Draught beer hygiene: a forcing test to assess quality

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Abstract

The quality of draught beer is important to consumers but can be inconsistent, ranging from excellent through to unacceptable. The few but dated studies of draught beer quality have focused on the number of microorganisms that are present in the product. Work reported here, suggests that this approach has its limitations and fails to relate to beer quality post-dispense. An alternative approach using the long-established 'forcing' method provides a better but still retrospective assessment of draught beer quality. Samples post dispense are 'forced' by static incubation at 30°C for four days and beer quality is ranked by the measurement of absorbance at 660nm. The increase in absorbance reflects the growth of beer spoilage microorganisms present in the beer at dispense. Four quality bands are proposed, where quality is described as excellent (absorbance increase of < 0.3), acceptable (0.3-0.6), poor (0.6-0.9) and unacceptable (> 0.9). The method is straightforward, requires no special skills and enables, for the first time, the robust quantification of draught beer quality. It is anticipated that the method will have widespread application in the measurement and improvement of the quality of draught beer.

Additional supporting information can be found in the online version of this article at the publisher's website.

Keywords: dispense; beer spoilage; quality; method

Short title: A simple method to assess draught beer quality

Introduction

The mix between the on-trade/on-premise (pubs, bars and restaurants) and the off-trade/off-premise (supermarkets, shops) varies widely across the world. In 2014 *(1)*, the major on-trade markets include Ireland and Spain (64%), the UK (50%), Japan and South Korea (48%) with the Czech Republic, Italy, Belgium and Australia accounting for 40-45%. Globally, draught beer in on-trade licensed premises accounts for 7% or more of the worldwide market although, in most countries, volumes are either static or in decline *(2).* This reflects a host of factors – political, economic, social and technological - that impact the on-trade *(3).* Of these, poor or compromised draught beer quality is an important consideration, which in turn, is exacerbated by comparatively high pricing.

For beer, quality has been defined as ‘meeting the customer requirements’ *(4)* and ‘the achievement of consistency and elimination of unwanted surprises’ *(5).* More specifically, from the perspective of consumers *(6)*, beer quality reflects parameters such as colour, aroma, alcohol content, haze, foam, flavour and gas content. For beers packaged into bottle and especially can, such criteria are broadly stable. However, draught beer in kegs and cask, which - on packaging are of excellent quality – deteriorates ‘on dispense’. This reflects a mix of dispense parameters including product temperature, gas management, throughput and poor hygienic practices. Of these, temperature (too high or too low) and dispense gases (wrong mixture, product pick-up, pressure too high or too low) impact on brand presentation, dispense delivery and losses of product.

Hygiene and the associated microbiological risk are managed by regular and effective alkaline line cleaning to remove microbial biofilms *(2).* In addition, keg couplers and taps *(2)* together with nozzles (7) should be cleaned and sanitised to minimize contamination of the system. Deteriorating dispense hygiene results in the growth and metabolism of beer spoilage microorganisms which contribute to changes in beer flavour and aroma together with the appearance - *in extremis* - of haze and ‘bits’.

Consumers ‘drink as much with their eyes as with their mouth’ *(8)* having expectations of appearance such as foam and clarity. Shifts in beer flavour and aroma due to the formation of compounds such as diacetyl, esters and acetic acid are viewed by consumers as being ‘different’, ‘off’ or ‘wrong’. Whatever the interpretation or understanding, some (but not all) consumers will ‘vote with their feet’, leave the outlet, tell their friends or blame the brand and move to a different beer. Indeed, it is reported that ‘nearly 95% of consumers would stop using a pub if beer quality was constantly poor’. Conversely from the same report, ‘for nearly 90% of consumers, beer quality is essential or very important when selecting a venue’ *(9).*

Intuitively, draught beer of excellent quality ‘sells’. Anecdotal reports suggest beer of high quality leads to an uplift in sales (reportedly 10%) over beer of indifferent to poor quality. Hard evidence though is limited. One report *(3)* from a UK retailer links the frequency of line cleaning with the commercial performance of licensed premises. In terms of volume growth, cleaning every two weeks results in marginal (0.1%) growth with weekly (UK best practice) cleaning resulting in almost 2% uplift. Conversely frequencies of between two and four weeks lead to about a 2% loss of volume. Despite the financial benefit, take up of best practice is patchy with an estimated third of UK draught beer dispensed through dirty lines *(9*).

