INTRODUCTION TO

YEAST & FERMENTATION

A LEARN2BREW PRESENTATION

BY

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Brewing Yeast

Yeast is a single celled fungus and is (or should be) the only living organism that comes into contact with the beer until it is drunk by the consumer.

Yeast acts as a catalyst in converting sugar to alcohol, but does form not part of the end product

Yeasts are found everywhere, in the atmosphere, on the surface of dead and decaying animals and plants.

Large number of yeast species which are adapted to a variety of environments.

Strain which is mainly used in fermentation is *Saccharomyces cerevisiae.*

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Brewing Yeast

1,000 separate strains of the species *Saccharomyces cerevisiae* which include brewing, baking, wine, distilling and laboratory cultures.

The two main types of beer, lager and ale, are fermented with strains belonging to the species *Saccharomyces pastorianus (uvarum, carlsbergensis)* and *Saccharomyces cerevisiae* respectively.

Distinguished on the basis of their ability to ferment the disaccharide melibiose (glucose-galactose).

Strains of *Saccharomyces pastorianus* possess *MEL* gene(s).They produce the extracellular enzyme a-galactosidase (melibiase) and are therefore able to utilise melibiose.

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Yeast Appearance

Yeast under the microscope:

- Bud scars indicate age
- Shape & density of vacuole - development stage
- Size 5 by 8 micron - strain dependent
- Some form chains - linked together
- Can be classified according to its fermentation performance - flocculent (settles out) or non-flocculent (does not settle out so readily)
- Now usually identified by "genetic finger print"

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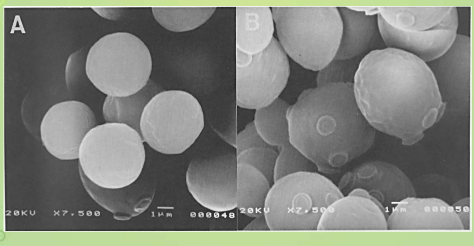
Yeast Under the Electron Microscope



Yeast cells magnified many thousands of times. Shows young yeast on left and old yeast with bud scars on right.

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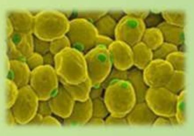
Lager Yeast

Saccharomyces Pastorianus
(*S. Uvarum / S.Carlbergensis*)



- Bottom cropping
- Temperature: 8 to 14˚C
- Slow: 5 to 14 days
- Low in esters
- Malt character dominates
- Traditionally long, cool maturation – 2 months.
- 2 strains: Frohberg and Saaz.

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Ale Yeast

Saccharomyces Cerevisiae

- Top cropping
- Fermentation: 16 to 26˚C
- Fast: 3 to 5 days
- Fruity (‘estery’); may be spicy and complex
- Ready to drink within a week
- 180 to 200 ale strains
- Different:
  - aroma / flavour profiles
  - Fermentation profiles

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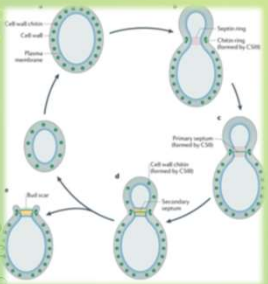
Budding Yeast Reproduction



Yeast cells multiply by budding, producing daughter cells. Up to 30 daughter cells during the cell life time have been recorded.

In a normal fermentation yeast will reproduce itself between 4 and 6 times.

If oxygen is limited it cannot produce sufficient cell membrane material for effective budding.

Insufficient yeast growth will produce a defective fermentation.

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Reproduction by Budding

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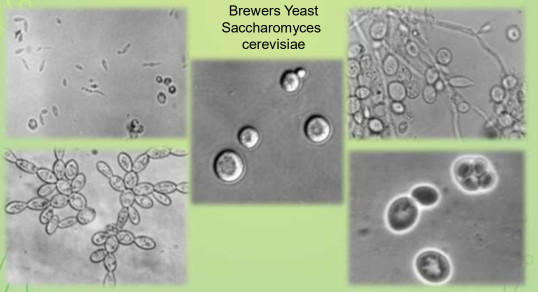
Yeast Forms



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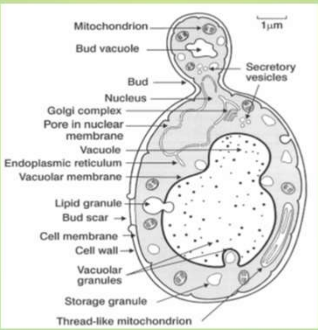
Cell Structure



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Key Cell Components

Yeast cell wall – Comprises mainly 1-3 and 1-6 beta glucans, chitin and mannoproteins. Dictates hydrophobicity, charge and flocculation character. 15-25% of cell mass is in the wall.

Periplasm – This is a small gap between the outer cell wall and inner cell membrane. Contains proteins mainly in the form of enzymes e.g. invertase and phosphatase.

