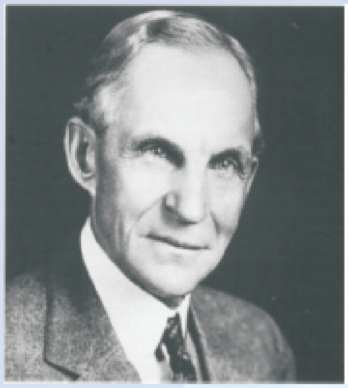

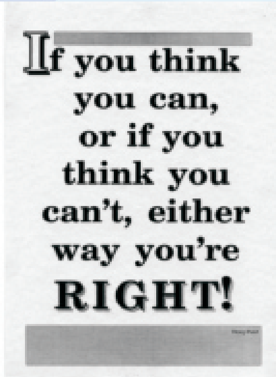

Corn to Ethanol:
Retrospect's and Prospects

Maize: Leading to a new Paradigm

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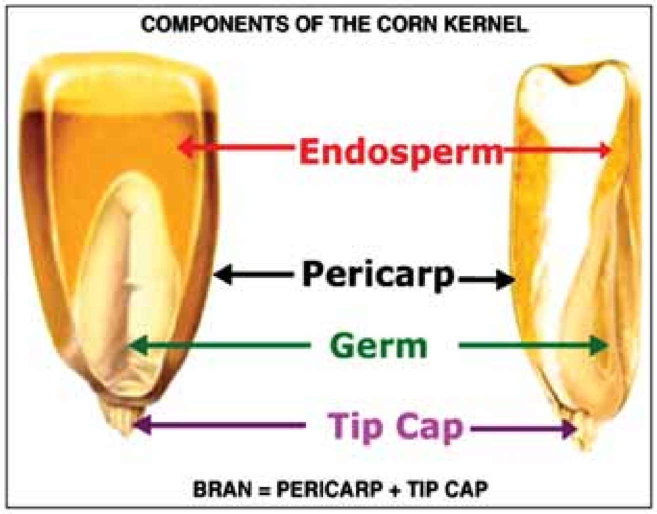
Background

Energy is the vital need of mankind and it is the priceless gift offered by the nature. Apart from food, we need lots of energy in various forms in our day-to-day life. At present, most of our energy requirements are fulfilled by non-renewable sources. The most extensively used non-renewable energy sources are the fossil fuels. Due to the unsustainable use of world's energy stores we had been encountered with numerous issues in the past and are fast approaching towards a similar crisis. The gasoline shortage of 1970's or the panic of fuel-price after Hurricane Katrina should be taken as wakeup calls so that we could remind ourselves from time to time regarding our ever depleting energy reserves. As we all know that energy can neither be created nor it can be destroyed, it can only be converted from one form to the other. So how long it will take us to realize importance of energy and the difficulty in tapping the energy from geological reserves? Man need to think of "renewable" as replenishing only in the right circumstances and within the laws of balance of nature so as to allow the natural ecological cycles to remain natural, and not mess with Mother Nature.

As need is the mother of discovery, the foreseeing energy crisis has induced interest in the synthesis of bio-fuel. The global consumption of the liquid petroleum will increase tremendously in coming years. It is estimated that if the present trend continues, the energy demand is projected to grow by more than 50% by 2025 (Ragauskas, 2006). Most importantly, unlimited demand for limited petroleum resources can not be a satisfactory option for a long time. Therefore, before the things slip out of our hands, we must start working towards bringing transition from the non-renewable carbon source to renewable bio-resources. Corn to ethanol concept can be a road-map in this regard.

Use of ethanol as a fuel is not a novel concept. Henry Ford Model T was run by ethanol because his vision was to introduce such a vehicle that could be affordable by rural masses (Kovarik, et al. 1998). In 1930's people used to use ethanol as a fuel source in cars, but the practice was brought to an end post world war-II due to the availability of petroleum and natural gas as cheap source of energy (Bothast and Schlicher, 2005). It burns more cleanly and also increases the octane level of gasoline. Only half the volume is required to produce the same oxygen level in gasoline by ethanol due to higher oxygen content as compared to Methyl tert-Butyl Ether (MTBE) (Dipardo, 2000). Presently, USA is the major exporter of maize and shifted 30% of its maize grains towards bio-fuel production to meet society's prospective hidden requirements (Ragauskas, 2006). Corn is the most important and economical source of starch, comprising about 68-72% of kernel weight, which is easily converted into glucose and fermented into ethanol.



Maize is the major crop of India after rice and wheat that provides food, feed and fodder to the livestock (Fig.1) and serves as a source of basic raw materials for a number of industrial products mainly starch, corn oil, corn syrup, alcoholic beverages, cosmetics, bio-fuel and many more. Earlier the research emphasis was laid on the development of composite maize varieties, double cross and three way cross hybrids. However, with the realization of the advantages of single cross hybrid over double cross, three way cross and composites, the research was focused towards the development of single cross hybrids. The Directorate of Maize Research, India played a pivotal role in this direction. As a result of single cross hybrid (SCH) technology, maize achieved

