# BEER

The word beer comes from the Latin *bibere* (to drink). It is a beverage whose history can be traced back between 6000 and 8000 years and the process, whilst increasingly regulated and well-controlled because of tremendous strides in the understanding of it, has remained unchanged for hundreds of years. The basic ingredients for most beers are malted barley, water, hops and yeast; indeed the 500+-year old Bavarian purity law (the *Reinheitsgebot*) restricts brewers to these ingredients for beer to be brewed in Germany. Most other Brewers worldwide have much greater flexibility in their production process opportunities, yet the largest companies are ever mindful of the importance of tradition.

Compared to most other alcoholic beverages, beer is relatively low in alcohol. The highest average strength of beer (Alcohol by volume [ABV] indicates ml of ethanol per 100 ml of beer) in any country worldwide is 5.1% and the lowest is 3.9%. By contrast, the ABV of wines is typically in the range 11–15%.

## History

In 1992, a research team from the Applied Science Center for Archaeology at the University of Pennsylvania made a discovery that was publicized within archaeology circles (and college campuses) around the world. These researchers had analyzed a small amount of organic residue from inside an ancient pottery vessel that had been retrieved from the Zagros Mountains of Western Iran. When the residue from this clay pot (which itself was dated circa 3500-3100 B.C.E.) was analyzed, the results revealed the presence of oxalate ion. Since oxalate, and calcium oxalate in particular, collect in only a few places, this finding could mean only one thing—this pot had been used to brew beer. The 5,000-year-old residue, the archaeologists concluded, was the earliest chemical evidence for the origins of beer in the world.

Although this discovery attracted its share of headlines, it was but one of many events in the remarkable historical record of beer making (Figure 1). Beers were widely prepared and consumed in Egypt and other Middle Eastern counties from ancient days and then spread west into Europe and the British Isles. Despite the introduction of Islam in the eighth century, and its prohibition against alcohol consumption, beer making and consumption continued to grow throughout Europe during the Middle Ages.

During that time, beer making was considered an art, performed by skilled craftsmen. Many of the early breweries were located within various European monasteries, which created a tradition of brewing expertise and innovation. The use of hops in beer as a flavoring agent, for example, was first practiced by monks, as was the use of bottom-fermenting yeasts. Although beer manufacture was practiced throughout Europe, it was of particular cultural and economic importance in Great Britain and Germany, which became the epicenters for brewing technology. By the late eighteenth century, and especially by the mid-1800s, beer making was one of the first food processes to become industrialized. Although monastery-based breweries were still common, many small breweries began to form, serving their product directly on the premises

(much like the brew pubs that are popular even today). Eventually, some of the larger breweries contained not only production facilities, but also the beginning of what we might now consider to be quality control laboratories. And although these breweries were located mainly in Europe and England, beer making had also spread to North America and the American colonies.

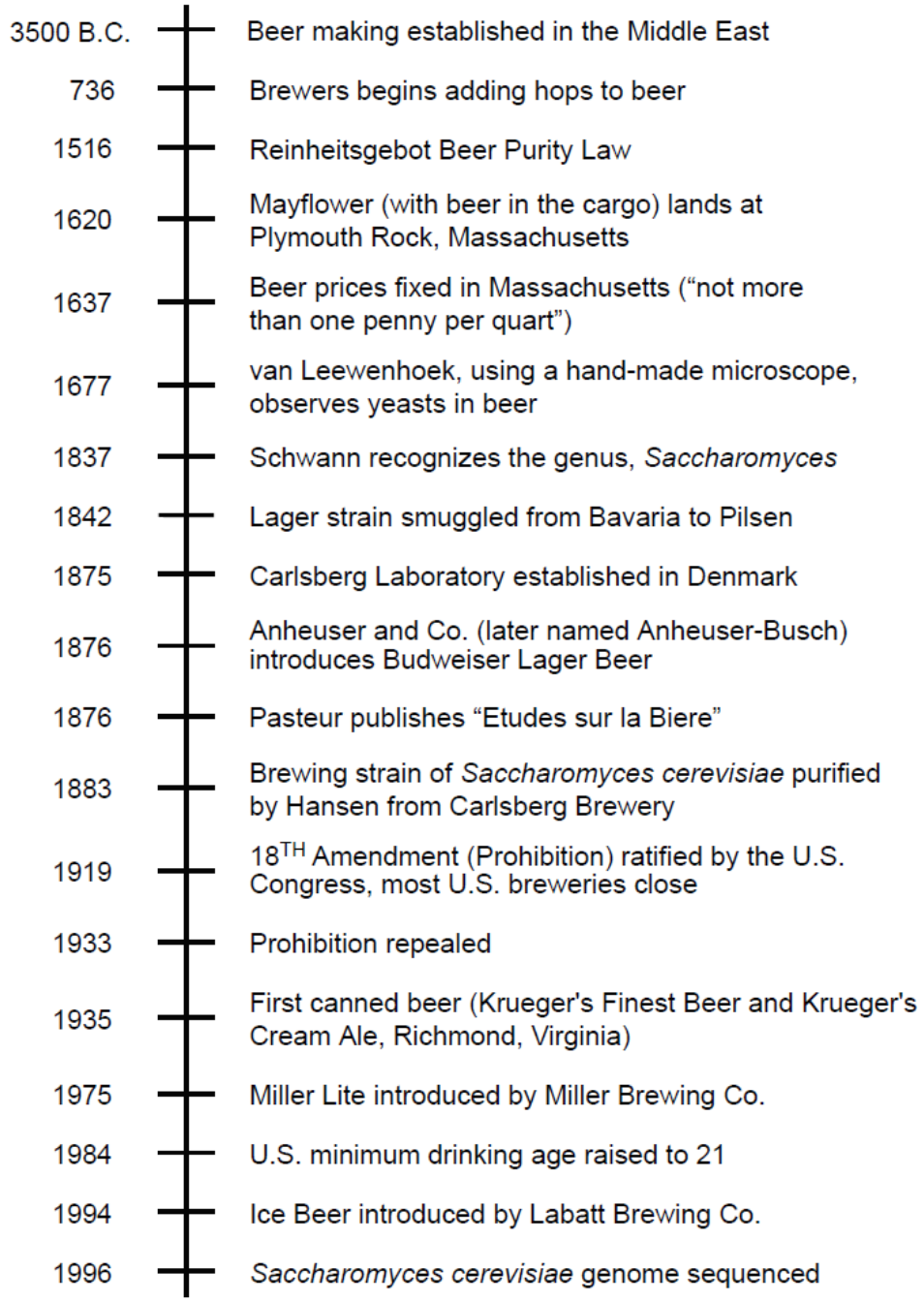



Figure 1. Milestones in the history of beer.

In fact, English ale-style beer had been among the provisions carried by Pilgrims on the Mayflower, and there are reports, written in the actual travel logs, of the ship running low on beer and the worries that caused the passengers. ("We could not now take time for further search . . . our victuals being much spent, especially our beer. . .") During the latter half of the nineteenth century, the number of breweries in the United States increased three-fold, from 400 to 1,300. Most of these breweries produced German lager-style beer (see below), reflecting the huge immigrant population from Germany during that era (including the brewers themselves, e.g.,Adolphus Busch, Eberhard Anheuser, Adolph Coors, Frederick Miller, Joseph Schlitz).

**Overview of Malting and Brewing**

An overview of the malting and brewing processes is shown in Figure 2. Brewers yeast *Saccharomyces cerevisiae* can grow on sugar anaerobically by fermenting it to ethanol:

$$C_6H_{12}O_6 \longrightarrow 2C_2H_5OH + 2CO_2$$

Whilst *malt* and *yeast* contribute substantially to the character of beers, the quality of beer is at least as much a function of the *water* and, especially, of the *hops* used in its production.

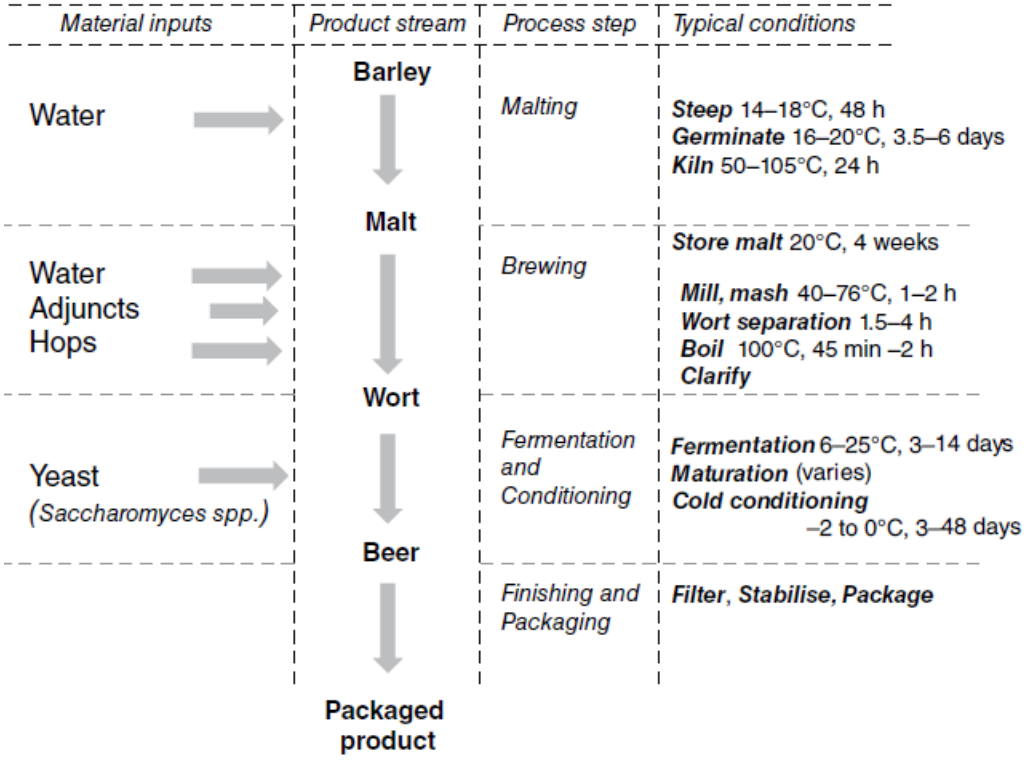



Figure 2. An overview of malting and brewing.

Barley starch supplies most of the sugars from which the alcohol is derived in the majority of the world's beers. Historically, this is because, unlike other cereals, barley retains its husk on threshing and this husk traditionally forms the filter bed through which the liquid extract of sugars is separated in the brewery. Even so, some beers are made largely from wheat, others

from sorghum or cassava, including gluten-free beers made with these and other cereals and pseudo-cereals.

The starch in barley is enclosed in a cell wall and proteins and these wrappings are stripped away in the malting process (essentially a limited germination of the barley grains), leaving the starch largely preserved. Removal of the cell wall framework softens the grain and makes it more readily milled. Not only that, unpleasant grainy and astringent characters are removed during malting.

In the brewery, the malted grain must first be *milled* to produce relatively fine particles, which are for the most part starch. The particles are then intimately mixed with hot water in a process called *mashing*. The water must possess the right mix of salts. For example fine ales are produced from waters with high levels of calcium whilst famous pilsners are brewed from waters with low levels of calcium. Typically mashes have a thickness of three parts water to one part malt and contain a stand at around 65 °C, at which temperature the granules of starch are converted by gelatinization from an indigestible granular state into a 'melted' form that is much more susceptible to enzymatic digestion. The enzymes that break down the starch are called the amylases. They are developed during the malting process, but only start to act once the gelatinization of the starch has occurred in the mash tun. Some brewers will have added starch from other sources, such as maize (corn) or rice, to supplement that from malt. These other sources are called *adjuncts*. After perhaps an hour of mashing, the liquid portion of the mash, known as *wort*, is recovered, either by straining through the residual *spent grains* or by filtering through membrane filter plates. The wort is run to the *kettle* (sometimes known as the *copper*, even though they are nowadays mostly fabricated from stainless steel) where it is boiled, usually for around one hour. *Boiling* serves various functions, including sterilisation of wort, precipitation of proteins (which would otherwise come out of solution in the finished beer and cause cloudiness), and the driving away of unpleasant grainy characters originating in the barley. Many brewers also add some adjunct sugars at this stage, at which most brewers introduce at least a proportion of their *hops*.

The hops have two principal components: resins and essential oils. The resins (so-called α-acids) are changed ('isomerised') during boiling to yield iso-α-acids, which impart the bitterness to beer. This process is rather inefficient. Nowadays, hops are often extracted with liquefied carbon dioxide and the extract is either added to the kettle or extensively isomerised outside the brewery for addition to the finished beer (thereby avoiding losses due to the tendency of the bitter substances to stick on to yeast and be lost due to over-foaming, or in the insoluble 'trub' formed during wort boiling). The oils are responsible for the 'hoppy nose' on beer. They are very volatile and if the hops are all added at the start of the boil then all of the aroma will be blown up the chimney (stack). In traditional lager brewing a proportion of the hops is held back and only added towards the end of boiling, which allows the oils to remain in the wort. For obvious reasons, this process is called *late hopping*. In traditional ale production, hops are added at the end of the process, enabling a complex mixture of oils to give a distinctive character to such products. This is called *dry hopping*. Liquid carbon dioxide can be used to extract oils as

well as resins and these extracts can also be added late in the process to make modifications to beer flavour.

After the precipitate produced during boiling (‘hot break’, ‘trub’) has been removed, the hopped wort is cooled and *pitched* with yeast. There are many strains of brewing yeast and Brewers carefully look after their own strains because of their importance in determining brand identity. Fundamentally brewing yeast can be divided into ale and lager strains, the former type collecting at the surface of the fermenting wort and the latter settling to the bottom of a fermentation (although this differentiation is becoming blurred with modern fermenters). Both types need a little oxygen to trigger off their metabolism, but otherwise the alcoholic *fermentation* is anaerobic. Ale fermentations are usually complete within a few days at temperatures as high as 20 °C, whereas lager fermentations at as low as 6 °C can take several weeks. Fermentation is complete when the desired alcohol content has been reached and when an unpleasant butterscotch/ popcorn flavour that develops during all fermentations has been mopped up by yeast. The yeast is harvested for use in the next fermentation.

In *traditional ale brewing* the beer is now mixed with hops, some priming sugars and with isinglass finings from the swim bladders of certain fish, which settle out the solids in the cask.

In *traditional lager brewing* the ‘green beer’ is matured by several weeks of cold storage, prior to filtering.