Assessment of draught beer quality in the UK on-trade has mostly focused on cask beer and is essentially qualitative. Cask Marque *(10)* provides an independent, empirical assessment of cask ale quality based on Assessors visiting subscribing outlets at least twice a year. Visits are unannounced and involve a yes/no measurement of temperature, clarity and, a sip test, to assess flavour and aroma. Accreditation to Cask Marque is communicated to consumers via a plaque or can be searched online via the CaskFinder app.

A recent review *(2),* reported that the brewing literature on draught beer quality is slight, with only 12 or so publications since the 1950's. However, reflecting the global reach of draught beer, these publications are from the UK, Finland, Germany, USA and Spain. Most studies on draught beer quality have used microbiological plate tests although ATP bioluminescence has also found application more recently.

Measurement of microbial loading does not easily relate to the consumer experience or to beer quality. The few studies *(2,11,12,13)* have reported a range of values for commercial draught beer ranging in quality from 'good' (< 1000 colony forming units (cfu) per millilitre of beer) to 'unacceptable' (> 50,000 cfu/ml). These figures are aligned to the recommendations of the Deutsches Institut für Normung (DIN) 6650 standard 'dispense systems for draught beverages' *(14)*. Part 6 of this German standard covers 'requirements for cleaning and disinfection' and provides guidelines for the extent of microbial loading. Here 'a typical guideline value for a positive result with respect to microbial contamination would be 1000 cfu/ml, a value of more than 50,000 cfu/ml being considered unacceptable. If the count is 10,000 or higher, cleaning is necessary'. No guidance is given as to testing methodology, as the standard is generic for draught beverages including beer, wine, water, carbonates etc.

In this work and for the first time, draught beer quality has been quantified by 'forcing' samples post dispense. Such accelerated shelf life testing by storage at elevated temperatures has long been used in the brewing industry to assess the microbiological stability of beer in process and more recently to assess the hygiene of dispense tap nozzles *(2)*. 'Forcing' was first developed by Horace Brown *(15)* in the early 1870's to predict the spoilage of Burton ales brewed between October and May for sale in the Summer when, in the absence of refrigeration, there was no brewing. Samples of beer could be stored 'under such conditions of temperature as would hasten the development of any of the adverse bacterial changes to which the beer was liable when stored under the ordinary conditions which rule in practice' *(15)*. The method involved newly racked beer being stored at (24-29°C) for between 10 days and three weeks. After forcing, beers were examined for flavour, clarity, present gravity, acidity and microscopically *(16).* As noted by Kulka *(17),* the environment during forcing with a mixed microflora gradually changes. Accordingly, the beer during forcing becomes a 'better medium for growth, allowing development of some organisms at the end of the forcing period which were initially incapable of growth' *(17).*

Here, forcing of draught beer samples was performed by incubating samples post dispense statically at 30°C for 4 days. The clarity of the samples - before and after incubation - was determined by measurement of absorbance.

Materials and methods

Microbiological media were obtained from Oxoid and cycloheximide (0.1%, w/v) from Sigma Aldrich.

Beer samples (250 ml) post dispense were purchased from on-trade licensed premises (pubs and bars) in Burton-on-Trent, Derby, Loughborough, Market Harborough and nearby villages. Beers were UK-wide keg brands including lagers (two categories of abv, ≤ 4.1% abv and > 4.1% abv), keg ale (≤ 4.1% abv and > 4.1% abv) and stout (4.2% abv). Other products included nationally available and local cask-conditioned beers, wheat beers, local craft/microbrewery beers and keg cider (abv ≤ 4.1% abv and > 4.1% abv). Table S1 (in the on-line supporting information) provides an overview of the licensed premises and the beers that were sampled as part of method development. Post sampling, all activities were performed aseptically.

Method for draught beer quality

The method was used to assess keg beers (lagers, ales and stouts), keg ciders, cask beers and 'craft' unfiltered and unfined keg beers. On-trade samples were transferred *ex* tap or from glassware to sterile 250 ml Duran bottles. Samples were kept cold in transit and either processed on the day or stored overnight (4-6°C) before processing. After thorough mixing, 2 x 25 ml was transferred to plastic Universal bottles, the cap located on top (but not tightened – to allow gas transfer) and incubated statically at 30°C. Cycloheximide (4mg/L) was added to cask, unfiltered and unfined beers to suppress the growth of primary *Saccharomyces* yeasts.

