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PERSPECTIVE OF MICROBIAL SPECIES USED IN LIGNOCELLULOSES BIOCONVERSION

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ABSTRACT

Lignocellulosic wastes are abundant, renewable and inexpensive sources of energy. This wastes contains large amount of residual plant biomass which is non edible material obtained from plant cell walls. Biomass could be obtained from crop, domestic liquid fuel, municipal solid waste and agricultural residuals. In nature, cellulose, hemicellulose and lignin are major component of plant biomass therefore, their recycling is essential for the carbon cycle completion. In many countries environmental problems arises due to improper utilization of valuable waste materials. Lignocelluloses degradation is a difficult process due to presence of lignin which is an important constituent of plant cell wall. Lignocelluloses waste is degraded by a variety of microorganisms which produce a battery of enzymes that work synergically. Lignin degrading microorganisms required enzymes to convert lignocellulosic biomass into valuable carbohydrate source for the production of bio-ethanol, biogas and many more industrial products. Bioconversion of lignocellulosic residues is initiated by microorganisms such as fungi and bacteria which are capable of degrading lignocellulolytic materials. Among lignin degrading microbial species, white rot fungus are main due to the adequate presence of lignin degrading enzymes. This group of fungus (Basidiomycetes) are known to be efficient lignocelluloses degraders and considered to be the primary degraders of lignin and lignocellulosic materials in terrestrial ecosystems. In addition to that it is also necessary to develop the strains for conversion of lignocellulosic biomass to useful product. This review focuses on the variety of microorganisms known to degrade lignocellulosics their characteristics and involved mechanics/action and future prospects in lignocellulose bioconversion.

KEYWORD: **Lignocellulose, Lignin, Hemicellulose, fungi**

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INTRODUCTION

Worldwide approximately 3480 Trillion grams/year of lignocellulose in the form of agricultural waste is accumulated every year [1]. Lignocellulose is a renewable organic material and is the major structural component of woody plants and non-woody plants such as grasses. It is composed of three major components: cellulose (35-50%), hemicelluloses (20-35%) and lignin (10-25%) [2]. In addition, small amounts of other materials such as ash, proteins and pectin can also be found in lignocellulosic residues [3]. Following are the composition and percentages of these polymers which varies from one plant species to another [4].

Table 1

Lignocellulose waste	Cellulose (wt %)	Hemicellulose (wt %)	Lignin (wt %)
Barley straw	33.8	21.9	13.8
Corn cobs	33.7	31.9	6.1
Corn stalks	35.0	16.8	7.0
Cotton stalks	58.5	14.4	21.5
Oat Straw	39.4	27.1	17.5
Rice Straw	36.2	19.0	9.9
Rye Straw	37.6	30.5	19.0
Soya Stalks	34.5	24.8	19.8
Sugarcane bagasse	40.0	27.0	10.0
Sunflower Stalks	42.1	29.7	13.4
Wheat straw	32.9	24.0	8.9

Moreover, the composition within a single plant varies with age, stage of growth, and other conditions. Cellulose is a high molecular weight linear homo polymer of repeated units of cellobiose (two anhydrous glucose rings joined together a β-1,4glycosidic linkage) [5]. The long-chain cellulose polymers are linked together by hydrogen and vander walls bonds, which cause the cellulose to be packed into micro fibrils [6]. Hemicellulose is a heteropoly saccharide composed of pentoses (D-xylose and D-arabinose), hexoses (D-mannose, D-glucose and D-galactose) and sugar acids. Lignin is a complex polymer of phenyl propane like *p*-coumaryl, coniferyl and sinapyl alcohol. This acts as a cementing agent, an impermeable barrier for enzymatic attack as well as resistance against microbial attack and oxidative stress and provides structural support to the plants [7]. These properties of lignin may be attributed to its amorphous nature, water insolubility and optical inactivity which also make it tough to be degraded [8].

Lignocellulose wastes include a variety of materials such as sawdust, poplar trees, sugarcane bagasse, waste paper, brewer's spent grains, switch grass, and straws, stems, stalks, leaves, husks, shells and peels from cereals like rice, wheat, corn, sorghum and barley etc. Significant effort many of which have been successful, have been made to convert these lignocellulosic residues to valuable products such as bio fuels, chemicals and animal feed [7, 9] and cheap energy sources for fermentation. However, due to the presence of lignin, lignocellulose is complex structure difficult for degradation. However, lignocellulose biomass digestibility may be enhanced by various pre-treatment like physical, chemical, physio-chemical and biological [10]. Some physio-chemical treatment such as dilute acid, hot water, ammonia fiber explosion, ammonia recycles percolation, and lime; but these methods are capital-intensive [11]. Whereas biological treatments have some advantages over other methods such as the high specificity, low energy consumption, no chemical requirement, and mild environmental conditions which avoid sugar degradation and resulting in high sugar yields [12]. Pre-treatment by high temperature and acid have been done as initially for chemical cellulose degradation and for lignocellulosic residues at industrial scales. However, this approach is expensive, slow and inefficient [13]. The overall yield of the fermentation process will be decreased because this pre-treatment releases inhibitors such as weak acids, furan and phenolic compounds [14].

MICROBIAL SPECIES INVOLVED IN LIGNOCELLULOSES DEGRADATION

Microorganisms are the primary degraders of lignocellulose, which is indigestible to most animals. The lignin component is the more recalcitrant to microbial degradation and can act as a barrier to biodegradation of the more readily degradable polysaccharides [15]. Thus, microbial degradation of the lignin component of lignocellulose is generally the rate-limiting step in degradation of the associated polysaccharides. Both bacteria and fungus is extensively used for lignocellulose degradation. Fungi especially white rot fungi are able to degrade all three major wood polymers: cellulose, hemicellulose, and lignin [16]. In general, because the substrates are insoluble, both bacterial and fungal degradation occurred exocellularly either in association with the outer cell envelope layer or extracellularly. Microorganisms have two types of extracellular enzymatic systems: the hydrolytic system, which produces hydrolases and which is degraded cellulose and hemicellulose; and a unique oxidative and extracellular ligninolytic system, which depolymerises lignin.

FUNGAL DEGRADATION OF LIGNOCELLULOSES: MICROORGANISMS AND THEIR MODE OF ACTION

Problems associated with chemical and physical treatment could be overcome by applying microorganisms such as fungi. It has been reported that most isolates of anaerobic fungi taken from animals fed on highly fibrous diets were able to degrade wheat straw *in vitro* environment [17]. Thermophilic fungal species such as *Sporotrichum thermophile* [18], *Thermoascus aurantiacus* [19] and *Thielavia terrestris* [20] have been proposed as good candidates for bioconversion of lignocellulosic residues to sugars and offer the great potential to be used at industrial scales. The lignocellulose-degrading enzymatic system is important for substrate colonization and carbon acquisition by wood rot fungi. Soft rot fungi produce cellulase and it has almost no effect on lignin [21]. Lignocellulolytic enzymes-producing fungi are widespread, these include several thousand species of fungi most of them are basidiomyceteous and a few are ascomyceteous, i.e. white-rot fungi (e.g. *Phanerochaete chrysosporium*) [22], brown-rot fungi (e.g. *Fomitopsis palustris*) and finally a few anaerobic species (e.g. *Orpinomyces* sp.), which also degrade cellulose in gastrointestinal tracts of ruminant animals [23, 24]. A few strains of *Pleirotus* sp. also reported to improve digestibility of straw on fungal mycelial growth or after mushroom harvest [25]. A large number of white rot fungi including strains of *Pleurotus, Ganoderma,* and *Stropharia* etc. have been also found to increase the *in vitro* dry matter degradability of wheat straw and sugarcane bagasse [26, 27] to a very significant extent. White rot fungi such as *Phanerochaete chrysosporium*, *Coriolus versicolor* and *Phlebia radiata* are the most active ligninolytic organisms studied so far [28]. Biomass degradation by these fungi is performed by complex mixtures of cellulases [29], hemicellulases [23] and ligninases [3, 30] reflecting the complexity of the materials. Some marine basidiomycetes such as *Digitatispora marina, Halocyphina villosa* and *Nia vibrissa* have been also shown as capable of white- rot type decay [31, 32 and 33]. Although other marine fungi principally the ascomycetes are commonly associated with salt-marsh detritus material at various stages of decomposition [34.] Some other marine ascomycetes such as *Buergenerula spartinae, Phaeosphaeria typharum*, and *Leptosphaeria obiones* are among the species most frequently observed growing on decaying *Spartina alterniflora* in salt marshes, including marshes on Sapelo Island. These reports suggest that these marine fungi may be capable of extensive degradation of lignocellulose of *Spartina alterniflora* detritus. The pure cultures of fungi and the mixed bacterial inoculums were all capable of degrading the uniformly labelled *Spartina alterniflora* material. Hydrolytic activity for the degradation of lignin of anaerobic fungal isolates obtained from wild ruminants is higher than from domestic ruminants [35].

