]. The disadvantage of SSF is the variation in the optimum temperature required for efficient performance of the cellulase and microorganisms during hydrolysis and fermentation, respectively. The high temperature required by the cellulase for hydrolysis might reduce the microorganisms such as yeast used for fermentation.

2.3.1.6 Simultaneous saccharification and co-fermentation (SSCF)

This involves carrying out the hydrolysis and saccharification in the same unit with co-fermentation of pentose sugars. Usually, genetically modified *Saccharomyces cerevisiae* strains that can ferment xylose are used since normal *Saccharomyces cerevisiae* cannot ferment pentose sugar [47]. Like SSF, SSCF has the advantages of lower cost, higher ethanol yield and shorter processing time [43]. In addition, SSCF helps to minimize the inhibition caused by sugars during the enzymatic hydrolytic process and increases xylose to glucose concentration ratio as most of the microorganisms consume xylose.

2.3.1.7 Consolidated bioprocessing (CBP)

This requires the enzyme production, hydrolysis and fermentation to be carried out in a single unit. The microorganism mostly used in this process is *Clostridium thermocellum* as it has the capacity to synthesize cellulase which degrades lignocellulose to monomeric sugars and produce ethanol [48]. Although, CBP is still at its nascent stage, the following advantages have been identified: less energy intensive, cheaper cost of enzyme, low cost of investment, less possibility of contamination.

2.3.2 Factors affecting bioethanol production

The factors which impact the bioethanol production include temperature, sugar concentration, pH, fermentation time, agitation rate and inoculum size [49]. High temperature could denature the enzymes and reduce their activity. The ideal temperature for the fermentation of biomass is 20–35°C [50]. The optimum yield of bioethanol production could be achieved using a concentration of 150 g/L [49]. The pH of the broth also affects the production of bioethanol because, it impacts on the bacterial contamination, yeast growth, fermentation rate and by-product formation. The optimum range of pH for the fermentation of the biomass using *Saccharomyces cerevisiae* is 4.0–5.0. When the pH is less than 4.0, a longer incubation period is required and at a pH above 5.0, ethanol concentration is significantly reduced. To optimize the yield of bioethanol, another factor to be considered is the agitation rate. The higher the agitation rate, the higher the quantity of ethanol produced. For fermentation using yeast cells, the commonly used agitation rate is 150–200 rpm. Excess agitation rate may limit the metabolic activities of the cells [49].

2.3.3 Integrated processes (IP)

This involves combining one or more processes in the bioethanol production processes from the lignocellulosic biomass for the purpose of optimization, resulting to the increase in yield and minimum production cost [33]. An example of IP is membrane reactor where both reaction and separation of products occur simultaneously [33]. The hydrolysis and fermentation processes can be integrated into separate hydrolysis and fermentation (SHF) and simultaneous saccharification and fermentation (SSF). As discussed earlier, SHF provides an opportunity for the
temperatures of the cellulases and *Saccharomyces cerevisiae* to be controlled separately for the efficient operation of each process. Operating the SHF at the optimum temperatures of 45–55°C for cellulase and less 32°C *Saccharomyces cerevisiae* provides favourable conditions for the pentose and hexose sugars to be fermented in a single-step process giving rise to a method known as separate hydrolysis and co-fermentation (SHCF), see Figure 7 [52]. As mentioned earlier, one of the disadvantages of the enzymatic hydrolysis is the inhibition of the cellulase caused by high concentration of glucose produced. This challenge can be solved by increasing the concentration of the enzyme or by using SSF [52]. SSF allows glucose obtained from the enzymatic hydrolysis to be converted directly to ethanol through fermentation in the same reactor. Some investigators have argued that SSF process is rather sequential and not simultaneous. Thus, saccharification coupled with co-fermentation (SCCF) is used (see Figure 8) [50].

The SSF has been developed further to a technology known as consolidated bioprocessing (CBP) by integrating enzyme production into the operation (Figure 9). As mentioned earlier on, enzyme production, hydrolysis and fermentation are conducted in a single unit [48].

### 2.4 Ethanol recovery

The fermentation of monomeric sugars is usually followed by ethanol recovery from the fermentation broth. Usually, the water content of the broth is reduced to approximately 0.5% by volume enabling the formation of anhydrous ethanol with a minimum of 99.5% by volume. This operation is constrained by the azeotropic nature of ethanol-water solution and can be carried out based on the principle of distillation (i.e. leveraging the difference in boiling point of the components of the solution). The problem with the azeotropic solution is overcome by using a separating agent which alters the relative volatility of the key component. The techniques used in the recovery of pure ethanol from the fermentation broth include adsorption distillation, azeotropic distillation, diffusion distillation, extractive distillation, vacuum distillation, membrane distillation and chemical dehydration. The conventional techniques include azeotropic distillation, liquid-liquid extraction and extractive distillation [53]. Extractive distillation is the most predominantly used for large scale operations. There are some other techniques that are gaining traction for future use especially due to less energy requirement. These are pervaporation and salt distillation [54].

