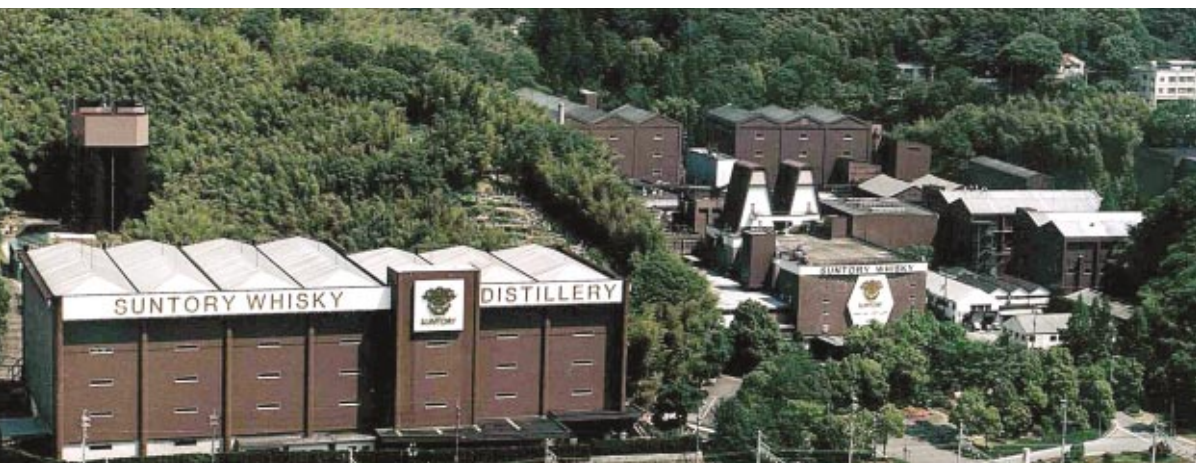


Lactic acid bacteria – the uninvited but generally welcome participants in malt whisky fermentation

Fergus G. Priest



How does malt whisky acquire its unique flavour? Fergus Priest explains how lactobacilli can play a crucial role.

● Scotch whisky – a multimillion pound industry

The production of Scotch malt whisky is governed by The Scotch Whisky Act (1988) which limits the ingredients to water, malted barley, whole grains of other cereals, yeast and caramel for colour adjustment. Interestingly, no mention is made of bacteria, yet we are beginning to think that lactic acid bacteria help refine the flavour of malt whisky in important ways. But first, a little background to the Scotch whisky industry.

The production of Scotch whisky employs around 41,000 Scottish residents and 65,000 people throughout the UK, generating about £1.3 billion of income for UK households. Sales of Scotch whisky topped £2.3 billion in 2002 and reached more than 200 countries, making it the largest matured spirit market in the world. Apart from the quality of the product, two related developments led to this phenomenal dominance of Scotch whisky. By the end of the 19th century the continuous still, patented by Aenas Coffey in 1827, enabled the prodigious production of grain whisky on a scale that could never be emulated in traditional pot stills. Second, this grain whisky provided the lighter base for blending with malt whisky to provide the consistency and quality of blended whisky. Blended whisky, which today often involves 30–40 individual whiskies, is the major component of the modern Scotch whisky market.

Malt whisky is prepared from a mash of malted barley which is fermented, distilled in traditional copper pot stills and matured in oak casks for not less than 3 years and generally a lot longer. Malt whisky is increasing in market share with a buoyant demand for 'boutique' products. However, despite increased export volume last year (by 9.3% to £268 million), malt whisky still represents only a little over 10% of the export market. About 85 malt distilleries operate today in Scotland, but their production is dwarfed by the dozen or so grain distilleries. Grain spirit is prepared from unmalted cereals as a source of starch, saccharified with malt enzymes, fermented and distilled in a continuous or