Cell membrane (plasma membrane) – Formed from lipids including sterols, fatty acids, unsaturated fatty acids (UFAs) and glycoproteins.

It acts as a sieve or screen to keep out unwanted extracellular products. It controls the flow and take up of ions, carbohydrates and amino acids. Cell requires molecular oxygen (O_2) for membrane completion and structure.

Restriction of cell budding will result due to lack of sterol and lipid synthesis if O_2 is unavailable or in low levels.

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Key Cell Components

Nucleus – The heart of the cell. Contains the chromosomes and DNA. It controls the growth, development and various metabolic processes within the cell. Also produces mRNA (messenger RNA) that contains the coding for protein synthesis.

Mitochondria – In the simplest terms they are the energy producing centres, they produce ATP and ADP (see below) from pyruvate. They mobilise and utilise the glycogen reserves during the lag phase to produce membrane sterols and fatty acids. Also produce Acetyl-CoA, a molecule important in cell metabolism.

Vacuoles – Vary in size and number throughout the life cycle. They act as repositories for nutrients and also provide a site for large molecule degradation e.g. proteolysis.

Golgi Complex – Appears to regulate protein flow and folding. Also acts as part of an excretion pathway.

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Selecting Yeast for Fermentation

As brewers we require our yeast to;

- Produce alcohol & desirable flavours
- Complete the fermentation
- Produce a consistent product
- Perform fermentation within a set time
- Retain genetic stability
- Be convenient to harvest - flocculation
- Retain viability & vitality during storage
- Reproduce adequately
- Be able to utilise raw materials used
- Withstand acid washing (if required)

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We all Want a Good Fermentation!

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Impact of Yeast Strain

Yeast Metabolic properties:

- Production of flavour compounds: esters, higher alcohols, aldehydes and ketones (VDKs), sulphur compounds.
- Flavour stability: ability to prevent oxidation flavours.

Yeast Physical properties:

- Attenuation ability
- Healthy yeast growth
- Flocculation and sedimentation: time and efficiency of yeast removal, beer filtration, beer loss
- Yeast vitality: autolysis, microbiological stability, flavour and haze

Ability to withstand:

- Lack of nutrients for an extended period of time
- High alcohol content
- High pressure in conditioning tanks
- Low pH

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Nutritional Needs of Yeast

These are provided, usually, by a 100% malt wort:

- Carbon source – sugars (for growth and energy)
- Nitrogen source – amino acids (for protein & enzyme synthesis
- Inorganic ions – Enzyme cofactors (Zn and Mg), pH control, flocculation (Ca)
- Oxygen – production of sterols and fatty acids (cell structure and reproduction)
- Vitamins

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Nutritional Needs of Yeast

Average composition of sweet wort (% solids) is:

- Carbohydrates 90-92%
- Nitrogen compounds 3-6%
- Lipids (fats) 0.2-1.0%
- Tannins (polyphenols) 1.5-2.0%
- Inorganic materials 1.5-2.0%
- Vitamins 0.05%

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Metabolism

Metabolism: Describes the entire set of chemical reactions that organic molecules undergo in living cells.

Metabolism can be further subdivided:

- **Catabolism -** Degradation of energy-rich molecules i.e. glucose
- **Anabolism -** Biosynthesis (creation) of new cell components.

In yeast catabolism, the chemical bonds of glucose are broken, and the cells capture some of the energy stored in these bonds.

Cells store this energy for use in the anabolic reactions that are responsible for growth and development.

Compound that serves as the main storehouse of this cellular energy is called ATP (adenosine triphosphate).

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Respiration vs Fermentation

Aerobic pathway (respiration) provides:

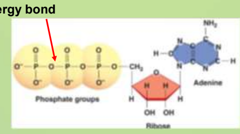
$C_6H_{12}O_6 + 6O_2 \longrightarrow 6CO_2 + 6H_2O$ + Energy (686Kcal) + 38ATP

Anaerobic pathway (fermentation) provides:

$C_6H_{12}O_6 \longrightarrow 2C_2H_5OH + 2CO_2$ +Energy (54Kcal) + 4ATP

ATP (adenosine tri-phosphate) is an energy storage molecule. The bond between the second and third phosphate group is a high energy bond.

High energy bond



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Sugar Uptake

Wort contains: sucrose, fructose, glucose, maltose and maltotriose plus dextrin material.

Brewing yeast strains (ale and lager) capable of utilising these in the above sequence.

Majority of brewing strains leave maltotetraose and other dextrins unfermented.

Saccharomyces diastaticus is able to utilise dextrin material; indicator of wild yeast infection.

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Sugar Uptake

Maltose and maltotriose are major wort sugars: Yeast's ability to use these two sugars is vital and depends upon the correct genetic complement.

Excess glucose (>1% w/v) suppresses maltose uptake.

Sucrose hydrolysis can leave surplus of fructose in finished beer.

