# Chapter 3 Biological Treatment of Agro-Industrial Waste



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Abstract Agro-industrial waste is mostly composed of lignocellulosic biomass, which is inexpensive, renewable, and abundant. It provides a unique natural resource for cost-effective bioenergy collection. Agro-industries generate a wide number of waste products either in the form of solid or liquid. The production of agro-industrial waste is growing worldwide and these wastes cannot be disposed of directly on the ground without any treatment, as they will cause serious environmental concerns. The problem of disposal and management of these wastes is a major issue especially in developing countries nowadays. Hence these agro-industrial waste must be treated before discharging or reuse for other purposes by effective methods. The conventional methods require the use of harsh and toxic chemicals with high processing cost and high waste management cost. In serious consideration of the worldwide economic and environmental pollution issues, there has been increasing research interest in the management of the agro-industrial waste proposing value-added green technologies. Biological treatment is seen as one of the promising green

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biotechnologies that gives less harm to the environment while balance out the ecosystem. The biological treatment utilizes microorganisms mainly from the bacterial and fungal species to cope with the issue raised and also act as bioremediation. This chapter begins with an overview of agro-industrial waste and further describes a number of biological treatments performed together with its advantages and disadvantages. This chapter finally deals with the possibility of creating a sustainable practice in industries processing agricultural products. Several suggestions and recommendations for future considerations are also thoroughly highlighted. The ultimate goal of this biological treatment chapter is to prepare the agricultural waste for a cleaner process toward a better and safer product.

Keywords Agro-industrial waste · Biological treatment · Sustainable · Green biotechnology · Bioremediation

### 3.1 Introduction

Agro-industrial activities could represent one of the primary sectors of the economy of a country, especially those that are characterized by high production rates of agricultural commodities (Pellera and Gidarakos [2017](#page-19-0)). Agricultural wastes are derived from agricultural production and products' processing, livestock breeding, and rural households while industrial wastes are residues, dust, and other wastes discharged in the process of industrial production (Mo et al. [2018\)](#page-18-0). Therefore, agroindustrial wastes refer to all by-products and residues generated from crops, livestock, forestry, and agro-based industries. Figure [3.1](#page-1-0) portrays the sources of these agro-industrial wastes as reported by Andri et al. [\(2018](#page-17-0)). Agricultural residues could be distributed into monosaccharide and disaccharide, starch, structural polysaccharide, and protein or lipid-rich sources. As for industrial residues, they could be

<span id="page-1-0"></span>

Fig. 3.1 Classifications of agro-industrial wastes according to different sources (Andri et al. [2018\)](#page-17-0)

classified as polymers rich sources, carbon compounds, and photosynthetic microorganisms.

Numerous industries produce massive amounts of wastes, consisting elevated levels of biomass, suspended solids, organic matter, effluent, and sludge which create disposal problems and cause environmental pollution in several ways (air, soil, and water) (Anastopoulos et al. [2017](#page-17-1)). Common agro-industrial biomass wastes include rice husk, sugarcane bagasse, wheat straw, spent coffee and tea, oil palm wastes, peanut hull, fruits and vegetable peels, seeds, and others. Meanwhile, relevant effluents of agro-industrial activities are wastewaters deposited from mills, winery, and starch industries as well as slaughterhouses. These wastes could be further categorized into solids, liquid, and mixed forms. Table [3.1](#page-3-0) demonstrates the agro-industrial wastes from diverse industries according to the respective types.

Most of the agro-industrial wastes are untreated and underutilized, for which majority reports it disposed of either by burning, dumping, or inappropriate landfilling (Sadh et al. [2018](#page-19-1)). These untreated wastes lead to the release of greenhouse gases that eventually resulted in environmental impact and climate change. The generated wastes might not only be biodegradable in nature but also rich in nutrient components such as carbohydrates, proteins, fibers, and minerals depending upon their sources (Andri et al. [2018;](#page-17-0) Panesar et al. [2015\)](#page-19-2). Despite being abundantly found and possessing nutritive values, however, the recalcitrant properties of these compounds impede the utilization of the fermentable components in the wastes. In addition, these wastes might also contain organic and/or inorganic (heavy metals and metalloids) pollutants and pathogens that render them unsuitable for further use (Markou et al. [2018\)](#page-18-1). Effective management/treatment is thus necessary to curb these arising problems. Instead of "wastes," these residues should be considered as "raw material" for other industrial processes. The presence of nutrients such as sugars, minerals, and proteins in the agro-industrial wastes offers suitable conditions for the prolific growth of microorganisms. The microorganisms could reuse the waste as raw materials or substrate for their growth via fermentation processes and subsequently produce an array of significant beneficial compounds. The utilization of these agro-industrial wastes would not only contribute to the recycling of waste but also alleviate the production cost and reduce pollution to a great degree (Sadh et al. [2018](#page-19-1)).

