



Review Paper

A review on production of biofuels from novel biomass

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Abstract

Nowadays, with a great increase in global population, converting the society to a comfortable feel good environment would be the only solution for stabling a permanent life on this planet. There is considerable potential to improve biomass fuels in producing suitable power sources such as combustible fuels, electricity and fuel gases, while abiding to provide conventional uses of biomass. The upgrading qualities and the industrial investment of biomass have already happening in many countries because biomass energy has specific amount of environmental and social benefits compared with fossil fuels. Biofuel is a fuel, derived from living organisms mostly from plants and other microbes. We can produce biofuels in an efficient and sustainable manner. The below review discusses various sources of biomass and their rate of production for different fuels (biomethanol, biobutanol, bioethanol, biodiesel and bio-hydrogen). Biomass energy production's global potential is large in absolute terms which could be realistically used to supply nearly 1,0,000Mega Watts (100quads) of electric capacity by 2020, and perhaps we can suspect the amount will be doubled by the year 2030. Sustainable economic growth along with industrial growth needs safe and feasible resources of energy. For the future re-arrangement of an encounter economy, we completely require new approaches in research and product development. In this review, we are mainly focusing on cost effective technologies and some mechanisms to convert biomass into useful liquid biofuels and bio products. We specifically focus on the production of alternative fuels from different feedstock and aiming to give comparative studies.

Keywords: Biomass, Biofuel, Feedstock, Fossil fuel, Bio product.

Introduction

Contemporarily, we are facing a global energy crisis as a cause of increased population rate, decreasing fossil fuel resources and environmental changes that concern an increasing demand for a substitute fuel source. Owing to less availability of fuels, many countries have raised the cost of fuels. The search for an alternative source for biofuel with less cost and slightest environmental effects has become the point of convergence. Biomass is known as the sustainable resource that can be used in a large scale fuel yield and causes less environmental effect. Biomass resources include agricultural wastes, municipality wastes, industrial wastes and forest residues. Biomass can be transformed into usable aspects of energy. As the achievability of biomass sources such as lifeless plants, composts and trash may not drop on comparing with limited fossil fuel, the different energy sources is considered to be inexhaustible. Carbon-neutrality is one of the leading advantages of biomass energy¹. Generating energy from biomass material has become the major issue throughout the world, which helps in waste management. By increasing the number of biomass energy plants, scraping wastes in landfills can be lessen. Thus biomass has a productive influence on the surrounding.

Because of the ample availability of biomass, biomass energy has a massive potential. As the production and consumption

levels tend to increase, biofuels can be produced effectively by using biomass as an alternative.

Biofuel is a biodegradable fuel derived from living organisms mostly from plants and microbes. Biofuels obtained from natural resources help to reduce greenhouse effect and global warming. Lignocellulosic biomass is used as the primary substance for burning and generating heat and electricity. The utilization of these wastes as the source of generating biofuel is an eco-friendly method. Another advantage of biofuel is to reduce the dependency on woody biomasses from forest, which results in lowering deforestation.

Some studies have aimed for biofuel production using industrial raw material and green processes of biomass. There are first- and second-generation biofuels. First generation biofuels refer to the fuel synthesized from some alimentations including carbohydrates obtained from corn, cane-sugar, sugar beet and wheat. There are three types of biofuel in first generation which is majorly used in transportation with huge production around the world. These fuels encompass bio-ethanol, bio-diesel and biogas. Second generation biofuels are highly manufactured from non-edible sources especially from biomass rich in lignin and cellulose. These biofuels liberate less volume of carbon dioxide into the earth's atmosphere on combustion, thereby reducing greenhouse gas. Plant biomass is an optimistic

feedstock for producing advanced biofuels. The disadvantages of biofuel include regional suitability, food security, and global warming. According to studies, fossil fuels will deplete in 2050. Over 100 billion tonnes of carbon is produced from the primary production of biomass. To know the importance of biofuels and its beneficial impact over environment a comparative review was made.

Biomass

Biomass is an organic material that can be produced using natural materials obtained from plants and animal wastes. These biomasses are primarily used to produce the energy (heat, electricity, liquid and gaseous fuels). Such energy production process is called Bio-energy. This has become a prominent energy source as it is one of the major alternatives for power generation over the past few decades. Biomass is initially found in the form of living or recently dead plant matters. On further biomasses are produced from biological wastes (plant materials, food and crops waste, agricultural waste, fishery waste, and animal waste) that can be turned into useful products like biofuel, industrial chemicals and fibers². The major energy feed stocks are from debris including agricultural, wood matters, municipality wastes and landfills gas. Woody biofuels contain very small amount of sulfur and contain considerably less fuel-bound nitrogen on comparison with oil and coal. Another source of biomass is sewage sludge. There are some knowledge-based researches involving in biomass derived from algae. Other feed stocks for biofuel production include enzymes and bacterial species from various sources (grown in cell cultures). All biomass is biologically-produced matter composed of carbon, hydrogen and oxygen. In 2020, various countries will use biomass as the only source of biofuel for domestic use³. Roughly 700 million metric tons of carbon per year is produced from estimated biomass but only 2% amount is transferred to biofuels (bio-ethanol). Energy produced from biomass are the most sustainable energy sources to meet up the day today energy requirements of human society. Production, conversion and utilization are performed efficiently with increased biomass on an availability basis which requires some support and development of new biomass units in an environmentally sustainable manner.