Yeast has separate transporter enzymes to carry the two sugars across the cell membrane.

Once inside the cell, both hydrolysed to glucose by a-glucosidase.

Transport, hydrolysis and fermentation of maltose is particularly important in brewing; maltose usually accounts for 50-60% of the fermentable sugar in wort.

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Wort Sugar Profile

Sugar profiles of wort produced at different mashing temperatures can vary greatly due to changes in malt enzyme action.

Wort Sugar Profiles - Percentages of sugar compounds at various mash temperatures (Data after G. Fix et al)

		60°C	65°C	70°C	80°C
Fermentable	**Monosaccharides**	10	9	8	3
Fermentable	**Disaccharides**	61	55	41	15
Fermentable	**Trisaccharide**	9	12	16	30
	Dextrins	20	24	35	52

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Typical Wort Composition (g/l)

Fructose	2.1
Glucose	9.1
Sucrose	2.8
Maltose	52.4
Maltotriose	12.8
Tetraose	3.0
Pentaose	1.0
Hexaose / Heptaose	4.0
Higher dextrins	22.0
Pentosan	0.4
Beta-glucan	<0.5
Soluble protein	2.8
Free amino acids	1.7
Phenolics	0.3
Lipids	0.05

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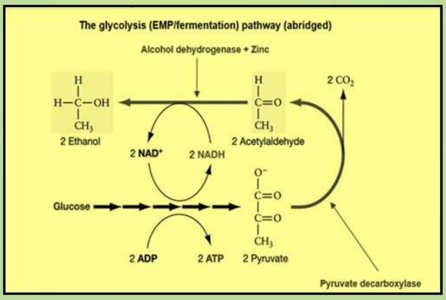
Fermentation Pathway



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The Crabtree Effect

Despite presence of oxygen if the glucose level in the wort (>0.5% w/v) it can direct the yeast to perform the fermentative pathway: this is known as catabolic repression.

This results in the oxygen being utilised for the production of sterols and unsaturated fatty acids (UFAs) which are required for cell membrane synthesis, growth and subsequent cell division and reproduction.

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The Role of Oxygen

- Yeast requires oxygen to synthesise Unsaturated Fatty Acids (UFAs) and sterols to maintain membrane integrity.
- Oxygen requirement is strain dependent
- Rule of thumb: 1ppm of O_2 required per degree Plato of gravity
- Sterol depletion is main limiting step for yeast reproduction
- Wort gravity and temperature affect solubility of O_2
- Aeration will give a max. solubility of O_2 of 8-9ppm in 1040OG wort at 15°C

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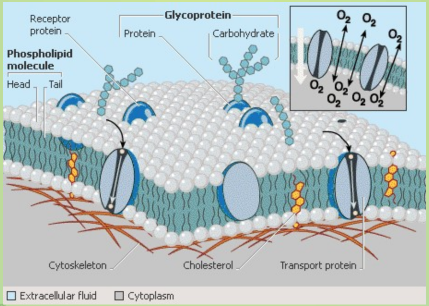
Cell Membrane Structure



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Wort Oxygenation

Factors affecting gas flow required for wort oxygenation:

1. Equipment used to inject/deliver the gas
2. Gas pressure: 3-4 bar?
3. Delivery system: Line length and flow rate, FV height
4. Gas: O_2 is 4.75 times more soluble in wort than air (21% O_2 by volume)
5. O_2 delivered on cold side only i.e. post PHE
6. Wort temperature: Hot or warm wort makes gases less soluble
7. Wort gravity: High gravity makes gases less soluble

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Wort Oxygenation

Gas flow required for wort oxygenation (1048OG & 20°C):

1. Compressed air: 0.34L/hl = 1ppm DO
   2.70L/hl = 8ppm DO
2. Pure O_2: 0.072L/hl = 1ppm DO
   0.570L/hl = 8ppm DO

Calculating gas flow rate example:

Using O_2 via 0.5micron sintered stone (efficiency factor 2.5) in 10BBL (1640 Litres) wort to generate approx. 8ppm DO over 45 minute cooling from copper to FV =

$$\frac{(1640/100)}{45} \times 0.57 \times 2.5 = 0.52\text{L/min of } O_2$$

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YAN & FAN: Nitrogen

YAN -Amino acid + ammonium content of wort
FAN – Free amino acid - proline

- Yeast requires nitrogen for growth. Strain dependent
- Amino acids are building blocks for proteins.
- Yeast prefers to build proteins intracellularly
- Less energy required to produce amino acids than to transport into cell and store from wort
- Typical FAN levels in 1040OG wort: 150-200mg/L
- High FAN levels lead to excessive yeast growth
- Low FAN levels lead to slow or stuck fermentations

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Minerals & Vitamins

- Zinc – Essential as cofactor for many enzymes, in particular alchohol dehydrogenase; final step towards alcohol in fermentation.
- Zinc requirement is strain specific: .045 – 1.10ppm
- Magnesium- Required by the yeast for growth and ATP synthesis. Protects cell from stress.
- Calcium – Metabolic signalling systems
- Potassium, Copper, Manganese
- Vitamins – Biotin, Thiamine, Riboflavin, Choline

