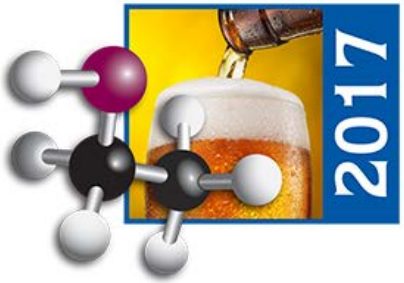


ASBC Annual Meeting

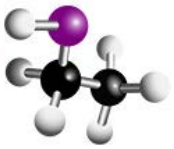
June 4–7 ■ Fort Myers, Florida

See what SCIENCE can brew for you

Brewer's Yeast Contribution to Flavor

Sylvie Van Zandycke, Ph.D.

Lallemand Brewing



Content

- Yeast Metabolism and Flavor Production
- Yeast Strains and Beer Styles
- Non-Traditional Yeast to Brew Beer

Contribution To Beer Flavor

Hops

Grist

Liquor

Contribution to Beer Flavor

Yeast?

Fermentation Parameter

Yeast Strain

Original Gravity

Yeast Viability and Vitality

Aeration

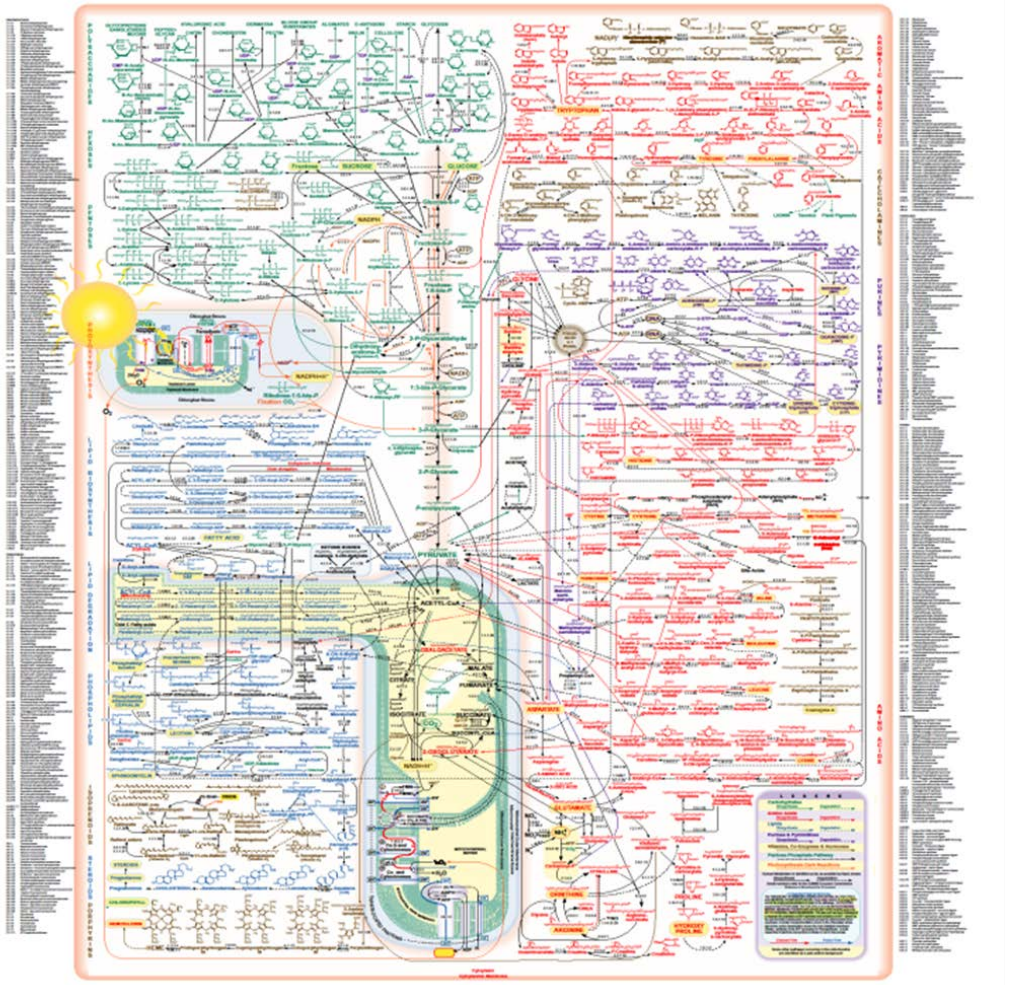
Yeast Nutrition

CO_2 Pressure

Fermentation Temperature

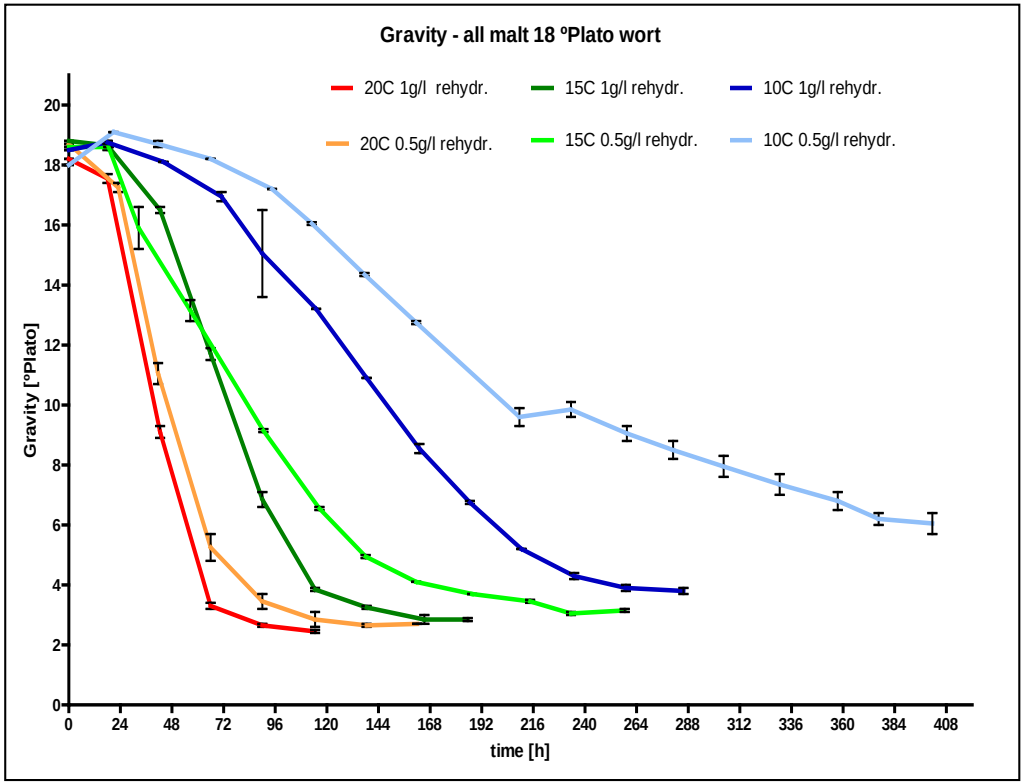
Geometry of fermentation vessel (convection)

Yeast Metabolism



Variables Affecting Fermentation

- Wort composition & pH
- Wort dissolved oxygen (DO)
- Yeast strain
- Yeast pitch rate
- Yeast quality
- Temperature
- Pressure
- Vessel geometry



Yeast in Fermentation

WORT

(sugars, amino acids, etc.)

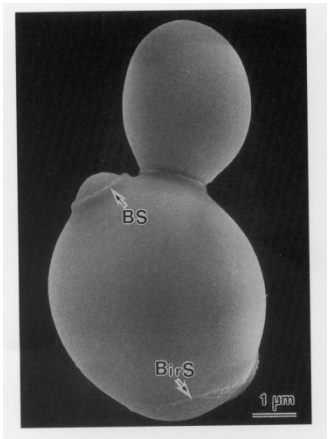
More yeast!