Biofuel

Biofuels, the fuels derived from organic biomasses. According to International Energy Agencies, biofuels that are produced meets nearly 1.5% of global transportation. To reduce Greenhouse Gases, biofuel production from biomass is chosen as the suitable method. There are various types of biofuels based on composition and properties such as biodiesel, biogas, biobutanol, biomethanol, biohydrogen etc., Generally biofuels are separated into first, second and third generation, whereas fourth generation biofuel is coming up from a fundamental research. The primary biofuels are manufactured from certain crops (wheat, rice, sugarcane, sugar-beet, soya bean oil etc.). Advanced biofuels are produced from viablere sources

including agriculture products, municipality wastes, used cooking oil, sewage sludge and industrial waste. Tertiary biofuels are produced from algae feedstock. Fourth grade biofuels, these are basically produced from photo-biological solar fuel and electoral fuel. The countries which are facing heaviest biofuel demand includes India, China, US, Brazil, and European Union accounting for nearly 90% of world biofuel demand up to 2035 (world energy outlook 2010). It is awaited that, global ethanol production will exceed by 14% during the considered period from 2017 to 2027, that is 120bln L to 131 bln L. Fifty percent of this increase is expected from Brazil, to fill domestic demand⁴. Further in this review we have given some brief explanation of various biofuels synthesized from different biomasses and its rate of production.

Biomethanol

Methanol is a clear liquid fuel which is water soluble and readily biodegradable one⁵. Methanol is one of the most flexible fuel commodities that have been produced from several feed stocks. Natural gas produces above 75% of Biomethanol, which was one of the alternative compounds. At present, it was said that more than 50 million tons of Bio-methanol was generated from 90 methanol plants. The techniques involved for Bio-methanol production are pyrolysis, gasterification, electrolysis, bio-synthesis and photo electrochemical processes⁶. Bio-methanol is most favorable and promising biofuel because of its ability to migrate CO₂ and being a liquid component it was easily transportable. It has a higher volumetric energy density in comparison with bio-ethanol.

In industry, various useful organic compounds are readily synthesized from raw material. It is used as a substitute for petroleum derived hydrocarbons and related chemicals as it is mainly composed of carbon. In comparison with pure gasoline fuel and other gasoline products, it has higher octane number. Table-1 discusses the various sources for bio-methanol production.

Table-1: Various sources for the production of bio-methanol.

Source of the biofuel	Microorganism used	Reference
Herbaceous plant, Crop Residues And Trees	Yeast	7
Goat Manure	Methanotrophic Bacteria	8
Agricultural Waste And Livestock Waste.	Anaerobic And Methanotrophic Bacteria	9
Microalgal Feedstock	Various Microalgae	10
Swine Manure	Taihu Blue Green Algae	11
Sugarcane Plant	Yeast	12