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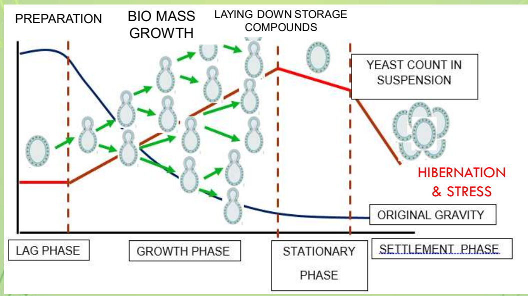
Summary of Yeast Growth



FEAST → FAMINE

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Fermentation Control

Two main aims of control:

1. To produce a beer of consistent quality and flavour that meets the required specification.
2. To ensure that the beer is produced within the required time frame thus ensuring that vessel and equipment utilisation is maximised.

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Factors Affecting Fermentation

- The quality (viability & vitality) of the yeast pitched.
- Pitching rate
- Wort sugar profile
- Oxygenation
- Pressure
- Lack of essential nutrients
- Temperature
- Fermenter design

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Yeast Pitching - Objectives

- Pitch correct yeast strain only
- Maintain microbiological stability
- Achieve target yeast cell count in wort
- Pitch early to minimise risk of infection
- Good dispersion of yeast in wort

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Typical Pitching Rates

Ales pitched at 8-10 million cells per ml

Lagers pitched at 12-15 million cells per ml

Higher rates may be required for high gravity worts (>1060).

Rule of thumb:

1 million cells per ml per degree Plato (4º Sacc)

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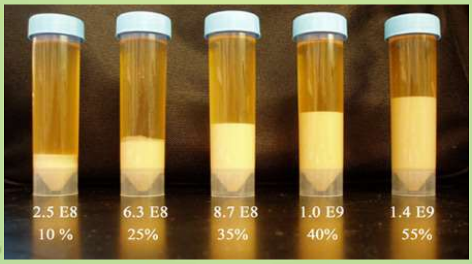
Typical Pitching Rates



Sedimentation Test

Courtesy of Wyeast Labs Inc.

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Calculation of Pitching Rates

As an example say you pitch normally 300g of dense yeast slurry, (50% solids) with 95% viability, per 100 litres of wort. Then for a 15hl FV volume you would pitch:

300g x 15 = 4500g

If the viability were only 90% then you would need to compensate and amend the pitching rate to:

$$\frac{4500 \times 95}{90} = 4750\text{g}$$

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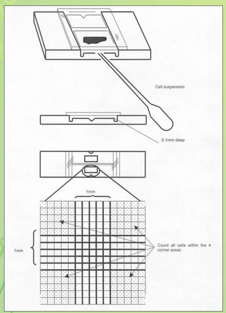
Yeast Pitching: Haemocytometer



Samples are taken and cells counted in a volumetric haemocytometer slide and yeast counts determined.

Viability measured using Methylene Blue stain

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Pitching Rates

Under-pitching can lead to:

- Excess levels of diacetyl
- Increase in higher/fusel alcohol formation
- Increase in ester formation
- Increase in volatile sulphur compounds
- High terminal gravities
- Stuck fermentations
- Increased risk of infection

Over-pitching can lead to:

- High ester production
- Very fast fermentations
- Beer which is thin or lacking body/mouthfeel
- Autolysis (Yeasty/meaty flavours)

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Effect of Temperature

Which is made up from:

- Initial wort temperature before the yeast is added.
- The rate of temperature increase during the growth phase
- The top heat which is the maximum temperature

Low temperatures result in longer fermentation times

High temperature: Speeds up chemical reactions; an increase of 10°C generally doubles the reaction rate:

- Various compounds form faster and are removed faster i.e. diacetyl.
- Increased formation of alcohol, acids, esters, higher alcohols.
- Increased yeast growth.
- Higher rate of attenuation through raised metabolism.
- Increased risk of autolysis and subsequent off-flavours.
- Increased loss of bitterness and colour due to biomass increase.
- More foaming during fermentation and subsequent reduction in head retention.
- Faster drop in pH and possible lower pH in final beer.

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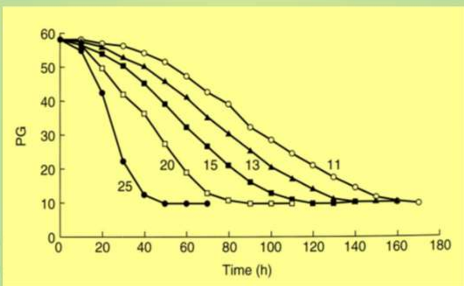
Effect of Temperature vs Time on Attenuation.



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Effect of Aeration on Fermentation.