### 3.2 Biological Treatment

Biological treatment is any kind of treatment, which involves the use of any microorganisms or the enzymes secreted from the particular microorganisms. Biological treatment contradicts conventional treatment which uses harsh and toxic chemicals for their treatment. Biological treatment is more specific and selective in nature and should be able to reduce the loss in carbohydrates significantly (Kusi et al. [2018\)](#page-18-2). The best biological treatment is where it can degrade most of the unwanted compounds while preserving most of the valuable compositions within a short span

<b>Types</b>	Industries	Sources	References
Solid	Palm oil milling	Empty fruit bunch (EFB), oil palm frond (OPF), palm press fiber (PPF), decanter cake (DC), and oil palm trunk (OPT)	Mamimin et al. (2019)
Liquid	Palm oil industry	Palm oil mill effluent (POME)	Jayakumar et al. (2017)
Solid	Coconut	Coir dust/pith, shell, and copra cake/meal	Go et al. (2019)
Solid	Paddy rice	Husk, bran	Go et al. (2019)
Solid	Pineapple canning	Core and peel	Sanguanchaipaiwong and Leksawasdi (2018)
Solid	Pineapple juice industry	Pomace along with peels, crown, and core	Banerjee et al. (2018)
Liquid	Pineapple juice industry	Liquid effluents	
Solid	Wine production; pro- duced during the crushing, pressing, and draining stages	Grape marc; mostly com- posed of stalks, skins, and seeds	Pellera and Gidarakos (2017)
Solid	Olive oil extraction	Olive pomace; olive pulp, peel, pieces of pit and an oil content	Carlini et al. (2015), Batuecas et al. (2019)
Mixture; liq- uid and solid	Olive oil manufacturing	Olive wet husk, two phases olive mill waste (TPOMW), or alperujo	Navas et al. (2015), Hernández et al. (2019)
Liquid	Olive oil manufacturing	Olive mill wastewater	Batuecas et al. (2019)
Liquid; high amount of total solids and starch	Potato processing	Potato processing wastewater	Singh et al. (2019)
Solid	Cotton processing	Burs, stalks, leaves, and immature cottonseed	Pellera and Gidarakos (2017)
Liquid	Cotton industry	Cotton seed oil	Tacin et al. (2019)
Solid	Sugarcane industry	Bagasse, filter cake	Saha and Ghosh (2019)
Solid	Sugarcane juice filtration	Press mud	Anastopoulos et al. (2017)
Liquid	Sugar manufacturing	Molasses	
Liquid	Sugar manufacturing	Sugar mill wastewater	
Solid	Corn milling	Corn bran (wet), fiber (dry)	Go et al. (2019)
Solid	Soybean oil extraction	Soybean meal	Gerliani et al. (2019)
Liquid	Dairy industry	Milk whey	Martínez-Ruano et al. (2019)
Liquid	Poultry slaughterhouses	Blood and wastewater washed off from poultry	Latifi et al. (2019)

<span id="page-3-0"></span>Table 3.1 List of industries producing agro-industrial wastes

of time. Nowadays, biological treatment is getting more attraction from around the globe, especially from the industries as they have the potential to reduce the wastewater treatment cost and more environmental-friendly in nature.

The increasing volume of agro-industrial waste has become a major problem for the agro-based industries. They are searching for a better solution to solve the issue as the waste tends to spoil easily due to the high moisture content. Moreover, agrowaste is an attractive alternative source for renewable energy without competing with the food sources (Yadav et al. [2019a](#page-20-1)). Asia region statistically produces the largest amount of rice and wheat straw while America generally produces the largest amount of corn straw and sugarcane bagasse (Kusi et al. [2018\)](#page-18-2). However, the recalcitrance lignin structure of agro-wastes impedes its potential to be utilized directly from the source. They have to undergo a certain treatment process prior to their viability of producing valuable products. This treatment is meant to open up the rigid structure of the cell walls and increase the accessibility of the cellulose and hemicellulose by the enzymes.

There are many types of biological treatment currently been studied and mostly utilizes bacteria and fungus or yeast to treat the agro-waste. Extensive studies have and still ongoing toward achieving the best biological treatment for solving the abundant agro-industrial waste issues while producing valuable products. However, due to the complex and complicated structure of these agro-wastes, it is a real challenge and quite impossible to obtain a single optimized biological treatment for all types of agro-wastes. Even the same type of agro-waste may result in different findings from the same treatment due to the inconsistency in the chemical composition. Factors such as the harvesting age, type of soil used, weather condition, parts of the plant taken, and many more will contribute to the difference in composition and structure. Additionally, improvement of the biological treatment has been carried out by metabolic engineering approach. This approach applies the combination of knowledge between synthetic biology, systems biology, and "omics" which consists of genomics, proteomics, transcriptomics, and metabolomics (Cristobal-Sarramian and Atzmüller [2018\)](#page-17-9).

### 3.2.1 Fungal Treatment

Fungal treatment is one of widely used biological treatment for agro-waste to replace the conventional chemical treatment (Carrere et al. [2016](#page-17-10)). The presence of lignin degradation enzymes such as laccase, lignin peroxidase, and manganese peroxidase in the fungi cell naturally helps to overcome the agro-waste recalcitrant structure (Rouches et al. [2016](#page-19-6)). The well-known fungi that have been used for biomass treatment are the white rot-, brown rot-, and soft rot fungi (Sindhu et al. [2016\)](#page-19-7). Among these three, the white rot fungi are the mostly studied fungi have been reported to produce high enzyme activities for ligninase, laccase, manganese peroxidase, and lignin peroxidase (Pramanik and Sahu [2017](#page-19-8)). These three types of fungi have their own respective treatments on the biomass, i.e., white rot fungi selectively

degrade lignin, brown rot fungi degrades polysaccharides and lignin while soft rot fungi simultaneously degrades polysaccharides and lignin (van Kuijk et al. [2015\)](#page-20-2). The efficiency of fungal treatment depends on several factors such as particle size of the biomass, temperature, moisture content, pH, inoculum concentration, and treatment time (Meehnian et al. [2016\)](#page-18-7). Prior to the fungal treatment with the biomass, the fungal growth condition, and media nutrient concentrations needs to be optimized first to enable maximum production of targeted hydrolytic enzymes (Chenthamarakshan et al. [2017\)](#page-17-11).