The core pools of enzymes responsible for cellulose breakdown comprises of cellulases, cellobiohydrolases, endoglucanases and β-glucosidase [36]. Endoglucanases can hydrolyze glycosidic bonds internally in cellulose chains whereas cellobiohydrolases act preferentially on chain

ends. Cellulases break cellulose into glucose or other oligosaccharide compounds [37]. The products of the enzymatic reaction are mostly a disaccharide known as cellobiose and to a lesser extent, cello-oligosaccharides [38]. A novel enzyme activity capable of an oxidative cleavage of the glycosidic bond is currently classified in the Glycoside Hydrolase family 61 (GH61) [39]. It has been identified as the oxidative process which is due to a variety of GH61-like proteins from different fungi as well as bacteria (GH61D from *Phanerochaete chrysosporium* [40, 41], GH61A from *Thermoascus aurantiacus* [42], several from *Neurospora crassa* [43] and CelS2 (CBM33) from *Streptomyces coelicolor* [44]) have been isolated and studied. Cellulases are inducible enzymes synthesized of these microorganisms can be aerobic, anaerobic, mesophilic or thermophilic. Among them, the genera of *Clostridium, Cellulomonas, Thermomonospora, Trichoderma,* and *Aspergillus* are the most extensively studied cellulose produce [13, 45, 46 and 47]. Most commercial glucanases (cellulases) are produced by *Trichoderma ressei* and β-D-glucosidase is produced from *Aspergillus niger* [48]. Fungi known to produce cellulases include *Sclerotium rolfsii, Phanerochaete chrysosporium* and various species of *Trichoderma, Aspergillus, Schizophyllum and Penicillium* [49, 50]. Among these fungi, *Trichoderma* species have been extensively studied for cellulase production [50]. 15,000 fungal species capable of degrading cellulose had been isolated, but only a few of them were subjected to in-depth studies [51]. Cellulolytic microorganisms can establish synergistic relationships with non-cellulolytic species in cellulosic wastes. It has been shown that white and brown-rot fungi produce low molecular weight chelators which are able to penetrate into the cell wall. For example *Gloeophyllum trabeum* produces a low molecular weight peptide (known as short fiber generating factor, SFGF) which can degrade cellulose into short fibers by an oxidative reaction [52, 53]. These fungi have been shown to produce cellulases and to degrade native cellulose; however, the enzyme activity in thermophilic organisms (e.g. *Sporotrichum thermophile*) is usually low compared to mesophilic fungi such as *Trichoderma reesei* [18]. These microorganisms represent the second important group of wood-colonizing fungi, after basidiomycetes. Certain ascomycetes cause soft-rot decay on wood with significant degradation of cellulose as compared to lignin [54]. Several mesophilic cellulolytic anaerobes have been isolated from common environments including soil and sediments, compost, sewage, sludge, and anaerobic digesters [55].

The cellulase systems of the mesophilic fungi such as *Trichoderma reesei* and *Phanerochaete chrysosporium* are the most thoroughly studied [56]. Rapid utilization of cellulose included several taxa. These are *Corollospora maritima* and *Monodictys pelagi* [57], *Julella avicenniae, Lignincola laevis, Nia vibrissa* and *Stagonospora* sp.[58]. The *Penicillium ochrochloron* Y5 has strong ability of wheat straw cellulose degradation, and its cellulase activities are higher than some published researches. The *Penicillium ochrochloron* Y5 strain has the great potential in research and development for inoculant of crop straw decomposition [59]. The anaerobic gut fungi have attracted widespread interest as the most active cellulose degrader in the biological world although their cultivation and maintenance are difficult [60]. Similarly to cellulases, aerobic fungi such as *Trichoderma* and *Aspergillus* secrete a wide variety of hemicellulases in high concentrations and these works in a synergistic manner [61]. Specific microorganisms or a suitable cocktail of enzymes known as hemicellulases can also promote the hemicellulose hydrolysis. Hemicellulases are also produced by many species of fungi. A number of hemicellulases, including xylanases and mannanases, have been identified in *Trichoderma reesei* [62]. Most commercial hemicellulase preparations are produced by genetically modified *Trichoderma* or *Aspergillus* strains. Xylan is the main carbohydrate found in hemicellulose. Its complete degradation requires the cooperative action of a variety of hydrolytic enzymes. In addition, hemicellulose biodegradation needs accessory enzymes such as xylan esterases, ferulic and p-coumaric esterases, α – l - arabinofuranosidases, and α-4-O-methyl glucuronosidases acting synergistically to

efficiently hydrolyze wood xylans and mannans [59]. Attempt has been made for the use of microbial enzymes in xylan hydrolysis, which are specific in action and are an environment friendly. Potential applications of xylanases include bioconversion of lignocellulosic material to fermentative products, clarification of juices, improvement in consistency of beer and the digestibility of animal feed stock etc. [63, 64]. Hemicellulose degradation by xylan and glucomannan utilisation has been observed in qualitative assays used for six marine fungi (4 mitosporic, 1 ascomycete, 1 basidiomycete) [65]**.** It has been reported that xylanase production can be increased by the supplementation of culture media with various aromatic compound, metal chloride. Increase in xylanase yield addition of tween-80 in *Thermoascus aurantiacus* [66], *Thermoascus cutaneum* SL 409 [67] and *Fusarium oxysporum* [68]. Xylanase production by *Aspergillus, Fusarium, Penicillium* and *Streptomyces* has also been shown to be markedly dependent on pH [69, 64]. The production of xylanase from this thermoalkalophilic actinomycete has been enhanced 1·44-fold [70].

The degradation of lignin has been discovered by microorganism which produce lignin degrading enzyme [71, 72 and 73] and by adsorption. Biological treatments, based on the use of brown, white and soft-rot fungi have been commonly used to degrade the lignin, being considered a cheap and effective method of delignification. Selective delignification for of wood and non wood lignocellulosic substrates has been reported by *Ceriporiopsis subvermispora* [74]. The recent research regarding the lignin degrading white rot and brown rot fungi is giving intriguing possibilities with aerobic fungal treatment followed by anaerobic digestion [75]. White-rot fungi are the microorganisms that most efficiently degrade lignin from wood. The lignin degrading enzymes consist of laccase, lignin peroxidase, manganese peroxidise and H_2O_2-supplying glucose oxidase for the peroxidise reactions. *Phanerochaete chrysosporium* is one of the most widely studied white-rot fungi with regards to lignin degrading enzymes [76]. Degradation of lignin by White-rot fungi such as *Phanerochaete chrysosporium, Trametes versicolor* [77]*, Trametes hirsuta*, *Bjerkandera adusta* and *Coriolus versicolor* [78] may be used to allow better access to the cellulose and hemicellulose components, besides to be also considered as an effective biological detoxification alternative. Main problems in using biological methods are that fungi may also attacks cellulose and hemicellulose, and hydrolysis rate in most biological materials is very low [12]. Lignins degrade fungi by secreting enzymes collectively termed “ligninases”. Two major families of enzymes are involved in ligninolysis by white-rot fungi: peroxidases and laccases. After peroxidises further divided two groups, lignin peroxidases (LiPs) and manganese-dependent peroxidases (MnPs) [28]. Lignin peroxidases (LiPs) were the first ligninolytic enzymes to be discovered Tien [79]. They occur in the frequently studied white rot fungi *Phanerochaete chrysosoporium, Trametes versicolor, Bjerkandera* sp., *Irpex lacteus*, and *Phlebia (Merulius) tremellosa* [80, 81, 82 and 83].

