Integration Approach of Anaerobic Digestion and Fermentation Process Towards Producing Biogas and Bioethanol with Zero Waste: Technical

Alrefai R*, Benyounis KY and Stokes J J
School of Mechanical and Manufacturing Engineering, Dublin City University, Dublin, Ireland

Abstract
The rapid increase in the world population has caused an enormous increase in the demand of energy. Growing demand has resulted in a shortfall in conventional energy resources. Due to that and because of the major negative impacts of fossil fuel on the environment and other aspects as well, the necessity toward finding alternative cheap, renewable, and environmentally friendly energy resources has significantly arose. Biomass as an energy resource has a potential to be a good alternative for non-renewable energy resources. Anaerobic digestion process is one of the most commonly biological conversion process used in converting biomass into biofuels. It has been extensively applied in many studies for converting several types of feedstocks and has proved its significant effectiveness. (AD) digestates are generally composed of solid and liquid streams. Those streams are rich in nutrients and contain undigested materials which have not been digested in the digestion process. Despite the significant effectiveness, it would contribute in major issues if it has been applied at large scale, as the amount of digestates which would be generated is quite high. Due to that and to take an advantage of the digestates in the production of biofuel and bioproducts as well, the interests in enhancing and utilizing anaerobic digestion residues have recently much increased. Bioethanol is one of the most promising liquid biofuel. It is eco-friendly alternative to fossil fuels. In recent years, number of studies have investigated the integration approach of producing biogas and bioproduct in which would result in zero waste. However, this paper discusses mainly an integration approach for producing two promising renewable energies can be utilized in many applications with no waste generated. This approach is still at an early stage and requires further studies to improve the properties of the biofuels and high-value bio-based products produced.

Keywords: Renewable energy; Anaerobic digestion; Digestate; Bioethanol; Integration approach

Introduction
Based on the estimation of the 2015 revision of the official United Nations population, the world population in mid 2015 was approximately 7.3 billion people. Which indicates that, the world has added about a billion people in the span of the last 12 years. United Nations has also projected the global population to reach 8.5, 9.7 and 11.2 billion in the year of 2030, 2050 and 2100 respectively [1,2]. According to the International Energy Agency (IEA), the world energy demand is estimated to grow up at an optimistic slow growth rate of average annual rate of about 1.2% up to 2035. Even that, the global energy demand will increase by 38% by 2035 reaching 16,934 million tons per year [3]. This increase in energy demand is expected to greatly contribute in the depletion of world's oil reserves by 2050 [4]. Among all of that and due to the major negative impacts of non renewable energy on the global environment, the attentions have increasingly turned to alternative renewable energy sources, such as solar, wind, thermal, hydroelectric, biomass, etc. However, biomass is the fourth largest primary energy resource. It is renewable, sustainable and clean resource of energy that has numerous benefits, locally and globally. For biomass to be further used as a renewable energy, it requires conversion process to convert it into a suitable form of energy such as biogas [5]. As well known, AD is one of the most effective biological conversion process of biomass into biofuel. The final output of anaerobic digestion are biogas and two by-products (nutrient- rich liquid digestate and the fiber-rich solid digestate). It is also known that, the organic matter is not fully degraded during anaerobic digestion. For instance, anaerobic microbes can only convert approximately 40–60% of carbon into methane in anaerobic digestion of animal manure. The remained portion of carbon stays in the digestate as cellulose, hemicellulose, lignin, and protein [6,7]. Although biogas is a promising, sustainable and renewable energy substitute, the sustainable development of AD process basically depends on the ability to deal with the excessive digestate produced [8,9].

As improper handling of digestate would cause serious environmental issues. Due to that and to fully utilize the digestate and convert it into useful high-value bio-based products as well, there is an urgent need of developing new biorefining processes. Several recent studies have focused on developing an integrated system including anaerobic digestion process and fermentation process to convert various feedstocks into biofuel and high-value bio-based products [10]. This paper has an objective to provide a description summary of integration approach of AD and fermentation process to produce biogas and bioethanol in which they can be utilized in many applications and ended up with zero waste generated.