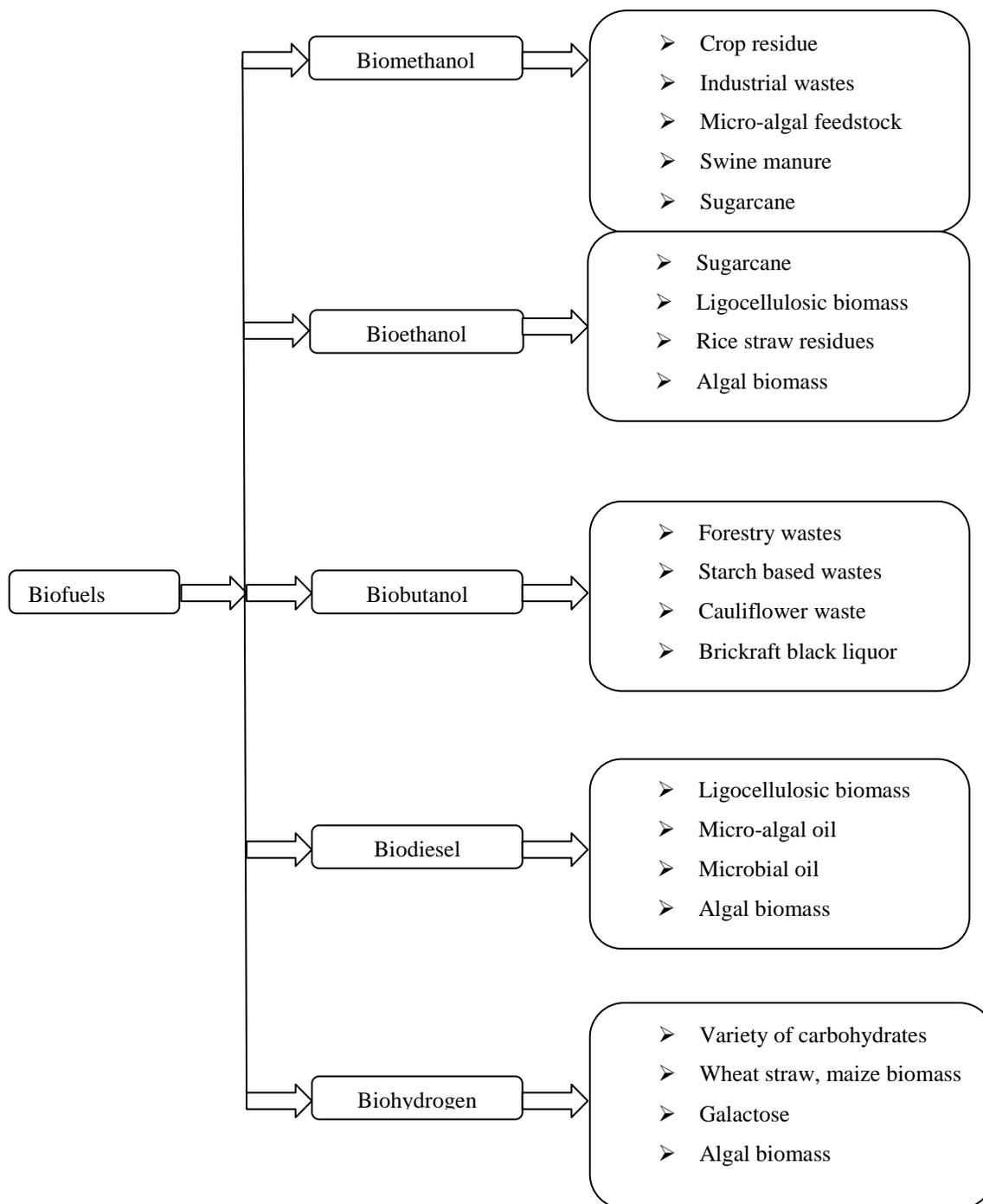


Figure-1: A simple flow diagram on production of different biofuels from various biomasses.

Goat manure: Another important source for the biomethanol synthesis is the goat manure. Cell concentration factors such as 0.4gL^{-1} and 0.36gL^{-1} were chosen and the process was carried out. The results are obtained and shown that higher concentration resulted in efficient biomethanol production without any particular conditions within 11 days whereas lower concentration resulted in production of biomethanol within 15 days with fermentation process⁸. A lab scale research was

carried out by M. Anitha et al. shown that skin of banana and well boiled straw feedstocks blended with cow manure inoculums are best source for biomethanol production. Cow dung, chicken wastes and goat manure inoculums are also adaptable for the production of biomethanol through bio-solid digestion⁹. The other studies show that biomethanol produces from goat manure gives a good result (2.48 wt% of biomethanol) through fermentation process, which is much

greater on comparison with cow dung (0.04wt%)¹⁵, from corn husk (2.07wt%), grasses (1.15wt%) and with leaves (1.25wt%).

Algal biomass: *Spirulina* species is the kind of algae which is known as the good source of algae for the production of methane, where growth rate of algae in photobioreactor is assumed to be 1kg/(m³d)^{10,16-18}. Several studies suggest that one tone algae produce 9.4 GJ of methane¹⁹⁻²². Growth of algae is significant. Nutrient rich sources are added to cultivate algae in photo bioreactor in order to yield high growth rate. The growth of algae can be achieved by using nitrogen supplement from poultry waste by continuous illumination. The work by X. Wang et al.⁶ it is assumed, 7776 GJ bio-methane can be produced in 180 days, the best method for cultivating algae is without greenhouse heating. This assumption is made according to PBR structure and algal characteristics.

Swine manure: The studies have shown that swine manure produces less amount of methane when compared to other biomasses. Fatty acids, amino acids and polysaccharides are also set on as a substrate for swine manure and blue green algae. In Anaerobic Digestion (AD) all the precursors of methane come from Volatile Solids (VS), where Volatile Solids are essential organic feedstock for anaerobic fermentation²³. H. Miao et al experiments scrutinizes that three enzymes (AK, protease, coenzyme F₄₂₀) are responsible for acidification, hydrolysis, and methanation respectively. C. Gonzalez-Fernandez et al. studies show that the highest methane benefits (246mL CH₄g COD_{in}⁻¹) was achieved by the 6th trial conducted by researchers. The studies show that, after 22 days the ISR (Inoculums Substrate Ratio) increases from 0.5 to 2.0 where the methane production rises from 48.1mLg⁻¹ VS to 212.6mLg⁻¹VS. Due to oversaturation of inoculums, the methane production reduced to 191.3mLg⁻¹VS. The same results were obtained by the other researchers using different substrate²⁴.

