Chapter 9
Plants as Sources of Energy

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Abstract This chapter is concerned with biotechnological applications involving the use of plants as sources of energy. Plants contain stored carbon captured from light-catalyzed carbon dioxide fixation via photosynthesis. This stored carbon from plants is available in oil and coal deposits that can be used as energy sources known as petrofuels. Living plants or plant residues can be used to generate biofuels such as methane from methane generators, wood fuel from wood chips, and alcohol from plant-based starch or cellulose in fermentation reactions. Topics that illustrate these applications include plant-based biofuels for engines – biodiesel and bioethanol; energy from woodchips (woodchip combustion, gazogen, or wood gasification); and methane (CH_4) or natural gas – methane gas production from landfills, methane gas produced in biodigesters using plant materials as substrate. We discuss the pros and cons of these applications with plant-derived fuels as well as the different types of value-added crops, including algae, that are currently being used to produce biofuels.

9.1 Introduction

Through the process of photosynthesis, plants have the capacity to capture and utilize energy, derived from the Sun, along with carbon from the Earth's atmosphere and nutrients from our soils to generate biomass. This biomass, in the form of roots, stems, leaves, fruits and seeds, is also consumed by animals and microorganisms, which in turn, generate their own forms of biomass. Manure, leaf litter, wood, garden waste, and crop residues are all common examples of biomass. Consequently, one definition of *biomass*is any organic/biological material which contains stored sunlight in the form of chemical energy. Typically, humans release this energy by burning the material, and humans have used biomass as an energy source in the form of solid biofuels for heating and cooking since the discovery of fire.

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A. Kirakosyan, P.B. Kaufman, *Recent Advances in Plant Biotechnology*,
DOI 10.1007/978-1-4419-0194-1_9,

Bioenergy is energy made available from organic materials and is often used as a synonym to biofuel. However, an important distinction between bioenergy and biofuel is that biomass is the fuel/biofuel and bioenergy is the energy contained in that fuel (Anderson, 2003; Agarwal, 2007; Drapcho et al., 2008). *Biofuel* can be broadly defined as any solid, liquid, or gas fuel derived from recently dead organic/biological material. This distinguishes it from fossil fuels such as coal, oil, and natural gas, which are derived from long dead, subterranean deposits of biological material. Unlike fossil fuel resources, which have an inevitable finite supply, biofuels are largely renewable energy sources based on a balance within the Earth's carbon cycle. As the human population continues to expand, and the demand for fossil fuels exceeds its supplies, pressure is mounting to find efficient and effective methods to produce renewable biofuels. Various plants and plant-derived materials are currently used for biofuel manufacturing, and biofuel industries are expanding in Europe, Asia, and the Americas. Agriculturally produced biomass fuels, such as biodiesel, bioethanol, and bagasse (often a by-product of sugarcane cultivation) can be burned in internal combustion engines and cooking stoves (Agarwal, 2007). However, there are many criticisms and concerns surrounding current practices for the production of biofuels. Consequently, research into more sustainable methods of generating biofuels will depend largely on the creation of environmentally responsible policies in farming, processing, and transporting of biofuels.

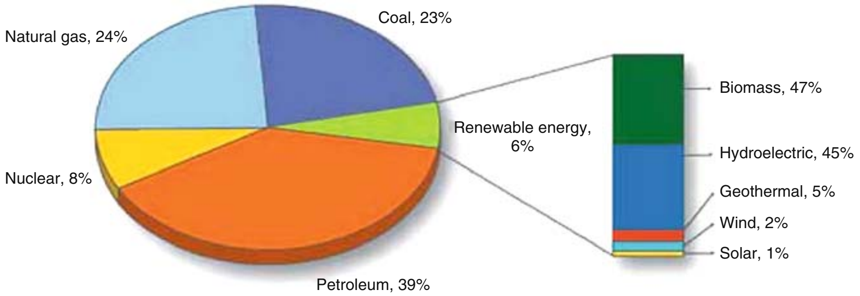
This chapter examines some of the pros and cons in the current methods used for generating various types of bioenergy, namely, energy derived from solid biomass, bioalcohol, biodiesel, biogas, and presents a critical look at how biotechnology can help to solve the world's current and future energy needs.

9.2 Energy Crisis and the Balance of Carbon

Biofuels were the first form of fuel used by human cultures around the world. Even up to the discovery of electricity and the start of the industrial revolution, fuels such as wood, whale oil, manure, and even alcohol were the primary sources of energy for heating, cooking, and lighting. However, the discovery and use of fossil fuels, including coal, oil, and natural gas dramatically reduced the emphasis on biomass fuel in the developed world (Peters and Thielmann, 2008). In the United States, for example, large supplies of crude oil were discovered in Pennsylvania and Texas in the mid- and late 1800s. This allowed petroleum-based fuels to become inexpensive. Because of these low costs, fossil fuels were widely used to promote the growing industrial age, especially for the production of power used to run factories and automobiles.

Despite the huge increase in the use of fossil fuels, most of the world continued to depend upon and make use of biofuels. Even in the United States, during the high-energy demand seen during wartime periods of World War II, biofuels were valued as a strategic alternative to imported oil. However, during the peacetime postwar period, inexpensive oil from the Middle East helped to trigger a worldwide shift away from biofuels. Since then, there have been a number of "energy crises" around the world, caused by a variety of social and political factors. An *energy crisis*is any large-scale bottleneck (including price rises) in the supply of energy resources to an economy. Two of the best known ones occurred in 1973 and 1979, when geopolitical conflicts in the Middle East caused *OPEC* (*Organization of Petroleum Exporting Countries*) to cut exports. Consequently, non-OPEC nations experienced a very large decrease in their oil supply. This crisis resulted in severe shortages and a sharp increase in the prices of high-demand oil-based products, most notably gasoline. Throughout history, the fluctuations of supply and demand, energy policy, military conflict, and environmental impacts have all contributed to a highly complex and volatile market for energy and fuel. On the other hand, such problems always resurrect the principles of *green energy* and sustainable living. This has led to an increasing interest in alternate power/fuel research such as bioethanol, biodiesel, biogas, fuel cell technology, hydrogen fuel, solar/photovoltaic energy, geothermal energy, tidal energy, wave power, wind energy, and fusion power. Heretofore, only hydroelectricity and nuclear power have been significant alternatives to fossil fuels, which still dominate as energy sources (Fig. 9.1).

Although technology has made oil extraction more efficient, the world is having to struggle to provide oil by using increasingly costly and less productive methods, such as deep sea drilling and developing environmentally sensitive areas such as the Arctic National Wildlife Refuge. In addition, the world's population continues to grow at a rate of ~250,000 people/day, and while a small part of the world's population consumes most of the resources, the people of developing nations continue to



Biomass Consumption	Million dry tons/year
Forest products industry	
Wood residues	44
Pulping liquors	52
Urban wood and food & other process residues	35
Fuelwood (residential/commercial & electric utilities)	35
Biofuels	18
Bioproducts	6
Total	**190**

Fig. 9.1 Estimated world energy use from different sources. From the state energy conservation office web site (http://www.seco.cpa.state.tx.us/re_biomass-crops.htm). Source: The US Department of Energy's (DOE) Energy Information Agency (EIA), used with their permission

adopt more energy-intensive lifestyles. Currently, the United States, with its population of 300 million people, consumes far more oil than China, with its population of 1.3 billion people. But, this is also beginning to change, leading to an ever increasing demand for energy around the world. Many energy experts have concluded that the world is heading toward an unprecedented large and potentially devastating global energy crisis due to a decline in the availability of cheap oil and other fossil fuels and a progressive decline in extractable energy reserves.

To add to this problem, carbon emissions, including greenhouse gasses like carbon dioxide (CO_2), have been increasing ever since the industrial revolution. It is well documented that atmospheric CO_2 concentrations have risen by $\sim$30% in the last 250 years. Data from monitoring stations, together with historical records extracted from ice cores, show that atmospheric CO_2 is now at a level higher than at any time in the last 650,000 years (Meehl et al., 2007). Such increases in CO_2 appear to be driven, in part, by the addition of 6–8 Pg (one Pg [petagram] = 1 billion metric tonnes = 1,000 × 1 billion kg) of carbon/year from human-derived sources, especially the burning of various fossil fuels which power our electricity and automobiles. Atmospheric CO_2 is predicted to continue to rise an additional 50% by 2050 (Meehl et al., 2007), and such rising levels of CO_2 are at the heart of the concerns over global warming and many of the associated environmental problems.

Biofuels and other forms of renewable energy aim to be carbon neutral or even carbon negative. *Carbon neutral* means that the carbon released during the use of the fuel is reabsorbed and balanced by the carbon absorbed by new plant growth during photosynthesis (Fig. 9.2). The plant biomass is then harvested to make the next batch of fuel, thus perpetuating the cycle of carbon in the Earth's atmosphere without adding to the problem. The Intergovernmental Panel on Climate Change (IPCC) estimates that between 46 and 56% of terrestrial carbon is found in forest biomes and that actions to preserve and enhance this carbon sink would likely increase the global terrestrial carbon by 60–87 Pg C by 2050, thereby offsetting ca. 15% of the anthropogenic emissions predicted for the same period (Saundry and Vranes, 2008). Using biomass to produce energy can reduce the use of fossil fuels, reduce greenhouse gas emissions, and reduce pollution and waste management problems (Agarwal, 2007). Therefore, carbon-neutral fuels, in theory, can lead to no net increases in human contributions to atmospheric CO_2 levels, thereby reducing the potential human contributions to global warming.

In addition to these arguments for biofuels, one of the strongest political drivers for the adoption of biofuel is "energy security." This means that a nation's dependence on oil is reduced and substituted with use of locally available sources, such as coal, gas, or renewable bioenergy sources. While the extent to which bioenergy can contribute to energy security and carbon balance will remain in active debate, it is clear that the dependence on oil is reduced. The US NREL (National Renewable Energy Laboratory) says that energy security is the number one driving force behind the US biofuels program (Bain, 2007) and the White House "Energy Security for the 21st Century" makes clear that energy security is a major reason for promoting bioenergy. Whether the driving forces behind a need for bioenergy is energy security, rising oil prices, concerns over the potential oil peak, greenhouse gas emissions

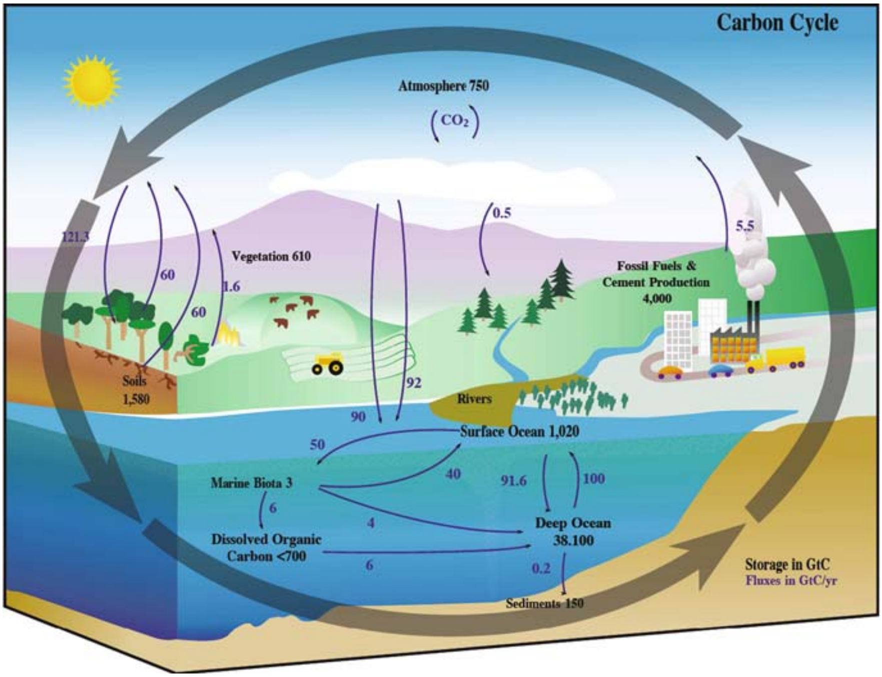


Fig. 9.2 The carbon cycle. Gigatons of carbon (GtC)/year, stored at various sites along the cycle. Illustration courtesy of NASA Earth Science Enterprise, available at Wikipedia public domain

(causing global warming and climate change), rural development interests, or instability in places such as the Middle East, it is clear that at some point, our global society is going to have to embrace the use of biofuels as a more stable, sustainable means of meeting our energy needs.

9.3 Disadvantages of Biofuels

While there are many potentially positive aspects to bioenergy and biofuels, there is growing international criticism because many biofuel energy applications take up large amounts of land, actually create environmental problems, or are incapable of generating adequate amounts of energy. While the plants that produce the biofuels do not produce pollution directly, the materials, farming practices, and industrial processes used to create this fuel may generate waste and pollution. Large-scale farming is necessary to produce agricultural biofuels, and this requires substantial amounts of cultivated land, which could be used for other purposes such as growing food, or left as undeveloped land for wildlife habitat stability. The farming of these lands often involves a decline in soil fertility. This is due to a reduction of organic matter, a decrease in water availability and quality due to intensive use of crops, and an increase in the use of pesticides and fertilizers (typically derived from petroleum). The need for more energy crop land has been cited to cause deforestation, soil erosion, huge impacts on water resources and is implicated in the dislocation of local communities. Proponents of biofuels, however, point out that while the production of biofuels does require space, it may also reduce the need for harvesting non-renewable energy sources, such as vast strip-mined areas and slag mountains for coal, safety zones around nuclear plants, and hundreds of square miles being strip-mined for oil/tar sands.

As an example of such issues, the current alcohol-from-corn (maize) production model in the United States has come under intense scrutiny. When one considers the total energy consumed by farm equipment, soil cultivation, planting, fertilizers, pesticides, herbicides, and fungicides made from petroleum, irrigation systems, harvesting, transport of feedstock to processing plants, fermentation, distillation, drying, transport to fuel terminals and retail pumps, and lower ethanol fuel energy content, the net benefit does little to reduce unsustainable imported oil and fossil fuels required to produce the ethanol in the first place. The June 17, 2006, editorial in the Wall Street Journal stated, “The most widely cited research on this subject comes from Cornell University’s David Pimental and University of Californa, Berkeley’s Ted Patzek. They’ve found that it takes more than a gallon of fossil fuel to make one gallon of ethanol from corn – 29% more. That’s because it takes enormous amounts of fossil-fuel energy to grow corn (using fertilizer and irrigation), to transport the crops and then to turn that corn into ethanol.” Ethanol is also corrosive and cannot be transported in current petroleum pipelines; so, more expensive over-the-road stainless-steel tank trucks need to be used. This not only uses fuel but increases the cost to the customer at the pump. In addition, the subsidies paid to fuel blenders and ethanol refineries have often been cited as the reason for driving up the price of corn, in farmers planting more corn, and the conversion of considerable land to corn production, which generally consumes more fertilizers and pesticides than many other land uses and also leads to serious environmental consequences such as dead zones in the Gulf of Mexico (Ahring and Westermann, 2007).

There are many concerns that, as demand for biofuels increases, food crops are replaced by fuel crops, driving food supplies downward and food prices upward. This is especially true for biofuels derived from food crops such as corn and soybean, which impacts food security and food prices, especially in poorer countries where the inhabitants have barely enough money to purchase their food let alone any fuel for cars or even stoves they cannot afford. There are those, such as the National Corn Growers Association, who say biofuel is not the main cause of food price increases and, instead, point to government actions to support biofuels as the cause. Others say increases are just due to oil price increases.

Some have called for a freeze on biofuels. Others have called for more funding for second generation biofuels which should not compete with food production. Alternatives such as cellulosic ethanol or biogas production may alleviate land use conflicts between food needs and fuel needs. Instead of utilizing only the starch by-products from grinding corn, wheat, and other crops, cellulosic ethanol and/or biogas production maximizes the use of all plant materials. Critics and proponents both agree that there is a need for sustainable biofuels, using feedstocks that minimize competition for prime croplands. These include farm, forest, and municipal waste streams; energy crops engineered to require less water, fertilizers, and pesticides; plants bred to grow on marginal lands; and aquatic systems such as algae used to produce alcohol, oil, and hydrogen gas (Ahring and Westermann, 2007). In short, biofuels, produced and utilized irresponsibly, could make our environmental/climate problems worse, while biofuels, done sustainably, could play a leading role in solving the energy supply/demand challenges ahead.

