

**The Institute of Brewing and Distilling
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**YEAST MANAGEMENT – CULTURE
HANDLING BETWEEN BREWING
FERMENTATIONS**



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Brewer's wort fermentation has two objectives:

Yeast management can be divided into a number of overlapping procedures:

- To consistently metabolise wort constituents into ethanol, carbon dioxide and other fermentation products in order to produce beer with satisfactory quality, drinkability and stability;
- To produce yeast crops that can be confidently repitched into subsequent wort fermentations.

Brewer's yeast recycling

- Brewing is the only major alcoholic beverage that recycles its yeast;
- Brewing alcoholic fermentation processes such as whisky production only use a yeast once;
- Recycling of brewer's yeast does reduce the total amount of the yeast required long term but it does introduce a degree of complexity and expense to overall process;
- The collective operation of brewer's yeast in between fermentations is designated as YEAST MANAGEMENT.

YEAST MANAGEMENT

STEP-BY-STEP

Yeast management step-by-step

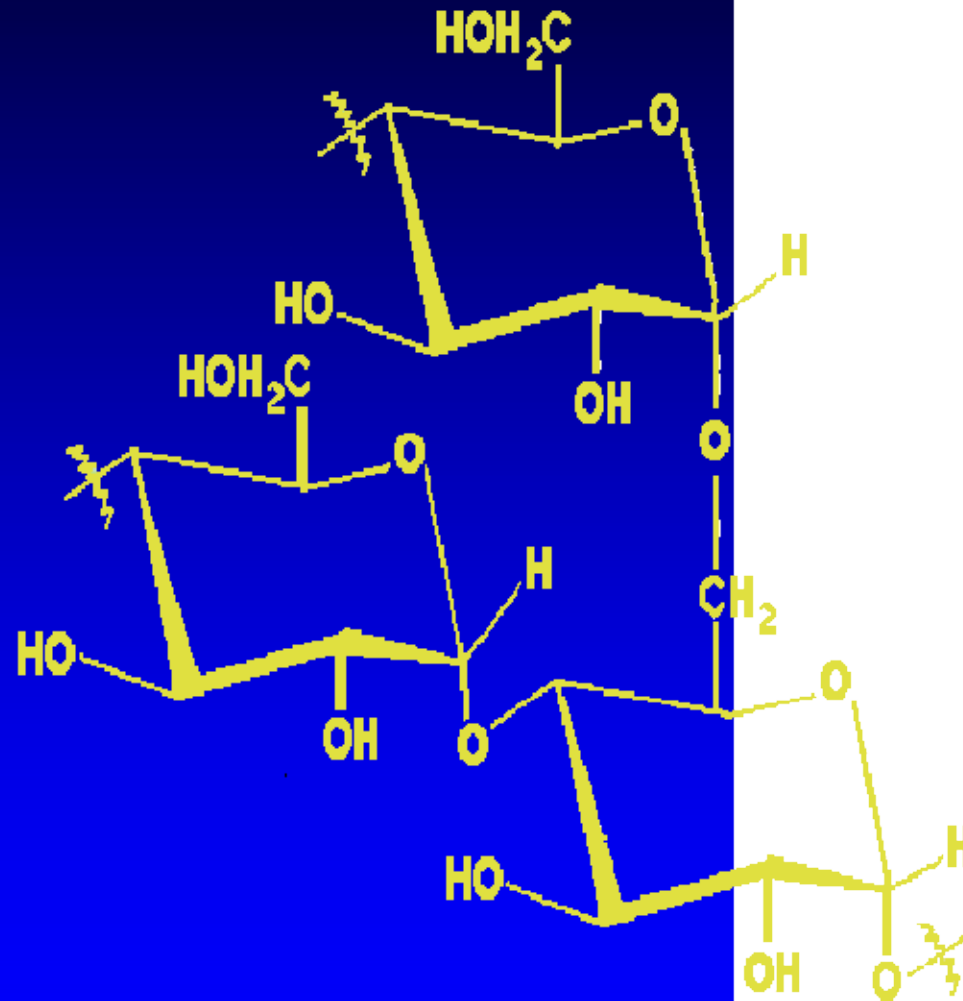
Yeast management can be divided into a number of overlapping procedures:

- Prior to propagation (the production of yeast biomass) after fermentation and cropping most (not all) yeast strains are stored under standard conditions in a brewery or in an accredited culture collection – sometimes both for security;
- Yeast propagation (biomass formation) in wort under aerobic conditions;
- Following propagation the yeast is pitched into wort. This is the first cycle (generation) of a multi-generation procedure;
- At the end of fermentation (attenuation), yeast cropping occurs followed by storage prior to re-pitching;
- In order to eliminate contaminating bacteria the yeast slurry can be acid washed. Also, sometimes, but less frequently these days, the yeast slurry is sieved to remove contaminating trub (coagulated protein-phenol solid material).