Nowadays, the majority of beers, both ales and lagers, receive a relatively short *conditioning* period after fermentation and before filtration. This conditioning is ideally performed at −1 °C or lower (but not so low as to freeze the beer) typically for three days, under which conditions more proteins drop out of solution, rendering the beer less likely to go cloudy in the package or glass.

The filtered beer is adjusted to the required carbonation before packaging into cans, kegs or glass or plastic bottles.

**Barley and Malt Production**

Although it is possible to make beer using raw barley and added enzymes (so-called ‘barley brewing’) this is extremely unusual. Unmalted barley alone is unsuitable for brewing beer because

i. it is hard and difficult to mill,
ii. it lacks most of the enzymes needed to produce fermentable components in wort,
iii. it contains complex viscous materials that slow down solid–liquid separation processes in the brewery and that may cause clarity problems in beer, and
iv. it contains unpleasant raw and grainy characters and is devoid of the pleasing flavours associated with malt.

Barley belongs to the grass family. Its Latin name is *Hordeum vulgare*, though this term tends to be retained for 6-row barley (see later), with *Hordeum distichon* being used for 2-row

barley. The part of the plant of interest to the brewer is the grain on the ear. Sometimes this is referred to as the seed, but individual grains are generally called kernels or corns. A schematic diagram of a single barley corn is shown in Figure 3.

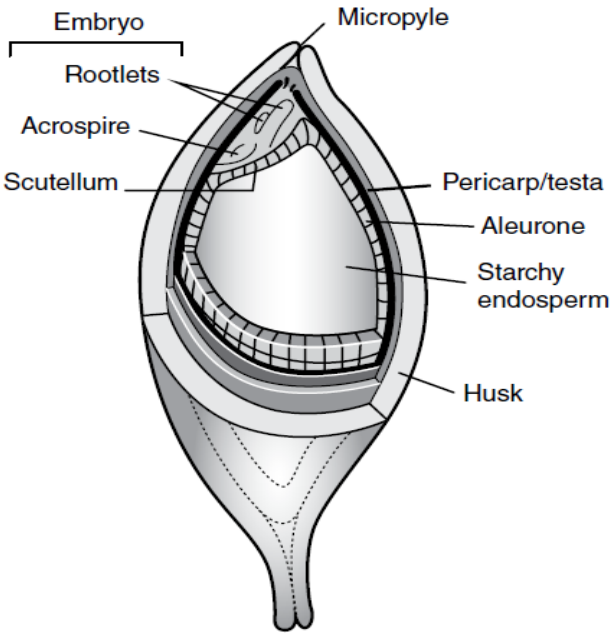



Figure 3. A barley corn.

Four components of the kernel are particularly significant:

- the embryo, which is the baby plant
- the starchy endosperm, which is the food reserve for the embryo
- the aleurone layer, which generates the enzymes that degrade the starchy endosperm
- the husk (hull), which is the protective layer around the corn. Barley is unusual amongst cereals in retaining a husk after threshing and this tissue is traditionally important for its role as a filter medium in the brewhouse when the wort is separated from spent grains.

The first stage in malting is to expose the grain to water, which enters an undamaged grain principally through the micropyle and progressively hydrates the embryo and endosperm. This switches on the metabolism of the embryo, which sends hormonal signals to the aleurone layer, which triggers that to switch on the synthesis of enzymes responsible for digesting the components of the starchy endosperm. The digestion products migrate to the embryo and sustain its growth.

The aim is controlled and consistent germination, to soften the grain, remove troublesome materials and expose starch without promoting excessive growth of the embryo that would be wasteful (*malting loss*). The three stages of commercial malting are:

- *steeping*, which brings the moisture content of the grain to a level sufficient to allow metabolism to be triggered in the grain.
- *germination*, during which the contents of the starchy endosperm are substantially degraded ('modification') resulting in a softening of the grain.
- *kilning*, a hot-air drying process in which the moisture is reduced to a level low enough to arrest modification.

The embryo and aleurone are both living tissues, but the starchy endosperm is dead. It is a mass of cells, each of which comprises a relatively thin cell wall (approx. 2 μm) inside which are packed many starch granules amidst a matrix of protein (see Figure 4). This starch and protein (and also the cell wall materials) are the food reserves for the embryo. However, the brewer's interest in them is as the source of fermentable sugars and assimilable amino acids that the yeast will use during alcoholic fermentation.

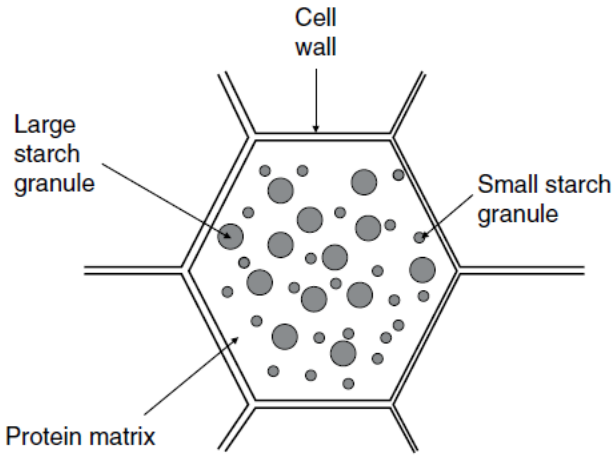



Figure 4. A single cell within the starchy endosperm of barley. Only a very small number of the multitude of small and large starch granules are depicted.

The wall around each cell of the starchy endosperm comprises (approximately) 75% β-glucan, 20% pentosan, 5% protein and some acids, notably acetic acid and the phenolic acid ferulic acid. The β-glucan comprises long linear chains of glucose units joined through β-linkages. Approximately 70% of these linkages are between C-1 and C-4 of adjacent glucosyl units (so-called β 1–4 links, just as in cellulose) and the remainder are between C-1 and C-3 of adjacent glucoses (β 1–3 links, which are not found in cellulose). These 1–3 links disrupt the structure of the β-glucan molecule and make it less ordered, more soluble and digestible than cellulose. Much less is known about the pentosan (arabinoxylan) component of the wall, and it is generally believed that it is less easily solubilised and difficult to break down when compared to the β-glucan. The cell wall polysaccharides are problematic because they restrict the yield of extract. They do this either when they are insoluble (by wrapping around the starch components) or when they are solubilised (by increasing the viscosity, thereby slowing the flow of wort from

spent grains during wort separation). Dissolved but undegraded β-glucans also slow down the filtration of beer. They are prone to drop out of solution as hazes, precipitates or gels. Conversely it has been claimed that β-glucans have positive health attributes for the human, by lowering cholesterol levels and contributing to dietary fibre, although as arabinoxylan is much less degraded than β-glucan it is rather more important in this regard.

The enzymic breakdown of β-glucan during the germination of barley and later in mashing is in two stages: solubilisation and hydrolysis. Several enzymes (collectively the activity is referred to by the trivial name 'solubilase') may be involved in releasing β-glucan from the cell wall, including esterases that hydrolyse ester bonds believed to cement polysaccharides, perhaps to the protein-rich middle lamella. The most recent evidence, however, is that the pentosan component encloses much of the glucan (Figure 5), and accordingly xylanases are efficient solubilases. This is despite the observations that pentosans are less digestible than glucans. B-Glucans are hydrolysed by endo β-glucanases (endo enzymes hydrolyse bonds within a polymeric molecule, releasing smaller units, that are subsequently broken down by exo enzymes that chop off one unit at a time, commencing at one end of the molecule). These enzymes convert viscous β-glucan molecules to non-viscous oligosaccharides comprising three or four glucose units. Less well understood enzymes are responsible for converting these oligosaccharides to glucose. There is little β-glucanase in raw barley, it being developed during the germination phase of malting in response to gibberellins. Endo-β-glucanase is extremely sensitive to heat, meaning that it is essential that malt is kilned very carefully to conserve this enzyme if it is necessary that it should complete the task of glucan degradation in the brewhouse. This is especially important if the brewer is using β-glucan-rich adjuncts such as unmalted barley, flaked barley and roasted barley. It is also the reason why brewers often employ a low temperature start to their mashing processes. Alternatively some brewers add exogenous heat-stable β-glucanases of microbial origin.

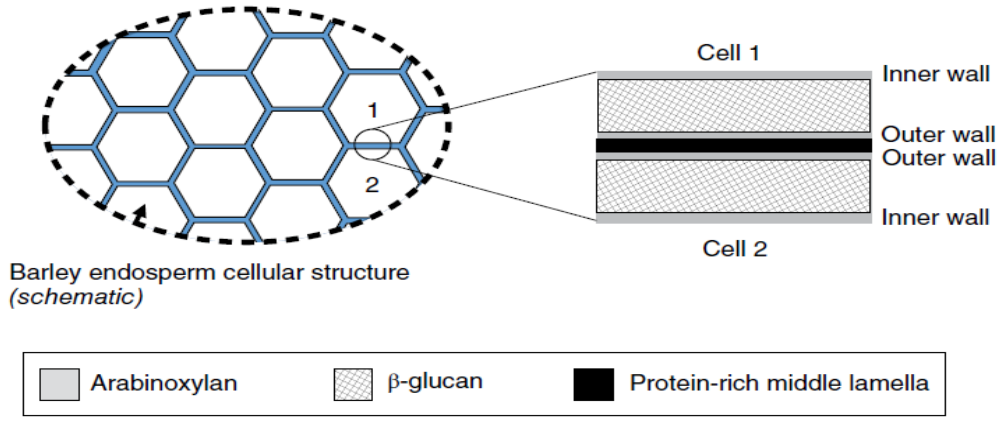



Figure 5. Current understanding of the structure of the cell walls of barley endosperm. Walls surrounding adjacent cells are cemented by a protein-rich middle lamella. To this is attached arabinoxylan, within which is the β-glucan.

The *starch* in the cells of the starchy endosperm is in two forms: large granules (approx. 25 μm in diameter) and small granules (5 μm). The structure of granules is quite complex, having crystalline and amorphous regions.

The *proteins* in the starchy endosperm may be classified according to their solubility characteristics. The two most relevant classes are the *albumins* (water-soluble) and the *hordeins*. In the starchy endosperm of barley the latter are quantitatively the most significant: they are the storage proteins. They need to be substantially degraded in order that the starch can be accessed and amino acids (which will be used by the yeast) generated. Their partial degradation products can also contribute to haze formation via cross-linking with polyphenols. Excessive proteolysis should not occur, however, as some partially degraded protein is required to afford stable foam to beer. Most of the proteolysis occurs during germination rather than subsequent mashing, probably because endogenous molecules that can inhibit the endo-proteinases are kept apart from these enzymes by compartmentalisation in the grain, but when the malt is milled this disrupts the separation and the inhibitors can now exert their effect. There may be some ongoing protein extraction and precipitation during mashing, and peptides are converted into amino acids at this stage through the action of carboxypeptidases. The endo-peptidases are synthesised during germination in response to gibberellin and they are relatively heat-labile (like the endo-β-glucanases). Substantial carboxypeptidase is present in raw barley and it further increases to abundant levels during germination. It is a heat-resistant enzyme and is unlikely to be limiting. Thus the extent of protein degradation is largely a function of the extent of proteinase activity during germination.

Much effort is devoted to breeding malting barleys that give high yields of 'extract' (i.e. fermentable material dissolved as wort). The hygiene status of the barley is also very important, and pesticide usage may be important to avoid the risk of infection from organism such as Fusarium. Barleys may be 2-row, in which only one kernel develops at each node on the ear and it appears as if there is one kernel on either side of the axis of the ear, or 6-row in which there are three corns per node. Obviously there is less room for the individual kernels in the latter case and they tend to be somewhat twisted and smaller and therefore less desirable. Farmers are restricted in how much nitrogenous fertiliser they can use because the grain will accumulate protein at the expense of starch in the endosperm and it is the starch (ergo fermentable sugar) that is especially desirable. Maltsters pay a 'malting premium' for the right variety, grown to have the desired level of protein. There must be some protein present, as this is the fraction of the grain that includes the enzymes and that is the origin of amino acids (for yeast metabolism) and foamable polypeptides. The amount of protein needed in malt will depend on whether the brewer intends to use some adjunct material as a substitute for malt. For example, corn syrup is a rich source of sugar but not of amino acids, which will need to come from the malt. Dead grain will not germinate, so batches of barley must pass viability tests.

Most barley in the Northern Hemisphere is sown between January and April and is referred to as Spring Barley. The earlier the sowing, the better the yield and lower protein levels because starch accumulates throughout the growing season. In locales with mild winters some

varieties (Winter Barleys) are sown in September and October. Best yields of grain are in locales where there is a cool damp growing season allowing steady growth, and then fine dry weather at harvest to ripen and dry the grain. Grain grown through very hot dry summers is thin, poorly filled, and with high nitrogen. Malting barley is grown in many countries (Table 2.1).

Table 1. World production of barley (2017).

| Countries | Production (1000 tonnes) |
|---|---|
| World | 137 470 |
| European Union | 59 500 |
| Canada | 7600 |
| Russian Fed. | 17 000 |
| Turkey | 5500 |
| USA | 3462 |
| Argentina | 3400 |
| Ukraine | 7400 |
| Australia | 8000 |

Grain arrives at the maltings by road or rail and, as the transport waits, the barley will be weighed and a sample tested for grain size distribution, viability, nitrogen content and moisture. Expert inspection at intake will also assess factors such as the appearance and smell of the barley, varietal purity and the absence of insect pests, signs of mould infestation or undue amounts of foreign bodies (e.g. weeds, stones). Once accepted, the barley will be cleaned and screened to remove small grain and dust, before passing into a silo, perhaps via a drying operation in areas with damp climates. Grain should be dry to counter infection and premature growth.

It is essential that the barley store is protected from the elements, yet it must also be ventilated, because barley, like other cereals, is susceptible to various infections, e.g. Fusarium, storage fungi such as Penicillium and Aspergillus, Mildew, and pests, e.g. aphids and weevils.

Steeping is probably the most critical stage in malting. If homogeneous malt is to be obtained (which will go on to ‘behave’ predictably in the brewery), then the aim must be to hydrate the kernels in a batch of barley evenly. Steeping regimes are determined on a barley-by-barley basis by small scale trials but most varieties need to be taken to 42–46%. Apart from water, barley needs oxygen in order to support respiration in the embryo and aleurone. Oxygen access is inhibited if grain is submerged for excessive periods in water, which phenomenon led directly to the use of interrupted steeping operations. Rather than submerge barley in water and leave it, grain is steeped for a period of time, before removing the water for a so-called ‘air-rest’ period. Then a further steep is performed and so on. Air rests serve the additional purpose of removing carbon dioxide and preventing the build-up of ethanol via anaerobic metabolism, either of which will suppress respiration. A typical steeping regime may involve an initial steep to 32-

38% moisture. The start of germination is prompted by an air rest of 10–20 hours, followed by a second steep to raise the water content to 40–42%. Emergence of the root tip (‘chitting’) is encouraged by a second air rest of 10–15 hours, before the final steep to the target moisture. The entire steeping operation may take 36–52 hours and procedures do vary depending on the steeping equipment used and the needs of a particular batch of barley. There is an increasing tendency for environmental and economic reasons to use barley varieties that demand shorter and fewer steeps.