Yeast autolytic compounds

Phenols

Sulphur compounds

Aldehydes & ketones



Esters

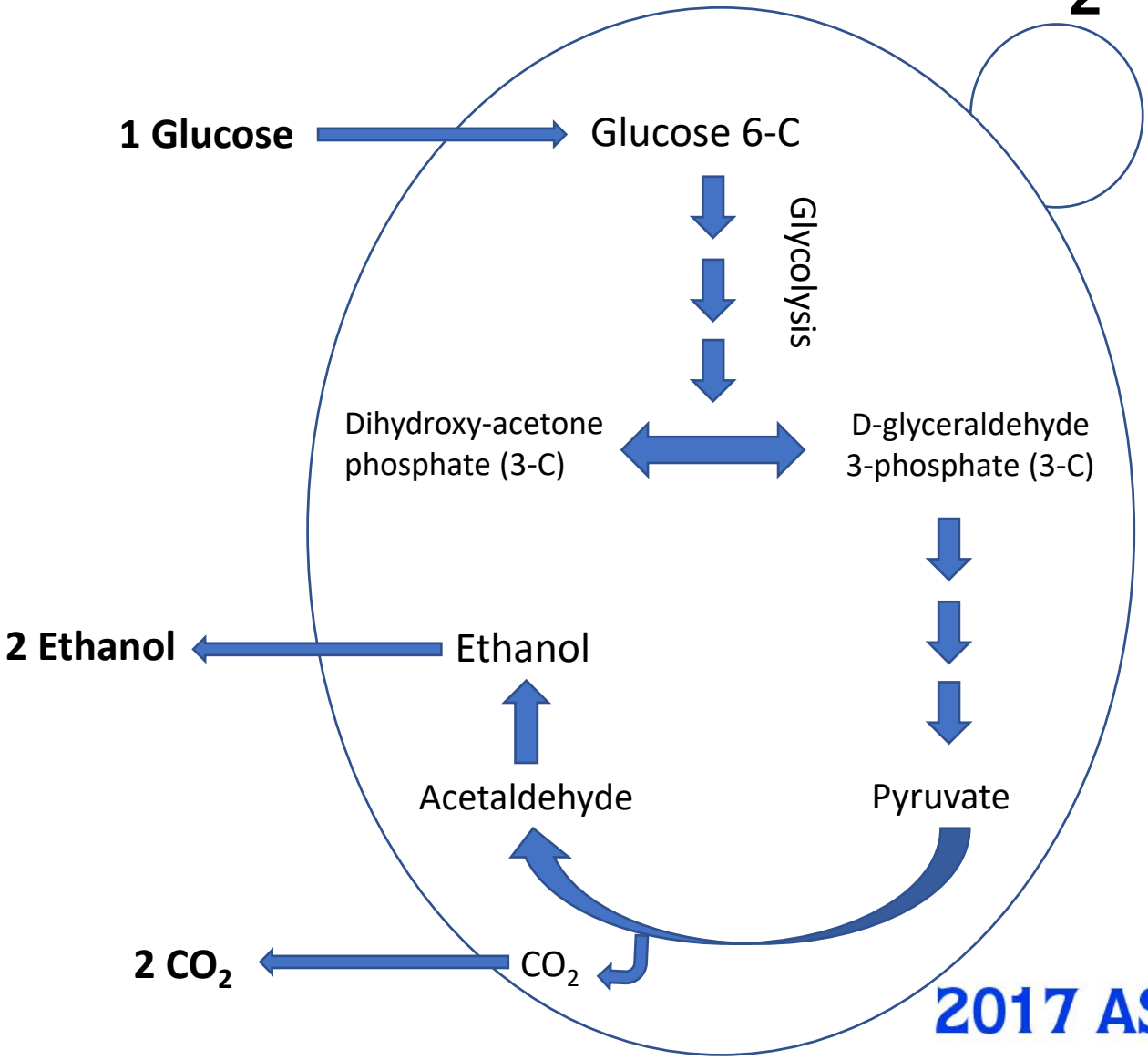
Acids (organic & fatty)

Alcohols (ethanol & higher alcohols)

CO_2

Glycerol

Ethanol & CO_2



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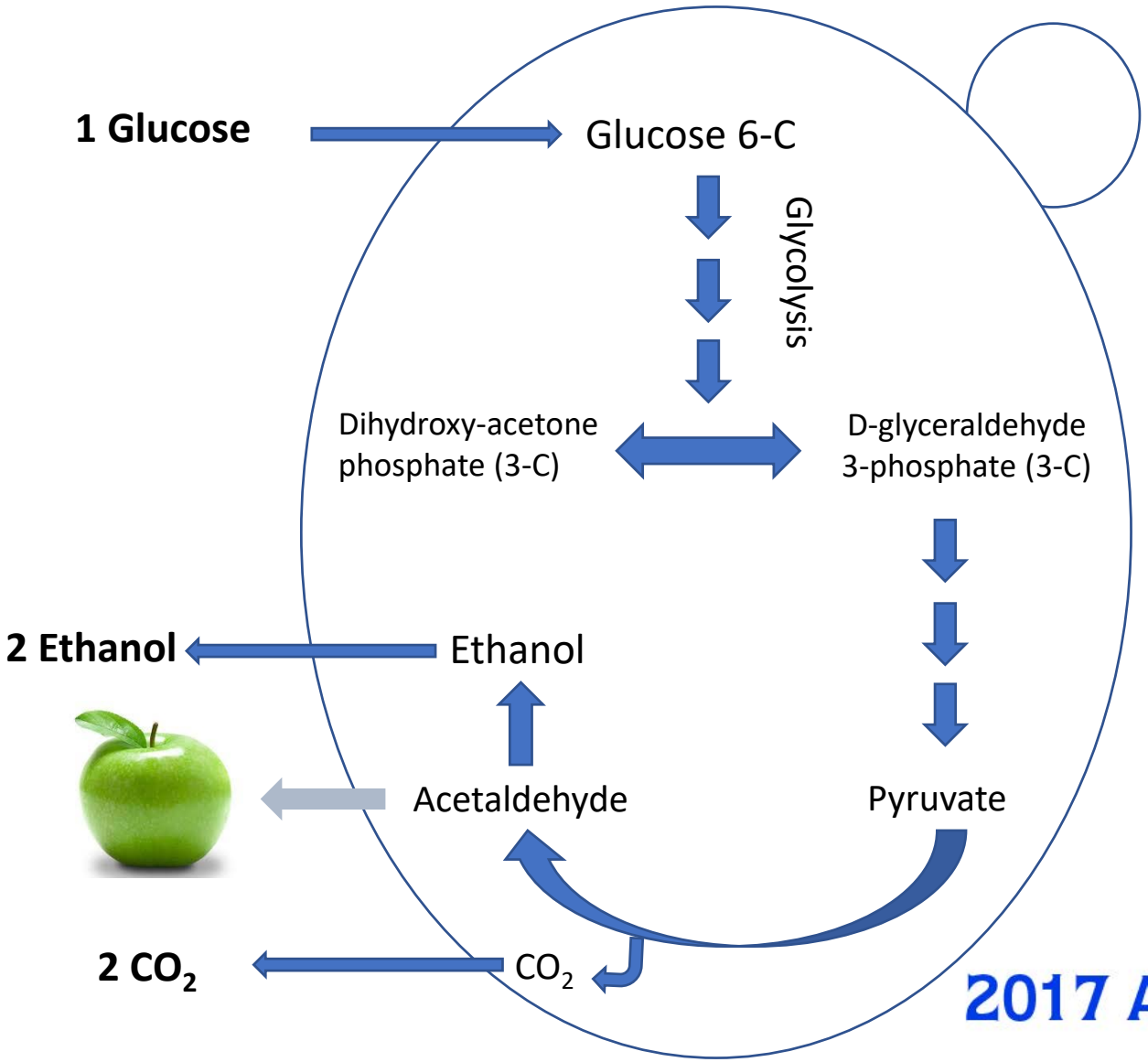
Acetaldehyde