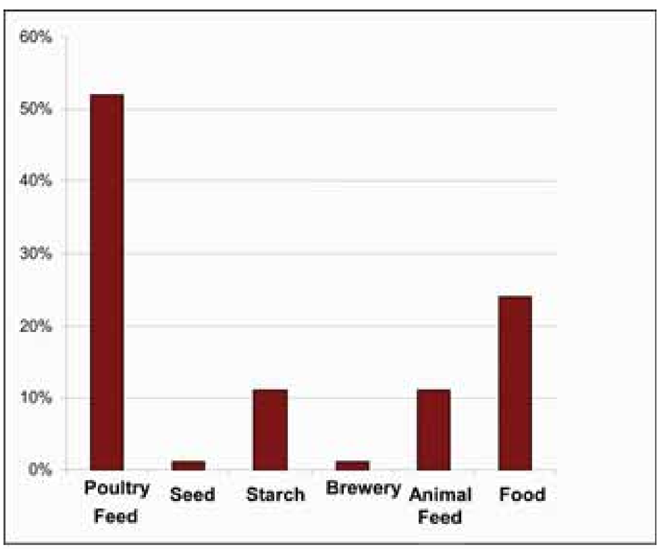


Figure 1: Maize Utilization Pattern in India

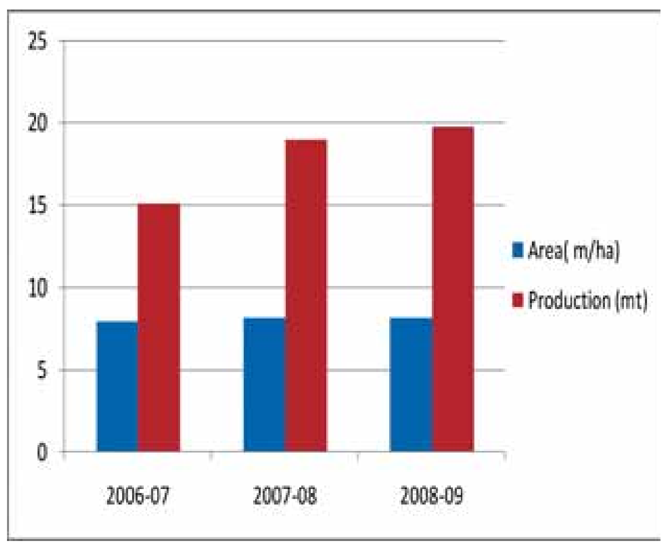


Fig. 2: Maize Growth Rate in India

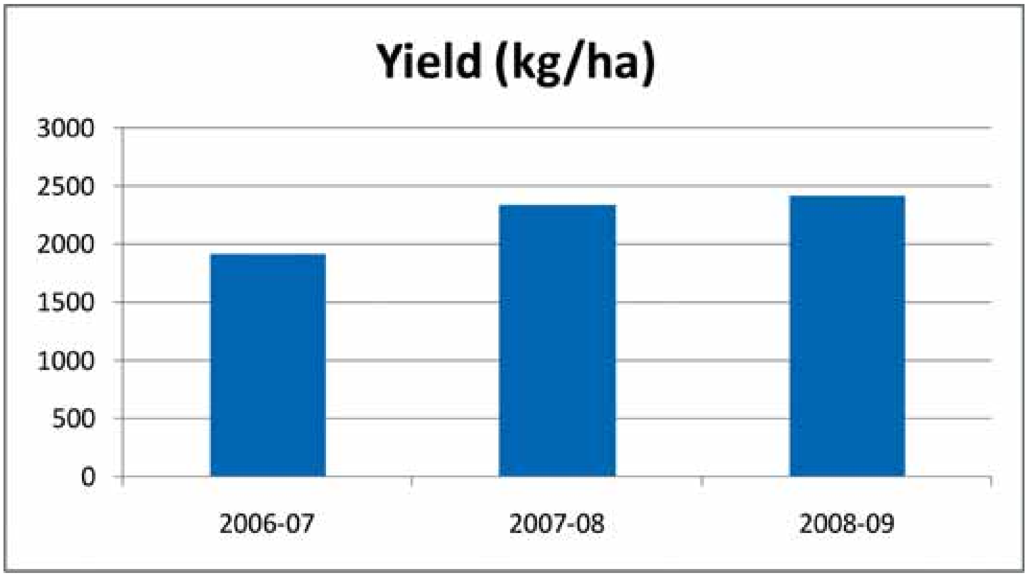


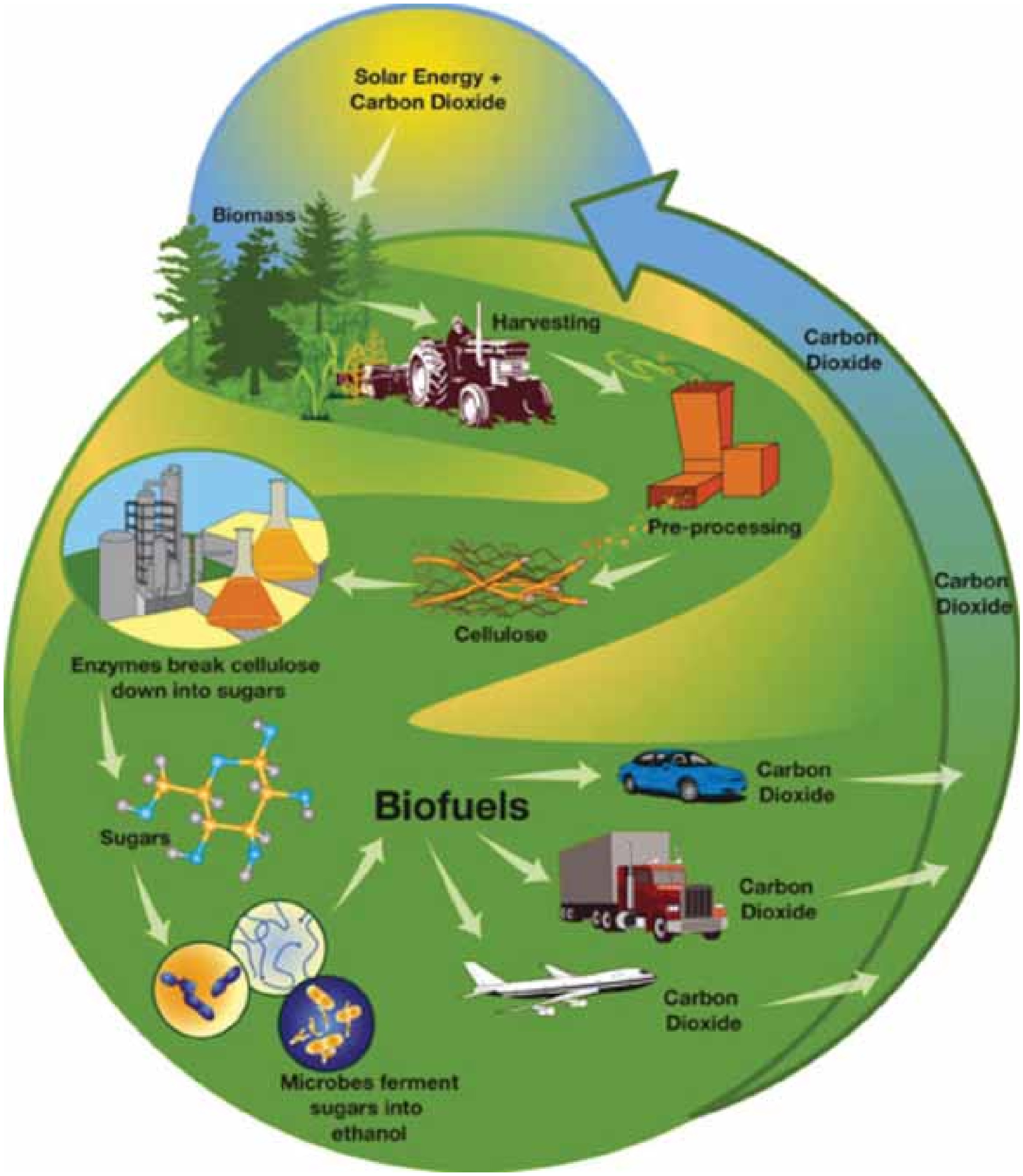
Fig. 3: Productivity pattern of India

the highest growth rate (6.7%) amongst cereals as against the required growth rate of 4.7% set by the XI planning commission of India (Fig.2 & 3) (DMR, Report). As a result India became importer to exporter of maize.

Crop yields kept pace with rapidly expanding production demand of global population and the available biomass can be much more innovative with superior and improved cellulosic technologies. Technological improvements in plant breeding hold promise to improve grain quality as well as quantity in India. Due to increase in area, production and productivity, India is producing surplus maize and this trend is likely to go on and the excess from the food basket can be used to secure us at the platform of energy and increasing future fuel demand of the country as an individual and world as a whole. Today, maize has set the stage to roll the dice and definitely ethanol will hit the right note.

Ethanol Production Processes

New Technologies are under experiment at various international research centers so as to provide more and better products to enhance the economics of the fuel ethanol. Further research in bio-fuel metrics is needed and the



Bio Fuel

major thrust areas are: Production of hybrids with higher starch content, Conversion of corn-kernel fiber fraction to ethanol and identification and development of new and higher value co-products. Many processes are employed like quick fiber (Singh et al. 1999), quick germ (Singh and Eckhoff 1996), COPE process (Cheryan 2002), enzymatic milling (Johnston et al. 2003), dry grind and wet milling. Both dry grind and wet milling processes are popular now-a-days but most of the ethanol is produced by dry grind method only because it is less capital and energy-intensive (Butzen and Haefele, 2008).

Dry-Grind Ethanol Process

Ethanol is commercially produced in by either of these ways, using either the wet mill or dry mill process. Wet milling involves separating the grain kernel into its constituents (germ, fiber, protein, and starch) prior to fermentation ((Butzen and Haefele, 2008). Whereas in dry mill process, the entire grain kernel is ground into flour. The starch in the flour is converted to ethanol during the fermentation process, creating carbon dioxide and distillers' grain. Fermentation is one of the oldest process known to man **(Bothast** and Schlicher**, 2005)**. In USA, ethanol is mainly produced by dry-grinding process (approximately 67%) and its percentage is increasing rapidly (Butzen and Haefele, 2008). The various procedure involved in this process are discussed below.

Mash Formation

The entire grains mass is screened to remove debris. The screened kernels are ground and then mixed with water to form slurry called ''mash''.

Cooking

In this process the starch in the flour is physically and chemically prepared for fermentation. The mash is further cooked and enzyme is added to convert the starch into sugar followed by the addition of yeast so as to accomplish the fermentation of sugar, leaving behind a mixture of ethanol and solids. The ethanol is extracted through distillation and dehydration and the solid remained are dried to produce distiller's dried grain solubles (DDGS).