Gibberellic acid (GA, itself produced in a commercial fermentation reaction from the fungus *Gibberella*) is added in some parts of the world to supplement the native gibberellins of the grain. Although some users of malt prohibit its use, GA can successfully accelerate the malting process. It tends to be sprayed on to grain at levels between 0.1 and 0.5 ppm as it passes from the last steep on its way to the germination vessel.

The hormones migrate to the aleurone to regulate enzyme synthesis, for the most part to promote the synthesis of enzymes that break down successively β-glucan, protein and starch. The gibberellin first reaches the part of the aleurone nearest to the embryo and therefore enzyme release is initially into the proximal endosperm. Breakdown of the endosperm (‘modification’) therefore passes in a band from proximal to distal regions of the grain.

Traditionally, steeped barley was spread out to a depth of up to 10 cm on the floors of long low buildings and germinated for periods up to 10 days. Rakes would be used to either thin out the grain (‘the piece’) or pile it up depending on whether the batch needed its temperature lowering or raising: the aim was to maintain it at 13–16 °C. Very few such floor maltings survive because of their labour intensity and a diversity of pneumatic (mechanical) germination equipment is now used, although in the current so-called ‘craft’ era there is something of a resurgence of these traditional approaches to malting. Newer germination vessels are circular, of steel or concrete, with capacities of as much as 500 t and with turning machinery that is microprocessor-controlled. Modern malting plant is often conveniently arranged in a tower format, with vessels vertically stacked, steeping tanks uppermost.

Germination in pneumatic plant is generally at 16–20 °C. Once the whole endosperm is readily squeezed out and if the shoot initials (the acrospire) are about three-quarters the length of the grain (the acrospire grows the length of the kernel between the testa and the aleurone and emerges from the husk at the distal end of the corn), then the ‘green malt’ is ready for kilning.

Through the controlled drying (kilning) of green malt, the maltster is able to

- arrest modification and render malt stable for storage;
- ensure survival of enzymes for mashing;
- introduce desirable flavour and colour characteristics and eliminate undesirable flavours; and
- render the rootlets brittle which facilitates their subsequent removal (mechanically) and avoidance of bitter off-flavours that would otherwise be imparted to beer.

Drying should commence at a relatively low temperature to ensure survival of the most heat-sensitive enzymes (enzymes are more resistant to heat when the moisture content is low). This is

followed by a progressive increase of temperature to effect the flavour and colour changes (Maillard reaction) and complete drying within the limited turnaround time available (typically under 24 hours). There are many different kiln designs, but most modern ones feature beds of malt supported on a wedge-wire floor that permits air to pass through the bed. Typical bed depths are 0.9–1.2 m and represent a balance between the homogeneity of malt produced, the power requirements to force drying air through the bed and the load capacity. They have a source of heat for warming incoming air, a fan to drive or pull the air through the bed, together with the necessary loading and stripping systems.

Newer kilns also use 'indirect firing', in that the products of fuel combustion don't pass through the grain bed, but are sent to exhaust, the air being warmed through a heater battery containing water as the conducting medium. Indirect firing arose because of concerns with the role of oxides of nitrogen present in kiln gases that might have promoted the formation of nitrosamines in malt. Nitrosamine levels are now seldom a problem in malt.

Lower finishing temperatures will give malts of lighter colour and will tend to be employed in the production of malts destined for lager-style beers. Higher temperatures, apart from giving darker malts, also lead to a wholly different flavour spectrum. Lager malts give beers that are relatively rich in sulphur compounds, including dimethyl sulphide (DMS). Ale malts have more roasted, nutty characters. For both lager and ale malts, kilning is sufficient to eliminate the unpleasant raw, grassy and beany characters associated with green malt.

When kilning is complete, the heat is switched off and the grain allowed to cool before it is stripped from the kiln in a stream of air at ambient temperatures. On its way to steel or concrete hopper-bottomed storage silos, the malt is 'dressed' to remove dried rootlets, which go to animal feed.

Some malts are produced not for their enzyme content but rather for use by the Brewer in relatively small quantities as a source of extra colour and distinct types of flavour. They may also be useful sources of natural antioxidant materials. There is much interest in these products for the opportunities they present for brewing new styles of beer.

**Mashing: The Production of Sweet Wort**

Sweet wort is the sugary liquid that is extracted from malt (and other solid adjuncts used at this stage) through the processes of milling, mashing and wort separation. Larger breweries will have raw materials delivered in bulk (rail or road) with increasingly sophisticated unloading and transfer facilities as size increases. Smaller breweries will have malt etc. delivered by sack. Railcars may carry up to 80 t of malt and a truck 20 t. The conscientious brewer will check the delivery and the vehicle it came in for cleanliness and will representatively sample the bulk. The resultant sample will be inspected visually and smelled before unloading is permitted. Most breweries will spotcheck malt deliveries for key analytical parameters to enable them to monitor the quality of a supplier's material against the agreed contractual specification. Grist materials are stored in silos sized according to brewhouse throughput.

### *1. Milling*

Before malt or other grains can be extracted they must be milled. Fundamentally the more extensive the milling the greater the potential there is to extract materials from the grain. However, in most systems for separating wort from spent grains after mashing, the husk is important as a filter medium. The more intact the husk, the better the filtration. Therefore milling must be a compromise between thoroughly grinding the endosperm whilst leaving the husk as intact as possible.

There are fundamentally two types of milling: dry milling and wet milling. In the former, mills may either be roll, disk or hammer. If wort separation is by a lauter tun, then a roller mill is used. If a mash filter is installed then a hammer (or disk) mill may be employed because the husk is much less important for wort separation by a mash filter. Wet milling, which was adopted from the corn starch process, was introduced into some brewing operations as an opportunity to minimise damage to the husk on milling. By making the husk 'soggy' it is rendered less likely to shatter than would a dry husk.

### *2. Mashing*

Mashing is the process of mixing milled grist with heated water in order to digest the key components of the malt and generate wort containing all the necessary ingredients for the desired fermentation and aspects of beer quality. Most importantly it is the primary stage for the breakdown of starch.

The starch in the granules is very highly ordered, which tends to make the granules difficult to digest. When granules are heated (in the case of barley starch beyond 55–65 °C) the molecular order in the granules is disrupted in a process called gelatinization. Now that the interactions (even to the point of crystallinity) within the starch have been broken down, the starch molecules become susceptible to enzymic digestion. It is for the purpose of gelatinization and subsequent enzymic digestion that the mashing process in brewing involves heating.

Although 80–90% of the granules in barley are small, they only account for 10–15% of the total weight of starch. The small granules are substantially degraded during the malting process, whereas degradation of the large granules is restricted to a degree of surface pitting. (This is important, as it is not in the interests of the brewer [or maltster] to have excessive loss of starch, which is needed as the source of sugar for fermentation.)

The starch in barley (as in other plants) is in two molecular forms: amylose, which has very long linear chains of α 1–4 linked glucose units, and amylopectin, which comprises shorter chains of glucose units that are linked via α 1–6 branch points. Several enzymes are required for the complete conversion of starch to glucose. α-Amylase, which is an endo enzyme, hydrolyses the α 1–4 bonds within amylose and amylopectin. β-amylase, an exo enzyme, also hydrolyses α 1–4 bonds, but it approaches the substrate (either intact starch or the lower molecular weight 'dextrins' produced by α-amylase) from the non-reducing end, chopping off units of two glucoses (i.e. molecules of maltose). Limit dextrinase ('debranching enzyme') is the third key activity, attacking the α 1–6 linkages in amylopectin.

α-Amylase develops during the germination phase of malting. Compared with other malt enzymes it is extremely heat-resistant, and also present in very high activity, therefore it is capable of extensive attack, not only on the starch from malt but also that from adjuncts added in quantities of up to 50% or more. β-Amylase is already present in the starchy endosperm of raw barley, in an inactive form through its association with Protein Z. It is released during germination by the action of a protease (and perhaps a reducing agent). β-Amylase is considerably more heat-labile than α-amylase, and will be largely destroyed after 30–45 minutes of mashing at 65 °C. Limit dextrinase is similarly heat-sensitive. Furthermore it is developed much later than the other two enzymes, and germination must be prolonged if high levels of this enzyme are to be developed. It is present in several forms (free and bound): the bound form is both synthesised and released during germination. As with the proteinases, there are endogenous inhibitors of limit dextrinase in grain, and this is probably the main factor that determines that some 20% of the starch in most brews is left in the wort as non-fermentable dextrins. Although it is possible to contrive operations that will allow greater conversion of starch to fermentable sugar, in practice many brewers seeking a fully fermentable wort add a heat-resistant glucoamylase (e.g. from *Aspergillus*) to the mash (or fermenter). This enzyme has an exo action like β-amylase, but it chops off individual glucose units.

There are several types of mashing that can broadly be classified as infusion mashing, decoction mashing and temperature-programmed mashing. Whichever type of mashing is employed, the vessels these days are almost exclusively fabricated from stainless steel. What stainless steel loses in heat transfer properties is made up for in its toughness and ability to be cleaned thoroughly by caustic and acidic detergents.

Irrespective of the mashing system, most (apart from in wet milling operations) incorporate a device for mixing the milled grist with the water (which some brewers call ‘liquor’). This device, the ‘pre-masher’, can be of various designs, the classic one being the Steel’s masher, which was developed for the traditional infusion mash tun (Figure 6).

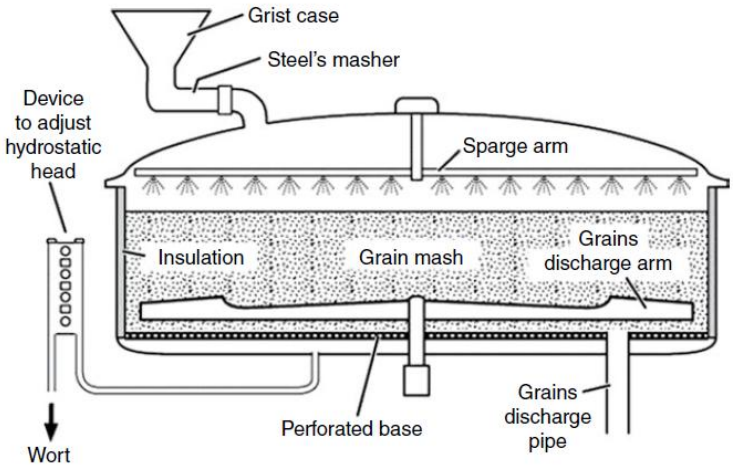



Figure 6. A mash tun.

*Infusion mashing* is relatively uncommon, but still championed by traditional brewers of ales. It was designed in England to deal with well- modified ale malts that did not require a low temperature start to mashing in order to deal with residual cell wall material (β-glucans). Grist is mixed with water (a typical ratio would be one part solid to three parts water) in a Steel's masher en route to the pre-heated mash tun, with a single holding temperature, typically 65 °C, being employed. This temperature facilitates gelatinization of starch and subsequent amylolytic action. At the completion of this 'conversion', wort is separated from the spent grains in the same vessel, which incorporates a false bottom and facility to regulate the hydrostatic pressure across the grains bed. The grist is sparged with further hot water ('liquor') to enable leaching of as much extract as possible from the bed.

*Decoction mashing* was designed on the mainland continent of Europe to deal with lager malts that were less well-modified than ale malts. Essentially it provides the facility to start mashing at a relatively low temperature, thereby allowing hydrolysis of the β-glucans present in the malt, followed by raising the temperature to a level sufficient to allow gelatinization of starch and its subsequent enzymic hydrolysis. The manner by which the temperature increase was achieved was by transferring a portion of the initial mash to a separate vessel where it was taken to boiling and then returned to the main mash, leading to an increase in temperature. This is a rather simplified version of the process, which traditionally involved several steps of progressive temperature increase.

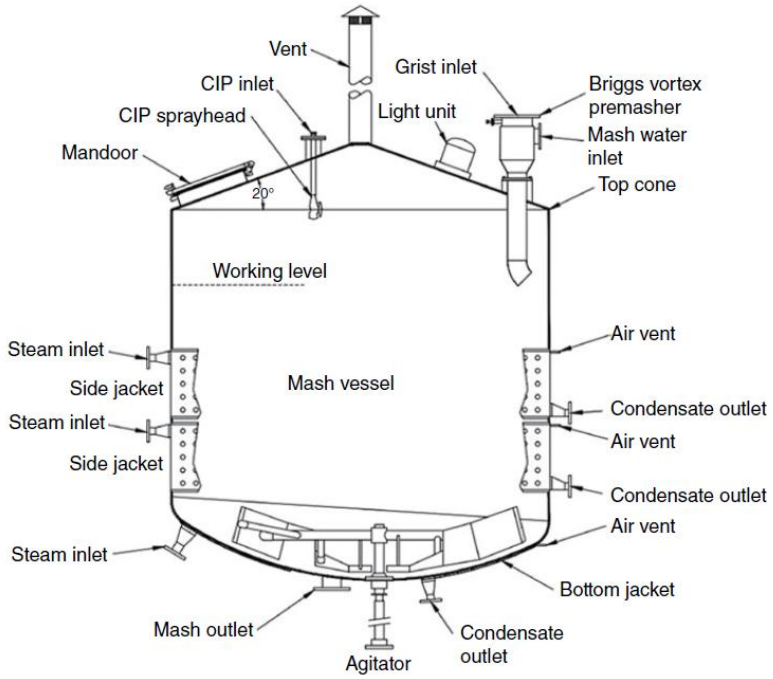



Figure 7. Layout of a modern design of mash conversion vessel (MCV).

*Temperature-programmed mashing*. Although there are some adherents to the decoction mashing protocol, most brewers nowadays employ the related but simpler temperature-programmed mashing. Again, the mashing is commenced at a relatively low temperature, but

subsequent increases in temperature are effected in a single vessel (Figure 7) by employing steam-heated jackets around the vessel to raise the temperature of the contents, which are thoroughly mixed to ensure even heat transfer. Mashing may commence at 45–50 °C, followed by a temperature rise of 1 °C per minute until the conversion temperature (63–68 °C) is reached. The mash will be held for perhaps 50 minutes to an hour, before raising the temperature again to the sparging temperature (76–78 °C). High temperatures are employed at the end of the process to arrest enzymic activity, facilitate solubilisation of materials and to reduce viscosity, thereby allowing more rapid liquid–solid separation.

