ABER YEAST MONITOR

HANDLE WITH CARE!

CHRISTOPHER BOULTON, E-MAIL: CHRISTOPHER.BOULTON@NOTTINGHAM.AC.UK, AND DAVID QUAIN, E-MAIL: DAVID.QUAIN@RED-TS.COM

Yeast is at the heart of the brewing process, yet is all too often treated as a peripheral concern.
But help is at hand, thanks to industry consultants Chris Boulton and David Quain, who offer here an invaluable guide to best practice in industrial-scale brewing yeast handling.

If I were a yeast cell, I would want to be looked after, cherished even! Regrettably yeast remains something of a commodity; being recycled from one fermentation to another with little, if any, comment. Perhaps things would be different if yeast was supplied from a third party, bought, if you will. Then, given the constructive two way interaction between the supplier and customer, things might be a little different. Yeast viability might well tighten, yeast solids would become of interest and, gosh, microbiology would become commercially important. Whilst it is good to dream, in the vast majority of breweries, yeast remains a bit of a 'poor relation', uncomplaining, but doing the job!

Of course, there is no such thing as a 'standard' brewery. Although brewing groups might have focussed on this or that equipment or process, global consolidation has resulted in a mish mash (no pun intended) of software, hardware, and processes to create beer.

From the viewpoint of yeast, a strain used by different breweries can be fermenting very different worts (raw materials, brewhouse operations) in a diversity of different fermenter types (size, geometry, and cooling). On top of this, yeast handling (cropping, storage, and acid washing) processes and approaches can vary. Accordingly for yeast, the philosophy of 'handle with care' may require some interpretation, but nevertheless it remains important to do so. Hopefully, this article will both reassure, but also challenge your approach to looking after your yeast.

YEAST SUPPLY

Whilst it is true that yeast must be cherished, it is obviously also a given that the recipient of all that care should be entitled to receive it. In other words, the supply system must be robust enough to deliver to the fermenter, the point of use, the right yeast strain in the right quantity and in the right condition.

As in any multi-step process, the whole cannot be any better than its weakest link. This means that storage of master cultures, assurance of culture identity, and propagation in both laboratory and brewery must be bullet proof procedures. It sounds like obvious stuff, and it is, but how many brewers can be totally confident that these basic requirements are actually achieved?

It is not all that uncommon to see laboratory propagation systems which have, obviously, been cobbled together on a shoestring budget, yet, somewhat optimistically, are expected to supply high quality cultures for shiny, expensive modern brewery handling systems. Similarly, many companies are apparently perfectly happy to invest enormous sums of money on huge cylindroconical fermentation tank farms, but leave the hapless brewer to service these with an inefficient, undersized propagation system and an insufficient number of too small yeast storage vessels. It is hardly surprising that in these circumstances beer quality is variable.

The gold standards for yeast supply are well known. Master cultures should be stored in liquid nitrogen and recovered using a properly audited supply system that guarantees that propagation cultures are pure and of the right identity. The laboratory

propagation system should be operated to pharmaceutical industry standards and must be capable of supplying sufficient high quality yeast to achieve an acceptable pitching rate in the first brewery propagation tank. The brewery propagation system must be the correct size for the fermenters that are required to be pitched and, most importantly, it must be designed to be capable of maintaining continuous aerobic conditions.

Understandably, brewers are more used to keeping oxygen out than thinking about how best to add it. Large scale cultures of actively growing yeast have an appetite for oxygen almost as voracious as marketing departments have for shares of company budgets. Satisfaction of this appetite requires growth vessels that are designed to deliver high rates of oxygen transfer. This means lots of oxygen and heavy duty mechanical rousers. The skills required for these designs are not generally to be found within the brewing industry and the use of external expertise in the early planning stages is probably a sensible course of action. It goes without saying that such capabilities cannot be retrofitted to existing plant.