Increased wort aeration

- Increased production of sterols and unsaturated fatty acids
- Increase in yeast growth
- Increase in higher alcohols
- Increase in H_2S levels
- Decrease in esters
- Decrease in short chain fatty acids (C6 to C10)

Decreased Aeration

- Less production of lipids will reduce cell growth and reproduction
- Reduced amino acid uptake and synthesis
- Fewer higher alcohols
- More esters
- More saturated fatty acids
- Less H_2S

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Using Dried Yeast?

Always rehydrate as per supplier’s instructions.

No need to aerate the wort: During production dried yeast stores up sufficient amounts of unsaturated fatty acids and sterols to produce enough biomass in the first stage of fermentation.

Generally a “pitch n ditch” system

If the slurry from dried yeast fermentation is re-pitched from one batch of beer to another, the wort has to be highly oxygenated. However, yeast performance will be different.

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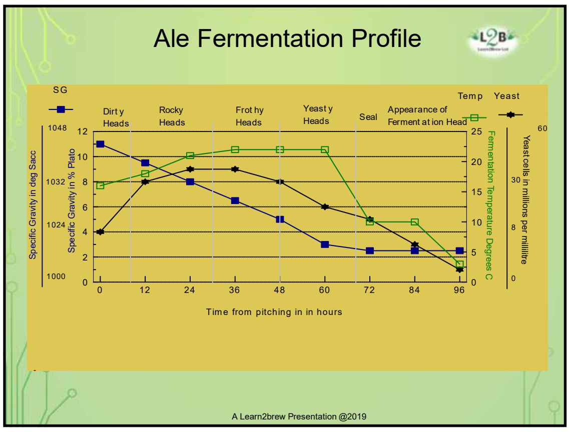
Ale Fermentation Profile
L2B
SG
Dirty Heads
Rocky Heads
Frothy Heads
Yeasty Heads
Seal
Appearance of Fermentation Head
Temp
Yeast
Specific Gravity in deg Sacc
Specific Gravity in % Plato
Fermentation Temperature Degrees C
Yeast cells in millions per millilitre
Time from pitching in in hours
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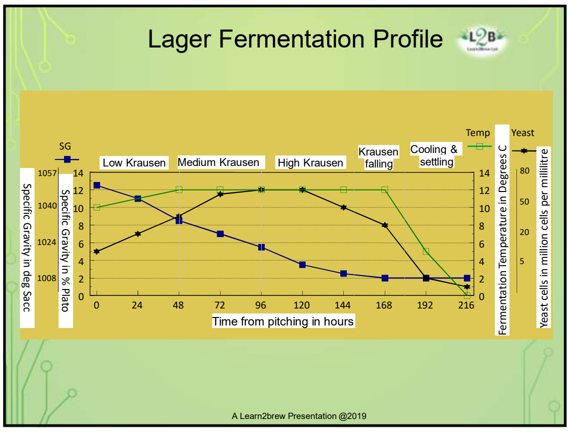
Lager Fermentation Profile
L2B
SG
Low Krausen
Medium Krausen
High Krausen
Krausen falling
Cooling & settling
Temp
Yeast
Specific Gravity in deg Sacc
Specific Gravity in % Plato
Fermentation Temperature in Degrees C
Yeast cells in million cells per millilitre
Time from pitching in hours
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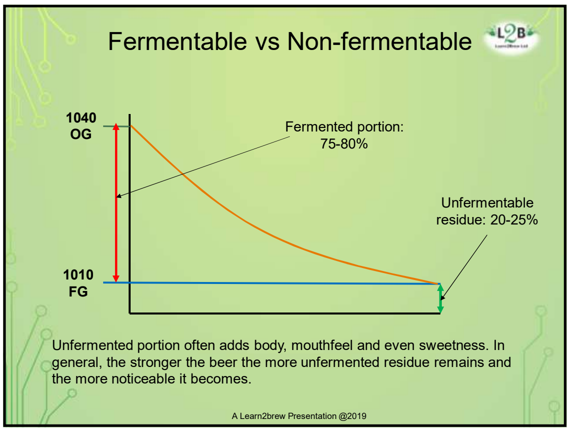
Fermentable vs Non-fermentable
L2B
1040
OG
Fermented portion:
75-80%
Unfermentable
residue: 20-25%
1010
FG
Unfermented portion often adds body, mouthfeel and even sweetness. In general, the stronger the beer the more unfermented residue remains and the more noticeable it becomes.
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Attenuation

The percentage figure used to give the amount of wort sugars turned into ethanol and CO_2. Majority of brewing strains attenuate in the range of 70 - 80%.

More accurately this is known as the "Apparent Attenuation" and is calculated by comparing the Original and Final Gravities of the beer.

