7 Microbial Intervention in Waste Remediation for Bio-Energy Production

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Abstract

The extensive exploitation of fossil fuels and the increasing global demand for energy entailed producing alternative fuels to swamp fossil fuels. Production of biofuels from biological, agricultural, municipal, and other waste products can be an alternative option to fossil fuels. Presently, biofuel production from waste products has marginally reduced the dependency on fossil fuels for energy. Eco-friendly renewable energy fuels such as biodiesel, bioethanol, biobutanol, biohydrogen, and biogas resulting from biomass conversion from agricultural waste, microalgae, or biological wastes have significantly contributed to the wellness of the economy as well as the environment. Biofuels are generated by biological processes such as fermentation via applications of suitable microorganisms from different genera with diverse biofuel production

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B. K. Kashyap, M. K. Solanki (eds.), *Current Research Trends and Applications in Waste Management*, https://doi.org/10.1007/978-981-99-3106-4_7

mechanisms. The effect of wastes on the environment, potential waste products which could be used as raw material for biofuels production, types of biofuels produced from the waste products, and potential microorganisms used in biofuel production have been discussed in the present chapter. Emphasis has been given to putative biochemical pathways involved in bio-energy production, along with recent research and updates on utilising different sustainable resources for bio-energy production. Finally, the chapter has concluded with prominent challenges encountered during biofuel production from waste materials and potential mitigation strategies for them.

Keywords

Biofuel · Bioethanol · Energy demand · Microbes · Sustainability

7.1 Introduction

The world population is expected to reach 9.7 billion by 2050 (FAO 2009). The increased population requires food and energy security, along with the augmentation and up-gradation of current technologies used to dispose of agricultural, food, and other wastes in an eco-friendly manner. The rapidly depleting fossil fuel sources, increasing energy demand, and rising environmental pollution levels have pushed the world to look for alternative, sustainable, and environmentally safe energy sources. Waste, an inevitable by-product of day-to-day human activities, could be an alternative source of energy. Due to the widening industrialisation and rapidly growing demand for food supply, waste generation will be an unavoidable threat in the near future. The emission of greenhouse gases and the accumulation of solid wastes are the associated risk factors with the waste. Hence, converting waste into energy could be an effective method to mitigate the energy crisis and pollution. The conversion of biodegradable (agriculture and food wastes) wastes into biofuel is a good choice, which is being explored extensively for energy production.

The initial biofuel production approaches had severe drawbacks and needed inevitable improvement. For example, the production of first-generation biofuel (bioethanol from the substrates with high starch content, such as corn, wheat, etc.) uses to demand food materials for biofuel production. The negative aspect of this approach was that it required food crops. Hence it was snatching the food reserve as well as agricultural land. This increased the pressure on crop production from 2000 to 2015 (FAO and OECD 2019). FAO reviewed the first-generation biofuel production and warned about its dangers in 2009 (FAO 2009). If this approach was followed, it would have resulted in a serious risk to food security for humans and overuse of agricultural land. Hence, agricultural diversification and alternatives to food crops were searched for biofuel production. Currently, extensive research works on third and fourth generation biofuels.

Various technologies are being used for biofuel production from biodegradable and non-biodegradable wastes, which can be classified into biochemical and thermochemical processes (Jeguirim and Limousy 2018; Bharati et al. 2020). In biochemical processes, microorganisms play a crucial role in transforming organic biomass into biodiesel, bioethanol, and biogas. Whereas in thermochemical processes, bio-hydrogen and bio-oil are produced by combustion, gasification, and pyrolysis. The selection of the processes for biofuel production primarily depends on the feedstock's nature and available pre-treatment methods (Singh and Das 2019).

Recent technologies have shown the potential of microorganisms in the production of bioethanol and biogas. The innovation in bioethanol production from first- and second-generation biofuel using yeast and genetically engineered bacterial strains has been well known for the past few years. Recent studies also reveal the high yields of alcohol from syngas using acetogenic bacteria in indirect fermentation (Liou et al. 2005; Maurya et al. 2020). Similarly, processing algal lipids is a promising and carbon-neutral approach to converting sunlight and CO_2 into biodiesel. Hence, in this chapter, the classes of biofuels and the potential of microorganisms in converting deteriorating wastes into beneficial biofuels have been described in detail.

7.2 Potential Biofuels Transformed from Wastes

7.2.1 Types of Biofuels

Biomass is one of the most valuable sources as it supplies food, feed materials, and energy in a human-dominated ecosystem of the Earth. In the context of a renewed return to a so-called biobased economy, as it was practised for many centuries before industrialisation, a new focus will be laid on the production of food, feed, bio-based materials, and bioenergy from biomass. Therefore, new value chains will have to be developed that include the primary production of biobased resources, their conversion to higher-value goods, and their energetic use after their lifespan or from wastes produced alongside the value chains (Zörb et al. 2018) (Fig. 7.1).

Biomass can be converted into usable energy such as fuel, electricity, and heat via three different conversion pathways: thermo-chemical, physio-chemical, and biochemical pathways (Madakka et al. 2020). Various biomasses can be converted into energy carriers in solid, liquid, and gaseous forms using either of these three pathways (Fig. 7.1). Thermochemical conversion includes the processes of carbonisation, gasification, or pyrolysis and will result in solid, gaseous, and liquid forms of bioenergy. In Physico-chemical conversion, the biomass is given mechanical and chemical treatment, resulting in the extraction of plant oils. The plant oils are converted into biofuels after their transesterification. In biochemical conversion processes, alcoholic fermentation and anaerobic digestion transform the biomass into liquid, and gaseous energy carries.