Conversion of Starch

(Primary Liquefaction, and Secondary Liquefaction)

Corn Starch is composed of glucose units joined through a linkage in chains by a 1-4 and in branches by a 1-6 glycosidic bonds. The linear starch molecules are called amulose, whereas the branched on is called amylopectin. Starch usually comprises of 25-30% amylose and rest is amylopectin. Yeast can not metabolize this starch directly but must be broken down into simple sugar i.e. glucose, prior to fermentation. The pH of the mash is adjusted at 6.0 so as to carry out this conversion. A thermo stable enzyme a-amylase is added to mash which breakdown the starch polymer (hydrolyze a 1-4 bond) and produce soluble dextrin.

Primary Liquefaction

The slurry is then pumped through a pressurized jet cooker at 221°F and held for 5 minutes. The mixture is then cooled by an atmospheric or vacuum flash condenser.

Secondary Liquefaction

After the flash condensation cooling, the mixture is held for 1–2 hours at 180–190°F to give the alpha-amylase enzyme time to break down the starch into short chain dextrins. The slurry is heated to 180–190°F for 30–45 minutes to reduce viscosity and to provide mechanical shearing to rupture starch molecule especially of high molecular weight. The mash is further liquefied for at least 30 min to reduce the size of starch polymer. In overall, this step required the addition of a-amylase and steam i.e. gelatinization and liquefaction. This dextrinized mash is further cooked and adjusted to pH 4.5 to facilitate the addition of gluco-amylase to convert liquefied starch into glucose. To accomplish the sacchrification of starch to glucose, gluco-amylase is added in enough quantity. Simultaneous Sacchrification and Fermentation (SSF) is again a better option to reduce the microbial interference and more yield per bushel of the corn.

Simultaneous Saccharification and Fermentation

Once inside the fermentation tanks, the mixture is referred to as mash. The gluco-amylase enzyme breaks down the dextrins to form simple sugars. Yeast is then added to convert the sugar to ethanol and carbon dioxide. The mash is afterwards allowed to ferment for 48–72 hours, resulting in a mixture that contains about 10% ethanol as well as the solids from the grain and added yeast.

Fermentation

After sacchrification, cooling is done and mash is transferred to fermenter and yeast is added. *Saccharomyces cerevisiae* is commonly used species due to its quick and efficient production of alcohol. Consequently, addition of protease is of immense importance as it breaks down the corn-protein to free amino acids to serve as an additional source of nitrogen for the yeast. The whole process requires 48-72 hours and can concentrate up to 10-12% of ethanol. CO_2 formed during this process lowers down the pH below 4.0 in order to enhance the activity of gluco-amylase and to check the surrounding infection. The Batch and Continuous fermentation systems can be used and out of both batch process is more popular. CO_2 released can be captured and sold for the use in carbonating soft drinks, dry ice and some beverages industries.

