Cellulose and Biomass Conversion Technology and Its Application to Ethanol Production from Corn

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SSF fermentation.

Transportation fuels are almost totally derived from petroleum and accounted for more than one-quarter of the total energy used in the U.S. in 1989, more than 81 quads.[1] Currently, almost 50% of all petroleum used in the U.S. is imported, and imports have risen markedly over the last few years.[1] Because OPEC controls about 75% of the world's oil reserves, and the U.S. has only about 5% of that total, it is likely that petroleum imports will continue to rise.

Few substitutes exist for petroleum-based transportation fuels, and the U.S. is extremely vulnerable strategically and economically to disruptions in its supply, as illustrated by our experience with Iraq's invasion of Kuwait. In addition, imported petroleum contributed about 40% of the balance-of-payments deficit for the U.S. in 1989.[2]

Evaporative losses during fueling of automobiles, losses from the fuel system, and nitrogen oxides and unburned hydrocarbons from automotive exhaust contribute to ozone formation in many cities, such as Los Angeles.[3] High altitude cities, such as Denver, experience excessive levels of carbon monoxide because of incomplete combustion of carbon-containing fuels. Petroleum derived transportation fuels are the source of up to two-thirds of carbon monoxide and one-third to one-half of smog causing emissions.[3,4,5,9]

For these reasons and others, interest in ethanol — and ethanol made from biomass — as a transportation fuel has increased.

Ethanol is a clean-burning liquid fuel that can be readily substituted for gasoline in our transportation sector. When ethanol is produced from renewable sources of cellulosic biomass, ethanol can improve energy security, reduce the balance-of-payments deficit, decrease urban air pollution, enhance agricultural income, and reduce the accumulation of carbon dioxide in the atmosphere.[7,8,9,10]

The Ethanol from Biomass Program, managed by the National Renewable Energy Laboratory (NREL) for the Biofuels Systems Division of the U.S. Department of Energy (DOE), is directed at lowering the cost of ethanol production to the point at which ethanol can compete with gasoline without tax incentives, so that the benefits of this unique fuel can be more widely realized. In this overview, technology for ethanol production from cellulosic biomass will be described. Opportunities for applying this technology to increase revenues from dry and wet milling processes for ethanol production from corn will also be discussed.

Cellulosic Biomass

Cellulosic biomass is a complex mixture of carbohydrate polymers from plant cell walls known as cellulose and hemicellulose, plus lignin and small amounts of other compounds known as extractives (see Figure 1). Examples include agricultural and forestry residues, municipal solid waste (MSW), herbaceous and woody plants, and underutilized standing forests. Cellulosic biomass is inexpensive to produce.

The cellulose fraction is composed of glucose sugar molecules bonded together in long chains that are held together in a crystalline structure. The hemicellulose portion of biomass is made of long chains of a number of different sugars and does not have a crystalline structure. For hardwoods and many grasses, the predominant component of hemicellulose is xylose, a five-carbon sugar that has historically been more difficult to convert into useful products than glucose has been.

For the U.S., it is estimated that the total amount of collectable underutilized wood, agricultural residues, short-rotation energy crops, and MSW could provide about 2,700 million dry tons of cellulosic biomass per year at prices from $20-70 per dry ton.[9,10] This quantity of feedstock can generate about 300-billion gallons of ethanol, more than double current U.S. gasoline market demand of 140-billion gallons of ethanol equivalent.

The price of these raw materials is low enough to provide a reasonable margin for conversion into ethanol. Even though the magnitude of the resource is subject to significant uncertainty, the potential availability of renewable feedstocks is projected to be substantial while the cost is reasonable. Thus, cellulosic biomass is a favorable feedstock for fuel ethanol production.

Ultimately, abundant feedstocks are required to achieve large-scale ethanol production. Oak Ridge National Laboratory (ORNL) manages a Biomass Production Program for DOE to develop technology for producing fast-growing herbaceous and woody crops that will provide an abundant and low-cost substrate for ethanol production.

Conversion of Cellulosic Biomass into Ethanol

Cellulosic biomass is an inexpensive feedstock, and acids or enzymes will catalyze the breakdown of the cellulose and hemicellulose chains into their component sugar molecules, which can be fermented into ethanol. Lignin is a complex phenolic polymer that cannot be fermented into ethanol. The challenge is to develop low-cost methods to convert the naturally resistant cellulose and hemicellulose into ethanol.