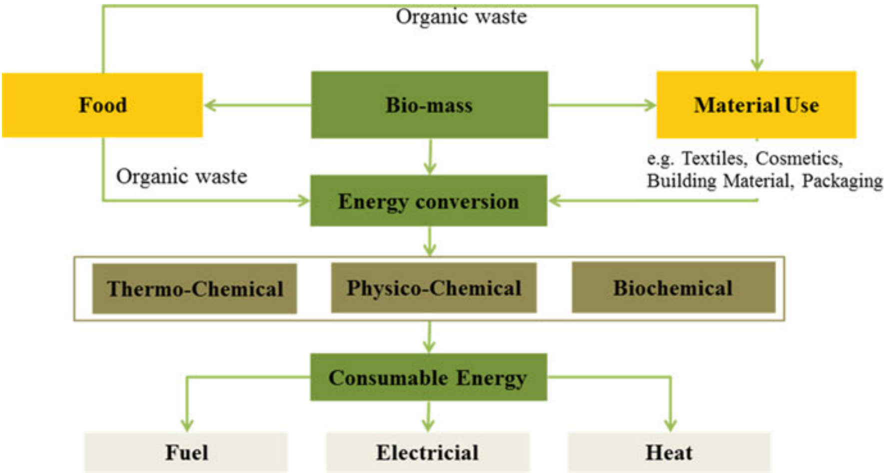

Fig. 7.1 Conversion paths from biomass to energy

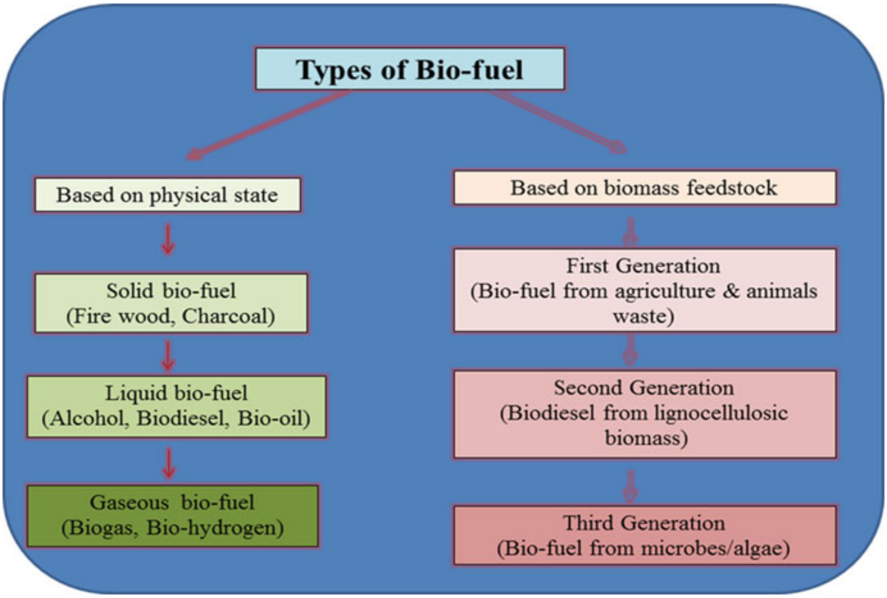

Fig. 7.2 Categorisation of biofuels based on their physical state and biomass Feedstock

Biofuels are renewable fuels derived from biomass through thermo-, physio-, or biochemical reactions. Depending on the feedstock used, three generations of biofuels are identified in the literature (Fig. 7.2). "First-generation" biofuels are based on food crops, such as wheat, barley, rapeseed, sugarcane, and corn, and thus have direct competition with food and feed. These raw materials have been the subject of much debate worldwide as their use may lead to food shortages. For this reason, the use of “second-generation” or “advanced” biofuels, based on non-food crops and lignocellulosic material that will have reduced or no food competition, increased. To avoid any competition with food or feed a “third-generation” of biofuels based on algae or other microorganisms has been the focus of research as those resources will have only little land requirements (Loeffler et al. 2018; Zörb et al. 2018; Kumar et al. 2019a). Nowadays, research on “fourth-generation” which consists of combining genetically engineered feedstock with genomically synthesised microorganisms, is also being carried out to increase the efficiency of biofuel production from biomass (Mansoori et al. 2021).

Biofuels are classified into solid, liquid, and gaseous energy forms according to their physical properties (Fig. 7.2).

7.2.1.1 Solid Biomass

The use of solid biomass to derive energy is known as solid biofuels and has been classified into four well-known types of solid biofuels.

1. **Firewood:** Wood is the ancient biofuel source being used for thousands of years for the production of heat and light and other domestic purposes. Before its use as firewood, the wood needed to be dried with its moisture content reduced to about 10–25%. Compared to green firewood, dried wood burns more quickly and efficiently. But, the burning of firewood or fuelwood also produces hazardous greenhouse gases, which cause a negative impact on the environment.
2. **Woodchips:** wood chips are a processed form of firewood that is easier to handle and faster to burn. It is mostly used in areas where mechanical forestry equipment is available.
3. **Wood pellets:** In the wood pellets, the wood is converted into sawdust and processed at high temperatures. At high pressure, the temperature rises, and the lignin melts and glues the sawdust into pellets. Afterwards, the pellets are broken into pieces of 2–3 cm in length. Nowadays, wood pellets made from seed husk, formed after oil extraction, have a high demand for animal feed.
4. **Charcoal:** Charcoal has a much higher energy content compared to the other forms of wood biofuels. Charcoal is produced after the wood materials are heated below 400 °C temperature in the absence of air.

7.2.1.2 Liquid Biofuels

Liquid biofuels are transport fuels obtained from biomass. They are refined products of biomass feedstock. Bioalcohols (bioethanol and biomethanol) and biodiesel formed from bio-oil are examples of liquid biofuels.

1. **Bioethanol:** Bioethanol is produced by direct and indirect fermentation processes. In direct fermentation, ethanol is made from simple sugars obtained from either first-generation (wheat, beetroot, corn, and sugar cane) or second-generation biofuels (Stover, straw, stem, and stalks) sources (Elshahed 2010). In first-generation biofuel, extraction of sugar syrup is relatively simple. Hence,

microbial and enzymatic treatments are not required for pre-treatments. Sugar syrup is converted into ethanol using genetically engineered yeast and bacterial strains. Due to increasing debates on fuel Vs food during the past few years, various countries have moved from the first-generation biofuel to second-generation biofuels. In the second-generation biofuels, the lignocellulolytic microbial (bacteria and fungi) strains are used for the initial hydrolysis of complex sugars (polysaccharides) into simple sugars (oligo, di, or monosaccharides). These simple sugars are then subjected to microbial fermentation for bioethanol production (Lau and Dale 2009). Indirect fermentation is a promising approach for ethanol production. In this process, plant material is converted into syngas by pyrolysis. Syngas contains CO, CO_2, and hydrogen (H_2), which are then transformed into ethanol by anaerobic acetogenic bacteria (Tanner 2008).

