Corn Grading, Cleaning, Milling and Cooking

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Topics of Discussion

- Feedstock Grading and Storage
- Cleaning and Milling
- Cooking

Feedstock Storage: Farm or Elevator

- Corn is harvested in the US once a year (July-Sept)
- Storage of corn is required
  - For year round operation
  - Minimize the carbohydrate losses due to spoilage or sprouting
- Storage time is dependent on
  - Moisture content and grain temperature
  - Drying is generally required at the time of harvest

Recommended Moistures for Safe Storage

Grain Type & Storage Time	Maximum Moisture, %
Shelled Corn & Sorghum	
Sold by Spring	15.5
Stored 6 - 12 mos.	14
Stored more than a year	13

Feedstock Quality and Grading

- Corn delivered at plant is measured for
  - Moisture content
  - Mycotoxins
  - BCFM (broken corn and foreign material)
  - Test Weight
  - Grade

Grade Requirements

Grade	Minimum test weight per bushel (Percent)	Maximum Limits of Damaged Kernels: Heat Damaged Kernels (Percent)	Maximum Limits of Damaged Kernels: Total (Percent)	Maximum Limits of Broken corn and foreign material (Percent)
U.S. No. 1	56.0	0.1	3.0	2.0
U.S. No. 2	54.0	0.2	5.0	3.0
U.S. No. 3	52.0	0.5	7.0	4.0
U.S. No. 4	49.0	1.0	10.0	5.0
U.S. No. 5	46.0	3.0	15.0	7.0

U.S. Sample grade is corn that:

(a) Does not meet the requirements for the grades U.S. Nos. 1, 2, 3, 4, or 5; or

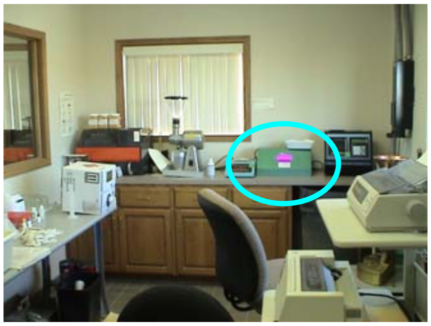
(b) Contains stones which have an aggregate weight in excess of 0.1 percent of the sample weight, 2 or more pieces of glass, 3 or more crotalaria seeds (Crotalaria spp.), 2 or more castor beans (Ricinus communis L.), 4 or more particles of an unknown foreign substance(s) or a commonly recognized harmful or toxic substance(s), 8 or more cocklebures (Xanthium spp.) or similar seeds singly or in combination, or animal filth in excess of 0.20 percent in 1,000 grams; or

(c) Has a musty, sour, or commercially objectionable foreign odor; or

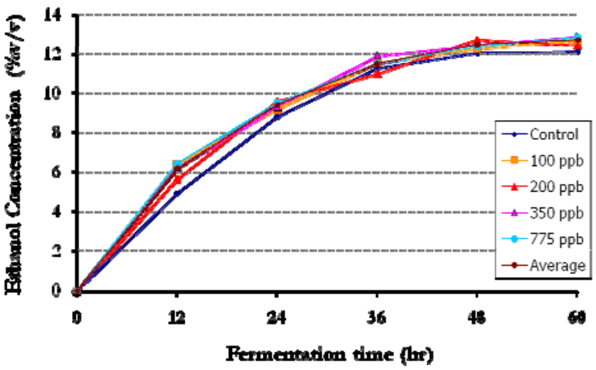
(d) Is heating or otherwise of distinctly low quality.

Mycotoxin Detection

- Use of black light to detect mold growth in corn
- It is the mold that fluoresces not mycotoxins
- Definitive detection of mycotoxins is done by HPLC
- In dry grind ethanol plant mycotoxins end up in DDGS (animal foodstuff)
- Mycotoxins potentially stress yeast and lower ethanol yields



Effect of Mycotoxins on Dry Grind Ethanol



Murthy, G.S., Townsend, D.E., Meerdink, G.L., Bargren, G.L., Tumbleson, M.E. and Singh, V. 2005. Effect of aflatoxin B1 on dry grind ethanol process. Cereal Chem. 82:302-304.

Effect of Mycotoxins on Dry Grind Ethanol

	Wet Grains	Thin Stillage
	%	
Control	-	-
100 ppb	55.2	44.8
204 ppb	47.2	52.8
342 ppb	58.1	41.9
772 ppb	74.4	25.6

Murthy, G.S., Townsend, D.E., Meerdink, G.L., Bargren, G.L., Tumbleson, M.E. and Singh, V. 2005. Effect of aflatoxin B1 on dry grind ethanol process. Cereal Chem. 82:302-304.

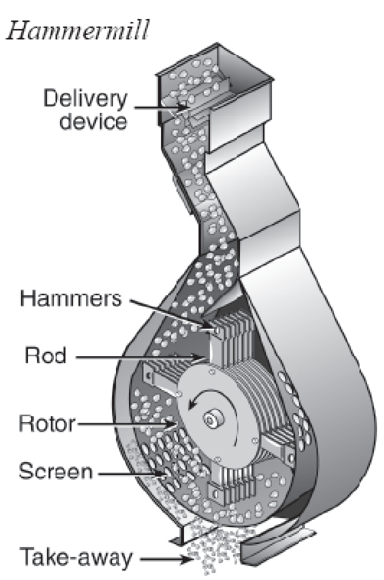
Cleaning and Milling

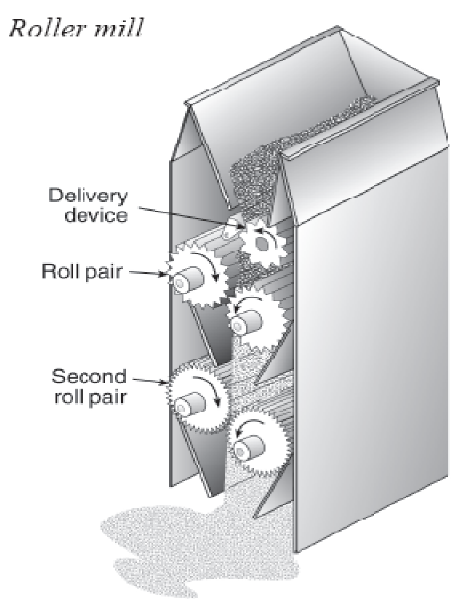
- Cleaning
  - Prior to processing corn is cleaned for BCFM
    - Stones, metals, other debris
    - Can cause problems with screens and other equipment
    - Dust problems
  - Cleaning can be done by sieving or blowing air

Cleaning and Milling

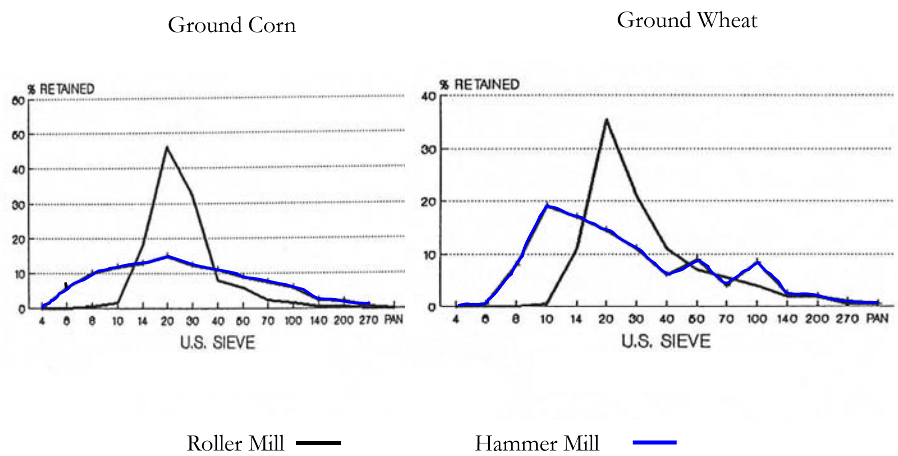
- Milling
  - Starch is present in corn in small cells. Milling exposes cells for hydration, liquefaction and conversion to glucose
  - The objectives of Milling are
    - Split and remove the pericarp which is cutinized from outside and impervious to penetration of water
    - Disintegrate the inner portion of corn kernel into small pieces to make all the constituents readily accessible to enzymatic action
    - Minimize the quantity of very fine flour
      - prevent balling during slurring
      - prevent increase in amount of soluble solids in thin stillage