Anaerobic Digestion (AD)
The anaerobic digestion process is a biological conversion process of biomass into biofuel. It has been utilized widespread in converting several types of biomass such as; Lignocellulose materials. It breaks down organic materials by micro-organisms in oxygen-depleted environment in order to produce biogas. In general, AD process passes...
Biogas

Biogas produced from AD is a promising biofuel. It composed of different gases in different quantities. As stated before, the composition of biogas is varied depending on several factors. It is similar to what naturally occurs in decomposing organic mud at the bottom of marshes or in landfills. The decomposition process is occurred in four stages, are: Hydrolysis/Liquefaction, Acidogenesis, Acetogenesis, and Methanogenesis. Figure 1, illustrates the decomposition process. In digestion process, the substrate must undergo all of these stages. As shown in Figure 1, hydrolysis is the first step. In this step, Micro-organism breaks down particulate organic substrate into liquefied monomers and polymers. In case the organic substrate were proteins, carbohydrates, and fats, they will be converted by Micro-organism into amino acids, monosaccharides and fatty acids respectively. Solubilized monomers resulted in the first step, would by further broken down by acidogenic bacteria into the following organic compounds: short chain volatile acids, ketones, methanol, alcohols, hydrogen and carbon dioxide. The last two steps usually run parallel, as symbiosis of two groups of organisms. In the Acetogenesis step, propionic acid and butyric acid are converted by acetogenic bacteria into hydrogen, carbon dioxide and acetic acid. The results are further converted by methanogens bacteria into methane and carbon dioxide. Approximately two-thirds of the methane produced from an anaerobic digestion, originates from acetate. While, the remaining amount produced are derived from conversion of hydrogen and carbon dioxide. However, AD is efficient, requires low energy and generally contribute in reducing greenhouse gases. The composition of the biogas produced varies to some extent depending on several parameters such as feedstock types, digestion systems, temperature, retention time, etc. This biogas can then be used as chemical feedstock or as biofuel [11-15]. While, the digestate produced comes in solid and liquid streams in which can be separated from each other and further used in several applications. Liquid digestate is commonly used in agriculture as fertilizers. While, solid residues can be composted, used for dairy bedding or applied directly to cropland. It can be also used in generating high-value bio-based products through biorefinery concepts i.e., Bioethanol [16-18]. Like any conversion process, AD has some disadvantages such as; it has poor operational stability thus, it is not widely commercialized [19]. Another drawback associated with AD if applied widely is the improper handling of the digestate. Such a drawback could make AD less attractive and cause some serious environmental issues [10] (Figure 1).

AD digestate

As previously mentioned, AD digestate comes in solid and liquids forms, rich in nutrients and contains undigested and suspended materials. The composition of the digestate depends basically on the feedstocks processed [26,27]. Table 1 depicts the typical nutrient values of digestate.

Anaerobic microbes are able to convert only a specific proportions of carbon into CH4 during digestion. Whereas, the large amount of carbon stays in the digestate in the forms of cellulose, hemicellulose, lignin, and protein [6]. In addition, any materials have not been digested in the digestion process will remain in the digestate. For further use of digestate, several pretreatment techniques can be applied. In the recent years, the attentions towards AD digestate has much increased for enhancing biogas yield or producing multiple biofuels and high-value bio-based products [26-28]. A study in 2015, was aimed to assessed the optimization of biogas produced from different substrates by using mechanical pretreatment only to the non-degraded digestate after the fermentation process for feeding it back into the process. Another aim of the study was to evaluate the losses of VFA in the digestate through warming by mechanical pretreatment, as well as the impacts on the comminuting intensities and on the degradation kinetics. In this study, ball milling pretratment as a mechanical pretreatment was applied to the digestate at four time periods. The study has concluded that, mechanical pretreatment resulted in maximum to a triplication of the methane yield and to a quadruplicating of the daily methane yield. Also, no losses of VFAs through warming was observed [29]. Furthermore, biorefinery concept in which used for making a large variety of bioproducts is quite similar to current petroleum refinery. It makes multiple bio-products including chemicals, fuels, polymers etc. from organic biomass [16]. It broadly aims to one of the two following end objectives, which are, 1) to be fractionated into three main elements for later conversion to bio-based products with maximal added-value, or 2) to produce basically biofuels, with digestates as co-bioproducts [30]. However, biorefinery concept has taken its shape lately, but still many studies and investigation should be implemented before an established system can prosper [17].
produce both ethanol and carbon dioxide [31,32]. Zymomonas mobilis metabolize sugars in oxygen-lean circumstances to certain species of yeast like saccharomyces cerevisiae or bacteria such as; are used as biofuel. Those bioethanol and the one used in alcoholic the acid-catalyzed hydration of ethylene. The majority of bioethanol is manufactured petrochemically through low toxicity and could cause only little environmental pollution if spilt. Industrial ethanol is basically manufactured petrochemically through the acid-catalyzed hydration of ethylene. The majority of bioethanol are used as biofuel. Those bioethanol and the one used in alcoholic beverages as well are typically produced by fermentation process where certain species of yeast like saccharomyces cerevisiae or bacteria such as; zymomonas mobilis metabolize sugars in oxygen-lean circumstances to produce both ethanol and carbon dioxide [31,32].