2. **Biomethanol:** The preparation of biomethanol involves the gasification of carbohydrates from biomass and their partial oxidation. Compared to producing methanol from fossil fuels, the production of biomethoanol from biomass is expensive. Hence only a tiny percentage of biomethanol is produced from biomass. Methanol is used as fuel, fuel additive, and an important base chemical for industries. Low flammability, high performance, and low emission of pollution are the advantages of using biomethanol (Pirola et al. 2018).
3. **Biodiesel:** Biodiesel consists of alkyl (C1-C4) esters of long-chain fatty acids. The production of biodiesel involves the transesterification of biological lipids (raw plant oil, animal fat, and waste oil) in the presence of methanol. A base is also used during the transesterification of lipids to form a liquid fuel. Biodiesel is used either as a substitute or as an additive for diesel. The lipids from photosynthetic algae are processed to produce biodiesel. This promising process is also popular as an eco-friendly and carbon-neutral process of biofuel production due to converting greenhouse gas CO_2 into biodiesel using sunlight. The process also has high carbon-fixation efficiency because the growth rate of microalgae is much faster than oil crops, and the extraction of oil exceeds about 80% of the dry biomass (Chisti 2007).
4. **Bio-oil:** Bio-oil is a pyrolysis product and comes along with other products such as biochar and syngas. Modification and optimising the conditions during pyrolysis can increase the amount of bio-oil. Bio-oil is a mixture of many compounds such as acids, alcohols, aldehydes, esters, ketones, sugars, alkenes, aromatic and nitrogen compounds, and many others. However, bio-oil is difficult to burn due to excess moisture. Moreover, it is also volatile, corrosive, and adhesive.

In recent studies, algae with high lipid profiles (e.g. arachidonic, eicosapentaenoic, and docosahexaenoic acids) have been used for the production of bio-oils. The major challenge in this process includes the development of low-cost extraction methods (Baskar et al. 2019).

7.2.1.3 Gaseous Biofuels

Gas and its products are extensively used for cooking, heating, transportation and electricity generation as they are very flexible in their use. Biogas, biohydrogen, and syngas are some types of gaseous biofuels.

1. **Biogas:** The anaerobic digestion of organic waste, sewage sludge, animal wastes, or energy crops using microorganisms leads to a mixture of gases known as biogas. This process works in four stages: hydrolysis, acidogenesis, acetogenesis, and methanogenesis. In the hydrolysis step, the microorganism ferment complex biomass into long-chain and short-chain volatile fatty acids. The product formed in acidogenesis is utilised by acetogenic bacteria to produce H_2, CO_2 and acetate, which is finally used up by methanogens to produce methane (Borja and Rincón 2017).
   Biogas is composed of approximately 60–65% methane (CH_4) and 30–35% carbon dioxide (CO_2). However, the exact composition depends upon the feed material. Other gases H_2, hydrogen sulphide, and water vapours are also in lower amounts. Following the purification and concentration of biogas, it can be combined with heat and power units to generate heat and electricity. In addition, biogas can be injected into the gas grid or liquefied using pressure for fuel purposes.
2. **Biohydrogen:** H_2 is an ecologically pure biofuel because it does not release any harmful gases upon combustion. Pyrolysis of biomass, such as waste, crop straw, municipal solid waste, crop grain residue, pulp waste, or manure slurry, results in the synthesis of biohydrogen. H_2 is also formed as a final product in the fermentation process by the H_2ase enzymes in microorganisms (Vignais and Billoud 2007).
   In photobiological H_2 production, photosynthetic microbes such as *Cyanobacteria* and green algae are also well known to produce low-cost H_2. These photosynthetic microbes split the water molecules into electrons and oxygen. The hydrogenase enzyme can convert the produced electron into H_2 (Prince and Kheshgi 2005).
3. **Syngas:** Synthesis gas (syngas) is produced by pyrolysis or gasification of plant biomass or biobased gases. Carbon monoxide (CO) and H_2 are the main components of syngas, accompanied by CO_2, CH_4, hydrogen sulphide, water vapours, etc., depending on the biomass composition. Power to Gas technologies such as catalytic and biological methanation is becoming increasingly important (Martín 2016).

The syngas can be injected into the grid, liquified for fuel, and used to produce other fuels such as diesel. Moreover, syngas is the leading source for producing various chemicals such as ethanol, methanol and ethane. The H_2 separated from syngas is used in fuel cells for electricity generation (Wu and Tu 2016).

7.3 Substrates for Biofuel Production

Due to the shortage of fossil fuels and environmental issues, renewable, environment-friendly fuels are becoming more important nowadays. Fuel crisis and treatment and proper usage of organic wastes are among the significant global challenges. Both challenges can be addressed by using organic wastes for biofuel production. Based on their origin, organic wastes can be classified into agricultural/ forestry and non-agricultural/forestry wastes (Table 7.1). Agricultural wastes (by-products, co-products) are usually defined as non-food or feed plant or animal residues generated from either harvesting crops/trees or rearing animals. Compared to agricultural waste, the non-agricultural organic wastes (biowastes) include all organic wastes from the domestic, food, municipal, and industrial sectors.

All these wastes can generally be used for the production of biofuels. Depending on their composition (content of carbohydrates, proteins, lipids, cellulose, hemicellulose, lignin) and their dry matter they can be used to produce specific kinds of biofuels.

7.3.1 Biofuels from Different Types of Biomass

Wastes with high content of dry matter like forestry residues and by-products from forest, straw, bagasse, solid animal waste, and other vegetal materials can be used to produce solid biofuels. These solid biofuels can substitute common wood-based biofuels. A homogenous fraction is a good choice for producing liquid biofuels from biowastes. Lipid-rich wastes from restaurants, catering, retail premises and food processing plants are suitable materials for producing liquid biodiesel. Waste biomass rich in starch, sugar, and lignocellulosic material is a good choice for the production of bioethanol and biomethanol (Yadav et al. 2020). However, this method is still in the infancy stage of development (Hirschnitz-Garbers and Gosens 2015). The production of bio-oils by pyrolysis of wastes is currently under optimisation at an industrial scale. Once optimised, this method can also use different biowastes to produce bio-oils (Karmee 2016). Gaseous biofuels (biohydrogen and syngas) are also released by pyrolysis or gasification of wastes.

Table 7.1 Classification of organic wastes (modified—according to Pimiä et al. 2014)

Types	Organic wastes
Agricultural/forestry wastes	Forestry and agricultural residues, Manure
Non-agricultural/forestry wastes	food and kitchen waste
• Food waste	Household waste, Restaurant waste, Catering waste
	Retail premises waste, waste from food processing plants
• Industrial waste	Nature textiles, paper, processed wood
• Municipal waste	Garbage, Biodegradable garden and park waste, sewage sludge

Unlike bio-oil production from waste, the production of biogas from waste is already an optimised method being practised worldwide. Biogas is another gaseous biofuel, produced utilising a variety of putrescible organic wastes, such as agricultural residues, manure, food wastes, industrial wastes, sewage, and the organic fraction of municipal solid waste (MSW). The high lignin and lignocellulosic contents lower the specific biogas yield (De Simio et al. 2008).