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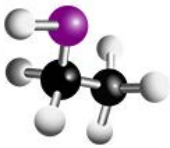

Acetaldehyde

- Healthy yeast -
- No CO_2 back-pressure -
- Over oxygenation +
- High pitching rate +
- Very high fermentation temperature -

Higher Alcohols

Catabolic pathway

Amino acids → keto acids → aldehyde + CO_2

Anabolic pathway

Glucose → pyruvate → acetaldehyde + CO_2

Higher alcohol

Ethanol

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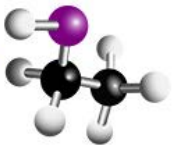
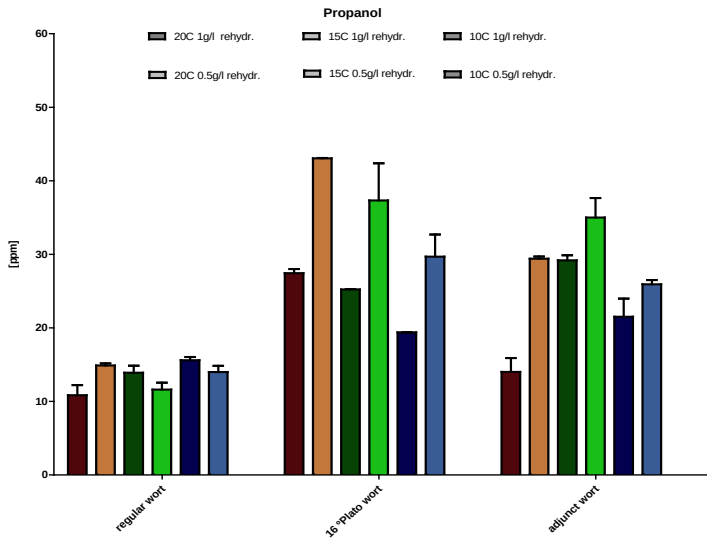
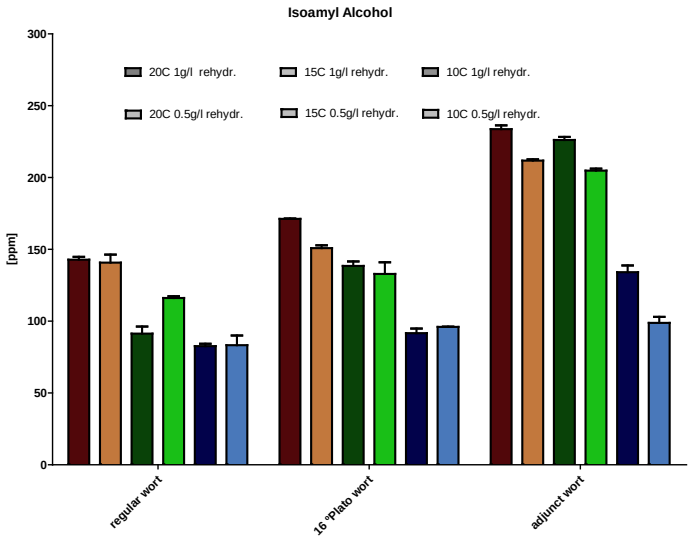
Amino Acid Uptake by Brewing Yeast

- GROUP A (fast)
  Glu, Asp, Asn, Gln, Ser, Thre, Lys, Arg
- GROUP B (intermediate)
  Val, Met, Leu, Isoleu, His
- GROUP C (slow)
  Gly, Phe, Tyr, Try, Ala, NH_3
- GROUP D (little or no)
  Pro



Higher Alcohols





- Higher gravity and higher temperature resulted in increased higher alcohol concentrations

Control of Higher Alcohols

- Good FAN supply -
- Flocculent yeast -
- Wort Aeration +
- Strong fermentation movement +
- High fermentation temperatures +
- Application of pressure -
- Yeast strain +-

Esters arise from the reaction of an alcohol and an acyl-CoA molecule

Esters

Glucose

↓

Pyruvate

↓

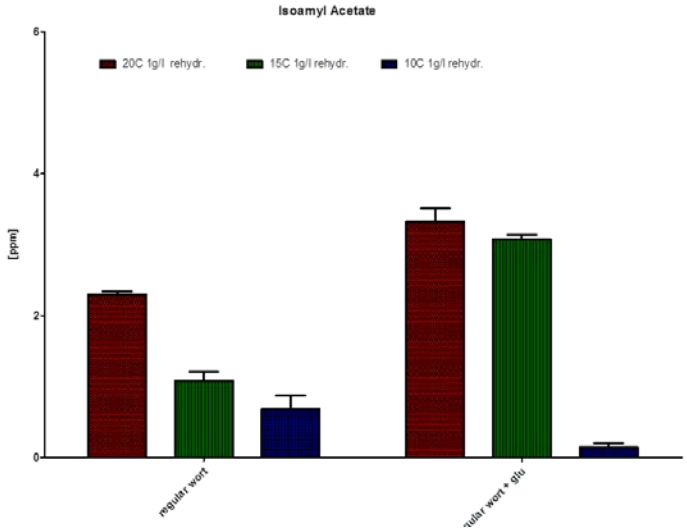
Esters ← **Acetyl CoA** → **TCA Cycle**

↓

Lipids
Amino Acids
Nucleic Acids

Control of Esters

- Increase glucose concentration in the wort:
  - adding 10g/l of sterile glucose solution to the wort
  - follow a specific mashing process to increase the ratio of glu