Forced samples were thoroughly mixed by inversion – recalcitrant sediments were resuspended with a sterile plastic loop and well mixed. The absorbance of the samples was measured in duplicate at 660 nm (Jenway spectrophotometer 7315) in duplicate (1 ml) at the beginning of incubation and after *ca*. 96 hours at 30°C.

The absorbance of the sample was proportional to the degree of light scattering by suspended particles (yeast and bacterial cells, flocs, flakes etc.) The chosen wavelength has long been used to quantify yeast cultures *(18).* Heavily contaminated dark beers were diluted (1:1) with water prior to measurement of absorbance.

There are numerous practical and operational unknowns in the on-trade that impact on draught beer quality. Accordingly, it is recommended that sampling of draught beers in a licensed account is performed more than once. To avoid 'first runnings' - which typically have a higher microbial load - samples were taken during busy trading sessions and not on opening.

Quality bands

The 'quality' of the beer post dispense was determined from the difference in absorbance of the two samples measured the beginning of incubation and after *ca*. 96 hours at 30°C. The increase in absorbance was used to classify the samples into four bands; A (0-0.3), B (> 0.3-0.6), C (> 0.6-0.9) and D (> 0.9) (Figure 1). The change in turbidity reflected the microbiological 'quality' at dispense, such that the A category ('excellent') with relatively little change in absorbance was superior to B ('acceptable') and which in turn was better than C ('poor') with D being of 'unacceptable' quality.

Quality index

For groups of samples (e.g. sampling all the taps on the bar), a 'quality index' was calculated from the sum of the individual quality bands (where A = 4, B = 3, C = 2, D = 1) divided by (number of samples x 4) x 100. If all samples are measured as excellent/quality band A, the quality index is 100%.

Microbiology

Samples post dispense from 24 public houses were diluted 10^{-1} and 10^{-2} and post forcing 10^{-4} and 10^{-5}. For each dilution, 0.1 ml (in duplicate) were spread onto selective agars. Raka Ray (with cycloheximide, 10 mg/L) plates were incubated anaerobically (Oxoid Anaerogen) for five days at 30°C and WLN (Wallerstein Laboratory Nutrient) incubated aerobically for two days at 25°C.

Results and discussion

Forcing in Universal bottles

Disposable Universal bottles are of a standard shape (9 x 2.5 cm, with a conical base) and volume (30 ml), sourced in either polystyrene or polypropylene with plastic screw caps and or with a proprietary 'flow seal' cap to provide 'excellent sample containment'.

Beer spoilage assessed through 'forcing' depends on the mix of the spoilage microbiome and sufficient nutrients in the beer to the support growth of contaminating microorganisms. The availability of oxygen supports the growth of aerobic microorganisms in the microflora. However, under conditions of static incubation, transfer of air into the forced beer would be modest. The use of Universals with a 'flow seal' cap resulted in a lower value for draught beer forcing compared to the same samples processed with a loosely positioned standard screwcap.

To explore this further, draught samples - stout and lager (> 4.1% abv) - from the same outlet were forced in triplicate in Universals made of different materials (glass, polypropylene and three sources of polystyrene, including the flow seal cap) with the cap either tight and loosely positioned. The mean OD_{660} difference for the stout was 1.405 (open) compared to 0.933 (closed) whereas for the premium lager was 1.006 (open) and 0.471 (closed). The Student's T-test (two tailed) showed the results between open and closed lids were significantly different at the $P < 0.001$ level confirming the need for the caps to be loosely positioned on the Universal.

Reproducibility

Whilst 'forcing' is a long-established method, its reproducibility in the context of draught beer quality needed to be validated. In all, 12 keg beers (7 x lager (abv ≤ 4.1%), 2 x lager (> 4.1% abv), 3 x ale (abv ≤ 4.1%)) from five public houses were assessed in quintuplicate. Table 1 reports the mean (± standard error) of the absorbance of individual samples post forcing at 30°C for four days. The reproducibility of the method is clear, with a consistent quality band for the five replicates of the 12 different samples of forced draught beer.

Incubation – static v mixed

Initially, the impact of mixing was assessed with 22 keg lagers (abv ≤ 4.1%) and 12 keg ales (abv ≤ 4.1%) from 16 public houses. Samples were forced for four days either statically or with daily inversion. A more detailed experiment was then performed with 11 draught lagers (abv ≤ 4.1%) and nine draught ales (abv ≤ 4.1%) from 10 public houses. The change in turbidity was assessed after four days with daily mixing and statically after four, five, six and eight days. A Student's T-test (two tailed) showed no significant difference between daily mixing and static incubation (P = 0.97).