Sugarcane: High solid fed batch saccharification and fermentation process are the two major process involved in biofuel production using sugarcane bagasse as a biomass source, next to evaporation, the residues acquired are used for bio methane production through anaerobic digestion²⁵. In this the pH of the anaerobic digester was maintained between 6.5 and 8.5. The bio-methane production in presence and absence of NaOH pretreatment was determined by using anaerobic bath digesters. The methane yield obtained from the two different bagasse varied between 236 to 326NmLgvs⁻¹, this is due to difference in the concentration of non-fiber carbohydrates. Hence, the production of bio-methane using sugarcane bagasse helps in reduction of CO₂ in environment.

Bioethanol

Bio-ethanol - a degradable biofuel which is a clear colorless liquid. Ethanol when burned will produce carbon dioxide and water. It causes less environmental pollution and low toxicity even when it splits. Ethanol acts as an octane enhancer in diesel.

Ethanol has a GGE value-gasoline gallon equivalent of 1.5 which proves to take the place of the energy gasoline. Biomass contains a difficult combination of polysaccharides from the cell wall-cellulose, hemicellulose and lignin. So bio-ethanol is primarily formed from agricultural feedstock. Crops which has high lignocellulosic compound such as rice, sugarcane, potato, cassava and corn are chosen for production. A large area is required for growing crops, energy maintenance and pollution balance, which resulted increase in food prices. Current growth with cellulosic ethanol production may diminish some of these concerns. There is an upcoming research on developing ethanol from the use of municipal solid wastes. Potentially 49:1 GL year⁻¹ of bio-ethanol could be probably produced from the wasted crops (73:9 teragram of dry crops) in the entire world²⁶. On comparison with other biofuels, the production cost is very low. Approximately the bio-ethanol provides 93% more energy per gallon compared to biodiesel. Nearly 450 gallons of ethanol could be produced per acre of energy plants. While comparing with other regular gasses, bio-ethanol emits less greenhouse gas (12%). The Table-2 had provided the various sources for synthesis of bioethanol.

Table-2: Various sources for the production of bioethanol.

Biofuel source	Microorganisms used	Reference
Sugarcane	Yeast <i>Ceriporiopsis subver mispora</i>	27
Lignocellulosic biomass	Yeast (<i>S. cerevisiae</i>)	28
Rice straw residues	<i>T.reesei</i>	29
Algal biomass, pineapple, fish biomass	<i>S. cerevisiae</i>	29

Sugarcane: The experiments done by Yunyun Liu et al., using sugarcane bagasse clearly shows that production of ethanol is high during 12 to 24hr in a rate 0.943g/L\$h, and have reached the peak value of 20.42g/L with 69.81% efficiency in conversion, through 2 days of fermentation (yeast) with 1 day of pre-hydrolysis. Thus, from this study we obtain the change in sugar production which apparently increases the bio-ethanol production³⁰. Chizuru Sasaki et al.²⁷ experiment shows that treatment (using *Ceriporiopsis subver mispora fungus*) in arrangement with microwave hydrothermolysis enhanced enzymatic reduction of higher carbohydrate to smaller monosaccharides and ethanol fermentation of sugarcane wastes. The bioethanol concentration after 24 hours was 15.3 and 10.2g/L for the feed stocks in presence and absence of the treatment with fungus, which correlates the overall ethanol yield of 35.6% and 27.1% respectively. This relatively indicates that the use of fungi for SSF increases the bio-ethanol production by 8.8%. Native (single-pressure) and double effect distillation columns are the two configuration for the distillation column chosen by Marina O.S. Dias et al.¹². Double distillation is chosen for production of bioethanol because 90% of bagasses

are available whereas only 76% of bagasses are available for conventional distillation¹².

Lignocellulosic materials: The work from A. Limayem et al.³¹ shows that bio-ethanol production processes are usually associated with thermophilic and cellulolytic microbes including organisms such as *T. reesei* along with *P. chrysosporium*, *K. marxianus* and *C. cellulolyticum* with some among them possess fermentative capability in addition to their hydrolytic potential. However, *S. cerevisiae* and ethanologenic *Z. mobilis* depicts the higher alcohol tolerance and ethanol yield. Devendra Prasad Maurya et al., discuss a variety of pretreatment technologies for lignocellulosic biomasses were discussed to improve ethanol production. Due to existence of lignin this is considered as a main technique which has the capacity to inhibit the unbinding of water from cellulose and hemicellulose. Increase in cost of finished products is due to loss of biomass which effects final yield i.e., bio-ethanol³². Piotr Oleśkiewicz-Popiel et al.²⁸ experimental results proves that using wet biomasses obvious quantity of ethanol is synthesised only from ensiled crops: production of bioethanol for maize (72.1-77.6%), for rye (72.6-80.8%) and for clover (75.5-79.6%) by solid state fermenter using cellulolytic proteins and yeast (*S. cerevisiae* and *K. marxianus*).