### *3. Adjuncts*

The decision whether to use an adjunct is made on the basis of cost (does it represent a cost advantageous source of extract, compared to malted barley?) and quality (does the adjunct provide a quality benefit, in respect of flavour, foam or colour?). Liquid adjuncts (sugars/syrups) are added in the wort boiling stage (see later). A series of solid adjuncts may be added at the mashing stage, because they depend on the enzymes from malt to digest their component macromolecules. Solid adjuncts may be based on other cereals as well as barley: wheat, corn (maize), rice, oats, rye, sorghum. In turn, these adjuncts can be used in different forms (Table 2).

Table 2. Cereal 'mash' adjuncts and their different processed forms.

| Form of adjunct | Typical cereals utilised |
|---|---|
| Raw (unmalted) grains | Barley, corn, sorghum, wheat |
| Grits (uncooked endosperm fragments) | Barley, corn, rice, sorghum |
| Flour/starch | Corn, rice, sorghum, wheat |
| Flaked (e.g. steam-cooked, flattened and dried) | Barley, corn, oats, rice |
| Torrefied or Micronised | Barley, corn, wheat |
| Malted cereals other than barley | Oats, rye, sorghum, wheat |

Table 3. Gelatinization temperatures of starches from different cereals.

| Source | Gelatinization temp (°C) |
|---|---|
| Barley | 61–62 |
| Buckwheat | 75–95 |
| Corn | 70–80 |
| Millet | 67–77 |
| Oats | 55–60 |
| Rice | 70–80 |
| Rye | 60–65 |
| Sorghum | 70–80 |
| Wheat | 52–54 |

A key aspect of solid adjuncts is the gelatinisation temperature of the starch (Table 3). The higher gelatinisation temperatures for corn, rice, buckwheat, millet and sorghum starches mean that these cereals need treatment at higher temperatures than do barley, oats, rye or wheat. If the cereal is in the form of grits (produced by the dry milling of cereal in order to remove outer layers and the oil-rich germ), then it needs to be 'cooked' in the brew house. Alternatively the cereal can be pre-processed by intense heat treatment in a micronisation or flaking operation. In the former process the whole grain is passed by conveyor under an intense heat source (260 °C), resulting in a 'popping' of the kernels (cf. puffed breakfast cereals). In flaking, grits are gelatinised by steam and then rolled between steam-heated rollers. Flakes do not need to be milled in the brewhouse, but micronised cereal does.

Cereal cookers employed for dealing with grits are made of stainless steel and incorporate an agitator and steam jackets. The adjunct is delivered from a hopper and the adjunct will be mixed with water at a rate of perhaps 15 kg per hectolitre of water. The adjunct will be mixed with 10–20% of malt as a source of enzymes. The precise temperature employed in the cooker will depend on the adjunct and the preferences of the Brewer. Following cooking the adjunct mash is likely to be taken to boiling and then mixed with the main mash (at its mashing- in temperature), with the resultant effect being the temperature rise to conversion for the malt starch (cf. decoction mashing). This is sometimes called *Double mashing*.

### *4. Wort Separation*

Traditionally, recovering wort from the residual grains in the brewery is perhaps the most skilled part of brewing. Not only is the aim to produce a wort with as much extract as possible, but many brewers prefer to do this such that the wort is 'bright', i.e. not containing many insoluble particles that may present difficulties later. All this needs to take place within a time window, for the mashing vessel must be emptied in readiness for the next brew.

Irrespective of the system employed for mash separation (traditional infusion mash tun, lauter tun, or mash filter), the science dictating rate of liquid recovery is the same and is defined by Darcy's equation:

$$\text{rate of liquid flow} = \frac{\text{pressure} \times \text{bed permeability} \times \text{filtration area}}{\text{bed depth} \times \text{wort viscosity}}$$

Thus the wort will be recovered more quickly if the device used to separate the wort has a large surface area, is shallow and if a high pressure can be employed to force the liquid through. The liquid should be of as low viscosity as possible, as less viscous liquids flow more readily. Also the bed of solids should be as permeable as possible. Perhaps the best analogy here is to sand and clay. Sand comprises relatively large particles around which a liquid will flow readily. To pass through the much smaller particles of clay, though, water has to take a much more circuitous route and it is held up. The particle sizes in a bed of grains depends on certain factors, such as the fineness of the original milling and the extent to which the husk survived milling (see earlier). Furthermore, a layer (teig or oberteig) collects on the surface of a lauter bed, this being a

complex of certain macromolecules, including oxidatively cross-linked proteins, lipids and cell wall polysaccharides, and this layer has a very fine size distribution analogous to clay. (The oxidative cross-linking of the proteins is exactly akin to that involved in bread dough. However particle size also depends on the temperature, and it is known that at the higher temperatures used for wort separation (e.g. 78 °C) there is an agglomeration of very fine particles into larger ones past which wort will flow more quickly.

*4.1. Lauter Tun*

Generally this is a straight-sided round vessel with a slotted or wedged wire base and Run-off pipes through which the wort is recovered (Figure 8). Within the vessel there are arms that can be rotated about a central axis. These arms carry vertical knives that are used as appropriate to slice through the grain bed and facilitate run off of the wort. Hot water can be sparged onto the grain to ensure collection of all the desired soluble material. Following completion of sweet wort collection, the insoluble remnants, 'spent grains', are shipped off site to be used as cattle food.

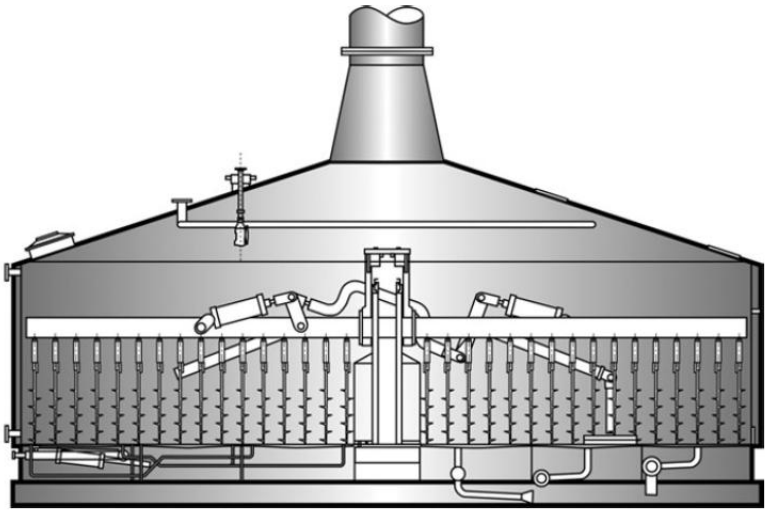


Figure 8. A lauter tun.

*4.2. Mash Filters*

Increasingly, modern breweries use mash filters. These operate by using plates of polypropylene fitted with cloths to filter the liquid wort from the residual grains. Accordingly, the husks serve no purpose as a filter medium and their particle sizes are irrelevant. Key steps in the operational cycle of a modern design of a Thin Chamber Mash Filter are shown in Figure 9. The high pressures that can be used in the squeezing of the plates together overcome the reduced permeability due to smaller particle sizes (the sand versus clay analogy used earlier). Furthermore, the grains bed depth is particularly shallow (2–3 in.), being nothing more than the distance between the adjacent plates.

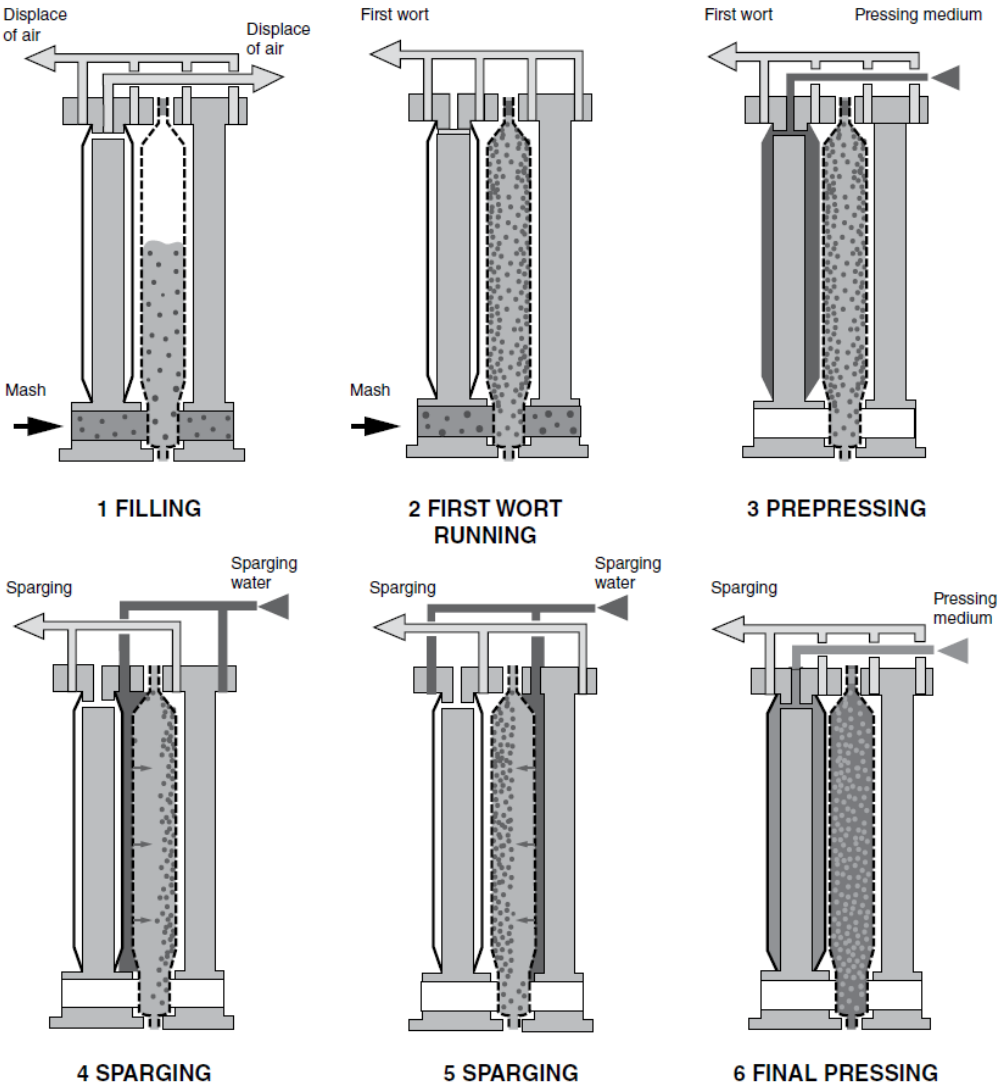



Figure 9. Steps in the operation of a Thin Chamber Mash filter.

**Water**

Because water represents at least 90% of the composition of most beers, it will clearly have a major direct impact on the product, in terms particularly of flavour and clarity. The nature of the water, however, exerts its influence much earlier in the process, through the impact of the salts it contains on enzymic and chemical processes, through the determination of pH etc.

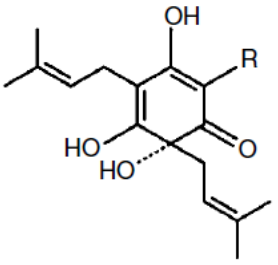
Water in breweries either comes from wells owned by the brewer or from municipal supplies; especially in the latter instance, the water will be subjected to clean-up procedures, such as charcoal filtration, to eliminate undesirable taints and colours.

The ionic composition of the water varies from place to place. Water may be very hard or soft. It is clear that the nature of the water has had some impact on the quality of the different beer styles traditionally produced in different places.

The water composition can be adjusted, either by adding or removing ions. Thus calcium levels may be increased in order to promote the precipitation of oxalic acid as oxalate, to lower the pH by reaction with phosphate ions ($3Ca_2+ + 2HPO_4^{2-} \rightarrow Ca_3(PO_4)^2+ 2H^+$) and to promote amylase action (the optimum pH for mashing is between 5.2 and 5.4). The alkalinity of water used for sparging (alkalinity is largely determined by the content of carbonate and bicarbonate) may be reduced to less than 50 ppm in order to limit the extraction of tannins. Ions such as iron and copper must be as low as possible to preclude oxidation. Furthermore water may need to be of different standards for different purposes. The microbiological status of water used for slurrying yeast or for use downstream generally is important. Water used for diluting high gravity streams must be of low oxygen content, and its ionic composition will be critical. When ions need to be removed the likeliest approach is either ion- exchange resin technology or reverse osmosis.

**Hops**

The hop, *Humulus lupulus*, is rich in resins (Figure 10) and oils (Figure 11), the former being the source of bitterness, the latter the source of aroma. The hop is remarkable amongst agricultural crops in that essentially its sole outlet is for brewing, although there is growing interest in hops as sources of anti-microbials and pharmacologically beneficial molecules. Hops are suited to cultivation between a latitude of 35–55° either north or south of the equator, with the United States and Germany the major producers.




| Name | Side Chain (R) |
|---|---|
| Humulone | —$CO•CH_2•CH(CH_3)_2$ isovaleryl |
| Cohumulone | —$CO•CH(CH_3)_2$ isobutyryl |
| Adhumulone | —$CO•CH(CH_3)•CH_2•CH_3$ 2-methylbutyryl |

Figure 10. The structures of alpha acids prevalent in hop resins.

Hops are hardy climbing herbaceous perennial plants grown in gardens using characteristic string frameworks to support them. It is only the female plants that are cultivated, as they are the ones that develop the hop cones (Figure 12). Their rootstock remains in the ground year on year and is spaced in an appropriate fashion for effective horticultural procedures (for example, spraying by tractors passing between rows). In recent years, so-called dwarf varieties have been bred, which retain the bittering and aroma potential of ‘traditional’ hops but which grow to a shorter height (6–8 ft as opposed to twice as big). As a result they are much

easier to harvest and there is less wastage of pesticide during spraying. Hop gardens featuring dwarf varieties are also much cheaper to establish.