Incubation – impact of time

Further work (Figure 2) suggests the impact of mixing though is marginal with a small shift in the mixed samples from quality band A to B compared to static. Importantly, the C and D quality bands were the same with or without mixing. However, extending the time of static incubation from four up to a maximum of eight days had a marked impact. As might be anticipated, increasing the time of incubation reduced the number of 'excellent' (A band) samples and progressively increased the 'poor' (C) and 'unacceptable' (D) categories. This is reflected by calculation of the quality index which declined with time from 80% after four days (81% static, 79% daily mixing) to 71% (five days), 70% (six days) and 64% (eight days).

Processing and overnight storage

Processing of trade samples on the same day as they were sampled was not always possible. On such occasions, bulk samples were stored overnight at 4°C. It would be anticipated that such a treatment would not impact significantly on the outcome of the forcing test. To confirm this (or not) the forcing of ten beers (and a cider) were compared with and without storage, 17 of the 20 results were unchanged with three changing by one quality band (two up and one down). A Student's T-test (two tailed) showed no significant difference between samples processed on the same day as sampling or after overnight cold storage (P = 0.53).

Suppressing the growth of brewing yeasts

The forcing test predicts quality by amplifying the indigenous yeasts and bacteria present in the beer post dispense. Unlike filtered keg products, cask and unfiltered/unfined draught beers will contain primary brewing yeasts because of the beer style and not because of poor dispense hygiene. Clearly, the presence of brewing yeasts can grow in a forcing test and distort the measurement of draught beer quality. To suppress the growth of primary yeasts, cycloheximide (*aka* actidione) was added prior to forcing. This antibiotic has long been used in microbiological media to suppress the growth of brewing yeasts whilst allowing the growth of 'wild' yeasts and bacteria *(19)*. As ever with brewing microbiology, things are not black and white. The inclusion of cycloheximide *(20)* can suppress the growth of *Saccharomyces* wild yeast (e.g. *S. diastaticus*) which have been reported to be 'prolific beer spoilage microorganisms' responsible for trade returns of draught beer *(21)*. However, the addition of cycloheximide does allow the growth of non-*Saccharomyces* contaminants *(20)* such as *Brettanomyces, Pichia, Candida and Hansenula,* which have been reported in draught beer *(22,23,24).*

Analysis of cask beers, wheat and unfiltered beers from four public houses with and without the addition of cycloheximide (4 mg/L) suggested that the inclusion of the inhibitor resulted in marginally more spoilage but this was dependent on the public house. Indeed, a Student's T-test (two-tailed) showed that addition of cycloheximide did result in a significant difference at the $P \leq 0.05$ level.

This is not surprising and reflects the likely diversity and mix of the spoilage microbiome within and between licensed premises. However, despite these complexities, it is suggested that the addition of cycloheximide should be routinely added to samples of cask, unfiltered and unfined beers post dispense and prior to using the forcing test.

Microbial loading post dispense and DIN 6650-6

The DIN standard recommendations for the microbiological loading of beverages (in this case beer), provides a framework for the assessment of post dispense quality. Four categories of loading are detailed in the standard (i) < 1000 cfu/mL, (ii) 1000-10,000 cfu/ml, (iii) 10,000-50,000 cfu/ml and (iv) > 50,000 cfu/ml. Table 2 reports the four categories, in terms of aerobic and anaerobic selective agars for the work reported here. Here, an average of 32.5% of the samples were in the 'positive result' category with 32% in the 'acceptable' category. However, 18% were in the 'cleaning is necessary' band, with 17.5% 'unacceptable'. The highest microbial count of the samples reported here (in the > 50,000 cfu/ml category), was 300,000 cfu/ml although up to 1,000,000 cfu/ml have been reported in draught beer samples in Germany *(11)* and Finland *(12)*.