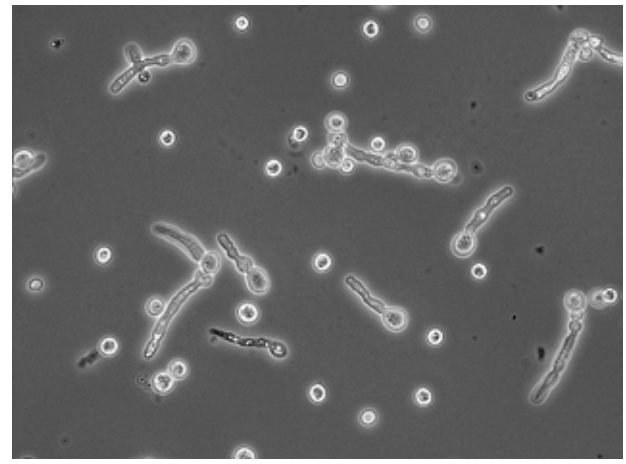
PITCHING AND FERMENTATION

Assuming the supply system is capable of delivering the right yeast, the next link in the chain is the requirement to pitch the right amount of yeast. At first sight, this procedure appears deceptively simple, yet the manner in which it is achieved can have far reaching effects on fermentation performance and beer quality. These relationships are not always appreciated and understood.

The requirements of pitching rate control systems are easy to define (in theory). The pitching rate is the viable yeast concentration suspended in wort at the start of fermentation. It follows that the pitching system must be capable of delivering the correct quantity of viable yeast.

At completion of pitching, the yeast should be homogeneously distributed throughout the entire volume of wort. For the sake of consistency, it is preferable that every yeast cell should be

> “PERHAPS THINGS WOULD BE DIFFERENT IF YEAST WAS SUPPLIED FROM A THIRD PARTY, BOUGHT, IF YOU WILL. THEN, GIVEN THE CONSTRUCTIVE TWO WAY INTERACTION BETWEEN THE SUPPLIER AND CUSTOMER, THINGS MIGHT BE A LITTLE DIFFERENT.”



simultaneously exposed to homogenous wort. It is these two requirements that have the capacity to cause the greatest degree of process variability.

The accuracy and repeatability of pitching rate control systems have improved enormously over the last several years. Undoubtedly, those systems that have e.g. the Aber Instrument yeast biomass probe at their heart represent the best that is currently available. With appropriate flow cells and properly designed controlling software, in-line yeast biomass based systems are probably capable of delivering a pitching rate that is plus or minus 5% of the desired value.

So what about the timing and manner of pitching? This has become a serious issue largely because of the now common use of very high capacity fermenters, each requiring several batches of wort to fill them. In this situation, the obvious question is how to manage the sequence and timing of pitching and oxygenation. The dilemmas are obvious:

- Should you pitch all the yeast early in collection of the first wort length?
- Should you distribute the yeast addition across the entire wort collection?
- Should you oxygenate all batches of wort and at what concentration?

Clearly, the possible permutations are endless and this only gets worse when the same beer quality is required to be made, using a variety of fermenters of varying capacity and with varying collection procedures.

Sadly, there are no simple answers. If all the yeast is added, in a single slug, at the start of collection of the first batch of wort, all cells undergo, more or less simultaneously, the transition from storage to fermenter. This satisfies one of the key requirements of pitching systems.

However, things may not be quite that simple. Usually, the yeast is added in-line in the form of a chilled slurry in beer. The wort, into which the yeast slurry is injected, is, obviously, →

considerably denser and several degrees warmer. Since fermenters are usually designed to be filled with a minimum of turbulence, there is a very good chance that the yeast slurry will take a long time to fully disperse. Indeed, it might even just float on the surface of the wort!

If the yeast is added over the whole time course of wort collection, it is relatively easy to make sure that it ends up evenly dispersed when the fermenter is full. Of course, very large fermenters can take several hours to fill. The consequences of this are almost certainly much more problematic.

The yeast added early in collection takes up oxygen and moves from the stationary (or maintenance) phase into active growth. The yeast added late in collection is not in such a fortunate situation. It is still in the stationary phase and, compared to the actively growing population, is not able to assimilate oxygen, or for that matter any other nutrients, with ease.