Distillation and Dehydration

Distillation

Distillation is the process of separating ethanol from the solids and water in the mash. The fermented mash is pumped into a multi-column distillation system where additional heat is added. The columns utilize the differences in the boiling points of ethanol (78^{o}C) and water (100^{o}C) as a milestone to boil off and separate the ethanol. By the time the product stream is ready to leave the distillation columns, it contains about 95% ethanol by volume (190-proof).

Dehydration

The 190-proof ethanol still contains about 5% water. Alcohol and water form an azeotrope at this point and can not be separated further. Rest of 5% water must be removed in order to blend

it with gasoline. To carry out this operation It is passed through a molecular sieve to physically separate the remaining water from the ethanol based on the different sizes of the molecules. This step produces 100% pure (200-proof) anhydrous (waterless) ethanol.

Ethanol Storage

Before the ethanol is sent to storage tanks, a small amount of denaturant (5% gasoline) is added to render it undrinkable (unfit for human consumption) and save the beverage alcohol tax. Most ethanol plants' storage tanks are sized to allow storage of 7–10 days' production capacity.

Co-Product Processing

The residue from this process, called ''whole-stillage'' contains non-fermentable starch, fiber, oil, and protein component of the grain and water. It is pumped out from the bottom of the columns into the centrifuges. During the ethanol production process, two valuable co-products are created: carbon dioxide and distillers grains.

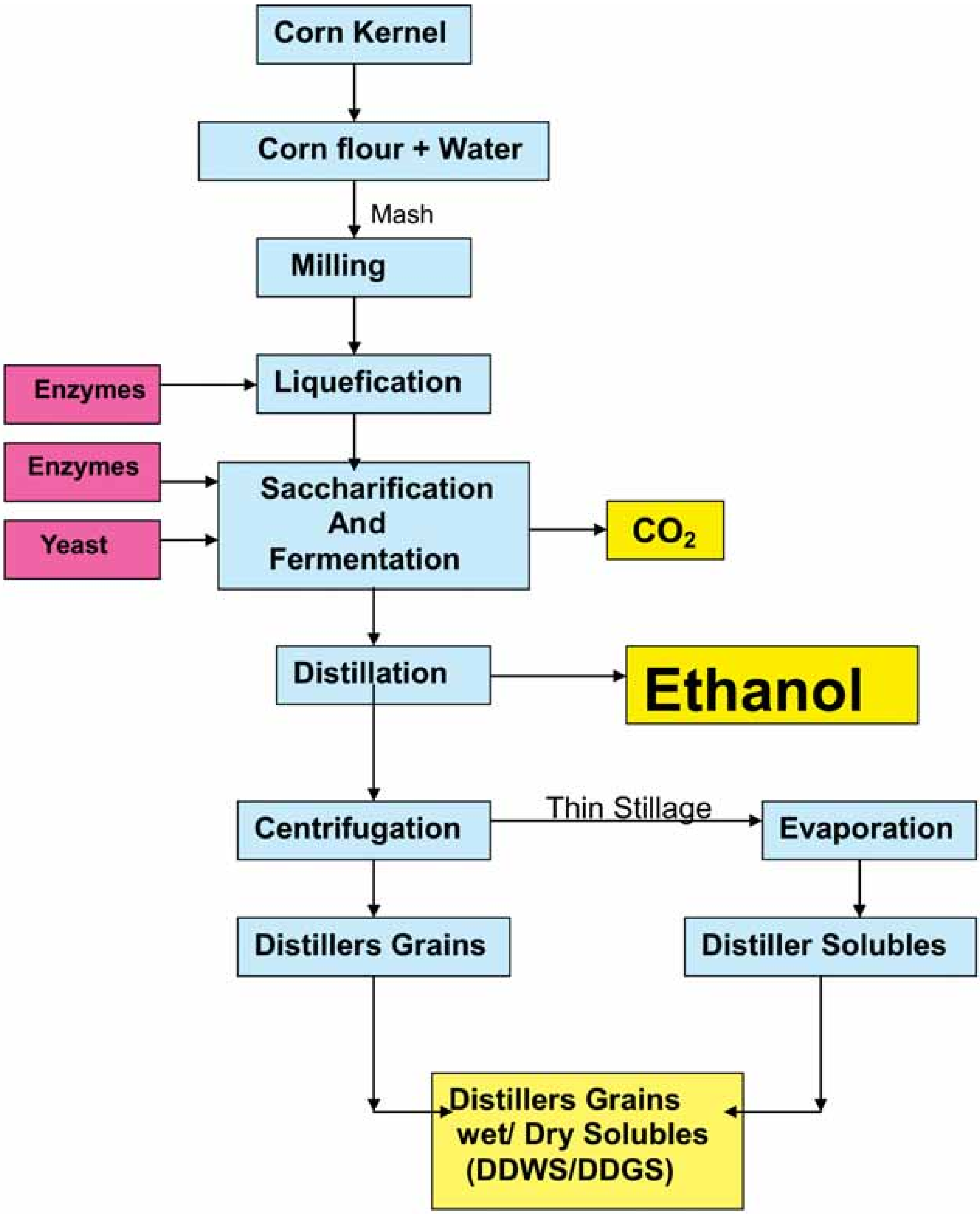
As yeast ferment the sugar, they release large amounts of carbon dioxide gas. This CO_2 can be released into the atmosphere, but it's generally captured and sold to food processing industries as discussed earlier. The stillage from the bottom of the distillation tanks contains solids from the grain and added yeast as well as liquid from the water added during the whole process. This is a valuable feed ingredient and can fed as such but it is usually sent to centrifuges for separation into thin stillage (a liquid with 5–10% solids) and wet distillers grain. Some of the thin stillage (15-30%) is routed back to the cook / slurry tanks as makeup water, reducing the amount of fresh water required by the cook process. The rest is concentrated further into syrup containing 25–50% solids. After drying (evaporation), the thick syrup, which is high in protein and fat content, is then mixed back to create feed product known as wet distillers grain (WDG).