Yeast management – the principal objectives

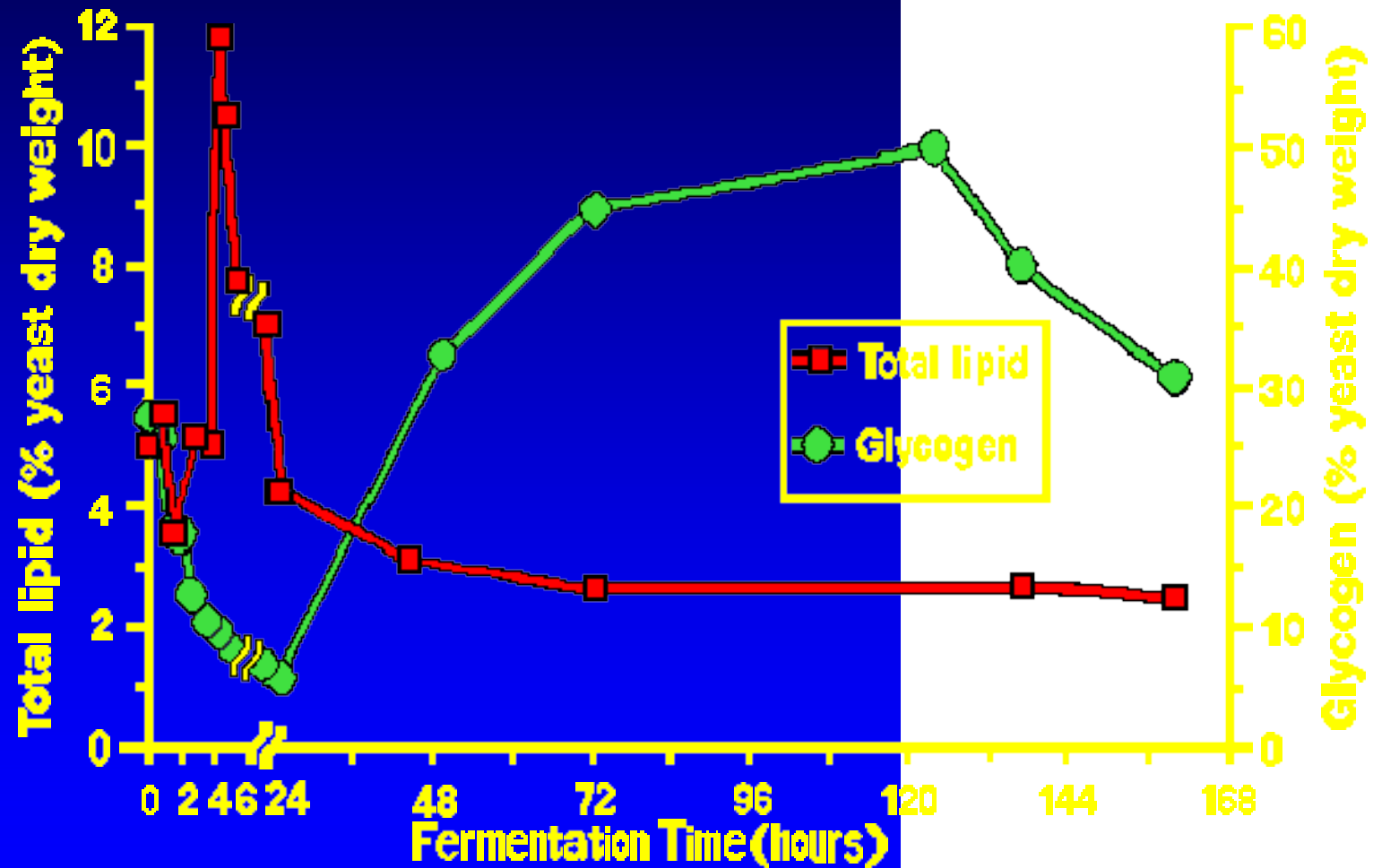
- To maintain the yeast viability above 90% (usually determined with methylene blue stain) and enhance the cultures' vitality;
- Ensure that the vitality remains elevated but vitality measurements are difficult:
 - Acidification Power Test;
 - Intracellular pH (flow cytometry);
 - Magnesium Release test;
- Preserve the cell's cellular integrity;
- Intracellular glycogen levels are critical as a store potential biochemical energy.

