Chapter 11

Potential of Cellulosic Ethanol to Overcome Energy Crisis in Pakistan

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Additional information is available at the end of the chapter

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Abstract

Liquid biofuel industry in Pakistan may become a promising source for saving our foreign exchange and environment. Currently, bioethanol production is dependent on cane molasses, a product of sugar industry. Harnessing of more bioethanol from lignocellulosic waste crop residue has potential to respond to the fuel scarcity. Lignocellulose exists in nature as a polymer and serves as the largest sink for fixed global carbon and could be used both as a carbon source for microbial growth-assisted bioethanol production and for fabricating enzymes for more energetic simultaneous production to represent an important segment of the renewable energy sector. An exciting aspect of this research is the development of new biorefining techniques that facilitate the extraction of sugar-derived biofuel by processing of waste crop residues by employing novel nature inspired lignolytic enzyme. Further research will explore more avenues for stabilization of system in terms of process parameters for optimum bioethanol yield from enzymatically hydrolyzed lignin waste streams. The chapter can be considered as an anticipatory work and exploration of new dimensions for promotion of nature-inspired enzyme-assisted lignocellulose-based bioethanol production industry, which maximizes sustainable development opportunities especially in energy sector.

Keywords: crop residue conversion into biofuel, agriculture waste bioethanol, enzymatic ethanol, lignin biofuel, sustainable ethanol

1. Introduction

There has been a universal consensus that greenhouse gases (GHG) such as methane (CH), carbon dioxide (CO) and nitrous oxide (NO) are the main cause of global warming. This extreme apprehension forced many nations of the world to reach treaty on Japanese Protocol.

Pakistan signed the Kyoto Protocol on Climate Change in 2004 and accredited the scientific invention's potential as a possible way to control the emissions of GHG [1]. Transportation consumes approximately 27% of primary energy [2]. In EU25 countries, transportation consumes about 28% of the energy and more than 80% is for lane transport [3]. The current oil requirement is around 12 million tons a day and there is forecasting that it may increase to about 16 million tons per day in 2030. The 30% of the world oil production is used to fulfill the fuel requirements for transport. In the current energy blend in Pakistan, the share of petroleum products is about 40%. Its use has grown up suddenly, mainly by fuel oil and the gasoline [4]. The immediate focus of this chapter is to review the current developments in the dimensions of waste crop residue-based bioethanol production and applications of nature-inspired enzymes efficient production of the liquid fuel in Pakistan.

Pakistan is facing severe energy crisis in these days which is resulting in many social and economic problems. Solution of this crisis might come from alternative(s), addressing cheap and eco-friendly fuel sources. This utmost and urgent requirement is likely to be achieved by biomass resources that have been mainly ignored previously, while they are accessible in enough quantities to solve the energy crisis in the country. Agriculture has remained the basis of Pakistan's economy as it provides employment to 45% of the population and provides feedstocks to agro-based industry. Clean energy supply is critical in agricultural areas in Pakistan where biofuels are currently not an option because of the lack of cost-effective and efficient biofuel production technologies, although villagers depend on conventional diesel for powering different agriculture machinery and gasoline for transporting agricultural goods from farm/market to end consumers. Fuel shortage has also led to a cut in electricity production. It is thus clear that the major limiting factor is energy which creates barriers for developing economies. Careful estimates show that by 2050, Pakistan's energy needs will increase three times without a concurrent increase in supply. Pakistan plans to cut natural gas supply by around 4,247,527 m^3/d to fertilizer plants and compressed natural gas (CNG) pumps to increase electricity supply to cities facing daily rolling blackouts. In 2012, Pakistan's natural gas supply had about 15 billion m^3/year deficit with increasing tendency. Large biofuel production plants that can contribute significant amounts of sustainable fuel are the only solution to supplement the power shortage in the country. Rise in conventional fuel price and its continuous depletion naturally brings great demand for the innovative biofuel production technologies as a Clean Energy Solution in Pakistan. The valuable progress starts with the beneficial use of the waste material and crop residues as feedstocks which otherwise represent environmental liabilities. The development in this sector will further provide opportunities to create multiple symbiotic partnerships among the farmers and the business community.