7.3.2 Pre-treatment of Waste Prior to Microbial Treatment

Biofuel usually starts with a preliminary feedstock preparation step involving cleaning and size reduction by milling, grinding, or chopping. All these steps consume a large amount of energy. Subsequently, the process follows four major steps: (1) pre-treatment, which involves degradation of the complex lignocellulosic network into smaller units, (2) Hydrolysis/saccharification to obtain fermentable sugars, (3) fermentation to convert sugars into ethanol, and (4) Purification (recovery and dehydration) to obtained good quality ethanol (Fig. 7.3).

7.3.2.1 Pre-treatment

Naturally occurring forms (crystalline structure) of cellulose have high resistance to hydrolysis. The presence of lignin also limits enzymatic hydrolysis by adsorption of enzymes. Pre-treatment performs de-lignification, degradation of hemicelluloses and reduction in cellulose content. Pre-treatments can be physical (e.g. milling, grinding, and microwave), chemical (acid, alkali, ozonolysis, organosolv, and ionic liquids), physicochemical (steam explosion, ammonia fibre explosion, CO_2 explosion, liquid

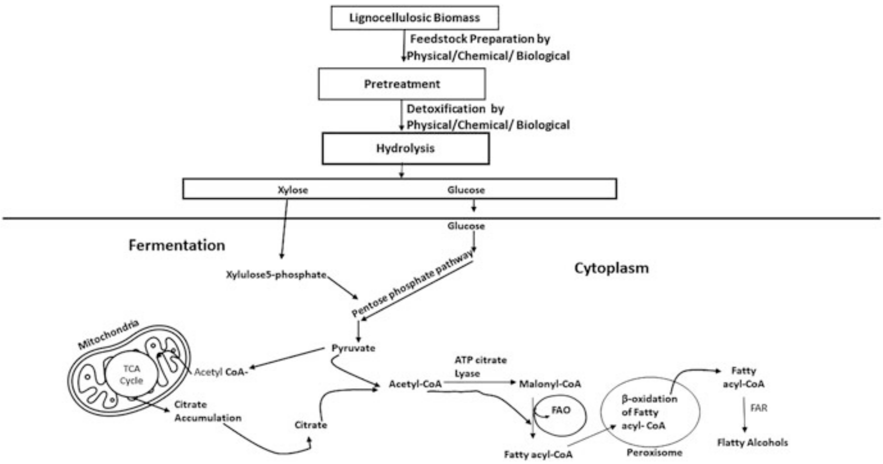

Fig. 7.3 Biochemical pathway of biofuel production from waste

hot water, and wet oxidation), or biological. During pre-treatment, lignocellulosic biomass several compounds such as (1) furfural and HMF (5-hydroxymethyl-2-furaldehyde), originating from the degradation of hexoses and pentoses, (2) acetic acid, originating from hemicelluloses, and (3) phenolic compounds originating from lignin are generated. These compounds are toxic to microorganisms, inhibit their growth, and extend the lag phase. So, several detoxification technologies are used to remove these toxic compounds.

7.3.2.2 Hydrolysis/Saccharification

It is a crucial step in which sulphuric acid or hydrochloric acid or enzymes are used to convert cellulose and hemicelluloses into their monomers, i.e. fermentable sugars using the process of acid or enzymatic hydrolysis at low temperature, followed by microbial fermentation for the production of biofuel (Branco-Vieira et al. 2018).

7.3.2.3 Fermentation

Different enzymes like xylanases, laccases, chitinases, cellulases, and proteases play a dedicated role in bioconversion. For example, xylan and cellulose as substrates are used for biofuel production. Bioconversion of the sugars to bioethanol occurs through fermentation, involving microorganisms (Adegboye et al. 2021; Soni et al. 2020).

7.3.2.4 Purification

Lastly, the product obtained needs to undergo the process of purification and distillation, which involves separating the bioethanol, in pure form, from the fermentation broth. The quantity of bioethanol obtained from the fermentation process mainly depends on the amount of sugar produced during pre-treatment and hydrolysis/saccharification. The total yield of bioethanol can be measured in terms of the volume of ethanol produced per dry weight of raw material (Adegboye et al. 2021).

7.4 Biological Agent in Biofuel Production from Waste

7.4.1 Bacteria

Microorganisms are considered alternative sources for the production of biofuels. Bacteria have significant advantages over higher plants and microalgae for synthesising intracellular as well as extracellular fatty acids to produce environment-friendly fuel oil (Kumar et al. 2020). Fast-growing bacteria can potentially use a wide range of feedstocks for biodiesel production. Bacteria effectively use agricultural by-products for their growth and utilise sugar and proteins pre-set in waste materials (Mihajlovski et al. 2020). Some of the well-known potential biofuel-producing strains of bacteria have been summarised in Table 7.2. Activated sludge contains a microbial population of heterotrophic bacteria responsible for wastewater treatment. These bacteria use the organic compounds in wastewater for their growth

Table 7.2 Microorganisms in biofuel production

Organisms	Biofuel type	References
Acinetobacter calcoaceticus	Lipid	Choi et al. (2014), Moshtagh et al. (2021)
Alkalibaculum bacchi	Ethanol	Allen et al. (2010), He et al. (2022)
Bacillus sp. (*B. mycoides*, *B. amyloliquefaciens*, *B. pumilus*)	Butanol	Kanno et al. (2013), Shabbir et al. (2022)
Clostridium acetobutylicum	Acetone, butanol, and ethanol	Ennis et al. (1986), He et al. (2022)
Clostridium beijerinckii	Isopropanol, butanol, and ethanol	Hettinga et al. (2009), Comwien et al. (2015), He et al. (2022)
Clostridium carboxidivorans	Ethanol, butanol	Fernández-Naveira et al. (2016), He et al. (2022)
Clostridium phytofermentans	Ethanol	He et al. (2022)
Clostridium ragsdalei	Ethanol	Devarapalli et al. (2017), He et al. (2022)
Clostridium thermocellum	Ethanol	Ng et al. (1981), He et al. (2022)
Costridium saccharoperbutylacetonicum	Butanol	Shukor et al. (2014), He et al. (2022)
Cryptococcus curvatus	Lipids	Yu et al. (2011), Kamal et al. (2022)
E. coli	Ethanol, 1-Propanol, 1-pentanol isobutanol, 1-butanol	Asghari et al. (1996), Zhang et al. (2008), Ku et al. (2022)
Lactobacillus brevis	Butanol	Russmayer et al. (2019), Esquivel-Hernández et al. (2022)
Lipomycesstarkeyi	Lipids	Yu et al. (2011), Zhang et al. (2022)
Pseudomonas putida	Butanol	Sahoo et al. (2019), Sarwar et al. (2022)
Rhodococcus opacus	Lipid	Le et al. (2017), Nair and Sivakumar (2022)
Rhodosporidium Toruloides	Lipids (Glucose and xylose)	Xie et al. (2012), Gao et al. (2022)
S. cerevisiae	Ethanol	Sharma et al. (2022)
S. stipitis	Ethanol	da Silva et al. (2022)
Zymomonas mobilis	Ethanol	Li et al. (2022)