Additionally, during the fungal treatment toward biomass, crude protein may be found due to the incorporation of nitrogen from the biomass into the fungal protein that causes nitrogen enrichment from the degradation of other nutrients. The efficiency of the fungal treatment is very hard to determine and is highly dependent on the type of biomass used. In order to justify the most efficient fungal treatment, several types of fungi must be treated with one similar biomass and evaluate the production yield for each treatment. The highest production yield achieved can then be claimed as the most efficient fungal treatment. Some of the fungal treatments were coupled with bacterial treatment and the result obtained showed an increase in production yield (Yadav et al. [2019b\)](#page-20-3). However, despite the environmental-friendly process of this fungal treatment on agro-waste, it is time consuming and some carbohydrates will be consumed for the metabolism of fungus and thus may lose some of the valuable products after the treatment (van Kuijk et al. [2015\)](#page-20-2). Nevertheless, the researcher is developing some genetically modified microorganisms, especially to enhance biomass degradation. Table 3.2 below shows the list of recent biological treatment using fungal treatment in various biomass. (Table [3.2](#page-5-0)).

<b>Biomass</b>	Type of fungus	Enzyme involved	Product(s) observed	References
Wheat straw and pearl millet straw (India)	Chaetomium globosporum	Laccase	Reducing sugar and biogas production	Yadav et al. (2019a)
Pearl millet and wheat straw (India)	Pleurotus ostreatus	Laccase	Reducing sugar and biomethanation production	Yadav et al. (2019b)
Hay	<b>Neocallimastix</b> frontalis	Lignocellulolytic enzymes	Biogas production	Dollhofer et al. (2018)
Rice straw	Coprinopsis cinerea	Laccase, cellu- lase, and xylanase	Enzymes' activity profile	Zhang et al. (2018)
Copra meal, sugar- cane bagasse, and rice straw	Aspergillus tubingensis NKBP-55	Cellulase	Fermentable sugars	Prajapati et al. (2018)

<span id="page-5-0"></span>Table 3.2 List of recent biological treatment using fungal treatment on various biomass

# 3.2.2 Yeast Treatment

Yeast treatment is normally been used for the fermentation process where it converts the sugar-containing hydrolysate from the hydrolysis process in bioethanol. The high carbohydrate and nutritional content make agricultural waste as promising biomass for microbial growth of yeast. Nevertheless, yeast has become one of the pioneer microorganism being used as an essential component for human activities such as in bread making, wine, beer, and other distilled beverages. It is also been used widely as a research model organism to study the molecular mechanisms within a cellular process. The Saccharomyces cerevisiae is the most well-known type of yeast among others and is being used industrially for the large-scale production of certain biochemicals. Normally, the utilization of yeast in bioprocessing is carried out subsequently after the pretreatment and hydrolysis process of the biomass. The main function of yeast is to convert the sugar(s) produced in the hydrolysate after the hydrolysis process into biogas in what is known as microbial fermentation process. The efficiency of the pretreatment and the hydrolysis process will determine the success of the yeast fermentation process. There is also a research that utilizes yeast to enhance the nutritive value in some biomass (Kewan et al. [2019\)](#page-18-8). This is one of the broad spectrums of the yeast applications. However, the utilization of yeast in a fermentation technology is not a simple process as it produces inhibitors in the lignocellulosic hydrolysate such as furfural or acetic acid which can interfere the cellular growth and gives diverse metabolic process (Cristobal-Sarramian and Atzmüller [2018\)](#page-17-9). Some approach by using a genetic engineering method has been used to enhance the yeast resistance toward these types of inhibitors and thus may improve the production yield. Table 3.3 shows the recent studies utilizing yeast as a biological treatment for agro-waste. (Table [3.3\)](#page-6-0).

# 3.2.3 Bacterial Treatment

Due to the massive investigation of fungal treatment on biomass, the ability of bacterial treatment has been overlooked. The longer time taken for fungi to grow

<b>Biomass</b>	Type of yeast	Product observed	References
Sida acuta (Thailand Weed)	Saccharomyces cerevisiae K35	<b>Biogas</b>	Siripong et al. (2019)
Wheat straw	Candida guilliermondii	Xylitol	Cristobal- Sarramian and Atzmüller (2018)
Moringa tree stalks	Saccharomyces cerevisiae (yeast) coupled with Trichoderma reesei (fungi)	Improved nutritive value for lamb feed	Kewan et al. $(2019)$