Ethanol is an important energy source which has the huge potential to lessen the sole dependency on fossil-derived fuels and alleviate hazards to our environment. Additionally, it is an ideal precursor molecule because of its promising potential as fuel, beverage, feedstock, antiseptic and industrial solvent. Currently, it is replacing around 3% of the gasoline that is derived from fossils throughout the world which is compatible with petroleum and recommended for transportation in both blended and pure forms. The consumption of ethanol is around 1.6 million tons and consumption of fuel ethanol can be increased up to 160,000 tons by 10% blending of bioethanol in petrol. According to Chris Somerville, director of the

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Energy Biosciences Institute, USA, annual production of ethanol from corn is around 13–14 billion gallons in the United States, which is equal to 10% of gasoline use. In Brazil, 40% of liquid transportation fuel is bioethanol and 15% of the nation's electricity is deriving from it. Therefore, it is assumed that current technology is healthy enough to produce ethanol as an alternative fuel to some extent for immediate partial replacement of oil. However, corn ethanol which is already in use has several drawbacks as corn is a food crop. Furthermore, when cost-to-benefit ratio regarding equipment and processing involved in planting, harvesting and transporting corn ethanol are considered, it becomes incomparable with gasoline. Therefore, hardy, fast-growing plants, like switchgrass, elephant grass and miscanthus, are more favorable feedstocks options. These grasses can grow up to 10 feet height, thrive on marginal land and can survive even with little or no fertilizer [5]. Moreover, cellulose-rich waste material of paper and other industries and waste crop residues can also be considered attractive options for cost-effective and even zero cost bioethanol productions by using nature inspired enzymatic processes.

2. Biomass: a cheap and sustainable biofuel resource

Pakistan is an agricultural country and huge capacity of biomass in the form of waste crop residues such as rice straw, wheat straw, cotton stalks, maize stalks, sugarcane tops and so on are available for bioethanol production. Pakistan annually generates around 69 million tons of field-based crop residues. Field-based crop residues are generally considered useless and it has been estimated that 50 million tons of residue/waste is produced every year from major crops (including 6.88 million tons of sugarcane bagasse). These are either burned in the fields/ homes or buried in the land. Direct burning of this field base left over emits carbon dioxide and smoke which are hazardous for health and a source of ozone with risks to the atmosphere. Excluding domestic consumption and commercial usage, the net available resource potential of four crops, that is (wheat, cotton, rice and corn) for biomass power generation, is estimated to be about 10.942 million tons [6]. These estimations showed that Pakistan is endowed with abundant availability of biomass resources, which can be economically exploited for developing a sustainable biomass energy system, because the country has been perennially facing power demand-supply gap, which is currently estimated at 4500–5500 MW [7]. The system is being maintained by resorting to load shedding, often extending up to 12–16 h. Pakistan has strategies to add 9700 MW of electricity generation capacity by 2030 as per the Medium Term Development Framework [8], which would partially take care of current fuel scarcities. In this context, power generation through biomass can also play an important role in bridging the overall demand-supply gap. It would be essential to expand and diversify the resource base, particularly in the context of continuous access to electricity in all regions of the country. Large numbers of industries in Pakistan are currently dependent on liquid fuels for meeting their captive demand for electricity and heat. The situation is therefore ideally suited for promoting biomass-based liquid biofuel production as a sustainable and renewable alternative for the industrial sector as well. If only field left over crop residues are used for production of bioethanol, even then a sufficient amount of bioethanol can be produced to cut oil import and improve the profitability of the farming sector.

It is known that some developed countries like the United States and Brazil are largest bioethanol producers and ethanol production in these countries is achieved by fermentation of corn glucose [9]. The production of ethanol from molasses is not new, but some areas need to be researched for enhanced yield. Until now, in Pakistan, sugar mills distilleries are operational for ethanol production using molasses, but in order to utilize molasses fully and get maximum benefit, it is important to increase the number of distillery units on one hand and assurance of possible involvement of efficient enzymes and engineered microbes on the other hand. Furthermore, application of mathematical imitations would be used to explore efficient way for optimized yield without intervention of any pilot plant [10, 11]. The major steps for large-scale microbial production of ethanol are fermentation of sugars, distillation and dehydration.