Michael Eder, New Brewer 2009

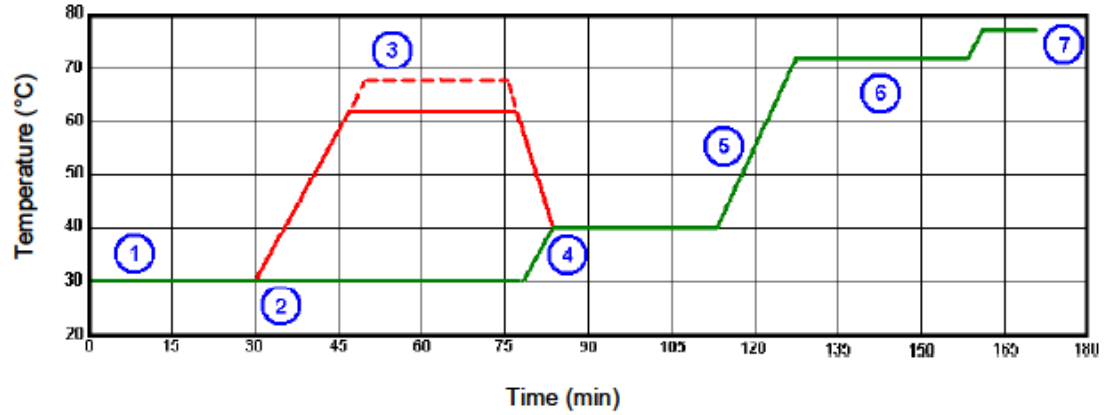
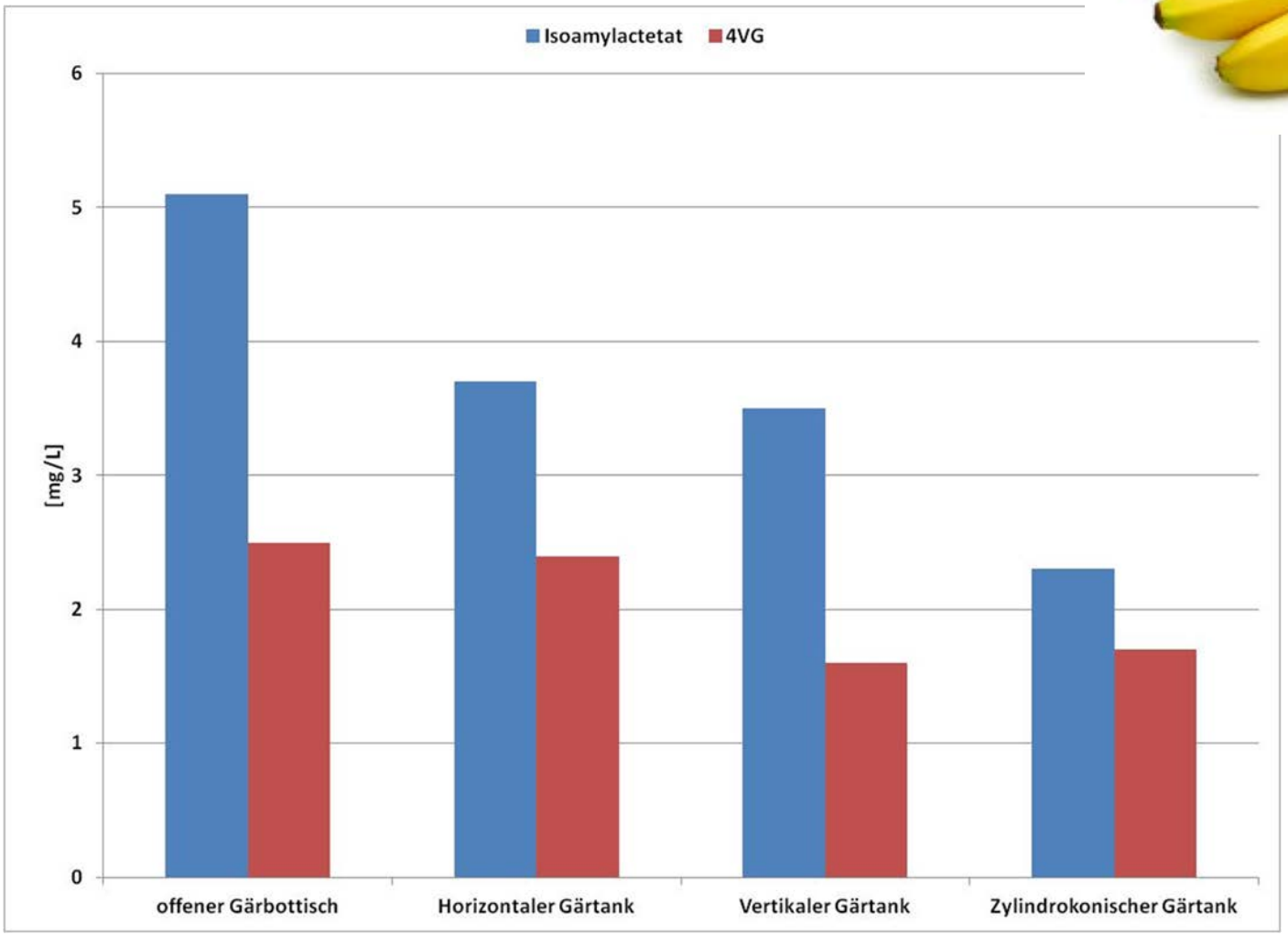


Figure 1: 1-Mashing with water/grist ratio of 5:1 2-Mash separation (25-30% of thick mash and 70-75% of thin mash) 3-Heating of the thick mash to 63° C and 30 min rest to activate ß-amylase 4-Mixing of both thin and thick mashes to achieve a temperature of 40° C and activate maltase 5-Heating to 72° C to perform iodine reaction 6-Heating to 78° C before transfer to lauter tun.

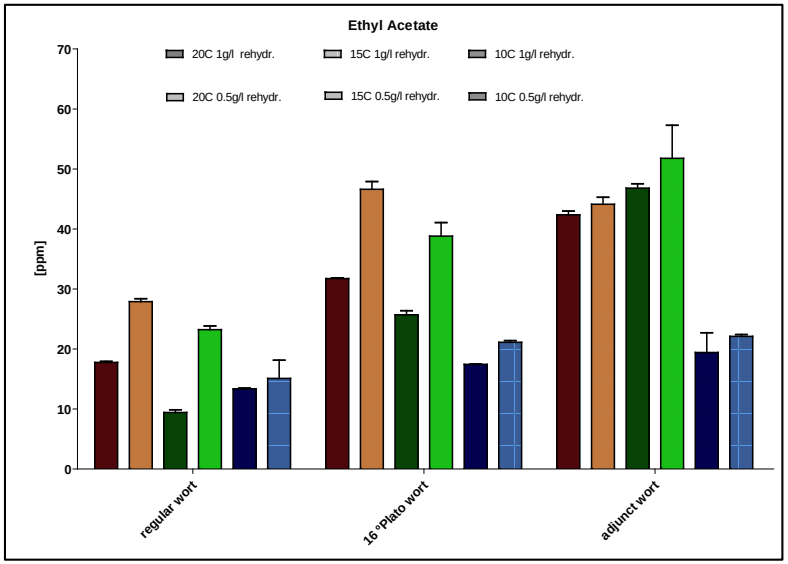
Control of Esters

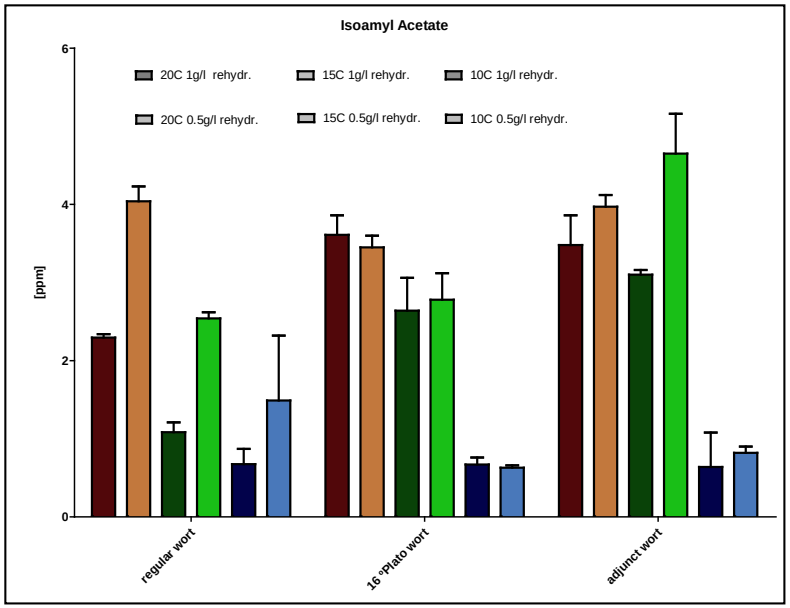
Fermentation Vessel



Back, W. Ausgewählte Kapitel der Brautechnologie, Fachverlag Hans Carl, Nürnberg, 2005.