9.4 What Are the Major Types of Biofuels (Solid, Liquid, and Gas)?

There are several common strategies of producing biofuels. Each strategy is derived from growing an "energy crop." This is a type of plant grown at low cost and low maintenance that is converted into solid, liquid, or gas biofuels. Where the energy crop will be burned directly to exploit its energy content, woody crops such as *Miscanthus*, *Salix,* or *Populus* are widely used. Liquid biofuels can be generated from energy crops that are high in sugars (sugarcane, sugar beet, and sweet sorghum) or starch (corn/maize) by using yeast (*Saccharomyces*) alcoholic fermentation to produce ethyl alcohol (ethanol). It is also possible to make cellulosic ethanol from non-edible plants (switchgrass, hemp, and timber) and plant parts (rice husks, corn stalks, or grass clippings). Other liquid biofuels are derived from plants that contain high amounts of vegetable oil, such as oil palm, soybean, *Jatropha* or even algae. When these oils are heated, their viscosity is reduced, and they can be burned directly in diesel engines or they can be chemically processed to produce fuels such as biodiesel (Agarwal, 2007). In fact, the diesel engine was originally designed to run on vegetable oil rather than fossil fuel. Finally, biogas (methane, CH_4) has been produced for hundreds of years from waste materials including manure and crop residues. If high carbohydrate content is desired for the production of biogas, whole-crops such as maize, sudan grass, millet, white sweet-clover, wood, and many others can be made into silage and also be converted into biogas.

Depending on geographic location in the world, the type of energy crop grown often varies. These include corn, switchgrass, and soybeans, primarily grown in the United States; rapeseed, wheat, and sugar beet primarily grown in Europe; sugarcane in Brazil; palm oil and *Miscanthus* grown in Southeast Asia; sorghum and cassava in China; and *Jatropha* in India. In many locations, biodegradable outputs from industry, agriculture, forestry, and households can also be used for biofuel production, either by the use of anaerobic digestion to produce biogas or by the use of second generation biofuels to make use of straw, timber, manure, rice husks, sewage, and food waste. It is unfortunate that most governments appear fixated on the liquid fuel paradigm. Refocusing and balancing policies and communications to support the development of other technologies, including biogas and methods to extract the most energy out of plant and waste material would be very prudent. How to use biotechnology to better access this stored energy is a hot topic in science these days.

9.4.1 Solid Biomass

As mentioned above, humans have used solid biomass as a fuel for cooking and heating since the discovery of fire. The most obvious examples are wood and grasses, which have been used in campfires for centuries. Many native cultures around the world have also used the burning of solid biofuels, not only to release stored energy in the form of heat but also to release stored nutrients used to fertilize fields for better plant growth. The Aborigines in Australia, for example, have routinely burned the native Spinifex grass (*Spinifex sericeus* R. Br.) to elicit better plant growth in the desert and to aid in hunting animals by driving them in a known direction. Other, more agricultural societies use burning to fertilize crop lands to this day. Cattle farmers in the United States still use fire to trigger the growth of new grasses for their cattle, not to mention their traditional uses of cow manure for fertilizer, heating, and cooking. In fact, cow manure is estimated to still contain two-thirds of the original energy consumed by the cow. Wood was the main source of energy in the United States and the rest of the world until the mid-1800s, and biomass continues to be a major source of energy in much of the developing world.

In modern societies, solid biomass continues to be used directly as a combustible fuel, producing 10–20 $\mathrm{MJ{\cdot}kg^{-1}}$of heat. Its forms and sources include wood, the biogenic portion of municipal solid waste, or the unused portions of field crops. In the United States wood and wood waste (bark, sawdust, wood chips, and wood scrap) provide only about 2% of the energy we use today. About 84% of the wood and wood waste fuel used in the United States is consumed by the forest industry, electric power producers, and commercial businesses. The rest is used in homes for heating and cooking.

In addition to wood as a fuel, field crops may be used as fuel sources. For example, not only the field crops be grown intentionally as an energy crop but also the remaining plant by-products be used as a solid fuel. Sugarcane residue (also called *bagasse*), wheat chaff, corncobs, rice hulls, and other plant matter can be, and are burned quite successfully. Processes to harvest biomass from short-rotation poplars (*Populus* spp.) and willows (*Salix* spp.), and perennial grasses such as switchgrass (*Panicum virgatum* L.), *Phalaris*, and *Miscanthus*, require less frequent cultivation and less nitrogen than from typical annual crops. Pelletizing *Miscanthus* and burning it to generate electricity is being studied and may be economically viable.

Heating by wood is a more attractive option these days because technological improvements have made wood burning safer, more efficient, and cleaner. Options range from traditional wood stoves to pellet- and wood chipburning systems. While pellet fuel is manufactured by compressing ground wood and biomass waste into small, cylindrical pellets; woodchip fuel requires very little processing. In a typical woodchip heating system, a motor-driven conveyor system moves the chip fuel slowly and steadily from a chip hopper into a very efficient combustion chamber where the chips are burned (Fig. 9.3). As the chips burn, a fan blows hot air into a heat exchange boiler where water-filled tubes are heated. The hot water then circulates in pipes to provide heat to homes. In some commercial operations, steam can also be produced to power turbines that generate electricity. Many manufacturing

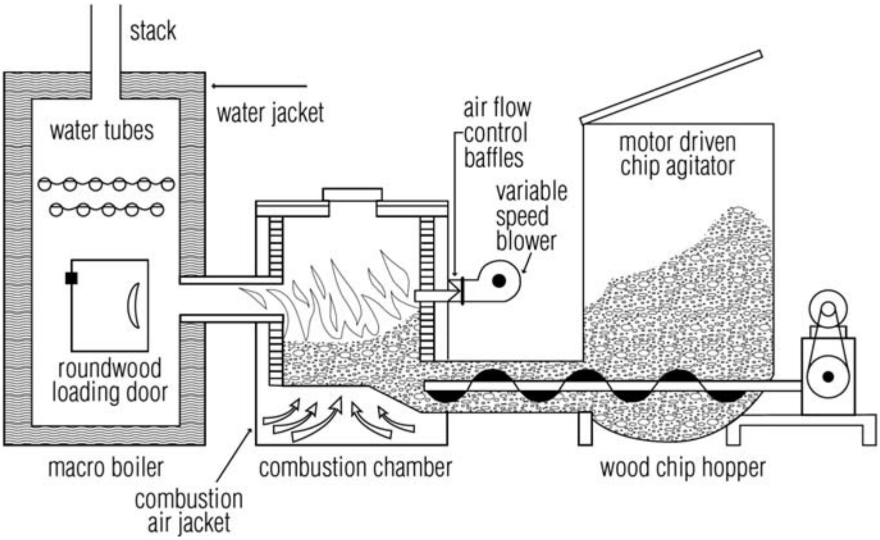


Fig. 9.3 An example of a modern woodchip heating system

plants in the wood and paper products industry use wood waste to produce their own steam and electricity. This saves these companies money because they do not have to dispose of their waste products and they do not have to purchase as much electricity.

Another advantage of solid biofuels is that the net carbon dioxide emissions that are added to the atmosphere by the burning process are only derived from the fossil fuels that were used to plant, fertilize, harvest, and transport the solid biomass. Likewise, chip combustion contributes less pollution and is a renewable resource. Modern woodchip combustion also gives the opportunity to use mill waste and lower grade wood from thinning operations. Wood chip fuel produced from such residues is cheaper than cordwood and pellet fuels. While the capital costs of wood chip heating systems are higher than oil-based systems, the operating costs are lower.

9.4.1.1 Combustion of Coal as a Biomass Energy Source: Pros and Cons

Coal is a solid fossil fuel formed in ecosystems where plant remains were preserved by water and mud during oxidization and biodegradation, thus sequestering atmospheric carbon present thousands or even millions of years ago. It is composed primarily of carbon and hydrogen along with small quantities of other elements, notably sulfur. Such elements are the primary source of pollution when the coal is finally burned. Since coal is the largest source of fuel for the generation of electricity worldwide, as well as the largest worldwide source of carbon dioxide emissions, its contribution to climate change and global warming is immense. In terms of carbon dioxide emissions, coal is slightly ahead of petroleum and about double that of natural gas. In addition, coal is extracted from the ground by coal mining, either by underground mining or by open pit mining (surface/strip mining). The practices of mining coal are deleterious to the local environment as seen in mountain top removal with strip mining, pollution of streams and rivers, and destruction of ecosystems.

In recent years, there has been talk about "clean coal". This is an umbrella term used in the promotion of the use of coal as an energy source by emphasizing methods being developed to reduce its environmental impact. These efforts include chemically washing minerals and impurities from the coal, gasification (see also IGCC), treating the flue gases with steam to remove sulfur dioxide, and carbon capture and storage technologies to capture the carbon dioxide from the flue gas. These methods and the technology used are described as *clean coal technology*, and such technology is a popular conversational topic for politicians. Clean coal can certainly be beneficial to the energy security of a country, but it is unlikely that coal will ever be truly clean. The same is true for most solid biofuels. Over 2 billion people currently cook every day and heat their homes by burning biomass, and this process is not "clean." In the nineteenth century, for example, wood-fired steam engines were common and contributed significantly to unhealthy air pollution seen during the industrial revolution. Today, the black soot that is being carried from Asia to polar ice caps appears to be causing them to melt faster in the summer.

9.4.1.2 Does Wood as a Solid Biofuel Offer Any Benefits as a Transportation Fuel?

With current technology, solid biofuels are not ideally suited for use as a transportation fuel. Most transportation vehicles require power sources with high-energy density, such as that provided by internal combustion engines. These engines generally require clean burning fuels, which are in liquid form, and to a lesser extent, compressed gases. Liquid biofuels are more portable, and they can be pumped, which makes handling much easier. This is why most transportation fuels are liquids. Non-transportation applications such as boilers, heaters, and stoves can usually tolerate the low-energy density contained in solid fuels, but technologies are being developed to make better use of solid fuels. Wood and its by-products can now be converted through process such as gasification into biofuels such as wood gas (synthesis gas), biogas, methanol, or ethanol fuel; however, further development may be required to make these methods affordable and practical.

Because solid fuels have inherent problems of relatively high costs, air pollution on combustion, and production inefficiency, one has to look at other, less polluting, more efficient, lower cost fuel sources. These include bioalcohol and biogas, which are covered in the next two sections. In contrast to the above, energy harvesting via bioreactors (methane generators) is a cost-effective solution, as for example, when applied to the animal solid waste product (manure) disposal issues faced by the dairy farmer. They can produce enough biogas/natural gas (methane, CH_4) to run a farm and work quite well in internal combustion engines (see Section 9.4.4)

9.4.2 Bioalcohol

The most abundant source of ethanol is the hydration of ethylene ($CH_2{=}CH_2$) derived from petroleum and other fossil fuels. While bioalcohols (especially bioethanol) have been in use for hundreds of years, it is only relatively recently that ethanol from biological sources has become more substantial. Ethanol fuel is now the most common biofuel worldwide, particularly in Brazil and the United States. Alcohol fuels are produced by fermentation of sugars derived from energy crops, such as corn, sugarcane, sugar beets, sorghum, wheat, or any sugar or starch that alcoholic beverages can be made from, including potatoes and fruit waste. Creation of ethanol starts with the energy of the Sun, carbon dioxide from the atmosphere and nutrients from soil, which allow the feedstocks to grow. Plants produce sugars such as glucose through the process of photosynthesis ($6CO_2 + 6H_2O$ + light $\rightarrow C_6H_{12}O_6 + 6O_2$). During ethanol fermentation, performed primarily by yeast (*Saccharomyces* spp.), glucose is decomposed into ethanol and carbon dioxide ($C_6H^{12}O_6 \rightarrow 2C_2H_6O + 2CO_2$+ heat). During combustion, ethanol reacts with oxygen to produce carbon dioxide, water, and heat ($C_2H_6O + 3O_2 \rightarrow 2CO_2 + 3H_2O$ + heat). Since two molecules of ethanol are produced for each glucose molecule, there are equal numbers of each type of molecule on each side of the equation, and the net reaction for the overall production and consumption of ethanol is simply (light → heat). The heat of the combustion of ethanol can be used to drive the piston of an internal combustion engine (Agarwal, 2007). Ethanol is considered "renewable" because it is primarily the result of conversion of the Sun's energy into usable energy.

The most common steps in the production of bioalcohols are as follows: (1) enzymatic digestion (to release sugars from stored starches); (2) fermentation of the sugars through the action of microorganisms (yeasts that generate alcohol in the process); (3) distillation (to concentrate the alcohol); and (4) drying (to remove residual water that can prevent the liquid from being used as a fuel). The distillation process, in particular, requires significant energy input as heat (often using natural gas from fossil fuels). Likewise, we have already discussed some of the concerns over the amount of land needed to produce ethanol fuel crops and how land used for this purpose seems to be adversely impacting usable land for food resources (see Sections 9.2 and 9.3).

More recently, attention has focused on making use of non-food crops or the waste biomass leftover from other crops. Plant biomass high in cellulose (including wood and paper waste) can also be tapped for its stored sugar content. Once the cellulose is broken down through the action of enzymes and microorganisms (e.g., cellulose-decomposing fungi), it can be used as a starting material for fermentation and alcohol production. However, since cellulose is extremely stable, it is very difficult to break apart. In addition, it is commonly linked to lignin (another support molecule found in the cell walls of plants), and the resulting "lignocellulose" is one of the toughest plant materials to decompose. One good example of a plant high in both sugars and cellulosic biomass is sugarcane. The cane can be pressed to extract its juice which has high levels of sugar. The leftover bagasse, the waste left after sugarcane is pressed, can also be dried and used as a solid biomass to provide heat for the distillation process after fermentation.

Ethanol can be used in automobile engines as a replacement for gasoline (Agarwal, 2007). It can be mixed with gasoline to any percentage; however, most existing automobile gasoline engines can only run on blends up to 15% bioethanol with petroleum/gasoline. Gasoline with ethanol added has a higher octane, which means that the engine can typically burn hotter, more efficiently, and more cleanly. In high-altitude (thin air) locations, some states mandate a mix of gasoline and ethanol as a winter oxidizer to reduce atmospheric pollution emissions (Agarwal, 2007). The top five producers of ethanol for fuel are the United States, Brazil, China, India, and France. Brazil and the United States accounted for ∼70% of all ethanol production, with total world production of 13.5 billion US gallons (40 million tonnes).

9.4.2.1 History of Bioalcohol Use

Throughout the history of its use as a fuel, bioethanol has been at the crux of supply, demand, and often subtle price variations between ethanol and other liquid fuels. Since ancient times, ethanol has been used for lamp oil and cooking, along with plant and animal oils. Before the US Civil War, many US farmers had alcohol stills that could turn crop waste into virtually free lamp and stove fuel. In 1826, Samuel Morey, experimented with a prototype internal combustion engine that used ethanol (combined with turpentine and ambient air then vaporized) as fuel. At that time, his discovery was overlooked, mostly due to the success of steam power. And while ethanol was known of for decades, it received little attention as a fuel until 1860, when Nicholas Otto began experimenting with internal combustion engines. Such a use would have meshed well with the farmers' alcohol stills. However, the Industrial Age caused many farmers to move to city jobs, leaving their farms and ethanol fuel stills behind. Despite this, alcohol remained popular for lighting, cooking, and industrial purposes. In 1862, and again in 1864, a tax on alcohol was passed in the United States to help pay for the Civil War. This increased the price of ethanol dramatically, causing farmers not to be able to sell their ethanol due to reduced demand. Consequently, farmers used the ethanol themselves. Later in the 1890s, alcohol-fueled engines were used in farm machinery, train locomotives, and eventually cars in the United States and Europe. Henry Ford's first car, the Quadracycle, was released in 1896 and ran on 100% ethanol. Thus ethanol was the first fuel used by American cars before gasoline.