Cellulolytic and lignolytic fungus especially *Aspergillus fumigatus, Aspergillus flavus* and *Fusarium oxysporum* are mainly involved to convert agricultural, kitchen wastes and all organic waste in to Biogas, Bio ethanol, Composting etc., by the activity of their degrading enzymes like Laccase [84]. Wood-rotting fungi are the main producers of laccases but this oxidase has been isolated from many fungi including *Aspergillus* and the thermophilic fungi *Myceliophora thermophila* and *Chaemotium thermophilium* [85]. The role of laccases in lignin biodegradation has been discussed recently [76]. White-rot fungi are currently being used not only in the biodegradation of lignin but also in bioremediation of other lignin related pollutants such as industrial White-rot dyes, aromatic pollutants [86, 87] as they secrete the low specificity and strong oxidative ligninolytic enzymes which could oxidatively degrade lignin and mineralize them into CO_2 and water [88, 89]. Molecular oxygen can be crucial in determining the rate of lignin degradation by *Phanerochaete chrysosporium* [90], as well as by certain other white rot fungi [91].

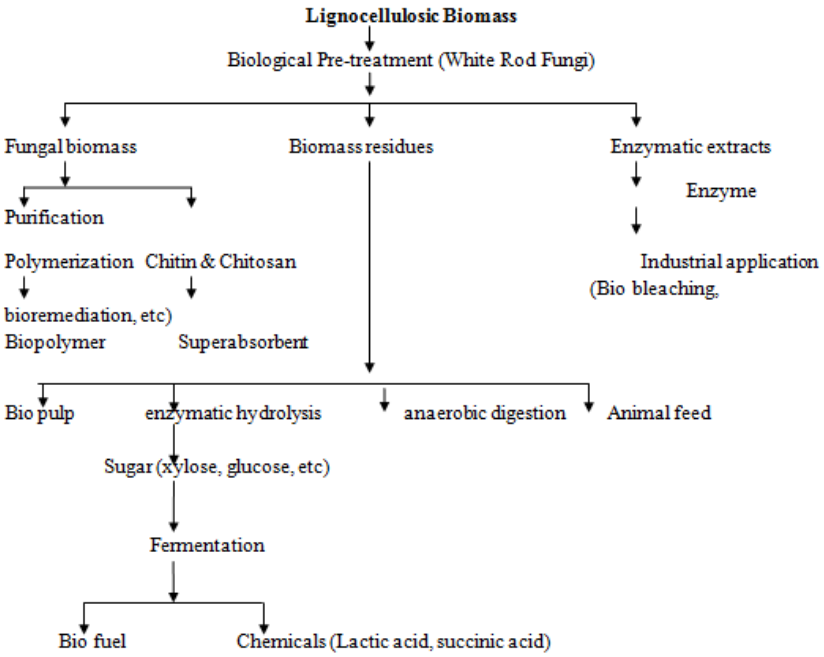

Figure 1
Biological pretreatment with White rod fungi for various applications [92]

BACTERIAL DEGRADATION OF LIGNOCELLULOSES

Lignocellulose degrading bacteria has an important role in energy supply for ruminants. Ruminants are able to convert low quality feed in rumen because role of the lignocellulolytic bacteria. Tropical buffalo can grow properly with low quality roughage, agricultural and industrial waste with basic structure high lignocellulose as main energy source [93]. Both bacteria and fungi can produce glucanases (cellulases) that hydrolyze of lignocellulosic materials. These microorganisms can be aerobic or anaerobic and mesophilic or thermophilic. The genome of *Clostridium phytofermentans,* (ATCC 700394) encodes for the highest number of enzymes for degradation of lignocellulosic biomass among sequenced clostridial genomes [94]. Bacteria belonging to genera of *Clostridium, Cellulomonas, Bacillus, Thermomonospora, Ruminococcus, Bacteriodes, Erwinia, Acetovibrio, Microbispora*, and *Streptomyces* are known to produce cellulose [95]. Anaerobic bacterial species such as *Clostridium phytofermentans, Clostridium thermocellum, Clostridium hungatei,* and *Clostridium papyrosolvens* produces cellulases with high specific activity [50, 95]. Anaerobic bacteria, mainly from the class Clostridia e.g. *Clostridium thermocellum* [96] inhabiting in the gastro-intestinal tract of ruminants and most non-ruminant herbivores produce a range of cellulolytic and hemicellulolytic enzymes in a multienzyme complex known as cellulosome. The cellulosome, however, was initially discovered in anaerobic bacteria such as *Clostridium thermocellum* in 1983 [97] and then first described in anaerobic fungi in 1992 *Neocallimastix frontalis* [98].

In anaerobic bacteria, the cellulosome is usually comprised of 20 or more different cellulolytic/ hemicellulolytic enzymes. Among aerobic cellulolytic bacteria, species from the genera *Cellulomonas*, *Pseudomonas*, and *Streptomyces* are the best studied [99]. Thermophilic xylanases have been described in actinobacteria such as *Thermomonospora* and *Actinomadura* [100]. Thermo stable xylanase has been isolated from the hyper thermophilic primitive bacterium *Thermotoga* [101]. Some actinomycete bacteria of the genus *Streptomyces* have been shown to produce extracellular lignin peroxidises [102] that can depolymerise/solubilise the lignin component of lignocelluloses [103]. Degradation of lignin and lignin-degrading enzymes has also been reported for actinobacteria from the

Streptomyces genus [104]. The known potential lignin-degrading bacteria are mostly derived from guts of wood-eating insects and include *Alphaproteobacteria, Gammaproteobacteria* and *Actinomycetes* [105], with the best-characterized being *Streptomyces viridosporus* [102]. Bacteria known to break down lignin are concentrated in the *Alphaproteobacteria, Gammaproteobacteria*, and *Actinomycetes* [105]. There is evidence that bacterial lignin degradation is more specific than fungal systems [106]. Bacterial degradation systems are efficient because they exhibit greater genetic adaptability and have very high reproductive rates [107, 108 and 109]. The ability of filamentous and non-filamentous bacterial species of *Acinetobacter* [110], *Arthrobacter* [111], *Bacillus* [112], *Branhamella*, *Brochothrix* [113, 114], *Micrococcus* [114], *Nocardia* [115], *Pseudomonas* [116], *Serratia* [117], *Streptomyces* [118] and *Xanthomonas* [119] to degrade lignin has been established. In some study it has been reported that *Serratia marcescens* exhaust unique catabolic potential for lignin degradation. Indulin (a Kraft pine lignin, practically free of carbohydrates) was used as a bait substrate. Since indulin is free of carbohydrates, it serves as the best source for efficient screening of lignolytic bacteria. Reports on the use of indulin as lignin substrate are well established [104, 120]. Earlier studies indicated that *Serratia marcescens* has lignolytic ability but it has been that during this activity there is no intermediates were not identified [117, 121]. This was the first report on the identification of intermediates involved in the bioconversion of indulin by *Serratia marcescens.* Bollag and co-workers [122] isolated a strain of *Streptomyces* (named strain PS1 by [123] that degraded the chloroacetanilide herbicide metolachlor. The bacteria *Agrobacterium*, *Athrobacteri*, *Bacillus*, *Flavobacterium,* and *Pseudomonas* are the most important microorganisms present in soil involved in the remediation of chlorophenols, but the presence of actinomycetes and fungi is also relevant bacterial degradation of lignin. Numerous bacteria have been reported to decompose lignin and convert it to CO_2 and assimilate lignin degradation products as a carbon source [121, 122, 124, 125]. Certain bacteria such as *Bacillus pumillus* (ATCC12905), *Bacillus stearothermophilus, Rhodobacter sphaeroides, Rhodomonas palustri, Steptococccus lactis, Pseudomonas purrocinna* ATCC 15958*, Pseudomonas flourescens* NRRL B-11, etc., are able to produce microperoxidases, which can mineralize lignin and lignin-containing compounds. *Pseudomonas ovalis, P. putida FK-1,* and *FK-2* are also found to degrade lignin [126].