<span id="page-6-0"></span>Table 3.3 Recent studies utilizing yeast as a biological treatment for agro-waste

and secrete its enzyme is not an industrial interest. Due to this limitation of fungal treatment, the potential of bacteria to be used as a treatment source for agro-waste can be explored further. The most popular bacteria used are from the Bacillus sp., Sphingomonas sp., Cellulomonas, and Zymomonas spp. Some bacterial strains are able to secrete lignin-degrading enzymes such as laccase and manganese peroxidase (Zhuo et al. [2018](#page-20-5)) even though the strength of treatment is lower than the fungal treatment. This is probably due to the low amount of degrading enzyme(s) secreted from the bacteria as compared to fungi and the fragility of the bacteria to the changes in the environment. Bacteria may pose different degradation reactivity toward lignin compared to the fungi. Furthermore, most of the bacterial treatment cannot act independently on the biomass due to the direct interactions occurring between the bacterial enzymes and the lignin of the biomass. However, they can slightly improve enzymatic digestion of the biomass after the outer structure breakdown by other physicochemical treatment. Other strategy to improve the performance of bacteria treatment is to extract out the enzyme(s) inside the bacterial cell and further purified the enzyme(s) prior to the treatment. Reports on the bacterial treatment for agrowaste still remain scarce as researchers did not put more effort toward using bacteria for their treatments. Table 3.4 shows the most recent studies utilizing bacteria for the treatment of agricultural waste. (Table [3.4\)](#page-7-0).

# 3.2.4 Fermentation Technology

Fermentation is the final process of converting agro-waste or biomass into bioethanol by various microorganisms such as bacteria and fungi. This conversion process of biomass involves metabolic breakdown that releases several compounds which are known as secondary metabolites. The secondary metabolites released can

Bacteria	Source	<b>Biomass</b>	References
Pandoraea sp. B-6	Bamboo slips	Corn stover	Zhuo et al. (2018)
Paenibacillus chitinolyticus CKS1, Streptomyces fulvissimus CKS7	Soil	Tobacco waste	Aneta et al. (2019)
Bacillus subtilis, Bacillus licheniformis, and Bacillus amyloliquefaciens	Fermented pig manure	Waste straw consortium	Shen et al. (2019)
<b>Bacillus</b> subtilis	Prawn and fish scales	Straw (wheat) and pearl millet)	Yadav et al. (2019b)
Enterococcus, Sporanaerobacter, Para clos- <i>tridium</i> , and Clostridium sensu stricto 1	Sludge (a municipal wastewater treatment plants), cow dung, pig manure, and camel dung	Corn straw, rice straw, corncob, and sorghum stalk	Li et al. (2018)

<span id="page-7-0"></span>Table 3.4 The most recent studies utilizing bacteria for the treatment of agricultural waste

consist of antibiotics, enzymes, and some growth factors. The fermentation process is highly dependent on the type of microorganism's strain used. Some of them are able to one type of sugar, e.g., either glucose or xylose while some are able to ferment both, e.g., glucose and xylose to produce bioethanol. Up to date, several microorganisms have been used for the fermentation process which includes genetically engineered yeast and bacteria, co-culturing and sequential use of yeast strains and also protoplasts fusant strains as glucose and xylose fermenting yeast (Pramanik and Sahu [2017\)](#page-19-8).

#### 3.2.4.1 Submerged Fermentation

Submerged fermentation (SF) is defined as a biological process of producing biomolecules in which microorganisms and enzymes together with other reactive components are submerged in a solution. The liquid solution may be in the form of alcohol, oil, or a nutrient broth. It also involves free-flowing liquid substrates such as molasses and broths. Microorganisms such as fungi will be placed in a closed container or tank containing these rich media. The submerged fermentation process can be divided into two types: (a) aerobic fermentation and (b) anaerobic fermentation with two methods of operation (a) fed-batch fermentation and (b) continuous fermentation. The first critical step in this submerged fermentation is to obtain an adequate number of microorganisms to be used as an inoculum for the subsequent process (Zhang et al. [2019\)](#page-20-6).

#### 3.2.4.2 Solid-State Fermentation

Solid-state fermentation (SSF) is defined as a biological process that involves the growth of microorganism on a dry medium (absence of water) of non-soluble substrate or solid biomass (Sadh et al. [2018\)](#page-19-1). The SSF has already been used by our previous ancestors for natural food processing and currently is regaining more attention due to the increasing number of agro-waste available. These agro-wastes which are rich in valuable composition can be considered as "raw materials" for the SSH instead of "wastes." The existence of these valuable and nutritional compound inside the agro-wastes makes them a suitable platform for the prolific growth of microorganisms. A successful SSF depends on many factors such as microorganism (s) used, biomass, aeration, and water activity. Different biomass may have better characteristics to be used as an immobilization carrier in the SSF because of the high water absorption capacity compared with other biomass. The microorganism used can consist of a single culture, mixed culture, or consortium of a mixed indigenous microorganisms. Molds or fungi are usually been used in the SSF process while bacteria and yeasts may also be used but with lower yield. Table 3.5 shows the comparison between submerge fermentation (SF) and solid-state fermentation  $(SSF)$ . (Table  $3.5$ ).