Milling Equipment

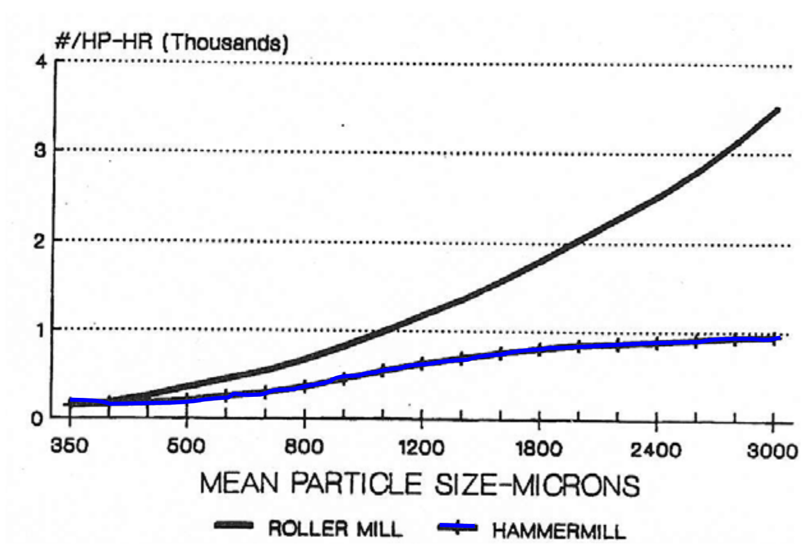




Comparison between Roller Mill and Hammer Mill for Size Reduction

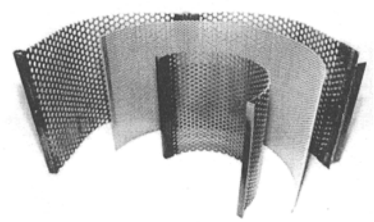


Efficiency Comparison between Roller Mills and Hammer Mills



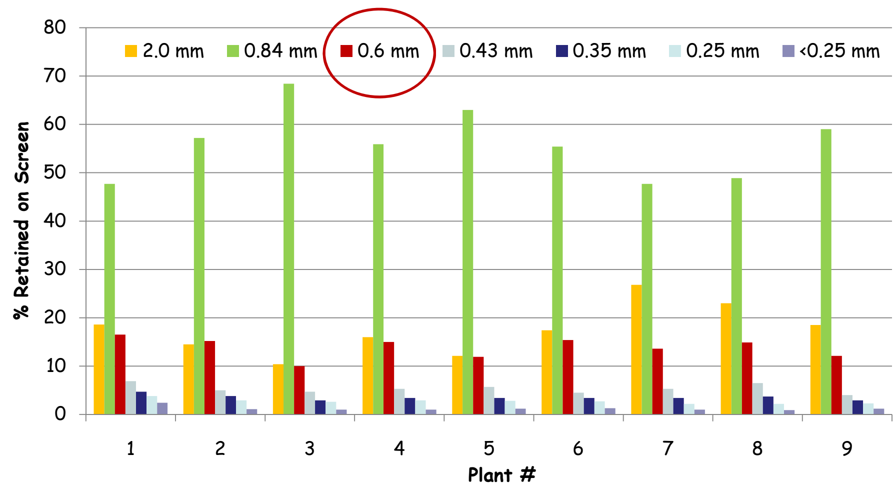
Control of Particle Size

- Mill outlet is controlled by retention screen
- The screens are normally in size range of 0.125 (6/64") - 0.172 inches (11/64")
- Sieve analysis is done to determine the particle size of the ground flour



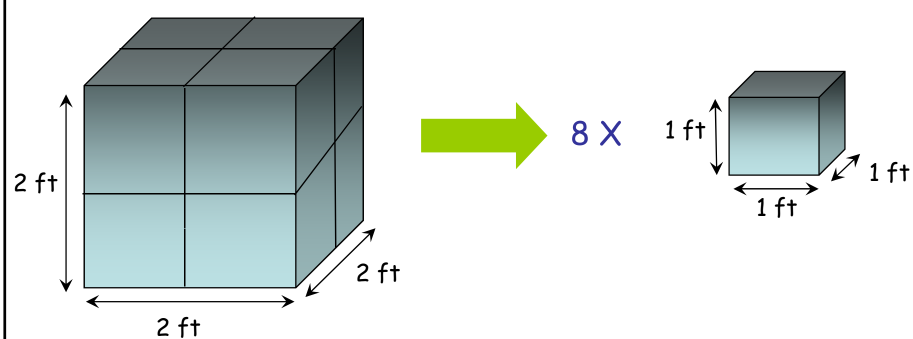
Hammer Mill Operation

Sieve Analysis of Corn Meal



Rausch et al. 2005. Trans of ASABE.