Over the years, a number of processes have been studied for conversion of cellulose-containing biomass into ethanol through reactions catalyzed by dilute acid, concentrated acid, or enzymes known as cellulases. In each of these options, the feedstock is pretreated to reduce its size and facilitate downstream processing, as shown in Figure 2. The cellulose fraction is hydrolyzed by acids or enzymes to produce glucose sugar, which is subsequently fermented to ethanol. The soluble xylose and other sugars derived from hemicellulose can also be fermented to ethanol, and the lignin fraction can be burned as fuel to power the rest of the process, converted into octane boosters, or used as a feedstock for the production of chemicals.

Acid-Catalyzed Processes

Several dilute acid hydrolysis pilot plants were constructed in the U.S. during World War II as part of an effort to produce ethanol for fuel use,[11] but the economics were too unfavorable to

Figure 1. Cellulosic biomass consists of cellulose, hemicellulose, lignin, and some extractives as shown here for representative examples of agricultural residues.

allow continued operation in a free market economy. Dilute acid-catalyzed processes are currently operated in Eastern Europe for converting cellulosic biomass into ethanol and single cell protein. Low yields of 50-70% typical of dilute acid systems make these processes unable to compete with existing fuel options.[12, 13]

Concentrated sulfuric or halogen acid options achieve the high yields required. However, because low-cost acids, (e.g., sulfuric) must be used in large amounts and more potent halogen acids are relatively expensive, recycling of acid by efficient, low-cost recovery operations is essential to achieve economic operation.[14, 15] The acids must be recovered at a cost substantially lower than that of producing these inexpensive materials in the first place — a difficult requirement. Several processes are being developed for concentrated-acid hydrolysis of biomass.

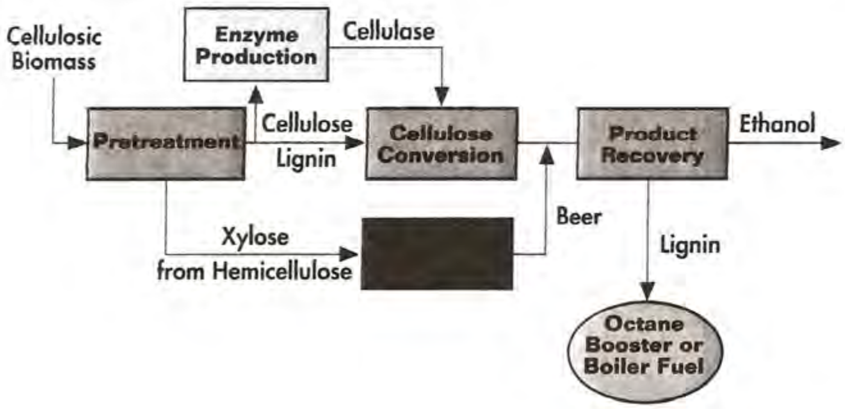
Enzyme-Catalyzed Processes

Enzyme-catalyzed processes achieve high yields under mild conditions with relatively low amounts of catalyst. Enzymes are also biodegradable and environmentally benign. Over the years, several enzyme-based processes have been studied at the laboratory scale, but only a few investigations have been expanded to a larger scale. The three leading processes considered are discussed below.

Separate Hydrolysis and Fermentation

In the separate hydrolysis and fermentation (SHF) process, the cellulosic biomass is first processed in a pretreatment device to open the biomass structure.A portion of the pretreated biomass is used in an enzyme production vessel to support growth of a fungus that produces cellulase enzyme, and the cellulase is added to the bulk of the pretreated substrate in a hydrolysis reactor. At this stage, the enzyme catalyzes the breakdown of the cellulose by a so-called hydrolysis reaction to form glucose sugar. The stream from the hydrolysis process passes on to a fermenter to which yeast are added to convert the glucose

Figure 2. The process flow diagram for acid- or enzyme-catalyzed conversion of cellulosic biomass to ethanol.



into ethanol. Finally, the ethanol is separated from the rest of the fermentation broth in a purification step.[16, 17]

Simultaneous Saccharification and Fermentation

The sequence of steps for the simultaneous saccharification and fermentation (SSF) process is virtually the same as for separate hydrolysis and fermentation except that hydrolysis and fermentation reactions are combined in one vessel.[18, 19] The presence of yeast along with the enzymes minimizes the accumulation of sugar in the vessel, and because the sugar produced during breakdown of the cellulose slows down the action of the cellulase enzymes, higher rates and yields are possible for SSF than for SHF. Additional benefits come from elimination of about half of the expensive fermentation vessels and a mixture that is less vulnerable to invasion by unwanted microorganisms due to its ethanol content.