Only few reports have been documented where actinomycetes contribute to lignocellulose degradation. The biochemistry and enzymes for lignin degradation remains poorly understood but probably extracellular peroxidises [127] such as lignin peroxidase type enzyme [128] facilitates this conversion. Actinomycetes cellulases which are inducible extracellular enzymes attack on cellulose just like a fungal hydrolytic cellulose and xylanases and express similar patterns of production and activities identify in other bacteria and fungi [129]. These lignin peroxidases produced at relatively high level in the presence of wheat straw in *Streptomyces* sp. EC1 [130]. A Novel actinomycete strains having ability to degrade lignocelluloses are isolated from termite gut [131]. Three actinomycetes such as *Streptomyces* sp. *EC22*, *Streptomyces viridosporus* T7A and *Thermomonospora fusca* BD25 are capable of degradation ball-milled wheat [132].

Table 2
List of some lignocelluloses degrading microorganism

Fungi	Bacteria
Sclerotium rolfsii	*Bacillus subtilis*
Sporotrichum thermophile	*Clostridium thermocellum*
Aspergillus niger	*Clostridium stercorarium*
Trichoderma reesei	*Escherichia coli*
Humicola insolvens	*Bacillus polymyxa*
Coriolus versicolor	*Pyrococcus furiosus*
Trichoderma longibrachiatum	*Bacillus pumilus*
Stropharia coronilla	*Clostridium phytofermentans*
Botrytis cinerea	*Thermomonospora*
Phanerochaete chrysosporium	*Ruminococcus*

APPLICATION

Lignocellulose biotechnology offers significant opportunities for utilisation of readily available residual plant biomass to produce numerous value added products. Lignocellulose converted into various different values added products including bio fuels, chemical, cheap energy sources for fermentation, improved animal feeds and human nutrients [133]. Bioconversion of lignocellulosic wastes could make a significant contribution to the production of organic chemicals. Over 75% of organic chemicals are produced from five primary base-chemicals: ethylene, propylene, benzene, toluene and xylene which are used to synthesis other organic compounds, which in turn are used to produce various chemical products including polymers and resins [134]. The production of ethanol from sugars or starch impacts negatively on the economics of the process, thus making ethanol more expensive compared with fossil fuels. So the technology development focus for the production of ethanol has shifted towards the utilisation of residual lignocellulosic materials to lower production costs. The aromatic compounds might be produced from lignin whereas the low molecular mass aliphatic compounds can be derived from ethanol produced by fermentation of sugar generated from the cellulose and hemicellulose. Currently a number of products such as organic acids, amino acids, vitamins and a number of bacterial and fungal polysaccharides such as xanthan are produced by fermentation using glucose as the base substrate but theoretically these same products could be manufactured from “lignocellulose waste”. Ribbons (1987) reported two monomeric potential products Vanillin and Gallic acids which could be derived from lignin [135] Vanillin is used for various purposes including being an intermediate in the chemical and pharmaceutical industries for the production of herbicides, anti-foaming agents or drugs such as papaverine, L-dopa and the antimicrobial agent, trimethoprim. It is also used in domestic products such as air-fresheners and floor polishes [136]. The high price and limited supply of natural vanillin have necessitated a shift towards its production from other sources [137].

Hemicellulose is a readily presented bulk source of xylose in the particular industrial interest from which can be derived xylitol and furfural. Xylitol used in its place of sucrose in food as a sweetener, has odontological applications such as teeth hardening, remineralisation, and as an antimicrobial agent, it is used in chewing gum and toothpaste formulations [138]. The yield of xylans as xylitol by chemical means is only about 50-60% making xylitol production expensive. Various bioconversion methods, therefore, have been explored for the production of xylitol from hemicellulose using microorganisms or their enzymes [139]. Furfural is used in the makeup of furfural phenol plastics, varnishes and pesticides [140] annually produced over 200 000 tones of furfural with a market price of about $1700 per ton [140, 141]. Biotechnology from a capital costs investment perspective is an attractive technology for developing countries since its biodegradation could follow solid-state fermentation comparable to silage or mushroom production, thus making such technology suitable for farms and small industrial plants without the need for large engineering infrastructure. It is also important to emphasize that in order for lignocellulose biotechnology to make meaningful impact on developing countries; suitable bioconversion processes need to be developed on a much wider scale and these countries should begin to pull their meagre resources and biological science expertise in a cooperative and integrated manner towards modern, advance genomics and proteomics technologies for identifying novel lignocellulolytic enzymes and engineering enzymes with improved activities suitable for industrial-scale application.

FUTURE INTERVENTIONS

Lignocellulosic biomass comprising forestry, agricultural and agro-industrial wastes are abundant, renewable and inexpensive energy sources. Lignocellulose wastes are accumulated every year in large quantities, causing environmental problems. Therefore, the development of processes for reuse of these wastes is of great interest. Various treatment methods (physical, chemical and biological) have been used to enhance digestibility of lignocellulosic biomass. In

biological treatment microorganisms are the primary degraders of lignocellulose, which is indigestible to most animals. Several species of bacteria and fungi produce lignolytic enzymes which degraded of lignocellulose. Biological treatments, based on the use of brown, white and soft-rot fungi have been commonly used to degrade the lignin. Basidiomycetes are known to be efficient lignocelluloses degraders and these are considered to be the primary degraders of lignin and lignocellulosic materials. After potential uses of these materials are in pulp and paper industries, production of fuel alcohol and chemicals, protein for food, and feed.

The genetic engineering approach is used recently to reduce lignin content or to modify its composition [142, 143 and 144]. Improvement of lignin degrading potential has been done by using *Agrobacterium* mediated gene transformation system that carrying the genes coding for β-glucuronidase, green fluorescent protein and hygromycin phosphotransferase to the nuclear genome of lignin degrading white-rot fungi such as *Phanerochaete chrysosporium, Ganoderma* sp. RCKK-02, *Pycnoporous cinnabarinus, Crinipellis* sp. RCK-1, *Pleurotus sajor-caju* [145]. Recombinant *Lactobacillus plantarum*, were constructed to express alpha amylase, and cellulase or xylanase genes and have potential to increase the digestibility and nutritive values of forage plants and crop residues [146, 147]. Recombinant enzyme was purified and characterized by the expression of an ascomycete laccase-encoding gene from *Melanocarpus albomyces* into *T. Reesei* [148]. This recombinant enzyme is potentially involved in the degradation of lignin and related aromatic compounds. MnP from white-rot fungi is considered the primary enzyme responsible for bio bleaching of kraft pulps. The main drawback in commercial applications of MnP is the unavailability of the enzyme in large quantities; this can be resolved with the use of DNA recombinant technology [149]. For example, wild-type MnP from white-rot fungi [150] and recombinant MnP (rMnP) expressed in *Pichia pastoris* [151] have been used to remove lignin from cellulose fibers in pulp bleaching experiments [151].