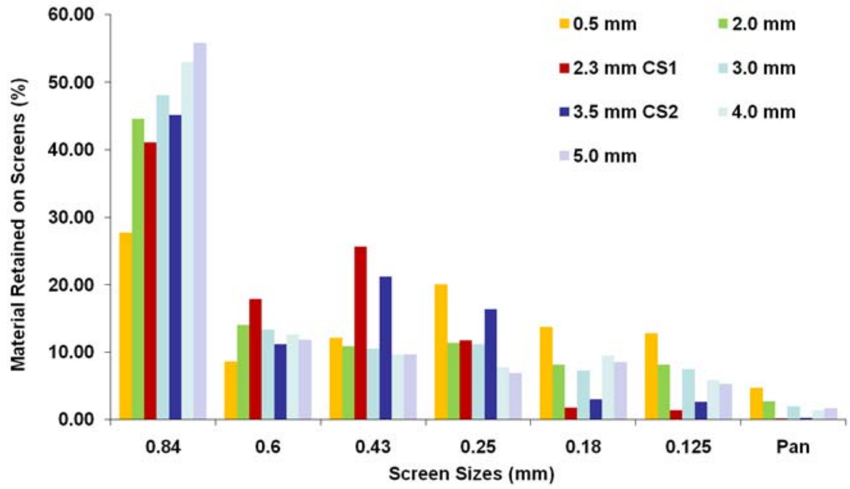
Effect of Grind Size on Ethanol Yield



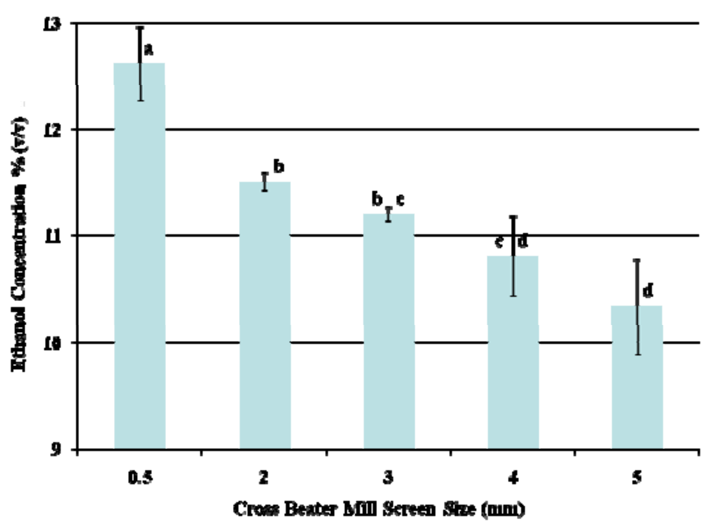
V = 8 ft³; SA=24 ft²

V = 8 ft³; SA=48 ft²

Effect of Grind Size on Ethanol Yield

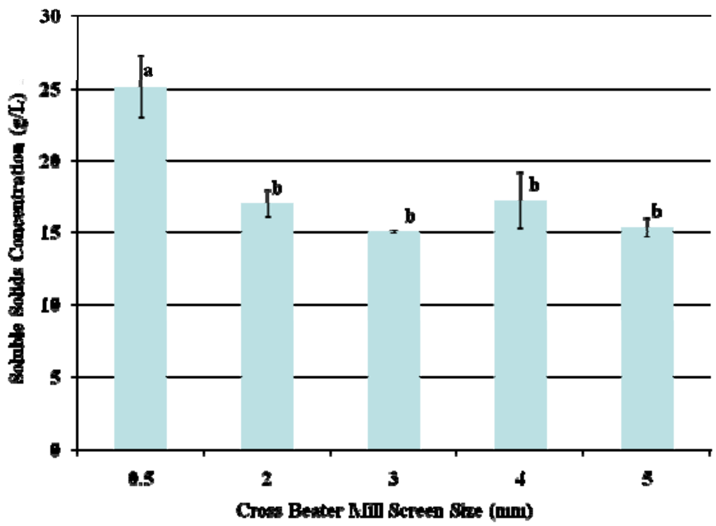


Effect of Grind Size on Ethanol Yield

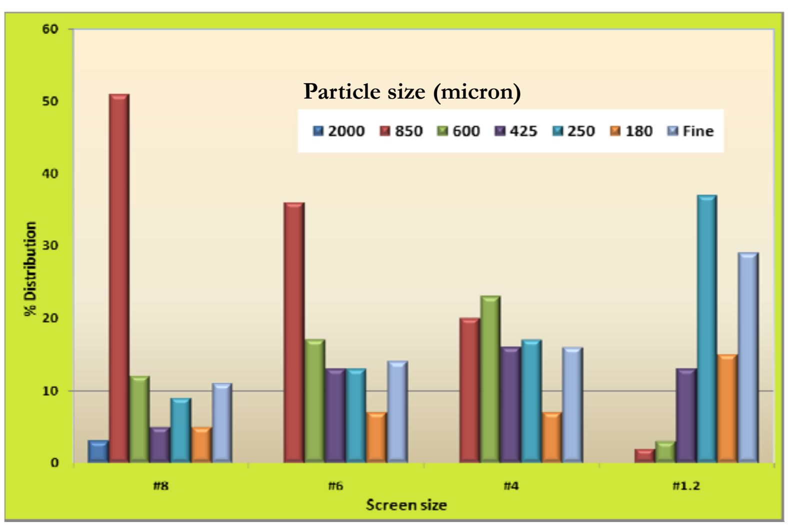


ILLINOIS

Effect of Grind Size on Thin Stillage



Effect of Grind Size



Screen size	mm
#8	3.18
#6	2.38
#4	1.59
#1.2	0.51

- Smaller screen sizes gave higher distribution of finer particle sizes

Soong et al. 2009. Novozymes NA. Grind size study in conventional process – Effects of α-amylase and glucoamylase doses on the processing parameters. Proceedings for International Starch Conference, Urbana, IL

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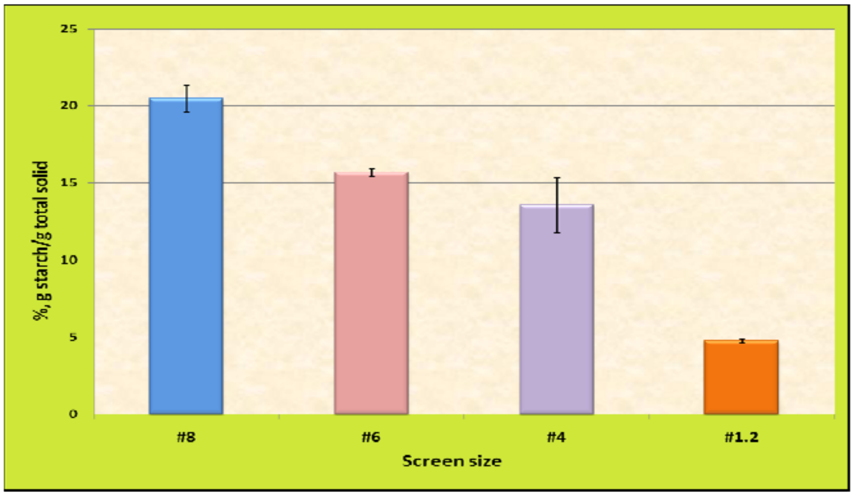
Post Liquefaction – Insoluble Starch



- Smaller screen sizes produced lower insoluble starch values after liquefaction
- Factors that contribute insoluble starch : a) Incomplete gelatinization; b) Inefficient liquefaction process

Soong et al. 2009. Novozymes NA. Grind size study in conventional process – Effects of α-amylase and glucoamylase doses on the processing parameters. Proceedings for International Starch Conference, Urbana, IL

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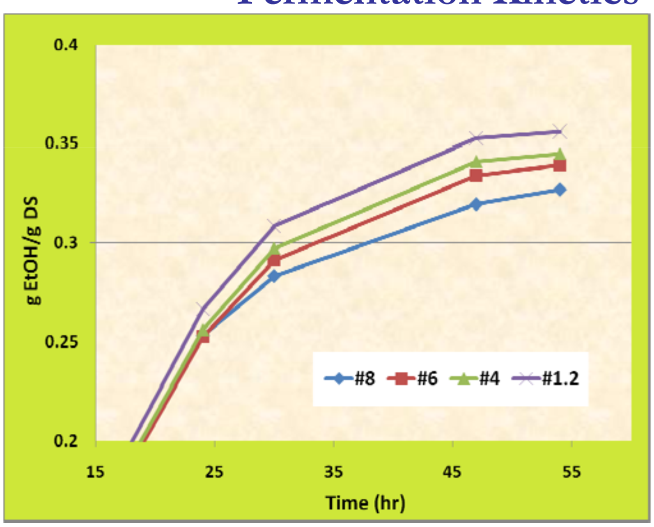
Effect of Grind Size on SSF – Fermentation Kinetics



- Smaller screen sizes gave faster fermentation kinetics and higher ethanol titers

Note: **Liquozyme dose, 0.024% w/w corn as is**
Spirizyme Fuel, 0.055% w/w corn as is (typical industry dose)

Soong et al. 2009. Novozymes NA. Grind size study in conventional process – Effects of α-amylase and glucoamylase doses on the processing parameters. Proceedings for International Starch Conference, Urbana, IL

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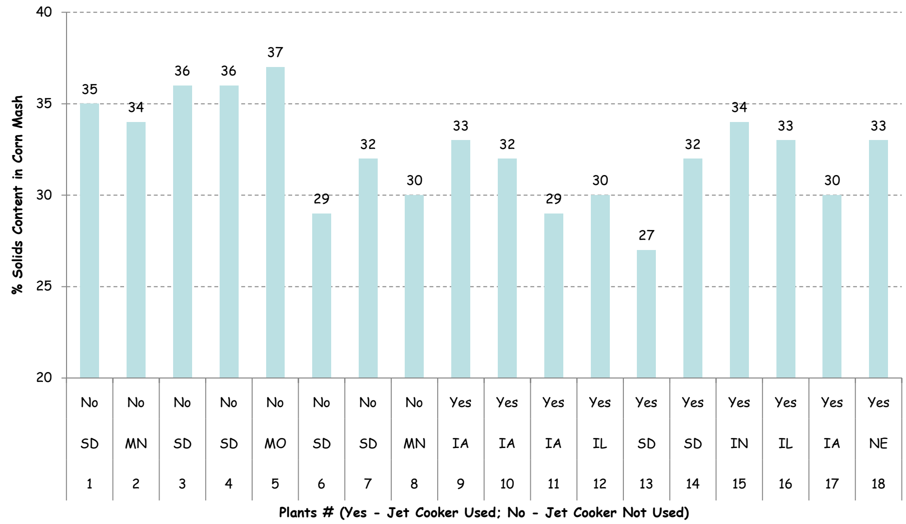
% Solids Content of Corn Slurry