and store the organic material in the form of lipid droplets. Oleaginous bacterial species belonging to the order Actinomycetales (*Mycobacterium*, *Streptomyces*, *Nocardia,* and *Rhodococcus*) can accumulate lipid up to 20% or more of their biomass (Cea et al. 2015). *Acidothermus*, *Bacillus*, *Clostridium*, *Pseudomonas,* and *Rhodothermus* degrade cellulose. A wide assortment of Gram-positive and

Gram-negative cellulose-degrading bacterial species includes *Clostridium thermocellum*, *Streptomyces* sp., *Ruminococcus* sp., *Pseudomonas* sp., *Cellulomonas* sp., *Bacillus* sp., *Serratia* sp., *Proteus* sp., *Staphylococcus* sp., and *Bacillus subtilis* (Kashyap et al. 2019; Khedr et al. 2019). *Geobacillus* is an obligate thermophilic bacteria which can generate and enhance the productivity of important bioenergy sources such as ethanol, isobutanol, 2,3-butanediol, biodiesel, and biogas at the temperature range of 35–75 °C (Novik et al. 2018).

Biogas is an effective source of renewable energy. Anaerobic microorganisms produce biogas by organic decomposition of domestic and agricultural waste as a substrate. CH_4 is the main combustible element of biogas, forming 50–75% volume of biogas. Remaining 25–50% volumes consists of non-combustible gaseous elements, such as CO_2, N_2 (<1%), O_2 (0–1%), and nitrogen siloxanes (0–0.02%), halogenated hydrocarbons (<0.6%), CO <0.6%, hydrogen sulfide (0.005–2%), and water vapours (5–10%) (Wellinger and Lindberg 1999). *Thermovirga*, *Soehngenia* and *Actinomyces* are H group-containing bacteria that have more capacity to generate CH_4 than the black group. These microbial communities (black and H group) have been categorised with the help of Illumina sequencing. Archaeal species like *Methanosaeta*, *Methanolinea*, *Ethanospirillum,* and *Methanoculleus* are reported in both groups (Wang et al. 2017). Bioaugmentation strategies for enhancing biogas production plays a crucial role during the anaerobic degradation of cow manure. These bacterial strains include *Rikenellaceae*, *Clostridiaceae*, *Porphyromonadaceae*, *Bacteroidaceae,* and *Ruminococcaceae*. *Flavefaciens* and *Ruminococcus albus* showed CH_4 production at 41 °C (Ozbayram et al. 2018).

Biodiesel, consisting of mono-alkyl esters, is produced by the transesterification of edible and non-edible oil/fat from plant and animal origin. The use of biodiesel over conventional fossil fuel-based diesel offers several advantages, such as less emission of greenhouse gases, other gaseous pollutants and particulate matter (Behera et al. 2019). Oleaginous bacteria *Rhodococcus opacus* produce 80% biodiesel of its cellular dry weight using wastewater from corn stover (Le et al. 2017). Moreover, *Serratia* sp., a chemolithotroph, uses municipal secondary sludge as growth media for biodiesel production. These bacteria apply several strategies for their adaptation to produce lipids, bioplastics, exopolysaccharides and fatty acids (Kumar et al. 2020).

Bioethanol is an important alternative to fossil fuels and contributes to the economy by using domestic and environmental wastes. It is a safe, efficient and non-toxic biofuel produced without any by-products (Younesi et al. 2005; Eriksson and Kjellström 2010). The organic fraction of MSW comprises 50% lignocellulose-rich material. *Zymomonas mobilis* and *Rhodococcus opacus* have the potential of producing ethanol from MSW (Dornau et al. 2020). Brigham (2019) reported that Knallgas bacteria produce different types of high-energy-density transportation fuels by utilising CO_2, H_2, and O_2. *Ralstonia eutropha* is a Knallgas bacterium, which has been genetically engineered to produce *n*-butanol, isobutanol, and terpene under chemolithoautotrophic conditions. Many extremophilic bacterial species, mainly thermophilic microorganisms, produce cellulase enzyme which increases the rates of cellulose hydrolysis. *Clostridium thermocellum*, *Thermoanaerobacter* *thermohydrosulfuricum,* and *Clostridium stercorarium* subsp. *thermolacticum* not only efficiently degrades cellulose and hemicelluloses through hydrolysis but also readily ferments the pentose and hexose sugars (Di Donato et al. 2019). Ethyl alcohol is produced using syngas fermentation, in which anaerobic microorganisms (*Clostridium ljungdahli*, *C. tetanomorpum,* and *Clostridium* strain P11) utilise accessible carbon and energy source to produce ethanol biofuels (Williams et al. 2015; Kundiyana et al. 2010).

7.4.2 Yeast/Fungi

Fungi degrade the biomass of agricultural waste through biochemical and thermo-chemical processes to produce biofuels. Biochemical conversion leads to bioethanol and biodiesel production (Maurya et al. 2020). Endophytic fungi produce compounds such as alkanes, cyclohexanes, cyclopentane, alkyl alcohols/ketones, benzenes, and polyaromatic hydrocarbons found in biodiesel (Raven et al. 2019; Kumar et al. 2023). *Rhizopus Oryzae* fungi have been demonstrated to efficiently catalyse the methanolysis of vegetable oils for biodiesel production in solvent-free systems (Nagaraj et al. 2010). Some of the fungi used for biofuel production have been presented in Table 7.3.