REFERENCES

1. Kim S and Dale BE, Global potential bioethanol production from wasted crops and crop residues. Biomass and Bioenergy, 26: 361-375, (2004).
2. Saha BC, Hemicellulose bioconversion. Journal of Industrial Microbiology and Biotechnology, 30: 279-91, (2003).
3. Sánchez C, Lignocellulosic residues: biodegradation and bioconversion by fungi. Biotechnol Adv, 27: 185-94, (2009).
4. P.S. Nigam, N. Gupta and A. Anthwal. Pre-treatment of agro-industrial residues. In: P. S. Nigam, A. Pandey (eds.), *Biotechnology for agro-industrial residues utilization,* 1st edition. Springer, Netherlands, 2009, pp. 13-33.
5. D. Klemm, B. Philipp, T. Heinze, U. Heinze and W. Wagenknecht. Comprehensive cellulose chemistry. Chichester: Wiley VCH, 1998.
6. Ha MA, Apperley DC, Evans BW, Huxham IM, Jardine WG, Viëtor RJ, Reis D, Vian B and Jarvis MC, Fine structure in cellulose microfibrils: NMR evidence from onion and quince. Plant J, 16: 183-90, (1998).
7. Howard RL, Abotsi E, Jansen van Rensburg EL and Howard S, Lignocellulose biotechnology: Issues of bioconversion and enzyme production. Afr. J. Biotechnol, 2: 602-19, (2003).
8. D. Fengel and G. Wegener .Chemical composition and analysis of wood. *In Wood: Chemistry, Ultra structure, Reactions.* Walter de Gruyter, Berlin, 1984, pp 26-65.
9. Gould JM, Alkaline peroxide delignification of agricultural residues to enhance enzymatic saccharifcation. Biotechnology and Bioengineering, 26: 46–52, (1984).
10. Taherzadeh MJ and Karimi K, Pre treatment of lignocellulosic wastes to improve ethanol and biogas production. Int J Mol Sci, 9: 1621-51, (2008).

11. Eggeman T and Elander RT, Process and economic analysis of pretreatment technologies. Bioresour Technol, 96: 2019-25, (2005).
12. Sun Y and Cheng J, Hydrolysis of lignocellulosic materials for ethanol production. Bioresource Technology, 83: 1-11, (2002).
13. Rubin EM, Genomics of cellulosic bio fuels. Nature, 454: 841-5, (2008).
14. Palmqvist E and Hahn-Hägerdal B, Fermentation of lignocellulosic hydrolysates. I: inhibition and detoxification. Bioresource Technology, 74: 17-24, (2000).
15. Benner R, Maccubbin AE and Hodson RE, Anaerobic biodegradation of the lignin and polysaccharide components of lignocellulose and synthetic lignin by sediment micro flora. Appl. Environ. Microbiol, 47: 998-1004, (1984).
16. Gold MH and Alic M, Molecular-biology of the lignin-degrading basidiomycete *Phanerochaete chrysosporium.* Microbiol. Rev, 57: 605–22, (1993).
17. M. K. Theodorou, W. Y. Zhu, A. Rickers, B. B. Nielsen, K. Gull and A. P. J. Trinci. Biochemistry and ecology of anaerobic fungi. In: M. Howard (ed.), *The mycota VI human and animal relationship*. Springer- Verlag, Berlin Heidelberg, 1996.
18. Bhat KM and Maheshwari R, *Sporotrichum thermophile* Growth, Cellulose Degradation and Cellulase Activity. Appl Environ Microbiol, 53: 2175-82, (1987).
19. Gomes I, Gomes J, Gomes DJ and Steiner W, Simultaneous production of high activities of thermostable endoglucanase and beta-glucosidase by the wild thermophilic fungus *Thermoascus aurantiacus*. Appl Microbiol Biotechnol, 53: 461-8, (2000).
20. Gilbert M, Yaguchi M, Watson DC, Wong KK, Breuil C and Saddler JN, A comparison of two xylanases from the thermophilic fungi *Thielavia terrestris* and *Thermoascus crustaceus.* Appl Microbiol Biotechnol, 40: 508-14, (1993).
21. Valásková V and Baldrian P, Estimation of bound and free fractions of lignocellulose-degrading enzymes of wood-rotting fungi *Pleurotus ostreatus*, *Trametes versicolor* and *Piptoporus betulinus*. Res Microbiol, 157: 119-24, (2006).
22. K. E. L. Eriksson, R. A. Blanchette and P. Ander, Microbial and enzymatic degradation of wood and wood components, Springer, Berlin, Germany, 1990.
23. Ljungdahl LG, The cellulose/ hemicellulase system of the anaerobic fungus *Orpinomyces* PC-2 and aspects of its applied use. Ann N Y Acad Sci, 1125: 308-21, (2008).
24. Yoon JJ, Cha CJ, Kim YS, Son DW and Kim YK. The brown-rot basidiomycete *Fomitopsis palustris* has the endo-glucanases capable of degrading microcrystalline cellulose. *J Microbiol Biotechnol*, 17: 800-5, (2007).
25. Flachowsky G and ØBrskov ER, Anwendung der Nylonbeutal- Met, liode zur Ermittlung des Trockeiisubstanzve~luste von Gersteiistroh vor wid nach & in Wachstum essbarer Pilze. Tierernahrung und Fiitterung, 244-48, (1987).
26. Kamra DN, Kewalramani N, Lall D and Pathak NN, Bio delignification and changes in *invitro* digestibility of sugarcane bagasse treated with white rot fungi. J. Appl. Aniin. Res, 4: 133-40, (1993).
27. Pal D, Kamra DN and Pathak NN, Changes in cheirical composition and *in sacco* degradability of wheat. Straw treated with *Plerrrotrts sajorcaju* and *P. ostreatrrs.* Int. J. Anim. Sci, 14: 61-67, (1999).
28. Martínez AT, Speranza M, Ruiz-Dueñas FJ, Ferreira P, Camarero S, Guillén F, Martínez MJ, Gutiérrez A and del Río JC, Biodegradation of lignocellulosics: microbial, chemical, and enzymatic aspects of the fungal attack of lignin. Int Microbiol, 8: 195-204, (2005).
29. Bayer EA, Chanzy H, Lamed R and Shoham Y, Cellulose, cellulases and cellulosomes. Curr Opin Struct Biol, 8: 548-57, (1998).