**Bioethanol**

Bioethanol or ethyl alcohol (C₂H₅OH) is one of the promising biofuel. It is eco-friendly alternative to fossil fuels and can be used as a petrol substitute for road transport vehicles. In addition to that, it is liquid, colorless, renewable, sustainable, biodegradable biofuel, has low toxicity and could cause only little environmental pollution if spilt. Bioethanol is one of the most crucial biofuel generated from bioenergy crops and biomass [33,34]. Bioethanol has a great octane number, great heat of vaporization, and small vapor pressure. Due to that, bioethanol can be the clean, renewable and green combustible fuel alternative to gasoline, and can be a replacement of it in internal combustion engines as well. Bioethanol has several advantageous properties such as; it can simply miscible and utilized as oxygenated portion in gasoline for greener emission [34,35].

Moreover, bioethanol stands at present in better position than biodiesel because of several factors such as; higher environmental sustainability of raw materials it is obtained from, better in mitigation of (GHG) emissions, less production cost and a more favorable future evolution [34,36-40]. In addition, ethanol has the potential to be blended with gasoline. This blending can greatly improve power output. In comparison, 66% of the energy produced from one liter of ethanol, can be obtained from the same quantity of gasoline. Ethanol has octane number rate (106-110) greater than gasoline (91-96) which increases the performance of gasoline when blended with ethanol [41]. It is oxygenated fuel has 34.7% oxygen, while gasoline is free oxygen. Subsequently, leads to approximately 15% increase in the combustion efficiency of ethanol than that of gasoline [42]. Indeed, all these advantageous properties and others as well are contributed in making bioethanol as one of the most crucial liquid biofuel.

**Production of bioethanol**

Bioethanol is mainly comes from biomass contain either free fermentable sugars or may be complex carbohydrate in which are able to be converted then into soluble sugars for making it fermentable. These biomass can be classified into three groups. These groups are extremely differ from each others in term of sugar solutions. The groups are; 1) sugars (sugar crops and byproducts of sugar refineries), 2) starchy crops, 3) lignocellulosic biomass (LCB) [43]. Moreover, production of bioethanol from edible foods contain sugars and starch is much easier than its production from LCBs. The cost of these sources, the environmental concerns and the debate on using of edible food for the bioenergy has created the reasons have made the production of bioethanol from edible sources less attractive [44]. Therefore, much interests have been grown recently by researchers and others in producing it from LCBs. Another reasons as well have drawn the attentions to lignocellulosic biomass, like; they are mostly waste materials, abundant, has low and stable cost, contain high carbohydrates, and non–competitive with food chain [45]. Furthermore, the production of bioethanol from LCBs contributes in reducing environmental pollution as its emissions of net greenhouse gas are low or insignificant [46].