Fermentation technology	Submerge fermentation (SF)	Solid-state fermentation (SSF)
Advantages	Can be run in either aerobic or anaerobic condition. Sort period, low cost, high yield easier purifica- tion process, and simpler fermenta- tion control	Requires simple fermenter, less efflu- ent produce, better aeration, and less attractive environment for competing bacteria, energy reduction for stirring, and sterilization
Disadvantages	Expensive media	Heat transfer problem, e.g., accumu- lation of heat inside the solid decreased the productivity by time
Microorganisms involved	Bacteria, fungi, and yeast Best suited for bacteria that require a high moisture content	Bacteria, fungi, and yeast Best suited for fungi that require less moisture content
Applications	Extraction of secondary metabolites that is needed to be in liquid form Production of microbial-derived enzymes The process requires agitation Widely used in the manufacturing industries	Enzymes, organic acids, biopesticides, biofertilizers, biosurfactants, animal feed, pigments, vitamin, and antibiotics The process does not involve agitation

<span id="page-9-0"></span>Table 3.5 Comparison between submerge fermentation (SF) and solid-state fermentation (SSF) (Sadh et al. [2018;](#page-19-1) Zhu et al. [2011](#page-20-7))

# 3.3 Advantages and Disadvantages of Biological **Treatments**

Circular economy is a sustainable concept which is based on minimizing waste generation and making use most of the resources. Biological treatment of agricultural biomass is seen as one of the methods that apply a circular economy concept. Traditionally, abundant agricultural wastes are burned in the field which causes the release of greenhouse gasses. Some are dumped in the landfill which causes foul odor and spread of disease. As science advances, in addition to the energy crisis, the world is facing nowadays, this biomass that was used to be called "waste" and "burden" now seems a blessing in disguise. Biomass is an abundant source of carbohydrate, which means food and energy. Turning the biomass into high-value products minimizes waste, hence in line with the circular economy concept.

Saccharification of lignocellulosic biomass is the main challenge to extract carbohydrates from the lignocellulose complex structure. Research is still on-going to find a feasible lignocellulosic pretreatment method that does not destroy the cellulose and hemicellulose, produces cellulose that is readily available for enzymatic hydrolysis, low in energy usage, less by-product, and does not inhibit the subsequent enzymatic reaction. Single or combination methods of physical pretreatment, chemical pretreatment, and physicochemical pretreatment have been widely used to release the cellulosic materials from biomass. Although it is effective in removing the recalcitrant lignin structure, these methods are expensive, require large inputs of energy, cause pollution, and some release by-products that inhibit the following enzymatic hydrolysis or fermentation process, leading to low yield of final

products (Chaturvedi and Verma [2013\)](#page-17-14). Biological pretreatment is an alternative method that is categorized as safe, eco-friendly and sustainable. Table [3.6](#page-10-0) summarizes the advantages and disadvantages of these methods.

Biological pretreatment that includes the use of microorganisms or enzymes provides an alternative for more effective method, eco-friendly, requires low energy and mild operation conditions, and most importantly does not produce inhibitor by-products. Cellulolytic and lignolytic microorganisms such as filamentous fungi are responsible for most of the biodegradation of wood materials in nature. These fungi produce cellulolytic and lignolytic enzymes and degrade cellulose, hemicellulose, and lignin. Therefore, in biological pretreatment of lignocellulosic biomass, various types of fungi were incorporated such as white-rot fungi, brown-rot fungi, and soft-rot fungi (Chaturvedi and Verma [2013\)](#page-17-14). Some bacteria and actinomycetes do not produce lignolytic enzyme but they degrade lignocellulosic biomass by the action of excreted xylanase enzyme. However, the significant side reaction of cellulose degradation by these microorganisms is a major concern (Singh et al. [2008\)](#page-19-12). Hence, the use of purified lignin-degrading enzymes, such as lignin peroxidase, manganese peroxidase, laccase, etc. is another alternative to ensure selective removal of lignin without affecting cellulose content (Kaur et al. [2010\)](#page-18-10). Biological pretreatment of biomass is very specific and selective. In addition, the difference in mechanisms of different species of microorganisms and enzyme reaction specificity offers a broad range of selection to match the desired process goal.

When the biological method is opted for pretreatment of lignocellulosic biomass prior to conversion of cellulose and hemicellulose to the value-added product, it is important to optimize the pretreatment conditions, especially when it involves microorganisms. This is because microorganisms consume carbohydrates. This will lead to reduced yields of cellulose and hemicellulose. Meanwhile, biofertilizer

<b>Treatment</b>	Advantages	Disadvantages
<b>Biological</b>	Eco-friendly	Slow reaction rate
	Mild operating condi-	Needs deep understanding of the mechanism
	tion	Needs optimization due to lack of standardized
	Does not produce inhib-	methods
	itor	Sensitive to inlet feedstock
	Specific	
	Low-energy demand	
Chemical	<b>Fast reaction</b>	Harsh chemicals
	Established standard	Expensive
	method	Cause environmental pollution
		Nonselective reaction; affects cellulose and
		hemicellulose
Physical	Does not require	Expensive equipment
	chemicals	Limited application
Physicochemical	Fast and established	High-energy demand
	method	Needs special equipment
		Produces toxic waste

<span id="page-10-0"></span>Table 3.6 Comparison between different types of lignocellulosic biomass pretreatment methods

does not require cellulose and hemicellulose retention. Therefore, composting using microorganisms is an advantage in the production of biofertilizer. Composting is seen as a great way to waste management. Composting not only minimizes agricultural waste, but it also helps to improve soil fertility and become a green alternative to chemical fertilizer which often destroys soil microdiversity. Although microbial degradation takes time, composting with right technique at controlled condition and optimum composition shorten the time, especially when fungi are involved. Many research has established the optimized method for composting and this could easily be applied to the field. Another drawback of biological treatment is the microorganisms are sensitive to the inlet feedstock and can be easily inhibited if the feedstock contains substances that are toxic to the microorganisms' growth (Basso et al. [2016\)](#page-17-15).