DOE/NREL has focused on the SSF process as a promising route to achieve low-cost fuel ethanol production within a reasonable time frame.[20]

Direct Microbial Conversion

The direct microbial conversion process combines the enzyme production, cellulose hydrolysis, and sugar fermentation steps in one vessel.[21, 22, 23] In the most extensively tested configuration, two bacteria are employed to produce cellulase enzymes and ferment the sugars formed by breakdown of cellulose and hemicellulose into ethanol. Unfortunately, the bacteria also produce a number of products in addition to ethanol, and yields are lower than for the SHF or SSF processes. Due to the simplicity of the process, the DMC option is very promising if yields and concentrations of ethanol can be improved.

Pretreatment of Cellulosic Biomass

Cellulosic biomass is naturally resistant to enzymatic attack, and a pretreatment step is required to overcome this resistance if the enzyme-catalyzed hydrolysis is to proceed at a reasonable rate with the high yields vital to economic viability. Several options have been considered for biomass pretreatment, including steam explosion, acid-catalyzed steam explosion, ammonia fiber explosion, organosolv, and dilute acid.

The dilute acid option has good near-term economic potential.[24] In this process, about 0.5% sulfuric acid is added to the feedstock, and the mixture is heated to about 140-160º C for 5-20 minutes.Under these conditions, most of the hemicellulose is broken down to form xylose sugars, leaving a porous material of primarily cellulose and lignin that is more accessible for enzyme attack. Evaluation of the dilute acid process with various agricultural residues, short-rotation hardwoods, and herbaceous energy crops has consistently shown that the conversion yields correlate well with the degree of hemicellulose removal.[25] Although this process has good near-term potential, significant benefit would result if a low-cost scheme could be devised that would also remove lignin because the solid lignin associated with the cellulose creates some processing difficulties in the hydrolysis and fermentation steps.

Hemicellulose Utilization

The hemicellulose polymers in cellulosic biomass such as hardwood, agricultural residues, and herbaceous plants can be readily broken down to form five-carbon sugars such as xylose during the pretreatment step. However, until recently, the xylose stream could not be utilized, and it was necessary to send this material to waste disposal. From an economic perspective, this costs the process twice: first, in payment for the xylose, and second in disposal costs.

Several options, outlined below, have been examined for xylose utilization:

Conversion of Xylose into Furfural

For dilute acid-catalyzed breakdown of cellulose to fermentation sugars, a significant fraction of the xylose will degrade into furfural.[26] Similarly, the xylose released by pretreatment can be reacted to furfural. This product is currently manufactured for use in foundry and other applications, so it could be sold as a by-product, generating additional revenues. However, the furfural market would be quickly saturated by the volume of furfural that would accompany large-scale production of ethanol as a fuel,[27] limiting the number of ethanol plants that furfural sales could support.

Yeast for Ethanol Production

Certain strains of yeast are known to ferment xylose into ethanol, such as Candida shehatae, Pichia stipitis, and Pachysolen tannophilus.[28,29,30,31] However, these strains require small amounts of oxygen in the fermentation broth to ferment xylose (microaerophilically). Large-scale production of ethanol fuels will probably require the use of huge fermenters with volumes approaching a million gallons each, and proper control of oxygen in such large vessels could be challenging. Furthermore, these yeast strains typically cannot yet achieve very high ethanol production rates or tolerate high ethanol concentrations.[32]

Other Microorganisms for Ethanol Production

Other microorganisms, such as thermophilic bacteria and fungi, can anaerobically ferment xylose into ethanol.[33,34,35,36,37,38] However, ethanol tolerance has not been satisfactorily demonstrated for bacteria, although some new evidence suggests that previous conclusions may have been premature.[39] New information indicates that the yields could be improved in continuous culture.[40] The fungi evaluated suffer from similar limitations in both ethanol tolerance and yield.