In general, the production of lignocellulosic ethanol involves several steps which are shown in Figure 2 and passes through five main stages [31]. These stages are; biomass pretreatment and/or detoxification, hydrolysis, fermentation and product recovery. So far, many pretreatment techniques have been evaluated on a wide range of lignocellulosic materials, either simple or technologically and logistically more intensive [47]. Pretreatment is the most important stage as it has a major influences on all the other steps in the process. It is the most complicated and costly stage in the conversion of LCBs into ethanol [48]. For selection of the most suitable and effective pretreatment of LCBs, number of criteria should be taken into account, such as; should require low capital and operational costs, require minimum size reduction of the biomass, toxic compounds produced under the pretreatment conditions should be minimum, etc. [49,50]. Physical pretreatments is one of the four pretreatment groups can be used in the production process. It usually helps in increasing the accessible surface area and pore volume, decreasing the degree of polymerization of cellulose and its crystallinity, hydrolysis of hemicelluloses, and partial depolymerization of lignin [51,52]. They are expensive techniques and in some cases require energy higher than the theoretical energy content available in the feedstock. Therefore, physical pretreatment usually not recommended to be used on a large scale [53]. Chemical pretreatments is one of the pretreatments groups. It mainly include alkali and acid pretreatment methods. Sodium hydroxide NaOH is the common chemical compounds used in alkali pretreatment. While, sulfuric acid H₂SO₄ is the most common acid used in acidic pretreatment. Basically, alkalis and acids help in delignification, decreasing the degree of polymerization and crystallinity of cellulose from biomass [47,49,50,54].

Detoxification stage is essential. It comes after pretreatment to remove inhibitors produced in pretreatment stage [55]. In hydrolysis stage, both cellulose and hemicellulose present in LCBs are converted into fermentable sugars [56]. These sugars produced are then fermented by the relevant microorganisms and converted into ethanol. Two fermentation methods can be applied in ethanol fermentation, which are: submerged or solid state fermentation [57]. Ethanol recovery from the fermentation broth is typically done by distillation or distillation combined with adsorption [47].

However, many research have been carried out on the investigation of the production of bioethanol from various renewable resources such as; waste biomass of lignocellulosic and starch-based origin, such as municipal solid waste, industrial waste (waste paper or coffee residues), livestock manure, and agricultural waste (wood biomass and agricultural crop residues) [58]. Also, numerous studies across the world have been investigated bioethanol production from different types of fruit wastes. Usually, large quantity of fruits are disposed at harvest and during marketing due to several problems make them unfit for human consumption (e.g. low quality or unacceptable physical appearance) [59,60]. A study was conducted in 2013 on the ethanol production from different fruit waste with use of microorganism called saccharomyces cerevisiae. In this study, the waste of the following fruits were the feedstocks used for making of bioethanol: apple, banana, papaya and...
grapes. The highest yield of bioethanol was achieved from grape wastes at pH 5.4, 30°C, specific gravity 0.860, and concentration of 6.21%. The study has concluded that, grape waste has higher efficiency than other fruit wastes have been investigated in this study or the mixed fruit wastes. It has been recommended that, fruit wastes contain fermentable sugar should be utilized, not disposed into our environment, and converted to useful bioproducts such as; bioethanol that are able to serve as an alternative energy source [61]. However, conversion of feedstocks into bioethanol are varied greatly depending on the nature of feedstock, primarily due to the variation in biochemical composition. Therefore, only a few feedstocks have been exploited commercially [62]. While, the final yield of bioethanol of biomass source depends considerably on the efficiency of overall conversion [63] (Figure 2).