Rice straw residues: Rice straw is the prospective substrate for bioethanol synthesis. This has large cellulose and hemicelluloses substance which is easily converted into fermentation sugar molecules. The outer straw mostly composed of cellulose (35-50%), hemicellulose (14-25%) and lignin (7-24%). Nearly, 731 billion tons of rice straw produces 206 billion liters of bioethanol per year²⁹. Four steps take part in bioethanol synthesis from several lignin and cellulosic biomasses. They are feedstock pre-treatment, enzymatic saccharification, fermentation and recovery of ethanol. According to the studies, it is known that rice straw is used to inactivate the *S. cerevisiae* and then it is fermented at 50°C by producing ethanol *S. cerevisiae* is inactivated in order to reduce xylitol production. Some studies with *M. indicus*, *R. oryzae* and *S. cerevisiae* show that *M. indicus* is capable to manufacture ethanol from pentose. These species is considered to be very effective for ethanol synthesis from lignin and celluloses (rice straw)³³.

Algal biomass: Algae is considered as the alternative renewable source of biomass for ethanol production, grouped under third-generation biofuel^{34,35}. The commercial advantages of algae includes, carbohydrate content in the algae cell is abundant which causes bio-ethanol synthesis. They don't have lignin and low hemicellulose levels that result in high yield fermentation and hydrolysis efficiency³⁶. Enzymatic hydrolytes are fermented through *S. cerevisiae* which produce high quantity of ethanol (14.90+0.3g/L) of productivity (0.9+0.02g/L/h) with (0.49+0.01 g/g) total yield³⁷. Biological treatment is the cost effective method for bio-ethanol production. Macro algae grow at higher proportions and gives high amount of biomass than microalgae.

Researchers said that bio-ethanol production from macro algae will be popular in future.

Pine apple: Pine apple contains dry matter, organic and inorganic components (10%, 96% 4%). There are three different methods for biologically produced ethanol from pineapple residue: direct fermentation (DF), separate hydrolysis and fermentation (SHF) in single reactor, instant aneoussac charification and fermentation (SSF). Initially pineapple residue was utilized as a source for bromelain extraction, cultivation of yeast and also for organic acids production³⁸⁻⁴¹. Enzymatic unbinding of water is the best method for ethanol synthesis. Pine apple canneries have significant amount of carbohydrates which helps in bioethanol production. The test determined by dry matter basis resulted in 3.6% and 3.8% of EY (enzymatic yield), in accordance to 89% and 96% of TY (theoretical yield)³³.

Bio-Butanol

Biobutanol which is known as 4C compounds are produced by fermentation process by using certain biomass as substrate. The properties of biobutanol and gasoline are similar whereas some gasoline powdered vehicles can also use biobutanol without any alternations. Biobutanol and petroleum fuels can be stirred together upto 11.5% of volume. 10-20% of energy content is available in biobutanol than that of gasoline. Biobutanol reduces carbon emission upto 85% in comparison with gasoline, then it is more feasible and very good alternative for gasoline and gasoline blended products. Bio-butanol has a large-spirited energy. With some energy of 105,000 british thermal unit per gallon, bio-butanol has energy nearly that of gasoline, which is abruptly 114,000 british thermal unit per gallon. Organic feed stocks from agricultural wastes, forestries, peanuts and mainly cauliflower are fermented and produce biobutanol. Acetone-butanol-ethanol (ABE) is the fermentation process which produces acetone, n-butanol and ethanol from carbohydrates and third ABE fermentation uses *Clostridium acetobutylicum* bacteria. But, this microbe easily can be poised, when the alcohol concentration increases larger than 2%. Biobutanol is ease in mixing and has low octane value, lesser energy density, small Reid Vapour Pressure (RVP), due to this property biobutanol is known as greater biofuel when compared to ethanol⁴²⁻⁴⁴. Lignocellulosic butanol is an adaptable one from a greenhouse gas (GHG) outlook as literature studies says that approximately 32.2-42.8 percentage reduction in green house gas as compared to conventional gasoline⁴⁵. The below Table-3 had provided the sources for production of bio-butanol.

Agriculture, forest, municipality solid and agro-industry waste: *Clostridium ijundahlia* is a bacterium which utilizes carbon dioxide and hydrogen. This bacterium can be used for butanol production, because forestry waste results in emission of gasses like hydrogen and carbon dioxide⁴⁶. Most commonly used raw materials in various industries are sugar and lignocellulosic biomasses. *Clostridium* is a natural biobutanol

producer because of this property it was widely used in many industries. Approximately 15.34 to 22 million gal/yr is produced using different feedstocks. The key factors for biobutanol production are raw materials, microorganism and fermentation process. The results from the study of Władysław Kamiński et al.⁴⁷ clearly states that *Clostridium* bacteria helps in separation of biobutanol from fermented broth with the help of ionic liquid compounds.

Table-3: Various sources for the production of bio-butanol.