With the added syrup, the WDG (65% moisture) still contains most of the nutritive value of the original feedstock plus the added yeast. It is preferred as an excellent cattle ration for local feedlots and dairies due to high moisture content. After the addition of the syrup, it's conveyed to a wet cake pad having a self-life of 1-2 weeks. So, it is loaded for transport as soon as possible. Unless the target destination is within the vicinity of plant, it becomes hard to handle the same. Many ethanol facilities do not have enough nearby cattle to utilize all of the WDG. To reduce this burden WDGS is often dried to 10-13% moisture to give rise to Dried Distiller's Grain Solubles (DDGS).This facilitates the removal of moisture and extends its shelf life. This dried distillers grain (DDG) is commonly used as a high-protein ingredient in cattle, swine, poultry, and fish diets.

DDGS

Wet-Milling Process

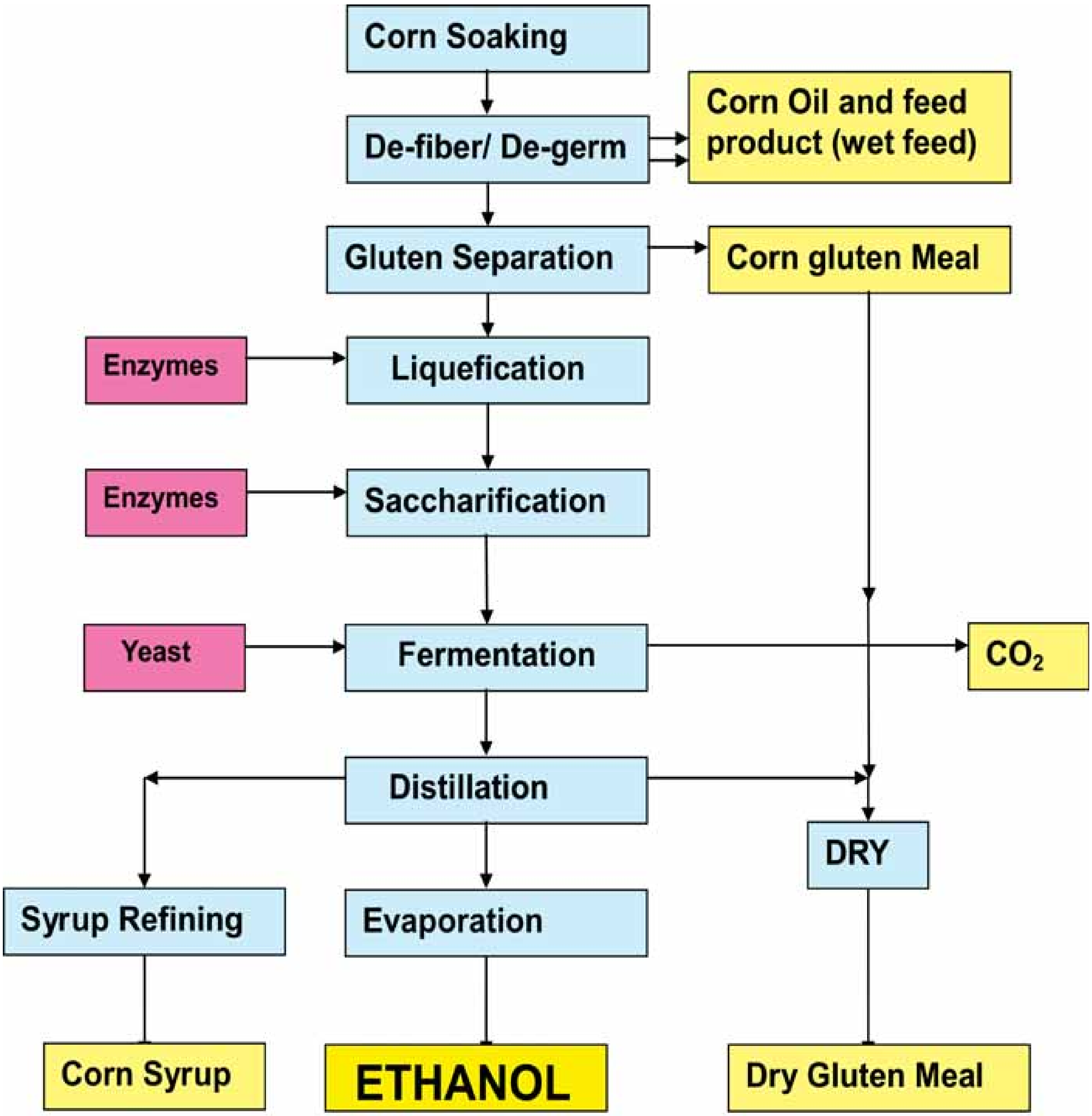
The major difference between the wet-mill and dry grind is the initial treatment of the grain. In wet-mill the grain is steeped or soaked into water and dilute sulfurous acid for 24-48 hours to facilitate the separation of no. of component parts of the grain including starch, germ, fiber and gluten. The germ is removed from the kernel by processing the corn slurry through a series of grinders. The germ is used either to extract the corn oil on the site or sold to crushers who extract the corn oil. Other remaining components like starch, fiber and gluten are segregated further as by-products by centrifuge, screen and hydroclonic separators. The germ meal is then added to fiber to form corn gluten feed and sold to livestock industry. Corn gluten meal is a very high-protein animal feed and sought after as a feed ingredient in poultry broiler operations. Moreover, heavy steep water can also be sold as either feed ingredients or as a part of Ice-Ban (an environmental friendly alternative to salt) for removing ice from the roads in various countries. In this process, fermentable sugars are produced from the starch and fermented into ethanol. Fate of this starch and remaining water can be either fermented into ethanol or dried and sold as dried or modified corn starch or processed into corn-syrup. Wet-mill facilities are considered as true ''Bio-refineries'' as this process is followed by various high-value co-products.