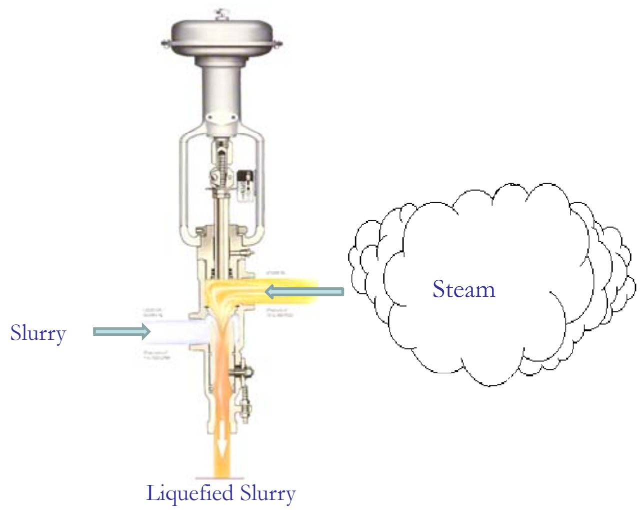


Johal and Deinhammer, Proceeding of Corn Utilization Tech Conf, 2006, p57-61

Liquefaction

- Jet cooked with split dosage
- Non jet cooked with single dosage

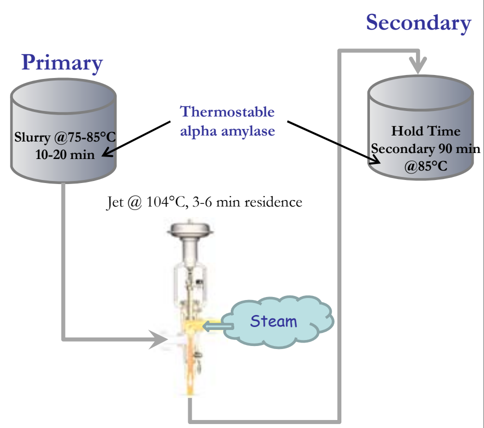
Jet cooking



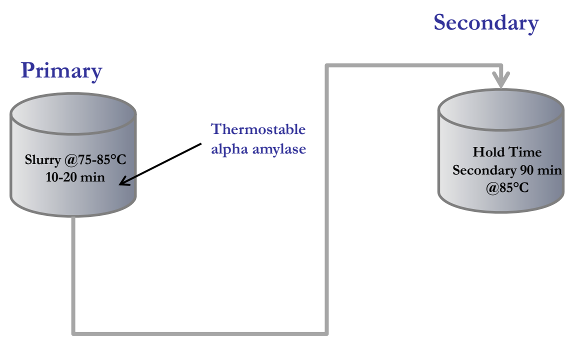
Courtesy: Hydrothermal Corp.

Jet Cooking with Split Dose

- Jet cooking with split dose
  - Primary 107°C for 3-6 minutes
  - Secondary 85°C for 90-120 minutes
    - Slurry Solids 32-35%ds
    - pH ideally 5.6-5.8
    - 30-35% thin stillage
    - Thermostable alpha amylase
    - 12-14 DE and <1500 cP viscosity



Flow Diagram for Liquefaction: Non Jet Cooked with Single Dosage



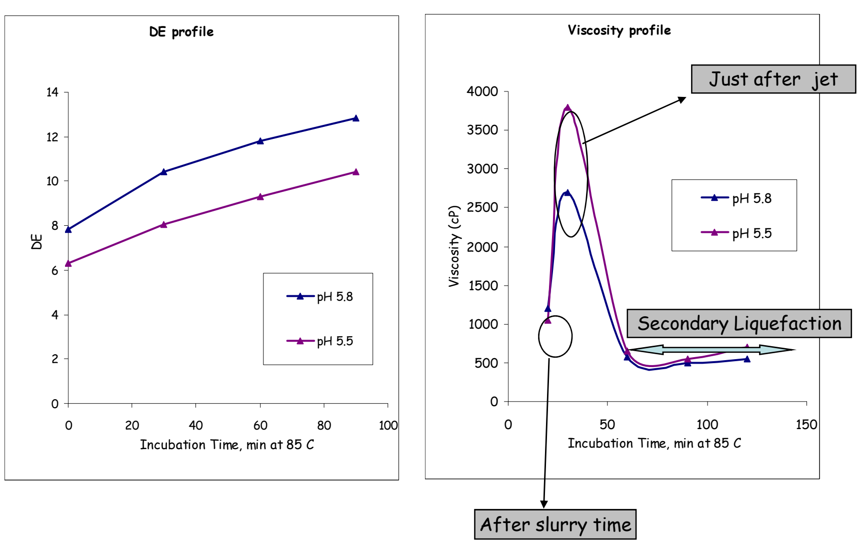
Non Jet Cooking with Single Dose

- Hold at 85°C for 60-120 minutes
  - Slurry Solids 32-35%ds
  - pH ideally 5.6-5.8
  - 30-35% thin stillage
  - Thermostable alpha amylase
  - 12-14 DE and <1500 cP viscosity

Liquefaction Process

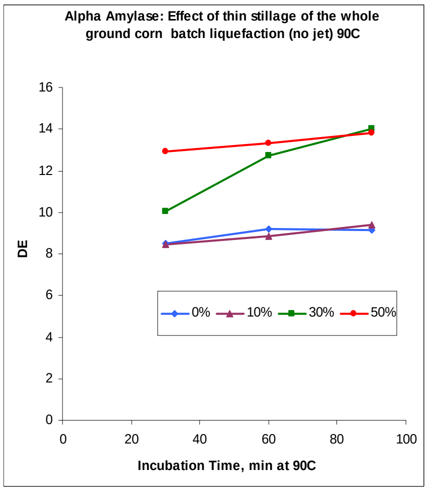
- Parameters
  - DE
    - Measure reducing ends of sugar
  - Viscosity
    - Flowability, thinning

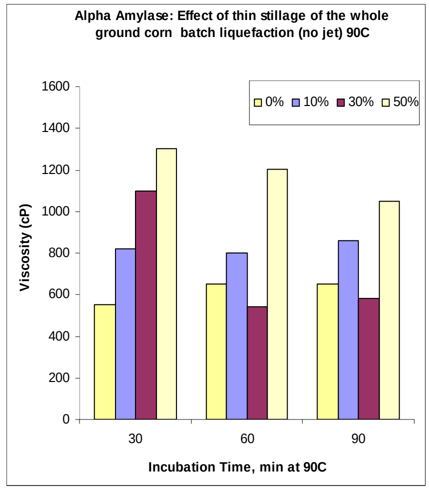
Typical DE and Viscosity Progression



Reference: Vivek Sharma, Genencor International, Dry Grind Ethanol Course, UIUC, 2008