The early 1900s were an important time in the history of how gasoline eventually overtook alcohol fuels as the fuel of choice for automobiles. In 1902, the Paris alcohol fuel exposition exhibited alcohol-powered cars, farm machinery, lamps, stoves, heaters, laundry irons, hair curlers, coffee roasters, and many household appliances that were powered by alcohol. A few years later, the United States repealed the alcohol tax while under Theodore Roosevelt, who was strongly against fossil fuels like oil. This allowed the price of ethanol (∼14 cents/US gallon) to fall below the price of gasoline (∼22 cents/US gallon). Unfortunately, in 1907, the discovery of new oil fields in Texas caused the price of gasoline to drop to between 18 and 22 cents/US gallon, and at the same time, alcohol fuel prices rose to around 25–30 cents/US gallon. Because of the struggle between the markets for alcohol and gasoline, Henry Ford introduced his Ford Model T in 1908. It had an engine that could run on either ethanol or gasoline or a mix of both. Ford continued to be an advocate for ethanol as a fuel, even during the prohibition. But in 1919, the prohibition police destroyed virtually all corn-alcohol stills, putting what appeared to be an end to the use of alcohol as a fuel in the United States.

It is interesting to note that in many other parts of the world, people believed that ethanol would be the fuel that would eventually replace petroleum. Experiments on the use of alcohol as fuel continued in these other parts of the world because there continued to be a battle between the prices of ethanol and gasoline. For example, in 1923, the price of alcohol from molasses was less than 20 cents/US gallon, while retail gasoline prices had reached an all-time high of 28 cents/gal. At about the same time, Standard Oil Co. experimented with a 10% alcohol/90% gasoline blend to increase octane and stop engine knocking. By the mid-1920s, ethanol blended with gasoline was standard in every industrialized nation except the United States. By 1925, France, Germany, Brazil, and other countries had already passed “mandatory blending” laws. During this time, Ford Motor Co. was building cars that could be changed slightly to run on gasoline, alcohol, or kerosene. It is noteworthy that the situation changed in the United States. In 2007, Portland, Oregon, became the first city in the United States to require all gasoline sold within city limits to contain at least 10% ethanol. As of January 2008, three states – Missouri, Minnesota, and Hawaii – require ethanol to be blended with gasoline motor fuel. Many cities are also required to use an ethanol blend due to non-attainment of federal air quality goals.

In 1933, faced with the 25% unemployment rate of the Great Depression, the US government considered tax advantages that would help ethanol production to increase employment among farmers. The “farm chemurgy” *movement*, supported by farmers, Republicans, and Henry Ford, searched for new crop-based products from farms (such as soybean-derived plastics) and supported alcohol fuel. From 1933 to 1939, The American Petroleum Institute argued that such government help would hurt the oil industry, reduce state treasuries, and cause an unhealthy criminal “bootlegger” atmosphere around fueling stations. They claimed alcohol fuel was in every way inferior to gasoline, and eventually, the government did not pass any alcohol fuel incentives. Pressure from the oil companies has also been blamed for the demise of various ethanol fuel companies. For example, in 1937, *Agrol*, an ethanol-gasoline blend, was sold at 2,000 service stations in the United States. Agrol plant managers complained of sabotage and bitter infighting elicited by the oil industry that resulted in cheaper gasoline prices. At this time, alcohol was 25 cents/gal, while gasoline was 17–19 cents/gal. In 1939, Agrol production shut down because of a lack of a viable market, and by 1940, the US Midwestern alcohol fuel movement had disintegrated.

Fuel pressures that arose during World War II resulted in yet another revival of alcohol as fuel, and new technologies were developed to make use of such a fuel. For example, on October 14, 1947, legendary test pilot Chuck Yeager became the first man to fly faster than Mach 1, the speed of sound. He was piloting the Bell X-1, a bullet-shaped rocket plane (powered by liquid oxygen and alcohol fuel) that was the first in a series of secret high-speed research aircraft that were flown out of California's Edwards Air Force Base in the late 1940s and 1950s. Another boost for ethanol came in 1973, when a worldwide energy crisis began. This caused ethanol to once again become cheaper than gasoline. Gasoline containing up to 10% ethanol has been increasing in use in the United States since the late 1970s. By the mid-1980s, over 100 new corn-alcohol production plants had been built, and over a billion US gallons of ethanol were sold for fuel each year. However, the tide would turn against ethanol again when, in the late 1980s and 1990s, new oil wells were discovered and the price of gasoline once again became much cheaper than alcohol fuel. This time, however, ethanol plants were able to get subsidies from the US government to support farmers who were growing energy crops.

Between 1997 and 2002, three million US cars and light trucks were produced which could run on E85, a blend of 85% ethanol with 15% gasoline (Agarwal, 2007). Ford, DaimlerChrysler, and GM are among the automobile companies that sell "flexible fuel" cars, trucks, and minivans that can use gasoline and ethanol blends that range from pure gasoline up to 85% ethanol (E85). Such *flex-fuel vehicles* are now having a significant impact on an attempted alcohol fuel transition because they allow drivers to choose different fuels based on price and availability. The primary problem, however, is that there are almost no gas stations that sell E85 fuel, and the ones that do are mostly located in the Midwest part of the-United States. During this time, the invasion of Iraq, and the subsequent turmoil it caused, allowed Americans to become aware of their dependence on foreign oil. In addition, the demand for ethanol fuel produced from field corn was spurred by the discovery that *methyl tertiary butyl ether* (*MBTE*) was contaminating groundwater. MBTE was the most common fuel oxygenate additive used to reduce carbon monoxide emissions. The groundwater contamination issue eventually led to MTBE being banned in almost 20 states by 2006. In 2003, California was the first state to start replacing MTBE with ethanol, and other states start switching soon afterward. This switch thus opened a new market for ethanol fuel, the primary substitute for MBTE. This event, coupled with worry over climate change, caused the leading alternative energy sources, including bioalcohol, solar and wind power, to expand ~20–30% each year (Agarwal, 2007). At a time when corn prices were around US $2 a bushel, corn growers recognized the potential of this new market and delivered accordingly.

Since 2003, crude oil prices have risen by as much as 80%, and gasoline and US diesel fuel prices have risen by as much as 50%, only to fall again in highly volatile markets. These rises are caused by hurricane damage to oil rigs in the Gulf of Mexico, attacks on Iraqi oil pipelines, disruptions elsewhere, and rising demand for gasoline in Asia, particularly as Asians buy more cars. Gasoline prices rise as ethanol prices stay the same, due to rapidly a growing ethanol supply and federal tax subsidies for ethanol production. In 2008, the United Nations urged that there be a cessation in the provision of subsidies for food-based biofuels, including ethanol, due to rising controversies over fuel price fluctuations, production costs, and supply/demand variables.

9.4.2.2 Advantages and Disadvantages of Bioalcohol: Can Corn Do the Job?

As mentioned above, one advantage of bioalcohol is that it can be produced from a variety of feedstocks, including sugarcane, bagasse, miscanthus, sugar beet, sorghum, grain sorghum, switchgrass, barley, hemp, kenaf, potatoes, sweet potatoes, cassava, sunflower, fruit, molasses, corn, stover, grain, wheat, straw, cotton, biomass in general as well as many types of cellulose waste and harvestings. As discussed in Section 9.2, the primary advantage of biofuels such as bioalcohol is that they are relatively "renewable" or carbon neutral as compared to fossil fuels. Carbon dioxide, a greenhouse gas, is emitted during fermentation and combustion. However, this by-product is canceled out by the greater uptake of carbon dioxide by the plants as they grow to produce the input material for the alcohol. The replacement of MTBE (an environmental toxin) with ethanol as an oxygenate in gasoline has also reduced carbon monoxide emissions (Agarwal, 2007). However, ethanol is not a completely clean burning fuel. When burned in the atmosphere, harmful nitrous oxide gases are produced, including nitrogen dioxide which contributes to the formation of "brown smog." Acetaldehyde and other aldehydes are also produced when alcohols are oxidized. When only a 10% mixture of ethanol is added to gasoline (as is common in E10 gasohol), aldehyde emissions increase by as much as 40%, and these components are not regulated in emissions laws.

The use of alcohol in various mixes with gasoline is also cited as the reason for reducing prices. According to a 2008 analysis by Iowa State University, the growth in US ethanol production has caused retail gasoline prices to be 29–40 cents/gal lower than would otherwise have been the case. However, because alcohol mixes with both gasoline and with water, ethanol fuels are often diluted after the drying process by absorbing environmental moisture from the atmosphere. Water in alcohol-mix fuels reduces efficiency, makes engines harder to start, causes intermittent operation (sputtering), and oxidizes aluminum and steel components (Agarwal, 2007). Ethanol itself is also corrosive to standard fuel systems, rubber hoses and gaskets, aluminum, and combustion chambers. It also corrodes fiberglass fuel tanks such as those used in marine engines. For higher ethanol percentage blends, and 100% ethanol vehicles, engine modifications are required. In addition, corrosive ethanol cannot be transported in gasoline pipelines, so more expensive stainless-steel tank trucks are required to deliver ethanol to customers. Perhaps even more problematic, ethanol fuel has less BTU energy content, which means it takes more fuel to produce the same amount of work. Even dry ethanol has roughly one-third lower energy content per unit of volume compared to gasoline.

Current interest in ethanol fuel in the United States mainly lies in bioethanol, produced from corn, but there has been considerable debate about how useful bioethanol will be in replacing fossil fuels in vehicles. As described in Section 9.3, concerns relate to the large amount of arable land required for energy crops as well as energy and pollution balance of the whole cycle of ethanol production.

Large-scale farming is necessary to produce agricultural alcohol and this requires substantial amounts of cultivated land. Farming may also involve a decline in soil fertility due to reduction of organic matter, a decrease in water availability and quality, an increase in the use of pesticides and fertilizers, deforestation, and potential dislocation of local communities. Likewise, "food vs. fuel" is the dilemma regarding the risk of diverting farmland away from food crops and toward the production of biofuels. The "food vs. fuel" debate is internationally controversial, with good arguments on all sides. Recent developments with cellulosic ethanol production and commercialization may allay some of these concerns.

One rationale given for extensive ethanol production in the United States is its benefit to energy security by shifting the need for some foreign-produced oil to domestically produced energy sources. In the United States, the number of ethanol factories has almost tripled from 50 in 2000 to about 140 in 2008. A further 60 or so are under construction, and many more are planned. The debates surrounding bioalcohol production are needed to prevent too many resources being placed into a technology that could have too many problems to make energy issues any better. Such projects are being challenged by residents at courts in Missouri (where water is drawn from the Ozark Aquifer), Iowa, Nebraska, Kansas (all of which draw water from the non-renewable Ogallala Aquifer), central Illinois (where water is drawn from the Mahomet Aquifer) and Minnesota. With large current unsustainable, non-scalable subsidies, ethanol fuel still costs much more per distance traveled than current high gasoline prices in the United States.

The United States produces and consumes more ethanol fuel than any other country in the world. This is partly due to energy crisis issues and price battles between ethanol and gasoline as explained in Section 9.4.2.1. However, one of the main incentives has been legislation that has been passed. A senior member of the House Energy and Commerce Committee, Congressman Fred Upton, introduced the legislation to use at least E10 fuel by 2012 in all cars in the United States. Likewise, the US Energy Independence and Security Act of 2007 requires American "fuel producers" to use at least 36 billion gallons of biofuel in 2022. This is nearly a five-fold increase over current levels. Such legislation is at the heart of the push to use corn as fuel and causing a significant shift of resources away from food production. Essentially all ethanol fuels in the United States are now produced from corn. As described above, the amount of land used to generate such large amounts of corn ethanol is a central concern behind the food vs. fuel debate and other environmental issues. Unfortunately, corn is a very energy-intensive crop. In the current alcohol-from-corn production model in the United States, considering the total energy consumed by farm equipment, cultivation, planting, fertilizers, pesticides, herbicides, and fungicides made from petroleum, irrigation systems, harvesting, transport of feedstock to processing plants, fermentation, distillation, drying, transport to fuel terminals and retail pumps, and lower ethanol fuel energy content, the net energy content value added and delivered to consumers is very small. And, the net benefit (all things considered) does little to reduce unsustainable imported oil and fossil fuels required to produce the ethanol.

The problem here is that current processes for the production of ethanol from corn use only a small part of the corn plant. The corn kernels are taken from the corn plant and only the starch is transformed into ethanol. Corn is typically 66% starch and the remaining 33% is not fermented. This unfermented component is called distillers grain, which is high in fats and proteins, and makes good animal feed. US corn-derived ethanol costs 30% more because the corn starch must first be converted to sugar before being fermented into alcohol. Here enzymes are required to first liquefy the starch. A second enzyme converts the liquefied starch to sugars, which are fermented by yeast into ethanol and carbon dioxide. The released CO_2 can also be captured and sold for use in carbonating beverages and in the manufacture of dry ice; however, this is not always done. Despite the cost differentials in production, in contrast to Japan and Sweden, the United States does not import much Brazilian ethanol because of US trade barriers corresponding to a tariff of 54-cent/gal – a levy designed to offset the 51-cent/gal blender's federal tax credit that is applied to ethanol no matter its country of origin.

9.4.2.3 Ethanol Derived from Sugarcane

Sugarcane or sugar cane (*Saccharum*) is a genus of 6–37 species (depending on taxonomic interpretation) of 2–6 m tall perennial grasses (family Poaceae, tribe Andropogoneae). They are native to warm temperate to tropical regions of the world, having stout, jointed, fibrous stalks that are very rich in sugar. Sugarcane is one of the most efficient photosynthesizers in the plant kingdom. It is able to convert up to 2% of incident solar energy into biomass. All of the sugarcane species interbreed, and all of the major commercial cultivars are complex hybrids. Sugarcane originated from tropical South and Southeast Asia. Different species likely originated in different locations with *S. barberi* originating in India and *S. edule* and *S. officinarum* from New Guinea. The thick stalk stores energy as sucrose in the sap. This sap can be extracted by pressing, and sugar is extracted by evaporating the water from the resulting juice. The use of crystallized sugar has been reported for over 5,000 years in India. The methods of growing sugarcane and processing sugar were transferred to China from India in the seventh century, and around the eighth century C.E., Arabs introduced sugar to the Mediterranean, Mesopotamia, Egypt, North Africa, and Spain. By the tenth century, there was virtually no village in Mesopotamia that did not grow sugarcane, and sugarcane was among the early crops brought to the Americas by the Spaniards.

Currently, about 200 countries grow sugarcane to produce $\sim$1,325 million tons of sugary biomass. As of 2005, the world's largest producer of sugarcane by far is Brazil, followed by India. Uses of sugarcane include the production of sugar, Falernum, molasses, rum, soda, cachaça (the national spirit of Brazil), and ethanol for fuel. Ethanol is produced most typically by yeast (*Saccharomyces* species) fermentation of the sugar extracted from the cane. The bagasse that remains after crushing the sugarcane may also be burned to provide heat both for distillation processes and for the production of electricity. Because of its high cellulose content, it may also be used as raw material for paper and cardboard, as a starting material for cellulosic ethanol, and is branded as "environmentally friendly" because it is a renewable by-product of sugar production.

Brazil has the largest and most successful sugarcane biofuel programs in the world, and it is considered to have the world's first sustainable biofuels economy. In 2006, Brazilian ethanol provided ~18% of the country's transportation fuel, and by April 2008, more than 50% of the fuel used as a replacement to gasoline was derived from sugarcane. As a result of the increasing use of ethanol, together with the exploitation of domestic deep water oil sources, Brazil reached complete self-sufficiency in oil supply in 2006, whereas years ago, the country had to import a large share of the petroleum needed for domestic consumption. Since 1977, the government made it mandatory to blend 20% of ethanol (E20) with gasoline, requiring just minor adjustments on standard gasoline engines (Agarwal, 2007). Today, the mandatory blend is allowed to vary nationwide between 20 and 25% ethanol (E25), and it is used by all normal gasoline vehicles. In addition, three million Brazilian cars run on 100% anhydrous ethanol and six million flexible fuel vehicles are now active in Brazil. Introduced to the market in 2003, these flex-fuel vehicles became a commercial success, representing around 23% of Brazil's standard motor vehicles. The ethanol-powered and flex vehicles have also been manufactured to tolerate even *hydrated ethanol*, an azeotrope comprised of 95.6% ethanol and 4.4% water.