Filamentous fungus *Aspergillus* sp. produces biodiesel with good fuel quality (acid number, 0.40 mg KOH/g of acid; iodine value, 11 g I_2/100 g oil; density, 0.8342 g/cm^3) using corncob waste liquor (CWL) as substrates (Subhash and Mohan 2011). Moreover, *Aspergillus niger* and *Trichoderma harzianum* have been reported to perform the alkali and enzymatic hydrolysis of rice husks (Solanki et al. 2019; Abbas et al. 2022). This hydrolysed husk can be used for bioethanol production via fermentation using *Saccharomyces cerevisiae* (Ahmad et al. 2017). Similarly, the co-culture of *Aspergillus niger* and *Saccharomyces cerevisiae* produce ethanol from the rice wastewater (Hatami et al. 2015; Gujjala et al. 2019). Furthermore, Subhash and Mohan (2015) reported that *Aspergillus awamori* uses CWL, paper mill effluent (lignocellulosic wastewaters) and cellulosic waste (de-oiled algae extract, DAE) as feedstock for single cell oil (SCO) production. DAE improvises biomass production by reducing production time; however, the high feedstock cost is a major limiting factor. Oleaginous fungi are cultured with lignocellulosic materials for lipid production, which produces biofuel at a comparatively lower cost due to the abundance of low-cost feedstock, such as glycerol, sewage water, whey and molasses. Oleaginous microorganisms have multiple advantages (Zheng et al. 2012), such as (1) capacity to accumulate 80% of lipid and increase the quality of fatty acids, (2) having good lipid profiles, suitable for making high-quality biodiesel, (3) capacity to utilise monosaccharides, glycerol, acetic acid, cereal, corncob, sweet sorghum, wheat straw, orange peel, apple pomace and oil for lipid production, (4) low capital cost and low energy expenditure is required for oil production, through solid-state fermentation, and (5) ease of oil harvesting from cell broth by using simple filtration after pellet formation, and reduction in the viscosity of the fermentation broth to

Table 7.3 Role of important microbes in fuel production from different feed stocks

Organism	Biofuel	Feedstock	References
Trichoderma asperellum	Biohydrogen	Sweet sorghum	Shanmugam et al. (2018)
Consortium of *T. viride* and *A. niger*	Biohydrogen	Oat straw	Zhao et al. (2019)
A. tubingensis, *Trichosporono idesspathulata*, *Candida tropicalis*, *Rhodotorula mucilaginosa*	Biodiesel	Palm empty fruit bunch	Intasit et al. (2020)
Mucor circinelloides	Biodiesel	Sugarcane bagasse, corn milling	Carvalho et al. (2018)
Penicillium citrinum	Biodiesel	*Musa balbisiana* cola peels	Bardhan et al. (2019)
Aspergillus awamori, *Aspergillus oryzae*	Biohydrogen, Bioethanol	Food waste	Han et al. (2016)
Gymnopus contrarius	Biohydrogen	Rice straw	Sheng et al. (2018)
Clostridium thermocellum	Biohydrogen	Waste date palm	Swathy et al. (2020)
Pleurotus ostreatus, *Trametes versicolor*	Biogas	Chicken manure with sawdust and wheat straw	Basinas et al. (2022)
Orpinomyces sp., *Piromyces* sp., *Anaeromyces* sp., *Neocallimastix frontalis*	Biogas	Animal manure	Yıldırım et al. (2017), Bhujbal et al. (2022)
Cladosporium sp., *Verticillium* sp.	Biogas	Feathers, biological sludgeslime	Wrońska and cybulska (2018)

improve the mixing and mass transfer performance, compared to traditional high-cost centrifugation methods.

Oleaginous yeast such as *Rhodotorula glutinis* accumulates 25% lipid of its biomass for biodiesel production from monosodium glutamate wastewater (Zheng et al. 2012). *Saccharomyces cerevisiae* can use hexose monosaccharides (glucose, mannose, and galactose) and disaccharides (sucrose and maltose) to produce bioethanol via fermentation of lignocellulosic hydrolysates (Branco et al. 2019). Yeast strains such as *Kluyveromyces fragilis*, *Candida* sp., *Rhodosporidium* sp., *Rhodotorula* sp., and *Lipomyces* sp. accumulate 70% triacylglycerols of their biomass (Subhash and Mohan 2011). Hemicellulose and lignins of plant cell walls are acetylated, which yield acetic acid after hydrolysis as an unavoidable component. Acetic acid is toxic to the fermenting microorganisms, negatively influencing sugar fermentation and, subsequently, biofuel yield. Additionally, *Trichosporon fermentans* could be used for microbial lipid production from detoxified rice straw acid hydrolysate. But the obtained lipid content was lower than glucose as the sole carbon source (Huang et al. 2012). Yeast, *Saccharomyces cerevisiae*, is widely used for the production of ethanol from corn and sugarcane, but it cannot metabolise xylose. But *Scheffersomyces stipitis* can convert xylose to xylulose by expression of nicotinamide adenine dinucleotide phosphate (NAD(P)H)-linked xylose reductase (XR) and nicotinamide adenine dinucleotide (NAD)-linked xylitol dehydrogenase (XDH) genes. This xylulose can be metabolised after its phosphorylation via the pentose-phosphate pathway (Wei et al. 2013). Moreover, endophytic fungal isolates *Colletotrichum* sp., *Alternaria* sp., and *Aspergillus* sp. have the ability of lipid accumulation, as whole-cell biocatalysts, under the nutrient optimum and nutrient-stressed conditions (Subhash and Mohan 2011).

Biogas production efficiency is influenced by the type and quality of the raw materials used. Waste products from the poultry industry, agricultural crop wastes, and animal residues fulfil the requirements of good raw materials due to having a significant proportion of fats and proteins (Wrońska and Cybulska 2018). Anaerobic fungi are known to produce plant carbohydrate hydrolysing enzymes for cell wall polysaccharide decomposition. Anaerobic fungi are promising candidates for mechanical and enzymatic degradation of plant polysaccharides to improve biogas production (Dollhofer et al. 2015). Anaerobic fungus *Piromyces rhizinflata* degrades volatile fatty acid and augments the lignocellulose biomass (corn silage and cattail) as feedstock for CH_4 and H_2 production (Nkemka et al. 2015). Similarly, the fungus *Auricularia auricula-judae* is used to decay sweet chestnut (*Castanea sativa*) leaves, hay and wood to decompose cellulose, hemicelluloses and lignin for the production of biogas (Mackuľak et al. 2012).