A 1.040 OG that ferments to a FG of 1.010 would have an apparent attenuation of 75%:

% apparent attenuation (AA) = (OG-FG)/OG = 40-10/40 = 75%

The "Real" attenuation is less (81.9% of AA). Ethanol has a gravity of about 0.800. A 1.040 OG beer with 100% real attenuation would give FG of about 0.990

Yeast attenuation ability can be expressed by the following percentages:

<70% = Low
71-75% = Medium
76-80% = High

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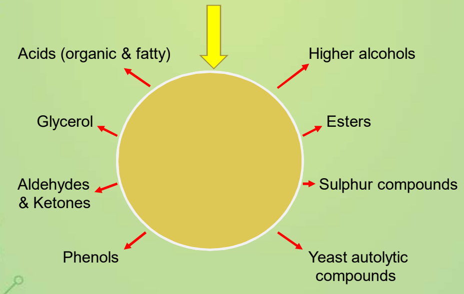
Secondary Metabolites



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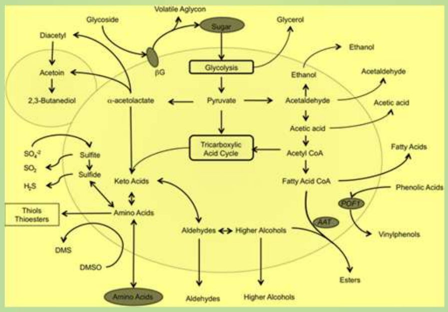
Overview of Saccharomyces Metabolic Activities Influencing Beer Quality.



Nicholas A. Bokulich, and Charles W. Bamforth Microbiol. Mol. Biol. Rev. 2013;77:157-172

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Main Reasons for Secondary Yeast Metabolites

- Overflow or "leaking" metabolism
- Cellular redox balancing - in general, redox reactions involve the transfer of electrons between various molecules.
- Maintenance of intracellular pH (pHi) - The pHi plays a critical role in the function of the cell and as such close regulation is required for cells to survive.
- Shock excretion - The phenomenon of amino acid release when cell surroundings change i.e. temperature drops, increased osmotic pressure.

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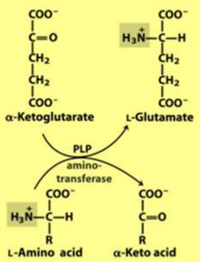
Transamination

- Typically, α-ketoglutarate accepts amino groups
- L-Glutamine acts as a temporary storage of nitrogen
- L-Glutamine can donate the amino group when needed for amino acid biosynthesis
- All aminotransferases rely on the pyridoxal phosphate (PLP) cofactor



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Fermentation Flavours

Higher alcohols: Larger than ethanol, >40. Have a warming and solvent like flavour. Major contributors to hangovers?
Most noticeable in strong beers. Include: n-propanol, isobutanol, 2-methyl-1-butanol and 3-methyl-1-butanol, 2-phenylethanol.

Produced by 2 pathways during primary fermentation: Amino acid synthesis (early) and from sugars (later).

End products; not reabsorbed.

- Yeast strain: Ranges greatly; 1-5x
- Pitching rate: High pitching = Less, underpitching = More
- Wort gravity: High = More
- Increased/excess aeration: More
- Low FAN: High adjunct use = More.
- Excess FAN = More
- FV design: Tall CCVs = More

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Fermentation Flavours

Esters form largest group of flavour active compounds, c.100 produced during fermentation. Give beer fruity flavour, more apparent in UK ales. Include: ethyl acetate, isoamyl acetate, isobutyl acetate, ethyl caproate and 2-phenylethyl acetate.

Yeast end products; not reabsorbed. Peak production at end of yeast growth phase.

- Yeast strain: Varies greatly
- Wort aeration: Too low = More.
- Excess CoA = More
- Wort sugar profile: High maltose = Less. High glucose = More
- FAN: Excess = More
- OG: Higher = More
- Temperature: Higher = More

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Fermentation Flavours

Diacetyl & Pentanedione (VDK): Gives fullness and sweetness at low levels. Can enhance flavour of some ales. At higher concentrations flavour is more butterscotch-like and the palate very full; considered an off-flavour.

By-product of protein anabolism i.e. formation. Excreted and reabsorbed.

Reaches peak at end of yeast growth phase.

Useful indicator for incomplete or faulty fermentation.

Can be produced by bacteria i.e. pediococcus

- Yeast strain
- FAN: Use well modified, low N malts and max. 30% adjunct
- Pitching rate: Under-pitching = More
- Aeration: Neither under nor over.
- Yeast viability: Healthy

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Fermentation Flavours

Diacetyl & Pentanedione (VDK):

Control of production, re-absorbtion & recovery of beer with high levels:

1. Forcing test to check pre-cursor level

2. Sufficient VDK rest time and temperature:

3. Rest days = $\frac{100 - (\text{sum daily ave FV Temp})}{\text{VDK rest Temp.}}$

4. Recovery using Krausen (10-15%)

5. If it's bacteria – Ditch it!

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Fermentation Flavours

Acetaldehyde (ethanal): Intermediate fermentation product, pre-curser to ethanol.