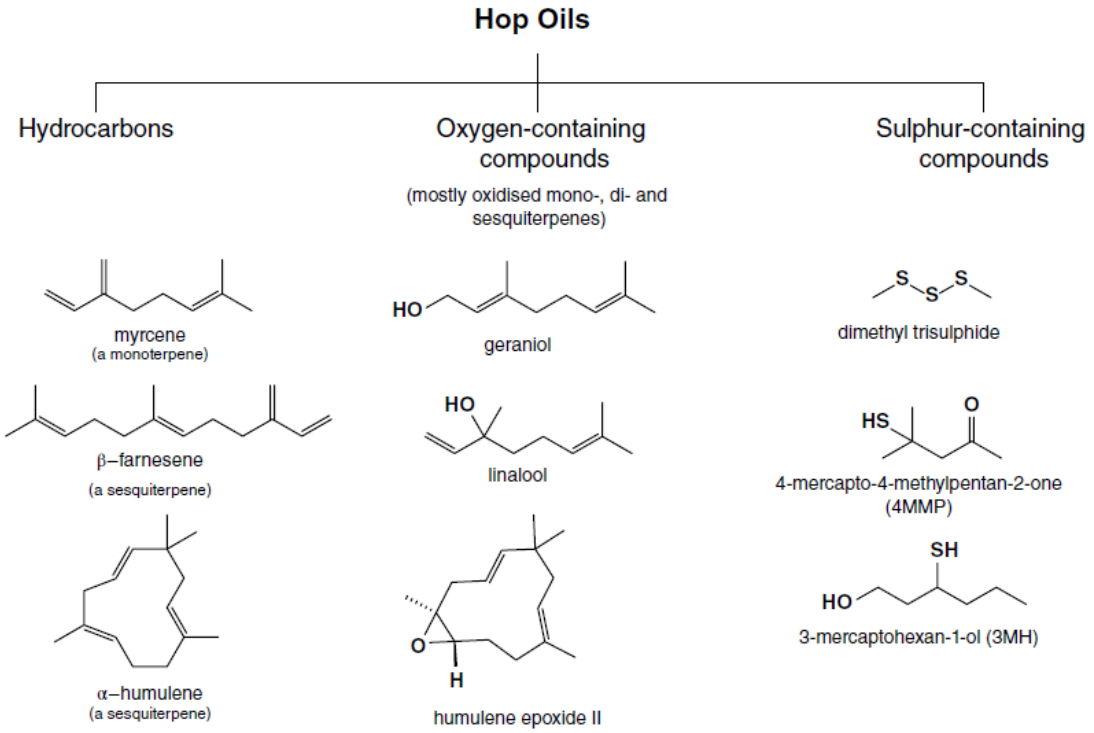



Figure 11. Selected components of hop oil fractions.

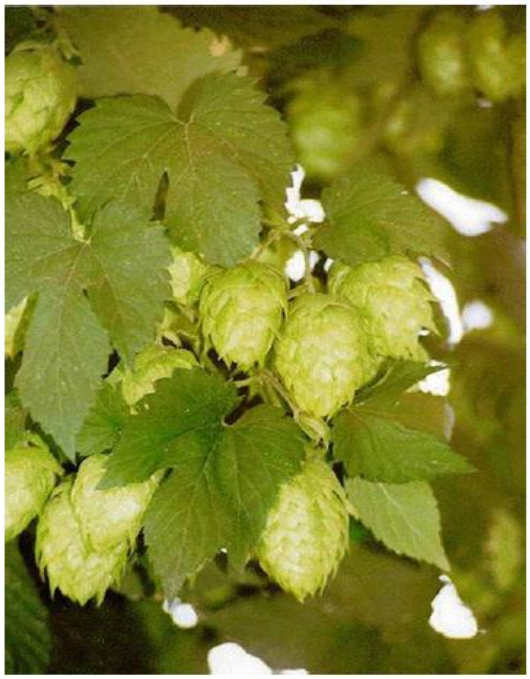


Figure 12. Hop cones.

Hops are susceptible to a wide range of diseases and pests. The most serious problems come from *Verticillium* wilt, downy mildew, mould and the damson-hop aphid. Varieties differ in their susceptibility to infestation and have been progressively selected on this basis. Nonetheless it is frequently necessary to apply pesticides, which are always stringently evaluated for their influence on hop quality, for any effect they may have on the brewing process and, of course, for their safety.

Hops are generally classified into two categories: aroma hops and bittering hops. All hops are capable of providing both bitterness and aroma. Some hops, however, such as the Czech variety Saaz, have a relatively high ratio of oil to resin and the character of the oil component is particularly prized. Such varieties command higher prices and are known as *aroma varieties*. They will seldom be used as the sole source of bitterness and aroma in a beer: a cheaper, higher α-acid hop (a *bittering variety*) will be used to provide the bulk of the bitterness, with the prized aroma variety added later for the contribution of its own unique blend of oils. Those Brewers requiring hops solely as a source of bitterness may well opt for a cheaper variety, ensuring its use early in the kettle boil so that the provision of bitterness is maximised and unwanted aroma is driven off.

The use of whole cone hops is comparatively uncommon nowadays. Many brewers use hops that have been hammer-milled and then compressed into pellets. In this form they are more stable, more efficiently utilised and they do not present the brewer with the problem of separating out the vegetative parts of the hop plant. Some use hop extracts that are derived by dissolving the resins in liquid carbon dioxide, followed in some cases by a chemical isomerisation to produce 'pre-isomerised' extracts. Recent years have been marked by an enormous increase in the use of such pre-isomerised extracts after they have been modified by reduction. One of the side-chains on the iso-α-acids is susceptible to cleavage by light; it then reacts with traces of sulphidic materials in beer to produce 2-methyl-3-butene-1-thiol (MBT), a substance that imparts an intensely unpleasant skunky character to beer. If the side-chain is reduced, it no longer produces MBT. For this reason, beers that are destined for packaging in green or clear glass bottles are often produced using these modified bitterness preparations, which have the added advantage of possessing increased foam-stabilising and antimicrobial properties.

**Wort Boiling and Clarification**

The boiling of wort serves various functions, primary amongst which are the isomerisation of the hop resins (α-acids) to the more soluble and bitter iso-α-acids, sterilisation, the driving off of unwanted volatile materials, the precipitation of macromolecular complexes (mostly protein, as 'hot break' or 'trub') and concentration of the wort. The extent of wort boiling is normally described in terms of % evaporation. Water is usually boiled off at a rate of about 4% per hour and the duration of boiling is likely to be one to two hours. Brew kettles are sometimes referred to as 'coppers', reflecting the original metal from which they were fabricated. Designs of kettle are encountered with either an internal (Figure 13) or external wort

boiler (Figure 14). One advantage of the latter is that siting the heat exchanger external to the vessel means there are no size limitations on the surface area for heat exchange. These days kettles are usually made from stainless steel.

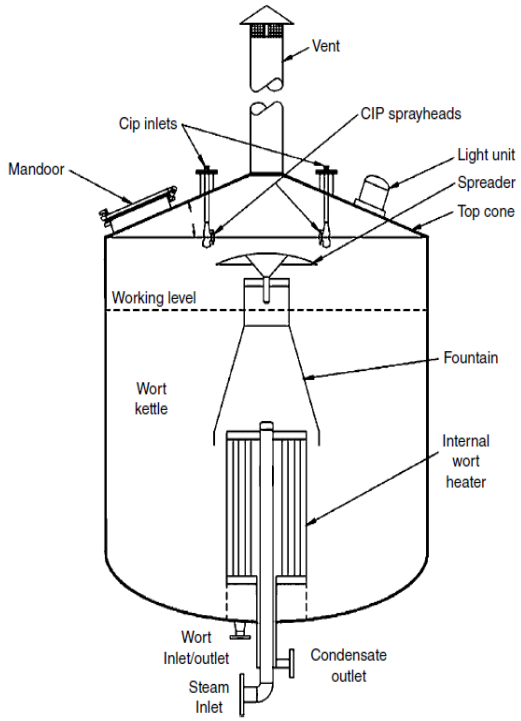



Fig. 13. Wort boiling kettle with an internal heater

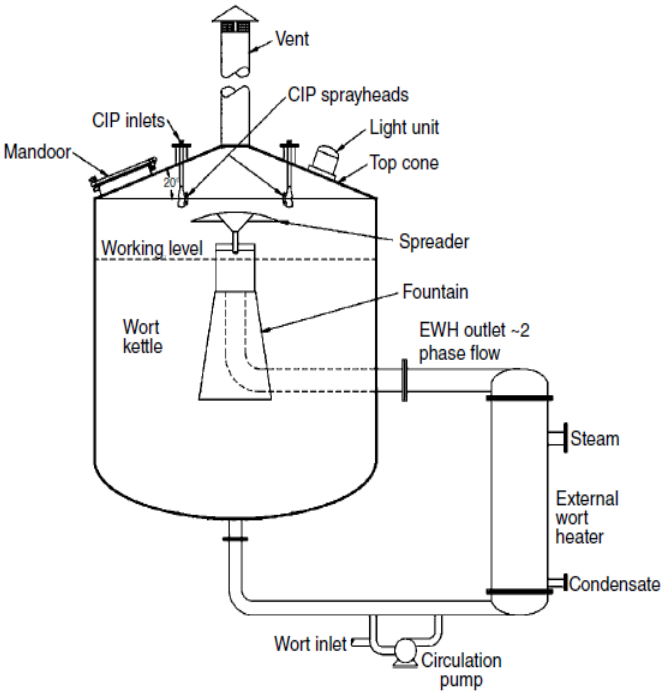



Fig. 14. Wort boiling kettle with an external heater

Table 4. Brewing sugars.

| Type | Carbohydrate Distribution (%) |
|---|---|
| Cane | Sucrose predominantly |
| Invert | Glucose (50), fructose (50) |
| Dextrose | Glucose (100) |
| High conversion (acid + enzyme) | Glucose (88), Maltose (4), maltotriose (2), dextrin (6) |
| Glucose chips | Glucose (84), Maltose (1), maltotriose (2), dextrin (13) |
| Maize syrup | Glucose (45), Maltose (38), maltotriose (3), dextrin (14) |
| Very high maltose | Glucose (5), Maltose (70), maltotriose (10), dextrin (15) |
| High conversion (acid) | Glucose (31), Maltose (18), maltotriose (13), dextrin (38) |
| High maltose | Glucose (10), Maltose (60), dextrin (30) |
| Low conversion | Glucose (12), Maltose (10), maltotriose (10), dextrin (68) |
| Maltodextrin | Maltose (1.5), maltotriose (1.5), dextrin (95) |
| Malt extract | Comparable to brewer's wort – also contains nitrogenous components |
| Candi sugar | Sucrose predominantly – but with caramelization-derived flavours |
| Lactose | Lactose predominantly |

The products dextrose through maltodextrin are customarily derived by the selective hydrolysis of corn (maize)-derived starch by acid and enzymes to varying extents.

Certain fining materials (e.g. a charged polysaccharide from Irish Moss) may be added to promote protein precipitation and ensuring sedimentation in the next stage. This is also the stage at which liquid sugar adjunct can be added (Table 4). Sugars added in the kettle are called ‘wort extenders’: they present the opportunity to increase the extract from a brew house without investment in extra mashing vessels and wort separation devices. Most sugars are derived from either corn or sugar cane. In the latter case the principal sugar is either sucrose, or fructose plus glucose if the product has been ‘inverted’. There are many different corn sugar products, differing in their degree of hydrolysis and therefore fermentability. Through the controlled use of acid but increasingly of starch-degrading enzymes, the supplier can produce preparations with a full range of fermentabilities depending on the needs of the Brewer; from 100% glucose through to high dextrin.

After boiling, wort is transferred to a clarification device. The system employed for removing insoluble material after boiling depends on the way in which the hopping was carried out. If whole hop cones are used, clarification is through a hop back, which is analogous to a lauter tun, but in this case the bed of residual hops constitutes the filter medium. If hop pellets or extracts are used, then the device of choice is the whirlpool, a cylindrical vessel, into which hot wort is transferred tangentially through an opening 0.5–1 m above the base (Figure 15). The wort is set into a rotational flux, which forces trub to a pile in the middle of the vessel.

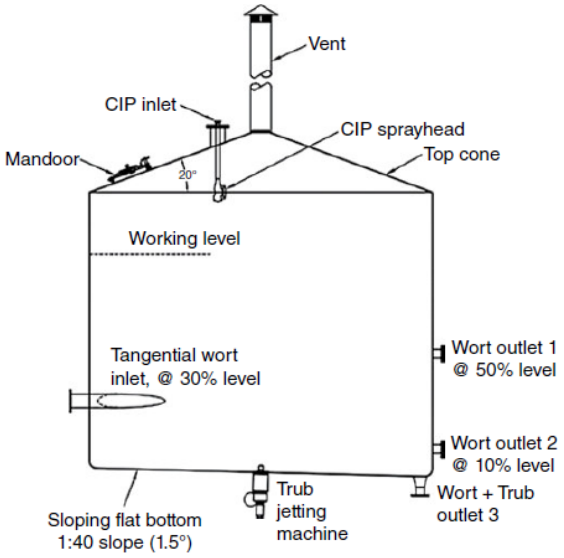



Figure 15. A ‘whirlpool’ (hot wort separation vessel).

**Wort Cooling**

Almost all cooling systems these days are of the stainless steel plate heat exchanger type, sometimes called ‘paraflows’ (Figure 16). Heat is transferred from the wort to a coolant, either water or glycol depending on how low the temperature needs to be taken. At this stage, it is likely that more material will precipitate from solution (‘cold break’). Brewers are divided on whether they feel this to be good or bad for fermentation and beer quality. The presence of this break certainly accelerates fermentation and therefore it will directly influence yeast metabolism.

As in so much of brewing, the aim should be consistency: either consistently ‘bright worts’ or ones containing a relatively consistent level of trub.

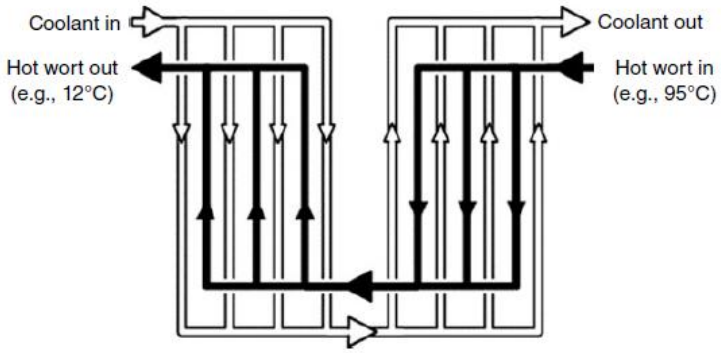



Figure 16. Product and coolant flows in a wort cooler (heat exchanger).