30. Weng JK, Li X, Bonawitz ND and Chapple C, Emerging strategies of lignin engineering and degradation for cellulosic biofuel production. Curr Opin Biotechnol, 19: 166-72, (2008).
31. Leightley LE and Eaton RA, *Nia vibrissa*- a marine white rot fungus. Trans Br Mycol Soc, 73: 35-40, (1979b).
32. R. Mouzouras. Patterns of timber decay caused by marine fungi. In: S. T. Moss (ed.), *The biology of marine fungi*. Cambridge University Press, Cambridge UK, 1986, pp. 341-53.
33. Mouzouras R, Decay of mangrove wood by marine fungi. Bot Mar, 32: 65-69, (1989b).
34. Gessner RV and Kohlmeyer J, Geographical distribution and taxonomy of fungi from salt-marsh *Spartina*. Can. J. Bot, 54: 2023-37, (1976).
35. Shyam S Paul, Devki N Kamra and Vadali RB Sastry, Archives of Animal Nutrition, 64: 279–92, (2010).
36. Lynd LR, Weimer PJ, van Zyl WH and Pretorius IS, Microbial cellulose utilization: fundamentals and biotechnology. Microbiol Mol Biol Rev, 66: 506-77, (2002).
37. Chellapandi P and Jani HM, Bra. J. Microbiol., 39: 122-127, (2008).
38. Kumar R, Singh S and Singh OV, Bioconversion of lignocellulosic biomass: biochemical and molecular perspectives. J Ind Microbiol Biotechnol, 35: 377-91, (2008).
39. Henrissat B and Davies G, Structural and sequence-based classification of glycoside hydrolases. Curr Opin Struct Biol, 7: 637-44, (1997).
40. Vaaje-Kolstad G, Westereng B, Horn SJ, Liu Z, Zhai H and Sørlie M, et. al, An oxidative enzyme boosting the enzymatic conversion of recalcitrant polysaccharides. Science, 330: 219-22, (2010).
41. Westereng B, Ishida T, Vaaje-Kolstad G, Wu M, Eijsink VG and Igarashi K, et. al., The putative endoglucanase PcGH61D from *Phanerochaete chrysosporium* is a metal-dependent oxidative enzyme that cleaves cellulose. PLoS One, 6: e27807, (2011).
42. Langston JA, Shaghasi T, Abbate E, Xu F, Vlasenko E and Sweeney MD, Oxidoreductive Cellulose Depolymerization by the Enzymes Cellobiose Dehydrogenase and Glycoside Hydrolase 61. Appl Environ Microb, 77: 7007–15, (2011).
43. Phillips CM, Beeson WT, Cate JH and Marletta MA, Cellobiose dehydrogenase and a copper-dependent polysaccharide monooxygenase potentiate cellulose degradation by *Neurospora crassa*. ACS Chem Biol, 6, 1399-406, (2011).
44. Forsberg Z, Vaaje-Kolstad G, Westereng B, Bunæs AC, Stenstrøm Y, MacKenzie A, Sørlie M, Horn SJ and Eijsink VG, Cleavage of cellulose by a CBM33 protein. Protein Sci, 20: 1479-83, (2011).
45. Sukumaran RK, Singhania RR and Pandey A, Microbial cellulases production, applications and challenges. Journal of Scientific and Industrial Research, 64: 832–44, (2005).
46. Kuhad RC, Gupta R and Khasa YP. Bio ethanol production from lignocellulosic biomass: an overview, in Wealth from Waste, B. Lal, Ed. Teri Press, New Delhi, India, 2010.
47. Kuhad RC, Manchanda M and Singh A, Hydrolytic potential of extracellular enzymes from a mutant strain of *Fusarium oxysporum*. Bioprocess Engineering, 20, 133–35, (1999).
48. Kaur J, Chadha BS, Kumar BA and Saini HS, Purification and characterization of two endoglucanases from *Melanocarpus* sp. MTCC 3922. Bioresour Technol, 98, 74-81, (2007).
49. L. T. Fan, M. M. Gharpuray and Y. H. Lee. *In, Cellulose Hydrolysis*, Springer, Biotechnol. Monographs. Berlin, 1987, pp. 57.
50. Duff SJB and Murray WD, Bioconversion of forest products industry waste cellulosics to fuel ethanol. Bioresour. Technol, 55: 1-33, (1996).
51. Mandels M and Sternberg D, Recent advances in cellulase technology. Ferment. Technol, 54: 267-86, (1976).

52. Wang W, Huang F, Mei Lu X and Ji Gao P, Lignin degradation by a novel peptide, GT factor, from brown rot fungus *Gloeophyllum trabeum*. Biotechnol J, 1: 447-53, (2006).
53. Yang W, Liu J, Wang W, Zhang Y and Gao P, Function of a low molecular peptide generated by cellulolytic fungi for the degradation of native cellulose. Biotechnol Lett, 26: 1799-802, (2004).
54. Savory JG, Breakdown of timber by ascomycetes and fungi *Imperfecti*. Ann Appl Biol, 1954, 41: 336–57, (1954).
55. Leschine SB, Cellulose degradation in anaerobic environments. Annu Rev Microbiol, 49: 399–426, (1995).
56. Kirk K and Cullen D. Enzymology and molecular genetics of wood degradation by white rot fungi. In: R. A. Young, M. Akhtar (eds.), *Environmental friendly technologies for pulp and paper industry*. Wiley, New York, 1998, pp. 273–307.
57. Rohrmann S and Molitoris HP, Screening for wood decay enzymes in marine fungi. Can J Bot, 70: 2116-23, (1992).
58. Pointing SB, Vrijmoed LLP and Jones EBG, A qualitative assessment of lignocellulose degrading enzyme activity in marine fungi. Bot Mar, 41: 293-98, (1998).
59. Yin ZW, Fan BQ and Ren P, Isolation and identification of a cellulose degrading fungus Y5 and its capability of degradating wheat straw. Huan Jing Ke Xue, 32: 247-52, (2011).
60. Wood TM and Wilson CA, Studies on the capacity of the cellulase of the anaerobic rumen fungus *Piromyces communis* P to degrade hydrogen bond-ordered cellulose. Appl Microbiol Biotechnol, 43: 572–8, (1995).
61. Shallom D and Shoham Y, Microbial hemicellulases. Curr Opin Microbiol, 6: 219-28, (2003).
62. Foreman PK, Brown D, Dankmeyer L, Dean R, Diener S and Dunn-Coleman NS et al., Transcriptional regulation of biomass-degrading enzymes in the filamentous fungus *Trichoderma reesei*. Journal of Biological Chemistry, 278: 31988-97, (2003).
63. Wong KK, Tan LU and Saddler JN, Multiplicity of beta-1, 4-xylanase in microorganisms: functions and applications. Microbiol Rev, 52: 305-17, (1988).
64. Kuhad RC, Singh A and Eriksson KE, Microorganisms and enzymes involved in the degradation of plant fiber cell walls. Adv Biochem Eng Biotechnol, 57: 45-125, (1997).
65. Leightley LE, Wood decay activities of marine fungi. Bot Mar, 23: 387-95, (1980).
66. Gomes DJ, Gomes J and Steiner W, Factors influencing the induction of endo-xylanase by *Thermoascus aurantiacus*. J Biotechnol, 33: 87–94, (1994).
67. Liu W, Zhu W, Lu Y, Kong Y and Ma G, Production, partial purification and characterization of xylanase from *Trichosporon cutaneum* SL409. Process Biochem, 33: 331–36, (1998).
68. Kuhad RC, Manchanda M and Singh A, Optimization of xylanase production by a hyperxylanolytic mutant strain of *Fusarium oxysporum*. Process Biochem, 33: 641–47, (1998).
69. Smith DC and Wood TM, Xylanase production by development of a medium and optimization of fermentation parameters for the production of extracellular xylanase and b-xylosidase while maintaining low protease production. Biotechnol Bioeng, 38: 883–90, (1991).
70. Ninawe S and Kuhad RC, Use of xylan-rich cost effective agro-residues in the production of xylanase by *Streptomyces cyaneus* SN32. Journal of Applied Microbiology, 99: 1141–8, (2005).
71. Revankar MS and Lele SS, Enhanced production of laccase using a new isolate of white rot fungus WR-1. Process Biochem, 41(3): 581-588, (2006).
72. Telke AA, Ghodake GS, Kalyani DC, Dhanve RS and Govindwar SP, Biochemical characteristics of a textile dye degrading extracellular laccase from