Together, Brazil and the United States lead the industrial world in global ethanol production, accounting for ~70% of the world's total production and nearly 90% of the ethanol used for fuel. However, Brazil's sugarcane-based industry is far more efficient than the US corn-based industry. Brazilian distillers are able to produce ethanol for less than 22 cents/l, compared with the 30 cents/l for corn-based ethanol. Sugarcane plantations cover 3.6 million ha of land for ethanol production, representing only 1% of Brazil's arable land, with a productivity of 7,500 l of ethanol/ha, as compared with the US maize ethanol productivity of 3,000 l/ha. However, as with corn in the United States, significant areas of land are likely to be dedicated to sugarcane in future years, as demand for ethanol increases worldwide. The expansion of sugarcane plantations is already placing pressure on environmentally sensitive native ecosystems, including rainforests in South America, where deforestation is contributing to the elevation of greenhouse gases, loss of habitat, and a reduction in biodiversity.

In some respects, it is good that sugarcane cultivation requires a tropical or subtropical climate, with a minimum of 24 in. of annual rainfall. This has limited its use in North America and has forced the development of technologies that are better suited to North America. However, sugarcane production in the United States is occurring in Florida, Louisiana, Hawaii, and Texas, and the first three ethanol plants to produce sugarcane-based ethanol are expected to go online in Louisiana by mid-2009.

9.4.2.4 Ethanol Derived from Biomass

Plant biomass is the most abundant renewable resource on Earth and is also a potential source of fermentable sugars for the production of bioalcohol. As in the production of other bioalcohols, fermentation of sugars derived from biomass can be accomplished through the action of microorganisms that generate alcohol, which then needs to be distilled and dried to remove residual water. However, conversion of plant biomass to fermentable sugars typically requires manual and/or chemical pretreatment and the hydrolysis of lignocellulose, a structural material that comprises most of the plant biomass. *Lignocellulose* is composed primarily of cellulose (a β-1,4-linked glucose polymer), hemicellulose (with various types of 5- and 6-carbon sugar polymers), and lignin (a polymer of phenolic compounds) (Table 9.1). Unfortunately, the use of lignocellulose as a fuel has been curtailed by its highly rigid structure. Consequently, an effective pretreatment is needed to liberate the cellulose from the crystalline structure of lignin so as to render it accessible for subsequent hydrolysis (also called *cellulolysis*).

In contrast to ethanol produced from corn and sugarcane starches and sugars, cellulose is contained in nearly every natural, free-growing plant, tree, and shrub, in every meadow, forest, and field all over the world. Since the components of lignocellulose cannot be digested by humans, the production of cellulosic ethanol does not have to compete with the production of food, and if marginal lands are used to grow cellulose-rich crops, it does not have to compete with the land used to grow food crops. According to US Department of Energy studies conducted by the Argonne National Laboratories and the University of Chicago, the major benefit of cellulosic ethanol is that it can reduce greenhouse gas emissions by as much as 85% over reformulated gasoline. By contrast, starch ethanol from corn most frequently uses natural gas to provide energy for processing and may not reduce greenhouse gas emissions at all, depending on how the starch-based feedstock is produced. In addition, cellulosic crops require fewer inputs, such as fertilizer, herbicides, and other

Table 9.1 Composition of various types of cellulosic biomass material (% dry weight)

Material	Cellulose	Hemicellulose	Lignin	Ash	Extractives
Softwood barks	18–38	15–33	30–60	0.8–1.0	4–6
Hardwood barks	22–40	20–38	30–55	0.8–1.0	6–8
Soft woods	42–44	27–29	28–31	0.5–0.6	3–5
Newspapers	40–55	25–40	18–30	–	–
Hard woods	45–47	30–35	20–24	0.6–0.8	5–8
Grasses	25–40	25–50	10–30	–	–
Wheat straw	37–41	27–32	13–15	11–14	7–9
Chemical pulps	60–80	20–30	2–10	–	–
Cornstalks	39–47	26–31	3–5	12–16	1–3
Cotton and flax	80–95	5–20	–	–	–
Algae	20–40	20–50	–	–	–

Modified from Demirbas et al. (2005).

chemicals that can pose risks to wildlife. Their extensive roots improve soil quality, reduce erosion, and increase nutrient capture. Herbaceous energy crops reduce soil erosion by greater than 90%, when compared to conventional food crop production. This can translate into improved water quality for rural communities. Additionally, cellulosic energy crops add organic material to depleted soils and can increase soil carbon as long as the land being used is not totally stripped of plant material. In addition, the price per ton of the raw cellulose material is much cheaper than for grains or fruits, and since cellulose is the main component of plants, the whole plant can be harvested. This results in much better yields per acre, up to 10 t, instead of 4 or 5 t for the best crops of grain. Thus, production of ethanol from lignocellulose has the advantage of having abundant and diverse resources that do not require agricultural effort or costs for growth; however, it does require a greater amount of processing to make the sugar monomers available to the microorganisms that produce ethanol during fermentation.

The first attempt at commercializing a process for ethanol from wood was undertaken in Germany in 1898. It involved the use of dilute acid to hydrolyze the cellulose to glucose and was able to produce 7.6 l of ethanol/100 kg of wood waste (18 gal/t). The Germans soon developed an industrial process optimized for yields of around 50 gal/t of biomass. This process soon found its way to the United States, where two commercial plants were put into operation in the southeast during World War I. These plants used what was called "*the American Process,*" a one-stage dilute sulfuric acid hydrolysis of wood products and waste. Although the yields were half that of the original German process (25 vs. 50 gal of ethanol/ton), the output of the American process was much higher. However, a drop in lumber production forced these ethanol plants to close shortly after the end of World War I. In the meantime, a small, but steady amount of research on dilute acid hydrolysis has continued at the USDA's Forest Products Laboratory in Madison, WI.

Currently, *corn stover* (leaves and stalks of maize left in the field after harvest), switchgrass, miscanthus, and woodchips are some of the more popular cellulosic materials for ethanol production. For example, switchgrass (*Panicum virgatum* L.) is a native prairie grass, known for its hardiness, rapid growth (from 2 to 6 ft tall), and high cellulose content. It can be grown in most parts of the United States, including swamplands, plains, streams, and along the shores and interstate highways. Since switchgrass yields twice as much ethanol per acre than corn, less land is needed for production, helping to prevent habitat fragmentation. It is unfortunate, however, that typical municipal practices discard the majority of cellulosic biomass. It is estimated that over 320 million tons of cellulose-containing raw materials, which could be used to generate ethanol, are thrown away each year. According to the International Energy Agency, this includes 36.8 million dry tons of urban wood wastes, 90.5 million dry tons of primary mill residues, 45 million dry tons of forest residues, and 150.7 million dry tons of corn stover and wheat straw. Likewise, organic waste makes up 71.5% of all landfill wastes deposited each day, consisting of large amounts of wood, envelopes, newsprint, grass, leaves, food scraps, office paper, corrugated cardboard, and agricultural composites as well as small amounts of manures, glossy paper, and paper ledger. All of these materials can be converted into fuels, and transforming such leftovers into ethanol can actually reduce solid waste disposal costs and provide as much as 30% of the current fuel consumption in the United States. Thus, the raw material to produce cellulosic ethanol is basically free, and it may actually have a negative cost, where ethanol producers can get paid to take it away.

To date, the available pretreatment techniques include acid hydrolysis, steam explosion, alkaline wet oxidation, ozone pretreatment, and ammonia fiber expansion. Besides effective cellulose liberation, an ideal pretreatment has to minimize the formation of degradation products because of their inhibitory effects on subsequent hydrolysis and fermentation processes. The presence of inhibitors will not only complicate ethanol production, but also, increase the cost of production by adding detoxification steps. Even though pretreatment by acid hydrolysis is probably the oldest and most studied pretreatment technique, it produces several potent inhibitors including furfural and hydroxymethyl furfural (HMF) which are toxic compounds present in lignocellulosic hydrolysate. Ammonia fiber expansion (AFEX) is currently the only pretreatment which features promising efficiency with no inhibitory effect in resulting hydrolysate, although experiments using fungal organisms that naturally breakdown the biomass are showing some promise for the release of cellulose polymer from lignocellulose. In the hydrolysis process, these polymers are broken down to free the sugar before it is fermented for alcohol production.

There are two primary approaches to cellulose hydrolysis (*cellulolysis*): a chemical approach using acids, or an enzymatic approach. In the traditional method, hydrolysis is performed by attacking the cellulose with an acid. Dilute acid may be used under high heat and high pressure or more concentrated acid can be used at lower temperatures and atmospheric pressure. The product from this hydrolysis is then neutralized and yeast fermentation is used to produce ethanol. As mentioned, a significant obstacle to the dilute acid process is that the hydrolysis is so harsh that toxic degradation products can be produced that can interfere with fermentation. In enzymatic hydrolysis, cellulose can be broken into glucose molecules by cellulase enzymes. Such enzymes are commonly found in the digestive systems of ruminants, such as cows, sheep, and termites, where a collection of enzymes are produced by bacteria. They are also found in naturally occurring fungi and soil bacteria that are part of the global carbon cycle. Using a similar enzymatic system, lignocellulosic materials can be enzymatically hydrolyzed under relatively mild conditions (50°C and $\mathrm{pH} = 5$), thus enabling effective cellulose breakdown without the formation of by-products that would otherwise inhibit enzyme activity. To be viable for large-scale fuel production, all major pretreatment methods, including dilute acid pretreatment, require some type of enzymatic hydrolysis step to achieve the high sugar yields required for ethanol fermentation. Various enzyme companies have already contributed significant technological breakthroughs in cellulosic ethanol production through the mass production of various cellulase enzymes at competitive prices. Iogen Corporation, for example, is a Canadian producer of enzymes for an enzymatic hydrolysis process that uses "specially engineered enzymes."

Traditionally, baker's yeast (*Saccharomyces cerevisiae*) has long been used in the brewery industry to produce ethanol from hexoses (6-carbon sugars). Yeast cells are especially attractive for cellulosic ethanol processes because they have been used in biotechnology for hundreds of years. They are tolerant to high ethanol and inhibitor concentrations, and they can grow at low pH values, which avoids bacterial contamination. Due to the complex nature of the carbohydrates present in lignocellulosic biomass, a significant amount of xylose and arabinose (5-carbon sugars derived from the hemicellulose portion of the lignocellulose) is also present in the hydrolysate. For example, in the hydrolysate of corn stover, approximately 30% of the total fermentable sugars are xylose. Thus, the ability of the fermenting microorganisms to utilize the whole range of sugars available from the hydrolysate is vital to increase the economic competitiveness of cellulosic ethanol.

In recent years, metabolic engineering for microorganisms used in bioethanol production has shown significant progress. Besides *Saccharomyces*, bacteria such as *Zymomonas mobilis* and *Escherichia coli* have been targeted for metabolic engineering to improve their fermentation abilities, and thus, improve cellulosic ethanol production. Likewise, genetically engineered yeasts have been described that efficiently ferment xylose and arabinose sugars. Some species of bacteria have also been determined to be capable of the direct conversion of cellulose into ethanol. One example is *Clostridium thermocellum*, which utilizes a complex cellulosome to breakdown cellulose and synthesize ethanol. However, *C. thermocellum* also produces contaminating by-products during cellulose metabolism, including acetate and lactate, in addition to ethanol. While this lowers the efficiency of the process, further research into the ethanol-producing pathways of such organisms holds great potential for future improvements in the generation of bioalcohol. Enzymes from thermophilic organisms are also particularly well suited for industrial applications because they are typically thermostable and relatively tolerant of other stresses such as pH extremes. Genes for a variety of thermostable cellulase enzymes from both bacteria and fungi are currently being assessed for their ability to improve cellulosic ethanol efficiency.

Similarly, much effort has been devoted to developing transgenic plants as bioreactors to produce heterologous proteins, including industrial cellulase enzymes (Park et al., 2003). Such plants, expressing genes from other species, are typically fertile and grow normally, and they supply easy access to the enzymes needed when cellulose is to be broken into sugars. Manufacturing heterologous cellulases in crop plant bioreactors could significantly reduce costs associated with enzyme production and could offer a potentially high-volume alternative to traditional enzyme production methods. Other plant biotechnology approaches aim to improve the lignocellulose characteristics of the biomass crops themselves. This has been done in switchgrass, where alteration of gene expression in the lignin biosynthesis pathway has both increased and reduced the amount of lignin within the plant (Fig. 9.4A). The reduction of lignin in plant tissues allows easier access to the cellulose; however, the amount of reduction has to be carefully tailored so as not to cause the growing plant to collapse due to lack of support structure. Other approaches to improved cellulosic crops include more traditional breeding programs that identify

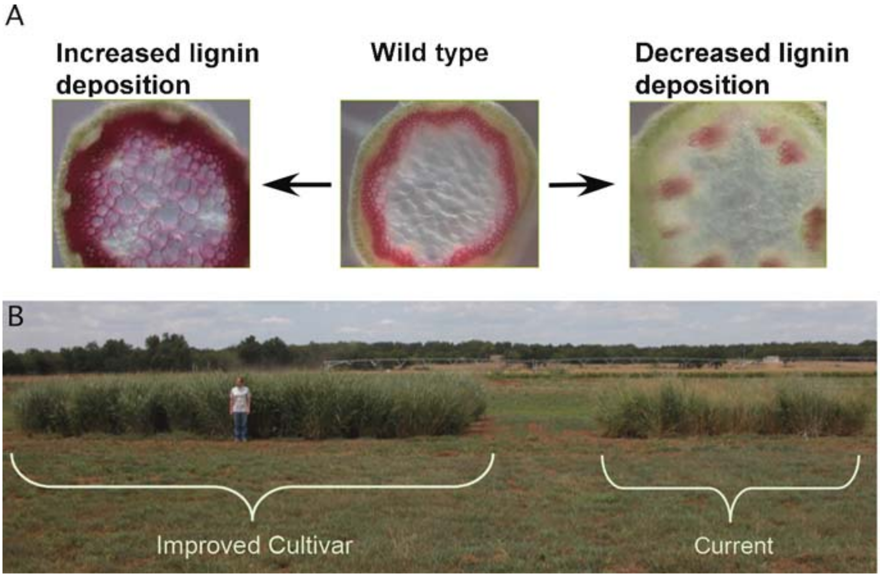


Fig. 9.4 Examples of improvements in cellulosic biomass. (**A**) Modifications in gene expression can result in both increased and decreased deposition of lignin in switch grass. (**B**) Genetic-based breeding programs can improve biomass in switch grass. Modified from Vermerris (2008) Genetic Improvement of Bioenergy Crops, Springer

useful traits to help develop superior varieties, including those that have enhanced biomass production (Fig. 9.4B).

It should be noted here that, while most efforts have focused on acid pretreatment and enzymatic hydrolysis of lignocellulose, gasification of the lignocellulosic raw material into gaseous carbon monoxide and hydrogen is also useful for ethanol production (Ahring and Westermann, 2007). The gasification process does not rely on chemical decomposition of the cellulose chain (cellulolysis). Instead of breaking the cellulose into sugar molecules, the carbon in the raw material is converted into *wood gas* (also called synthesis gas), using what amounts to partial combustion. The resulting carbon monoxide, carbon dioxide, and hydrogen may then be fed into a special kind of fermenter. Instead of sugar fermentation with yeast or bacteria, this process uses a bacterium named *Clostridium ljungdahlii*. *C. ljungdahlii* will ingest (eat) carbon monoxide, carbon dioxide, and hydrogen and produce ethanol and water. The ethanol can then be distilled and dried as usual. More recently, *C. thermocellum* (a thermophilic bacterium) has been found to be twice as efficient in making ethanol from carbon monoxide as *C. ljungdahlii*. Alternatively, the synthesis gas from gasification may be fed to a catalytic reactor where the synthesis gas is used to produce ethanol and other higher alcohols through a thermochemical process. Such technology development and the use of biotechnology will likely be key to the development of truly sustainable fuel sources in the future.