Simultaneous Isomerization and Fermentation of Xylose to Ethanol

Several groups have studied the use of xylose isomerase enzyme to convert xylose into an isomer called xylulose that many yeasts can ferment into ethanol under anaerobic conditions .[41,42] Researchers at NREL genetically engineered the common bacteria Escherichia coli to produce large quantities of xylose isomerase for such a process, and ethanol yields of 70% of theoretical have been achieved in the simultaneous isomerization and fermentation of xylose process.[43] In this configuration, the enzyme and yeast are employed together to drive the equilibrium-limited fermentation to completion, with the primary yield loss resulting from xylitol formation.[44,45] This process has the advantage of employing anaerobic yeast that are easier to use on a large scale, but the need to provide xylose isomerase enzyme and adjust for differences in pH optima between the yeast and enzyme complicate the technology.

Genetically Altered Bacteria

Researchers at the University of Florida have successfully introduced the genes from *Zymomonas mobilis* into the common *bacterium E. coli, Klebsiella oxytoca*, and others so that they can now ferment xylose directly into ethanol.[46,47,48,49,50] This approach has the advantage that a single organism can carry out the fermentation of xylose, and initial data suggest that high yields are possible. These bacteria currently require operation at near-neutral pH while production of by-product acids tends to drive the pH down, thus requiring addition of bases. Invasion by other bacteria could be problematic.

At this time, promising options for xylose conversion are the use of genetically engineered bacteria and microaerophilic yeast. However, full integration of these technologies into the overall conversion process is required to evaluate and improve their performance. Advantages would also result if these options could be improved further by genetic modifications or other approaches.

Lignin Conversion

As shown in Figure 1, lignin generally represents the third largest fraction of cellulosic biomass and is not significantly different in quantity than hemicellulose. Thus, it is important to derive value from the lignin fraction if the economic production of ethanol from biomass is to be achieved. Three options, discussed below, lead the possibilities for lignin use:

Lignin as a Boiler Fuel

Lignin has a high energy content and can be used as a boiler fuel.[51,52,53] The amount of lignin in most feedstocks is more than enough to supply all the heat required for the entire conversion process and to generate enough electricity to meet its demands. In fact, excess electricity beyond all of these needs is generated, and additional revenue can be generated from electricity exports from the plant. The electricity sold for current plant designs is equivalent to about 8% of the energy value of the ethanol product, and greater revenues are likely as the technology is improved to require less process heat and electricity.

Producing Octane Boosters from Lignin

Lignin is a complex phenolic polymer that can be broken down to form a mixture of monomeric phenolic compounds and hydrocarbons. The phenolic fraction can be reacted with alcohols to form methyl or ethyl aryl ethers, which are good octane boosters.[54] Because gasoline additives are more valuable than boiler fuel, this option for lignin use would generate more revenue. However, the technology must be improved to provide high product yields, and the conversion costs must be low enough to provide a net income gain for the ethanol plant.

Producing Chemicals from Lignin

A number of chemicals could be produced from lignin including phenolic compounds, aromatics, dibasic acids, and olefins.[55] Such materials could have a high value that would augment the total revenue for the ethanol plant. However, just as for the conversion of lignin into octane boosters, the cost of the conversion process must be low enough to ensure a net gain in revenue. In addition, high yields of target products will likely be necessary to achieve economic viability, and significant markets must exist to be compatible with large-scale ethanol production.

Progress and Potential for Improvement

Progress on enzyme-catalyzed processes to convert cellulosic biomass into fuel ethanol has been substantial during the last 10 years, with projected selling prices dropping from about \$3.60/gal. in 1980[56] to only about \$1.27/gal. now.[57] This reduction in selling price is a result of improved rates and yields from the SSF process, improvements in enzymes to achieve high yields with lower loadings, selection of better fermentative microbes, and advances in xylose fermentations through genetic engineering.