7.4.3 Photosynthetic Microorganisms

Photosynthetic microorganisms, as a platform for biofuel production, have gained substantial recognition as an option that could significantly reduce environmental pollution by using CO_2 emitted from various sources (Machado and Atsumi 2012). These photosynthetic microorganisms directly fix CO_2 as their primary carbon source for biofuel production and replace the requirement of fermentable sugars. Algae and cyanobacteria are the pioneer and desired organisms for this strategy of biofuel production. Both these groups of organisms can grow much faster than plants, do not need arable land for their production and can be grown in submerged water (Dismukes et al. 2008). Research on algae has centred on enhancing their potential to produce large amounts of lipids pertinent to biodiesel production (Pate et al. 2011; Kumar et al. 2017). Cyanobacteria coupled with prokaryotic organisms such as *E. coli* is beneficial to both as a photosynthetic microorganism and naturally transformable host. Studies reveal that cyanobacteria have already been manipulated to produce a number of different biofuels (Dismukes et al. 2008; Machado and Atsumi 2012; Gao et al. 2016). For instance, *Synechococcus elongatus* sp. strain PCC 7942 was successfully manipulated for ethanol production via the external addition of enzymes such as pyruvate decarboxylase and alcohol dehydrogenase, redirecting the carbon from pyruvate (Deng and Coleman 1999). Continuous research works have significantly improved the production of ethanol using cyanobacteria (Gao et al. 2012, 2016). Further researches are being conducted worldwide on other photosynthetic microorganisms to improve and strengthen the ability of biofuel production from waste.

7.5 Waste Product Impact on Climate

Wastes are all the by-products released from industries, households, or other sources humans cannot use further. Waste management is a more significant challenge for both the small and big cities of developing countries. Urbanisation and increasing population are the major issues responsible for increasing the burden of waste. According to the Global Waste Management Outlook 2015 (GWMO), 2.0 billion tonnes/year of waste is produced by MSW and 7–10 billion tonnes from households, commerce, industries and construction site (Everett 2012; Al-Dhrub et al. 2017). These wastes may be in solid, liquid or gaseous forms whose disposal improperly leads to negative consequences on the health of humans, animals and the environment (Misra and Pandey 2005). Improper and uncontrolled disposal generates heavy metal pollution in the water, air, and soil. Open burning causes the release of CO_2, SO and other air pollutants in the atmosphere. The release of waste in the water bodies also affects the aquatic ecosystems enhancing eutrophication (Ferronato and Torretta 2019). In the present climate change scenario, the melting of glaciers, increasing temperatures, seasonal variations, the emergence of various pathogens, and adverse consequences on agricultural production are the major threats to human society. Further, these wastes and their mismanagement will boost the future climate change rate. Nowadays, the conversion of different waste materials to generate energy and its use for societal welfare along with a significant positive impact on the environment is one of the top priorities (Tabasová et al. 2012; Kumar et al. 2019b). These strategies are required to control the rate of climate change and mitigate its adverse consequences.

Due to recent anthropogenic activities, the degree and amount of waste are increasing. The considerable increase in a waste generation began due to population explosion and industrialisation (Wilson 2007; Pikoń and Czop 2014). It has been reported that approximately 1.3 billion tonnes of MSW is generated per year, and it could rise to approximately 2.2 billion tonnes/year by the end of 2025 (Hoornweg and Bhada-Tata 2012). There are various waste management techniques through which the wastes can be transformed for the production of manures for agriculture purposes, eco-friendly energy sources, and pollution reduction (Widmer et al. 2005; Aljaradin and Persson 2012).

7.5.1 Impacts of Waste Disposal on the Environment

The waste material could be in solid, liquid or gaseous form and biodegradable or Non-biodegradable in nature. Food production through agriculture and its consumption is one of the main factors related to environmental impacts in the world. Food production involves using resources such as fuels, land, water and raw materials linked to economic and environmental impacts. Most food packaging materials are made up of non-biodegradable plastics which are obstinate towards microbial disintegration and hence do not meet the requirements of compost forming (Pikoń and Czop 2014). Disposal of food wastes into water bodies affects the aquatic ecosystem, causing eutrophication and algal blooms due to increased nutrient concentration in water bodies (Scherhaufer et al. 2018).

In developing nations, there is a major problem with management of solid waste (sewage and industrial sludge) due to several constraints; hence, landfilling with waste products in low-level areas is preferable. Sewage contains a large number of toxic substances which are harmful to human and animal health, as well as to the environment. MSWs majorly hold solid matter and are subject to landfilling for its management. The degradation of MSWs in landfills leads to the formation of different hazardous gases. The level of CO_2, which usually remains high, regularly drops as the CH_4 concentration builds up if the degradation procedure is shifted from aerobic to anaerobic conditions. Other gases, including H_2, nitrogen, etc., are produced in minor amounts during the degradation process. Burning solid waste at the landfilling site produces toxic gases that pollute the air, causing respiratory problems. These gases contribute to global warming and climate change. Solid waste undergoes a sequence of complex biochemical and physical processes, leading to the production of leachate and gaseous emissions. When leachates reach the water resources, they pollute surface water and groundwater (Aljaradin and Persson 2012).

7.5.2 Non-biodegradable Wastes

Hazardous and non-biodegradable solid wastes, which enter from the municipal waste directly disposed-off in the environment, play a significant role in environmental degradation. The majority of plastics are composed of polyaromatic hydrocarbon compounds and produce greenhouse gases, which cause a negative impact on the environment. Plastic restricts the water absorption in the soil due to seized soil capillaries and simultaneously affects the microbial diversity, water holding capacity, and loss of moisture content in the soil. More plastic waste in the soil environment triggers the process of soil infertility (Andreeßen and Steinbüchel 2019). Now a day's, the world is facing plastic waste pollution in the marine ecosystem also. Rivers are the indirect key carrier of plastic waste. Plastic waste harms many aquatic animals, and plastic pollution also decreases the aesthetic value of any water body.

The waste of glass industries is another unremarkable waste posing many challenges due to the high greenhouse gas emissions, rigorous energy use, and the intensive use of the Earth's natural resources. Discarding the glass waste in landfills is not offering environment-friendly management due to the non-biodegradable nature of glass waste and is triggering severe environmental soil pollution (Jani and Hogland 2014). Apart from municipal or industrial waste, E-waste comprises harmful materials that need proper management and recycling approaches to avoid environmental pollution (Gabra et al. 2019). E-waste is chemically and physically different from other forms of waste. The chemical composition of E-waste differs depending on the age and quality of the discarded items. Most E-wastes contain a mixture of metals, particularly Cu, Al, and Fe, which are used in several kinds of plastics and ceramics. Discarded personal computers, laptops, washing machines, refrigerators and electrical wires are comprised of metal, plastics, electronic components and glass. Disposing of all this E-waste in the environment is polluting the water, soil, and air (Robinson 2009).