9.4.2.5 Biobutane as an Alternative Fuel

Biobutanol (also called *biogasoline*) has a longer hydrocarbon chain than ethanol. This causes it to be fairly non-polar, making it more similar to gasoline than ethanol. It is often claimed to provide a direct replacement for gasoline, because it can be used directly in internal combustion engines without modification. Butanol better tolerates water contamination and is less corrosive than ethanol, making it more suitable for distribution through existing pipelines for gasoline. In blends with diesel or gasoline, butanol is less likely to separate from the fuel than ethanol if the fuel is contaminated with water. There is also a vapor pressure co-blend synergy with butanol and gasoline containing ethanol. This better facilitates ethanol blending, thus allowing better storage and distribution of blended fuels. Butanol also has a high octane rating (over 100) and high energy content, is only about 10% lower than gasoline, and subsequently is about 50% more energy dense than ethanol (100% more so than methanol). Butanol's only major disadvantages are its high flashpoint (95°F or 35°C), potential toxicity (but not necessarily more than gasoline), and the fact that the distillation process requires a large energy input.

The feedstocks for biobutanol are the same as for bioethanol, including energy crops such as sugar beets, sugarcane, corn grain, wheat, and cassava as well as agricultural by-products such as straw, corn stalks, and various other biomass. Biobutanol is formed by acetone/butanol/ethanol fermentation (*ABE fermentation*) through the activity of the bacterium, *Clostridium acetobutylicum*, also known as the Weizmann organism. This process was first delineated by Chaim Weizmann in 1916 for the production of acetone from starch for making *cordite*, a smokeless gunpowder. At the time, the butanol was a by-product of this fermentation, forming twice as much butanol as acetone. The process also creates a recoverable amount of hydrogen gas and a number of other by-products, including acetic, lactic and propionic acids, acetone, and isopropanol.

Experimental modifications of the process have shown potentially high net energy gains with biobutanol as the only liquid product. However, the key research challenge that must be resolved is that butanol production inhibits microbial growth even at low concentrations. The Weizmann organism can only tolerate butanol levels up to 2%, compared to 14% for ethanol from yeast. Thus, the overwhelming constituent of the fermentation broth is water; so, an energy-intensive distillation step is required for purification. This may be acceptable if the goal is to produce butanol for use as a solvent, but if butanol is to gain traction as a fuel, energy inputs need to be minimal. Currently, biobutanol is far to expensive (∼$4/US gallon) to be viable as a fuel. However, a number of companies are working on the problem. For example, DuPont and British Petroleum (BP) are working together to help develop biobutanol as a fuel source. According to DuPont, existing bioethanol plants can cost-effectively be retrofitted to produce biobutanol. Similarly, a Swiss company, Butalco GmbH, uses a special technology to modify yeasts in order to produce butanol instead of ethanol. Yeasts as production organisms for butanol production have decisive advantages compared to bacteria because they are much more tolerant to alcohol and contaminants that may inhibit fermentation.

9.4.2.6 Future Perspectives for Bioalcohol

In the United States, crops grown for biofuels are the most land- and water intensive of the renewable energy sources. In 2005, about 12% of the nation's corn crop (covering 11 million acres (45,000 km^2) of farmland) was used to produce 4 billion gallons of ethanol, which equates to about 2% of annual US gasoline consumption. For biofuels to make a much larger contribution to the energy economy, the industry will have to accelerate the development of new feedstocks, agricultural practices, and technologies that are more land- and water-efficient. The 200-page scientific roadmap cites recent advances in biotechnology that have made cost-effective production of ethanol from cellulose, or inedible plant fiber, an attainable goal, with federal loan guarantees for new cellulosic biorefineries. The report outlines a detailed research plan for developing new technologies to transform *cellulosic ethanol*– a renewable, cleaner burning, and carbon-neutral alternative to gasoline – into an economically viable transportation fuel. The US Department of Energy (DOE) has invested in research on enzymatic, thermochemical, acid hydrolysis, hybrid hydrolysis/enzymatic, and a variety of other technologies that are aimed toward achieving success in discovering an efficient and low-cost method of converting cellulose to ethanol. Already, the efficiency of biofuels production has increased significantly, and there are new methods being developed to boost biofuel production through the use of genetic engineering of both microorganisms and the plant feedstocks themselves (see Section 9.5). Many analysts suggest that, whichever ethanol fuel-production strategy is used, conservation efforts are also needed to make a large impact on reducing fossil fuel use, and biotechnology will likely play a central role in such conservation efforts by improving our ability to generate alternative fuels while also reducing energy inputs.

9.4.3 Biodiesel

*Biodiesel*is another type of liquid biofuel, commonly produced by the *trans*-esterification of the vegetable oil or animal fat feedstocks. This biofuel can be used directly in modern diesel engines. However, it is common to use various percentages of biodiesel blended with petroleum diesel (also called *petrodiesel*) so that modifications to the diesel engines can be avoided. Much of the world uses a system known as the "B" *factor* to state the amount of biodiesel in any fuel mix. Fuel containing 20% biodiesel is labeled B20, while pure biodiesel is referred to as B100. Blends of 20% biodiesel with 80% petrodiesel (B20) are generally used in unmodified diesel engines. Biodiesel can also be used in its pure form (B100), but may require certain engine modifications to avoid maintenance and performance problems. In many European countries, a 5% biodiesel blend is widely used and is available at thousands of gas stations (Agarwal, 2007).

A variety of plant oils can be used to produce biodiesel. Currently, rapeseed or canola (*Brassica napus* L.) and soybean (*Glycine max* L.) oils are most commonly used, where soybean oil alone accounts for about 90% of all biodiesel in the United States. Other plant crops can also be used, including oil palm (*Elaeis guineensis* Jacq. and *Elaeis oleifera* Jacq.), sunflower (*Helianthus annus* L.), flax (*Linum usitatissimum* L.), mustard (*Brassica*spp.), mahua (*Madhuca longifolia*) (J. Konig, J.F. Macbr.), *Jatropha*, cotton (*Gossypium spp.)*, hemp (*Cannabis sativa* L.), field pennycress (*Thlaspi arvense* L.). Waste vegetable oil is also a useful starting material for biodiesel, as are animal fats including tallow, lard, yellow grease, chicken fat, and the by-products derived from the production of omega-3 fatty acids from fish oil. Each of these oils can, in theory, be used as fuel; however, to ensure that the fuel injectors atomize the fuel in the correct pattern for efficient combustion, vegetable oils must be heated to reduce its viscosity to that of diesel (Agarwal, 2007). This typically is done with electric coils or heat exchangers.

The *trans-esterification* step used to produce biodiesel generates a lower viscosity fuel that has combustion properties very similar to those of petroleum diesel. Chemically, *trans*-esterified biodiesel is a mix of mono-alkyl esters of long chain fatty acids. Thus, its chemical name is fatty acid methyl (or ethyl) ester (*FAME*). There are several methods for carrying out the *trans*-esterification reaction (Lachenmaier-Koelch and Meyer-Pittroff, 2005). These include the common batch process, supercritical processes, ultrasonic methods, and even microwave methods. In the most commonly used method, oils are mixed with sodium hydroxide and methanol (or ethanol), and the resulting *trans*-esterification reaction produces biodiesel and glycerol. Methanol (converted to sodium methoxide in the reaction) is normally used to produce methyl esters, as it is the cheapest alcohol available. However, ethanol can be used to produce ethyl ester biodiesel, and higher alcohols such as isopropanol and butanol have also been used. In addition, one part glycerol is produced for every 10 parts biodiesel, and this by-product can be used as a starting material for other processes. The glycerol by-product can be used as a humectant (hygroscopic moistening agent), solvent, sweetener, and food preservative and as a starting material in the production of nitroglycerin.

Biodiesel can also be used as a heating fuel in domestic and commercial boilers, where it is sometimes known as *bioheat*. In countries such as the United States, where more than 80% of commercial trucks and city buses run on diesel, biodiesel offers a promising alternative to petroleum-derived diesel. Since the feedstocks contain very little sulfur, biodiesel is a cleaner burning fuel than petrodiesel. Likewise, the solvent characteristics of biodiesel tend to keep engine deposits from forming, thus maintaining cleaner operation.

9.4.3.1 History of Biodiesel vs. Petrodiesel Production

Trans-esterification of a vegetable oil was conducted as early as 1853 by scientists E. Duffy and J. Patrick, many years before the first diesel engine became functional. It was not until 40 years later when Rudolf Diesel's invention, a single 10 ft (3 m) iron cylinder with a flywheel at its base, ran on its own power for the first time in Augsburg, Germany, on August 10, 1893. A few years later, Mr. Diesel also demonstrated his diesel engine. This engine was engineered to run on peanut oil (at the request of the French government) and built by the French Otto Company. It was shown at the World Fair in Paris, France, in 1900, where it received the Grand Prix (highest prize). This engine stood as an example of Diesel's vision because it was powered by peanut oil – a biofuel. While peanut oil is not *trans*-esterified to a true diesel fuel, Rudolf Diesel believed that the utilization of biomass fuel was the real future of his engine. In a 1912 speech, Diesel said, "the use of vegetable oils for engine fuels may seem insignificant today but such oils may become, in the course of time, as important as petroleum and the coal-tar products of the present time."

During the 1920s, diesel engine manufacturers altered their engines to utilize the lower viscosity of petrodiesel, a fossil fuel, rather than vegetable oil, a biomass fuel. The petroleum industries were able to penetrate the fuel markets because their fuel was much cheaper to produce than the biomass alternatives at the time. The result, for many years, was a near elimination of the biomass fuel production. Only recently, environmental impact concerns and decreasing price differences have made vegetable oils an appealing alternative. Despite the widespread use of fossil petroleum-derived diesel fuels, interest in vegetable oils as fuels in internal combustion engines is reported in several countries during the 1920s and 1930s.

Later, during World War II, Belgium, France, Italy, the United Kingdom, Portugal, Germany, Brazil, Argentina, Japan, and China have been reported to have tested and used vegetable oils as fuels. As mentioned above, operational problems were reported due to the high viscosity of vegetable oils as compared to petroleum diesel fuel, which resulted in poor atomization of the fuel in the fuel spray and often leads to deposits and coking of the injectors, combustion chamber, and valves. Attempts to overcome these problems included heating of the vegetable oil, blending it with petroleum-derived diesel fuel or ethanol, pyrolysis, and catalytic cracking of the oils (Agarwal, 2007).

On August 31, 1937, G. Chavanne of the University of Brussels in Belgium was granted a patent for a "Procedure for the transformation of vegetable oils for their uses as fuels" (fr. 'Procédé de Transformation d'Huiles Végétales en Vue de Leur Utilisation comme Carburants' Belgian Patent 422,877). This patent described the *trans*-esterification of vegetable oils using methanol and ethanol in order to separate the fatty acids from the glycerol by replacing the glycerol by short linear chain alcohols. This appears to be the first account of the production of what is known as "biodiesel" today. More recently, in 1977, Brazilian scientist Expedito Parente produced biodiesel using *trans*-esterification with ethanol and filed a patent for the same process. Research into the use of *trans*-esterified sunflower oil, and refining it to low viscosity diesel fuel standards, was initiated in South Africa in 1979. By 1983, the process for producing fuel-quality, engine-tested biodiesel was completed and published internationally.

Since then, the benefits of the technology have been spreading. An Austrian company, Gaskoks, obtained the technology from the South African Agricultural Engineers. This company erected the first biodiesel pilot plant in November 1987 and the first industrial-scale plant in April 1989 (with a capacity of 30,000 t of rapeseed/year). Throughout the 1990s, biodiesel plants were opened in many European countries, including the Czech Republic, Germany, and Sweden. France launched local production of biodiesel fuel (referred to as *diester*) derived from rapeseed oil, which is mixed into regular diesel fuel at a level of 5%, and into the diesel fuel used by some public transportation at a level of 30% (Agarwal, 2007). During the same period, nations in other parts of the world also saw local production of biodiesel starting up, and by 2000, over 21 countries had commercial biodiesel projects.

In September 2005 Minnesota became the first US state to mandate that all diesel fuel sold in the state contain part biodiesel, requiring a content of at least 2% biodiesel. The world's first biofuel-powered commercial aircraft took off from London's Heathrow Airport on February 24, 2008, and touched down in Amsterdam on a demonstration flight, hailed as a first step toward "cleaner" flying. The "BioJet" fuel for this flight was produced by Seattle-based Imperium Renewables, Inc.

In summary, Biodiesel is a clean burning fuel for diesel engines made from domestically produced, renewable fats and oils such as soybean oil. Biodiesel has no sulfur or aromatic compounds and already meets the new Environmental Protection Agency (EPA) ultra-low sulfur diesel fuel mandated for introduction in 2006. Biodiesel can be used in existing diesel engines without modification. Biodiesel burns substantially cleaner than petroleum-based diesel fuel. It is a powerful option for improving our environment while reducing dependence on foreign oil, stretching our fossil fuel reserves, and providing value-added markets for agricultural products.

9.4.3.2 Sources of Plant Oils

European production of biodiesel from energy crops has grown steadily in the last decade, principally focused on rapeseed used for oil and energy. In North America rapeseed was renamed *Canada Oil* or *Canola*. Production of oil/biodiesel from rapeseed covers more than 1.2 million ha in Germany alone and has doubled in the past 15 years. Typical yield of oil as pure biodiesel may be as much as 1,000 l/ha or more. This makes biodiesel crops economically attractive. They also provide sustainable crop rotations that are nutrient balanced and preventative of the spread of disease.

Soybeans are by far the main source of vegetable oil production in the United States and likewise biodiesel production. While US biodiesel is being produced from a diverse array of feedstocks, soybean oil is still used for up to 80% of US biodiesel production. Based on US Bioenergy program requirements, the Renewable fuel Standards (RFS) for biomass-based diesel is 500 million gallons in 2009 and ramps up to 1 billion gallons by 2012. Some experts estimate that if the biodiesel industry keeps its current momentum, over 10% of US soybean oil could be used for biodiesel production in the next few years. Biodiesel yield of soybeans is significantly lower than that of rape, as can be seen in Table 9.2.

Since none of the current crop plants produce enough oil to completely replace fossil fuel usage, there is ongoing research into finding more suitable crops and improving oil yields. For example, it is estimated that it would require twice the land area of the United States to be devoted to soybean production, or two-thirds to be devoted to rapeseed production, to meet current US heating and transportation needs. The use of alternative crops that do not make use of prime cropland may be one way to curb problems associated with crops that overlap with food demands.

Table 9.2 Amount (%) of oil in various crop plants

Crop plant	Scientific name	% Extractable oil
Copra	*Cocos nucifera*	62
Castor bean	*Ricinus communis*	50
Sesame	*Sesamum indicum*	50
Groundnut kernel	*Arachis hypogaea*	42
Jatropha	*Jatropha*spp.	40
Rapeseed	*Brassica napus*	37
Palm kernel	*Elaeis*spp.	36
Mustard seed	*Brassica*spp.	35
Sunflower	*Helianthus communis*	32
Palm fruit	*Elaeis*spp.	20
Soybean	*Glycine max*	14
Cotton seed	*Gossypium hirsutum*	13

Non-food crops, such as mustard, *Camelina*, and *Jatropha*, are used for biodiesel and can thrive on marginal agricultural land where many trees and crops will not grow, or would produce only slow growth yields. Specially bred mustard varieties can produce reasonably high oil yields and are very useful in crop rotations with cereals. Mustards have the added benefit that the meal leftover after oil has been extracted can act as an effective and biodegradable pesticide. Camelina (*Camelina sativa* L. Crantz) is virtually 100% efficient. It can be harvested and crushed for oil and the remaining parts can be used to produce high-quality omega-3-rich animal feed, fiberboard, and glycerin. Most camelina is grown in areas that were previously not utilized for farming. For example, camelina can be grown in areas that receive limited rainfall that cannot sustain corn or soybeans without the addition of irrigation.