Production of bioethanol from AD digestate as a feedstock

As stated before, AD digestate can be separated into solid and liquid digestate. In case of integration approach, liquid digestate is separated to be further used in several applications. While, solid digestate is fully utilized to produce bioproducts [16-18]. Teater C et al. [6], have accomplished a study on the production of bioethanol from anaerobic digestion solid digestate as feedstock. The study has two major objectives, which are, evaluation and comparing the suitability of AD fiber from a completely stirred reactor (CSTR) for ethanol production with more commonly researched feedstocks; switchgrass and corn stover. The second objective of the study was to statistically specify the optimal dilute alkali pretreatment conditions (temperature, retention time and NaOH concentration). Overall glucose conversion of CSTR AD fiber resulted from this study was consistent to those of switchgrass and corn stover. However, this study is the first study has reported pre-treating of AD fiber to produce fuels/chemicals. It has ended up with that, NaOH pretreatment improved the enzymatic digestibility of AD fiber and enhanced the glucose and ethanol yields from enzymatic hydrolysis and fermentation. For further conclusions, the study has suggested an investigation of other pretreatment techniques, enzyme loading tests, scale-up, economic analysis, and life-cycle analysis of the conversion processes. Which indicates that, the production of bioethanol from anaerobic digestion solid digestate is still at early stage and require more assessments to much improve the process and enhance the properties of the product produced. In this study, AD digestate was firstly separated by commercial screw press separator into two streams: nitrogen and phosphorus- rich liquid digestate and fiber- rich solid digestate. AD fiber, switchgrass and corn stover were then pretreated by NaOH. In order to neutralize the pretreated mixtures to certain PH values, sulfuric acid solution at was applied at appropriate percentage. After that, the mixtures were centrifuged and rinsed using de-ionized water. For analyze the solid residue for dry matter and fiber content, wet solid samples were freeze at certain temperature. After mixing the frozen wet solid samples in a flask, they were autoclaved with deionized water. Following that, cellulase and a buffer solution was added to the previous mix. The flask was then placed in a shaker inside an incubator for 72 h, the sample was boiled and filtered using filter paper to analyze gloucose and other sugars as well. Afterward, enzymatic hydrolysis at appropriate % dry matter concentration was performed to the most effectively pretreated samples. Lastly, for the fermentation of ethanol, an appropriate microorganism should be applied.

Moreover, Dianlong W et al. have assessed Methane yield of AD at mesophilic and thermophilic conditions and the production of ethanol from solid digestate [8]. In the study, the pretreatment of ozone combined with aqueous ammonia was applied in order to recover residual organic carbon from solid digestate after AD of rice straw as a feedstock. The study has resulted that, CH4 yield at thermophilic condition was 72.2% higher than that at mesophilic condition under the same conditions (24 days and 17% solid concentration). It resulted also that, ethanol production efficiency of solid digestate after thermophilic condition was 24.3% higher than that mesophilic condition. In addition, the study concluded that pretreatment with ozone combined with aqueous ammonia is better approach to improve ethanol production from solid digestate of both mesophilic and thermophilic ADs. The study has revealed that, an integration of AD with ethanol fermentation process was an efficient biorefinery process, achieving a successive utilization of organic carbon.

Discussion

One of the major drawbacks which can minimize the uses and effectiveness of AD process is the improper handling of digestate. This drawback would lead to serious environmental issues. Due to that, the demand to find the right way to get rid of the digestate has became very important. Where the amount of digestate produced would be huge if AD has been applied wider. An integration approach is one of the most efficient and ideal solutions to these issues. This approach mainly aims to make full use of the biomass to produce biofuel and bioproducts simultaneously with minimum or no waste. That’s would significantly contribute in improving the industrial value of biofuel productions. In addition to that, number of studies on the development of integration approach have been recently made use of biogas produced from AD to power fermentation process in order to develop an energy-positive process. However, this approach is still new and required further studies and assessments to enhance the quality of the approach and improve the properties of the bioproducts produced.

Conclusion

As a liquid biofuel, Bioethanol is a promising, eco-friendly and
good alternative to fossil fuel. Integration approach for producing biofuel and high-value bio-based products from anaerobic digestion solid digestate as a feedstock is relatively a new approach. It significantly helps in mitigating the issues of producing large amounts of digestate in which associated with anaerobic digestion (AD) when it is applied in a large scale. In addition, this approach greatly contributes to: reduce or produce zero waste, fully utilization of biomass and produce biofuel and high-value bio-based products. However, this integration approach still require more investigations regarding to several aspects such as: pretreatment technique, economic analysis, etc. For future study, an investigation of production of another bioproducts such as: biodiesel and bioplastic from AD digestate is suggested.

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