Dry-Grind Ethanol Process

CORN GLUTEN MEAL

Wet Distiller Grains



Wet-Mill Ethanol Process

FERMENTER

Components of fermenter

1. Basic component includes drive motor, heaters, pump, etc.
2. Vessels and accessories
3. Peripheral equipment (reagent bottles)
4. Instrumentation and sensors

Various components of an ideal fermenter for batch process are:

1 Top plate cover (made of steel)

2 Clamp top plate compressed onto vessel using clamp

3 Seal separates top plate from vessel (glass) to prevent air leakage

4 Vessel glass, jacketed, steel with ports for various outputs, inputs, probes etc

5 Drive motor used to drive mixing shaft

6 Drive shaft mixes the medium evenly with its impeller

7 Marine impeller for plant tissue culture

8 Baffles prevent sedimentation on sides and proper mixing

9 Sparger air supplier / after filtration via membranes – ensures efficient dispersal – by attached to impeller

10 Exit gas cooler like condenser remove as much moisture as possible from exhaust

11 Inoculation needle port to add inoculum

12 Feed pumps regulates the flow rates of additives (medium, nutrients) variable speed

13 Peristaltic pumps fixed speed pumps – used for continuous sampling

14 Syringe pump using a syringe – mostly used in batch

15 Exit gas analysis CO_2 analyzer, O_2 analyzer, mass spectrometer

16 Sample pipe through which samples are drawn

17 Three way inlet to insert different probes

Monitoring and controlling parts of fermenter are:

1 Pt100 temperature sensor (platinum resistance electrode)

2 Foam probe kept above the medium level to sense foam formation

3 pH electrode senses pH

4 O_2 sensor Monitors dissolved oxygen level

5 Heater pad directly heats the medium

6 Cold finger after direct heating – used to cool the vessel contents (closed coil/pipe to pass cool water)

7 Rotameter variable air flow meter – indicates rate of air flow into vessel – attached to air sparger

8 Pressure valve attached to rotameter for safer operation

9 Air pump supply of air

10 Peristaltic pump to pump in medium, acids, bases, antifoam

TYPES OF FERMENTERS

The main function of a fermenter is to provide a controlled environment for the growth of microorganisms or animal cells, to obtain a desired product. Few of the bioreactor types are discussed below (J.E Bailey and D.Ollis):

STIRRED TANK FERMENTER

Microbial fermentations received prominence during 1940's namely for the production of life saving antibiotics. Stirred tank reactor is the choice for many (more than 70%) though it is not the best. Stirred tank reactor's have the following functions: homogenization, suspension of solids, dispersion of gas-liquid mixtures, aeration of liquid and heat exchange. The Stirred tank reactor is provided with a baffle and a rotating stirrer is attached either at the top or at the bottom of the bioreactor. The typical decision variables are: type, size, location and the number of impellers; sparger size and location. These determine the hydrodynamic pattern in the reactor, which in turn influence mixing times, mass and heat transfer coefficients, shear rates etc. The conventional fermentation is carried out in a batch mode. Since stirred tank reactors are commonly used for batch processes with slight modifications, these reactors are simple in design and easier to operate. Many of the industrial bioprocesses even today are being carried out in batch reactors though significant developments have taken place in the recent years in reactor design, the industry, still prefers stirred tanks because in case of contamination or any other substandard product formation the loss is minimal. The batch stirred tanks generally suffer due to their low volumetric productivity. The downtimes are quite large and unsteady state fermentation imposes stress to the microbial cultures due to nutritional limitations. The fed batch mode adopted in the recent years eliminates this limitation. The Stirred tank reactor's offer excellent mixing and reasonably good mass transfer rates. The cost of operation is lower and the reactors can be used with a variety of microbial species. Since stirred tank reactor is commonly used in chemical industry the mixing concepts are well developed. Stirred tank reactor with immobilized cells is not favored generally due to attrition problems; however by separating the zone of mixing from the zone of cell culturing one can successfully operate the system.

AIR-LIFT FERMENTER

Airlift fermenter (ALF) is generally classified as pneumatic reactors without any mechanical stirring arrangements for mixing. The turbulence caused by the fluid flow ensures adequate mixing of the liquid. The draft tube is provided in the central section of the reactor. The introduction of the fluid (air / liquid) causes upward motion and results in circulatory flow in the entire reactor. The air / liquid velocities will be low and hence the energy consumption is also low. ALFs can be used for both free and immobilized cells. There are very few reports on ALFs

for metabolite production. The advantages of Airlift reactors are the elimination of attrition effects generally encountered in mechanical agitated reactors. It is ideally suited for aerobic cultures since oxygen mass transfer coefficient are quite high in comparison to stirred tank reactors. This is ideal for SCP production from methanol as carbon substrate. This is used mainly to avoid excess heat produced during mechanical agitation.

FLUIDISED BED BIOREACTOR

Fluidised bed bioreactors (FBB) have received increased attention in the recent years due to their advantages over other types of reactors. Most of the FBBs developed for biological systems involving cells as biocatalysts are three phase systems (solid, liquid & gas). The fundamentals of three phase fluidization phenomena have been comprehensively covered in chemical engineering literature. The FBBs are generally operated in co-current upflow with liquid as continuous phase and other more unusual configurations like the inverse three phase fluidized bed or gas solid fluidized bed are not of much importance. Usually fluidization is obtained either by external liquid re-circulation or by gas fed to the reactor. In the case of immobilized enzymes the usual situation is of two-phase systems involving solid and liquid but the use of aerobic biocatalyst necessitate introduction of gas (air) as the third phase. A differentiation between the three phase fluidized bed and the airlift bioreactor would be made on the basis that the latter have a physical internal arrangement (draft tube), which provides aerating and non-aerating zones. The circulatory motion of the liquid is induced due to the draft tube. Basically the particles used in FBBs can be of three different types:

1. Inert core on which the biomass is created by cell attachment.

2. Porous particles in which the biocatalyst is entrapped.

3. Cell aggregates / flocs (self-immobilization).

In comparison to conventional mechanically stirred reactors, FBBs provide a much lower attrition of solid particles. The biocatalyst concentration can significantly be higher and washout limitations of free cell systems can be overcome. In comparison to packed bed reactors FBBs can be operated with smaller size particles without the drawbacks of clogging, high liquid pressure drop, channelling and bed compaction. The smaller particle size facilitates higher mass transfer rates and better mixing. The volumetric productivity attained in FBBs is usually higher than in stirred tank and packed bed bioreactors. There are several successful examples of using FBBs in bioprocess development.