Effect of Thin Stillage on Alpha Amylase



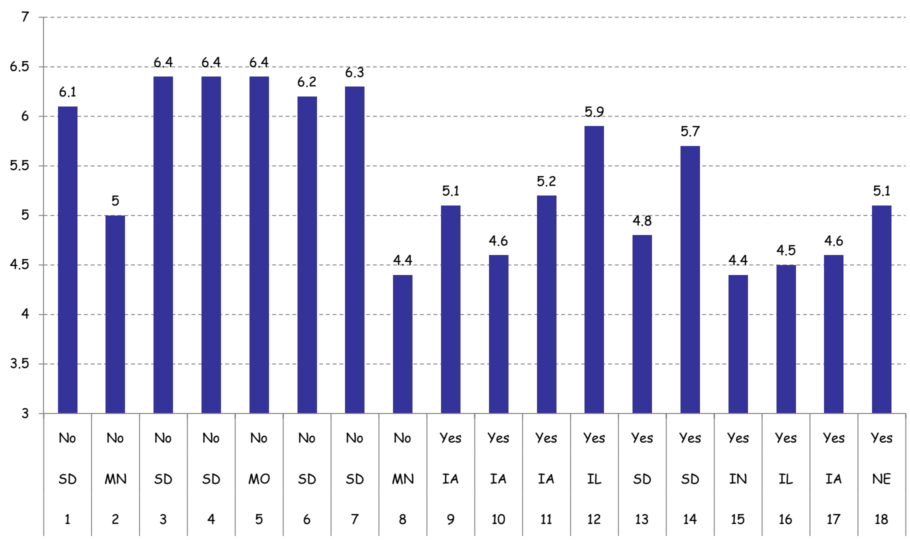


Reference: Vivek Sharma, Genencor International, Dry Grind Ethanol Course, UIUC, 2008

Evaluation of Liquefaction Process

- Iodine test
- DE measurement
- Sugar Profiles

pH of Corn Mash



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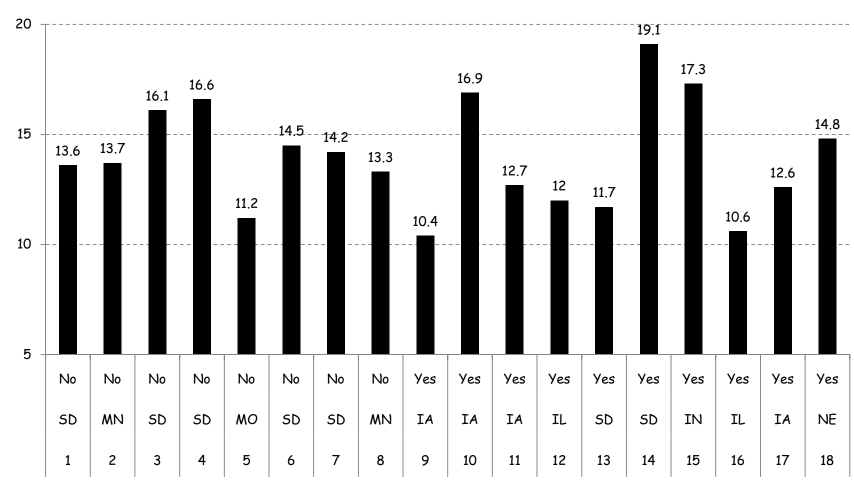
% DE of Corn Mash



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Saccharification/Fermentation Systems

- Complete Saccharification (Continuous and Batch)
- Pre-Saccharification followed by Simultaneous Saccharification/Fermentation
- Simultaneous Saccharification/Fermentation

Complete Saccharification

- Liquefact is hydrolyzed completely prior to starting the fermentation process (Glucose concentration > 93% w/w)
- Typical Operating Conditions and Parameters:
  - Temperature 140 - 144°F (60 - 62°C)
  - pH 4.0 - 4.5
  - Dry Substance 27 - 37%
  - Time 1 - 6 hr
  - Dose 0.08 - 0.4 GAU/g dry solids

Pre-Sacc Followed by SSF

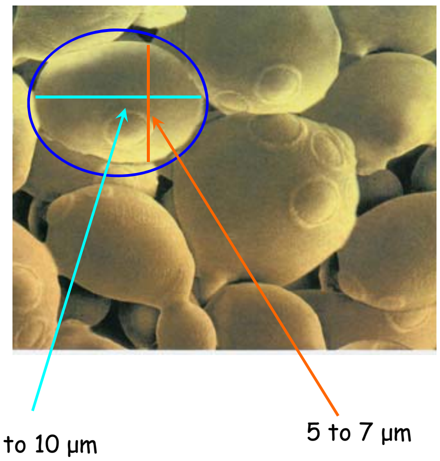
- Liquefact is saccharified to a glucose level 5-25%w/w before being cooled and sent to fermentation.
- Pre-Sacc Typical Operating Conditions and Parameters:
  - Temperature: 140 - 150°F (60 - 66°C)
  - pH: 4.0 - 4.5
  - Dry Substance: 27 - 37%
  - Time (in pre-sacc): 1-24 hours
  - Dose: 0.13- 0.45 GAU/g dry solids

Simultaneous Saccharification/Fermentation (SSF)

- Saccharification to glucose and fermentation occur simultaneously
- Typical Operating Conditions and Parameters
  - Temperature: 86 - 92°F (30 - 33°C)
  - pH: 4.0 – 5.5
  - Dry Substance: 27 - 37%
  - Time: 30 - 70 hr
  - GA Dose: 0.3 - 0.6 GAU/g dry solids

Saccharomyces Cerevisiae

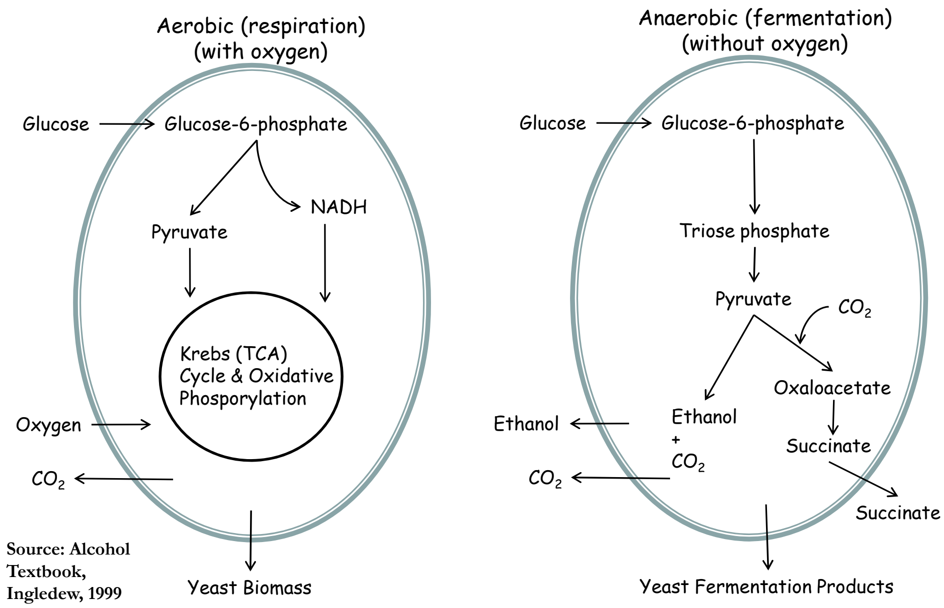
- View of typical fermentation yeast with microscope
- Yeast are identified partially by appearance
- Also identified by list of compounds used (sugars, vitamins, etc.)