**Yeast**

Brewing yeast is *Saccharomyces cerevisiae* (ale yeast) or *Saccharomyces pastorianus* (lager yeast). There are many separate strains of brewing yeast, each of which is distinguishable phenotypically (e.g. in the extent to which it will ferment different sugars, or in the amount of oxygen it needs to prompt its growth, or in the amounts of its metabolic products [i.e. flavour spectrum of the resultant beer], or its behaviour in suspension [top vs. bottom fermenting, flocculent or non-flocculent]) and genotypically, in terms of its DNA fingerprint.

The fundamental differentiation between ale and lager strains is based on the ability or otherwise to ferment the sugar melibiose: ale strains can’t whereas lager strains can because they produce the enzyme (α-galactosidase) necessary to convert melibiose into glucose and galactose. Ale yeasts also move to the top of open fermentation vessels and are called top-fermenting yeasts. Lager yeasts drop to the bottom of fermenters and are termed bottom-fermenting yeasts. Nowadays it is frequently difficult to make this differentiation, when beers are widely fermented in similar types of vessel (deep cylindro-conical tanks) which tend to equalise the way in which yeast behaves in suspension.

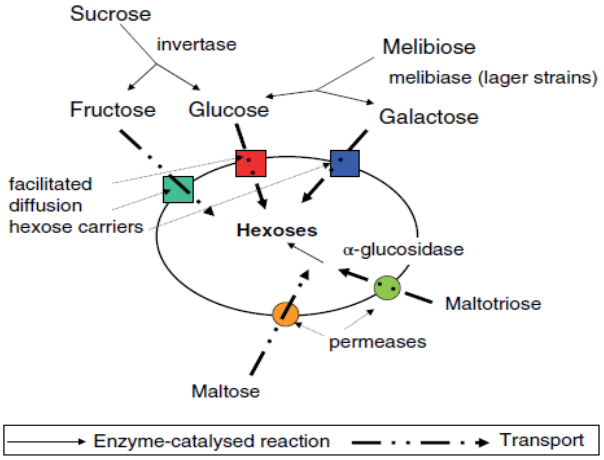



Figure 17. The uptake of sugars by brewing yeast.

When presented with wort, yeast encounters a selection of carbohydrates which, for a typical all-malt wort will approximate to maltose (45%), maltotriose (15%), glucose (10%), sucrose (5%), fructose (2%), and dextrin (23%). The last of these fractions (maltotetraose and longer oligomers) is unfermentable. The others will ordinarily be utilised in the sequence sucrose, glucose, fructose, maltose, and lastly maltotriose, although there may be some overlap (Figure 17). Sucrose is hydrolysed by an enzyme (invertase) released by the yeast outside the cell, and then the glucose and fructose enter the cell to be metabolised. Maltose and maltotriose also enter, through the agency of specific permeases. Inside the cell they are broken down into glucose by an α-glucosidase. Glucose represses the maltose and maltotriose permeases.

The principal route of sugar utilisation in the cell is the Embden-Meyerhof-Parnas (EMP) pathway of glycolysis (see Chapter 1). Brewing yeast derives most of the nitrogen it needs for synthesis of proteins and nucleic acids from the amino acids in the wort. A series of permeases is responsible for the sequential uptake of the amino acids. It is understood that the amino acids are transaminated to keto acids through reaction with α-ketoglutarate to produce glutamate, which comprises the internal store of nitrogen. It is used to transaminate keto-acids derived from intermediary metabolism to produce the required amino acid. The amino acid spectrum and level in wort (free amino nitrogen) is significant as it influences yeast metabolism leading to flavor-active products.

Oxygen is needed by the yeast to synthesise the unsaturated fatty acids and sterols needed for membranes. This oxygen is introduced at the wort cooling stage in the quantities that the yeast requires – but no more, because excessive aeration or oxygenation promotes excessive yeast growth and the more yeast is produced in a fermentation, the less alcohol will be produced. Different yeasts need different amounts of oxygen.

Yeast uses its stored reserves of carbohydrate in order to fuel the early stages of metabolism when it is pitched into wort, e.g. the synthesis of sterols. The main reserve glycogen is similar in structure to the amylopectin fraction of barley starch. The glycogen reserves of yeast build up during fermentation and it is important that they are conserved in the yeast during storage between fermentations. Trehalose (a disaccharide comprising two glucose units linked with an α1–1 bond between their reducing carbons) protects against the stress of starvation. It certainly seems to help the survival of yeast under dehydration conditions employed for the storage and shipping of dried yeast.

Pure yeast culture was pioneered by Hansen at Carlsberg in 1883. By a process of dilution he was able to isolate individual strains and open up the possibility of selecting and growing separate strains for specific purposes. Nowadays Brewers maintain their own pure yeast strains. Whilst it is still a fact that some brewers simply use the yeast grown in one fermentation to ‘pitch’ the following fermentation and that they have done this for many tens of years, it is much more usual for yeast to be re-propagated from a pure culture every 5–15 generations, a frequency dictated by company policy. When brewers talk of ‘generations’ they mean successive fermentations; strictly speaking yeast advances a generation every time it buds, and therefore there are several generations during any individual fermentation. Large quantities of yeast are

needed to pitch commercial scale fermentations. They need to be generated by successive scale-up growth from the master culture (Figure 18), a process called ‘propagation’. Higher yields are possible if fed-batch culture is used.

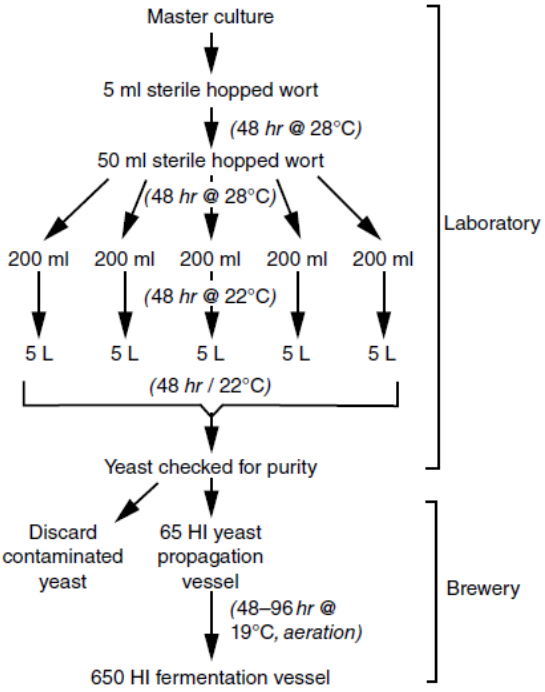



Figure 18. Yeast propagation.

This is the type of procedure used in the production of Baker’s Yeast. It takes advantage of the Crabtree effect, in which high concentrations of sugar drive the yeast to use it fermentatively rather than by respiration. When yeast grows by respiration it captures much more energy from the sugar and therefore produces much more cell material. In fed-batch culture, the amount of sugar made available to the yeast at any stage is low. Together with the high levels of oxygen in a well-aerated system, the yeast respires and grows substantially. The sugar is ‘dribbled in’ and the end result is a far higher yield of biomass, perhaps four fold more than is produced when the sugar is provided in a single batch at the start of fermentation.

The majority of brewing yeasts are resistant to acid (pH 2.0–2.2) and so the addition of phosphoric acid to attain this pH is very effective in killing bacteria with which yeast may become progressively contaminated from fermentation to fermentation. Many Brewers use such acid washing of yeast between fermentations.

There are two key indices of yeast health: viability and vitality. Both should be high if a successful fermentation is to be achieved. Viability is a measure of whether a yeast culture is alive or dead. Whilst microscopic inspection of a yeast sample is useful as a gross indicator of that culture (e.g. presence of substantial contamination), quantitative evaluation of viability needs a staining test. The most common is the use of methylene blue: viable yeast decolorizes it, dead cells do not. Although a yeast cell may be living, it does not necessarily mean that it is

healthy. Vitality is a measure of how healthy a yeast cell is. Many techniques have been advanced as an index of vitality, but none has been accepted as definitive.

Preferably yeast is stored in a readily sanitised room that can be cleaned efficiently and that is supplied with a filtered air supply and possesses a pressure higher than the surroundings in order to impose an outward vector of contaminants. Ideally it should be at or around 0 °C. Even if storage is not in such a room, the tanks must be rigorously cleaned, chilled to 0–4 °C and have the facility for gently rousing (mixing) to avoid hot spots. Yeast is stored in slurries ('barms') of 5–15% solids under 6 in. of beer, water or potassium phosphate. An alternative procedure is to press the yeast and store it at 4 °C in a cake form (20–30% dry solids). Pressed yeast may be held for about 10 days, slurried in water or beer for 3–4 weeks or slurried in 2% phosphate at pH 5 for 5 weeks.

Brewers seeking to ship yeast normally transport cultures for re-propagation at the destination. However greater consistency is achieved when it is feasible to propagate centrally and ship yeast for direct pitching. Such yeast must be contaminant-free and of high viability and vitality, washed free from fermentable material and cold (0 °C). The longer the distance, the greater the recommendation for low moisture pressed cake.

Apart from the importance of pitching yeast of good condition, it is also important that the amount pitched is appropriate. The higher the pitching rate, the more rapid the fermentation. As the pitching rate increases, initially so too does the amount of new biomass synthesised until at a certain rate the amount of new yeast synthesised declines. The rate of attenuation and the amount of growth directly impacts on the metabolism of yeast and the levels of its metabolic products (i.e. beer flavour), hence the need for control. Yeast can be quantified by weight or cell number. Typically some $10^7$ cells per ml will be pitched for wort of 12° Plato (1.5–2.5 g pressed weight per litre). At such a pitching rate lager yeast will divide four to five times in fermentation. Yeast numbers can be measured using a haemocytometer, which is a counting chamber loaded onto a microscope slide. It is possible to weigh yeast or to centrifuge it down in pots that are calibrated to relate volume to mass, but in these cases it must be remembered that there are usually other solid materials present, e.g. trub.

Another procedure that has come into vogue is the use of capacitance probes that can be inserted in-line. An intact and living yeast cell acts as a capacitor and gives a signal whereas dead ones (or insoluble materials) do not. The device is calibrated against a cell number (or weight technique) and therefore allows the direct read-out of the amount of viable yeast in a slurry. Other in-line devices are available which quantify yeast on the basis of light scatter.

**Brewery Fermentations**

Primary fermentation is the fermentation stage proper in which yeast, through controlled growth, is allowed to ferment wort to generate alcohol and the desired spectrum of flavours. Increasingly brewery fermentations are conducted in cylindro-conical vessels (Figure 19). The fermentation is regulated by control of several parameters, notably the starting strength of the wort (°Plato, which approximates to per cent sugar by weight, or Balling or Brix), the amount of

viable yeast (‘pitching rate’), the quantity of oxygen introduced and the temperature. Fermentation is monitored by measuring the decrease in specific gravity (alcohol has a much lower specific gravity than sugar).

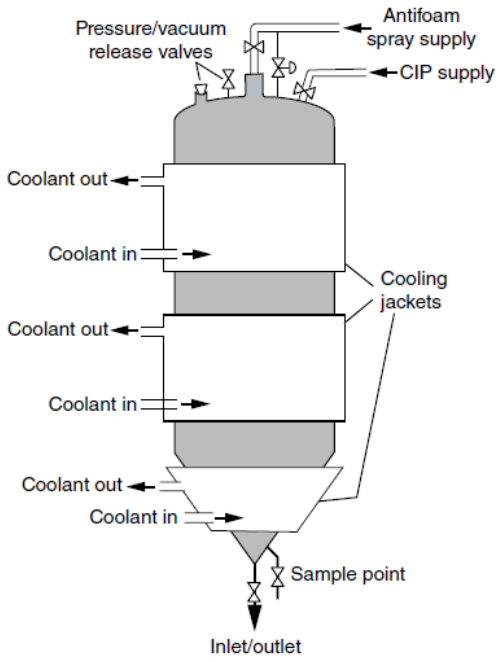



Figure 19. A cylindro-conical fermentation vessel.

Ales are generally fermented at a higher temperature (15–20 °C) than lagers (6–15 °C) and therefore attenuation (the achievement of the finished specific gravity) is achieved more rapidly. Thus an ale fermenting at 20 °C may achieve attenuation gravity in two days, whereas a lager fermented at 8.5 °C may take 10 days. Fermentation temperature has a substantial effect on the metabolism of yeast, and the levels of a flavor substance like iso-butanol might be 16.5 and 7 mg l−1 respectively for the ale and the lager. Some Brewers add zinc (e.g. 0.2 ppm) to promote yeast action – it is a cofactor for the enzyme alcohol dehydrogenase. During fermentation the pH falls, because yeast secretes organic acids and protons. Figure 20 depicts the time course of fermentation.

Surplus yeast will be removed at the end of fermentation, either by a process such as ‘skimming’ for a traditional square fermenter employing top fermenting yeast, or from the base of a cone in a cylindro-conical vessel. This is in order to preserve the viability and vitality of the yeast, but also to circumvent the autolysis and secretory tendencies of yeast that will be to the detriment of flavour and foam. There will still be sufficient yeast in the beer to effect the secondary fermentation.

The ‘green’ beer produced by primary fermentation needs to be ‘conditioned’, in respect of establishment of a desired carbon dioxide content and refinement of the flavour. This is called secondary fermentation. Above all at this stage there needs to be the removal of an undesirable butterscotch or popcorn flavour character due to substances called vicinal diketones (VDKs).

Traditionally it is the lager beers fermented at lower temperatures that have needed the more prolonged maturation (storage: 'lagering') in order to refine their flavour and develop carbonation. The latter depends on the presence of sugars, either those (perhaps 10%) which the brewer ensures are residual from the primary fermentation or those introduced in the 'krausening' process, in which a proportion of freshly fermenting wort is added to the maturing beer. Many brewers are unconvinced by the need for prolonged storage periods (other than for its strong marketing appeal) and they tend to combine the primary and secondary fermentation stages. Once the target attenuation has been reached, the temperature is allowed to rise (perhaps by 4 °C) which permits the yeast to deal more rapidly with the VDKs. Carbonation will be achieved downstream by the direct introduction of gas.