Obviously, the yeast that is pitched in between the first and last brew length faces a physiology intermediate between these two extremes. The result is that the population of yeast in the fermenter at the completion of collection is totally heterogeneous. The effects of this are difficult to quantify, but, at the very least, it is probable that the yeast added late cannot compete and the net outcome is under-pitching. What is certainly true is that the outcome of the fermentation is not in the control of the brewer.

Take this scenario slightly further and consider this type of collection procedure applied to two fermenters with very different capacities and filling times, and it becomes easy to see why product matching can sometimes be difficult.

Certainly, the best way to get round many of these problems is to take the option of pitching early and over as short a time as possible, but also to consider fitting fermenters with some form of efficient forced mixing.

No doubt, it is an accident of history, but the beverage industry seems reluctant to grasp the nettle of agitation. When most fermenters were less than 100 hectolitres, this was not much of a problem. With the advent of the enormous vessels in common use today, this laissez-faire attitude is really not good enough. Very few, if any, other industries would trust to convection currents and CO_2 bubbles as a means of ensuring homogeneous conditions during the key stage of formation of their product. Neither should the prudent brewer. There is ample evidence that agitation reduces fermenter cycle times and, more importantly, eliminates a lot of inconsistency. As Werner von Braun used to say, it is not rocket science.

CROPPING

Yeast cropping makes the fermentation world go around. It is a process prerequisite to crop sufficient yeast from fermenter for one or, ideally, two or more subsequent fermentations. This gives the brewer some options on what yeast batch to use next, allowing for consideration of factors such as sluggish performance, yeast viability, infection, and generation number. Above all, perhaps the key factor in cropping is for the yeast to be in the right place (the cone) at the right time, having hit racking gravity and, where appropriate, having achieved diacetyl reduction. Of course, things are often not as simple as that. Those non-conforming yeast strains for whom flocculence is late, cajoled through cooling, or stubbornly non-flocculent, requires cropping through a centrifuge.

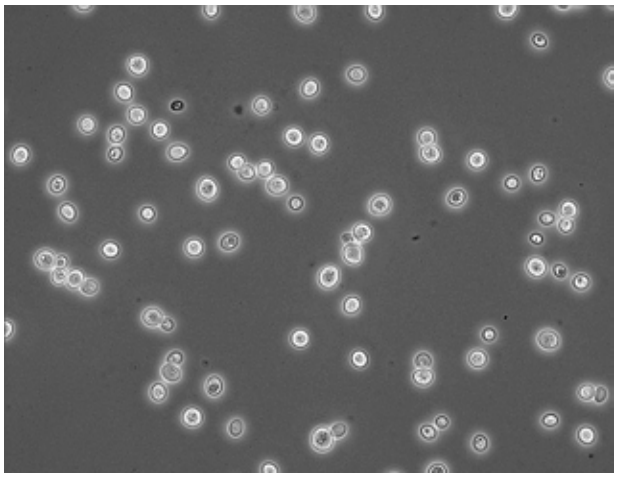
> “THE RESULT IS THAT THE POPULATION OF YEAST IN THE FERMENTER AT THE COMPLETION OF COLLECTION IS TOTALLY HETEROGENEOUS.”

Whatever the pre-cropping scenario, best practice requires removing the yeast from the vessel as soon as is practically possible. Whilst cone cropping is undeniably a convenient process, the cone is usually a less than hospitable environment for yeast. This compact plug of yeast acts as an insulator and is difficult, if not impossible, to cool effectively from the vessel wall through to the centre of the crop.

This presupposes the presence of cone cooling and, if so, encompassing most of the cone surface. Temperature control is further challenged if the top temperature of the fermentation is up to 20°C and the intent is to crop the yeast at 2 or 4°C. Frequently, it is easier and quicker to cool the cone to 6 or 8°C prior to cropping and apply in-line cooling to the desired lower temperature. If this is not enough, FV size and geometry impacts on the management of crop temperature. For example, vessel cone angles can vary greatly from 60° through 90° to 120°, with concomitant impact on cone volume, depth, and width.