Biological treatment is considered proper waste management that converts waste into high-end products with a process that produces low greenhouse gases (Basso et al. [2016](#page-17-15)) and toxic by-products. Conversion of this waste into bioenergy is one of the successful processes that could solve waste accumulation as well as providing sustainable alternatives for solving the energy crisis. Several European countries especially Germany, Denmark, Austria, and Sweden have increased their interest in these renewable energy sources and have invested to promote the development of biogas plants (Weiland [2006](#page-20-8)). Besides, biological treatment has also been proven effective to convert lignocellulosic biomass into biofuel, biofertilizer, animal feed, and bioactive compounds. Despite the disadvantages of the biological treatment process as mentioned above, research regarding this topic has been progressing and has reach maturity stage, whereby the process drawbacks could actually be catered by proper optimization. And now, it leaves a challenge for the engineers to upscale the process for large-scale application and convinces the policy makers around the world that it is time to change to a greener process for a better world.

# 3.4 Products from the Biological Treatments of Agro-Industrial Wastes

Commonly, the major constituents of agro-industrial wastes can be accounted for by several elements such as complex polysaccharide/proteins, carbohydrates, and polyphenolic constituents, which is considered as high strength of organic pollutants that could cause an adverse effect on the environment. One of the most efficient means of *lowering* environmental issues in agro-industries is through recycling and clean technology. Execution of this practice targets to constrain environmental pollution and other associated problems in addition to encourage economic benefits, such as the conversion of wastes to added value by-products (Prasertsan et al. [2007\)](#page-19-13). Assuming that the availability and the lavish quantity of nutritious agro-industrial wastes throughout the year, there is a great interest on the recycling of these wastes for the production of other value-added compounds with the expectancy of reducing the production costs. The potentials of agro-industrial wastes are presented in Table [3.7.](#page-12-0)

Application	Agro-waste	References
<b>Biofuel</b>	Rice straw, wheat straw, corn straw, and sug- arcane bagasse	Kim and Dale $(2004)$ , Georgieva et al. (2008), Li et al. (2010)
Enzyme production	Banana peel, wheat bran, rice bran, orange peel, and sugar cane bagasse, corn cob	Ravindran et al. (2018), Bharathiraja et al. (2017)
<b>Bioactive</b> compounds	Tomato, olive, apple, banana, guava, mango- steen, and jackfruit	Kumar et al. $(2017)$ , Deng et al. $(2012)$ , Mahmoud et al. $(2018)$
Mushroom cultivation	Rice straw and bran, coffee pulp, sawdust, wheat straw, cotton straw, tea leaves, and banana leaves	Kamthan and Tiwari (2017), Salama et al. (2016), Josephine (2014)

<span id="page-12-0"></span>Table 3.7 The potentials of agro-industrial wastes

Population and industrialization may lead to an increase in global energy consumption and a shortage of fossil energy sources such as oil, coal, and natural gas. In this regard, many agree that bioethanol is a good alternative to fossil energy as a renewable source. As a consequence of widespread interest and demand at a worldwide level, the global market share for bioethanol has entered a quick transitional growth phase. In addition, reducing crude oil reserves forced many countries to shift their focus from fossil energy to renewable sources for power production (Georgieva et al. [2008](#page-17-16); Sarkar et al. [2012](#page-19-14)). The use of bioethanol in the transport fuel market can *reduce dependency* on costly gasoline and exhaust *greenhouse gas emissions*, in particular,  $CO<sub>2</sub>$ . Thus, the necessity of using alternative raw materials in addition to conventional crops such as corn and sugarcane, which are currently the main feedstock used for the making of bioethanol, has increased (Sarkar et al. [2012;](#page-19-14) Gupta and Verma [2015](#page-17-17)). Lignocellulosic biomass contains a wide range of plantbased waste materials from agriculture industries such as wheat and rice straw, corn stover, and sugar cane bagasse. These feedstocks, which are cost effective, renewable and abundant, are scientifically tested and proposed for bioethanol production. Hence, bioethanol from agro-industrial wastes is very encouraging and seem to be an attractive alternative technology (Saggi and Dey [2016\)](#page-19-15). Pretreatment process is important to modify the structure of hemicellulose, lignin, and cellulose contain in lignocellulosic biomass for higher accessible to further chemical or biological treatment (enzymatic attack) thus increase the conversion yield of cellulose into monomeric sugars (Demirbas [2005](#page-17-18)). Production of bioethanol from lignocellulosic biomass faces some major challenges and limitations, for example, effective pretreatment methods to enhance the yield of delignification and hydrolysis of lignocellulosic biomass (Kucharska et al. [2018\)](#page-18-11). Therefore, considerable effort in recent years has been devoted to investigating efficient and cost-effective methods for hydrolysis and conversion of lignocellulosic biomass into fermentable sugars for biofuels production (Priyanka et al. [2018\)](#page-19-16).