Source of biofuel	Microorganisms used	Reference
Agriculture, forest, municipality solid and agro-industry waste	<i>Clostridium, Acetobutylicum</i>	47
Peanuts and agricultural wastes	<i>C.beijerinckii</i>	48
Cauliflower waste	<i>C.acetobutylicum</i>	49
Birch Kraft black liquor	<i>C.acetobutylicum</i>	50

Table-4: Various sources for the production of bio-diesel.

Source of Biofuel	Microorganisms used	Reference
Lignocellulosic Biomass	<i>Oleaginous Microorganisms</i>	50
Microalgal oil	<i>Chlorella protothecoides</i>	51
Microbial oil	<i>Oleaginous yeast</i>	52
Algal biomass	<i>Nannochloropsis</i>	53

Peanuts and agricultural wastes: *Clostridium acetobutylicum* B-527 was used for utilization of pea pods for biobutanol production which resulted in release of higher amount of sugar (30-48g/L) and aided efficient ABE fermentation⁵⁴. The toxicity free pea pod slurries are fermented (ABE Fermentation) that resulted in 4.26-5.99g/L of total solvent and 50% of sugar is utilized and activated charcoal detoxifier is used to remove compounds like phenol and acetic acid⁵⁵. From Table, it is known that *C.beijerinckii* BA101 utilizes waste substrate effectively for production. In some cases, starch and packing peanuts are chosen and experiment was carried. From that, experiment which used starch as a substrate shown higher results when compared to peanuts. The yields are 24.7g⁻¹ from starch, 21.7g⁻¹ from peanuts and 20.2g⁻¹ from agricultural wastes. From the studies we understood that starch can produce efficient amount of butanol⁴⁸.

Cauliflower: India is the second largest country in cauliflower production which shows 35% of total production in the world.

About 45-60 percentage of cauliflower wastes are generated while harvesting and marketing⁵⁶. Cauliflower waste contains 16.6 percentage cellulose content, 15 percentage crude protein, 8.2 percentage hemicelluloses, 16.9 percentage sugar, 6.27 percentage phenolics, 13.81 percentage ash and 10-20.5 percentage of minerals on non wetbasis⁵⁷. To improve final product detoxification is carried out to remove inhibitors⁵⁸. The hydrolyzed products obtained from cauliflower were well dried at different temperature and detoxification and acid hydrolysis are carried out in batch fermentation of *C. acetobutylicum* in production of bio butanol. It is essential to add nutrients in detoxified slurries which helps in growth rate and production of solvent^{59,60}. At 80°C, 4.3 and 5.3gL⁻¹ of butanol is obtained from dried and non-dried cauliflower which is detoxified and hydrolysed⁵³.

Birch kraft black liquor: The biobutanol production from xylan which is derived from *hardwood liquor* (by the *kraft*-based dissolving pulp production process) that acts as the renewable substrate by *C. acetobutylicum*. CO₂ filter cake was precipitated from brichkraft black liquor which was endowed using a lingo-boost carbon dioxide precipitation process with minor modification (from Smurfit kappa, Pitea, Sweden)⁶¹. Solventogenic *clostridia* are well adapted for fermenting sugars derived from lignocellulose⁶². Butanol was produce from agricultural wastes using isolated mixture of *Clostridium* species with different methods of fermentation and pretreatment took place⁶³. From the studies, other hydrolysis methods such as enzymatic hydrolysis (not generating inhibitors) can be considered. Biobutanol production from the hardwood kraft liquor is not much effective due to its commercial application⁵⁰.

Bio-Diesel: Biodiesel is a substitute fuel as like as its conventional form. Biodiesel has positive cetane ratings and lubricating ability when compared to diesel fuels (low sulfur). Nearly 37.27MJ/kg is the calorific capacity of biodiesel which is about 9% lesser than regular gasoline fuels. Shift in energy compactness is less dependence on mass production than the supplement used. It has also been asserted that biodiesel provides finer lubricity and more proper combustion. It is vaguely miscible with water. In comparison with other traditional fuels, biodiesel has a less vapor pressure and large boiling point. The biodiesel gives off sufficient vapour at 130°C to ignite air which is larger when compared to petrol (52°C). Biodiesel has a density of nearly 0.85gram per centimeter cube, higher than gasoline (~0.85g/cm³). Biodiesel is mainly synthesized from Lignocellulosic biomass, Microalgal oil, Microbial oil, Algal biomass. Biodiesel have several eco-friendly properties. The prime advantage is carbon neutrality. Biodiesel is increasingly bio-degradable and is completely non-toxic. In which the spillages represent far less risk than of diesel spillages. When compared to other gasoline fuels, biodiesel has high flash point which is considered as safe while any crash. In the below, we had discussed the production of bio-diesel from different sources as mentioned in Table-4.