Of course, the focus of optimising cone cropping is to minimise the inevitable damage to yeast of sitting in the cone. Most damaging of all is temperature. Prior to the application of cooling, the cone might be expected to be about the same temperature as the fermenting beer above it.

The reality is somewhat worse than this; with the yeast in the cone generating heat through metabolism that can only be poorly dissipated within the slurry. To make matters worse, with the exhaustion of fermentable sugars, the yeast begins to consume its considerable intracellular reserves of the polysaccharide, glycogen. The upshot of all this is that two important factors →



in yeast well-being – temperature and ethanol level – increase sufficiently to damage yeast physiology and viability.
The yeast in the cone is, by no stretch of the imagination, mixed and homogeneous. It has long been accepted that wort trub and big, fat dead yeast cells accumulate in the cone early in fermentation. There is also evidence that subsequent layers reflect heterogeneity in flocculation, cell size, and/or replicative cell age. The cone angle also has some say in things, as the layering and association with the cone wall will be very different at a cone angle of 60° to one at 120°. On top of all these variables, the temperature and ethanol hot spots within the yeast cone will contribute further complexity and heterogeneity.
To sum up, yeast cropping is – in HACCP terms – a critical control point. To minimise the inevitable yeast damage (yeast does not get any better in the cone!), crop as early as possible. Some yeast strains that crop 'well' can be recovered early and warm with downstream cooling prior to or in storage vessel. It is worth remembering that yeast in the cone plays no role in the reduction of VDK, so there is no value leaving it here if there is sufficient to crop. Finally, although heterogeneity is unavoidable, the first few hectolitres (dependent on cone volume) from the cone should be treated as waste and not recovered for storage and subsequent pitching on.

STORAGE

Having been cropped, yeast is stored prior to pitching. On the face of it, a simple enough statement which belies its importance! As with cropping, yeast does not improve with storage, but measures can (and should) be taken to minimise damage to cell physiology and viability.
In terms of best practice, there is a hierarchy of factors that are desired in yeast storage. At the top of the leader board, is the requirement to store yeast as slurry, rather than as a cake. At joint first, is the need to store the slurry cold and keep it mixed. Commendably close at number three, is the growing practice of diluting the yeast slurry to reduce the extra- (and intra-) cellular concentration of ethanol. Finally, where a top gas is applied to the storage tank, it is preferable that it is inert (nitrogen or carbon dioxide) rather than oxygen, which – for process consistency – is best added downstream in fermenter.
Whilst yeast storage as slurry at 50% solids, or less, is pretty much the norm, temperature is not always as well controlled. Although 2°C is favoured by many, marginally lower temperatures can be found.
Higher temperatures up to 8°C or so are often experienced where storage tanks are not independently cooled and are in a temperature cooled room. However, in this case, slurry temperature is also influenced by factors such as transfer temperature, ambient temperature of the room, and storage time. Of course, the driver behind cold storage is to minimise the same yeast activity that occurs in the fermenter cone, i.e. glycogen breakdown through fermentation to ethanol and accompanying metabolic heat. The comparatively poor temperature control in cold rooms can result in a vicious cycle where, as the slurry temperature increases, this kick-starts further metabolism and heat which becomes increasingly self perpetuating.
Effective mixing via an internal rouser or external recirculation loop is an important weapon against the build up of yeast 'hot spots'. As ever, a balance must be struck between mixing speed and efficacy against the damaging effects to the cell of shear. Whilst a contributor to yeast autolysis, susceptibility to 'shear' requires careful assessment, both in terms of process (tank size, shape, rouser positioning) and yeast (susceptibility to autolysis).