Structure of glycogen



The store of potential biochemical energy

Intracellular concentration of glycogen and lipids in a lager yeast strain during fermentation of a 15° Plato wort



**STORAGE OF
YEAST STOCK CULTURES
BETWEEN PROPAGATIONS**

Emil Christian Hansen (1842-1909)



Culture yeast management

Storage conditions investigated:

- Low temperature as a result of storage in liquid nitrogen (-196°C). With the advent of -70°C refrigerators in the 1980s, liquid nitrogen has been largely replaced for this purpose with similar results;
- Lyophilisation (freeze drying);
- Storage in distilled water;
- Storage under oil;
- Repeated direct transfer on solid culture media (subcultured once a week for two years);
- Long term storage at 21°C on solid nutrient medium – subcultured every six months for two years;
- Long term storage at 4°C on solid nutrient medium – subcultured every six months.

YEAST

PROPAGATION

Yeast propagation

- Brewer's yeast cultures do not last forever and should be regularly replaced;
- Yeast propagation in breweries is not fully understood because of the yeast's ability to metabolise wort sugars in both anaerobic and aerobic circumstances – Crabtree Effect;
- Propagation is carried out with wort in a batch reactors with constant aeration (or oxygenation);
- Wort (not molasses/ammonium ions) limits aerobic growth consequently theoretical quantities of biomass are not produced.

The principal functions of oxygen in brewing

It is interesting to note that oxygen is **ONLY** required at the following stages in the malting and brewing process:

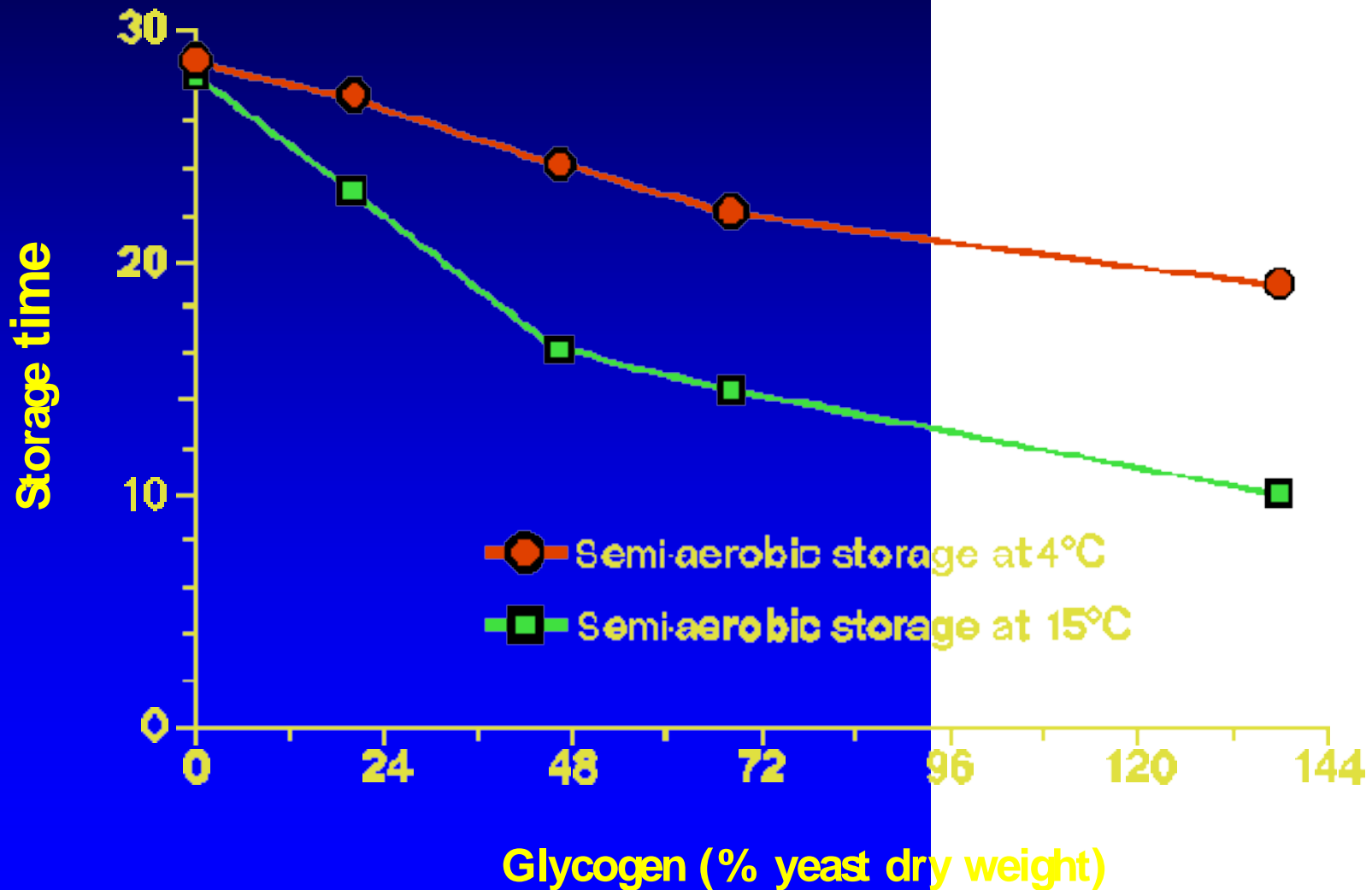
- During barley germination during malting;
- For biomass formation during yeast propagation;
- At the beginning of fermentation when the yeast is pitched into wort.