Esters





- Lower pitching rate resulted in higher ester production
- Higher temperature led to higher ester production
- Higher gravity resulted in higher ester concentrations

Control of Esters

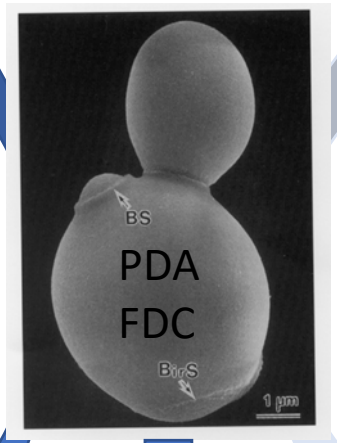
- High original gravity +
- Increased biomass production -
- High glucose concentration +
- Vigorous fermentation -
- Pressure during fermentation -
- High Fermentation temperatures +
- High concentration of unsaturated fatty acids -
- Wort aeration +-
- Yeast strain +-

Phenols

Ferulic Acid

Coumaric Acid

Cinnamic Acid



4-Vinylphenol
(Brettanomyces)

4-Vinylguajacol

Vinyl benzene (Styrene)

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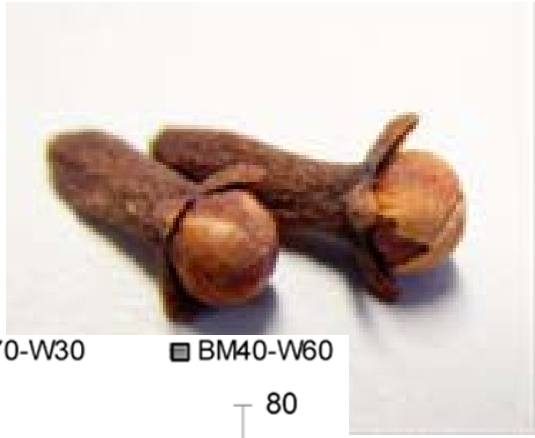
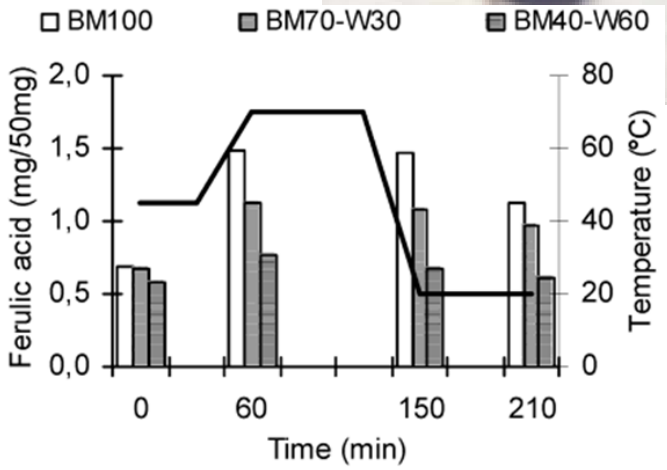
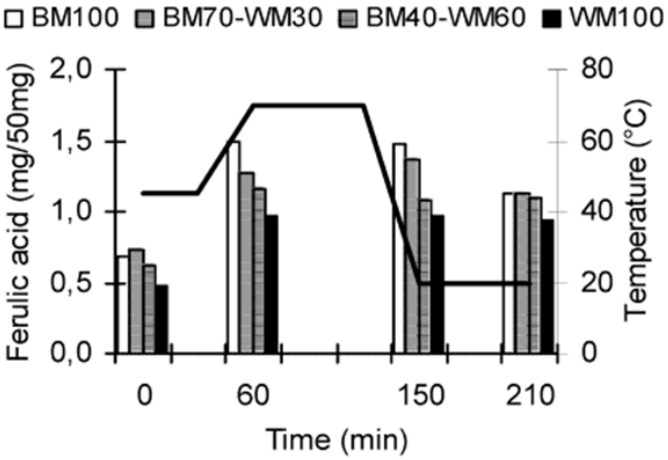
4VG (Barley : Wheat Ratio) Fermentation



- In general contains wheat (0.05-0.06%) more ferulic acid than barley (0.04-0.06%)
- Better ferulic acid extraction from barley than from wheat
  - higher Feruloylesterase- and Xylanase activity (Arabinoxylan break down)
  - Wheat contains proteins, that can inhibit xylanases





COGHE, S. : Ferulic Acid Release and 4-Vinylguaiacol Formation during Brewing and Fermentation: Indications for Feruloyl Esterase Activity in *Saccharomyces cerevisiae*, J. Agric. Food Chem. 2004,

Control of 4-Vinyl Guaiacol

- Mash in at 30-45 ºC +
- Malt ratio barley : wheat +-
- Fermentation vessel (open fermenters) +
- Use the appropriate yeast strain +-

Organic Acids

pyruvate

Acetyl-CoA

CoA

oxaloacetate

citrate

isocitrate

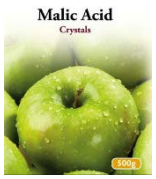
α-ketogluterate

Succinyl-CoA

succinate

fumerate

malate



bitter

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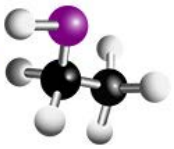
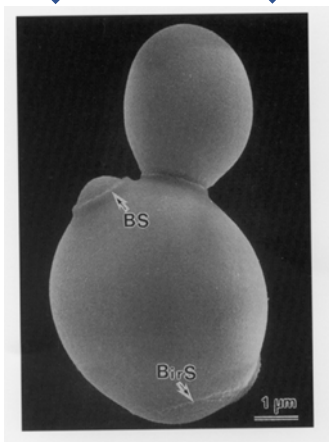


Control Organic Acids

- Healthy yeast -
- Interruption of TCA cycle +
- Yeast strain +-

Fatty Acids

Palmitic (16:0) Stearic (18:0) Oleic (18:1) Linoleic (18:2)



Caproic (6:0) Caprylic (8:0) Capric (10:0)

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Fatty acids

- Fatty acids are bad for foam

- Short chain
  fatty acids (C8-C14)

Control fatty acids

- Increase yeast growth +
  - Wort oxygenation +
  - High lipid content in wort +
  - High temperature +
  - High pitching rate +
- Yeast health
  - Autolysis +

Sulfur

Production

- Intermediates in amino acid metabolism
- When yeast needs to make sulfur containing amino acids

H_2S (Hydrogen Sulphide)

SO_2 (Sulfur dioxide)

Control of SO_2

- High oxygen -
- High OG +
- High lipid concentration -
- Poor yeast health +
- Pantothenic acid -
- Yeast strain +-

Control of H_2S

- Vigorous fermentation (CO_2⬆) -
- Poor yeast health +
- Pantothenic acid -
- Addition of Serine -
- Yeast strain +-

Manipulation Of Variables

A painter's pallet

Lager Yeast

- Bavarian origin.
  - 1400s in Munich - cool fermentations (selective pressure)
  - Taken to Pilsen and Copenhagen in 1840s
- Became very popular - displaced ale yeast
- Popularity fueled by advances of Industrial Revolution
  - Steam power, refrigeration, railroads, pasteurization and filtration technology
- Cool fermentation temperatures: 5 to 12 ºC
- Natural Hybrid

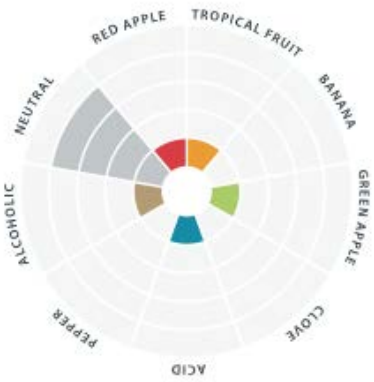
Characteristics of Lager Beer

- Strains are closely related - common origins
- Beers are more delicate, clean, drinkable, and less aromatic.
- Low bitterness, simple grist composition.