Although microbiological loading is the metric that the DIN standard uses to assess the quality of draught beverages, there are no recommendations regarding media or incubation conditions. Accordingly, as noted above, there are different interpretations of draught beer 'quality' in the trade depending on whether the media is – for example - WLN incubated aerobically or Raka Ray incubated anaerobically. The major draught beer spoilage organisms *(2,12)* have a mixed response to the presence of oxygen, and include the aerotolerant anaerobic bacteria (*Lactobacillus*, *Pediococcus*), aerobic bacteria (*Acetobacter*, *Gluconobacter*), facultatively aerobic yeasts (*Saccharomyces*, *Brettanomyces*) and aerobic yeasts (*Pichia*, *Candida*). However, which predominate to spoil draught beer reflects the microbial loading/mix together with the beer composition (nutrients, iso-alpha acids, pH etc), the concentration of carbon dioxide and availability of dissolved oxygen. In terms of processing, keg beer is assumed to be effectively anaerobic whilst cask beer can pick up oxygen as the container is dispensed. Despite this, container couplers and connectors together with taps and nozzles are hot spots for contamination *(2)* and are aerobic environments.

Microbial loading post dispense v forcing

The microbiological loading and the forcing test are quantified by the measurement of cell numbers. Both approaches require a period of incubation of two to seven days (microbiology) or four days (forcing). The methods differ, in that plate counts reflect the viable organisms detectable on

selective agars on sampling, whereas the forcing approach amplifies the number of yeast and bacteria able to grow in the beer *ex* dispense. Despite these differences, the DIN microbiological approach and the forcing test, both categorise beer quality through microbiological loading (directly or indirectly) from low (good quality) to high (bad quality).

In addition to reporting the DIN classification for trade samples, Table 2 also the same trade samples assessed using the forcing test. Of the 52 samples, Table 2 shows that 35% of the population were in quality band A, 44% in B, 17% in C and 4% in D. Subjectively, 4% of the samples being assessed as 'unacceptable' is a more realistic measure than the 18% flagged by the DIN approach. Linear regression analysis of the two approaches shows the best correlation (R^2 = 0.8737, y=2.0024x-25.059) with the combined results (aerobes and anaerobes) from the DIN categories against that of forcing. Despite this, the relationship is skewed such that the DIN approach of microbiological loading underestimates quality bands A (excellent) and B (acceptable) but overestimates bands C (poor) and D (unacceptable). A likely explanation is that microbiological testing quantifies a mix of microorganisms some of which are 'environmental' and accordingly are unable to spoil beer.

Microbial loading post dispense v forcing – an explanation

Conventional 'traditional' microbiological testing has its limitations. One limitation is that there is no universal microbiological medium, so to build a picture, different agars are used to 'select' for different microorganisms. In this work, WLN is selective for aerobes (yeast and acetic acid bacteria) whereas Raka Ray is used to quantify anaerobes (lactic acid bacteria). However, this is complicated by being unable to confidently extrapolate growth on a plate to the spoilage of beer. Microbiological testing is directional but offers no guarantees of robustness and accuracy. This is further compromised by considerations which contribute to microorganisms being unable to grow on selective agars. This can be due to poor growth rate, the recovery and growth of nutritionally fastidious microorganisms, viable but non-culturable organisms such as *Lactobacillus* species *(25)* and the loss of support (e.g. trading nutrients, quorum sensing etc) from the microbiome of the sample. Accordingly, quantification of microorganisms on agar plates does not necessarily mean those organisms can grow in/spoil beer *in situ*. Indeed, these results challenge the relationship between conventional microbiological analysis in breweries and the relevance of such analysis to possible spoilage.

The forcing test quantifies the increase in absorbance due to the growth of microorganisms in beer. This may be compounded by the cell size and shape of spoilage microorganisms that impact on light scattering and therefore absorbance. Although spoilage is (invariably) from a consortium of diverse yeasts and bacteria, there will be occasions where sample turbidity is skewed by the mix of large (yeast) and small (bacterial) cells.

Application of the method

Whilst 'best practice' in draught beer dispense is increasingly defined and communicated *(2),* measurement of quality has attracted little attention. Indirect measurement of poor quality through 'losses' can be commercially

relevant and can result in the loss of business and ultimately closure. The method reported here is simple (although taking four days) but provides real differentiation of beer quality based on forcing the indigenous microorganisms present at the point of dispense. Accordingly, this method provides a tool to assess beer quality in the on-trade/on-premise against a variety of parameters both routine and in response to changing practices. Obvious comparisons include (i) public houses within retailer groups, (ii) brands within and between public houses and (iii) the impact of outlet factors. Beyond the routines, the method will add value in assessing and validating the impact of innovation such as (i) line cleaning solutions, (ii) line cleaning frequency and technologies, (iii) dispense line composition (including FOB detectors) and (iv) end to end cooling of beer from keg to tap. The method would also lend itself to a quantifiable (rather than qualitative yes/no) assessment of beer quality in the on-trade licensed premises. Handled appropriately this method could add real value to consumers and their understanding of the importance of beer quality.