3. Microbial fermentation

Different ethanologenic microbes have been known to have promising qualities like limited growth requirements, genetically amenable, higher sugar and ethanol tolerance. Bioethanol production by two strains (mutant and wild) of yeast *Kluyveromyces marxianus* have been documented [12]. Wild strain showed maximum specific growth rate at 40°C while mutant showed maximum specific growth and ethanol formation rates at 45°C from fermentation of diluted molasses. Results of this study anticipated that large-scale production may also be economically feasible by employing these microbes. Yeast-assisted bioethanol production process is more common and commercially applicable method in Pakistan [13]. *Zymomonas mobilis* is also attracting more attention for bioethanol production due to less process limitations [14, 15]. Different experimental studies in this regard revealed that optimum ethanol production up to 55.8 g L^{-1} can be achieved by *Zymomonas mobilis* at 30°C after 48 h of retention time [16, 17].

Sugar beet, molasses and sugarcane juice are one of the most vital and easily accessible raw materials for the fermentative production of alcohol. The increased cost of molasses has triggered many distilleries to search alternate sources of feed stocks for the production of bioethanol in Pakistan. In starch industry, a by-product called enzose hydrol contains 5, 12, 56 and 5% of oligosaccharides, maltose, glucose and maltotriose, respectively, and is a cheap and good source of fermentable sugars. Mostly oligo-saccharides and maltotriose are not completely consumed by ethanologenic microbes and therefore need pretreatment [18]. Similarly, bioconversion of cellulose into ethanol can be accompanied by various microbes as well as by some filamentous fungi, including *Neurospora crassa* [19, 20] *Zymomonas* [21], *Trichoderma viride* [22], *Paecilomyces* sp. [23], *Zygosaccharomyces rouxii* [24] and *Aspergillus* sp. [25], termites' gut enzymes, genetically engineered bacteria such as *Escherichia coli* [26] and thermophilic, anaerobic bacteria, such as *Clostridium thermocellum* [27]. Thus, certain possible methods need to be designed for economical production of ethanol from agricultural farm residues by employing most effective microbes [28]. Among such agro-based wastes, wheat straw is one of the most plentiful crop residues which has broadly been studied and is abundantly available too [29].

Current investigations are focusing on pretreatment of the hard biomass, that is, lignocellulosic sugarcane bagasse, rice straw and wheat straw and subsequent production of ethanol from the pretreated biomass using ethanologenic microorganism. Different fungal species have promising potential for breaking down lignin and therefore may be applied for efficient ethanol fermentation. Hypothetical yield of ethanol is 0.511 g per gram of glucose consumed. Practically, this yield cannot be achieved because part of fermentable glucose is consumed for cell maintenance, for synthesis of by-products like glycerol and lactic acid and therefore is not completely converted into ethanol. Nevertheless, at the manufacturing level, under ideal conditions, it remains 90–95% of the hypothetical yield [30]. Ethanol formation represents a specific loop of the general cellular metabolism; however, its general production route is shown in **Figure 1** [31].

Generalized bioethanol production is as follows [31].

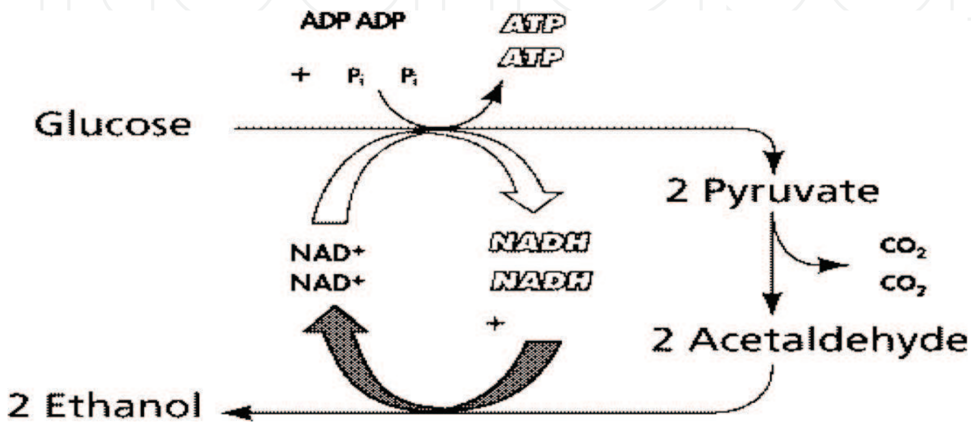

Figure 1. Part of the fermentative metabolism directly involved in the ethanol production.