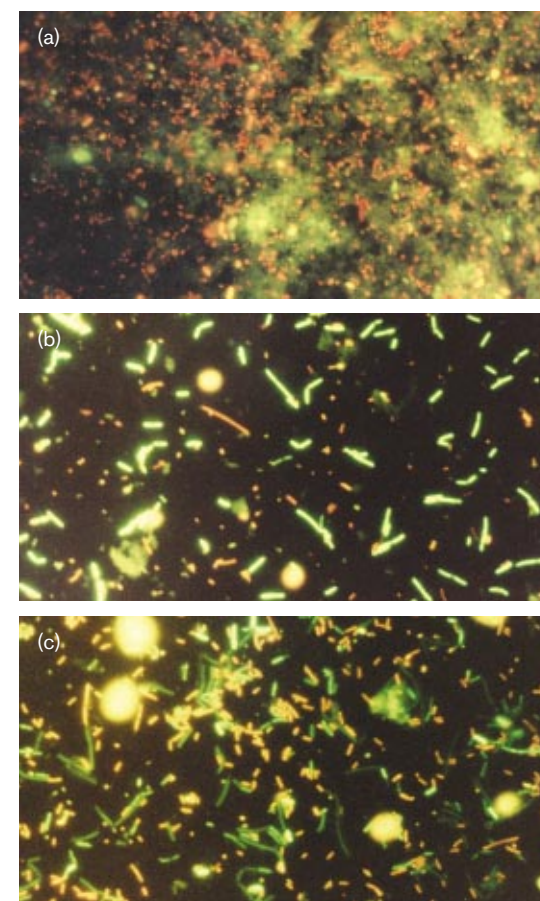
Coffey still. Like malt whisky, it must be matured for a minimum of 3 years. Grain whisky is the base of the standard blends such as Famous Grouse, Teachers, and Johnnie Walker which will contain between 15 and 30% malt whisky.

Both products use a similar process, but here we will focus on malt whisky. Malted barley is milled, infused in water at about 64 °C for some 30 minutes to 1 hour and the

wort is drained off into a fermenter or washback. The grain bed is rinsed with a second water at a higher temperature (typically about 70 °C) to remove residual nutrients from the fermentation. Finally, it is rinsed with a third water at about 80 °C which is used as the mashing water for the next mash. The spent grains are removed for cattle feed. You will note that the wort is not boiled as it is in a brewery. This is to permit the enzymes from the malted barley to continue to operate during the fermentation and to ensure complete hydrolysis of starch into glucose, maltose and other fermentable sugars. Two types of yeast are generally used: a pure-culture distiller's yeast obtained from a commercial yeast supplier and spent brewer's yeast. The practice of adding brewer's yeast is increasingly rare, but it is thought by some to impart important positive flavour characteristics to the spirit. Interestingly, Scotch whisky distillers are not so possessive of their yeasts as brewers and do not develop and maintain their own yeast strains with particular fermentation and flavour properties. Instead, they rely on the distillation process to govern the flavour profile of the finished product. The fermentation is conducted in wooden or stainless steel washbacks and is not temperature controlled. Consequently, the temperature can rise to over 30 °C and the fermentation is complete within 2 days, reaching a little under 10% alcohol by volume (abv). The wash, as it is known, is first distilled in the wash still to 21% abv. It is then distilled a second time in the spirit still to over 70% abv. Finally, it is reduced to 60% abv for maturation in used (generally ex sherry or bourbon) oak casks.

● Lactic acid bacteria

Malted barley carries a varied microbial load with a predominance of lactic acid bacteria. These Gram-positive bacteria are strictly fermentative organisms that cannot respire using an exogenous electron acceptor. They produce either lactate (homofermentative) or a mixture of lactate, acetate and carbon dioxide (heterofermentative) from glucose catabolism. Consequently,



diversity resulting in *Lactobacillus fermentum* and *Lactobacillus paracasei* as commonly dominant species by about 40 hours. During the final stages when the yeast is dying, a homofermentative bacterium related to *Lactobacillus acidophilus* often proliferates and produces large amounts of lactic acid.