Excreted early in fermentation but reabsorbed later in maturation.

Indicator of immature or stale beer

Can be produced by bacteria i.e. zymomonas

- Yeast strain
- FAN: Low levels; possible increase.
- Wort nutrient shortage i.e. lack of zinc = More
- Over pitching = More
- Excess aeration = More
- Insufficient maturation = More

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Fermentation Flavours

Sulphur Compounds:

Make a significant contribution to beer flavour. When in excess, they can give rise to unpleasant off flavours; fermentation should be managed. This is especially important for the more volatile compounds such as:

Hydrogen sulphide (H_2S) which smells of bad eggs
Sulphur dioxide (SO_2) which smells of burnt matches.

Both of these sulphur compounds are produced by yeast from sulphate and are by-products in the synthesis of sulphur-containing amino acids.

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Fermentation Flavours

Dimethyl Sulphide (DMS):

Principally derived from malt but can be from yeast action: Reduction of DMSO to DMS.

Smells and tastes of cooked vegetables, baked beans or tinned sweetcorn.

Rarely detectable in ales; usually removed during kilning to produce ale malts: Higher temperature.

Can be present at detectable levels in some lagers.

- Ensure sufficient evolution of CO_2 to purge these from the beer.
- Extended maturation time; allow these sulphur compounds to escape).
- More vigorous fermentation processes result in lower levels of the volatile sulphur compounds.

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Other Fermentation Flavours

Glycerol:

Occurs in all beers at differing levels: Sweet and adds mouthfeel at higher levels. 5%ABV beer contains around 0.2%v/v. Can reach 1%.

Organic acids:

Make minor contribution to beer flavour contributing acidic and salty notes. Important in lowering pH i.e. the acidity of the beer.

Fatty acids:

At high concentrations which can be tasted are undesirable giving soapy/fatty/goaty flavours.

Aldehydes:

Generally considered as off-flavours. Often associated with problem fermentations. Grassy, hay-like.

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Wild Yeast: Effect on Beer

Category of Yeast	Effect on wort or beer
Pichia.	Grows rapidly in the presence of air. Forms a film on the surface of the beer.
Candida mycoderma.	Grows rapidly in the presence of air. Forms a film on the surface of the beer.
Saccharomyces diastaticus.	Continues to ferment all carbohydrates so there is no control of attenuation. i.e. very low FG. Also causes off flavours i.e. 4VG
Torulopsis.	Fails to sediment and causes hazes.
Brettanomyces.	Very slow growing but it produces acid and causes off flavours.
Hansenula.	Grows rapidly in the presence of air. Forms a film on the surface of the beer.

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Importance of Maturation

At the end of fermentation three principal flavour faults might remain:

- *Acetaldehyde*
- *Diacetyl*
- *Sulphur compounds*

Largely result of poor yeast growth possibly due to poor oxygenation, yeast viability or vitality, temperature, nutrient status etc. Or poor brewing technique.

Yeast must be present during the maturation stage.

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Maturation

Various regimes dependent on beer style

Main targets:

- Reduce undesirable flavours and achieve desired flavour profile
- Colloidal (chill) stability through formation and sedimentation of polyphenol-protein complexes plus yeast and other insoluble compounds
- Conditioning; production of CO_2 via secondary fermentation and removal (scrubbing) of volatiles

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Flocculation Theory

- Yeast flocculation characteristics gene controlled; some strains show a preponderance to floc, others don't.
- Yeast split into 2 phenotypes: FLO1 and NEWFLO strains.
- Most brewing strains (both ale and lager) are of the NEWFLO type, the distinguishing feature between the two being the sugars that inhibit their flocculating ability.
- A third phenotype identified; "MI Type" (mannose-insensitive).
- Flocculation pattern impacts to a great extent on fermentation performance. An ideal strain completes the fermentation process in a regular manner i.e. same length of time and then leaves the beer rapidly for ease of collection.
- Chain forming is not flocculation.

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Flocculation Theory

Two other considerations:

- Cell Surface Hydrophobicity (CSH)
- Cell Surface Charge (CSC).

Ale strains <negative cell surface charge, display an aversion to water (hydrophobicity), more hydrophobic than lager strains hence form a cluster round CO_2 bubbles and rise to the surface of the wort; "top cropping".

CSH increases rapidly during exponential growth phase, settles back to a lower level in the stationary phase.

Young yeast cells display less CSH than older mature ones.

CSH thought to affect flocculins; more active during stationary phase?

Phosphate groups on the cell surface seem to play a big role in CSH levels.

CSC varies over the yeast cell life. Seems to decrease just prior to flocculation thereby reducing the electrostatic repulsion of two like charges.