4. Bioethanol production potential of industrial sector in Pakistan

Few industries in Pakistan are already involved in bioethanol production from by-products or industrial effluents, but it is necessary to develop nature inspired bioethanol production on a large scale that may not only provide a solution to Pakistan's power shortages but can also be profitable enough to render their viability in local conditions. The biomass like rice straw, sugarcane molasses, bagasse and wheat stubble are the chief resources of lignocellulose feedstocks worldwide [32]. One of the largest available biomass is rice straw which is about 7.31×10^{14} rice stubbles per year in world and 90% of its annual global production comes from the Asia [33]. Another abundantly available biomass, a by-product of sugarcane processing, is the sugarcane bagasse which represents important source for fuel generation systems and ethanol production due to its high easily accessible sugar contents for fermentation [34].

Sugar industry is the biggest agro-industry in Pakistan after textile and has been playing key role in the production of ethanol. There are about 76 sugar mills in Pakistan already which are producing seasonal ethanol from around 2.5 million metric tons (MMT) of molasses. However, being an agricultural country, the best option is second-generation ethanol. However, for this, the complex lignin-cellulose-hemicelluloses matrix of the biomass has to be broken and the carbohydrate polymers need to undergo hydrolyses to yield fermentable sugars. The important source of the livelihood of farmers is sugar industry and their 70% population is dependent on it. The yield of sugar in Pakistan is about 85.95 kg per 100 kg of sugarcane. The molasses production from sugarcane is approximately 40 kg per ton of cane from which ethanol production is approximately10 L. There is 270,000 tonnes per annum current production capacity for ethanol of fuel grade in our country which can readily be increased up to 400,000 tonnes per annum through the rise in employments and feedstock like waste crop residue [18, 35]. This molasses-to-bioethanol conversion process is conducted in distilleries. But most of the distilleries are located onsite in sugar mills which make the production cycle an integrated one. The mills, after processing sugarcane, store the molasses in storage tanks on-site and then pass it on to the distilleries for bioethanol production. Simple molecular sieve technology is used for bioethanol production in most of these mills which requires 1.5 million USD capital expenditure and can be completed in 5–6 months.

AL-Abbas sugar mill production plant is situated exactly in the center of one of the huge sugarcane growing areas of province Sindh at Mirwah Gorchani. This area is also known to be the most fruitful regarding sugarcane cultivation in Pakistan, assuring the supply of good crop of sugarcane throughout the entire season for the sugarcane plant. The plant is linked to the national highway by means of a mile of metal led road and is also accessible by a web of many other roads from different directions which facilitate transport of sugarcane from the plantation sites to the sugarcane plant. Total crushing potential of this sugarcane crushing plant is about 7500 M ton per day. AL-Abbas sugar mill established the largest ethanol distillery plant in 1999. The plant design is equipped with highly advanced French technology using multieffect vacuum distillation. The ethanol production capacity of unit-I is approximately 87,500 L per day. The growing demand of ethanol has urged the management to set up unit-II with the same capacity of ethanol production. The bioethanol-based power plant of AL-Abbas sugar mill has 15 MW electricity production capacity.

Shakarganj sugar mill is located in Jhang, Pakistan. They are producing three different types of ethanol, that is, concentrated ethyl alcohol, denatured spirit and methylated spirit for industrial and alternate source of energy usage. The mill is exporting approximately 90% of its total ethyl alcohol and is a four-time award winner for the highest export of ethyl alcohol. The unit produces anhydrous alcohol employing eco-friendly dry dehydration technology. The denatured and methylated spirit is in high demand in local wood product and paint industries.