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**Figure 7.** Separate hydrolysis and co-fermentation (SHCF) [51].
3. Life cycle assessment (LCA)

This assessment is usually carried out to measure the environmental impact of bioethanol production using different feedstocks. The LCA tool helps for the identification of potential impacts during a process design and for decision making in order to improve the process prior to scaling-up [55]. The LCA methodology consists of four main stages including definition of goal and scope, Life Cycle Inventory Analysis (LCIA), Impact assessment, and Interpretation of the results [1]. The LCIA can be conducted using methodologies such as CML 2002, Eco-indicator 99, ReCiPe, LIME, Lucas and TRACI depending on the impact categories and selection of indicators [56]. Numerous investigations have been conducted on the environmental impact of bioethanol and allied chemical products from different lignocellulosic feedstock (Table 3). The table indicates that bioethanol has the capacity to reduce the greenhouse gases emission and global warming potential substantially and hence facilitates the protection of the ozone layer.

4. Techno-economic analysis (TEA)

TEA is an effective tool used in assessing the economic feasibility of different processes pertaining to bioethanol production. This analysis provides the opportunity to evaluate the technical and economic efficiencies of different process routes leading to bioethanol production with an overarching objective of choosing the best route(s) [60]. The technical aspect of the analysis involves the development of the process flow diagram, and rigorous material and energy balance calculations using simulation software such as Aspen Plus and SuperPro. The economic aspect involves the capital and project cost estimation, discounted cash flow and determination of the minimum ethanol selling price (MESP). This may be carried out using the Aspen Economic Evaluator package (Aspen Technology, Inc., USA). The MESP can be used for comparing the differences in technology between processes.
| Feedstock                          | Method of production                                                                 | Environmental analysis                                                                 | Ref.                                                                                                                                 |
|-----------------------------------|--------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------|
| Cattle manure (CM)                | Drying, milling, pretreatment, solid phase separation, Separated Hydrolysis and Fermentation (SHF), and distillation | SimaPro software v7.3.2 was used with ReCiPe method and EcoInvent libraries             | 1. Results from midpoint indicators with normalized data showed that the main impacts were on human toxicity, freshwater eutrophication, terrestrial and marine ecotoxicity and fossil depletion |
|                                   |                                                                                      |                                                                                        | 2. Endpoint indicators showed that the main impacts were climate change, human toxicity, particulate matter formation and fossil resource depletion |
| Wheat straw                       | Steam pretreatment, hydrolysis, Fermentation, distillation, enzyme recycling, C5 sugars drying, and lignin pelletizing | Simplified LCA approach according to European Renewable Energy Directive (RED)        | Up to 87% GHG potential mitigation                                                                                                                                                      |
| Sweet potato                      | Cultivation, and conversion of sweet potato to bioethanol                            | EcoInvent 3.1database, literature and field data. SimaPro software was used for the impact assessment with CML IA baseline 3.02 method | Reduction of global warming potential (GWP) of 44%                                                                                                                                         |
| Loblolly pine, eucalyptus, unmanaged hardwoods, forest residues, and switchgrass | Thermochemical conversion                                                              | SimaPro 7.3 was used with the US Life Cycle Inventory dataset. The Tool for the Reduction and Assessment of Chemical and Other Environmental Impacts (TRACI) impact assessment method was used to calculate the life cycle environmental and human health midpoint impacts | Reduction in the GHG emissions by more than 60% compared to gasoline.                                                                                                                     |

Table 3. LCA of bioethanol production from different feedstock.
or for carrying out sensitivity analyses which helps to determine where economic or process performance improvement is required. Numerous investigators have studied the techno-economic analysis of bioethanol production using lignocellulose as feedstocks. For instance, Quintero et al. conducted a techno-economic analysis of bioethanol production from sugar cane bagasse, coffee cut-stems, rice husk, and empty fruit bunches for the Colombian case [61]. These researchers used Aspen Plus and Aspen Process Economic Analyzer for the process simulation and economic analysis, respectively. The results obtained showed that considering the four lignocellulosic biomasses assessed, the production cost of bioethanol from the empty fruit bunches was the lowest (0.49 US$/L).