Lager Yeast

Lager	Pilsner	Helles	Vienna	Bock
Schwartzbier	Märzen			

Characteristics of West Coast Ale Yeast



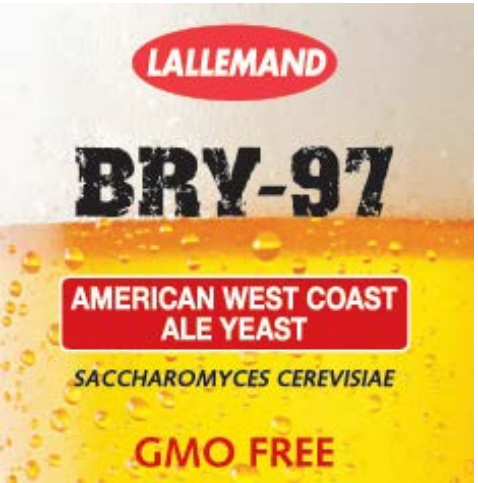
- Aroma: Neutral with a slight ester
- High attenuation
- Fermentation range: 15 – 22°C
- Flocculation: Medium to High
- Popular modern style

Characteristics of West Coast Ale

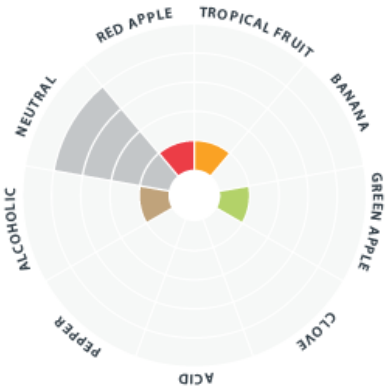
- 4.5 - 5.5% abv
- Straw like golden to deep amber colour
- Complex malty, bready/biscuity
- Moderate/strong USA hops, citrus & pine
- Medium bodied, moderate/high carbonation

American West Coast Ale Yeast

American Barleywine	American Pale Ale	American Amber Ale	American Brown Ale	American IPA
American Wheat	Blonde Ale	Cream Ale	Kölsch	Imperial IPA
Irish Red Ale	ESB	Scottish Ale	Strong Scottish Ale	Strong Ale



Characteristics of English Ale Yeast



- Flavor: moderate ester, well balanced
- Attenuation: Medium
- Fermentation temp: 18-22°C
- Flocculation: Medium - High

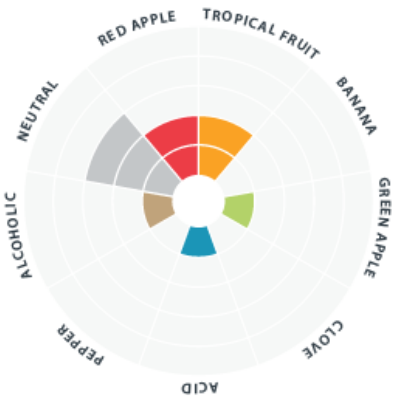
Characteristics of English Ale (ESB)

- Strong (5-6% abv),
- Full-bodied, mahogany-colored
- Mellow bitterness
- Complex malty notes- biscuit flavors and soft malt toffee, brewed with Pale Ale and Crystal malts

English Ale

ESB	Cream Ale	Blonde Ale	Bitter	Special/Best Bitter
Scottish Ale	Irish Red Ale	Brown Ale	Porter	Sweet Stout
English IPA	Old Ale	Mild		

Characteristics of British-Style Beer Yeast



AROMA

fruity, estery

ATTENUATION

medium

FERMENTATION RANGE

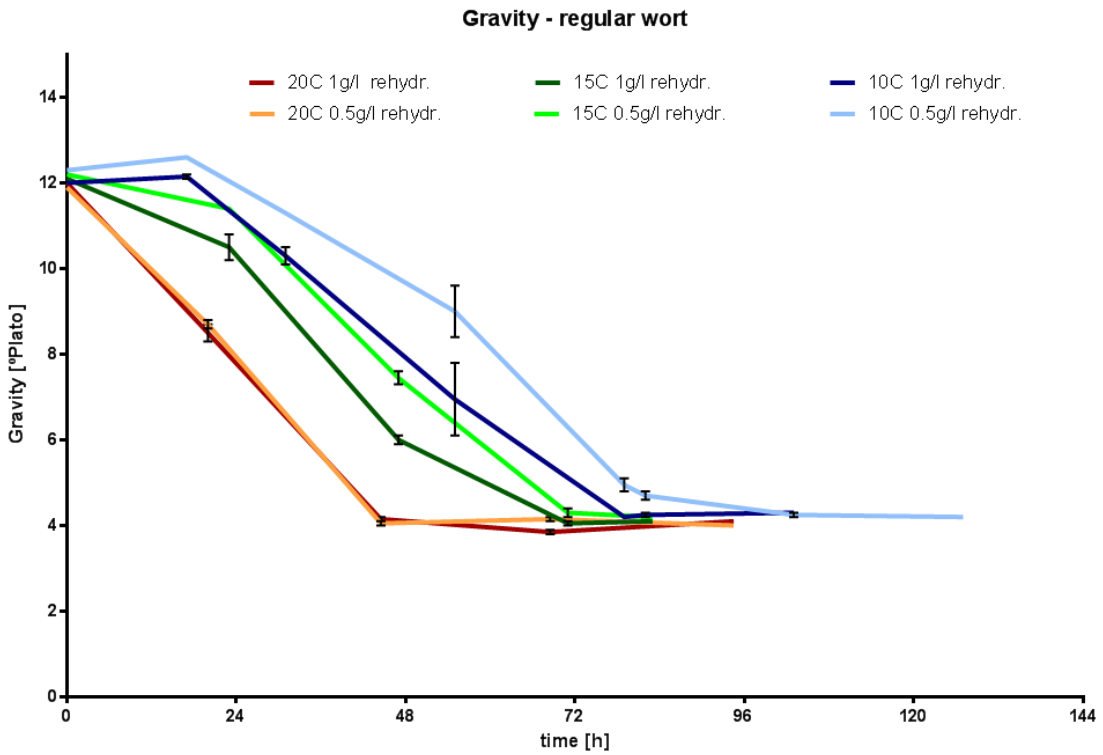
15 - 22°C (59 - 72°F)

FLOCCULATION

low

ALCOHOL TOLERANCE

9% ABV



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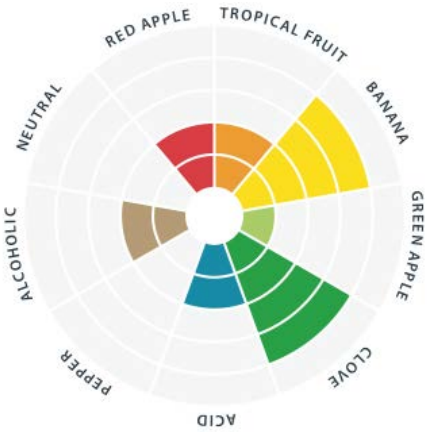
Characteristic of Sweet Stout

- Dark brown to black color
- Aroma of coffee, chocolate, cacao, low hoppiness
- Flavor of roasted grain with chocolate and hop bitterness moderate
- Creamy head
- Also known as milk/cream stout
- Sweetness

British-Style Beer Yeast

Mild Ale	Cream Ale	American style Hefeweizen	American style Wheat Ale	English style Pale Ale
Scottish style Ale	Amber Ale	Red Ale	Strong Scotch Ale	English style Brown Ale
Porter	Sweet Stout	Cream Stout	New England IPA	

Beer Styles Based around Yeast: Weissbier/Hefeweizen



- Origins in C16th Bavaria.
- Favored by royals, later gaining widespread popularity.
- Nearly dies out by C19th but revived by G.Schneider.
- Top fermenting ale yeast, fermented at warm temps.

Characteristcs of Bavarian Wheat Beers

- Unique yeast with complex flavour profile.
- Prominent esters – Banana, vanillia, bubblegum, apple.
- Phenols often present – 4VG (clove, ferulic acid), spices.
- Low hopping, simple grist composition.