Another bioethanol-producing sugar mill is located in Nankana, Sheikhupura, Pakistan. The ethanol production potential of this distillery is 125,000 L/day. Besides this capacity, the distillery also produces ethanol of fuel grade with 99.8% purity from the mill molasses. State-of-the-art distributed control system (DCS) which not only promises for increased steadiness and but also approachability of plant is used. The distillery system is established with fewer number of devices and lesson wiring. The distillery can cut in half the costs related to applying and sustaining the loops by incorporating the transmitter controllers into the process and by opting not to tie any critical loops back into the DCS. The distillery is equipped with ultra-modern machinery and is working on International Standard Operating processes to carry them to produce high-quality products and is meeting the demands of end users.

Crystalline Chemical Industries (Pvt.) Ltd (CCI) also practices sugarcane molasses fermentation for ethanol production, located in Sargodha, Pakistan. This unit of distillation exports about 90% of its ethanol produce.

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Habib sugar mills Ltd. has industrial alcohol production capacity up to 142,500 L/day. Pinnacle distilleries (Pvt.) Ltd. is producing rectified spirit for portable applications, technical alcohol, anhydrous ethanol 99.7% minimum for manufacturing use. The fuel grade alcohol is produced up to 30,000 tons per year.

Almost all sugar industries in Pakistan are producing bioethanol mainly from molasses containing feasible level of fermentable sugars. Presently, the biomass proportions which can be economically converted into ethanol are sugar (sugarcane) and starch (e.g. corn). In future, there will be plentiful industrial scale progress in the subject of lignoethanol where the hard part of a plant (cellulose) will be converted into fermentable sugars and consequently converted to bioethanol. After microbial fermentation, the produce is subjected to distillation, dehydration and then is condensed for quality improvement and water and other impurities removal. However, due to high cost in the form of energy input, this traditional process is replaced with some energy saving processes (molecular sieve) mainly to avoid distillation completely for dehydration. This process involves the use of ethanol vapors under pressure and allows these vapors to pass through molecular sieve beads bed. The energy saving by this technology of dehydration accounts for 3000 btus/gallon (840 kJ/L) than that of azeotropic distillation.

If all raw sugarcane molasses is converted to bioethanol, then it has the potential to substitute 5–7% consumption of gasoline. This will be a very important contribution in future to lessen the burdens on Pakistan economy. The government of Pakistan should make policy to endorse the blending of ethanol in transportation fuels as early as it becomes conceivable [18]. With the production of bioethanol from Pakistan's own raw molasses, about 600 million of precious foreign exchange can be saved [36]. Besides this, other advantages of ethanol usage are good engine performance and better yields; it burns more efficiently and keeps our environment clean and more easily biodegradable, as well as consistent with the global focus on biofuel. No doubt this is a most effective way for production of bioethanol from raw/waste material; however, involvement of a variety of waste biomass or crop residue will be more optimistic for solution of energy issues. The main factor in ethanol production is the content of lignocellulose present in substrates which will be hydrolyzed by different hydrolyzing agents to provide fermentable glucose [37, 38]. The nature-inspired enzymes from wood fungus and termite may be used as an extra bonus in the presence of exiting bioethanol production technologies, which can convert the long chains of polysaccharides into monosaccharaides. Different industries like forestry, pulp and paper, agriculture and food processing including municipal solid waste (MSW) and animal wastes are major producers of lignocellulosic waste materials [39, 40].

5. Present challenges for bioethanol production from lignocellulosic feedstocks

Currently, lignocellulolytic enzymes are derived from fungus, gut of termite and certain bacteria [41]. Established technology for bioethanol in Pakistan is relatively of low-tech approach to meet some needs by employing molasses and some selective biomass. Such limitations with biomass make the process and yield profit limited. At the same time, the farmers and agribusinesses cannot access recent technologies that may greatly expand the use of bioethanol to meet the demand for power in many applications.