PACKED BED BIOREACTOR

Packed bed or fixed bed bioreactors are commonly used with attached biofilms especially in wastewater engineering. The use of packed bed reactors gained importance after the potential of whole cell immobilization technique has been demonstrated. The immobilized biocatalyst is packed in the column and fed with nutrients either from top or from bottom. One of the disadvantages of packed beds is the changed flow characteristic due to alterations in the bed porosity during operation. While working with soft gels like alginates, carragenan etc the bed compaction which generally occurs during fermentation results in high pressure drop across the bed. In many cases the bed compaction was so severe that the gel integrity was severely hampered. In addition channeling may occur due to turbulence in the bed. Though packed beds belong to the class of plug flow reactors in which backmixing is absent in many of the packed beds slight amount of backmixing occurs which changes the characteristics of fermentation. Packed beds arc generally used where substrate inhibition governs the rate of reaction. The packed bed reactors are widely used with immobilized cells. Several modifications such as tapered beds to reduce the pressure drop across the length of the reactor, inclined bed, horizontal bed, rotary horizontal reactors have been tried with limited success.

BUBBLE COLUMN FERMENTER

Bubble column fermenter is a simplest type of tower fermenter consisting of a tube which is air sparged at the base. It is an elongated non-mechanically stirred fermenter with an aspect ratio of 6:1. This type of fermenter was used for citric acid production.

TYPES OF FERMENTATION

Any fermentation process can be carried out as Batch, Continuous and Fed Batch.

Batch:

Fermentation process allowed proceeding for limited time with required controlled conditions. After the stipulated incubation period the process stopped and yield recovered.

Continuous:

Fermentation process allowed to proceed continuously with continuous supply of required nutrients and sample withdrawn regularly.

Fed batch:

Fermentation process allowed to proceed continuously and sample withdrawn discontinuously with intermittently addition of medium

Uses of ethanol

Drinks

The "alcohol" in alcoholic drinks is simply ethanol.

Industrial methylated spirits

Ethanol is usually sold as industrial methylated spirits which is ethanol with a small quantity of methanol added and possibly some colour. Methanol is poisonous, and so the industrial methylated spirits is unfit to drink. This avoids the high taxes which are levied on alcoholic drinks (mainly in the UK!).

As a fuel

Ethanol burns to give carbon dioxide and water and can be used as a fuel in its own right, or in mixtures with petrol (gasoline). "Gasohol" is a petrol / ethanol mixture containing about 10 - 20% ethanol.

Because ethanol can be produced by fermentation, this is a useful way for countries without an oil industry to reduce imports of petrol.

As a solvent

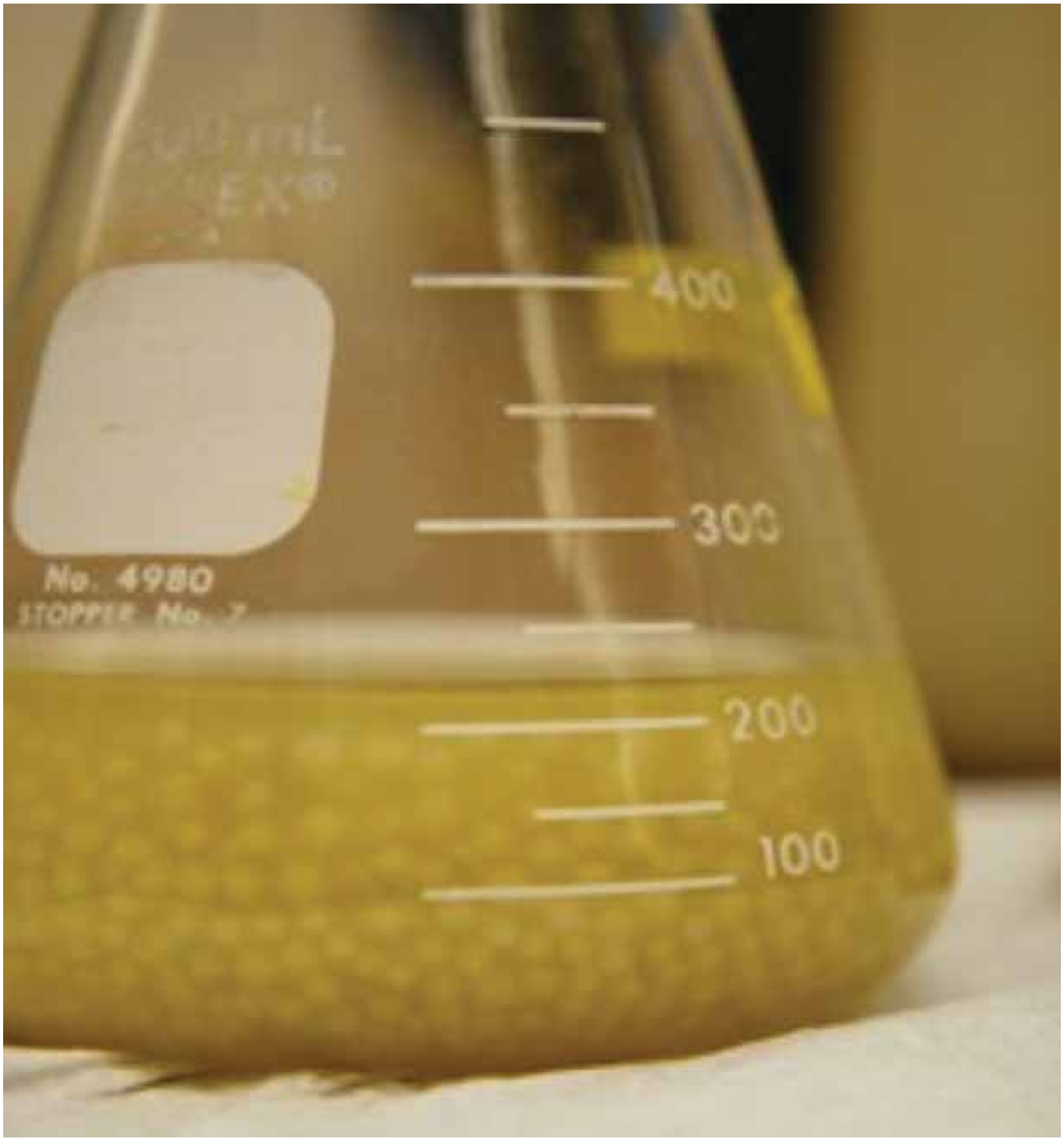
Ethanol is widely used as a solvent. It is relatively safe, and can be used to dissolve many organic compounds which are insoluble in water. It is used, for example, in many perfumes and cosmetics.