Chemical Composition of Yeast

- Yeast cell is 75% water and 25% dry matter
  - Carbohydrate 18-44%
  - Protein 36-60%
  - Nucleic Acids 4-8%
  - Lipids 4-7%
  - Total inorganics 6-10%
    - Phosphorus 1-3%
    - Potassium 1-3%
    - Sulfur 0.4%
  - Vitamins Trace amounts

Glucose Uptake by Yeast

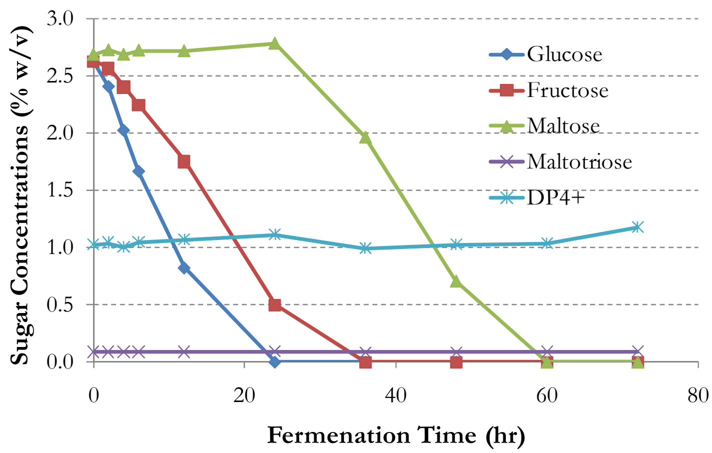


Source: Alcohol Textbook, Ingledew, 1999

Glucose Uptake by Yeast

- Yeast can grow in presence or absence of oxygen
- When O_2 is present and not too much sugar is present
  - Yeast use all sugar for growth
- When O_2 is not present and amount of sugar present is high
  - Yeast produces ethanol
- Yeast cell mass produced under anaerobic conditions is less than 5% of the weight of initial sugar

Preferential Sugar Utilization by Yeast



Fermentation

- Fermentation is the process of yeast converting glucose to ethanol and carbon dioxide in presence of oxygen
  - Process is moderated by several enzymes and coenzymes
  - Simplified equation

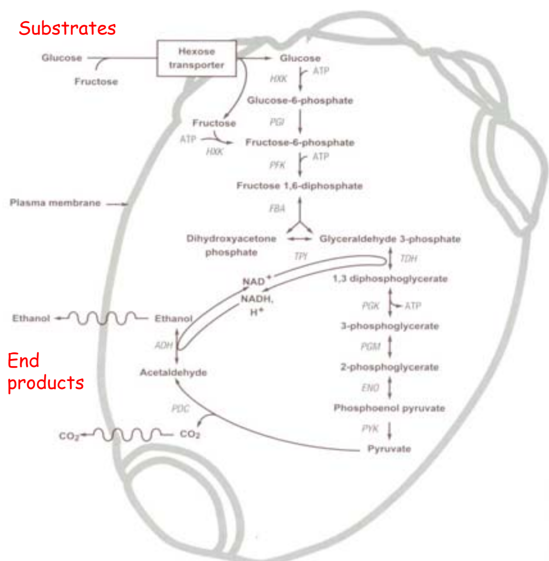
$$H(C_6H_{10}O_5)_nOH \xrightarrow[\text{water}]{\text{amylolytic enzymes}} n\,C_6H_{12}O_6 \xrightarrow{\text{yeast}} 2n\,CH_3CH_2OH + 2n\,CO_2$$

Starch	Glucose	Ethanol	Carbon dioxide
MW = 162	MW = 180	MW = 2 x 46	MW = 2 x 44

Calculate Amount of Ethanol Produced

- How many kg of starch is used to produce 150 kg of ethanol?
  - 150/0.511 = 293.54 kg glucose
  - 293.54/1.111 = 264.21 kg starch

How Yeast Makes Ethanol



Glycolysis, the degradation of sugar to alcohol with production of energy (ATP) for cell growth

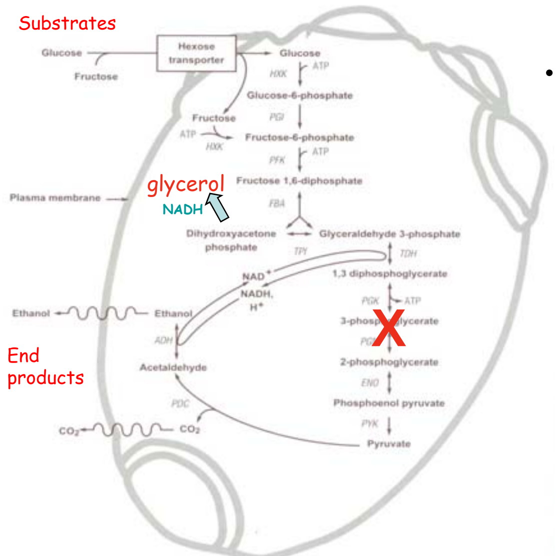
12 Enzymes convert ~90% of all produced glucose to ethanol and CO_2 under conditions of controlled oxidation/reduction balance

Source: Alcohol Textbook, Ingledew, 2009

Ethanol Production: Gycolysis Pathway

- Ethanol production is proportional to growth
- As sugar is used, energy (ATP) and pyruvic acid is made. The energy is used for cell production (growth), and the pyruvic acid is used as a sink for the reduced enzyme cofactor made in glycolysis which otherwise would accumulate
- Growing cells make ethanol 33 times faster than non-growing
- Pyruvic acid is converted through acetaldehyde to ethanol using reduced NADH + H+ and regenerating NAD
- Glycerol is made due to the accumulation of excess reducing power

How Yeast Makes Ethanol



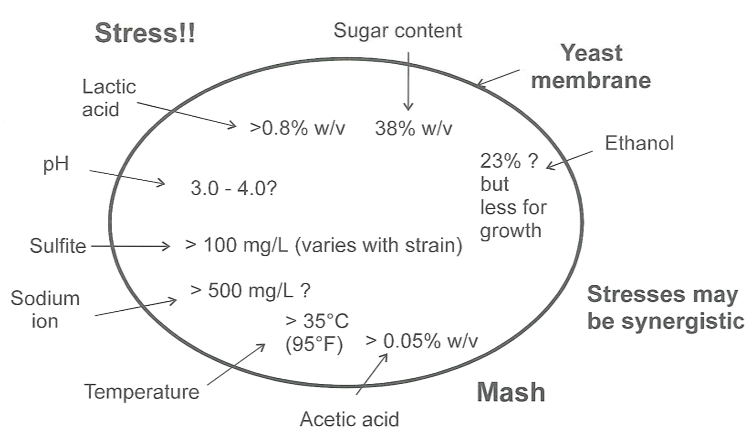
- When a few molecules of these are taken by the cell for growth, this leads to increases in NADH and lower ethanol (not noticed)

Source: Alcohol Textbook, Ingledew, 2009

Differences in Yeast

- Yeast vary in maximum temperature for growth from about 25 to 39°C
- Yeast vary in morphology, size of 5 to 10 microns, roundness, chain formation
- Yeast vary in production of cogeners, especially esters and higher alcohols
- Yeast vary in growth response
- Yeast characteristics are checked by lab fermentations

Yeast Stress



Source: Alcohol Textbook, Ingledew, 1999

Osmotic Stress

- Due to high sugar concentration
- Increase in glycogen synthesis
- Increase in glycerol synthesis (instead of ethanol)
- If starch is left in form of dextrins and slowly converted to glucose by enzymes, osmotic pressure will be lowered

Loss of Viability in Stressed Yeast

- Always seems to occur in same three stages no matter what the cause
  - Loss of ability to form new cells
  - Loss of membrane integrity
  - Loss of intracellular enzymes able to chemically reduce many chemicals

Signs of Yeast Aging

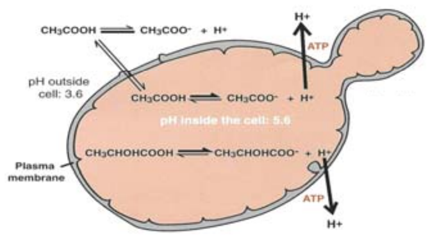
- Increase in bud scar number
- Cell size
- Surface wrinkles
- Granularity
- Daughter cell being retained