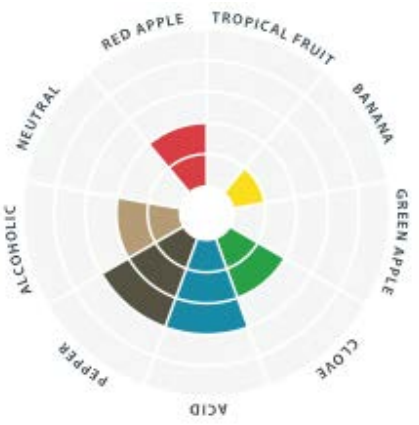
Wheat Beer Yeast

Weizen	Hefeweizen	Dunkelweizen	Weizenbock	American Style Hefeweizen

Beer Styles Based around Yeast: Saison

- French/Belgian origins (Wallonia).
- Farm house ales traditionally brewed in winter, stored until summer.
- Seasonal farm workers ‘Saisonniers”.
- Top fermenting, warm temps.

Characteristics of Saison Beers

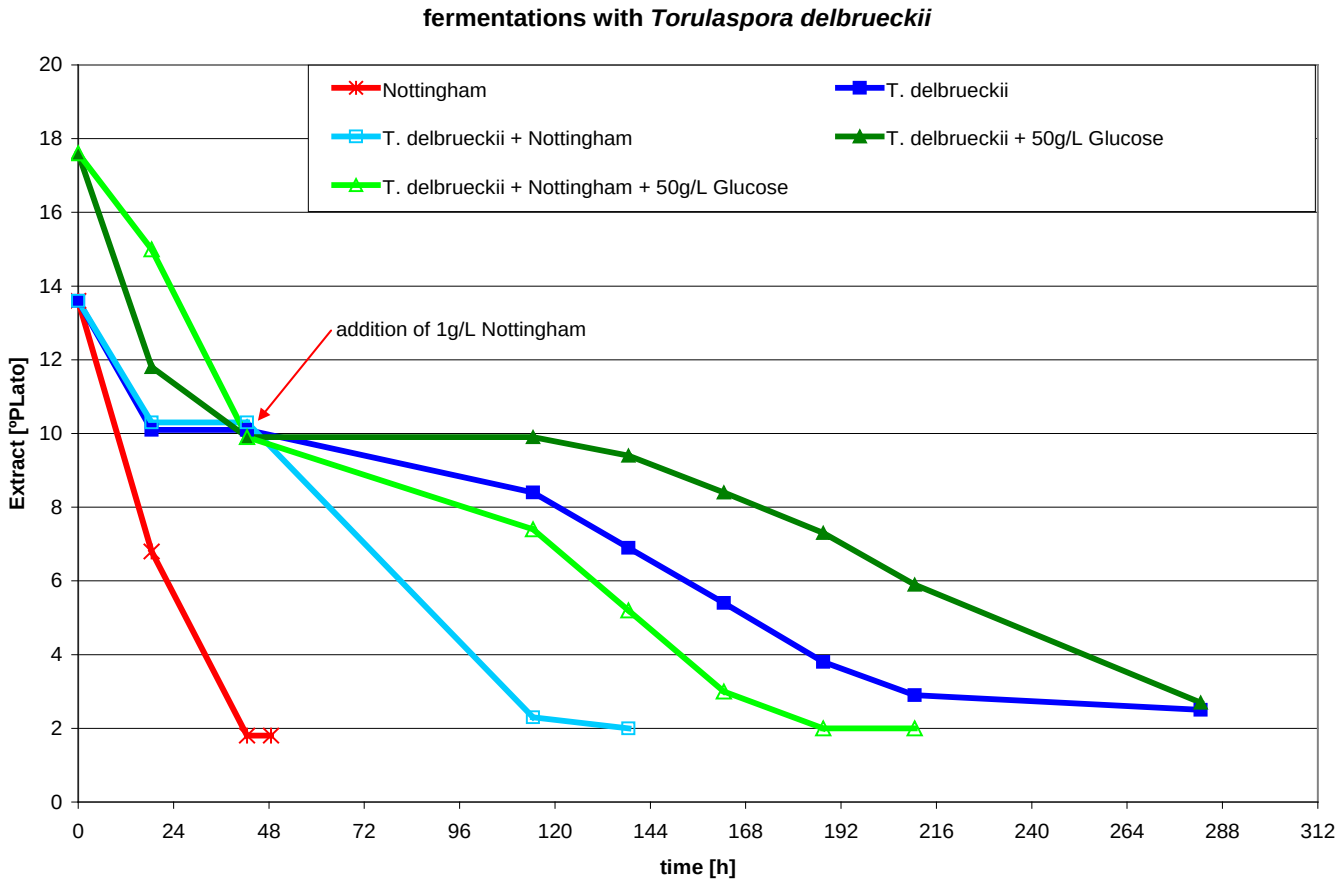


- Robust yeast with complex ester flavor profile (lemon/orange).
- Prominent ‘earthy’ yeast notes and spices (pepper).
- Very dry finish; high attenuation.
- Low hopping, simple grist composition.

Belgian Saison-Style Beer yeast

Saison	Bière de Garde	Belgian style beers		

Mixed Strains – *Torulaspora delbruckeii* + *Saccharomyces cerevisiae*

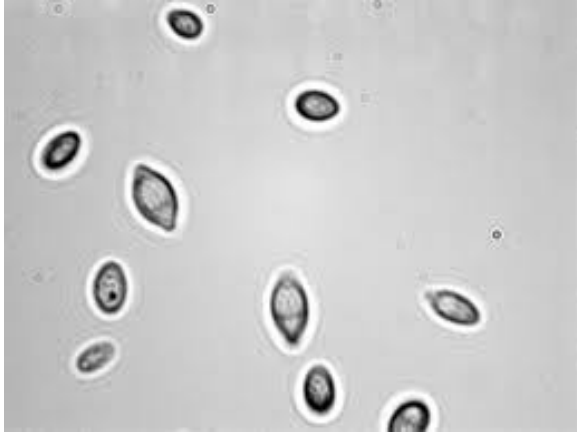


Non *Saccharomyces* strains

- *Torulaspora delbrueckii*
  - reduce volatile acidity in high-sugar fermentations
  - complexity and floral/fruity aroma
  - Bioflavoring or fermentation
  - Michel *et al* 2016

- *Candida zemplinina*
  - Osmotolerant
  - Reduces acetic acid, increases glycerol
  - Good fermentation
  - Estela-Escalante *et al* 2016