Corn to Ethanol: Economics

The bio-fuel concept is gaining momentum slowly. Bio-fuel is garnering larger and larger shares of the fuel markets. In the USA, ethanol production tripled in last few years from 2.8 billion gallons in 2003 to over 9 billion gallons in 2008 (Ling Tao and Andy Aden 2009). Under the Energy Independence and Security Act of 2007 (EISA 2007), 36 billion gallons of renewable fuel is required by 2022. Out of which, 15 billion gallons are corn-based ethanol. Most of the ethanol produced in USA is derived from corn only. Dry-grind process is more prevalent (RFA 2008). As a result, ethanol produced from wet-mill are comparatively lower (2.5 gal per bushel) than from dry-grind processes (2.8 gal per bushel) (Ling Tao and Andy Aden 2009).

Dry-grind Ethanol process Products are: ethanol (2.8 gal.), DDGS (18 lbs.), CO_2 (18 lbs.) and 150 bushel corn yields 413 gallons of ethanol per acre, 2700 pounds of DDGS (Douglas G. Tiffany). These high value co-products have potential to accelerate with the cost of ethanol production. Establishment of large scale industries can make ethanol production more economical. The variation of Ethanol production potential ranges up to 7% depending upon different corn hybrids (Pioneer's report)

Enormous debate over the potential benefits of bio-fuel has taken place for the concept of net energy. Various studies have accounted that ethanol and co-products manufactured from corm yielded a positive net energy and this can be of about 4 MJ/l to 9 MJ/l (Farrell 2006). The study that ignored co-products but used recent data found a slight positive net energy for corn ethanol. Studies that reported negative net energy ignored the co-products and used some of the obsolete data incorrectly.



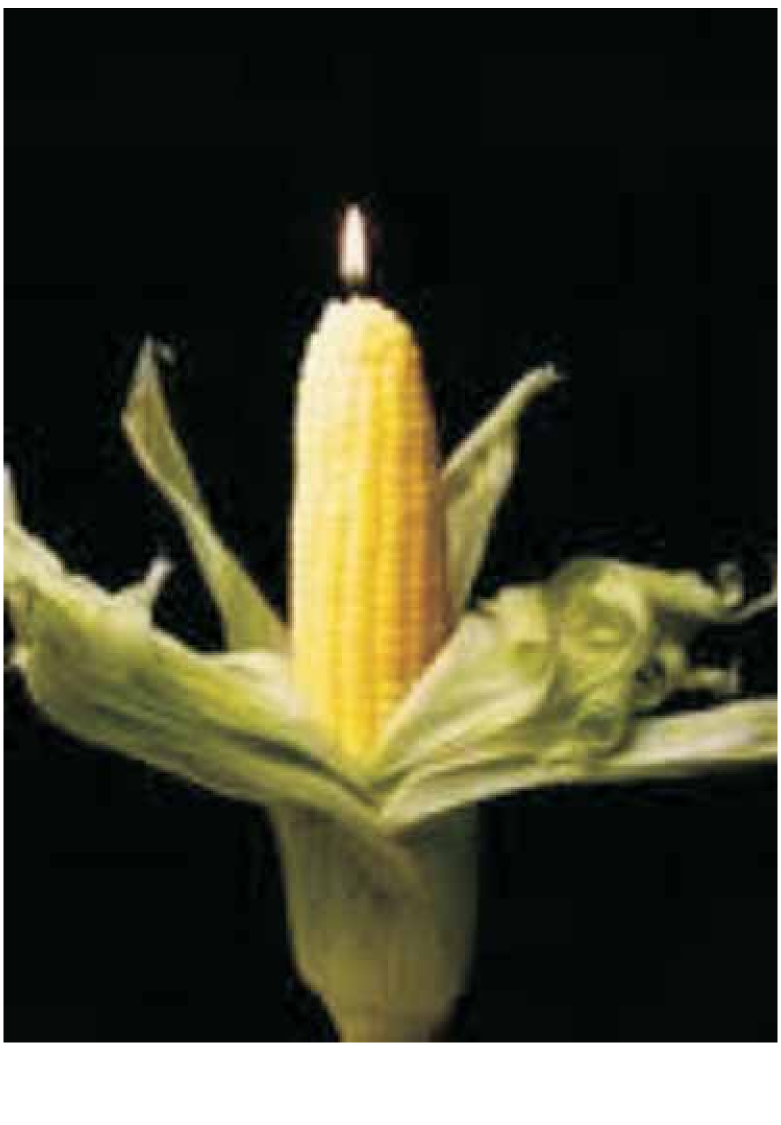
India and Ethanol Production

India now has a surplus of corn and sorghum that can be economically processed into ethanol so as to meet the emerging energy demands of the nation and the world (India and Ethanol Production.mht). International-Planners is planning the construction, of 20 regional grain to ethanol conversion facilities in India, to help the national petroleum ministry, fulfill its mandate, and provide drivers with auto, bus, truck & train fuels that must contain at least 5% ethanol by the year 2012. They will produce 6,000,000 gallons of ethanol a month, Ending India's reliance upon foreign fuel additives and doubling the income of the average farm workers family so as

to make them economically independent. These ethanol production centers can be built in the 20 different states producing corns, sugar cane & sorghum. India now has to import 15 million barrels of ethanol a year. The Indian government has recently offered to underwrite 75% of the development costs of ethanol bio-mass production centers in order to stimulate national ethanol production. The ethanol production centers will produce those 15 million barrels of ethanol for India's needs, which will save India each year more than Rupees 9,000,000,000~$760,000,000.

Future Prospects

In view of changing world energy requirements, a research road-map for the bio-refineries is mandatory. This will contribute towards sustainability in terms of energy. Various researchers are trying to come up with cellulosic approaches to enhance the economics of the corn to ethanol process. Progress towards attaining these goals requires new technologies, better quality hybrids, accelerated plant domestication programmes, improved management practices and enough research funding to develop a future renewable energy source. The demands in future bio-refineries will stimulate further advancement in the agriculture to produce more and more improved biomass for ethanol production. Such approaches can lead to a bio-fuel industry that will satisfy improved vehicle efficiency and in overall, will meet the energy security and climate change imperatives of the nation and the world because energy and environment implications of ethanol production are more important than others. Nonetheless, giving up humanity's dependence on decreasing non-renewable energy source is a big challenge in itself that need to be considered by researchers in order to win the end game.



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