Jatropha is a genus of approximately 175 succulent plants, shrubs, and trees (some are deciduous, like *Jatropha curcas L.*) from the spurge family, Euphorbiaceae. The name is derived from (Greek iatros = physician and trophe = nutrition), hence the common name physic nut. These plants are drought resistant and can share space with other cash crops such as coffee, sugar, fruits, and vegetables. It is well suited to semi-arid lands and can contribute to slowdown desertification, according to its advocates. The hardy *Jatropha* produces seeds containing up to 40% oil. When the seeds are crushed and processed, the resulting oil can be used in standard diesel engines, while the residue can also be processed into biomass to power electricity plants (Agarwal, 2007). However, estimates of *Jatropha* biodiesel yield vary, primarily due to a lack of research data, genetic diversity of different species, the range of environments in which the plants are grown, and *Jatropha's* perennial life cycle. Seed yields under cultivation can range from 1,500 to 2,000 kg/ha, corresponding to extractable oil yields of 540–680 l/ha (58–73 US gallons/acre).

Jatropha is native to Central America and has become naturalized in many tropical and subtropical areas, including India, Africa, and North America. Originating in the Caribbean, *Jatropha* was disseminated as a valuable hedge plant to Africa and Asia by Portuguese traders. Cultivation and fruit picking by hand is labor intensive and needs ca. 1 person/ha. So, in parts of rural India and Africa, this provides much-needed jobs. About 200,000 people worldwide now find employment through the production of *Jatropha*. Moreover, villagers often find that they can grow other crops in the shade of Jatropha trees. Currently, the oil from *Jatropha curcas* seeds is used to make biodiesel in the Philippines, as promoted by a law authored by Philippine senators Miriam Defensor-Santiago and Miguel Zubiri. Likewise, *Jatropha* oil is being promoted as a biofuel crop in hundreds of projects throughout India and other developing countries. One good example of its ability to grow on marginal lands is its use along the railway lines between Mumbai and Delhi in India, where the train itself runs on 15–20% biodiesel. In Africa, cultivation of *Jatropha* is also being promoted and it is grown successfully in countries such as Mali.

Since food crops are not efficient sources for oil-based fuels, often having less oil content and requiring more input energy for growth, energy crops that can be grown on marginal lands may have higher oil content and thus be a much better choice.

9.4.3.3 Advantages and Disadvantages of Biodiesel

One of the primary advantages of biodiesel comes from feedstocks used to create it. Fossil fuels, including petrodiesel, contain minor contaminants, such as salts and sulfur compounds that end up in the refined diesel. When the fuel is burned, these compounds build up in the atmosphere, where they have been causing environmental problems for years. Since the feedstocks used to make biodiesel contain virtually no sulfur, biodiesel is a cleaner-burning fuel than petrodiesel. Pure biodiesel (B100) is by far the lowest emission diesel fuel, and it is often used as an additive to ultra-low sulfur diesel (ULSD) fuel (Agarwal, 2007). However, while B100 biodiesel has a viscosity similar to petrodiesel, it may become more viscous at lower temperatures, depending on the feedstock used, thus requiring vehicles to have fuel line heaters.

Biodiesel is also an oxygenated fuel, meaning that it contains a reduced amount of carbon and higher hydrogen and oxygen contents than fossil diesel. This improves the combustion of fossil diesel and reduces the particulate emissions from unburnt carbon (Agarwal, 2007; Armas et al., 2008). Similarly, biodiesel has better lubricating properties than other diesel fuels. Thus, the addition of biodiesel to various blends of petrodiesel fuels reduces engine wear, increases the life of the fuel injection systems, and effectively cleans the engine combustion chamber of carbon deposits, helping to maintain efficiency.

On the other hand, while biofuels are generally considered to improve emissions and engine efficiency, biodiesel still produces local air pollution, including nitrogen oxides, the principal cause of smog. Since biodiesel is an effective solvent and cleans residues deposited by fossil diesel, engine filters may need to be replaced more often, as the biofuel dissolves old deposits in the fuel tank and pipes. Likewise, while older furnaces can burn biodiesel without any required conversion, the biodiesel may cause problems because rubber parts are adversely affected by the solvent properties of this fuel.

Perhaps the most profound problem with biodiesel is that worldwide production of vegetable oil and animal fat is not yet of sufficient magnitude to replace liquid fossil fuel use. As described in Section 9.3, some people object to the vast amount of farming required for such crop-based biofuels and the resulting fertilization, pesticide use, and land use conversion that would be needed to produce the additional vegetable oil. Transitioning fully to biodiesel would require immense tracts of land if traditional food crops (such as rapeseed (canola) or soybean) are used. The problem would be especially severe for nations with large economies, where energy consumption is proportional to economic output. If using only traditional food plants, most of such nations do not have sufficient arable land to produce biofuel for the nation's vehicles. Nations with smaller economies (hence, less energy consumption) and more arable land may be in better situations. However, many regions cannot afford to divert agricultural land away from food production.

In some regions of the world, a combination of increasing demand for food, and increasing demand for biofuel, is causing deforestation and threats to biodiversity. The best reported example of this is the expansion of oil palm plantations in Malaysia and Indonesia, where rainforest is being destroyed at alarming rates to establish new oil palm plantations to keep up with growing biodiesel demand in Europe and other markets. It is an important fact that 90% of the palm oil produced in Malaysia is also used by the food industry in a wide variety of food products; therefore, biofuels cannot be held solely responsible for this deforestation. Palm oil is also used in the manufacture of detergents and in electricity and heat generation both in Asia and around the world. So, there is a pressing need for sustainable palm oil production for both the food and fuel industries. Fortunately, many organizations, such as the Roundtable on Sustainable Biofuels, are working to define criteria, standards, and processes to promote sustainably produced biofuels.

Many biodiesel advocates suggest that waste vegetable oil and animal fats are the best sources of oil to produce biodiesel, but since the available supply of these oils is drastically less than the amount of petroleum-based fuel that is burned for transportation and home heating in the world, this local solution can only account for a very small percentage of petrodiesel usage. It is likely that biodiesel sources that make use of marginal lands (where food crops cannot be grown) would make much more sense as a solution to land use issues (e.g., palm oil nuts grown along roads or *Jatropha*grown along rail lines, see Section 9.4.3.2).

9.4.4 Biogas

Unlike natural gas derived from fossil fuels, *biogas* is a renewable "natural gas" that is produced from organic/biological materials as they decay. There are two primary types of biogas. The most common biogas is produced by anaerobic digestion or fermentation of organic/biological materials such as manure or sewage, municipal waste, and energy crops through the action of anaerobic bacteria. The resulting biogas is comprised primarily of methane (also called *biomethane*) and carbon dioxide.

Methane bacteria are responsible for such biological sources of methane, including symbiotic relationships within other life forms such as termites, ruminants, and cultivated crops. Another type of biogas is *wood gas* (also called synthesis gas), created by the gasification of wood, wood chips, or other carbon-rich biomass. This type of biogas requires a gasifier or wood gas generator. This biogas is comprised primarily of nitrogen, hydrogen, and carbon monoxide, with trace amounts of methane. These gasses can be combusted with oxygen present in the atmosphere to release energy, thus allowing wood gas to be used as a fuel.

Both types of biogas can be utilized for cooking, space heating, and water heating. They can also be utilized in modern waste management facilities to run heat engines that generate either mechanical or electrical power (Fig. 9.5). If compressed, they can replace compressed natural gas for use in vehicles, where they can fuel internal combustion engines or fuel cells. Biogas can be produced easily and cost-effectively from current waste streams, such as paper production, sugar production, sewage, and animal waste. It is most commonly produced using agricultural waste, such as plants and manure. The gas can also be produced by separating organic materials from waste that otherwise goes to landfills. Using materials that would otherwise generate no income, or even cost money to dispose of, improves the profitability and energy balance of biogas production. Similarly, the solid by-product or digestate derived from the biogas process can typically be used as a biofuel or

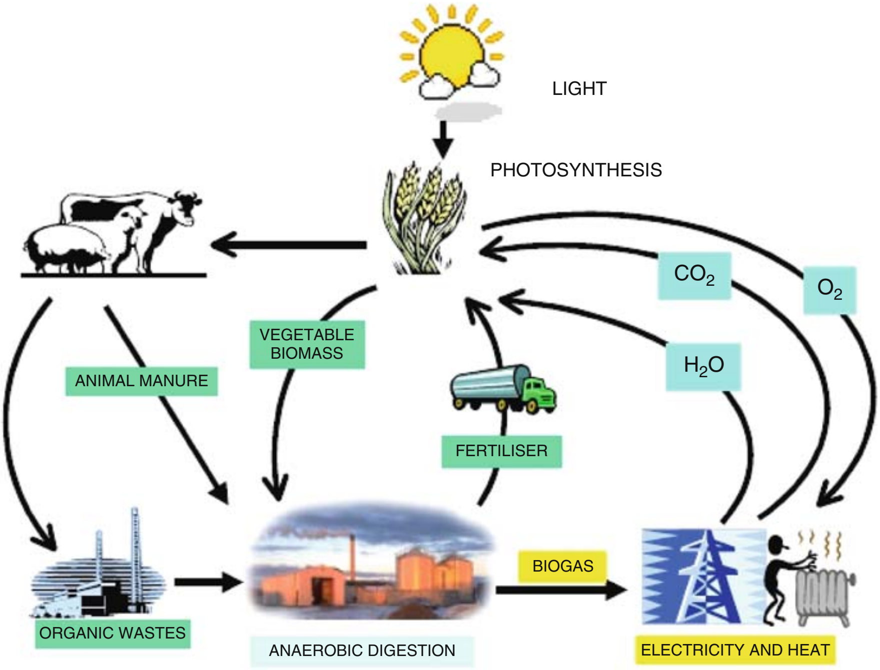


Fig. 9.5 A schematic of biogas formation. From www.makinemekanik.com

natural fertilizer. It is important to point out; however, that both carbon monoxide and methane are potent greenhouse gasses. Methane in particular is 21 times more reactive as a greenhouse gas than carbon dioxide (CO_2), and it is normally released into the atmosphere at most waste treatment facilities and landfills. Modern methods of biogas production have the advantage of keeping such gasses contained, avoiding potential environmental issues. Thus, biogas is considered to be one of the most climate-friendly sources of fuel.

9.4.4.1 History of the Use of Biogas

Some types of biogas, such as that derived from manure, have been used as a low-cost fuel for heating and cooking for hundreds of years. Anecdotal evidence indicates that biogas was used for heating bath water in Assyria during the tenth century BCE and in Persia during the sixteenth century CE. Some ancient Chinese literature also suggests that biogas was generated from sewage 2,000 to 3,000 years ago. However, Jan Baptita Van Helmont, in the seventeenth century, was credited as the first to determine that flammable gases could evolve from decaying organic matter. Likewise, Count Alessandro Volta concluded in 1776 that there was a direct correlation between the amount of decaying organic matter and the amount of flammable gas produced. In 1808, Sir Humphrey Davy determined that methane was present in the gases produced during the anaerobic decomposition of cattle manure.

It was not until the mid-1800s that the first biogas digesters were built. India has a quite long history of biogas development. The first unit usually referred to in literature is a biogas unit at the Mantunga Homeless Lepers Asylum near Mumbai in 1859. However, methane was first recognized as having practical and commercial value in England, where a specially designed septic tank was first used to generate gas for the purpose of lighting in the 1890s (Cheremisinoff et al., 1980). One other important development came from the development of microbiology as a science. This led to research by Buswell and others in the 1930s to identify anaerobic methane bacteria and the conditions that promote methane production by these organisms. Once the process became fairly well understood, the stage was set for the expansion of the use of biogas around the world. Such trends continue to this day, where India remains one of the biggest investors in biogas, making use of small biogas digester units installed in millions of homes.

9.4.4.2 Biogas Derived from Biodigesters

In India, biogas produced from the anaerobic digestion of manure in small-scale digestion facilities is called *gober gas*. This biogas is predominantly composed of methane and carbon dioxide. It is generated in more than 2 million household facilities. A typical gober gas digester consists of an airtight circular pit made of concrete with a pipe connection. The manure is directed to the pit, usually directly from a cattle shed, and the pit is then filled with a required quantity of wastewater. In this milieu, anaerobic bacteria create an oxygen-free environment in which they efficiently degrade the organic material and generate gas. The gas pipe can then be connected to a kitchen fireplace through control valves, and the flammable methane gas generated out of this apparatus is largely odorless and smokeless. The residue left after the extraction of the gas is commonly used as fertilizer for crop plants. Owing to its simplicity in implementation and use of cheap raw materials in the villages, it is often quoted as one of the most environmentally sound energy sources for rural needs.

Similar biogas digesters are now used extensively in rural regions around the world, including China, Costa Rica, Nepal, and Vietnam. The Government of Pakistan provides 50% of funds needed for the construction of moveable gas chamber biogas plants. Farmers around the world are making use of such small-scale units to convert plant and animal wastes into a useful combustible gas like methane. In Colombia, experiments with diesel engine generators partially fuelled by biogas demonstrate that biogas can reduce electricity costs by 40% as compared with purchase from regional utilities.

Although based on the same principles as employed in the simple digesters above, larger municipal plants generate biogas in more advanced anaerobic digesters that recover the recyclable elements of household waste, sewage sludge, food wastes, farm wastes, or energy crops (such as maize silage) and process the biodegradable fraction in the anaerobic digesters. The methane contained in the resulting biogas can be concentrated to the same quality standards as fossil natural gas (typically called *biomethane*). If the local gas utility network grants permission, the producer of the biogas may then be able to utilize the local gas distribution networks to deliver the gas to consumers. Such biomethane, however, must be very clean and be of the correct composition to reach pipeline standards. Carbon dioxide, water, hydrogen sulfide, and particulates must therefore be removed if present. Biomethane can also be concentrated and compressed for use in vehicle transportation. Compressed biogas is becoming widely used in Sweden, Switzerland, and Germany as a renewable fuel source.

Sweden is cited as a particularly good example of a global leader in converting biowaste (largely agricultural material and residues) into usable biomethane. Facing oil shortages, waste management problems, and lacking any natural gas reserves of its own, Sweden was motivated to develop its biomethane industry under the Kyoto Accords. The resulting biogas is now used to generate electricity, residential heating, and transportation fuel. According to the Swedish Gas Association, more than 50% of the methane used to power Sweden's natural gas vehicles now comes from biological sources. More than 8,000 vehicles in Sweden are powered by a combination of natural gas and biomethane. The vehicles include transit buses, refuse trucks, and more than 10 different models of passenger cars. There are more than 25 biomethane production facilities in Sweden and 65 filling stations. A biogas-powered train has even been in service since 2005.

9.4.4.3 Biogas Derived from Landfills

Biogas is also produced in landfills from organic waste decomposing under anaerobic conditions. When a clay cap is placed atop the compacted waste materials within the landfill, this prevents oxygen from penetrating the waste. As a consequence, anaerobic microorganisms like methane bacteria thrive under such conditions. One problem with such clay-capped landfills is that the resulting biogas builds up and is slowly released into the atmosphere. This gas contains a large portion of methane, which is 21 times more potent as a greenhouse gas as carbon dioxide. Therefore, uncontained landfill gas which escapes into the atmosphere may significantly contribute to the effects of global warming. In addition, *volatile organic compounds* (*VOCs*) contained within landfill gas contribute to the formation of unhealthy photochemical smog. However, if engineered properly, landfill sites can be made to capture such gases via pipes inserted into the clay cap that deliver the gases to gas clean-up and combustion facilities.

The European Union presently has some of the strictest legislation regarding waste management at landfill sites called the *Landfill Directive*. The United States legislates against landfill gas as it contains these VOCs. The US Clean Air Act and Title 40 of the Code of Federal Regulations (CFR) require landfill owners to estimate the quantity of *non-methane organic compounds* (*NMOCs*) emitted. If the estimated NMOC emissions exceeds 50 t/year, the landfill owner is required to collect the landfill gas and treat it to remove the NMOCs, which is usually done though combustion.