YEAST STORAGE

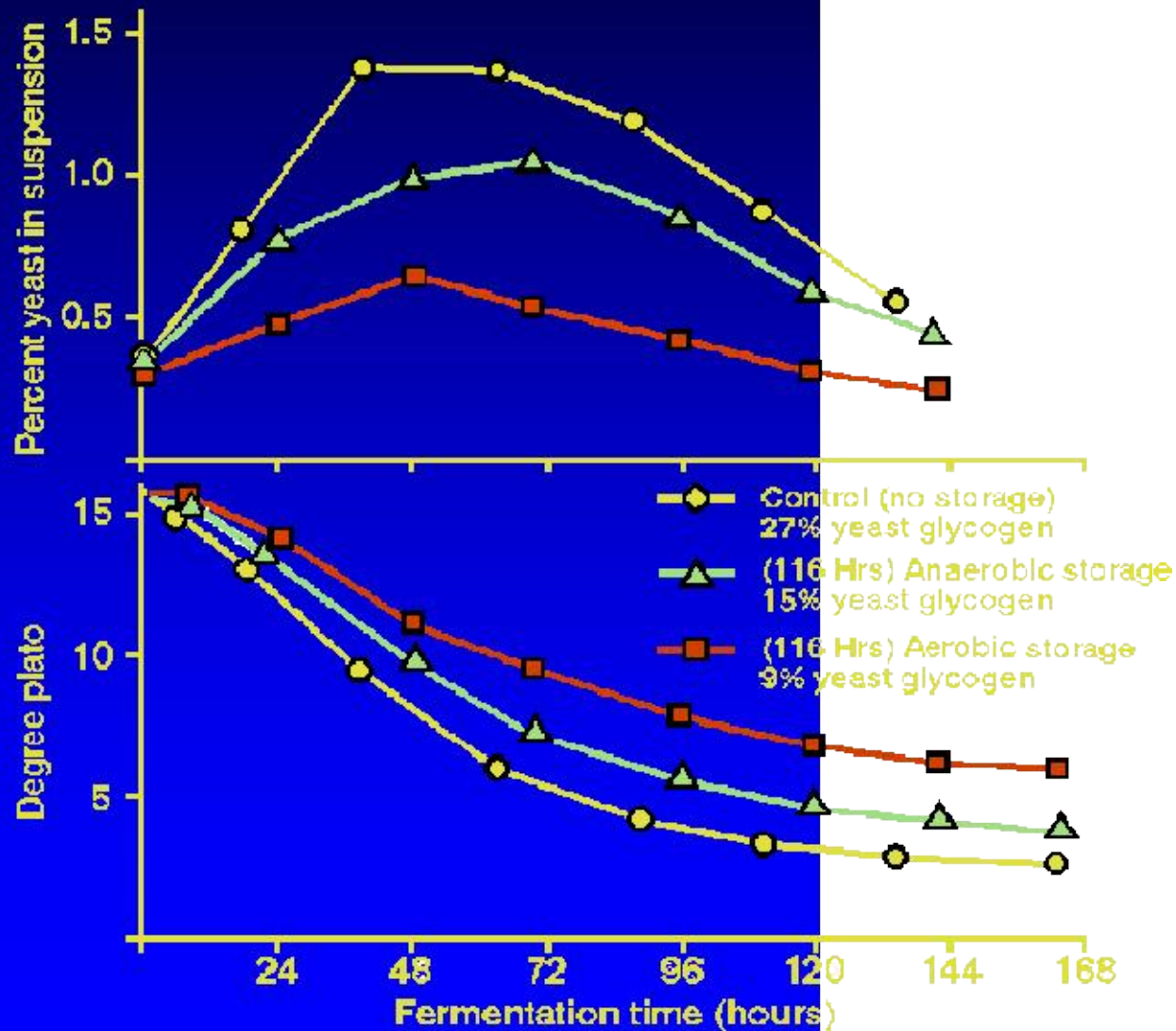
Storage of cropped yeast

- Cropped yeast should be stored between fermentations in sterile, cold (2-4° C) conditions;
- Cultures are usually stored in beer [4-6%(v/v) ethanol];
- Stored yeast slurries should be intermittently stored with low shear devices;
- Yeast cropping with centrifuges can present its problems!