a Bacillus sp. A. D. R. Bioresour.Technol, 102(2): 1752-1756, (2011).
73. Gorman MJ, Sullivan LI, Nguyen TDT, Dai H, Arakane Y, Dittmer NT, Syed LU, Li J, Hua DH, and Kanost MR, Kinetic properties of alternatively spliced isoforms of laccase-2 form *Tribolium castaneumand* Anopheles gambiae. Insect Biochem. Molec, 42(3): 193-202, (2012).
74. Blanchette R, Krueger E, Haight J, Akhtar M and Akin D, Cell wall alterations in loblolly pine wood decayed by the white-rot fungus, *Ceriporiopsis subvermispora*. J Biotechnol, 53: 203–13, (1997).
75. Makela M, Galkin S, Hatakka A and Lundell T, Production of organic acids and oxalate decarboxylase in lignin-degrading white rot fungi. Enzyme and Microbial Technology, 30: 542–49, (2002).
76. Tien M and Tu CP, Cloning and sequencing of a cDNA for a ligninase from *Phanerochaete chrysosporium*. Nature, 326: 520-3, (1987).
77. Moredo N, Lorenzo M, Domínguez A, Moldes D, Cameselle C and Sanroman A, Enhanced ligninolytic enzyme production and degrading capability of *Phanerochaete chrysosporium* and *Trametes versicolor.* World J Microb Biotechnol, 19: 665–69, (2003).
78. Wang L, Yan W, Chen J, Huang F and Gao P, Function of the iron-binding chelator produced by *Coriolus versicolor* in lignin biodegradation. Sci China C Life Sci, 51: 214-21, (2008).
79. Tien M and Kirk TK, Lignin-degrading enzyme from the hymenomycetes *Phanerochaete chrysosporium.* Science, 22: 661–63, (1983).
80. Kirby N, Marchant R and McMullan G, Decolourisation of synthetic textile dyes by *Phlebia tremellosa*. FEMS *Microbiol Lett,* 188, 93-6, (2000).
81. Kirk TK and Farrell RL, Enzymatic "combustion": the microbial degradation of lignin. The microbial degradation of lignin. Annu Rev Microbiol, 41: 465-505, (1987).
82. Novotny C, Erbanova P, Cajthaml T, Rothschild N, Dosoretz C, Sasek V and *Irpex lacteus*, A white rot fungus applicable to water and soil bioremediation. Appl. Microbiol. Biotechnol, 54: 850–53, (2000).
83. Tuor U, Wariishi H, Schoemaker HE and Gold MH, Oxidation of phenolic arylglycerol beta-aryl ether lignin model compounds by manganese peroxidase from *Phanerochaete chrysosporium* oxidative cleavage of an alpha-carbonyl model compound. Biochemistry, 31: 4986–95, (1992).
84. Rahman MM, Begum MF and Khan MSI, Isolation, identification and cultural optimization of native bacteria isolates as a potential bioconversion agent. Journal of Applied Science Research, 5: 1652-1662, (2009).
85. Leonowicz A, Cho NS, Luterek J, Wilkolazka A, Wojtas-Wasilewska M, Matuszewska A, Hofrichter M, Wesenberg D and Rogalski J, Fungal laccase: properties and activity on lignin. J Basic Microbiol, 41: 185-227, (2001).
86. Vargas-García MC, Suárez-Estrella F, López MJ and Moreno J, Effect of inoculation in composting processes: modifications in lignocellulosic fraction. Waste Management, 27: 1099–107, (2007).
87. Tamagawa Y, Hirai H, Kawai S and Nishida T, Removal of estrogenic activity of endocrine-disrupting genistein by ligninolytic enzymes from white rot fungi. FEMS Microbiology Letters, 244: 93–8, (2005).
88. Cohen S, Belinky PA, Hadar Y and Dosoretz CG, Characterization of catechol derivative removal by lignin peroxidase in aqueous mixture. Bioresource Technology, 100: 2247–53, (2009).
89. Huang DL, Zeng GM, Feng CL, Hu S, Zhao MH and Lai C, et al, Mycelial growth and solid-state fermentation of lignocellulosic waste by White-rot fungus *Phanerochaete chrysosporium* under lead stress. Chemosphere, 81: 1091–7, (2010).

90. Srinivasan C, Dsouza TM, Boominathan K and Reddy CA, Demonstration of Laccase in the White Rot Basidiomycete *Phanerochaete chrysosporium* BKM-F1767. Appl Environ Microbiol, 61: 4274-7, (1995).
91. Gill K and Arora S, Effect of culture conditions on manganese peroxidase production and activity by some white rot fungi. J Ind Microbiol Biotechnol, 30: 28-33, (2003).
92. Isroi et al, Biological pretreatment: Review. BioResources, 6(4): 5224-5259, (2011).
93. Wanapat M, Comparative study between swamp buffalo and native cattle in feed digestibility and potential transfer of buffalo rumen digest into cattle. Asian- Aust. J. Anim. Sci, 16: 504-10, (2003).
94. Weber C, Farwick A, Benisch F, Brat D, Dietz H and Subtil T et al., Trends and challenges in the microbial production of lignocellulosic bio alcohol fuels. Appl. Microbiol. Biotechnol, 87: 1303-15, (2010).
95. V. S. Bisaria. Bio processing of agro-residues to value added products. In: A. M. Martin (ed.), *Bioconversion of Waste Materials to Industrial Products*, 2nd edition. Chapman and Hall, UK, 1998, pp. 197-246.
96. Raman B, Pan C, Hurst GB, Rodriguez M Jr, McKeown CK and Lankford PK et al., Impact of pre-treated Switch grass and biomass carbohydrates on *Clostridium thermocellum* ATCC 27405 cellulosome compositions: a quantitative proteomic analysis. PLoS ONE, 4: e5271, (2009).
97. Lamed R, Setter E and Bayer EA, Characterization of a cellulose- binding, cellulase-containing complex in *Clostridium thermocellum.* J Bacteriol, 156: 828-36, (1983).
98. Wilson CA and Wood TM, The anaerobic fungus *Neocallimastix frontalis*. Isolation and properties of a cellulosome- type enzyme fraction with the capacity to solubilise hydrogen-bond-ordered cellulose. Appl. Microbiol. Biotechnol, 37: 125-29, (1992).
99. Béguin P and Aubert JP, The biological degradation of cellulose. FEMS Microbiol Rev, 13: 25-58, (1994).
100. George SP, Ahmad A and Rao MB, A novel thermostable xylanase from *Thermomono spora* sp.: influence of additives on thermostability. Bioresour Technol, 78: 221, (2001).
101. Simpson HD, Haufler UR and Daniel RM, An extremely thermostable xylanase from the thermophilic *Eubacterium Thermotoga.* Biochem J, 277: 413-7, (1991).
102. Ramachandra M, Crawford DL and Hertel G, Characterization of an extracellular lignin peroxidase of the lignocellulolytic actinomycete *Streptomyces viridosporus*. Appl Environ Microbiol, 54: 3057-63, (1988).
103. Crawford DL and Crawford RL, Enzyme Microbiol, Technol, 2: 11-22, (1980).
104. Berrocal M, Rodriguez J, Ball AS, Pe´rez-Leblic MI and Arias ME, Solubilisation and mineralisation of ^{14}C lignocelluloses from wheat straw by *Streptomyces cyaneus* CECT 3335 during growth in solid-state fermentation. Appl Microbiol Biotechnol, 48: 379–84, (1997).
105. Bugg TD, Ahmad M, Hardiman EM and Singh R, The emerging role for bacteria in lignin degradation and bio-product formation. Curr Opin Biotechnol, 22: 394-400, (2011).
106. Vicuna R, Bacterial degradation of lignin. Enzyme Microb. Technol, 10: 646–55, (1988).
107. Furukawa K and Miyazaki T, Cloning of a gene cluster encoding biphenyl and chlorobiphenyl degradation in *Pseudomonas pseudoalcaligenes.* J Bacteriol, 166: 392-8, (1986).
108. Cespedes R, Salas L, Calderon I, Gonzales B and Vicuna R, Microbial and biochemical characterization of a bacterial consortium isolated from decaying wood by growth on ft-0-4 lignin-related dimeric compound. Arch. Microbiol, 15: 162–70, (1992).
109. Vasudevan N and Mahadevan A, Utilization of complex phenolic compounds by *Acinetobacter* sp. Appl.