● Effects of lactobacilli on the flavour of malt whisky

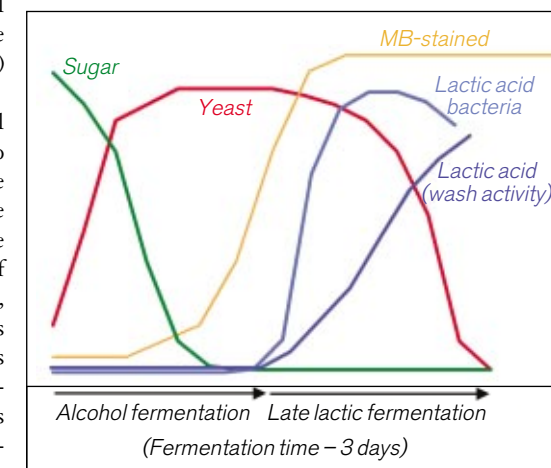
The bacteria can affect the flavour of the spirit in two ways. First, they will reduce the pH of the fermentation through the production of acetic and lactic acids. This will lead to a general increase in esters following distillation, a positive feature that has traditionally been associated with the late lactic fermentation. This general effect is apparent in the data presented in Fig. 3 in which the concentrations of various esters are increased in non-matured (new-make) spirits from laboratory-scale fermentation/distillations with and without lactobacilli. However, lactobacilli might also produce specific flavour compounds that contribute in a unique way to the flavour of the spirit. We have investigated these effects

they do not require oxygen for growth and flourish at relatively low pH (pH 6 to about 3.5). This fits them well for growth in alcoholic beverages in which they have both beneficial (e.g. the malolactic fermentation of wine) and spoilage effects.

While most of these bacteria from malted barley will be killed by the mashing process, some will survive to enter the fermentation (Fig. 1a). Many will also colonize the pipework, heat exchangers and other parts of the distillery plant. If too many occur at the early stage of the fermentation (generally more than 10^6 per ml of wash) they will grow strongly in the fermentation, inhibit the yeast and reduce the alcohol yield. This early lactic fermentation is to be avoided since it reduces the distillery efficiency; these are unwelcome participants in the fermentation. Attention to plant cleanliness is generally sufficient to avoid the early lactic fermentation. In a well-maintained distillery, the numbers of bacteria entering the fermentation are relatively few and bacterial growth is hardly evident during the initial ethanol fermentation stage (Fig. 2). Then, as the yeast dies, after about 36 hours, the bacteria proliferate, growing at the expense of yeast autolysis products, malto-oligosaccharides and pentose sugars (Fig. 1b). Finally, after about 80 hours even the bacteria begin to die (Fig. 1c). The wash is normally distilled after about 60–80 hours, and everything is discharged to the still, wash, yeast and bacteria.

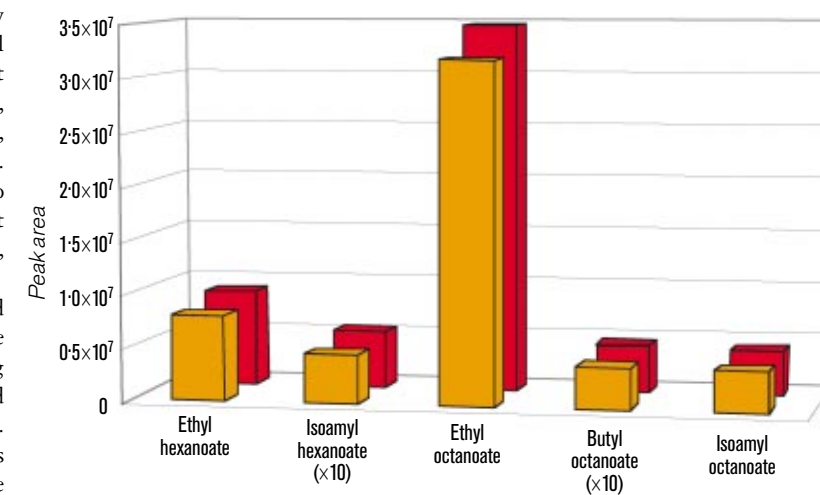
For many years it was thought that the lactic acid bacteria grew homogeneously throughout the fermentation. However, recent studies using denaturing gradient gel electrophoresis (DGGE) have revealed changes in the population as the fermentation proceeds. Initially, a mixed flora of various cocci and rods enters the fermentation. This is followed by a reduction in the