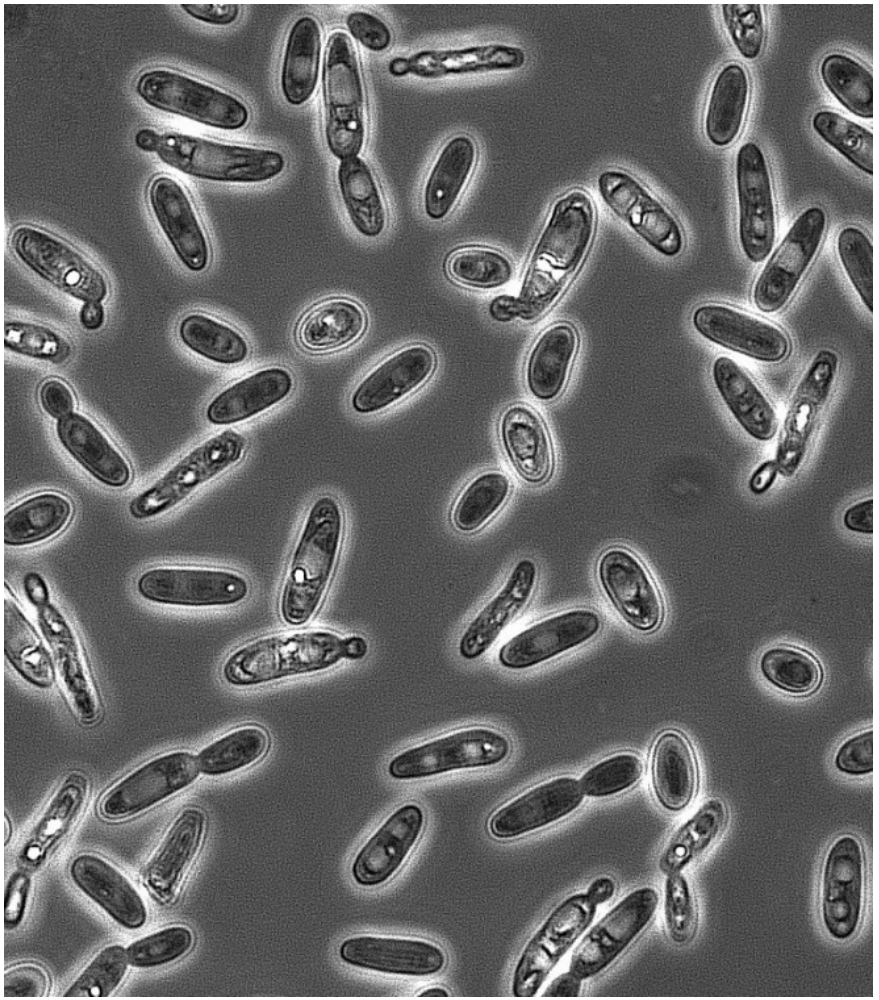
Non *Saccharomyces* Strains



- *Saccharomycodes ludwigii*
  - Non alcoholic beers
  - Only use glucose
  - Michel *et al*, 2016

Wild Yeast: Brettanomyces

- Can utilise broad range of sugars (inc. dextrins)
- Diverse sub species
- Does not contribute a lot of acidity on its own
- Phenolic, fruity
- Slow acting
- Secondary Fermentation

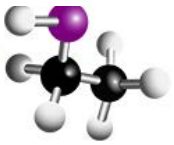


Yeast Hybrids

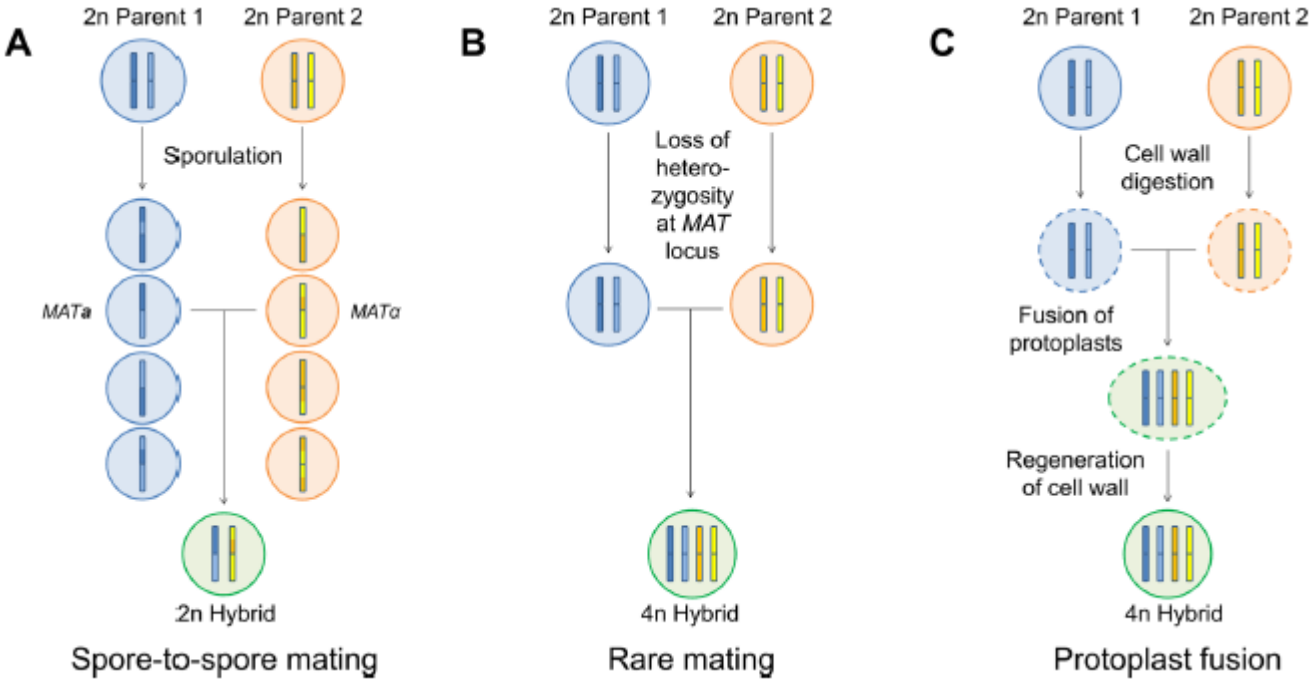
Table 1 A summary of studies published since the year 2000 investigating the use of de novo yeast hybrids in beer fermentation

Parental strains		Key results	Reference
S. cerevisiae ale strain	*S. cerevisiae* sake strain	The hybrid had an increased fermentation rate and produced increased concentrations of certain aroma compounds	Mukai et al. 2001
S. cerevisiae ale strain	*S. cerevisiae* strain (syn *S. cerevisiae* var. *diastaticus*)	Hybrids had higher attenuation levels (i.e., utilized a higher ratio of the original wort carbohydrates) and ethanol yield than the brewing parent strain	Choi et al. 2002
S. cerevisiae ale strain	Cold-tolerant *S. bayanus* strain	Hybrids had greater fermentation rates than the ale parent in low temperature wort fermentations	Sato et al. 2002
S. cerevisiae ale strain	Saaz-type *S. pastorianus* strain	Hybrids showed improved osmo- and temperature tolerance and fermentation performance compared to the lager parent strain	Garcia Sanchez et al. 2012
Various *S. cerevisiae* ale, bakery, sake, and wine strains		Hybrids with higher acetate ester formation than the parent strains were attained. Best-parent heterosis with regards to aroma formation was more common in outbred hybrids than in inbred hybrids	Steensels et al. 2014
S. cerevisiae laboratory strain	*S. eubayanus* type strain	The hybrid had improved sugar utilization and fermentation rate compared to the parent strains in synthetic wort	Hebly et al. 2015
S. cerevisiae ale strain	*S. eubayanus* type strain	Hybrids exhibited increased fermentation rates and aroma compound formation compared to parent strains	Krogerus et al. 2015
Various *S. cerevisiae* ale and wine strains	*S. eubayanus*	Hybrids produced a greater diversity of aroma compounds compared to traditional lager yeast and parent strains	Mertens et al. 2015
S. cerevisiae ale strain	*S. eubayanus* type strain	Hybrids exhibited increased fermentation rates and aroma compound formation compared to parent strains. Fermentation performance and aroma formation of the hybrids increased with ploidy. The aroma profile of de novo lager yeast hybrids can be controlled based on the relative contribution of parental DNA	Krogerus et al. 2016

Krogerus *et al*, 2016



How to Create Hybrids?

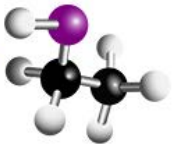


Krogerus *et al*, 2016

Yeast Hybrids

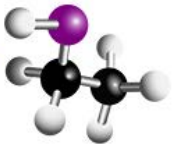
- Greater fermentation efficiency
  - + Fermentation speed
  - + Thermal tolerance
  - + Ethanol tolerance

- Greater diversity in sensory expression
  - + Flavor
  - + Aroma



Are there New Styles to be developed?

- Yes! Beer styles are changing as we speak
- Brewers (and Marketers) like to try new things
- Driven by home-brewers, beer enthusiasts and pro brewers
- Brewers want to sell more beer, want to keep consumers interested in their brands
- Consumers want an experience in drinking, not just beer as a thirst quencher
- Brewers must still bear in mind “drinkability”



Questions?

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- Yeast samples, literature, questions…