Lignocellulosic biomass: Biomass rich in lignin and cellulose is the largest and very good potential source of biomass all over the earth which is the main raw material for microbial oil cultivation. Cellulose and hemicelluloses are produced at a maximum of about 85×10^9 ton/annum in the entire world; by many cereal straw which is calculated approximately 2.89×10^9 tones per annum was stated in the research work of Sun et al.⁶⁴. The article by A. Yousuf et al. simply compares and examines which microbes can facilitate the gathering of high amount of lipids based on the basic materials and the surroundings at which fermentation occurs for biodiesel production⁶⁵. Studies from Holdsworth and Ratledge explains that *Lypomycesstarkeyi* has the capability of storing large quantities of lipids compared to other lipid-accumulating yeasts⁵¹. Studies of Holdsworth et al. says that this species have shown low level of stored fatty acids reutilization⁶⁶.

Microalgal oil: The study done by X. Miao, Q. Wu gives a unified method for the biodiesel production from heterotrophic microalgal oil. Miao et al. scrutinizes that *Chlorella protothecoides*, microphytes can be photo-autotrophically or heterotrophically grown under various condition of culture¹¹. Heterotrophic growth of *Chlorella protothecoides* resulted in synthesis of huge content of lipid in cells as high as 55.20%⁶⁷. Biondi G, et al. explains that microphytes sequestrate carbon dioxide from exhaust gases emitted from fossil fuel, thereby decreasing emissions of harmful greenhouse gases. One kilogram of algal biomass requires nearly 1.9kg of carbon dioxide. Oil content in microphytes can be greater than 80% by dry weight of biomass⁶⁸. On comparison with different crops for biodiesel production, Algae produces nearly 5000–20,000 gallons of oil gal/acre/year.

Algal biomass: Trans – esterification plays an important role in conversion of algae to biodiesel. *Chlorella vulgaris*, lipid rich algae is used as a primary commodity in production by two steps i.e., sequential photo and heterotropic growth stages⁶⁹. Approximately 7.63+0.48 weight H₂O and 20.10+0.4 weight C₆H₁₄- extractable oil is present in algal biomass. Currently, microphytes which grown in either photosynthetic or in the fed-batch heterotropic formats were known as good source of lipid for biodiesel production^{52,70-73}. Green cells were grown on glucose for 7 days in the dark, which turned into yellow, and biomass grown from 2 gram per liter to 4 gram per liter. The fuel quality an important aspect can be determined from fatty acid (FA) profile of biodiesel feedstock^{74,75}. Karl Fischer titration is used to measure the water content in the feedstock like algal biomass. Hence, to produce biodiesel from an algal biomass large area is required.

Microbial oil: Oleaginous microorganisms have a rich lipid content and it is known as single cell oil (SCO) which is popular all around the world. Only a specific microorganisms are used for the biodiesel production, which will be a good source for production. Various oil-containing yeast and microalgae are grown and produced lipids which are synonymous to vegetable

oil^{76,77}, soaps⁷⁸ by utilizing sole carbon as a energy source. According to some studies, oleaginous yeast and fungi are well known alternatives for biodiesel production. As an outcome, biodiesel derived from oleaginous microorganism are well good which can replace petroleum diesel and the biodiesel from certain plant resources.

Biohydrogen

Bio-hydrogen -production of hydrogen through biological techniques. Interest in this technology is very high as H₂ is one of the clean fuels and can be effortlessly produced from particular kinds of biomass. Hydrogen is a colorless and odorless gas which is extremely reactive with oxygen and other oxides. Bio-hydrogen has low ignition energy and high flame temperature. One gram (1g) of combustion provides nearly 30,000 calories as compared to gasoline that gives only 11,000 calories. Bio-hydrogen has ability both as an energy carrier and source for chemical industry. Many provocations characterize this technology, mainly due to H₂, such as storage and transportation of a non-condensable gas. Hydrogen producing organisms are highly poisoned by O₂. There is an only small yield of H₂. As hydrogen production from conventional fossil fuels provides notably to greenhouse gas emissions. An alternative and well sustainable energy resource is biomass for production. In production of biohydrogen, a single cycle releases less than 1kg of carbon dioxide equivalent to per kg of hydrogen produced. Bio-hydrogen is over-priced in comparison with natural gas. So, credits are needed to make bio-hydrogen a competitive fuel with natural gas. Bio-hydrogen is one of the pollution free biofuel which has the ability to lessen the dependence of fossil fuels especially in energy and transportation sectors. Among the known gaseous fuels, molecular hydrogen is the best which has large energy content of 143GJ tones/wt and is the only zero-carbon fuel which ultimately produces combustion product water as a result of oxidation. Research on Biogenic hydrogen has demonstrated its promise and preference over other routes⁷⁹. However, it is absolute that the arrival of hydrogen as a renewable energy source will have supreme economic implications which provide the scientific and technological challenges to overcome. In the below, we had discussed the production of bio-hydrogen from different sources as mentioned in Table-5.

Table-5: Various sources for the production of bio-hydrogen.