LEFT: Fig. 1. Fluorescence photomicrographs of whisky fermentation samples: green cells are viable, red cells are dead. (a) Wort on entering the washback, (b) after fermentation for 55 hours and (c) after fermentation for 95 hours. (a) AND (b) ARE REPRODUCED WITH PERMISSION FROM VAN BEEK & PRIEST (2002, *APPL ENVIRON MICROBIOL* 68, 297–305); (c) COURTESY F. G. PRIEST



LEFT: Fig. 2. General progress of malt whisky fermentation. 'MB-stained' cells refers to yeast cells that stain with methylene blue and are thus dying or dead. DATA COURTESY HISATO IKEMOTO & TOSHIHIKO TAKATANI, SUNTORY LTD

BELOW: Fig. 3. Effects of *Lactobacillus* in the fermentation on ester concentrations in new-make spirit as detected by GC-MS.



ABOVE: The Suntory Distillery, Osaka, Japan. COURTESY SUNTORY DISTILLERY

Table 1. Effects of some *Lactobacillus* strains on the organoleptic qualities of new-make spirit evaluated by sensory analysis

Species	Strain	Sensory evaluation				
		Estery	Fatty	Fruity	Leafy	Other
<i>L. acidophilus</i>	1		+			Green
<i>L. brevis</i>	1		+			Strong character
<i>L. casei</i>	1		+			Cereal
	2	+				Cereal
	3					Diacetyl-like
<i>L. fermentum</i>	1		+	+	+	
	2	+				
<i>L. pentosus</i> /	1	+		+		Husky
<i>L. plantarum</i>	2	+		+		



PHOTO: JAM ATHERTON, SGM

Further reading

Russell, I. (editor) (2003). *Whisky: Technology, Production and Marketing*. London, San Diego: Academic Press.

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Simpson, K. L., Pettersson, B. & Priest, F. G. (2001). Characterization of lactobacilli from Scotch malt whisky distilleries and description of *Lactobacillus ferintoshensis* sp. nov., a new species isolated from malt whisky fermentations. *Microbiology* 147, 1007–1016.

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by preparing laboratory-scale fermentations with different bacterial strains present and analysing the new-make spirit by gas chromatography-mass spectrometry (GC-MS). The presence of bacteria increased the concentrations of damascenone in the new-make spirit from 24 (peak area) in the control to 41–72 (peak areas) in *Lactobacillus*-containing fermentations with *Lactobacillus acidophilus* having the greatest effect. Damascenone has a floral, herbal, tobacco-like aroma and has been reported to be an important flavour component of whisky. Exactly how the lactobacilli effect these changes are unknown.

However, the flavour changes are introduced, they are noticeable in the spirit as it emerges from the still. This new-make spirit from a series of laboratory fermentations containing various *Lactobacillus* strains isolated from malt whisky fermentations was analysed by a sensory panel. The flavour notes associated with various bacteria are described in Table 1 where it is evident that characteristics like estery, fruity and leafy are among the positive attributes.

Conclusions

Malt whisky flavour involves an enormously complex chemistry derived from the raw materials, the yeast, the distillation process and the oak cask in which the spirit is matured. That lactic acid bacteria can play a minor role in this process has been appreciated for generations, but the organisms involved and their contributions are only now being discovered. Some American Bourbon distillers embraced this many decades ago by adding their own mixtures of lactic acid bacteria to their fermentations to provide consistency of flavour. Perhaps it is time for Scotch distillers to appreciate their uninvited guests and consider a similar practice.

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