Agro-industrial wastes are the cheapest, lavishly available, and highly nutritious carbon source which can facilitate the growth of a wide range of microorganisms. The large composition of the agro-industrial wastes particularly from lignocellulosic waste is the carbohydrates thus have a great potential to be utilized for the production of numerous value-added products including industrially important enzymes (Ravindran et al. [2018\)](#page-19-17). wheat bran and straw, rice bran, corn cob, and sugarcane bagasse are the frequently studied agro-industrial waste for the production of numerous enzymes with various industrial processes applications such as food, drug, textile, and agriculture (Bharathiraja et al. [2017](#page-17-19)). Different fermentation strategies are applied including solid-state fermentation and submerged fermentation methods in which the former approach was preferred to the latter. Solid-state fermentation becomes an attractive and preferable process due to the direct utilization of crude fermented products as enzyme source which could potentially lessen the production cost as well as low amount of energy required, high product yield, and simple downstream processing. Recent studies have shown that a number of microorganisms including fungi, bacteria, and actinomycetes have been reported for enzyme production using lignocellulosic waste. Among them, a group of filamentous fungal species including Aspergillus (Bhavna and Magar [2010;](#page-17-21) Kang et al. [2004\)](#page-18-18), Trichoderma, Botrytis, and Penicillium has been widely investigated and commercially employed for cellulase and hemicellulase production using biomass from agro-industrial wastes (Soliman et al. [2013\)](#page-19-19). Several attention have also been focused on studying the production of other industrially important enzymes such as amylases, xylanase, lipases, mannanase, β-glucanase, lactase, invertase, pectinase, and many more by using the action of a wide range of microorganism (Ravindran et al. [2018](#page-19-17)). Exploitation of agro-industrial wastes has good potentials in significant cost reduction and improving the enzymes' demand for industrial purposes.

Many studies have demonstrated the importance of bioactive compounds in human health. Bioactive compounds exhibit numerous therapeutic effects through several cellular mechanisms and are known to possess properties such as anticancer, antioxidant, antidiabetic, and anti-cardiovascular activities. Recently, natural bioactive compounds are being studied for their potential in the prevention and therapy of a number of human diseases/disorders such as diabetic, heart disease, and cancer (Joana Gil-Chávez et al. [2013;](#page-18-19) Yusuf [2017](#page-20-9)). Particularly, the *potential* of agroindustrial wastes as inexpensive and readily available resources of natural bioactive compounds is considered which could lessen operational cost and reduce environmental impact. The recovery of the bioactive elements, especially the phenolic compounds and vitamins, offers great potential for food, cosmetics, and pharmaceutical industries such as for high-value products development (Kumar et al. [2017\)](#page-18-14). The investigation of antioxidant properties and phenolic contents of several fruit residues such as apple, banana, guava, mangosteen, and jackfruit exhibited the different antioxidant level in different fruits and from the analysis, the major bioactive constituents in fruits residues were detected which is composed of catechin, cyanidin 3-glucoside, epicatechin, galangin, gallic acid, homogentisic acid, kaempferol, and chlorogenic acid (Deng et al. [2012\)](#page-17-20). In addition, several therapeutic bioactive compounds extracted from olive and tomato waste exhibited outstanding antimicrobial antioxidant and anticancer efficacy (Kumar et al. [2017;](#page-18-14) Mahmoud et al. [2018](#page-18-15)).

Reprocessing of agro-industrial wastes with implementations in the agro-food industry is one of the big challenges in the biotechnology field nowadays. Yearly, the

accumulation of massive volumes of lignocellulose wastes resulted from agroforestry and agro-industrial production causes serious environmental damages. Due to the high nutritional content in them, disposal as a compost is challenging as the leaching process will occur. Thus, most of the time these waste will be disposed by incineration lead to a serious environmental pollution. Hence, recycling of these organic wastes via mushrooms cultivation becomes one of the most efficient solutions to overcome the environmental issues raised by the accumulation of these organic wastes (Kamthan and Tiwari [2017\)](#page-18-16). As a consequence of many investigations performed, the cultivation of edible and medicinal mushrooms was done using both solid-state fermentation and controlled submerged fermentation. These fermentation approaches are using a wide range of lignocellulose by-products which provide a rapid growth together with high biomass yield of the studied mushroom strains (Petre and Teodorescu [2012](#page-19-20)). Mushroom are capable to breakdown the complex lignincellulosic components in agro-industrial wastes thus this fleshly and spore-bearing fruiting bodies fungi can be generated from lignocellulosic waste materials. Mushrooms are regarded as nutritious food product rich with protein, folic acid along with vitamin B12. The bioconversion of agro-industrial wastes into a value-added products is observed as an environmentally friendly practice with potential economic advantage (Oyedele et al. [2018](#page-19-21)). Generally, numerous edible mushroom strains are cultivated such as Agaricus sp., (button mushroom), Pleurotus sp. (oyster mushroom), Lentinula edodes (shiitake mushroom), Volvallella volvacea (straw mushroom), and Ganoderma sp. (Chinese mushroom) using varied agroindustrial wastes cultivation medium including rice straw and bran, coffee pulp, sawdust, wheat straw, cotton straw, tea leaves, and banana leaves (Kamthan and Tiwari [2017;](#page-18-16) Salama et al. [2016](#page-19-18); Josephine [2014\)](#page-18-17).