Tests for Viability

- Viable yeast count
- Methylene blue viability test
  - Dead cells lose ability to keep methylene blue outside
  - Dead cells lose ability to reduce the dye to a colorless form very quickly
- Ability to form new cells on a thin layer of agar medium on a microscope slide is considered the reference method
- Flourescein diacetate dyes measure intracellular enzymes and give a good viability measure

Antinutritive Factors: Weak Acids

- Weak acids can have inhibitory effects on yeast if the free, unionized acid can enter the yeast cell
- Acids which enter the cell in the unionized form may ionize at the higher pH inside the cell
- Acetic acid is a potent anti yeast factor



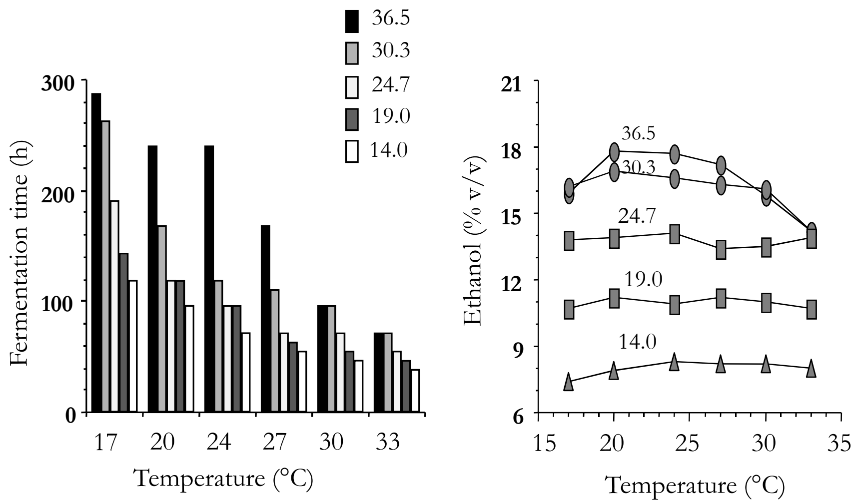
Source: Narendranath et al. 2001. Journal of the Am. Soc. of Brewing Chemists 59:187-194
Narendranath et al. 2001. Journal of Ind. Microbiology and Biotechnology 26:171-177

Nitrogen Requirements

- Amount of urea used
  - 8 mM (480 mg urea/L) to 16 mM urea (960 mg urea/L)
- Amount used depends upon "gravity" of mash (solids content)
- Virtually none remains after fermentation

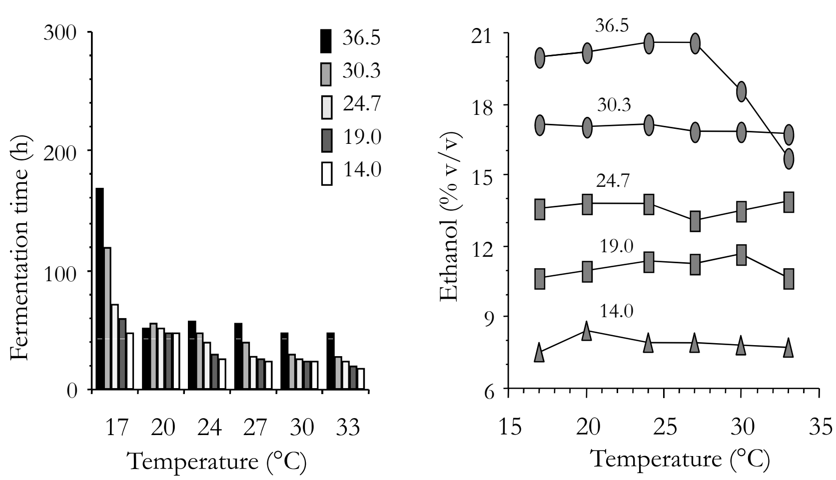
NH_2
|
C=O
|
NH_2

No Urea Nitrogen Added



Source: Alcohol Textbook, Ingledew, 2009

Urea Nitrogen Added



Source: Alcohol Textbook, Ingledew, 2009

Fermentation

- Continuous
- Batch

Continuous Fermentation

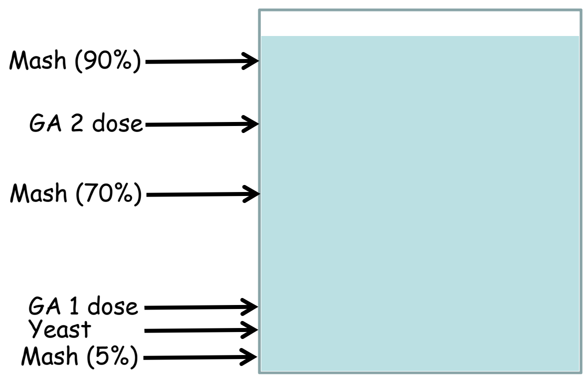
- Fresh substrate added and product removed at the same rate
- Reduce time required for filling, emptying and cleaning
- Simplify Control
- Operate continuously for several months without constant shutdown, cleaning and decontamination
- In reality many problems with contamination have been observed
  - Source of problem difficult to locate

Batch Fermentation

- Each fermentor is individually controlled
- Each fermentor is processed, cleaned and restarted with new batch of yeast
- Additional tanks are needed to maintain productivity
- Most of dry grind ethanol continuous fermentation plants have converted to batch fermentation

Yeast Propagation Tank

Typical Fermentor Fill



Fermentation: Factors Affecting Ethanol Yield

- Losses in Mashing
- Cell growth
- Minor end products
- Infections
- Struck Fermentation
- Nutritional Deficiencies

Factors Affecting Ethanol Yield

- Losses in Mashing
  - Unconverted starch (due to poor milling)
  - Retrogradation of starch
  - Starch blinding by protein
  - Less than complete enzymatic hydrolysis
  - Starch found in DDGS (indicator of losses in mashing)
  - Use of heat damaged grains

Factors Affecting Ethanol Yield

- Higher Alcohols (Fusel Oils)
  - N-propanol
  - Amyl Alcohol
  - Iosamyl Alcohol
  - Iosbutanol
  - Phenethyl Alcohol
- These can be formed from amino acids
- Yeast strain, high temperature, increased aeration increased agitation and composition of medium leads to production of higher alcohols

Factors Affecting Ethanol Yield

- Infections
  - Recycle streams provide nitrogen and nutrient source but also cause infections
  - Wet milling process
    - Light steep water is used in fermentation
  - Dry grind process
    - Backset/Thinstillage is used in fermentation
  - Major reasons of use of recycle steams is for water recycling/pollution control
    - However, inhibitory levels of sodium ions, sulfite ions, lactic acid and acetic acid lead to yeast stress

Struck (Sluggish) Fermentations

- Rate of sugar utilization becomes extremely slow (especially towards the end of fermentation)
- Caused due to nutritional deficiency especially nitrogen and oxygen

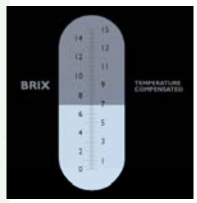
Very High Gravity Fermentation

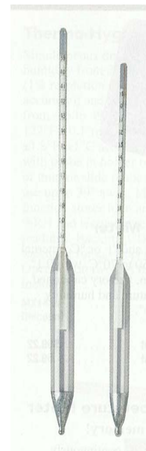
- Higher concentration of solids (glucose) in fermentation broth
- Yeast nutrition and oxygen play an important role
- In lab final ethanol concentration as high as 23% have been achieved in fermentation broth
- More ethanol produced per batch
- Less evaporation and dehydration required
  - Less energy usage

Specific Gravity Measurement

- °Brix
- °Balling
- °Plato

Instruments Used for Measuring Specific Gravity





Refractometers

Hydrometers

Percent Alcohol Determination

- Proof of Alcohol is calculated by multiplying “alcohol volume %” by 2
  - Example: 40% alcohol by volume = 80 degree proof