The effect of yeast storage temperature on intracellular glycogen concentration



The effect of yeast glycogen at pitching on lager fermentation characteristics



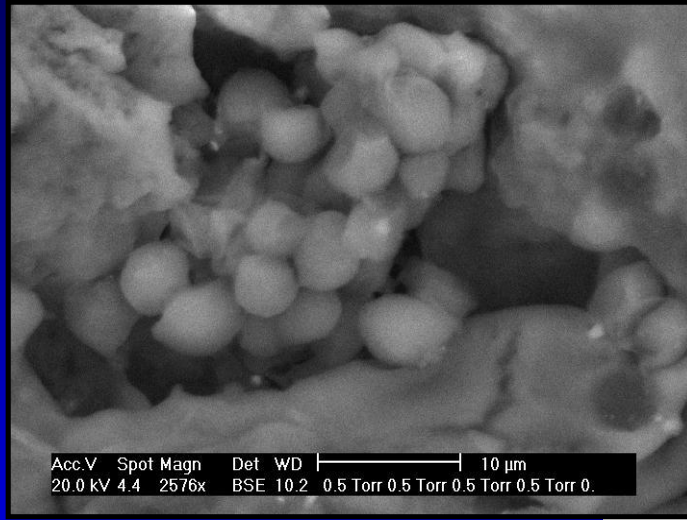
Disc stack centrifuge



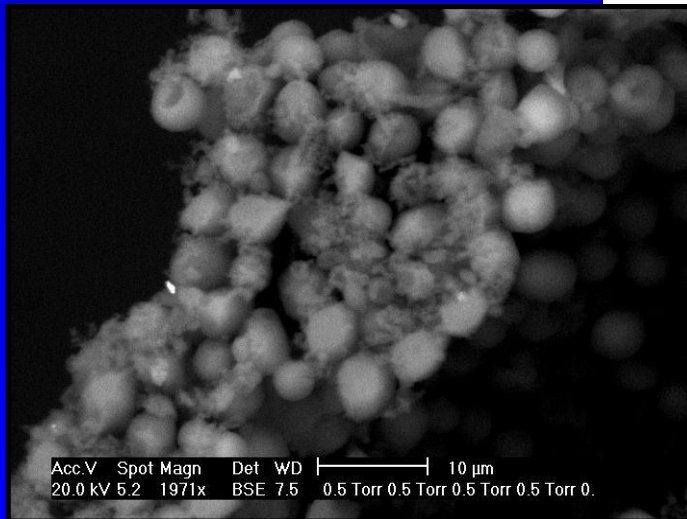
Scanning Electron Microscopy of an ale strain A.