Validation of the method

The method reported here has been successfully used to assess the quality of draught beer in > 65 public houses and > 500 samples of beer.

Conclusions

Quantitative assessment of draught beer quality that relates to the consumer experience has not been reported. The method provides a direct assessment of the microbiological status of beer at the point of dispense which is then subsequently amplified by forcing. Beer of excellent quality contains low numbers of beer spoilage organisms which on incubation at 30°C develop little turbidity (A_{660} < 0.3, quality band A). Conversely forcing beer of poor microbiological quality results in high turbidity with A_{660} > 0.9 (quality band D).

The method does not identify the source of contaminating microorganisms but reflects the total dispense system. Accordingly, there is no insight into potential hotspots of contamination although the dispense line, fob detector and nozzles are typically the primary candidates *(2).* In addition, the beer itself and, possibly, glassware may also contribute to the microbial mix that presents on forcing.

The forcing method presents a different picture to the measurement of microorganisms on agar plates. It is proposed, that despite the advocacy of the DIN standard, microbiological testing is an unsatisfactory approach to describe draught beer quality. This reflects complex factors that may – on the one hand - exaggerate the microbial loading through the use of selective microbiological media or – on the other - underestimate viable but non-culturable organisms. The forcing method described here quantifies those microorganisms in draught beer that can grow and spoil beer. Accordingly, the forcing test is recommended as a simple method to quantity draught beer quality.

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Supporting information

Additional supporting information on the public houses and beers sampled in this work may be found in the online version of this paper at the publisher's web site.

Table 1: Reproducibility of the forcing method.

On-premise	Beer	Mean A_{660}	± sem	Quality band
B1	Lager (≤ 4.1% abv) (SL 3)	0.120	0.012	5 x A
	Lager (≤ 4.1% abv) (SL 6)	0.535	0.008	5 x B
B2	Lager (≤ 4.1% abv) (SL 3)	0.146	0.028	5 x A
	Ale (≤ 4.1% abv) (SKA 5)	0.517	0.012	5 x B
B6	Lager (≤ 4.1% abv) (SL 3)	0.419	0.026	5 x B
	Lager (≤ 4.1% abv) (SL 4)	0.314	0.005	5 x A
B8	Lager (>4.1% abv) (PL 1)	0.810	0.010	5 x C
	Lager (>4.1% abv) (PL 5)	0.091	0.005	5 x A
	Lager (≤ 4.1% abv) (SL 3)	0.154	0.006	5 x A
	Ale (≤ 4.1% abv) (SKA 5)	0.713	0.014	5 x B
B9	Lager (≤ 4.1% abv) (SL 3)	0.073	0.032	5 x A
	Ale (≤ 4.1% abv) (SKA 1)	0.680	0.026	5 x B

- Public houses were sampled in Burton-on-Trent (B). Keg beers are described as lager (≤ 4.1% abv) (SL), lager (>4.1 abv) (PL) and ale (≤ 4.1% abv) (SKA).
- An overview of the brands and licensed premises can be found in Table S1 (in the on-line supporting information).

Table 2: Microbial loading v forcing

Microbiology					Forcing		
cfu/mL	DIN Description*	% Aerobes	% Anaerobes	% Combined	Band	Description	%
< 1000	Positive	25	40	32.5	A	Excellent	35
1-10,000	Acceptable	35	29	32	B	Acceptable	44
10-50,000	Cleaning required	19	17	18	C	Poor	17
> 50,000	Unacceptable	21	14	17.5	D	Unacceptable	4

- Samples (52) post dispense were taken from 24 licensed premises and represent 15 different brands of keg lager, ale and cider together with cask ale (details are reported in Table S1 in the on-line supporting information).
- The DIN standard 6650-6 defines the microbial counts (as cfu/ml) (i) < 1000 as a ‘positive result’, (ii) 1-10,000 as ‘acceptable’, (iii) 10-50,000 ‘cleaning is necessary’ and (iv) > 50,000 as ‘unacceptable’.
- ‘Aerobes’, ‘anaerobes’ and ‘forcing’ are as defined in the Materials and Methods.