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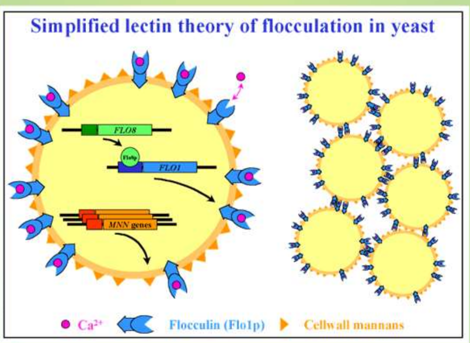
Yeast Flocculation



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Yeast Cropping

Ale yeasts are cropped generally on day 3-4 of the fermentation when internal glycogen reserves have built up again and viability is high.

Lager yeasts are cropped at the end of fermentation and need to be collected carefully to avoid contamination by trub and dead yeast cells if not using:

a) CCVs

a) Traditional secondary fermentation in lagering tanks which is akin to the ale dropping fermentation system

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Ale Yeast Cropping

Top cropping yeast is removed during the fermentation and saved for re-pitching.

Removing the yeast also helps control attenuation.

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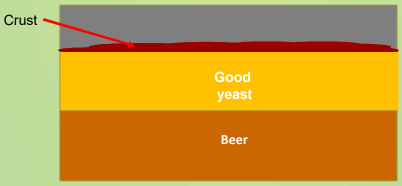
Ale Yeast Cropping

Cross section at top of FV



1. Gently move crust to one side revealing good clean healthy yeast layer – should be thick and creamy.
2. Crop carefully using sterilised ladle, jug etc. and transfer immediately to sterilised bucket.
3. Close/seal bucket and refrigerate immediately: <4ºC.

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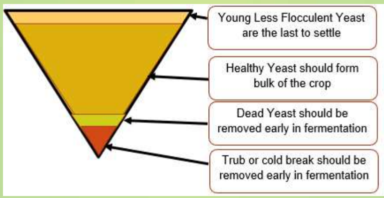
Lager Yeast Cropping

The yeast cone is an inhomogeneous system



In conical vessels the first part of yeast crop is discarded and the middle portion is kept as the most representative part of the crop. Best practice is to remove a small portion of bottoms from the FV in the first 12 - 24 hours to take off dead cells and cold break that may have settled

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Yeast Storage: Best Practice

For best quality yeast and fermentation performance:

- Chill to storage temperature rapidly
- Maintain temperature at 2-4ºC (Dried yeast 10ºC)
- Keep Yeast slurries homogeneous: mix but don't aerate.
- Avoid yeast aeration or the yeast will start to grow
- Keep storage time to a minimum 2 days (max. 5 days)
- Slurry with sterile water or liquor if high gravity brewing
- Comprehensive CIP regime
- Thorough sampling & inspection regime i.e. counts and viability

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Acid Washing of Yeast

Equipment:
- A clean and sterilised container (10-20 litres)
- A clean and sterilised whisk or brewing spoon for stirring
- Food grade Phosphoric Acid (usually 85% concentration)
- pH meter or accurate pH strips.
- PPE (Goggles, gauntlets etc.)

Process:
Ensure acid and yeast slurry are chilled <4ºC
Put the goggles and gauntlets on!
- Dilute the phosphoric acid; 1 part acid in 9 parts water to give 8-9% conc. NB. Always add acid to water.
- Sanitise the whisk or spoon thoroughly with peracetic acid.
- Whilst stirring the yeast slurry add, slowly, a small amount of the diluted phosphoric acid solution. Then continue stirring for at least 3 minutes to allow the acid solution to mix in evenly and thoroughly.
- Take a pH reading. If it is between 1.9 and 2.1, then it is OK. If pH is >2.2 continue to add small amounts of the acid solution and mix well.
- Rest for 1 hour in fridge at <4ºC, then pitch.

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Trouble Shooting

Fermentation not proceeding as expected or is stuck:

- Insufficient aeration at time of pitching – Rouse and aerate vigorously if within 24 hours of pitching yeast
- Insufficient yeast pitched – Rouse and add additional yeast at 50% of pitching level if within 24 hours of first pitching
- Poor yeast viability – Ensure yeast slurry is used within 3-4 days of cropping and stored at <4ºC. Ensure all dried yeast is from good sealed packets.
- Wort temperature wrong at time of pitching: 18-20ºC for ales and 10-12ºC for lagers.
- Lack of key nutrients e.g. zinc – Particular risk when using high additions of sugar syrups, rice or maize. Add yeast food and rouse.
- Fermentation temperature too low – Ale yeasts become almost dormant at <12ºC. Raise FV temperature slowly and evenly. If need be rouse yeast off bottom of vessel but no need to aerate.
- Yeast has flocculated and dropped out of wort – Rouse daily if the strain has this tendency. Don't aerate.
- Check calcium level: 150 to 200mg/l recommended
- Incorrect mashing regime resulting in too many dextrins/higher sugars – Check mash temperature, 65-66ºC is normal.

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Thank you.

Any questions?

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