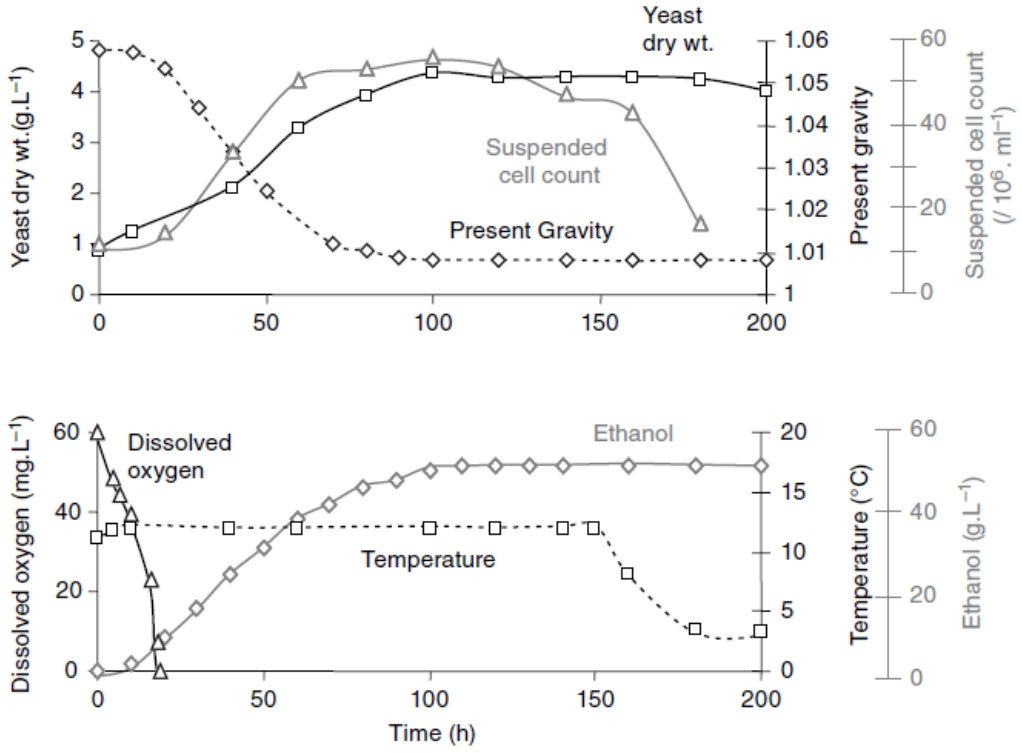



Figure 20. Changes occurring during a brewery lager fermentation.

Once the secondary fermentation stage is complete (and the length of this varies considerably between brewers), then the temperature is dropped, ideally to −1 or −2 °C to enable precipitation and sedimentation of materials that would otherwise cause a haze in the beer. The sedimentation of yeast is also promoted in this 'cold conditioning' stage, perhaps with the aid of isinglass finings. These are solutions of collagen derived from the swim bladders of certain species of fish from the South China Seas. Collagen has a net positive charge at the pH of beer, whereas yeast and other particulates have a net negative charge. Opposite charges attracting, the isinglass forms a complex with these particles and the resultant large agglomerates sediment readily because of an increase in particle size. Sometimes the isinglass finings are used alongside

'auxiliary finings' based on alginate or silicate, the combination being more effective than isinglass alone. Some brewers centrifuge to aid clarification.

For the most part fermenters these days are fabricated from stainless steel and will be lagged and feature jackets that allow coolant to be circulated (the heat generated during fermentation is sufficient to effect any necessary warming – so the temperature is regulated by balancing metabolic heat with cooling afforded by the coolant in the jacket, which many be water, propylene glycol or ammonia depending on how much refrigeration is demanded). Modern vessels tend to be enclosed, for microbiological reasons. However across the world there remain a great many open tanks. Cylindroconical vessels can have a capacity of up to 13 000 hectolitres and are readily cleaned using cleaning-in-place (CIP) operations.

Only one company, in New Zealand, practices continuous fermentation. Many brewers nowadays maximise the output by fermenting wort at a higher gravity than necessary to give the target alcohol concentration, before diluting the beer downstream with deaerated water to 'sales gravity' (i.e. the required strength of the beer in package). This is called 'high gravity brewing'. There are limits to the strength of wort that can be fermented. This is because yeast becomes stressed at high sugar concentrations and when the alcohol level increases beyond a certain point. Brewing is unusual amongst alcohol production industries in that it re-uses yeast for ensuing fermentations. Excluded from this are those beers in which very high alcohol levels are developed (e.g. the barley wines). The yeast is stressed in these conditions and will not be re-usable. This is the reason why wine fermentations, for instance, involve 'one trip' yeast. This is also the reason why, in the production of sweeter fortified wines, alcohol is added at the start of fermentation in order to hinder the removal of sugars.

**Filtration**

After a period of typically three days minimum in 'cold conditioning', the beer is generally filtered. Diverse types of filter are available, including the plate-and-frame filter, which consists of a series of plates in sequence, over each of which a cloth is hung. Latterly there is more common use of leaf and candle filters. The beer is mixed with a filter aid – particles which both trap particles and prevent the system from clogging. Two major kinds of filter aid are in regular use: kieselguhr (diatomaceous earth) and perlite. The former comprises fossils or skeletons of primitive organisms called diatoms. These can be mined and classified to provide grades that differ in their permeability characteristics. Particles of kieselguhr contain pores into which other particles (such as those found in beer) can pass, depending on their size. Perlites are derived from volcanic glasses crushed to form microscopic flat particles. They are better to handle than kieselguhr, but are not such efficient filter aids. Filtration starts when a pre-coat of filter aid is applied to the filter by cycling a slurry of filter aid through the plates. This pre-coat is generally of quite a coarse grade, whereas the filter-aid (the body feed) which is dosed into the beer during the filtration proper tends to be a finer grade. It is selected according to the particles within the beer that need to be removed. If a beer contains a lot of yeast, but relatively few small particles, then a relatively coarse grade is best. If the converse applies, then a fine grade with

smaller pores will be used. Cross-flow filtration using membranes and without the involvement of filter aids is gaining more attention.

**The Stabilisation of Beer**

Apart from filtration, various other treatments may be applied to beer downstream, all with the aim of enhancing the shelf life of the product. A haze in beer can be due to various materials, but principally it is due to the cross-linking of certain proteins and certain polyphenols. Therefore if one or both of these materials is removed then the shelf life is extended. Brewhouse operations are in part designed to precipitate out protein-polyphenol complexes. Thus, if these operations are performed efficiently, then much of the job of stabilisation is achieved. Good, vigorous, 'rolling' boils, for instance, will ensure precipitation. Before that, avoidance of the last runnings in the lautering operation will prevent excessive levels of polyphenol entering the wort. The cold conditioning stage also has a major role to play, by chilling out protein-polyphenol complexes, enabling them to be taken out on the filter. Control over oxygen and oxidation is important because it is particularly the oxidised polyphenols that tend to cross-link with proteins. For really long shelf lives, though, and certainly if the beer is being shipped to extremes of climate, additional stabilisation treatments will be necessary. Polyphenols can be removed with polyvinylpolypyrrolidone (PVPP). Protein can be precipitated by adding tannic acid, hydrolysed using papain (the same enzyme from paw paw that is used as a meat tenderizer) or prolyl endoproteinase, and most commonly, adsorbed on silica hydrogels and silica xerogels.

**Gas Control**

Final adjustment will now be made to the level of gases in the beer. As we have seen, it is important that the oxygen level in the bright beer is as low as possible. Unfortunately, whenever beer is moved around and processed in a brewery there is always the risk of oxygen pick-up. For example, oxygen can enter through leaky pumps. A check on oxygen content will be made once the bright beer tank (filtered beer is bright beer) is filled and, if the level is above specification (which most Brewers will set at 0.1 ppm or less), oxygen will have to be removed. This is achieved by purging the tank with an inert gas, usually nitrogen, from a sinter in the base of the vessel. The level of carbon dioxide in a beer may either need to be increased or decreased. The majority of beers contain between 2 and 3 volumes of $CO_2$, whereas most brewery fermentations generate 'naturally' no more than 1.2–1.7 volumes of the gas. The simplest and most usual procedure by which $CO_2$ is introduced is by injection as beer is transferred from the filter to the Bright Beer Tank. If the $CO_2$ content needs to be dropped this is a more formidable challenge. It may be necessary for beers that are supposed to have a relatively low carbonation and, as for oxygen, this can be achieved by purging. However concerns about 'bit' production have stimulated the development of gentle membrane-based systems for gas control. Beer is flowed across membranes, made from polypropylene or polytetrafluoroethylene, that are water-hating and therefore don't 'wet-out'. Gases, but not liquids, will pass freely across such membranes, the

rate of flux being proportional to the concentration of each individual gas and dependent also on the rate at which the beer flows past the membrane.

**Packaging**

The packaging operation is the most expensive stage in the brewery, in terms of raw materials and labour. Beer will be brought into specification in the Bright Beer Tank (sometimes called the Fine Ale Tank or the Package Release Tank). The carbonation level may be higher (e.g. by 0.2 vol.) than that specified for the beer in package, to allow for losses during filling.

Although beer is relatively resistant to spoilage, it is by no means entirely incapable of supporting the growth of micro-organisms. For this reason most beers are treated to eliminate any residual brewing yeast or infecting wild yeasts and bacteria before or during packaging. This can be achieved in one of two ways: pasteurisation or sterile filtration. Pasteurisation can take one of two forms in the brewery: flash pasteurisation for beer pre-package, and tunnel pasteurisation for beer in can or bottle. The principle in either case, of course, is one that heat kills micro-organisms. One Pasteurisation Unit (PU) is defined as one minute at 60 °C. The higher the temperature, the more rapidly are micro-organisms destroyed. A 7 °C rise in temperature leads to a 10-fold increase in the rate of cell death. The pasteurisation time required to kill organisms at different temperatures can be read off from a plot. Typically a brewer might use 5–20 PU – but higher 'doses' may be used for some beers, e.g. low alcohol beers, which are more susceptible to contamination. In flash pasteurisation, the beer flows through a heat exchanger (essentially like a wort cooler acting in reverse), which raises the temperature typically to 72 °C. Residence times of between 30 and 60 seconds at this temperature are sufficient to kill off virtually all microbes. Ideally there won't be many of these to remove: good brewers will ensure low loadings of micro-organisms by attention to hygiene throughout the process and ensuring that the prior filtration operation is efficient. Tunnel pasteurizers comprise large heated chambers through which cans or glass bottles are conveyed over a period of minutes, as opposed to the seconds employed in a flash pasteurizer. Accordingly, temperatures in a tunnel pasteurizer are lower, typically 60 °C for a residence time of 10–20 minutes. An increasingly popular mechanism for removing micro-organisms is to filter them out by passing the beer through a fine mesh filter. The rationale for selecting this procedure rather than pasteurisation is as much for marketing reasons as for any technical advantage it presents: many brands of beer these days are being sold on a claim of not being heat-treated, and therefore free from any 'cooking'. In fact, provided the oxygen level is very low, modest heating of beer does not have a major impact on the flavour of many beers, although those products with relatively subtle, lighter flavour will obviously display 'cooked' notes more readily than will beers which have a more complex flavour character. The sterile filter must be located downstream from the filter that is used to separate solids from the beer. Sterile filters may be of several types, a common variant incorporating a membrane formed from polypropylene or polytetrafluoroethylene and with pores of between 0.45 and 0.8 μm.

***1. Filling Bottles and Cans***

Bottles entering the brewery's packaging hall are first washed and, if they are returnable bottles (that is, they have been used previously to hold beer), they will need a much more robust cleaning and sterilisation, inside and out, involving soaking and jetting with hot caustic detergent and thorough rinsing with water. The beer coming from the Bright Beer Tanks is transferred to a bowl at the heart of the filling machine. Bottle fillers are machines based on a rotary carousel principle. They have a series of filling heads: the more heads, the greater the capacity of the filler. The bottles enter on a conveyor and, sequentially they are individually raised into position beneath the next vacant filler head, each of which comprises a filler tube. An air-tight seal is made and, in modern fillers, a specific air evacuation stage starts the filling sequence. The bottle is counter-pressured with carbon dioxide, before the beer is allowed to flow into the bottle by gravity from the bowl. The machine will have been adjusted so that the correct volume of beer is introduced into the vessel. Once filled, the 'top' pressure on the bottle is relieved, and the bottle is released from its filling head. It passes rapidly to the machine that will crimp on the crown cork but, en route, the bottle will have been either tapped or its contents 'jetted' with a minuscule amount of sterile water in order to fob the contents and drive off any air from the space in the bottle between the surface of the beer and the neck (the 'headspace'). Next stop is the tunnel pasteurizer if the beer is to be pasteurised after filling but if sterile filtration is used the filler and capper are likely to be enclosed in a sterile room. The bottles now head off for labelling, secondary packaging and warehousing.

Putting beer into cans has much in common with bottling. It is the container, of course, that is very different – and definitely one trip. Cans may be of aluminium or stainless steel, which will have an internal lacquer to protect the beer from the metal surface and vice-versa. Cans arrive in the canning hall on vast trays, all pre-printed and instantly recognisable. They are inverted, washed, and sprayed, prior to filling in a manner very similar to the bottles. Once filled, the lid is fitted to the can basically by folding the two pieces of metal together to make a secure seam past which neither beer nor gas can pass.

***2. Filling Kegs***

Kegs are manufactured from either aluminium or stainless steel. They are containers generally of 100 L or less, containing a central spear. Kegs, of course, are multi-trip devices. On return to the brewery from an 'outlet' they are washed externally before transfer to the multi head machine in which successive heads are responsible for their washing, sterilising and filling. Generally they will be inverted as this takes place. The cleaning involves high pressure spraying of the entire internal surface of the vessel with water at approximately 70 °C. After about 10 seconds, the keg passes to the steaming stage, the temperature reaching 105 °C over a period of perhaps half a minute. Then the keg goes to the filling head, where a brief purge with carbon dioxide precedes the introduction of beer, which may take a couple of minutes. The discharged keg is weighed to ensure that it contains the correct quantity of beer and is labelled and palleted before warehousing.

## The Quality of Beer

### *1. Flavour*

The flavour of beer can be split into three separate components: taste, smell (aroma) and texture (mouthfeel).

There are five basic taste qualities: sweet, sour, salt, bitter and umami. They are detected on the tongue. A related sense is the tingle associated with high levels of carbonation in a drink: this is due to the triggering of the trigeminal nerve by carbon dioxide. This nerve responds to mild irritants, such as carbonation and capsaicin (a substance largely responsible for the 'pain delivery' of spices and peppers).

Carbon dioxide is also relevant insofar as its level influences the extent to which volatile molecules will be delivered via the foam and into the headspace above the beer in a glass.

The sweetness of a beer is due, of course, to its level of sugars, either those that have survived fermentation or those introduced as primings.

The principal contributors to sourness in beer are the organic acids that are produced by yeast during fermentation. These lower the pH: it is the $H^+$ ion imparted by acidic solutions that causes the sour character to be perceived on the palate. Most beers have a pH between 3.9 and 4.6. It is now accepted that pH is a less accurate gauge of sourness than is titratable acidity.