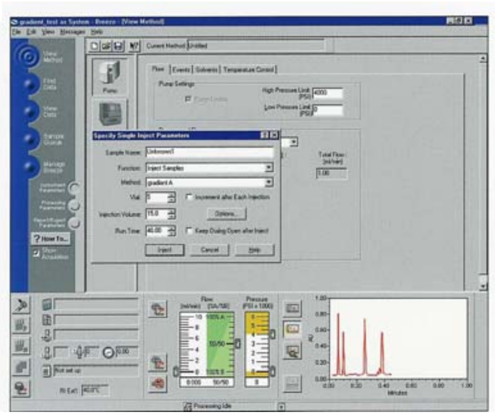
Ethanol Yield

- Ethanol producers like to look at gallons of ethanol produced from a bushel of corn
  – Generic weight of bushel of corn = 56 lb
  – Moisture content in corn = 14%
  – Starch content is 70%
  – Assuming 100% of starch is converted into ethanol
    - Ethanol yield will be 2.9 gallons/bushel
  – Typically yield is around 2.65 gallons/bushel (in dry grind ethanol plant)
    - In terms of efficiency of starch conversion, this would be 100 x 2.65/2.9 = 90% efficiency

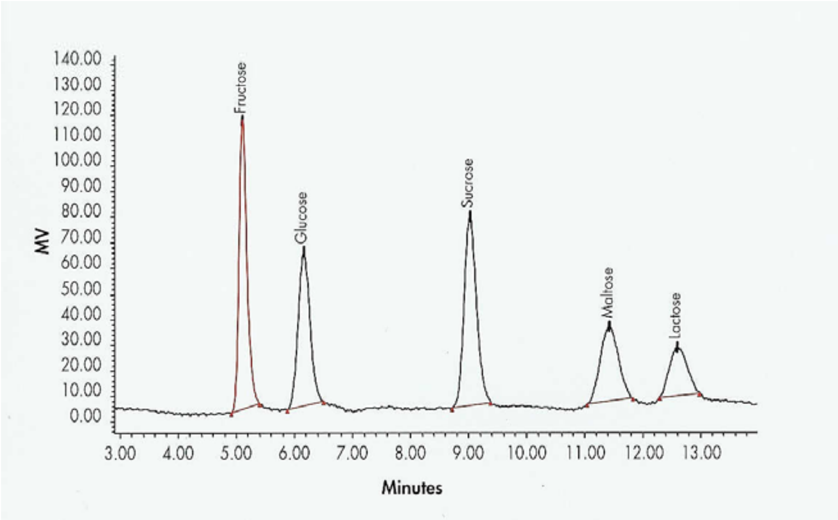
Monitoring Rate of Fermentation

- Alcohol level at a given time using HPCL
- Time to a repeat Brix using refractometer
- Rate of pH change and final levels using pH meter
- Volumetric Productivity

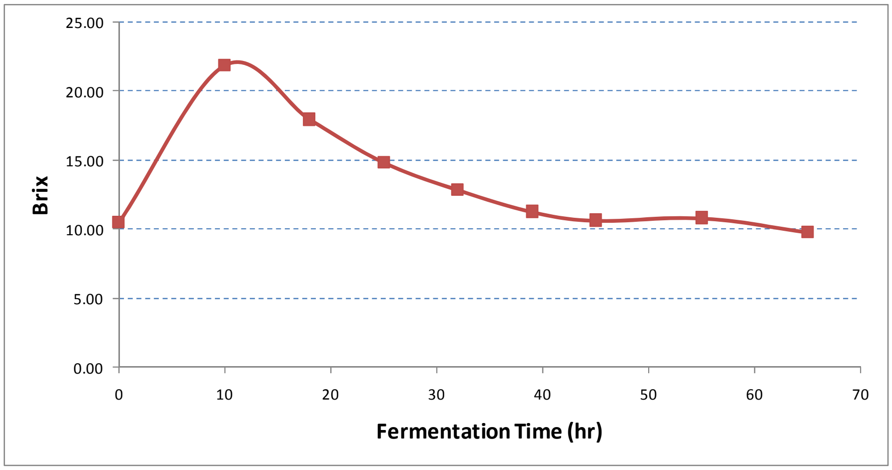
Monitoring Rate of Fermentation



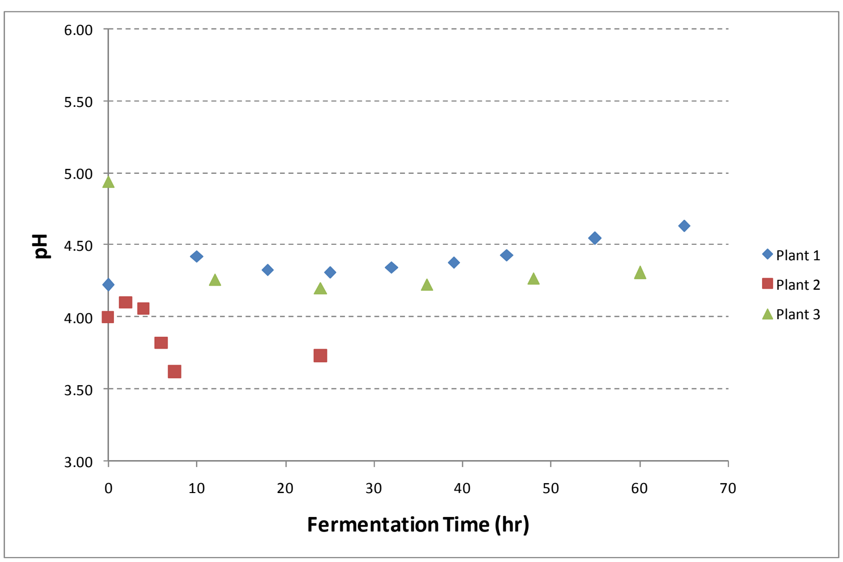
Monitoring Rate of Fermentation HPLC Chromatogram



Monitoring Brix During Fermentation



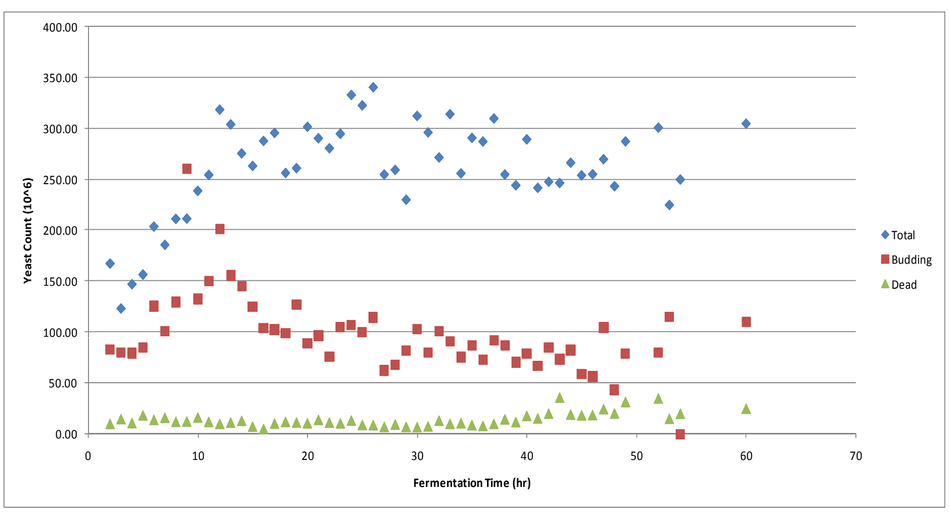
Monitoring pH During Fermentation



Monitoring Yeast Performance

- Cell Count
- Viability
- Budding Cells

Cell Count



ILLINOIS