# 3.5 Creating a Sustainable Practice for Industries Producing Agricultural Waste

### 3.5.1 Improve Plant Profitability

Over the years, agro-industrial waste has received huge attention from different researchers worldwide and is being explored as a plentiful source for bioactive compounds and promotes microbial growth. Most of the agricultural wastes have carbohydrates as a major component and lignocellulosic in nature (Ravindran et al. [2018\)](#page-19-17). Production of value-added products using agricultural residues is a promising and smart strategy from an industrial point of view to produce enzymes (Sadh et al. [2018\)](#page-19-1). Utilization of agro-industrial wastes as low-cost raw materials for the production of the value-added product can help to minimize the production cost. Instead of using an expansive substrate as core nutrients, using agro-industrial waste provides a cheaper and more sustainable process in which they can be used as a sole nutrient source during production. In addition, the pollution load from the

environment can be greatly reduced and less cost required to create and maintain a waste management system. Intense research has been carried out in order to investigate different fermentation strategies for the production of enzymes to enable the use of various agro-industrial wastes such as sugarcane bagasse, corn cob, and rice bran during the process (Singh et al. [2012\)](#page-19-22).

Through a comparison of the impact on enzyme production via different fermentation strategies, solid-state fermentation showed greater potential than submerged fermentation utilizing agricultural waste for enzyme production (Ravindran et al. [2018\)](#page-19-17). This is due to the physical–chemical nature of many lignocellulosic substrates naturally lends itself to solid-phase culture, hence, implies a means to procure the acknowledged potential of this fermentation method. Recent studies proved that the enzyme yield could be significantly improved using pretreatment technologies (Ravindran et al. [2018\)](#page-19-17). Currently, starch is the foremost cost element in fermentation to generate bioethanol. Agricultural by-product such as wheat bran, rice bran, sugarcane bagasse, corn cob, and wheat straw can be used to get fermentable starch and sucrose easily (Singh et al. [2012](#page-19-22)). The use of low-value agricultural wastes in fermentation process provides the reduction of negative impact costing on production (Molina et al. [2018\)](#page-18-20).

### 3.5.2 Suggestions and Recommendations

Agro-industrial wastes are produced during the industrial processing of agricultural products. These waste products and the by-products are being created in abundance which rising economic losses issue and worsen the low-margin of profitability in the food industry and high costing of raw materials, which should make the caution step to use the residues beneficial to the agricultural industry. Thus, the profitability of the industry could be momentously enhanced. The valorization and following value addition of carbohydrates that obtained easily from lignocellulosic food industry wastes are the cheapest. Numerous enzymes can be generated by both bacterial and fungal species using agricultural wastes. Economics value of enzyme production can be greatly enhanced by pre-treatments that enable high saccharification rates at lower enzyme loadings (Ravindran et al. [2018\)](#page-19-17). The potential of exploiting agricultural by-product is being limited as high host in drying or storage purpose as well as tend to be easily spoiled by the bacteria. The extensively exploiting of agro-industrial coproducts alleviate the environmental problems and add nutritional value to the food products. Due to these coproducts constitute up to 70% of fresh fruit and poor ability to discard these materials. Thus, the pollution of environmental issues is aroused. On the other hand, it is possible to utilize the content of bioactive compounds such as polyphenols, proteases, amd dietary fibers with appropriate processing to improve the nutritional properties in health and pharmaceutical sectors. These nutritional properties can be improved by providing more nutrients such as proteins, carbohydrates, or fats. In addition, it is reported by previous researcher that

chronic diseases can be evaded by consumption of these nutrients with processed products.

Agro-industrial wastes or residues are naturally rich in nutrient composition and bioactive compounds (Beltrán-Ramírez et al. [2019](#page-17-22)). Sugars, minerals, and proteins are the compositions constituted in agro-industrial waste. Therefore, it presumes to consider as "raw material" rather than "wastes" for use in other industrial processes. The availability of nutrients in these residues contributes to suitable productive growth conditions for the microorganisms. Through the fermentation process, the microorganisms have enormous potential to reutilize the waste as raw materials for growing purpose (Sadh et al. [2018](#page-19-1)). A wide range of beneficial bioactive compounds can be produced significantly by utilization of agro-industrial wastes as solid support in the solid-state fermentation process (Lizardi-Jimenez and Hernandez-Martinez [2017\)](#page-18-21). Extensively utilizing agricultural and agro-based industrial wastes as raw materials can minimize production cost and contribute in reprocessing of waste. Therefore, environmental pollution problems can be mitigated (Beltrán-Ramírez et al. [2019](#page-17-22)).

### 3.6 Conclusion

Biological treatments have immense potential to relieve the alarming issue on the disposal of the escalating amount of agro-industrial wastes. The present study has listed the sources and types of these wastes, along with the various industries producing them. Understanding that most agro-industrial wastes are left untreated or underutilized, it is imperative to seek for effective measures to avoid further environmental damages. A number of possible biological treatments have been highlighted including fermentation, solid-state fermentation, fungal, and bacterial treatments. Any of these methods could be applied according to the suitability of the waste conditions. The advantages and disadvantages have also been discussed for which a comparison between biological treatments and other treatments were deliberated. Aside from being economical, biological treatments are straightforward and easy to implement, for which they will benefit the human and environment simultaneously. This study strengthens the idea that closing the loop by valorizing the agro-industrial wastes through biotransformation significantly contributes to the reduction of waste deposition into the environment. Useful products like biogas, bioactive compounds and mushrooms are prospective outcomes of biological treatments. Lastly, several recommendations on creating a sustainable practice for agroindustries have been outlined. These efforts aim at overcoming probable issues like nutrient imbalance, rapid acidification, and inhibiting compounds, and ultimately ensuring plant profitability. This study provides an important insight into applying a cleaner process on unworthy materials toward a better and safer product.

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