5. Exergy analysis

Exergy is the maximum amount of work that can be obtained when a mass or energy stream is brought to equilibrium with a reference environment. Exergy analysis helps in: identifying the location, source, and the magnitude of true thermodynamic losses; determining the exergy losses in each process step which reduces the performance of the system, and comparing various process configurations to determine the most efficient route for maximum productions. Exergy analysis can be used to evaluate the thermodynamic efficiency (\(\eta\)) of a system, Eq. (2) [62].

\[
\eta = \frac{\text{Exergy of useful products}}{\text{Input Exergy}}
\]  

Eq. (2) can be adapted to evaluate the overall efficiency (\(\eta\)) of the production of ethanol through biochemical process, Eq. (3)

\[
\eta = \frac{E_{X,et} + P_{net} + E_{X,rest}}{E_{X,biom} + \sum E_{X,ch} + E_{X,L,T}}
\]

where; \(E_{X,et}\) = chemical exergy of ethanol, \(P_{net}\) = net electricity produced by the system, \(E_{X,rest}\) = exergy of the lignin-enriched residue, \(E_{X,biom}\) = input chemical exergy of biomass, \(\sum E_{X,ch}\) = sum of the chemical exergies of all inputs to the process, and \(E_{X,L,T}\) = exergy of a potential low temperature heat source supplied to the system.

Exergy balance can be applied to the system boundary of a unit operation of a process to evaluate the thermodynamics losses, Eq. (4). This equation shows that contrary to energy, exergy is not conserved.

\[
\sum_{in} E_X = \sum_{out} E_{X,prod} + \sum_{out} E_{X,wsp} + I
\]

where; \(\sum_{in} E_X\) = total input exergy flow; \(\sum_{out} E_{X,prod}\) = total output exergy flow in the products; \(\sum_{out} E_{X,wsp}\) = total output exergy flow in the waste products from the unit processes, and \(I = \text{exergy destruction due to internal irreversibility} (I \neq 0 \text{ for an irreversible process})\).

From Eq. (4) exergy loss associated with the unit process = \(\sum_{out} E_{X,wsp} + I\).

The exergy analysis can be combined with life cycle assessment (LCA) to form exergetic life cycle assessment (ELCA) which helps to account for all environmental issues as well as the depleting natural resources [62]. This involves closed material and energy balances and can be carried out by determining the exergy destruction during the process.
Several works have been carried out by investigators in the area of applying the exergy tools to evaluate the process performance in the bioethanol production from biomass. Hurtado et al. used exergy analysis to evaluate the efficiency of the bioethanol production processes using rice husks as feedstock \[63\]. Aspen Plus software was used to simulate the process and the results of the exergy analysis showed that the pretreatment stage required improvement of either mass or energy as the stage gave the lowest exergetic efficiency and highest irreversibilities.

### 6. Process simulation

This is the pictorial representation of chemical, physical, biological, other technical processes and unit operations in a simulation software. The software helps: in the design of environmental-friendly and safe processes, reduction of capital and operating costs, to provide functionality and flexibility needed for modelling efficient biofuel processes, to enhance heat recovery processes, reconcile data, verify operating conditions, efficient and optimal process design, regulatory compliance, and operational analysis of the biofuels process \[64\]. With the simulation software, engineers can work virtually, thereby avoiding expenses and time delays associated with testing the process in the real world \[64\]. Examples of simulation software used in simulating the bioethanol production include Aspen plus, Chemcad, Prosimplus, Hysys and PRO/II \[1, 64\]. The most commonly used software in biorefinery is the Aspen plus \[1\]. The simulation of bioethanol production from lignocellulosic biomass requires interconnecting the various unit operations: pretreatment, hydrolysis, fermentation and distillation involved. This has been demonstrated by Peralta-Ruiz et al. by simulating the bioethanol production process using residual microalgae biomass as the feedstock \[65\]. These investigators evaluated the most effective route from three technologies: simultaneous saccharification and co-fermentation (SSCF), simultaneous saccharification and fermentation (SSF) and separate saccharification and fermentation using acid hydrolysis (SHF) leading to the highest yield of bioethanol. The simulation was carried out using Aspen Plus 7.1 and the results obtained showed that SSCF gave the highest yield of 23.6% and SHF the lowest yield of 18.5%. With these results, they concluded that enzymatic technologies could be used for microalgal production of bioethanol.

### 7. Conclusions

The quantity of bioethanol produced globally is increasing (110 billion liters in 2018 and could be 140 billion liters in 2022) with US and Brazil currently the highest producers. These countries produce bioethanol (1G bioethanol) from corn and sugar cane, respectively. Due to high cost of production with 40–70% contribution from the feedstocks, other sources of feedstocks are being considered leading to the production of the 2G, 3G and 4G bioethanol, respectively. The high cost of processing lignocellulosic biomass into bioethanol still makes the route unattractive compared to 1G bioethanol. The 2G bioethanol constitutes less than 3% of the total bioethanol production and has a higher GHG reduction potential compared to 1G bioethanol. Results show that bioethanol has the capacity to reduce the greenhouse gases emission and global warming potential substantially and hence facilitates the protection of the ozone layer.

In order to increase the yield of bioethanol and minimize the cost of production, different processes maybe combined through integrated processes, for example SHCF, SCCF and CBP.
Based on the LCA of the bioethanol production, the environmental impacts depend on the feedstock availability and the technology used for converting them to bioethanol.

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