The composition of biogas varies depending upon the origin of the anaerobic digestion process. There are literally thousands of different types of anaerobic bacteria living in landfills. Their differing interactions, types of available waste, and resulting products generate different amounts of usable gas. Landfill gas typically has methane concentrations around 50%; however, advanced waste treatment technologies can produce biogas with 55–75% methane. This gas can be used to heat on-site buildings or to power engines for the generation of electricity, which can, in turn, be sold back to electricity utility companies for renewable energy subsidies. However, because of the remoteness of landfill sites, it is sometimes not economically feasible to produce electricity from this biogas. Still, it is estimated that a 3 MW landfill power plant can power 1,900 homes and at the same time prevent 6,000 t/year of methane from entering the environment. This is equivalent to eliminating 18,000 t/year of CO_2 derived from fossil fuel use or removing 25,000 cars from the road, planting 36,000 acres (146 km^2) of forest, or not using 305,000 barrels (48,500 m^3) of oil/year.

9.4.4.4 Wood Gas Derived from Carbon-Rich Biomass

Wood gas (or synthesis gas) is another form of biogas produced by thermal gasification of woody biomass or other carbon-rich materials in a gasifier or wood gas generator. Usable materials include wood chips, sawdust, charcoal, coal, rubber, or similar materials which are burned incompletely in a fire box that produces wood gas, tars, solid ash, and soot. The latter three by-products have to be removed periodically from the gasifier. In this case, the wood gas is filtered from tars and soot/ash particles, then cooled and directed to an engine or fuel cell to produce mechanical or electrical power. The gas is the result of two high-temperature reactions (above

700°C or 1,292°F): an *exothermic reaction,* where carbon burns to CO_2; and an *endothermic reaction,* where carbon reacts with steam to produce carbon monoxide (CO), hydrogen (H_2), and carbon dioxide (CO_2). Wood gas is flammable mainly because of the carbon monoxide and hydrogen content, but it also contains nitrogen and small amounts of methane, which is also flammable (Ahring and Westermann, 2007).

The first wood gasifier was apparently built by Bischof in 1839, and as a result, gasification became an important and familiar nineteenth and early twentieth century technology. "Town gas," produced by centralized gasifiers, was once quite popular and used primarily for lighting purposes. By the time World War II arrived in United States, large numbers of such generators were constructed and commercial generators were in production both before and after the war. The applicability of wood gas to the internal combustion engine was well understood from its earliest days of development, and the first vehicle powered by wood gas was built by Parker in 1901. Internal combustion engines were initially fueled by town gas during the nineteenth century; however, wood and wood chips can also be used to power cars with ordinary internal combustion engines if a wood gasifier is attached. This was actually quite popular during World War II in several European and Asian countries because the war prevented easy and cost-effective access to oil. Many of these early gas generators had the problem of generating soot and tar, which would in turn clog the engines if not first removed from the gas. This problem has only recently been solved through the use of modern heat-resistant filters that can separate practically all the particles, allowing easy disposal of clean, dry ash.

Modern gasifiers, especially those used to power gas turbines or fuel cells for the production of electricity, are quite efficient. The gasification process in modern designs is usually preceded by *pyrolysis*, where the biomass or coal is initially converted to char, releasing methane (CH_4) and tar rich in polycyclic aromatic hydrocarbons (PAH). This pyrolyzed char is then fed into another gasifier to generate the gasses described above. Such staged gasifiers, where pyrolysis and gasification occur separately, can also be engineered to produce essentially tar-free gas (less than 1 $mg{\cdot}m^{-3}$), while single reactor fluid-bed gasifiers may exceed 50,000 $mg{\cdot}m^{-3}$tar.

Contrary to general belief, exhaust gas emission levels of internal combustion engines are significantly lower for wood gas than for gasoline. The efficiency rate of the gasifier system is relatively high: it converts about 75% of fuel energy content into combustible gas that can be used directly as fuel for the engine. Based on long-term studies, comparing otherwise unmodified vehicles during real transportation and under similar driving conditions, the energy consumption for wood gas has been determined to be 1.54 times greater as compared to the energy demand of the same car powered by gasoline (not including the energy needed to extract, transport, and refine the oil from which gasoline is derived).

The primary *disadvantages* of wood gas generators are their typically large size and relatively slow starting speeds. In addition, while the carbon monoxide is an intentional fuel-product that is subsequently burned to safer carbon dioxide in the engine, it is poisonous to humans, even in small to moderate concentrations. However, if the system is well maintained, the chance of exposure to CO is extremely low. Wood gas generators have several key *advantages* over other sources of fuel. They are relatively easy and inexpensive to build and can be used directly to run internal combustion engines using wood and other forms of carbon-rich biomass. This can reduce dependency on fossil natural gas, gasoline, and oil. Such generators have a closed carbon cycle. This means that the carbon released from the generator, in the form of CO_2, is absorbed by plants that via photosynthesis convert it to biomass. Gasifiers are also far cleaner burning than equivalent burning processes, such as a wood fire or even a gasoline-powered engine (without emissions controls).

In more recent times, wood gas has been suggested as a clean, cheap, and efficient method to heat and cook in developing countries or even to produce electricity when used in internal combustion engines, gas turbines, or fuel cells for maximal efficiency. Gasifiers have been built for remote Asian communities using rice hulls, which in many cases has no other use except for being utilized as a strengthener type additive in concrete. One installation in Burma uses an 80 kW modified diesel engine to supply power for about 500 people who are otherwise without power. The ash is also used as an efficient fertilizer.

So, in summary, it is clear that wood gas generators represent a promising technology based on its utilization of renewable biomass. Once improvements in this technology result in improved efficiency, their uses are expected to expand greatly.

9.4.4.5 Future Perspectives for Biogas

Whatever its source, biogas presents us with viable renewable energy opportunities for the following reasons: Anaerobic digesters allow us to sequester a potentially dangerous and environmentally unfriendly waste (biomethane) by using it as major biofuel. Of all the greenhouse gas emissions, biomethane is 21 times more harmful to the atmosphere than carbon dioxide. It is generated during natural processes of decay; so it is essentially free. Biogas is closer to carbon-neutral than any other source of fuel, making it a near perfect fuel. As biogas technologies such as anaerobic digesters and biomass gasification development increases and becomes more common, one of the fundamental questions is, what is the size of the potential biomass resource supply in the United States?

In April 2005, the US Department of Energy (DOE) and the US Department of Agriculture (USDA) co-published a report assessing the potential of the land resources in the United States for producing sustainable biomass entitled, “Biomass as Feedstock for a Bioenergy and Bioproducts Industry: The Technical Feasibility of a Billion-Ton Annual Supply.” Looking at forest land and agricultural land, the two largest potential biomass sources, this study estimated that the United States can sustainably produce up to 1.3 billion tons of biomass feedstock by mid-century (2050). This would be enough feedstock to produce 60 billion gallons of B100 biodiesel and E100 ethanol with existing technologies. This is an impressive number. However, the study does not address the opportunities for biogas production from biomass feedstock or biomass gasification technologies. Some recent estimates indicate that biogas could replace up to 50% of present natural gas consumption in the United States. In some countries, such as Iceland, biogas already provides 100% of the natural gas resources. Sweden currently obtains 51% of its methane from biogas, and Switzerland, 37%. Countries such as France, Norway, Germany, and Austria use smaller amounts for vehicles. China, India, Korea, the Ukraine, Spain, and Italy are other examples of countries now initiating projects where biogas will be used as a vehicle fuel.

When viewed in a broader perspective, biogas has an outstanding potential to help solve current environmental problems, including urban and agricultural waste management, water purification, and the need for cleaner air. By converting biomass waste, such as municipal solid waste, sewage sludge, crop residues, energy crops, and manure, into biogas, governments can address energy and environmental problems in a sustainable manner (Ahring and Westermann, 2007). However, most governments, including the United States, are perhaps too focused on liquid fuels to support the development of a biogas infrastructure. It is very unlikely that any one technology will come close to solving global energy needs. A combination of all technologies will therefore be required to address such problems, and the development of new technologies will clearly play an important role in this approach.

9.5 Future Technologies in Biofuels: Algae for Energy

The idea of using algae as a source of fuel is not new, but it is now becoming a promising technology because of the escalating price of petroleum and, more significantly, the emerging concern that burning fossil fuels is contributing to global warming (Chisti, 2007; Briggs, 2004). Like plant tissues, algae can provide several different types of renewable biofuels. These include methane produced by anaerobic digestion of the algal biomass; bioethanol fermented from algal cell walls; biodiesel derived from algal oil; and photobiologically produced biohydrogen. Unlike other plant energy crops, algae species can grow extremely rapidly and many are exceedingly rich in oil. Microalgae (single-celled algae) commonly double their biomass within 24 h, and biomass doubling times during exponential growth are commonly as short as 3.5 h. Thus, algae can develop huge amounts of biomass usable as starting material for both bioethanol and biogas production. An added benefit here is that algae do not produce lignin (Table 9.1); however, the amount of cellulose and hemicellulose is relatively low compared to other plants.

It is not yet feasible to collect algal biomass from natural sources. However, the US Department of Energy's Aquatic Species Program has experimented with "raceway ponds" for the cultivation of algae. These artificial, shallow ponds are divided into a rectangular grid, with each rectangle containing one channel in the shape of an oval, like an automotive raceway circuit. Each rectangle contains a paddle wheel to make the water flow continuously around the circuit. Under such conditions, nutrients can be delivered to algae crops, and biomass doubling rates can be optimized.

Another promising technology for algae growth involves photobioreactors. Unlike open raceways, photobioreactors permit essentially single-species culture of algae for prolonged durations, and they have been successfully used for producing large quantities of microalgal biomass. A tubular photobioreactor consists of an array of straight transparent tubes that are usually made of plastic or glass. This tubular array, or the solar collector, is where algal broth is circulated from a reservoir and where sunlight is captured. Such reactors can be constructed in areas that are not suitable for plant growth (such as deserts), and they hold great promise for the large-scale production of algae oil and biohydrogen.

9.5.1 Biodiesel and Biopetroleum from Algae

Oil content in some algae species can exceed 80% by weight of dry biomass. Since many common algae (e.g., *Chlorella vulgaris*) have a natural oil content greater than 50%, they have a high potential to be low-input, high-yield feedstocks useable for biofuel production (Chisti, 2007). Oil-based algae fuel, also called *oilgae* or sometimes third generation biofuel, is a biofuel derived from oil content of algal biomass. A self-published article by Michael Briggs, at the UNH Biodiesel Group, offers estimates for the realistic replacement of all vehicular fuel with biodiesel by utilizing algae, which Briggs suggests can be grown on algae ponds at wastewater and sewage treatment plants. These oil-rich algae can then be extracted from the system and processed into biodiesel, with the dried remainder further reprocessed to create ethanol.

Algae fuel yields have not yet been accurately determined; however, the US Department of Energy is reported as saying that algae yield 30 times more energy per acre than land crops such as soybeans. The DOE estimates that if algae fuel replaced all of the petroleum fuel in the United States, it would require only 15,000 square miles (38,849 square km), which is roughly the size of Maryland. These estimates are very promising; however, algal oil is difficult to extract, and more research needs to focus on the development of efficient extraction protocols.

Many companies are pursuing algae bioreactors for various purposes, including high-yield oil production that can scale biodiesel production up to commercial levels. Alternative fuel companies such as Solazyme (South San Francisco, CA) and Solix Biofuels (Fort Collins, CO) are using algae to produce biodiesel (Fig. 9.6). While the cultivation of algae to harvest oil for biodiesel has not yet been undertaken on a commercial scale, one of the most appealing aspects of alga-culture is that – unlike crop-based biofuels – it does not entail a decrease in food production, since it requires neither farmland nor fresh water. Unfortunately, like ethanol, biodiesel (including that extracted from algae) attracts water and thus cannot be shipped in existing pipelines. Both ethanol and biodiesel also have lower energy density than traditional gasoline and diesel fuels.

Some companies, such as Sapphire Energy (formally launched in May of 2007) have developed molecular platforms that converts sunlight and CO_2 into renewable, carbon-neutral alternatives to conventional fossil fuels without the numerous downsides of current biofuel efforts. The end product is not ethanol – and not biodiesel. The end product is “green crude,” which is chemically identical to molecules in

Fig. 9.6 Colorado's Solix Biofuels tackles the difficult task of harvesting algae for oil with a field of bioreactors that take a kind of painter's drop cloth (inset) to bubble CO through its system. From Popular Mechanics online article "Pond-Powered Biofuels: Turning Algae into America's New Energy" 2007 (http://www.popularmechanics.com/science/earth/4213775.html)

crude oil, making the products entirely compatible with the current energy infrastructure. Such green crude is said to have the same energy density as gasoline and can be shipped in existing pipelines and refined the same way gasoline and diesel are. Such technologies are at the forefront of an entirely new industry with the potential to profoundly change America's energy and petrochemical landscape. However, the use of such technology will be dependent on the enactment of government policies that pressure oil companies to turn their attention away from foreign oil.

9.5.2 Biohydrogen

Hydrogen is regarded as the fuel of the future, being easy to handle and extremely clean burning. The efficiency of conversion of hydrogen to useable energy is especially high in fuel cells for the production of electricity, with water being the sole end product. Presently, hydrogen is produced from fossil reserves with the concomitant release of anthropogenic carbon dioxide. Therefore, new hydrogen production technologies, making use of renewable resources such as the production of biohydrogen from biomass through the action of microorganisms, are being pursued.

In 1939, a German researcher named Hans Gaffron, while working at the University of Chicago, observed that the algae he was studying, *Chlamydomonas reinhardtii* (a green alga), would sometimes switch from the normal production of oxygen to the production of hydrogen. Gaffron never discovered the cause for this change and for many years other scientists failed in their attempts at its discovery. However, in the late 1990s professor Anastasios Melis, a researcher at the University of California at Berkeley, discovered that if the algae culture medium is deprived of sulfur it will switch from the production of oxygen (normal photosynthesis) to the production of hydrogen. He found that the enzyme responsible for this reaction is hydrogenase, but that the hydrogenase is not functional in the presence of oxygen.

As a result of these findings, photobioreactors, harboring various algae species, are currently being used for biological hydrogen production (Rupprecht et al., 2006). Initially, the yields were not very high; however in 2006, researchers from the University of Bielefeld and the University of Queensland used genetic modifications to changed *C. reinhardtii* in such a way that it produces an especially large amount of hydrogen, producing five times the volume made by the wild form of the algae with up to 2.0% energy efficiency. Later in 2007 while studying mutants of *C. reinhardtii*, Anastasios Melis achieved 15% energy efficiency, demonstrating that truncated chlorophyll antenna size would minimize wasteful dissipation of sunlight by individual cells. Such results hold great promise for alternative fuels, but other areas of research need to be address, including the development of oxygen-tolerant hydrogenases and increasing hydrogen production rates through improved electron transfer.

Biohydrogen can and is also produced in bioreactors that utilize feedstocks other than algae, the most common feedstock being waste streams. The hydrogen is produced by bacterial species such as *Rhodobacter sphaeroides* and *Enterobacter cloacae* through dark fermentation either by mixed cultures of hydrogen-producing sludge or pure cultures of anaerobic bacteria, such as *Clostridium butyricum*. This process involves allowing the bacteria to feed on hydrocarbons under anoxic conditions so that they release hydrogen and CO_2. Under natural conditions, this hydrogen is usually consumed as soon as it is being produced by methanogenic bacteria, releasing methane as the end product. However, under bioreactor conditions, the CO_2can be sequestered successfully by several methods, and the methanogens can be omitted from the system, allowing hydrogen gas to be captured. A prototype hydrogen bioreactor using waste as a feedstock is in operation at Welch's grape juice factory in North East, Pennsylvania.