Microbiol. Biotechnol, 37: 404–07, (1992a).
110. Vasudevan N and Mahadevan A, Degradation of lignin and lignin derivatives by *Acinetobacter* sp. J. Appl. Bacteriol, 70: 169–76, (1991).
111. Kerr TJ, Kerr RD and Benner R, Isolation of bacterium capable of degrading pea nut hull lignin. App. Environ. Microbiol, 46: 1201–06, (1983).
112. Gurujeyalakshmi G and Mahadevan A, Dissimilation of ferulic acid by *Bacillus subtilis. Curr. Microbiol*, 16: 69–73, (1987a).
113. Gurujeyalakshmi G and Mahadevan A, Degradation of guaicol glyceryl ether (GGE) by *Bacillus subtilis.* Appl. Microbiol. Biotechnol, 26: 289–93, (1987b).
114. Kumar L, Rathore V and Srivastava H, 14C-[lignin]-lignocellulose biodegradation by bacteria isolated from polluted soil. Indian J Exp Biol, 39: 584-9, (2001).
115. Trojanowski J, Haider K and Sundman V, Decomposition of 14C-labelled lignin and phenols by a *Nocardia* sp. Arch Microbiol, 114: 149-53, (1977).
116. Kaplan DL and Hartenstein R, Decomposition of lignins by microorganisms. Soil Biology and Biochemistry, 12: 56–75, (1980).
117. Perestelo F, Falcon MA, Camicero A, Rodriguez A and De la Fuente G, Limited degradation of industrial, synthetic and natural lignins by *Serratia marcescens.* Biotechnol. Lett, 16: 299–302, (1994).
118. McCarthy AJ and Broda P, Screening for lignin degrading actinomycetes and characterization of their activity against 14C- labelled wheat lignocelluloses. J. Gen. Microbiol, 130: 2905–13, (1984).
119. Kern HW, Bacterial degradation of dehydropolymers of coniferyl alcohol. Arch Microbiol, 138: 18-25, (1984).
120. Vasudevan N and Mahadevan A, Degradation of indulin by *acinetobacter* sp. Indian J. Expt. Biol, 31: 252–55, (1993).
121. Rhoads TL, Mikell AT Jr and Eley MH, Investigation of the lignin-degrading activity of *Serratia marcescens*: biochemical screening and ultra structural evidence. Can J Microbiol, 41: 592-600, (1995).
122. Saxena A, Zhang RW and Bollag JM, Microorganisms capable of metabolizing the herbicide metolachlor. Appl Environ Microbiol, 53: 390-6, (1987).
123. Speedie MK, Pogell BM, MacDonald MJ, Kline R Jr and Huang YI, The actinomycetes. 20: 315-35, (1988).
124. Haider K, Trojanowski J and Sundman V, Screening for lignin degrading bacteria by means of 14C-labelled lignins. Arch Microbiol, 119: 103-6, (1978).
125. Gradziel K, Haider K, Kochmanska J, Malarczyk E and Trojanowski J, Bacterial decomposition of synthetic ^{14}C-labeled lignin and lignin monomer derivatives. Acta Microbio Pol, 27: 103-09, (1978).
126. K. Kawakami. Bacterial degradation of lignin compounds I, II and III. In: Dickinson and G. J. F. Pugh (eds.), *Mokkuzai Gakkaishi*. Academic Press, New York, 1975.
127. Mercer DK, Iqbal M, Miller P and McCarthy AJ, Screening actinomycetes for extracellular peroxidase activity. Appl Environ Microbio, 62: 2186-90, (1996).
128. Adhi TP, Korus RA and Crawford DL, Production of major extracellular enzymes during lignocellulose degradation by two *Streptomycetes* in agitated submerged culture. Appl Environ Microbiol, 55: 1165-8, (1989).
129. McCarthy A.J, Lignocellulose-degrading actinomycetes. FEM Microbiology Letters, 46: 145–63, (1987).
130. Godden B, Ball AS, Helvenstein P, McCarthy AJ and Penninckx MJ, Towards elucidation of the lignin degradation pathway in actinomycetes. J. Gen. Microbiol, 138: 2441-8, (1992).
131. Pasti MB, Pometto AL 3rd, Nuti MP and Crawford DL, Lignin-solubilizing ability of actinomycetes isolated from termite (*Termitidae*) gut. Appl Environ Microbiol, 56: 2213-8, (1990).
132. Trigo C and Ball AS, Is the solubilized product from the degradation of lignocellulose by actinomycetes a

precursor of humic substances? Microbiology, 140: 3145-52, (1994).
133. Malherbe S and Cloete TE, Lignocellulose biodegradation: fundamentals and applications: A review. Environ. Sci. Biotechnol, 1: 105-14, (2003).
134. Coombs J, EEC resources and strategies. Phil. Trans. R. Soc. Lond. Ser. A, 321: 405-22, (1987).
135. Ribbons RW, Chemicals from lignin. Phil. Trans. R. Soc. Lond. Ser. A, 321: 485-94, (1987).
136. Walton NJ, Mayer MJ and Narbad A, Molecules of interest: Vanillin. Phytochemistry, 63: 505-15, (2003).
137. Priefert H, Rabenhorst J and Steinbüchel A, Biotechnological production of vanillin. Appl Microbiol Biotechnol, 56: 296-314, (2001).
138. Roberto IC, Mussatto SI and Rodrigues RCLB, Dilute-acid hydrolysis for optimization of xylose recovery from rice straw in a semi-pilot reactor. Indust. Crops Prod, 17: 171-6, (2003).
139. Nigam P and Singh D, Processes for fermentative production of xylitol – a sugar substitute: A review. Process Biochem, 30: 117-24, (1995).
140. Montané D, Salvadó J, Torras C and Farriol X, High-temperature dilute-acid hydrolysis of olive stones for furfural production. Biomass Bioenergy, 22: 295-04, (2002).
141. K. J. Zeitch, The chemistry and technology of Furfural and Its Many By-Products. In: Zeitch (ed.), Elsevier, 2000, pp. 358.
142. Ralph J, Akiyama T, Kim H, Lu F, Schatz PF and Marita JM et. al., Effects of coumarate 3- hydroxylase down-regulation on lignin structure. J Biol Chem, 281: 8843-53, (2006).
143. Chapple C, Ladisch M and Meilan R, Loosening lignin's grip on biofuel production. Nat Biotechnol, 25: 746-8, (2007).
144. Chen F and Dixon RA, Lignin modification improves fermentable sugar yields for biofuel production. Nat Biotechnol, 25: 759-61, (2007).
145. Sharma KK and Kuhad RC, Genetic transformation of lignin degrading fungi facilitated by *Agrobacterium tumefaciens*. BMC Biotechnol, 10: 67, (2010).
146. Scheirlinck T, Mahillon J, Joos H, Dhaese P and Michiels F, Integration and expression of alpha-amylase and endoglucanase genes in the *Lactobacillus plantarum* chromosome. Appl Environ Microbiol, 55: 2130-7, (1989).
147. Scheirlinck T, DeMeutter J, Arnaut G, Joos H, Claeyssens M and Michiels F, Cloning and expression of cellulase and xylanase genes in *Lactobacillus plantarum*. Appl. Microbiol. Biotechnol, 33: 534–41, (1990).
148. Kiiskinen LL, Kruus K, Bailey M, Ylösmäki E, Siika-Aho M and Saloheimo M, Expression of *Melanocarpus albomyces* laccase in *Trichoderma reesei* and characterization of the purified enzyme. Microbiology, 150: 3065-74, (2004).
149. Xu H, Scott GM, Jiang F and Kelly C, Recombinant manganese peroxidase (rMnP) from *Pichia pastoris*. Part 1: Kraft pulp delignification. Holzforschung, 64:137–143, 2010.
150. Harazono K, Kondo R and Sakai K. Bleaching of Hardwood Kraft Pulp with Manganese Peroxidase from *Phanerochaete sordida* YK-624 without Addition of MnS0 (inf4). Appl Environ Microbiol, 62:913–917, 1996.
151. Gu L, Lajoie C and Kelly C, Expression of a *Phanerochaete chrysosporium* manganese peroxidase gene in the yeast *Pichia pastoris*. Biotechnol Prog, 19:1403–1409, 2003.