Although research progress has been substantial, significant opportunities still exist to reduce the selling price of ethanol from cellulosic biomass to \$.67/gal. at the plant gate. Key target areas include further improvements in glucose and xylose yields from pretreatment; increased ethanol yields to 90% or greater from cellulose and xylose fermentations; decreased stirring and pretreatment power requirements; better productivities through continuous processing and biocatalyst immobilization; low-cost production of octane enhancers or chemicals from lignin; increased ethanol concentration; and reduction of fermentation times. Because feedstock costs are a significant fraction of the final product selling price, improvements in feedstock production, collection, and genetics could provide additional cost reductions through economies of scale for larger ethanol plants, decreased feedstock costs, and less nonfermentable feedstock. Many of these goals have been met individually; the evidence that the rest can be achieved is great. The primary need is to meet these goals simultaneously. It is also encouraging that enough options exist to lower the selling price of ethanol that not all the technical goals must be achieved to reach the target selling price.

Applications to Corn Ethanol Production

The cost of the feedstock is an important portion of the projected selling price of ethanol from cellulosic biomass. At the current projected \$1.27/gal. selling price, \$0.46 is for feedstock at a price of \$42/ton delivered to the plant gate. For example, if a free feedstock could be obtained, ethanol could be produced for about \$0.81/gal. Because ethanol from corn now sells for about \$1.20-1.30/gal, use of an inexpensive feedstock could be advantageous. Several possible low-value or waste streams could prove attractive, including the carbohydrate fractions of corn gluten feed from corn wet milling; distillers' dried grains and solubles (DDGS) from dry milling from corn; agricultural waste streams such as corn cobs, corn stover, or wheat straw; commercial processing waste streams such as white water in paper manufacture; and domestic wastes such as MSW. Although such feedstocks are of limited availability, they provide an opportunity to establish the technology early.

Utilization of the Fiber From Wet Milling Processes

About two-thirds of the ethanol made from starch in the U.S. is made by wet milling processes.[58,59] Typically about 2.5 gal. of ethanol, 13 lb of an animal feed coproduct called corn gluten feed, 3.4 lb of another animal feed coproduct called corn gluten meal, and 1.7 lb of corn oil are produced from a bushel of corn (47.3 lb dry weight). Corn gluten feed, an animal feed containing 21% protein, results from combining three by-product streams: fiber, germ meal, and corn steep liquor. The fiber and germ meal product streams contain significant amounts of cellulose, hemicellulose, and starch.

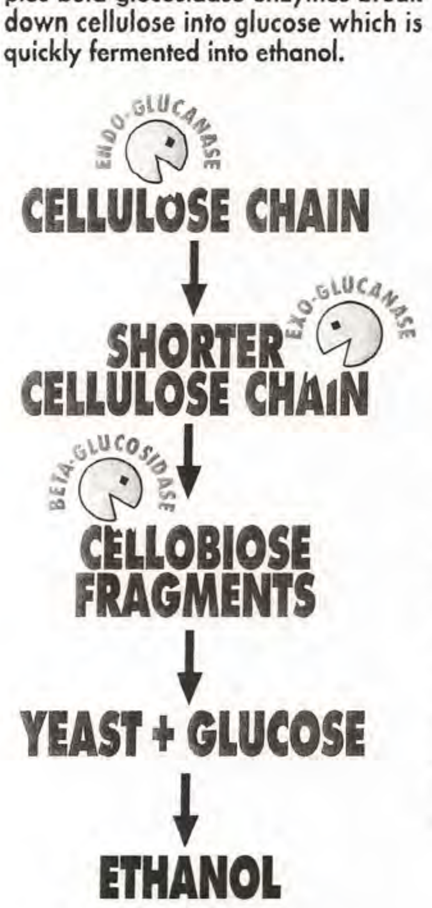
The price of corn gluten feed and other animal feeds seems to be set largely by their protein content, giving less value to the cellulose, hemicellulose, and other carbohydrate materials. In fact, since most of the corn gluten feed is sold to the European Economic Community, the cellulose, hemicellulose, and other carbohydrate materials add to shipping costs. A process that could convert cellulose, hemicellulose and other carbohydrates into valuable products such as ethanol could enhance the revenue for the plant while reducing most shipping costs.