Saltiness in beer is afforded by sodium and potassium, whilst of the anions present in beer chloride and sulphate are of particular importance. Chloride is said to contribute a mellowing and fullness to a palate, while sulphate is felt to elevate the dryness of beer. Perhaps the most important taste in beer is bitterness, primarily imparted by the iso-α-acids derived from the hop resins.

Many people believe that they can taste other notes on a beer. In fact they are detecting most of these with the nose, the confusion arising because there is a continuum between the back of the throat and the nasal passages. The smell (or aroma) of a beer is a complex distillation of the contribution of a great many individual molecules. No beer is so simple as to have its 'nose' determined by one or even a very few substances. The perceived character is a balance between positive and negative flavour notes, each of which may be a consequence of one or a combination of many compounds of different chemical classes. The 'flavour threshold' is the lowest concentration of a substance that is detectable in beer.

The substances that contribute to the aroma of beer are diverse. They are derived from malt and hops and by yeast activity (leaving aside for the moment the contribution of contaminating microbes). In turn there are interactions between these sources, insofar as yeast converting one flavour constituent from malt or hops into a different one, for example.

Various alcohols influence the flavour of beer (Table 5), by far and away the most important of which is ethanol, which is present in most beers at levels at least 350 - fold higher than any other alcohol. Ethanol contributes directly to the flavour of beer, registering a warming character. It also influences the flavour contribution of other volatile substances in beer. Because it is quantitatively third only to water and carbon dioxide as the main component of beer it is unsurprising that it moderates the flavour impact of other substances. It does this by affecting the

vapour pressure of other molecules (i.e. their relative tendency to remain in beer or to migrate to the headspace of the beer). The higher alcohols in beer are important as the immediate precursors of the esters, which are proportionately more flavour active (see Table 6). And so it is important to be able to regulate the levels of the higher alcohols produced by yeast if ester levels are also to be controlled.

Table 5. Some alcohols in beer.

| Alcohol | Flavour threshold (mg $l^{-1}$) | Perceived character |
|---|---|---|
| ethanol | 14000 | pungent |
| propan-1-ol | 800 | sharp, musty |
| butan-2-ol | 16 | |
| iso-amyl alcohol | 50 | alcohol, banana, vinous |
| tyrosol | 200 | bitter |
| phenylethanol | 40–100 | roses, perfume |

Table 6. Some esters in beer.

| Ester | Flavour threshold (mg $l^{-1}$) | Perceived character |
|---|---|---|
| Ethyl acetate | 33 | pear |
| Iso-amyl acetate | 1.0 | banana |
| ethyl octanoate | 0.9 | apples, sweet, fruity |
| phenylethyl acetate | 3.8 | roses, honey, apples |

The higher alcohols are produced during fermentation by two routes: “catabolic’ and anabolic. In the catabolic route yeast amino acids taken up from the wort by yeast are transaminated to alpha-keto-acids, which are decarboxylated and reduced to alcohols.

Various esters may make a contribution to the flavour of beer (Table 6). Conceptually, for as many acids and alcohols as are produced by yeast, the esters produced by the various combinations of them might be produced. In reality most focus has been on the acetyl esters. These esters are produced from their equivalent alcohols (ROH), through catalysis by the enzyme alcohol acetyl transferase (AAT), with acetyl-coenzyme A being the donor of the acetate group.

Whereas the esters and higher alcohols can make positive contributions to the flavor of beer, few (with the possible exception of some ales) are helped by the presence of the vicinal diketones (VDKs), diacetyl and (less importantly) pentanedione (Table 7). Elimination of VDKs from beer depends on the fermentation process being well-run. These substances are offshoots of the pathways by which yeast produces the amino acids valine and isoleucine (and therefore there is a relationship to the anabolic pathway of higher alcohol production).

Table 7. Vicinal diketones in beer.

| Vicinal diketone | Flavour threshold (mg $l^{-1}$) | Perceived character |
|---|---|---|
| diacetyl | 0.1 | butterscotch, popcorn |
| pentanedione | 0.9 | honey, buttery |

In many ways the most complex flavour characters in beer are due to the sulphur containing compounds. There are many of these in beer (Table 8) and they make various contributions. Thus many ales have a deliberate hydrogen sulphide character, not too much, but just enough to give a nice 'eggy' nose. Lagers tend to have a more complex sulphury character. Some lagers are relatively devoid of any sulphury nose. Others, though, have a distinct DMS character, while some have characters ranging from cabbagy to burnt rubber. This range of characteristics renders substantial complexity to the control of sulphury flavours.

Table 8. Some sulphur-containing substances in beer.

| S-containing compound | Flavour threshold (mg $l^{-1}$) | Perceived character |
|---|---|---|
| hydrogen sulphide | 0.005 | rotten eggs |
| sulphur dioxide | 25 | burnt matches |
| methanethiol | 0.002 | drains |
| ethanethiol | 0.002 | putrefaction |
| propanethiol | 0.0015 | onion |
| dimethyl sulphide | 0.03 | sweetcorn |
| dimethyl disulphide | 0.0075 | rotting vegetables |
| dimethyl trisulphide | 0.0001 | rotting vegetables, onion |
| methyl thioacetate | 0.05 | cooked cabbage |
| diethyl sulphide | 0.0012 | cooked vegetables, garlic |
| methional | 0.25 | cooked potato |
| 3-methyl-2-butene-1-thiol | 0.000004–0.001 | lightstruck, skunky |
| 2-furfurylmercaptan | | rubber |

Hydrogen sulphide (H2S) can also be produced by more than one pathway in yeast. It may be formed by the breakdown of amino acids such as cysteine or peptides like glutathione, or by the reduction of inorganic sources such as sulphate and sulphite (Figure 21).

The addition of hops during beer production not only contributes much of the resulting bitterness, but also imparts a unique so-called 'hoppy' aroma. This attribute comes from the complex volatile oil fraction of hops. Most of the component substances do not survive the brewing process intact and are chemically transformed into as yet poorly defined compounds. Certainly, there does not appear to be one compound solely responsible for hop aroma in beer, although several groups (e.g. sesquiterpene epoxides, cyclic ethers, and furanones) have been strongly implicated.

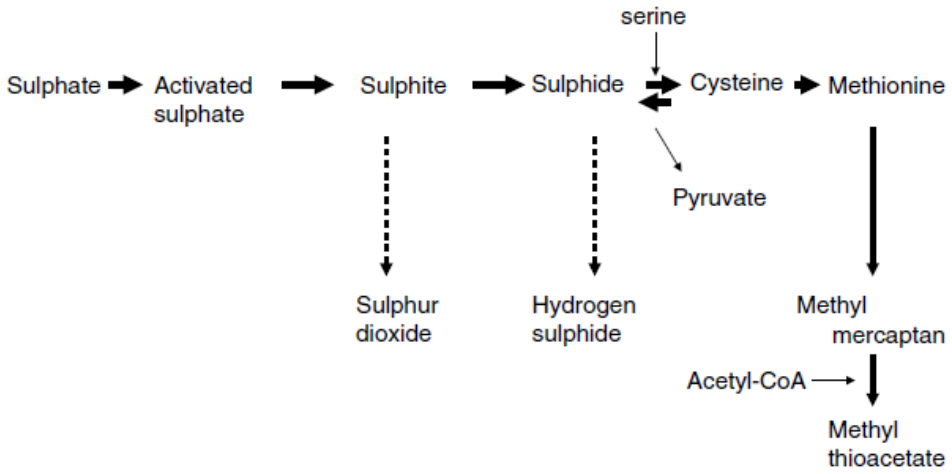



Figure 21. The origins of other sulphur-containing volatiles in beer

The point at which hops are added during beer production determines the resulting flavour that they impart. The practice of adding aroma hops close to the end of boiling (late hopping) still results in the substantial evaporation of volatile material, but of the little that remains, much is transformed into other species (e.g. the hop oil component humulene can be converted to the more flavor-active humulene epoxide). Further changes then occur during fermentation, such as the transesterification of methyl esters to their ethyl counterparts. The resultant late hop flavour is rather floral in character and is generally an attribute more associated with lager beers.

In a generally distinct practice, hops may be added to the beer right at the end of production. This process of dry hopping gives certain ales their characteristic aroma. The hop oil components contributed to beer by this process are very different to those from late hopping, with mono- and sesquiterpenes surviving generally unchanged in the beer.

Malty character in beer is due in part at least to isovaleraldehyde, which is formed from leucine in the Strecker degradation. The toffee and caramel flavours in crystal malts and the roasted, coffee- like notes found in darker malts are due to various complex components generated from amino acids and sugars that cross-react during kilning – the Maillard reaction.

Acetaldehyde, which is the immediate precursor of ethanol in yeast, has a flavor threshold of between 5 and 50 mg l−1 and imparts a ‘green apples’ flavour to beer. High levels should not survive into beer in successful fermentations, because yeast will efficiently convert the acetaldehyde into ethanol. If levels are persistently high, then this is an indication of premature yeast separation, poor yeast quality, or a Zymomonas infection.

The short chain fatty acids (Table 9) are made by yeast as intermediates in the synthesis of the lipid membrane components. For this reason, the control of these acids is exactly analogous to that of the esters (see earlier): if yeast needs to make fewer lipids (under conditions where it needs to grow less), then short chain fatty acids will accumulate.

Table 9. Some short-chain fatty acids in beer.

| Fatty acid | Flavour threshold (mg $l^{-1}$) | Perceived character |
|---|---|---|
| acetic | 175 | vinegar |
| propionic | 150 | acidic, milky |
| butyric | 2.2 | cheesy |
| 3-methyl butyric | 1.5 | sweaty |
| hexanoic | 8 | vegetable oil |
| octanoic | 15 | goaty |
| phenyl acetic | 2.5 | honey |

Some beers (for example some wheat beers) feature a phenolic or clove-like character. This is due to molecules such as 4-vinylguiacol (4-VG), which is produced by certain *Saccharomyces* species, including *Saccharomyces diastaticus*. Its unwanted presence in a beer is an indication of a wild yeast contamination. 4-VG is produced by the decarboxylation of ferulic acid by an enzyme that is present in *S. diastaticus* and other wild yeasts, but not in brewing strains other than a few specific strains of *S. cerevisiae*, namely the ones prized in Bavaria for their use in wheat beer manufacture.

The flavour of beer changes with time. There is a decrease in bitterness (due to the progressive loss of the iso-α-acids), an increase in perceived sweetness and toffee character and a development of a cardboard note. It is the latter that most Brewers worry about in connection with the shelf life of their products. Cardboard is due to a range of carbonyl compounds, which may originate in various precursors, including unsaturated fatty acids, higher alcohols, amino acids and the bitter substances. Most importantly, their formation is a result of oxidation, hence the importance of minimising oxygen levels in beer and, perhaps, further upstream.

***2. Foam***

A point of difference between beer and other alcoholic beverages is its possession of stable foam. This is due to the presence of hydrophobic (amphipathic) polypeptides, derived from cereals, that cross-link with the bitter iso-α-acids in the bubble walls to counter the forces of surface tension that tend to lead to foam collapse.

***3. Gushing***

Foaming can be taken to excess, in which case the problem that manifests itself in a small pack is ‘gushing’, i.e. the spontaneous generation of foam on opening a package of beer. This is due to the presence of nucleation sites in beer that cause the dramatic discharging of carbon dioxide from solution. These nucleation sites may be particles of materials like oxalate or filter aid, but most commonly gushing is caused by intensely hydrophobic peptides that are produced from moulds such as Fusarium that can contaminate barley unless precautions are taken.

**Spoilage of Beer**

Compared with most other foods and beverages beer is relatively resistant to contamination. There are several reasons for this, namely the presence of ethanol, a low pH, the relative shortage of nutrients (sugars, amino acids), the anaerobic conditions and the presence of antimicrobial agents, notably the iso-α-acids.

The most problematic Gram-positive bacteria are lactic acid bacteria belonging to the genera *Lactobacillus* and *Pediococcus*. At least 10 species of lactobacillus spoil beer. They tolerate the acidic conditions. Some species (e.g. *Lactobacillus brevis* and *Lactobacillus plantarum*) grow quickly during fermentation, conditioning and storage, whilst others (e.g. *Lactobacillus lindner*) grow relatively slowly. Spoilage with lactobacilli is especially problematic during the conditioning of beer and after packaging, resulting in a silky turbidity and off-flavours. Pediococci are homofermentative. Six species have been identified, the most important being *Pediococcus damnosus*. Such contamination generates lactic acid and diacetyl. The production of polysaccharide capsules can cause ropiness in beer.

Many Gram-positive bacteria are killed by iso-α-acids. These agents probably disrupt nutrient transport across the membrane of the bacteria, but only when they are present in their protonated forms (i.e. at low pH). This is one of the reasons why a beer at pH 4.0 will be more resistant to contamination than one at pH 4.5. Some Gram-positives are resistant to iso-α-acids and most Gram-negatives are.

Important Gram-negative bacteria include the acetic acid bacteria (*Acetobacter*, *Gluconobacter*); Enterobacteriaceae (*Escherichia*, *Aerobacter*, *Klebsiella*, *Citrobacter*, *Obesumbacterium*); *Zymomonas*, *Pectinatus* and *Megasphaera*. Acetic acid bacteria a vinegary flavour to beer and a ropy slime. It is most often found in draft beer, where there is a relatively aerobic environment close to the beer, e.g. in partly-emptied containers. Enterobacteriaceae are aerobic and cannot grow in the presence of ethanol. They are a threat in wort and early in fermentation and they produce cabbagy/vegetable/eggy aromas. *Zymomonas* is a problem with primed beers (it uses invert sugar or glucose, but cannot use maltose). Although it has a metabolism reminiscent of *Saccharomyces* (it is actually used to produce alcoholic beverages in some countries, for example pulque), it does tend to produce large amounts of acetaldehyde.

A wild yeast is any yeast other than the culture yeast used for a given beer. As well as *Saccharomyces*, wild yeast may be *Brettanomyces*, *Candida*, *Debaromyces*, *Hansenula*, *Kloeckera*, *Pichia*, *Rhodotorula*, *Torulaspora* or *Zygosaccharomyces*. If the contaminating yeast is another brewing yeast then the risk is a shift in performance to that associated with the 'foreign' yeast (i.e. you will not get the expected beer). If the contaminant is another type of yeast the risk is off-flavour production (e.g. clove-like flavours produced by decarboxylation of ferulic acid) or a problem such as over-attenuation as might be caused by a diastatic organism such as *S. diastaticus*.