ACID WASHING

Acid washing might be a 'short and sharp shock', but it has the potential to deliver as much – if not more – damage to yeast than cropping or storage! Returning to our mantra, if I were a yeast cell, acid washing would worry me the most!
Get it right and its phosphoric acid, pH 2.1, preferably 2°C with mixing, for a couple of hours maximum. Get it wrong, particularly time and/or temperature, and cell physiology will be struggling and viability will inevitably drop. Regrettably in today's world, poor acid washing is either missed or is the 'way things are' and its impact on yeast quality is not known nor compensated for. So from the perspective of 'handle with care', it is best not to acid wash yeast. If hygiene issues dictate, acid wash as necessary, rather than as a batch to batch routine.
Remember acid washing only targets bacteria (and only those that are acid sensitive) and not contaminating yeasts. Indeed, it ➜

is a salutary lesson that non-Saccharomyces wild yeast tolerates acid washing better than brewing yeasts.

YEAST GENERATION NUMBER

From a yeast perspective, the modern brewery is a hostile environment. Even if every last page of the manuals of best practice is read and applied, some deterioration of yeast is bound to occur.

Of course, the effects can be minimised, but the truth is that contrary to much popular opinion, brewing yeasts are not stable. They are prone to genetic drift and the procedures that are used to ferment, crop, and store pitching yeast can select for genetic variants.

The visible effects of this variation can be very apparent. For example, sudden and permanent shifts in flocculence are by no means rare.

If this occurs, the offending variant is relatively easy to identify and eliminate. The effects of other genetic shifts might be much more subtle and, therefore, not as easy to spot.

Who is to say what the effects of such variability on product and process consistency might be?

In this case, prevention is better than cure. The maximum number of permissible re-pitchings needs to be set in relation to the degree of hostility of the process being used. In the case of very large capacity and deep conicals, fermenting high gravity worts at warm temperatures probably fewer rather than more generations is prudent.

How many re-pitchings is a tricky question. Typically these days, lager strains can be capped with as few as four generations or as many as 12-15.

CONCLUSIONS

Many companies are proud to declare that they are 'Investors in People'. It is obviously sensible to have a committed, skilled, and motivated workforce. Of course, in brewing, it is the yeast that actually pays everyone's wages. We hope that we have shown that having an 'Investors in Yeast' sign displayed outside the brewery gates might not be a bad idea.

Chris Boulton and David Quain can be found agitating for better yeast processing at red.ts Ltd, a drinks industry consultancy based in Burton-on-Trent, England.

CLIP AND SAVE

B&Q's top tips for looking after your yeast

Supply	• *Master cultures stored in liquid nitrogen (in house or outsourced) and supplied as slopes/plates (not freeze dried!).* • *QA approach to assure the yeast is the right strain and free from contaminants.* • *Use slopes once.*
Propagation	• *Demonstrably aerobic with mixers and forced aeration/oxygenation.* • *Controlled to maintain minimal, but detectable levels of dissolved oxygen.* • *First generation wort 'sized' to ensure normal pitching rate (and thereby minimise stress).*
Pitching	• *Ensure consistent and assured viable yeast pitching rate.* • *When collecting multi-brew lengths, pitch all yeast as early as possible.*
Fermentation	• *Mixing is good. Where possible, use in situ rousers or recirculation loops.* • *In conventional vessels without 'forced' mixing, optimise temperature and pitching rate to achieve vigorous fermentation and, therefore, good mixing.*
Cropping	• *Minimise storage in the vessel cone by warm cropping as early as possible post attenuation, particularly in large vessels with steep cone angle (60°).*
Storage	• *Control yeast slurry (<50% solids) and store at 2-4°C with continuous mixing.* • *Reduce ABV by dilution with microbiologically sound water.* • *Where a top pressure gas is used, ensure it is inert (i.e. not oxygen).*
Acid washing	• *Used only when the yeast is contaminated by bacteria, rather than a routine treatment.* • *Use phosphoric acid (pH 2.1-2.4) at 2-4°C with continuous mixing for a maximum of two hours.* • *If at all possible, adjust ABV by dilution with water prior to acid washing to <6%v/v.*
Generations	• *Minimise the opportunity for genetic variants (seen and unseen!) to accumulate by limiting the number of generations. How many? Two schools of thought: 4-6 generations and 12-15 generations.*