7.6 Challenges in Biofuels Production from Waste

World socio-economic developments are mainly progressed by energy. Presently, the world's fuel demand of around 75% is compensated by non-renewable sources like petroleum and its derived fuel. As per the International Energy Report 2014, the global energy demand is expected to elevate by 37% by 2040 (Joshi et al. 2017). Therefore, research is being carried out in different parts of the world with a special focus on renewable sources to meet anticipatory growing energy demand. Hence, biofuels from waste biomasses could be a probable source to meet the global anticipatory energy demand.

There are several procedures and technologies by which renewable resources can generate biofuels (Joshi et al. 2017). The biofuels could be produced from enriched biochemicals produced by either microbiological agents such as bacteria, fungi, and microalgae or animals (Rodionova et al. 2017; Kumar and Banerjee 2019). For the last few decades, agriculture production has increased several folds. Simultaneously, food and agricultural waste also increased proportionally; hence, this waste production has been known to be the potential source of biofuels. However, algal biomass has recently been known to be a potential bioresource for producing different types of biofuels (Dragone et al. 2010; Rodionova et al. 2017).

There are several prospects for the production of biofuels from wastes product that have been well recognised and exploited. Among them, biofuels by cyanobacteria or microalgae have been highly acknowledged (Demirbas et al. 2016; Heimann 2016; Rodionova et al. 2017; Chintagunta et al. 2020). Scott et al. (2008) have reported several benefits of using microalgae for biofuel production owing to high productivity compared to other bioresources. Besides the benefits of microalgae-based biofuels production, several challenges are still to be considered for commercial production of biofuels, such as ease and continuous accessibility of waste products, pre-treatment and processing of waste products that could be subjected to biofuel production. Appropriate selection of bioreactors for large-scale production of microalgal biomass, maintenance of contamination-free medium during the reaction, selection of superior microalgae strains and most important continuous supply of sterile medium as well as CO_2 for microalgae growth are the other aspects that need optimisations (Scott et al. 2008).

Food waste is the anon consumable source of lipids, carbohydrates, amino acids and phosphates. On average, food waste materials contain around 30% lipid and 50% carbohydrate (Pleissner et al. 2014, 2016). The waste food can be hydrolysed enzymatically, and the food wastes abundant in carbohydrates and lipids can be subjected to bio-ethanol and biodiesel production, respectively. In the past few decades, focused research on the application of food wastes for producing biofuels has been going on globally. Sulaiman (2014) proposed a halal biorefinery to produce biofuels in Malaysia. Chinese Academy of Sciences reported using food waste to produce hydrolysates for bioethanol production (Yan et al. 2011; Karmee and Lin 2014). In Europe, potato peel has been utilised to produce bioethanol using environmentally benign biocatalytic methods with the involvement of liquefaction, saccharification and fermentation of peel (Arapoglou et al. 2010; Yan et al. 2011; Wang et al. 2017). The prime drawback of pre-treatment methods of waste products included the production of specific inhibitors for microbes that may interfere with the processing and production of biofuels. These inhibitors are formic acid, acetic acid, phenolic compounds, furan aldehydes, ionic lipids, and levulinic acid (Wang et al. 2018; Zhang et al. 2016).

Recent economics estimates that the costs of biofuel production from waste are 2–3 folds more expensive than petroleum fuels on an energy-equivalence basis (Lynch et al. 2016; Bušić et al. 2018). To lower the production cost of biofuel, several challenges are to be taken into consideration while converting waste biomass to biofuels, such as feedstock production, feedstock logistics, development of energy-efficient technologies (pre-treatment, enzyme hydrolysis, and microbial fermentation), separation of by-products (lignin and hemicelluloses), product development, the establishment of biofuel and biochemical standards, biofuel distribution and environmental impact minimisation. Some of the major drawbacks of pre-treatment procedures include the generation of by-products that works as inhibitors for microbial growth and fermentation. These compounds are formic acid, acetic acid, and levulinic acid (Wang et al. 2018; Zhang et al. 2016). The acetic acid in growing media potentially reduces the specific growth rate and biomass yield of *Saccharomyces cerevisiae* during ethanol production waste biomass (Pampulha and Loureiro-Dias 2000; Wang et al. 2018).

Similarly, phenolic compounds, furan aldehydes and ionic lipids also act as inhibitors to *S. cerevisiae* by decreasing specific cell growth rate and ethanol yield (Lin et al. 2015; Banerjee et al. 2019). All these constraints for biofuel production from wastes require high skill in agronomy, biomass logistics, biomass conversion, process engineering, chemistry, conversion technology, genetic engineering, microbial fermentation, economics, and environmental science (Rai et al. 2020; Kumaraswamy and Kashyap 2021). It is challenging to produce biofuel from waste and economically expensive over fossil fuel. However, developing recombinant strains through genetic engineering with high commercial potential, redefining effective pre-treatment processes, and increased access to waste bioresources could be a promising strategy for sustainable biofuel production.

7.7 Conclusion and Future Prospects

Presently, developed and developing nations are encountering several challenges pertinent to climate change, depletion of natural resources, environmental sustainability and energy security, and all of these directly or indirectly affect the environment. Hence, biofuels are supposed to be the most important to alleviate such energy crises sustainably. Furthermore, several biofuels of various classes could be produced from available indigenous resources and waste products generated from agriculture and food processing. Biomass generated as waste after processing agriculture and food is a potential feedstock for biofuel production. These biomasses are potentially converted into several biofuel products through the application of different microbes of the different genera (bacteria, fungi, and photosynthetic microbes). However, biofuel productions from waste products also have several constraints that must be overcome with an integrated application of technological advancement pertinent to strain improvement, adoption of improved protocol for pre and post-processing of biomasses, and control of microbial inhibitors to improve the yield and quality of biofuels. A combination of all these approaches and further researches in the area are expected to provide remedies for the existing energy crisis due to the depletion of non-renewable sources.

Acknowledgement The corresponding author expresses sincere gratitude to the Director, ICAR-Indian Institute of Seed Science, Mau, Uttar Pradesh, Director, MACS' Agharkar Research Institute, Pune, Maharashtra, for providing the working platform. Uma Chaurasiya thanks the University Grant Commission, New Delhi, India, for the award of the Junior Research Fellowship.

Conflicts of Interest The authors declare no conflict of interest.

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