The current energy scenario warrants the demand for research and development of biomass-based biofuel production systems. Biomass, due to its renewable nature and abundance, is becoming an increasingly attractive fuel source. Lignin, the second most abundant biomass constituting aromatic biopolymer on Earth, is highly recalcitrant to depolymerization. Lignin serves as bonding for hemicellulose and cellulose and creates an obstacle for penetration of any solution or enzyme to lignocellulosic structure which is the major structural component of all plants and can be depolymerized to fermentable sugars. Microbes enhance the conversion of lignin into fermentable sugars but there are some hurdles which need to be removed first. Recalcitrant nature of lignin could be tackled through different biocatalysts due to their non-hazardous and eco-friendly nature. Therefore, lignocellulolytic microorganisms like fungi and some bacteria are considered as promising biomass degraders especially for large-scale applications due to their potential yields of extracellular synergistically acting enzymes into the environment. These enzymes can contribute significantly in degradation of lignocellulosic material by converting long chain polysaccharides into their 5- and 6-carbon sugar components [42, 43]. Although lignin resists attack by most microorganisms, basidiomycetes, white-rot fungi are able to degrade lignin efficiently [44, 45]. Lignocellulolytic enzymes-producing fungi are widespread and include species from the ascomycetes (e.g. *Trichoderma reesei)* and basidiomycetes phyla such as white-rot (e.g. *Phanerochaete chrysosporium)* and brown-rot fungi (e.g. *Fomitopsis palustris)* [46, 47]. Few basidiomycetes, for example, *P. eryngii, P. chrysosporium* and *T. versicolor* can act as biocatalysts for ethanol production by having potential for lignin degradation/depolymerization. Ethanol fermentation requires high concentration of sugar solutions; therefore, biocatalytic conversion of lignocellulosic material into hydrolysate containing high concentration of sugar will be incentive for decreasing production cost. Therefore, variety of lignocellulytic material (wheat straw, rice straw and rice husk) could be degraded by basidiomycetes and subject to ethanologenic fermentation for ethanol production cost-effectively. However, some strains of white-rot fungi have promising potential to degrade lignin by simultaneous attack on lignin, hemicellulose and cellulose, whereas few can selectively work just on lignin. It is pertinent here to note that synergistic biocatalytic ability of white rot fungi would be source of efficient depolymerization method and will be helpful in proving that the heteropolymer lignin represents an untapped resource of renewable aromatic chemicals [48, 49]. Lignocellulosic biofuel production is not yet economically competitive with fossil fuels; therefore, to ensure successful utilization of all sugars is important for improving the overall economy especially in terms of maximum theoretical yield. Xylose is one of the most abundant sugars in lignocellulosic hydrolysate. Therefore, over expression of xylose isomerase will facilitate complete utilization of xylose present in hydrolysate which otherwise remains to varying extent in spent culture [18, 50]. Another matter of concerns regarding lignin depolymerization and its conversion into biofuels/bioethanol is repolymerization of lignin-derived low molecular weight sp. into high molecular weight molecules which are not easy to be degraded by microbes. Repolymerization is observed to occur within

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few hours after onset of lignin volarization. For this purpose, organization of most effective microbial sink for immediate utilization of low molecular species for bioethanol production is the most appealing option [51, 52].

For overcoming this bottleneck, microbial sink/consortium of different microbes with xylose overexpression is an offered strategy. Preventing repolymerization of low molecular weight lignin species into high molecular weight lignin compounds and ensuring the complete utilization and conversion of available sugars into bioethanol can make the bioprocess cost-effective. The description in this chapter will lead to development of technologies that can be helpful in efficient depolymerization of lignin and its simultaneous conversion into high-valued microbial-assisted advanced biofuel. The chapter represents need for development of road map for advanced level of biofuel production from waste crop residues. Nature-inspired enzymes' involvement is the most effective way for enhanced bioethanol production from biomass. The enzymes convert the long chains of polysaccharides into monosaccharaides. Currently, lignocellulytic enzymes used for ethanol production from cellulosic biomass are obtained from fungus, gut of termite and certain bacteria [40]. Present restrictions of enzymatic breakdown of lignocellulose-based biomass are mostly due to concern of enzymatic steadiness and vulnerability to inhibitors or by-products [53, 54]. Continuous bioengineering efforts and prospecting should provide novel enzymes with lower susceptibility to inhibitors and relatively higher specific activity [55]. Few insects such as termites have very efficient approaches to break the lignocellulose-based substrates as potential mean of bioenergy [56]. In case of lower termites, activity (cellulolytic) is normally dependent on enzymes produced by endosymbiotic, flagellated protists [57], while in case of higher termites, their guts contain lignocellulytic enzymes which combine with cellulases secreted by certain endosymbiotic gut bacteria [58, 59].