Source of biofuel	Microorganisms used	Reference
Variety of carbohydrates	<i>Bacillus firmus</i>	80
Wheat straw, maize biomass.	<i>Caldicellulosiruptor Saccharolyticus</i>	81,82,83,84
Galactose	Red algae	85
Algal biomass	<i>Thermoanaerobacterium</i>	86

Variety of carbohydrates: In a large number of studies, glucose is mentioned as the most important substrate in the

production. The research done by Ren N et al.⁸⁷ indicates the maximum collective hydrogen yield and H₂ production rate in glucose.

Table-6: Hydrogen production by various bacteria.

Substrate	Bacteria	H ₂ yield	H ₂ produced	Ref.
Glucose	<i>W16 strain</i>	2.42 mol/mol glucose	12.9 mol H ₂ /L.h	87
Glucose	<i>CMC-1</i>	Not specified	1.82 mol /mol glucose	
Xylulose	<i>Clostridium acetobutylicum</i>	2.0 mol H ₂ /mol glucose	Not specified	88
Glucose	<i>Clostridium butyricum</i>	2.3 mol H ₂ /mol glucose	Not specified	89
Reducing sugar	<i>NMBL-03</i>	0.38±0.17 mol H ₂ per mole	Not specified	90

From the Table-6 it is shown that various bacteria are used for hydrogen production. According to studies, *W16 strain* yield high amount of hydrogen.

Seaweeds: Generally, See-weeds can be categorized into three groups such as green, red and brown algae (*chlorophyta* & *Charophyta*, *Rhodophyta* and *Phaeophyceae* respectively)⁸⁵. The species belonging to red algae are generally polysaccharide complex consist of agar and fibres. Hydrogen production was conducted in 7-L fermentor in presence of red algae (*Gelidium amansii*). Dry algae of 54.4 dry weights per litre yield 53.5 ml of hydrogen per gram. This has reported a maximum value of 21-29 ml/gm when compared with the biomasses obtained from land such as wheat bran, food wastes, rice bran etc.,⁹¹⁻⁹³. In the case of red algae, galactose is the suitable sugar source for hydrogen fermentation. Galactose has a maximum production rate of 2.46L H₂ per gram and a yield of 2.03mol H₂/mol galactose added to this byproduct is well suited for hydrogen fermentation. Thus, Red algae are considered as the suitable source for production of hydrogen.

Algal biomass: Biomasses obtained from algae are very good feedstock source for bio-hydrogen synthesis using direct or indirect photolysis of water⁹⁴. The volatile fatty acid production is always associated with hydrogen production⁹⁵. Potential feedstock for hydrogen production can be obtained from different environments such as naturally grown algal biomass, algal biomass grown in wastewater or exhaust gas from thermal power plants, remaining biomass after lipid and valuable component extraction. Less than one-third of energy in glucose gets turned into hydrogen in dark fermentation, example: *C. sorokiniana* (algal biomass). Moreover, the peak production of hydrogen is 2.15m³ and production of hydrogen is 0.34m³m⁻³h⁻¹.

On the whole, measures were observed in 200 dm³m⁻³HCl treated biomass⁸⁶.

Crop biomass: The bagasses of wheat straw, maize and sweet sorghum biomass plays a major part in bio-hydrogen production as a reason of its carbohydrate source and mostly importantly it is produced using dark fermentation. These feedstocks are economically feasible hydrogen technology and cheap, widely available substrate. *C. saccharolyticus* is a fermentative anaerobe, extreme thermophile, gram-positive microbe utilizes lots of substrates including carbohydrates such as cellulose, hemicelluloses, pectin⁸¹⁻⁸⁴. *C. saccharolyticus* is one of the most markable thermophilic bacteria, carrying out hydrogen generation process efficiently with considerable degree of metabolic byproducts. The uses of clean substances such as sugars in hydrogen production were justified with mapped metabolic routes. When compared to mesophilic bacteria, thermophilic bacteria yield 1.89 to 3.8 mole hydrogen per mole glucose. Hence, we conclude that hydrogen is produced enormously at high temperature.

Conclusion

This paper mainly discussed several biofuel productions (first, second and third generation biofuels) from various renewable biomasses using microorganisms. Biofuels are highly used in power generation (heating and lighting), transportation etc., there are huge number of bio-based products like wood, sugar bagasses, grasses etc., for production of bio based fuels. Substituting fossil fuels with biofuels has clearly shown reduction in greenhouse gasses. So, attention is needed for such profitable benefits including co-products of the biofuels. It is estimated that in 2030, there will be an immense potential for production of biofuels from non-edible plant leftovers. It is known that the biofuel production helps in managing wastes, environmental issues and economy which includes improved management in landfills, creation of jobs, surplus agricultural lands were used in many industrialized countries, providing some of the modern energy carriers for rural communities which helps in carbon dioxide ratio reduction, controlling of wastes, recycling of nutrients and so on. Some biological research will be necessary for contributing in the improvement of biofuel production by energy plants breeding, hydrolysis techniques using enzymes, specialized strains for fermentation and in treatment of waste. Thus, commercialization of biomass power may well serve to meet global energy needs and future economic goals. Hence biomasses played a great role in biofuel production and have good outcomes.

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