By uncoupling methane production from hydrogen production, more hydrogen can be harvested. In such systems, hydrogen and acetic acid are first produced from biomass by thermophilic bacteria. Then in a second stage, the acetic acid in the effluent is converted to hydrogen by purple non-sulfur bacteria (Zheng et al., 2008). Biohydrogen can also be produced from glucose in upflow biofilm reactors with plastic carriers under extreme thermophilic conditions (>70°C). Biological hydrogen production derived from these systems is also promising but needs to address the following issues: (1) conversion of biomass from an energy crop or an organic waste stream to fermentable feedstock; (2) selection of thermophilic and/or photoheterotrophic microorganisms and design of optimal growth and production conditions; (3) design of an integrated bioprocess; and (4) development of recovery and purification methods for upgrading the gas to fuel cell specifications.

9.6 How Can Plant Biotechnology Contribute to Bioenergy?

Technology tends to change in cycles that last approximately 50 years. The Russian economist Nikolai Kondratiev (1892–1938) was the first to bring this observation international attention in his book *The Major Economic Cycles* (1925), and others have expanded upon his observations (Fig. 9.7). Starting with the industrial age, when the expansion of steam power changed the world, this source of power (fueled by biomass) led to the era of the railroads. Railroads in turn promoted the development of petrochemicals (including coal and oil), which gave ample energy required to develop information technology. Currently, the information age is in full swing, and without this technology, modern biology and biotechnology would not be possible. Computers play an essential role in the generation and analysis of the gene sequences used in most modern biotechnological applications. With the invention of new processes for large-scale gene sequencing (e.g., *pyrosequencing* approaches) and even faster computer technology, it appears that we have only scratched the surface of the potential held within a new biological age. This new age will likely be driven by the human need for food and energy, and plant biotechnology will play an essential role in meeting these human needs while finding ways to address land and water usage issues and potential environmental problems including global carbon balance.

The use of biotechnology for improving plant-derived materials for bioenergy production can be achieved at different levels. At the front end of potential improvements are improvements to the plant feedstocks themselves. High-energy yield can

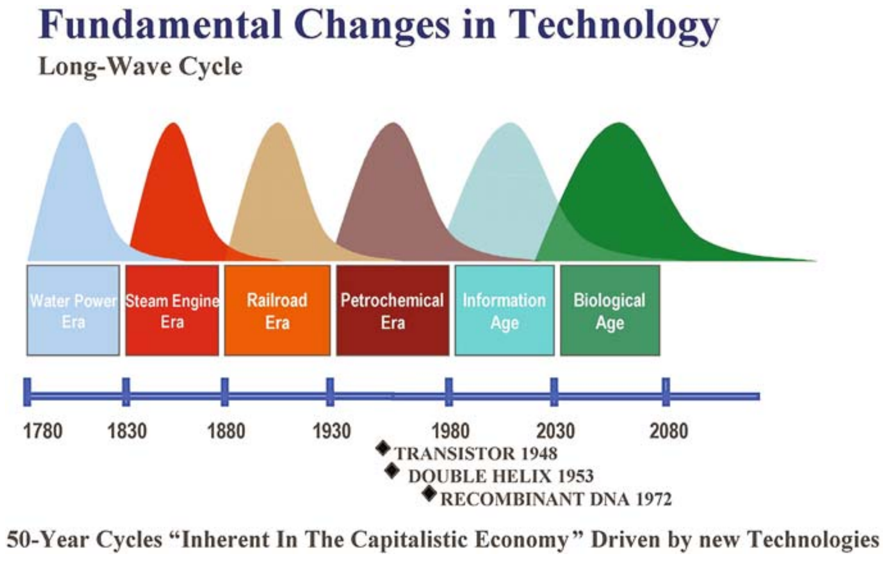


Fig. 9.7 Progressive cycles of technology: 1780–2080 CE based on the work of Nikolai Dimitriyevitch Kontratyev (CE refers to "common era" that replaces "AD")

be realized through plant breeding programs designed to identify genetic markers for beneficial traits and to enhance these traits in existing energy crops, especially sugarcane, sorghum, switchgrass, and non-traditional oil crops having especially high levels of oil (e.g., *Camelina*, mustard, and *Jatropha*). Genetic modification of biosynthetic pathways (metabolic engineering) and the application of biotechnology will likely play the biggest role because of the promise of significant and rapid improvements. Plants will very likely be manipulated to create greater yields of oils and biomass; show greater resistance to abiotic and biotic stresses; require less water and grow in harsher environments; and reduce associated costs of growth and processing. Some such modifications have already been accomplished, such as the reduction of lignin biosynthesis, allowing easier extraction of cellulose and hemicellulose polymers in preparation for processing (see Section 9.4.2.4) or altering plant oil profiles so that specific types of plant oils are produced.

Improvements in bioenergy can also be achieved at the level of processing plant materials. This can include (1) improvements to extraction and pretreatment stages of plant oil and ethanol production; (2) improvements to chemical and enzymatic processes used to extract stored energy (including mass production of tailor-made cellulase enzymes that are more efficient or by genetically engineering plants and fungi to produce desired cellulase, xylanase, and/or other hydrolase enzymes that can help degrade plant polymers); or (3) improvements to the efficiency of collecting and transporting the plant oil tissues or cellulosic biomass to the biofuel refinery. Efforts are also being directed toward improving the efficiency of the biosynthetic pathways of the microorganisms (yeast and bacteria) that perform ethanol fermentation or ABE fermentation (see Section 9.4.2.4), as well as methanogen processes that generate biogas.

Improvements can also be made at the tail end of bioenergy production. This may include improvements to distillation efficiency by engineering microorganisms to be more resistant to bioalcohol (allowing higher concentrations of ethanol or butanol) or by simply improving the ability of the biofuels to be used in existing networks of pipes for delivery to the consumer (see Section 9.5.1). The point here is that there is room for biological improvement at virtually every stage of bioenergy production. Almost all of these improvements, however, are dependent on an in-depth understanding of the biological processes and biosynthetic pathways that control each of these stages.

Sorghum (*Sorghum spp.*), which is related to corn and sugarcane, provides a good example because it has a number of characteristics that make it an attractive biomass crop for ethanol production. Sorghum requires little water and little fertilizer compared to traditional bioenergy crops. It is tolerant to heat and drought, has high biomass yield, and bioalcohol can be produced from sugars stored in the stems, starch from the grain as well as the lignocellulose from the leftover stalks. Two traits of particular interest are the “sweet sorghum trait,” which results in the accumulation of fermentable sugars in the juice of the stems (similar to sugarcane), and the “brown midrib trait,” which changes the chemical composition (and hence color) of the vascular tissue resulting in higher yields of fermentable sugars obtained after enzymatic processing of the lignocellulosic biomass. The genetic basis of these traits, however, is poorly understood and impedes the full exploitation of sorghum for bioenergy production.

Consequently, research is being directed toward identification of regions of the genome (through quantitative trait loci [QTL] and genomics) that contain genes associated with the sweet sorghum trait and other traits of interest for bioenergy production. In addition, identification of the genes that are responsible for the accumulation of sugars in the stem (through genetic mapping, cloning, and elucidation of biochemical functions) will aid in the understanding of the brown midrib trait. Without an understanding of the biochemical and molecular mechanisms behind such traits, improvements to subsequent ethanol production are limited.

Perhaps the most major improvements can be made at the front end of bioenergy production, i.e., improvement of cellulosic biomass/cell wall production in herbaceous grasses such as sorghum, sugarcane, and switchgrass. Likewise, improvements to woody biomass may include the manipulation of wood productivity and carbon sequestration as well as improved cellulose biosynthesis in poplar trees (*Populus* species) for use as energy crops (Joshi and Mansfield, 2007). Attention to front end production is especially important because it can benefit multiple types of biofuels at the same time, including the production of solid biomass, bioalcohol and biogas.

There are a number of traits that are especially beneficial for the biological improvement of bioenergy. An ideal energy crop plant would have a combination of (1) a high yield of biomass resulting from a high rate of growth and high photosynthetic capacity; (2) a higher fuel to mass ratio resulting from efficient nutrient uptake from the soil and high levels of stored sugars or oils; (3) reduced need for soil nutrients and soil moisture resulting from the growth of deep roots; (4) resistance to pests, disease, and the extremes of cold and hot temperatures; (5) tolerance to salt, high and low pH, and heavy metal contaminants in the soil; (6) a perennial grow habit to allow collection of biomass each year without fresh planting; and (7) the silencing of flowering to help avoid the spread of foreign genes to native populations (Fig. 9.8). Many of these traits have already been studied in model plant species such as *Arabidopsis*, rice, soybean and poplar trees, and some of the genes that are orchestrating molecular events behind such traits have been identified (Fig. 9.9). Therefore, the stage is already set for the use of plant biotechnology to genetically engineer plants.

While genetic modification of algae has received little attention until just recently, metabolic engineering will also likely have an impact on improving the economics of biomass, oil production, and biohydrogen formation from algae species. Molecular level engineering can be used to potentially (1) increase the photosynthetic efficiency to enable increased biomass yield and rate of production; (2) improve temperature tolerance to reduce the expense of cooling within photobioreactors; (3) eliminate the light saturation phenomenon so that growth continues to increase in response to increasing light level; (4) reduce photoinhibition that actually reduces growth rate at midday light intensities; and (5) reduce the susceptibility to photooxidation that can damage the cells; and (6) increase oil content in biomass.

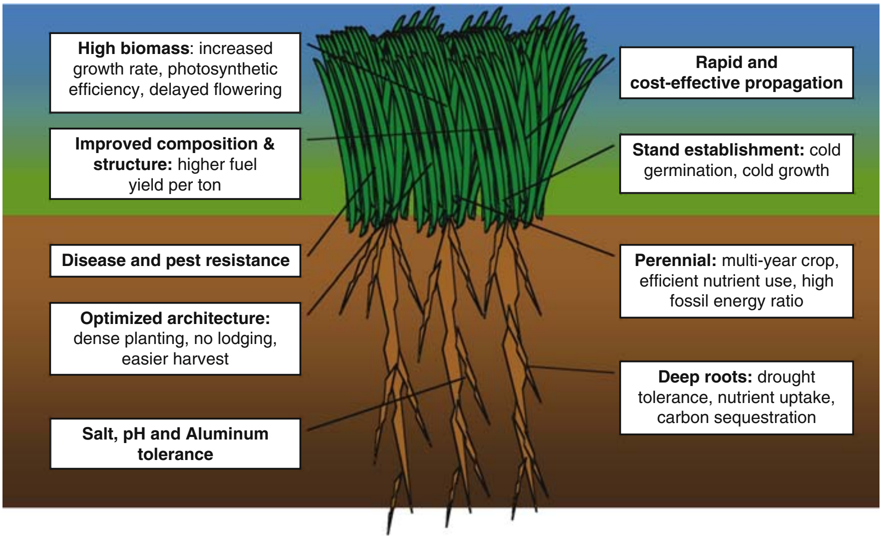


Fig. 9.8 Traits of the perfect energy crop. Modified from Ceres Technologies "The Promise of Dedicated Energy Crops" 2008

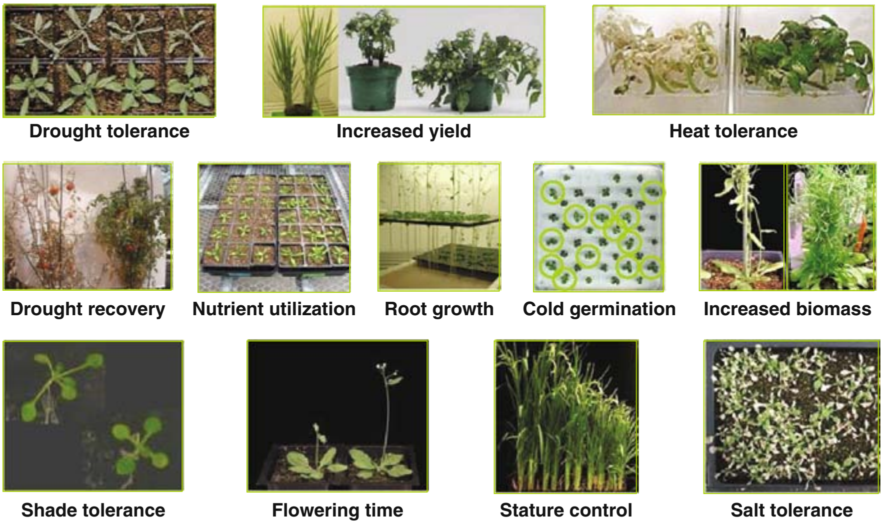


Fig. 9.9 Traits for which genes have been identified that offer improvements in biofuel production. Modified from Ceres Technologies "The Promise of Dedicated Energy Crops" 2008

As far as plant oil is concerned, development of knowledge-based approaches to generate high-yielding oil crop species is key to manipulating oil biosynthesis through plant biotechnological methods (Abbadi et al., 2004; Dyer et al., 2008). Based on our current understanding of seed oil production, improvements can be made for increasing oil content and composition in oil crops. For example, triacylglycerol (TAG) is the plant oil product that is chemically most similar to fossil oil and therefore has the most potential to replace petroleum-based oil (Agarwal, 2007). Thus, manipulation of the biosynthetic pathways leading to TAG is a useful endeavor. In addition, the study of high-yield oil crops that can be grown on marginal lands, such as *Camelina*, mustard, and *Jatropha,* will also be important (Table 9.2). These crops offer benefits to small-scale farmers, especially those in under-developed countries, by providing new sources of income using land that they would otherwise not have developed. Since oil biosynthesis pathways and genes that can be manipulated to improve oil yield and composition have been tested in model crops such as soybean and castor bean, the application of altered oil biosynthesis can now be implement in new marginal land energy crops.

While there are still limitations on the widespread application of plant biotechnology for bioenergy production, expanding information on the genes involved in plant biosynthetic pathways, compartmentation of intermediates within plant cells, and the genomics of bioenergy-producing plant species are helping to design new approaches for improving yields and composition in a variety of plant species (Allen et al., 2007). It remains to be seen if such a push can be accomplished. However, it is clear that the world is moving in the direction of bio-based energy, focusing on renewability and efficiency in energy consumption. Genetic improvements of energy crops require knowledge of desired quality attributes; the relative economic value of the quality parameters in relation to yield, genetic variation for the desired traits, or for molecular breeding; knowledge of genes to suppress or add; and knowledge of any associated negative consequences of biomass quality manipulation. Finding the optimal direction forward in the new biological age will

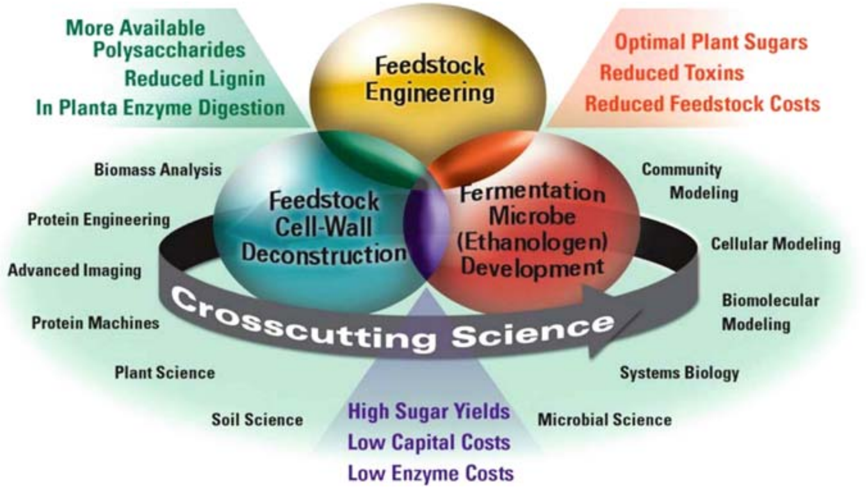


Fig. 9.10 A multidisciplinary approach to improvements in bioenergy. DOE's Bioenergy Roadmap for Biomass. From the DOE Genome Program (http://doegenomes.org)

certainly require a multidisciplinary approach that combines and spans many different fields of research (Fig. 9.10). Because plant bioenergy technology is still under development, desirable plant feedstock characteristics have not yet been completely delineated, but there is no question that plant biotechnology and modern biological techniques (such as genomics, proteomics, metabolomics, and fluxomics) will be at the heart of the improvements to bioenergy.

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