Hence, establishment of large-scale bioethanol production plant by treating waste crop residues with such novel enzyme will enhance the production and can successfully provide support to deteriorating economy of Pakistan (**Figure 2**). The resulting cleaner environment is another benefit that has monetary values that the government may be financially and ethically interested in. Such multiple positive benefits will attract different interested parties to involve in replication of process, each with resources and benefits to sustain and multiply. Additionally, the greenhouse gas emission will be reduced by burning bioethanol, as the net CO_2 emission is zero because the amount of CO_2 emitted on burning is equal to the amount of it, which is absorbed from the atmosphere by the process of plant photosynthesis which will be used for production of bioethanol [60]. In Pakistan, the current domestic production of raw oil presently satisfies only almost 25% of the country's consumption and remaining demands are met by importing fuels from abroad. This make Pakistan's economy vulnerable to different social and economic issues; however, incorporation of biofuel/bioethanol will reduce burden on country's economy significantly.

Federal Cabinet, Economic Coordination Committee (ECC) of Pakistan has decided to permit marketing of Ethanol 10 as motor vehicle fuel on the trial basis. Anhydrous ethanol can be blended with gasoline in different proportions having less than 1% water content. Many of the motor vehicles having gasoline engines operate well with ethanol blend of 10% in their fuels (E 10). The Government of Pakistan enacted a 15% duty on export of molasses to prefer the use of molasses for ethanol production rather than export [61]. The government of Pakistan should make policy to enforce the blending of ethanol in transportation fuels as early as it becomes conceivable [18]. Successful implementation of large-scale waste crop residue-based bioethanol production concept will attract private sector investment and company-farm partnership to accelerate the development and commercialization of new bioenergy solution to improve emerging economies and transform the lives of at least small farmers. The concept is readily adoptable by different agricultural regions as the essential supply of the feedstock is available in the form of agricultural residues that are sustainable and typically available abundantly and locally.