A. Cells prior to passage through a disc centrifuge. **B.** Cells following passage through a disc centrifuge.

A



B



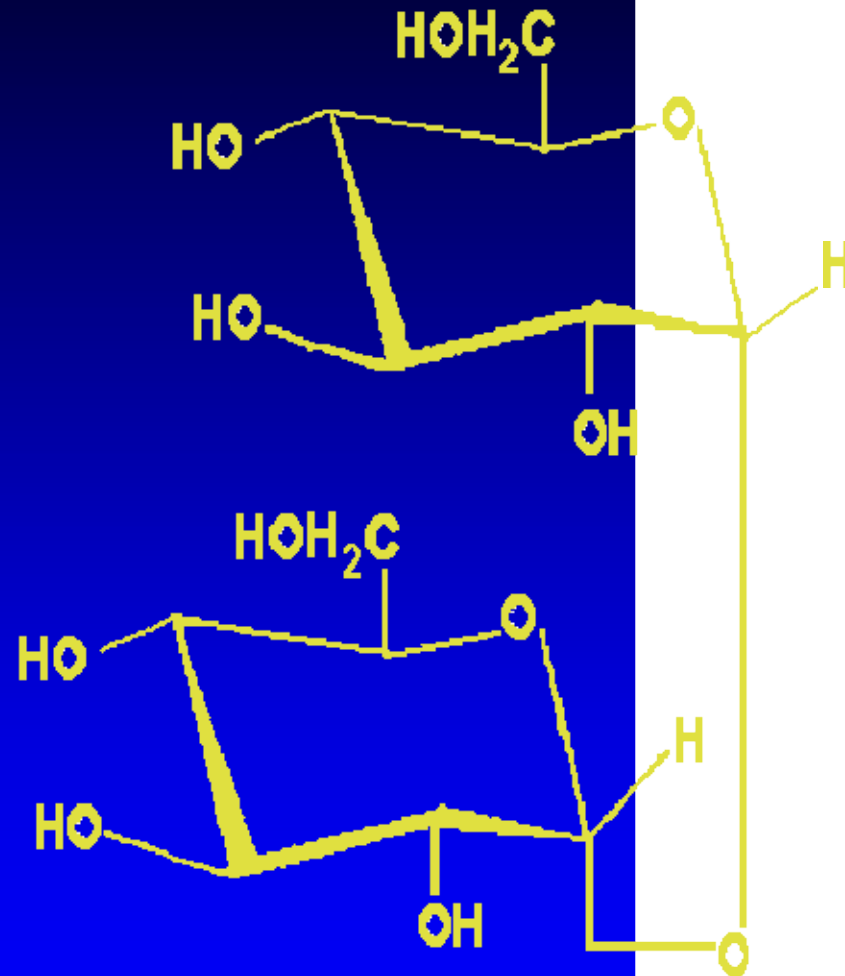
Yeast condition following centrifugation from a 16° Plato wort fermentation

Yeast exit temperature	16-18°C	28-30°C
Viability (%)	95	80
Respiratory deficient cells (%)	5	25
Glycogen (mg/ g dry weight)	18.6	12.2

Wort gravity and yeast cycles (generations)

- 12° Plato wort - > 20 yeast cycles
- 14° Plato wort - 16 yeast cycles
- 16° Plato wort - 12 yeast cycles
- 18° Plato wort - 8 yeast cycles

Structure of trehalose



Protects yeast from stress - osmotic pressure, ethanol, temperature, etc.

**Concentration of trehalose and glycogen
in lager yeast after one, four and eight cycles following
fermentation in 15° Plato wort**

	Cycles (generations)		
	One	Four	Eight
Trehalose^a	8.8	9.2	11.6
Glycogen^b	14.6	12.6	9.2

^a $\mu\text{g/g}$ dry weight of yeast

^b mg/g dry weight of yeast

YEAST WASHING

Yeast washing

Do's and do not's for yeast acid washing :

The Do's of acid washing:

- Use food grade acid;
- Chill the acid and the yeast slurry before use to less than 5°C;
- Wash the yeast as a beer slurry or as a slurry in water;
- Ensure constant stirring whilst the acid is added to the yeast and preferably throughout the wash;
- Ensure that the temperature of the yeast slurry does not exceed 5°C during washing;
- Verify the pH of the yeast slurry; and
- Pitch the yeast immediately after washing.

Yeast washing (cont'd)

The Do Nots of acid washing:

- Do not wash for more than two hours – very important;
- Do not store washed yeast;
- Do not wash unhealthy yeast; and
- Avoid washing yeast from high gravity fermentations prior to dilution.

Yeast washing (cont'd)

There are a number of options to acid washing brewer's yeast:

- Never acid wash yeast;
- Low yeast generation (cycle) specification;
- Discard yeast when there is evidence of contamination (bacteria and/ or wild yeast);
- Avoid washing yeast from high gravity fermentations prior to dilution;
- Acid wash every cycle, this procedure can have adverse effects on yeast; or
- Acid wash when bacteria infection levels warrant the procedure.

Summary

It is important to jealously protect the quality of the cropped yeast between fermentations because it will be used to pitch a later fermentation and will have a profound effect on the quality and stability of the beer produced with it.

A cknowledgements

Thanks are due to Anne Anstruther for her assistance with the development of this presentation and the written version.