The composition of the fiber stream is about 22.5% starch, 12.5% protein, 3% fat, 10% cellulose, 30% hemicellulose, 0.5% lignin, and 2% ash. The germ meal contains 26% protein, 20% starch, 5% fat, 13% cellulose, 32% hemicellulose, 1% lignin, and 4% ash. From every bushel of corn, 5.4 lb of fiber and 1.9 lb of germ meal are produced. Thus, from one bushel of corn, these combined streams contain 2.2 lb of hemicellulose, 0.79 lb of cellulose, and 1.0 lb of starch.[59]

These streams could be converted into 0.26 gal of ethanol from hemicellulose and cellulose, and 0.13 gal of ethanol from starch, or a total of 0.39 gal of ethanol, an increase of 16% over the quantity of ethanol typically produced from a bushel of corn. This increased production would increase ethanol revenue by 16% while decreasing shipping costs of corn gluten feed by as much as 42%. For every 1 billion bushels of corn pro-cessed, total ethanol production from the carbohydrate fraction of the fiber and germ meal streams could be 400 million gallons, which represents revenues of \$480 million at an ethanol price of \$1.20/gal.

On the other hand, the protein content of the corn gluten feed would

Figure 3
SSF process: Endo- and exo-glucanase plus beta-glucosidase enzymes break down cellulose into glucose which is quickly fermented into ethanol.



be higher after removal of the carbohydrates. As a result, revenues from protein sales could remain the same if the product is valued by its protein content while shipping costs of the corn gluten feed could drop by as much as 42%.

Utilization of the Carbohydrates in DDGS for the Dry Milling Process

The remaining ethanol made in the U.S. is from the dry milling of corn.[58,59] In this process, 2.5-2.6 gal of ethanol are produced per bushel of corn processed, along with about 17 lb of an animal feed coproduct called DDGS, and 17 lb of carbon dioxide. DDGS contains at least 27% protein. Essentially, all the cellulose and hemicellulose from corn ends up in the DDGS product, along with some starch. The price of DDGS seems to be set largely by its protein content, giving little value to the cellulose and hemicellulose. In fact, these carbohydrates add to shipping costs. As for wet milling, a process that converts the cellulose and hemicellulose into a valuable product such as ethanol could enhance revenues for the plant while lowering shipping costs for DDGS.

Because the cellulose and hemicellulose comprise 9.5% of the dry weight of corn, 4.3 lb of these materials are expected in 17 lb of DDGS from one bushel of corn. If converted into ethanol, the hemicellulose and cellulose in DDGS could yield 0.37 gal of ethanol from every bushel of corn processed. This is an increase of 14-15% over the ethanol typically produced from a bushel of corn. Sale of this increased ethanol would raise revenues similarly, while possibly maintaining animal feed coproduct revenues as a higher value protein feed and decreasing shipping costs by as much as 25%. For every 100 million bushels of corn processed, total ethanol production from the hemicellulose and cellulose content of DDGS could be 37 million gallons.

Conclusions

Ethanol is a clean-burning, high-octane fuel that can be used in today's transportation sector to improve air quality. Cellulosic biomass is an abundant resource in the United States that could support large-scale production of ethanol for fuel use. This means that using ethanol derived from biomass could reduce the strategic vulnerability of the United States, prevent sudden changes in price, and lower our trade deficit.

Both acid- and enzyme-catalyzed reactions have been evaluated for conversion of cellulosic biomass into ethanol, and research has been focused on enzymatic hydrolysis technology because of its potential to achieve high yields of ethanol under mild conditions. In particular, the SSF process is favored for ethanol production from the major cellulose fraction of the feedstock because of its low cost potential. Technology has also been developed for converting the second-largest fraction, hemicellulose, into ethanol, and the remaining lignin can be burned as boiler fuel to power the conversion process and generate extra electricity for export. Together, developments in conversion technology have reduced the selling price of ethanol from about $3.60/gal ten years ago to $1.27/gal now. Additional technical targets have been identified that could bring the selling price down to $0.67/gal with aggressive research and development.

Technology for conversion of cellulosic biomass to ethanol could be applied to both wet and dry milling processes to substantially enhance ethanol revenues, while likely maintaining revenues from the sale of animal feed coproducts with higher protein content. Because the carbohydrates in these products have low value, the application of cellulose conversion technology to these processes could result in lower ethanol costs for the plant. Other low-cost materials could also be converted into ethanol at prices competitive in today's fuel market.