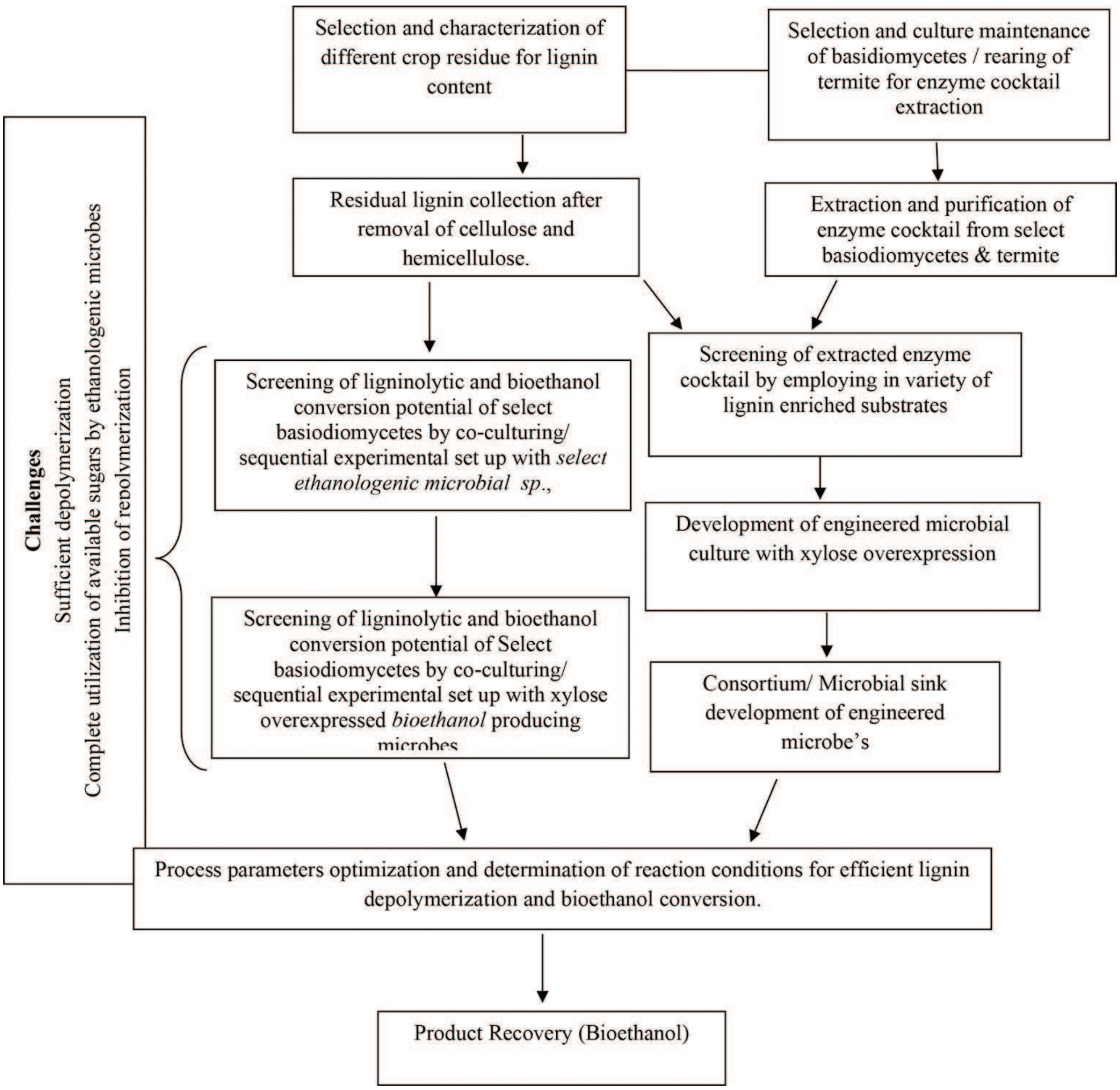

Figure 2. Schematic representation of nature inspired enzyme assisted bioethanol production process.

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6. Concluding remarks

To import conventional energy resources, Pakistan is spending around 7 US$ billion equivalent to 40% of total imports. Careful estimates show that by 2050 Pakistan's energy needs will be increased three times while the supplies are not very inspiring. In 2012, Pakistan natural gas supply had about 15 billion-m^3/year deficit with increasing tendency. Rise in natural gas price (0.51 $/$m^3$) brings great potential to promote biomass-based biofuel production in Pakistan.

Pakistan being an agricultural country produces wheat, sugarcane and potatoes as one of its biggest crops [62–64]. Consequently, large amounts of wheat straw and sugarcane bagasse are obtained as by waste products. Due to high ambient temperature in most part of the year, poor post-harvest processing and storage of thousands of tons of biomass are wasted each year. In short access of promising process for the initiation of sustainable strategies for waste crop residue-based bioethanol production while consuming starch, cellulosic and lignin loads of effluents of respective origins in the country, bestowed with suitable biomass and temperature optima for successful cultivation of ethanologenic microbes, is expected to provide sustainable supplies of biofuels. Additionally, more than three billion acres worldwide which are not suitable for agriculture purpose due to dryness could be utilized for growing drought-hardy plants for biofuels production. The only disadvantage of using these crops is that they contain lignocellulose, a hard plant material, that needs more treatment than either corn or sugarcane to be converted into alcohol. Therefore, search for ways to make the overall process more efficient by reusing materials, changing the fermenting agent and searching for better and nature-inspired enzymes will be milestone in this regard. The process development of nature inspired enzyme-assisted conversion of agricultural and food waste into bioethanol that can be used as clean biofuel is demand of time. Adoption of these new dimensions for bioethanol production will definitely reduce pressure on energy and transportation sector, entire dependence on conventional fuels and can triumph fight against climate change.

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