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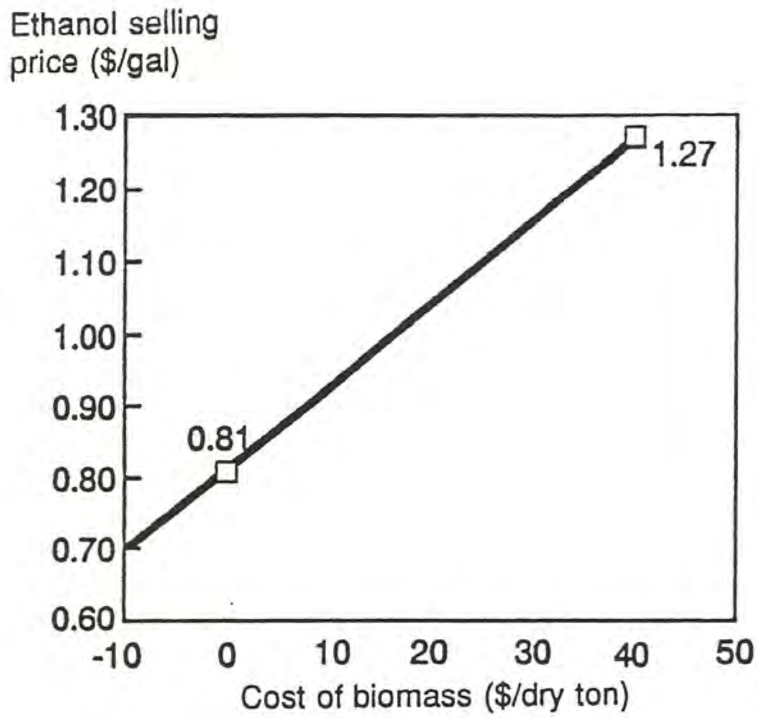

Figure 4. The current price to ethanol from cellulosic biomass is $1.27/gal when the cost of the feedstock is $42/ton. Lower cost feedstocks result in a lower cost of ethanol production, as shown here.

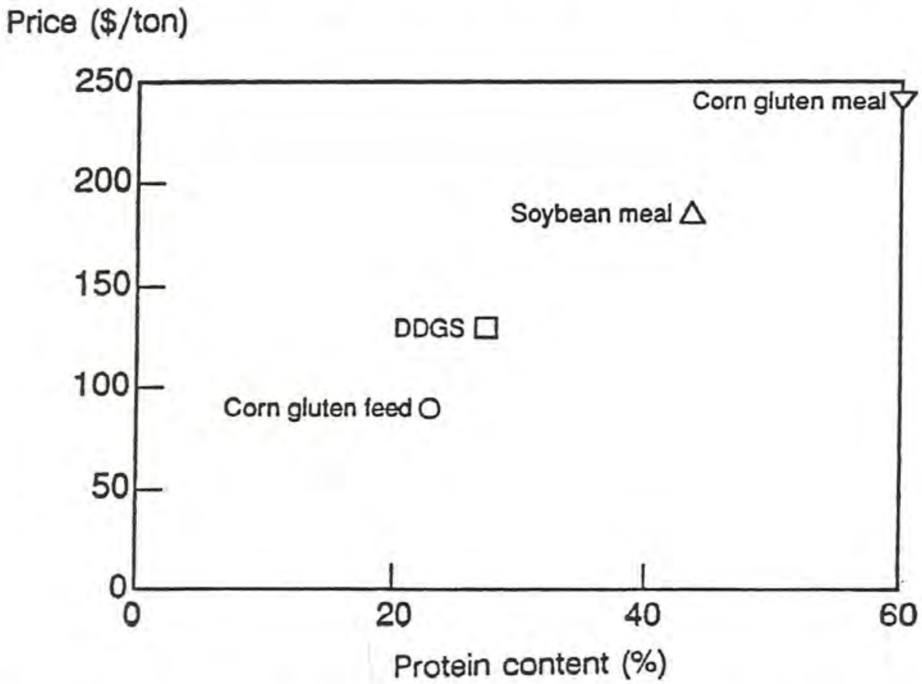

Figure 5. Relationship among the market prices of various animal feeds and their protein content, based on recently reported prices.